UNIVERSIDADE FEDERAL DE UBERLÂNDIA
INSTITUTO DE BIOTECNOLOGIA
PROGRAMA DE PÓS-GRADUAÇÃO EM BIOTECNOLOGIA

FLÁVIO CÉSAR THIAGO

IDENTIFICAÇÃO E CARACTERIZAÇÃO DE miRNAs E GENES ENVOLVIDOS NA VIA DE PROCESSAMENTO DE miRNAs EM *Pomacea canaliculata* E *Pomacea maculata*

PATOS DE MINAS – MG

AGOSTO - 2021

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Dissertação de mestrado apresentada ao Programa de Pós-graduação em Biotecnologia como requisito final para obtenção do título de mestre.

Orientador Prof. Dr. Matheus de Souza Gomes
Coorientador Prof. Dr. Fábio Ribeiro Queiroz

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T422
2021

Thiago, Flávio César, 1977-
miRNAS EM Pomacea canaliculata E Pomacea maculata: IDENTIFICAÇÃO, CARACTERIZAÇÃO E MAQUINARIA DE PROCESSAMENTO [recurso eletrônico] / Flávio César Thiago. - 2021.

Orientador: Matheus de Souza Gomes.
Dissertação (Mestrado) - Universidade Federal de Uberlândia, Pós-graduação em Biotecnologia.
Modo de acesso: Internet.
Disponível em: http://doi.org/10.14393/ufu.di.2021.417
Inclui bibliografia.
Inclui ilustrações.

1. Biotecnologia. I. Gomes, Matheus de Souza,1981-, (Orient.). II. Universidade Federal de Uberlândia. Pós-graduação em Biotecnologia. III. Título.

CDU: 60

Bibliotecários responsáveis pela estrutura de acordo com o AACR2:

Gizele Cristine Nunes do Couto - CRB6/2091

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Programa de Pós-Graduação em:	Biotecnologia				
Defesa de:	Dissertação de Mestrado Acadêmico				
Data:	20 de agosto de 2021	Hora de início:	9:30	Hora de encerramento:	11:53
Matrícula do Discente:	41912BTC007				
Nome do Discente:	Flávio César Thiago				
Título do Trabalho:	IDENTIFICAÇÃO E CARACTERIZAÇÃO DE miRNAs E GENES ENVOLVIDOS NA VIA DE PROCESSAMENTO DE miRNAs EM Pomacea canaliculata E Pomacea maculata				
Área de concentração:	Biociência				
Linha de pesquisa:	Bioinformática e Biologia Molecular aplicada à genômica, transcritômica e proteômica				
Projeto de Pesquisa de vinculação:	Bioinformática e Biologia Molecular aplicada à genômica, transcritômica e proteômica				

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Dissertação de mestrado apresentada ao Programa de Pós-graduação em Biotecnologia como requisito parcial para obtenção do título de mestre.

Aprovação: ___/______/_____

BANCA EXAMINADORA

____
Prof. Dr. Matheus de Souza Gomes
(Presidente)

____
Prof^{a}. Dr^{a}. Enyara Rezende Morais
(Membro interno)

____
Prof^{a}. Dr^{a}. Fernanda Janku Cabral
(Membro externo)

AGRADECIMENTOS

A Deus por ter me concedido a graça de chegar até aqui nesta caminhada.

Ao meu pai Joaquim Thiago por ser exemplo de luta, determinação, força e ter nos dado a oportunidade de estudar. Estar sempre presente nos momentos de alegria e tristeza. Homem de rotina inflexível que dedicou a vida em prol da família.

A minha eterna mãe: Marina (*in memorian*), como eu gostaria que a senhora estivesse aqui comigo neste momento. Devo a senhora a minha educação, as cobranças das tabuadas, as leituras dos ditados na pré-escola e todo o seu esforço para nos formar homens.

Aos meus irmãos incansáveis batalhadores Alex e Lucas. Toda inspiração, exemplo e toda a minha gratidão também. Alex por ter sido o pioneiro, Lucas por ter dado continuidade ao sonho que nos une.

A minha filha Ana Júlia, meus sobrinhos e afilhados João Victor e ao pequeno Miguel, a qual com certeza herdarão o nosso legado e também minhas cunhadas.

Minha esposa Gaby a qual agradeço de coração. Obrigado por ter vivido cada minuto desta jornada. Compartilhado cada pequena vitória, cuidado de minha saúde mental, abdicado seu tempo para viver este momento ímpar. Se teria palavra para definir o amor, essa palavra seria abdicação, cuidado, dedicação; essa palavra seria Gaby.

Aos colegas da Coordenação de Assistência Farmacêutica da Superintendência Regional de Saúde de Patos de Minas. Todo esforço para manter a equipe coesa na minha ausência e também pelas orações.

A professora Enyara Rezende, pelos conselhos e por me guiar a mudar o rumo da minha trajetória acadêmica. A professora Cristina Ribas a qual levarei seus ensinamentos por toda a minha vida. Raphael, pela ajuda e compartilhamento do artigo. Carlos, pelas “lives” produtivas. Aos mestres e doutores que publicaram artigos que nos inspiravam Fábio Queiroz, Thaís Cunha e finalmente o professor Matheus de Souza Gomes, idealizador do projeto, que me acolheu e se colocou à disposição 24 horas, pela humildade, conhecimento, positividade, artigos. A UFU por ter proporcionado ensino de qualidade e gratuito. A todos muito obrigado, gratidão....

RESUMO

Os miRNAs são pequenos RNAs não codificadores de proteínas que auxiliam e atuam no controle da expressão gênica impedindo a tradução de diversos genes (em sua maior parte). É consenso na literatura científica que os mecanismos de controle da expressão gênica são essenciais para o desenvolvimento do organismo orquestrando vários processos biológicos. A análise de miRNAs em moluscos, sobretudo em caramujos, é muito importante já que possuem poucos miRNAs identificados, quando comparado ao vasto número de espécies de moluscos. Os caramujos do gênero *Pomacea* são considerados espécies invasoras prejudiciais ao planeta incluindo *P. canaliculata* e *P. maculata*. A família Ampullariidae em moluscos constitui um modelo emergente para estudos evolutivos devido à alta diversidade, história antiga e ampla distribuição geográfica. Dessa forma, o objetivo deste trabalho foi identificar e caracterizar através de análises *in silico* miRNAs maduros conservados e seus respectivos precursores, as prováveis proteínas envolvidas na via de processamento de miRNAs e seus respectivos genes, utilizando banco de dados públicos de genoma e transcriptoma de *P. canaliculata* e *P.maculata.* Foi possível identificar 269 pré-miRNAs preditos, 296 miRNAs maduros e 7 prováveis proteínas da via de miRNAs em *P. canaliculata.* Já em *P. maculata* foram identificados 299 precursores, 316 miRNAs maduros e 8 prováveis proteínas da biogênese de miRNAs. A caracterização foi realizada baseada na conservação filogenética, na estrutura primária e secundária característica dos precursores e das proteínas da via de miRNAs. Os dados obtidos embasarão estudos de filogenia, divergência populacional, especiação e padrõesde diversidade na família Ampullariidae.

Palavras chave: miRNAs. Análise Computacional. Caramujos. Ampullariidae. Genoma.

ABSTRACT

miRNAs are small non-protein coding RNAs that help and act in the control of gene expression, preventing the translation of several genes (for the most part). There is a consensus in the scientific literature that gene expression control mechanisms are essential for the development of the organism, orchestrating several biological processes. The analysis of miRNAs in molluscs, especially in snails, is very important since they have few identified miRNAs, when compared to the vast number of mollusc species. The snails of the genus *Pomacea* are considered invasive species harmful to the planet, including *P. canaliculata* and *P. maculata*. The Ampullariidae family in molluscs is an emerging model for evolutionary studies due to its high diversity, ancient history and wide geographic distribution. Thus, the objective of this work was to identify and characterize, through in silico analysis, conserved mature miRNAs and their respective precursors, the putative-putative proteins involved in the miRNA processing pathway and their respective genes, using public database of genome and transcriptome of *P. canaliculata* and *P. maculata*. It was possible to identify 269 predicted pre-miRNAs, 296 mature miRNAs and 7 probable miRNA pathway proteins in *P. canaliculata*. In *P. maculata*, 299 precursors, 316 mature miRNAs and 8 probable miRNA biogenesis proteins were identified. The characterization was performed based on phylogenetic conservation, primary and secondary structure characteristic of precursors and proteins of the miRNA pathway. The data obtained will support studies of phylogeny, population divergence, speciation and diversity patterns in the Ampullariidae family.

Keywords: miRNAs. Computational Analysis. Snails. Ampullariidae. Genome.

LISTA DE FIGURAS

LISTA DE TABELAS

SUMÁRIO

1 INTRODUÇÃO

1.1 PROBLEMA DE PESQUISA

Os miRNAs exercem mecanismos de controle gênico orquestrando vários processos biológicos. Diante disso, quais miRNAs poderão ser identificados nas espécies do gênero *Pomacea* em estudo? Se é possível a identificação de miRNAs nas espécies citadas, quais os genes envolvidos na biogênese dos miRNAs?

1.2 HIPÓTESES

Os miRNAs, precursores e maduros, e os genes envolvidos na via de processamento de pequenos RNAs estão presentes no genoma e transcriptoma de *P. canaliculata* e *P. maculata* e esta descoberta poderá embasar estudos futuros a respeito de doenças e pragas agrícolas nos quais os caramujos estão envolvidos.

1.3 OBJETIVOS

1.3.1 Objetivo Geral

O objetivo deste trabalho foi a identificar e caracterizar miRNAs e genes envolvidos em sua via de processamento, tanto no genoma quanto no transcriptoma das espécies de moluscos *P. canaliculata* e *P. maculata*.

1.3.2 Objetivos específicos

a) Identificar e caracterizar miRNAs conservados, maduros e precursores, no genoma e transcriptoma de *P. canaliculata*.
b) Identificar as prováveis proteínas envolvidas na via da biogênese de miRNAs em *P. canaliculata* e *P. maculata*
c) Caracterizar as prováveis proteínas da via de miRNAs em *P. canaliculata* e *P. maculata,* através do alinhamento de proteínas, análise de domínios conservados, sítios ativos e filogenia.

1.4 JUSTIFICATIVA

As duas espécies de caracóis *P. canaliculata* e *P. maculata* são muito semelhantes em características morfológicas e adaptativas, apresentando crescimento rápido, alta taxa de reprodução, tolerância ao estresse ambiental, resistência a altas temperaturas e seca. As duas espécies de moluscos são consideradas pragas para algumas culturas agrícolas.

Para tanto, os estudos relativos aos miRNAs contribuirão para uma melhor compreensão a respeito da adaptabilidade e biologia de *P. canaliculata* e *P. maculata.* Isto é possível devido a capacidade dos pequenos RNAs não codificantes de proteínas de interferir no perfil de expressão de genes específicos relacionados a possíveis processos de resistência e plasticidade ambiental da espécie.

P. canaliculata tem a propriedade de realizar o controle biológico de *B. glabrata*, podendo alterar a transmissibilidade da esquistossomose em humanos. A maneira que os miRNAs de *P. canaliculata* interferem no perfil de expressão de genes poderá fornecer informações importantes para o desenvolvimento de estratégias mais eficientes para o controle da transmissão da esquistossomose. O estudo de miRNAs em *P. canaliculata* poderá conferir inúmeras respostas ao processo de interação do molusco com o nematódeo *A. cantonensis*, fornecendo mecanismos para compreensão da transmissão da meningite eosinofílica em humanos e prováveis instrumentos de controle da patologia.

Identificar miRNAs em *P. canaliculata* e *P. maculata* fornecerá embasamento para trabalhados futuros, dentre estes o possível mecanismo de controle que os miRNAs destes moluscos exercem na invasividade das culturas agrícolas. Nossos resultados ampliarão o estudo de miRNAs em moluscos, propiciando novos conhecimentos para o entendimento da regulação de miRNAs na família Ampullariidae e nas espécies *P. canaliculata* e *P. maculata.*

2 REFERÊNCIAL TEÓRICO

2.1 MOLUSCOS

Moluscos são animais de corpo mole invertebrados não segmentados. O corpo é organizado em um pé musculoso, cabeça e massa visceral que contém a maioria dos sistemas orgânicos. Alguns têm um manto que secreta a concha calcária (BROWN; LYDEARD, 2010). O filo Mollusca é o segundo com maior diversidade de espécies dentre os metazoários e o mais rico em subgrupos. Os habitats podem ser marinhos, de água doce ou terrestres. As espécies podem viver em temperaturas de até 40°C ou ambientes com gelo permanente (HASZPRUNAR; WANNINGER, 2012).

O filo Mollusca é o mais morfologicamente distinto dentre os outros filos, sendo composto por oito classes Chefalopoda (polvos, lulas, etc), Gastropoda (caracóis, lesmas, etc) Bivalvia (ostras, mexilhão,etc), Monoplacophora (moluscos marinhos que vivem em altas profundidades), Solenogastres, Caudofoveata, Polyplacophora e Scaphpoda (GOMES-DOS-SANTOS et al., 2020; SIGWART; LINDBERG, 2015).

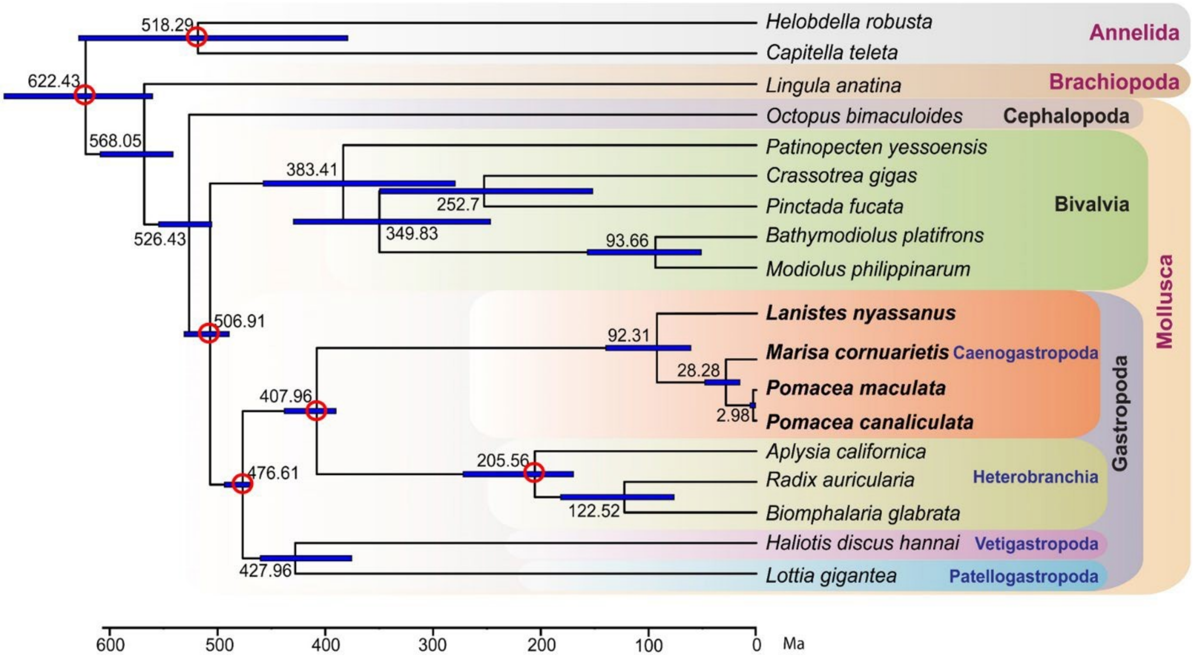
A classe gastrópode é a única classe de moluscos que tem representantes tanto em ambientes marinhos como de água doce e também terrestres, possuindo três subclasses: Prosobranchia, Opisthobranchia e Pulmonata (BROWN; LYDEARD, 2010). A classe gastrópode tem mais de 100.000 espécies que compreendem cerca de 80% de todos osmoluscos. É o grupo mais diversificado dentro do filo Mollusca. Alguns moluscos gastrópodesde água doce são hospedeiros intermediários de vários trematódeos patogênicos em seres humanos como na esquistossomose (HASZPRUNAR; WANNINGER, 2012). Os gastrópodesquando com concha são univalves e possuem uma boca com rádula (língua com vários dentículos) (BROWN; LYDEARD, 2010).

Os moluscos são usados como fonte de proteína em todo mundo, dentre estes bivalves (mexilhões), cefalópodes (polvos e lulas) e gastrópodes (abalones, lapas, búzios, caracóis terrestres). Há matérias-primas de valor econômico e cultural para humanidade como conchas, opérculos e pérolas, usadas como decoração, joias e moedas (HASZPRUNAR; WANNINGER, 2012). Os moluscos são utilizados para monitoramento da poluição ambiental, sendo espécies-chave do ecossistema que promovem a reciclagem de nutrientes e filtração de água (FIGUERAS *et al.*, 2019; RENAUT *et al.*, 2018).

Os moluscos da família Ampullariidae apresentam várias adaptações morfológicas e fisiológicas aos seus habitats que os tornam candidatos ideais para estudo de divergência populacional, especiação, adaptação e padrões de diversidade em maior escala incluindo a biogeografia de populações nativas e invasivas (IP *et al*., 2018). Há quatro ampularídeos com genomas sequenciados abrangendo os clados do Velho mundo (*Lanistes nyassanus*) e Novo mundo (*P. canaliculata, P. maculata* e *Marisa cornuarietis)* que podem ser objeto de estudos genômicos comparativos e de análise de miRNAs.

A divisão entre as linhagens do Novo e Velho Mundo (figura 1) pode ter ocorrido há cerca de 92,3 milhões de anos atrás (intervalo de confiança de 95% de 61,3–139,0 milhões de anos atrás) que é condizente com a época da separação de Gondwana, em torno de 120 milhões de anos atrás (JOKAT *et al*., 2003; SUN *et al*., 2019). *Marisa cornuarietis* e o gênero *Pomacea* divergiram a cerca de 28,3 milhões de anos atrás. As duas espécies de *Pomacea* parecem ter separado há cerca de 3,0 milhões de anos (intervalo de confiança de 95% de 2,0 - 5,0 milhões de anos atrás). O grau de semelhança entre as espécies *P. canaliculata* e *P. maculata* é dimensionado a ponto de poderem hibridizar e produzir progênie F1 viável (JOKAT *et al*., 2003; MATSUKURA *et al*., 2013; SUN *et al*., 2019).

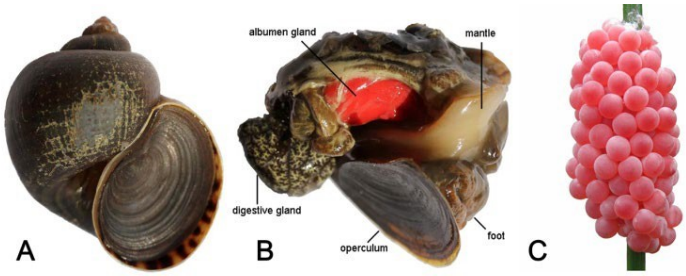
Figura 1: Árvore filogenética de *P. canaliculata, P. maculata e* espécies, construída usando 1.357 genes ortólogos de cópia única com um total de 455.177 aminoácidos.



Adaptado de Sun *et al*. (2019)

A espécie *P. canaliculata* (figura 2) é um caramujo de água doce da classe Gastrópode pertencente à família Ampullariidae. Apesar de aquático e possuir brânquias, precisa ir à superfície para realizar as trocas gasosas nos pulmões (JOSHI, 2005). O molusco é popularmente conhecido como caracol maçã, possuindo fertilização interna e são ovíparos (ACCORSI *et al*., 2013). O gênero *Pomacea* deposita os ovos acima da superfície da água em objetos sólidos, como rochas e caules de vegetação (HAYES *et al*., 2009). O fluido perivitelino em torno do embrião em desenvolvimento contém não apenas todos os nutrientes e doadores de energia para o desenvolvimento embrionário (HERAS; GARIN; POLLERO, 1998), mas também muitas macromoléculas que protegem os ovos de estressores ambientais e predadores (DREON *et al*., 2004; HERAS *et al*., 2008). Cerca de 21 genes do fluido perivitelino são específicos para a glândula albúmen, indicando que este órgão é a origem de muitas proteínas deste fluido (SUN *et al.,* 2012). A espécie *P. canaliculata* tem alta variação interpopulacional em relação ao tamanho, forma e espessura da concha e habita ampla gama de ecossistemas como pântanos, lagoas, lagos e rios (ESTEBENET; MARTIN, 2003).

Figura 2: A) Fotos fêmea de *Pomacea canaliculata* com uma concha intacta (comprimento da concha 4,0 cm). B) O mesmo animal com concha removido para mostrar estruturas corporais, incluindo glândula albumen. C) Ninhada de ovos (cada um com aproximadamente 2,5 mm de diâmetro).



Adaptado de Sun *et al*. (2012)

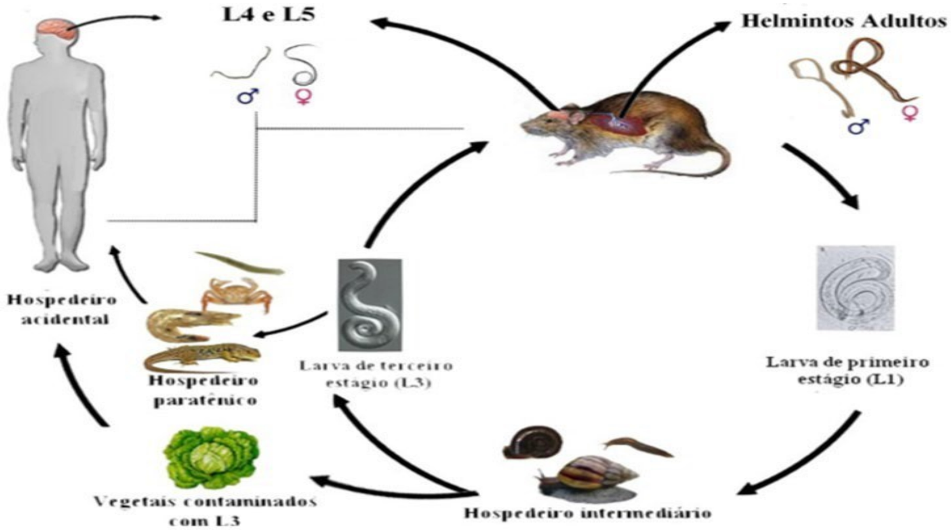
P. canaliculata é o Ampulariídeo com a maior distribuição geográfica conhecida, sendo originário da América do Sul. Está presente na Argentina, Uruguai, Paraguai e Bolívia, sendo abundante nas bacias dos rios da Prata, Uruguai e Paraguai (SILVANA THIENGO, 1993). O molusco também é encontrado na América Central e parte da América do Norte (HALWART, 1994). No Brasil, ocorre principalmente nas regiões Sul, Sudeste, floresta Amazônica ePantanal (SILVANA THIENGO, 1995). A introdução do molusco na Ásia ocorreu na década de 1980 através de empreendedores das Américas do Sul e do Norte, como iguaria para alimentação. No entanto, houve uma migração para agricultura onde se tornou inesperadamenteuma praga do arroz (HALWART, 1994). Na Indonésia, quando introduzido em plantações de arroz, causa uma queda de 15% da produção (NOVARINO, 2011). É descrita como uma espécie invasora e foi classificada entre as 100 maiores pragas em todo mundo (LUQUE et al., 2014). *P. canaliculata* tem crescimento rápido, alta taxa de reprodução, tolerância e adaptação ao estresse ambiental e distribuição geográfica em todos os continentes. Por estes motivos é considerado uma praga para agricultura, constituindo ameaça grave para o equilíbrio de um ecossistema (LIU *et al.*, 2018).

O molusco tem várias propriedades adaptativas e alta plasticidade ecológica. A espécie é resistente a altas temperaturas e seca (MATSUKURA *et al.*, 2009; YUSA; WADA; TAKAHASHI, 2006). *P. canaliculata* é capaz de sobreviver em ambientes com metais pesados ocasionando grande concentração nas brânquias (KRUATRACHUE *et al.*, 2011). Na microbiota intestinal do molusco, diversos parasitos reduzem os efeitos tóxicos destes metais (LIU *et al.*, 2018). A lectina neurotoxina presente nos ovos do caramujo defende os embriões dos predadores (DREON *et al.*, 2013). Os ovos de *P. canaliculata e P. maculata* são depositados na terra, diferente de outros moluscos que os depositam na água. Os ovos depositados em terra evitam o parasitismo e predação. A transição do meio aquático para o terrestre exigiu adaptações morfológicas tais como alongamento das vias respiratórias e aumento do tamanho do pulmão em adultos das espécies *P. canaliculata* e *P. maculata* (HAYES *et al.*, 2009). Há presença de um sistema imune neuroendócrino com memória imune específica em *P. canaliculata* (OTTAVIANI *et al.*, 2013). O sistema de biotransformação de substâncias tóxicas está desenvolvido em moluscos onde foram identificados 157 citocromos P450s nestes animais. A expansão dos genes na família do citocromo P450 em *P. canaliculata* contribui para resistência ao estresse ambiental. Foram encontradas enzimas antioxidantes (superóxido dismutase, catalase, peroxidase e glutadionaperoxidase) que são fatores inibidores dos radicais livres e condições patológicas (LIU *et al.*, 2018).

P. canaliculata está envolvido na transmissão do helminto *A. cantonensis*, causador da meningite eosinofílica em humanos. O ciclo biológico do nematódeo *A. cantonensis* (Figura 3) é heteroxênico, sendo os roedores hospedeiros definitivos. Nos animais contaminados, a fêmea do helminto adulto faz a postura de seus ovos nas artérias pulmonares dos roedores. Em seguida, ocorre a eclosão dos ovos surgindo a larva de primeiro estágio (L1), que são engolidas e excretadas pelas fezes. Moluscos são os hospedeiros intermediários que se contaminam pela ingestão da Larva L1, pela penetração dessas larvas no poro respiratório ou parede corporal. Ocorre duas mudas larvais dentro dos moluscos, passando pelo estágio larvar L2 originando L3. As larvas de terceiro estágio são transmitidas a hospedeiros paratênicos. Pode ocorrer a contaminação de seres humanos por estes nematódeos através de três maneiras: i) ingestão de hospedeiros paratênicos infectados (caranguejos, lagartos); ii) consumo de hospedeiros intermediários contaminados (moluscos crus ou malcozidos) e; iii) vegetais infestados pelas larvas infectantes. Os humanos são hospedeiros acidentais e no caso de infecção humana, a larva infectante se move até o Sistema Nervoso Central (SNC). O helminto não completa o ciclo de vida em humanos, embora permaneça no SNC causando meningite eosinofílica ou no globo ocular causando angiostrongilíase ocular (LUESSI et al., 2009; WANG et al., 2012, 2008). O episódio aconteceu pela primeira vez no leste da Ásia, sendo *P. canaliculata* hospedeiro intermediário para infecção em humanos (LV *et al*., 2008).

A meningite eosinófilica em humanos geralmente apresenta baixa gravidade. A resposta imune produzida pelo hospedeiro contribui para os sinais e sintomas da doença. Os pacientes apresentam meningite caracterizada pela presença de eosinófilos no líquido cefalorraquidiano (ESPÍRITO-SANTO *et al*., 2013; LUESSI *et al*., 2009; TSAI *et al*., 2001). Pode acontecer alguns casos graves da doença com sequelas neurológicas severas, coma e mortes, principalmente em crianças (CHEN et al., 2014a; LINDO et al., 2002; TSAI et al., 2001).

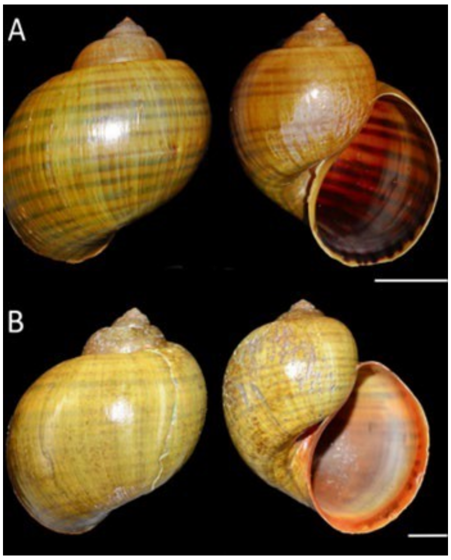
Figura 3:Ciclo biológico de *A. cantonensis.*



Adaptado de Monte (2014)

P. canaliculata realiza o controle biológico de *B. glabrata*, consumindo a desova deste molusco. Em suma, *P. canaliculata* pode ser uma bioferramenta de controle do hospedeiro intermediário da esquistossomose no ambiente em que coexistem (OLIVEIRA *et al*., 2012).

Figura 4: Conchas de *P. canaliculata* (A) e *P. maculata* (B).



Adaptado de Ng, Tan e Yeo (2017)

P. maculata (Figura 4) é um grande caracol de água doce nativo da América do Sul que coloniza desde o sul da Amazônia até bacia do Rio de da Plata (HAYES *et al*., 2012). Ele apresenta características fisiológicas e adaptativas semelhantes a *P. canaliculata,* sendo ambas as espécies reconhecidas por sua rápida propagação e aumento da população, constituindo ameaça grave para o equilíbrio de um ecossistema. Os custos econômicos do dano, controle e reparação podem ser imensos (PIMENTEL; ZUNIGA; MORRISON, 2005).

O sequenciamento de genes esclareceu muito a distribuição de *P. maculata* e *P. canaliculata* nos Estados Unidos, que apesar de serem parecidos fisiologicamente são substancialmente diferenciados geneticamente (HAYES *et al*., 2012). Levantamentos de uma série de populações sugerem que *P. maculata* é a espécie predominante em relação a *P. canaliculata* em pelo menos sete estados nos Estados Unidos: Carolina do Sul, Geórgia, Flórida, Alabama, Mississippi, Louisiana e Texas (MUECK *et al*., 2018). A extensão da destruição das regiões invadidas causada por *P. maculata* e *P. canaliculata* variaram consideravelmente, apesar das semelhanças ambientais e ecológicas entre os habitats afetados (HORGAN; STUART; KUDAVIDANAGE, 2014). Essas diferenças sugerem que pode haver variação fisiológica significativa entre espécies individuais (*P. maculata* e *P. canaliculata*) e entre populações das mesmas espécies (MATSUKURA *et al*., 2016).

2.2 SEQUENCIAMENTO DE DNA

O sequenciamento de DNA ganhou notoriedade em 1975 com o desenvolvimento de tecnologia amplamente utilizada, que ficou conhecida como Sequenciamento de Sanger (SANGER; COULSON, 1975). Este sequenciamento é altamente preciso e permaneceu por décadas como principal forma de sequenciamento de DNA, até o surgimento da nova tecnologia *Next Generation Sequencing* (NGS). Essa técnica devido ao menor custo e esforço necessário, permitiu que pesquisadores gerassem grande conjunto de dados genômicos (SCHULTZ; ADEMA, 2017). A enorme quantidade de dados gerados por essa tecnologia impulsionou o desenvolvimento, em paralelo com a bioinformática, de novas ferramentas e técnicas de análise (ZHANG *et al.*, 2011).

2.2.1 Sequenciamento em moluscos

O primeiro sequenciamento de genoma de metazoário ocorreu em 1998 no nematoide *Caenorhabditis elegans*. Desde então, várias sequências genômicas foram identificadas (TAKEUCHI *et al.*, 2012). Há uma tendência crescente no número de publicações que incluem dados genômicos aplicados a moluscos. A maioria dos recursos foram aplicados em gastrópodes e bivalves. A maior parte das abordagens está associada a estudos genéticos populacionais ou mapeamento de características por locus gênico (GOMES-DOS-SANTOS *et al.*, 2020). No banco de dados *National Centre for Biotechnology Information* (NCBI - http://.ncbi.nlm.nih.gov/), em maio de 2020, há registro de 48 genomas de espécies de moluscos sequenciados.

Moluscos apresentam níveis elevados de diversidade genética (WANG et al., 2017), a taxa de Polimorfismos de Nucleotídeo Único (SNPs) e polimorfismos observados em ostras do pacífico é de 6 a 10 vezes maior que observado em humanos (HOLLENBECK; JOHNSTON, 2018). Outro fator que contribui para a diversidade gênica é a alta taxa de mutação, que pode ser elevada devido ao grande número de eventos necessários na meiose, para produzir milhões de ovos na reprodução (PLOUGH; HEDGECOCK, 2016). Esta variabilidade genética está sendo revelada nos genomas sequenciados e um importante passo foi dado no sequenciamento de genoma completo de moluscos. Em um breve resumo, descreve-se o sequenciamento de genoma completo de alguns moluscos gastrópodes, cefalópodes e bivalves.

O sequenciamento de genoma completo do caramujo de água doce: *B. glabrata* (classe: gastrópode) foi um marco devido a importante relevância clínica para os humanos. A alteração genética do molusco poderá modificar a transmissibilidade da esquistossomose, doença negligenciada em países subdesenvolvidos. *B. glabrata* é hospedeiro intermediário do

Schistosoma mansoni causador da esquistossomose. O genoma tem um tamanho estimado de 916,4 megabases compreendidas em 18 cromossomos, nos quais 14.423 genes foram identificados. A caracterização deste genoma define melhor o clado Lophotrochozoa quando comparado ao clado Ecdysozoa e aos animais deuterostômios (ADEMA *et al*., 2017).

O genoma completo sequenciado do molusco *Lottia gigantea* (gastrópode marinho) foi publicado em 2013 e tem o tamanho estimado em 359,5 megabases (SIMAKOV *et al*., 2013). Foi verificada a presença de genes que codificam neuropeptídios e neurohormônios neste molusco. Quatro genes codificam peptídeos relacionados à insulina. Foi encontrado um peptídeo semelhante ao GnRH (hormônio liberador de gonadotrofina) e pequenos peptídeos cardioativos, revelando similaridades com outros moluscos e insetos (VEENSTRA, 2010).

O molusco da classe gastrópode, *Achatina fulica* (caramujo gigante africano), foi o primeiro molusco terrestre a ser sequenciado de forma completa. A extensão do genoma é de 2.120 megabases, com o conteúdo de repetições de até 71% (GUO et al., 2019). Estas repetições são parecidas (70%) no caramujo gastrópode pulmonar marinho *Radix auricularia*, devido principalmente aos transposons de DNA (SCHELL *et al*., 2017). No total 23.726 genes codificadores de proteínas foram previstos em *A. fulica*, compreendidos em 31 cromossomos e também a presença de 675 genes ortólogos relacionados a outros moluscos. O sequenciamento do genoma completo revela inferências filogenéticas entre este caramujo e o molusco *P. canaliculata*. Ambos são gastrópodes com ampla adaptabilidade ambiental, alta taxa de crescimento e capacidade reprodutiva. As espécies são pragas invasoras de culturas agrícolas que causam queda na produção, prejuízos econômicos e também são hospedeiros intermediários para patologias humanas (GUO et al., 2019)

Os genomas de *P. canaliculata* e *P. maculata* e foram montados após o sequenciamento de bibliotecas de DNA de inserção curta e longa usando plataformas de sequenciamento Illumina. Os genomas das espécies são considerados compactos quando comparado com outros moluscos, *P. canaliculata* contém 447,7 megabases e *P. maculata* 432,3 megabases (Tabela 1).

Tabela 1: Principais estatísticas de montagem de genomas de moluscos.

Classe	**Espécie**	**GC Conteúdo**	**Tamanho Montagem**	**No. Scaffolds**	**Scaffold mais longo**
Cephalopoda	*Octopus bimaculoides*	0,31	2.338,2 Mb	151.674	4,1 Mb
Bivalvia	*Crassostrea gigas*	0,3	557,7 Mb	7.659	1,96 Mb

Bivalvia	*Pinctada fucata*	0,33	815,3 Mb	29.306	1,26 Mb
Gastropoda	*P. canaliculata*	0,41	447,7 Mb	3.131	45,35 Mb
Gastropoda	*P. maculata*	0,4	432,3 Mb	3.914	2,52 Mb
Gastropoda	*Marisa cornuarietis*	0,41	535,5 Mb	665	24,67 Mb
Gastropoda	*Lanistes nyassanus*	0,42	510,0 Mb	34.401	1,79 Mb
Gastropoda	*Radix auricularia*	0,34	909,8 Mb	4.823	2,97 Mb
Gastropoda	*Aplysia californica*	0,32	927,3 Mb	4.332	610,3 kb
Gastropoda	*Biomphalaria glabrata*	0,35	916,4 Mb	331.401	2,18 Mb
Gastropoda	*Lottia gigantea*	0,28	359,5 Mb	4.475	9,39 Mb
Gastropoda	*Haliotis discus hannai*	0,38	1.865,4 Mb	80.032	2,21 Mb
Gastropoda	*Lymnaea stagnalis*	0,37	833,2 Mb	328.378	95,6 kb

Adaptado de Sun *et al*. (2019)

Os genomas de *P. canaliculata, P. maculata* e *Lanistes nyassanus* foram montados após o sequenciamento de bibliotecas de DNA de inserção curta e longa usando plataformas de sequenciamento Illumina. Esses três genomas variam de 432,3 a 510,0 megabases de comprimento total com os dois genomas de *Pomacea* sendo menores, *P. canaliculata* com 447,7 megabases e *P. maculata* com 432,3 megabases. O genoma de *Marisa cornuarietis* foi montado usando leituras curtas Illumina e leituras longas Nanopore, com o comprimento estimado de 535,5 megabases. Os valores finais de scaffold/contig N50 de *P. canaliculata* é de 32,6 Mb e de *M. cornuarietis* de 4,4 Mb, excedem a maioria dos outros genomas de moluscos sequenciados, mostrando a alta continuidade das montagens (Tabela 1) (SUN et al., 2019).

O sequenciamento de genoma completo em moluscos cefalópodes foi realizado primeiramente no polvo *Octopus bimaculoides* no ano de 2015. O tamanho do genoma foi estimado em 2.338,2 megabases (maior genoma conhecido do mundo dos invertebrados) e foram identificados 33.638 genes codificadores de proteína. Quase 45% do genoma é composto por elementos repetitivos, com presença de elementos de transposição. Há elementos sugestivos de duplicação de genoma inteiro, embora não possa ser confirmado. Este fato permite que genes adicionais adquiram novas funções. O desenvolvimento genético neuronal em cefalópodes é semelhante ao encontrado em bilatérios vertebrados. A família de genes protocaderinas está desenvolvida, sendo necessária para o aprimoramento normal dos circuitos neurais. Acreditava-se que as protocaderinas eram desenvolvidas somente em vertebrados. A enorme edição do RNA mensageiro gera diversidades de proteínas envolvidas na excitabilidade neuronal. Justifica-se a maior inteligência dos polvos em relação a outros moluscos. Foram identificados centenas de genes cefalópodes específicos, muitos destes mostraram elevados níveis de expressão na pele (mudança de coloração), ventosas e sistema nervoso (ALBERTIN *et al.*, 2015). No molusco *Lymnae stagnalis* foi identificado o miR-124 (WALKER *et al.*, 2018) que é também verificado no polvo *O. bimaculoides*. Este miRNA é bastante abundante no Sistema Nervoso de vertebrados e invertebrados, que contribui para desenvolvimento e diferenciação neuronal (LIU et al., 2011). A análise de microRNAs conservados revela funções parecidas em espécies distintas, as quais estão ligadas evolutivamente.

O sequenciamento do genoma completo do molusco bivalve *Pinctada fucata martensii* (ostra de pérola), com a extensão de genômica de 1.150 megabases, identificou 32.937 modelos de genes codificadores de proteínas. A análise filogenética de genomas sequenciados de *P. fucata martensii* (bivalve), *Crassostrea gigas* (bivalve) e *L. gigantea* (Gastropoda) forneceu estimativas de divergências em árvores filogenéticas. *P. fucata martensii* divergiu de seu ancestral comum ao bivalve descritos a 316 milhões de anos atrás, enquanto divergiu do gastrópode descrito a 485 milhões de anos atrás (DU *et al.*, 2017). Já no mexilhão dourado *Limnoperna fortunei*, molusco de água doce da classe bivalve, o sequenciamento de seu genoma total apresentou o tamanho de 1.600 megabases. Foram identificados 60.717 modelos de genes (58% destes com evidencias transcricionais baseadas em RNA). Estes moluscos são invasores agressivos prejudiciais as espécies nativas na região que colonizam, possuindo alta taxa de reprodução e causam prejuízos econômicos na agricultura. Estas espécies podem detectar produtos químicos na água e fechar suas válvulas como resposta defensiva, sendo tolerantes ao estresse ambiental. A partir do seu genoma sequenciado poderão ser apresentadas as bases moleculares desta invasividade e os mecanismos para controlar as pragas aquáticas (ULIANO *et al.*, 2018).

2.2.2 Sequenciamento nas espécies *P. canaliculata* e *P. maculata*

A escassez de genomas sequenciados de moluscos foi apontada pela Aliança Global Genômica de Invertebrados (*Global Invertebrate Genomics Alliance*) como um problema. A espécie *P. canaliculata* foi designada como espécie prioritária para sequenciamento de genoma total em 2017 (VOOLSTRA *et al.*, 2017). A importância do sequenciamento genômico deste molusco refere-se a um organismo modelo para estudo, apresentando diversas características inerentes para investigações ecológicas, evolutivas e funcionais. Dessa forma, grande esforço foi realizado para o sequenciamento da espécie, que foi publicado em 11 de maio de 2018 (LIU *et al.*, 2018).

Há um estudo de sequenciamento de genoma total para *P. maculata* (SUN *et al.*, 2019) e dois estudos para *P. canaliculata* (LIU *et al.*, 2018; SUN *et al.*, 2019). O primeiro trabalho em 2018 para *P. canaliculata* revela sequenciamento de alta qualidade com montagem genômica próxima ao nível cromossómico. O tamanho médio do genoma foi estimado em torno de 440 megabases, sendo considerado compacto quando comparado a outros moluscos como por exemplo, *Achatina fulica* (2.120 megabases) e *Octopus bimaculoides* (2.338,2 megabases). Os genomas ampularídeos tem um conteúdo de repetições menores do que a maioria dos outros genomas de moluscos (24,2 a 61,1%) (SUN et al., 2019). O baixo conteúdo de elementos repetidos em *P. canaliculata*, contra 71% em *A. fulica* e 45% em *O. bimaculoides* torna o ampularídeo com genoma compacto quando comparado com outros moluscos (ALBERTIN *et al.*, 2015; GUO *et al.*, 2019; LIU *et al.*, 2018). Essa analogia aplica-se também a *P. maculata* com presença de genoma denso (432,3 megabases). Além disto, o comprimento dos íntrons de ampularídeos é semelhante ao de outros gastrópodes, mas é mais curto do que outros moluscos (SUN *et al.*, 2019).

O genoma de *P. canaliculata* (Tabela 2) foi montado após sequenciamento de bibliotecas de DNA em plataformas de sequenciamento Illumina. Os valores finais de *scaffold* / contig N50 de *P. canaliculata* (32,6 Mb) excede a maioria dos outros genomas de moluscos sequenciados, mostrando alta continuidade das montagens (SUN *et al.*, 2019). No entanto, este estudo tem mais *scaffolds* (3131) que o trabalho de LIU *et al.*, 2018 (24). O cariótipo haploide é composto por 14 cromossomos e foram estimados 21.533 genes em uma região codificadora de 32,2 mega bases, que corresponde a 7,3% do genoma (LIU *et al.*, 2018). Foi encontrado menor quantitativo de genes (18.263) identificados no estudo de Sun *et al.* (2019). Os valores menores neste estudo podem ser devido a mais dados transcriptômicos e proteômicos usados para refinamento da predição do modelo de genes na espécie.

Tabela 2: Resumo da montagem e anotação genoma *P. canaliculata* e *P. maculata.*

	Pomacea maculata	***Pomacea canaliculata***

Contéudo de GC	0,41	0,41
Tamanho montagem	432,3 MB	447,7 MB
Número de Scaffolds	3.914	3.131
Tamanho de Scaffolds	2,52 MB	45,35 MB
Número de Scaffolds 1MB	38	14
Scaffold N50	375,9 MB	32,6 MB
Contig N50	91,9 MB	81,4 KB
Número de modelos de genes	23.464	18.263

Adaptado de Sun *et al.* (2019)

Grande parte das abordagens genômicas em moluscos está relacionada ao mapeamento de características por *locus* gênico (GOMES-DOS-SANTOS *et al.*, 2020). Após o sequenciamento genômico das espécies, foi facilitada a identificação de alguns genes em *P. canalicula* e *P. maculata*. Estudos futuros poderão identificar quais miRNAs interagem com a 3' UTR dos mRNAs alvos codificados por estes genes para verificar se ocorre regulação da expressão gênica (HA; KIM, 2014). Em seguida, há alguns genes de *P. canaliculta P. maculata* de características evolutivas identificadas, que poderão ser objetivo de estudo de miRNAs.

O gene Pca_154_3.36 de *P. canaliculata* codifica a proteína CaBP (ligação de cálcio proteína), que pode desempenhar um papel importante na formação da casca dura do ovo, protegendo fisicamente e evitando a dessecação. A proteína pode ter contribuído para a transição evolutiva da deposição de ovos aquática para terrestre nas espécies de ampularídeos. O molusco próximo evolutivamente a *P. canaliculata, Marisa cornuarietis* (da mesma família) não tem CABP e deposita ovos gelatinosos de forma subaquática. Apesar de haver uma presença generalizada de CaBP no filo molusca, ocorreu uma expansão da família de genes CaBP após a divergência de ampularídeos de outros moluscos. Além disso, a análise de dados proteômicos mostram que o CaBP é altamente abundante no fluido perivitelínico de duas espécies (*P. canaliculata e P. maculata*), que põem ovos terrestres com uma casca calcária em ampularídeos (HERAS *et al.*, 2008; SUN *et al.*, 2019).

O gene Pca_1306_1.39 de *P. canaliculata* e o gene Pma_3499_0.31 de *P. maculata* codificam a proteína MAPCF. Esta proteína é uma subunidade das perivitelinas tóxicas (PV2), as quais compõem o fluido perivitelínico (PVF) nos ovos de *P. canaliculata* e *P. maculata*. As PV2 demonstraram ser altamente neurotóxicas para camundongos. É considerada uma proteína defensiva para proteger os ovos ricos em nutrientes de predadores terrestres.

O complexo (PV2) é composto por duas proteínas, a MACPF (provavelmente a porção toxica) e a taquilectina. A análise filogenética mostrou que os genes do tipo MACPF de ampularídeos estão agrupados em dois grupos. O grupo A compreende sete sequências ampularídeas instaladas entre as não ampularídeas, indicando uma possível origem antiga desses genes remontando ao ancestral comum de todos os moluscos. O grupo B é específico para quatro ampularídeos (*M. cornuarietis, Lanistes nyssanus, P. maculata e P. canaliculata*), compreendendo 17 sequências, indicando que ocorreu extensos eventos de duplicação de genes. Estes fatos aconteceram após a divergência de Ampullariidae de outros moluscos. Dentro do Grupo B, há uma sequência que codifica a subunidade MACPF de *P. canaliculata* (Pca_1306_1.39) e Pma_3499_0.31 de *P. maculata*. Estas aquisições podem ter contribuído para a transição da postura aquática para terrestre de ovos em ampularídeos (DREON *et al*., 2013; SUN *et al*., 2019).

Há relatos de expansão das famílias de genes relacionados à detecção ambiental e digestão de celulose, o que pode ter influenciado alguns ampularídeos serem pragas invasivas notórias. Na detecção ambiental, há sete genes de *P. canaliculata* (Pca_112_8.10, Pca_112_8.16, Pca_112_7.37, Pca_112_8.52, Pca_112_8.32, Pca_112_8.45, Pca_112_8.8)que codificam a família das proteínas G (GRL101) (SUN *et al*., 2019), as quais estão envolvidasna quimiorrecepção em caracóis aquáticos (SCHULTZ; ADEMA, 2017). Em Ampullariidae ocorreu expansão massiva da família GRL101, com pelo menos dois principais eventos de duplicação de genes nesta família, após a divergência de ampuláriddeos de outros moluscos. Muitos desses genes GRL101 são altamente expressos nos tentáculos cefálicos e palpos labiais de *P. canaliculata*. Esta quimiorrecepção sensorial é extremamente importante para ocupação de ambientes transitórios e colonização de territórios, traços inerentes da invasividade e plasticidade ambiental de *P. canaliculata* e *P. maculata* (MENDOZA; SEBÉ-PEDRÓS; RUIZ-TRILLO, 2014; LIU *et al*., 2018; SUN *et al*., 2019).

2.3 PEQUENOS RNAS NÃO CODIFICADORES DE PROTEINAS

2.3.1 Histórico

Os três tipos principais de pequenos RNAs não codificadores de proteínas em animais são microRNAs (miRNAs), pequenos RNAs interferentes (siRNAs) e RNAs que interagem com PIWI (piRNAs) (OKADA *et al*., 2009). Os siRNAs têm aproximadamente 21 nucleotídeos de comprimento, são derivados de RNAs em formato de grampo longos (hairpins) e processados pela RNAase Dicer. Os siRNAs medeiam a supressão pós-transcricional através da clivagem de mRNA e contribuem para a defesa antiviral (GHILDIYAL; ZAMORE, 2009).

Os piRNAs têm de 24 a 30 nucleotídeos de comprimento e não dependem de proteínas do tipo RNase III para processamento e são obtidos a partir de precursores de fita simples por uma endonuclease. A expressão gênica é inibida por piRNAs através da clivagem do mRNA (IPSARO *et al*., 2012; NISHIMASU *et al*., 2012; SIOMI *et al*., 2011). A principal função dos piRNAs é silenciar elementos transponíveis em células germinativas.

Os miRNAs foram descobertos em 1993 por Victor Ambros e colaboradores ao estudar o nematódeo *C. elegans*. Foram verificados indícios de controle da expressão genica por pequenos RNAs, com funções diferentes até então conhecidas. No gene lin-4 foi observada a presença de pequenos RNAs não codificantes de proteínas. Estes miRNAs continham algumas sequências complementares à região 3'UTR do mRNA transcrito do gene lin-14, que reprimia a expressão pós-transcricionalmente impedindo a tradução (LEE; FEINBAUM; AMBROS, 1993). Desde então, miRNAs foram detectados em vários organismos e mostraram ser altamente conservados entre as espécies (FRIEDLÄNDER *et al*., 2014).

Os miRNAs são pequenos RNAs não codificantes de proteínas com cerca de 22 nucleotídeos. A maioria dos miRNAs é transcrita de sequências de DNA em miRNAs primários (pri-miRNAs), processados em miRNAs precursores (pré-miRNAs) e subsequentemente em miRNAs maduros. Na grande parte dos casos, os miRNAs interagem com a 3′ UTR dos mRNAs alvos para suprimir expressão gênica (HA; KIM, 2014). A interação de miRNAs com outras regiões, incluindo a 5' UTR , a sequência de codificação e genes promotores também foram relatados para inibição gênica (BROUGHTON *et al*., 2016). Os miRNAs também demonstraram ativar a expressão gênica em determinadas situações (VASUDEVAN, 2012).

Cerca de metade de todos os miRNAs identificados são intragênicos e processados principalmente a partir de íntrons, os demais são intergênicos regulados por seus próprios promotores. O domínio na extremidade 5′ dos miRNAs que se estende da posição 2 a 7 é crucial para o reconhecimento do alvo (mRNA) e foi denominada *seeds,* "semente de miRNA ou resíduos-semente". Os nucleotídeos 13 a 16 também contribuem com o emparelhamento de mRNAs alvos (RIE *et al*., 2017; HA; KIM, 2014; KIM; KIM, 2007).

É consenso na literatura científica os mecanismos reguladores dos miRNAs tais como: diferenciação celular, apoptose, proliferação e tumorigênese, hematopoiese, organogênese; desenvolvimento das células tronco pluripotentes e testicular (FABIAN; SONENBERG; FILIPOWICZ, 2010; KIM, 2005; LEONARDO *et al*., 2012; RAKOCZY *et al*., 2013). Os miRNAs desempenham papéis importantes na plasticidade cerebral e memória e em algumas situações se comportam como hormônios, sendo encapsulados em exossomas e transportados a distância (ANFOSSI *et al*., 2017; BREDY *et al*., 2011). Da mesma forma, como elemento regulador, o próprio miRNA é modulado de maneira coordenada por diversos fatores (FABIAN; SONENBERG; FILIPOWICZ, 2010).

Os miRNAs têm papel regulatório em processos patológicos (KIM, 2005). No câncer há miRNAs envolvidos na metástase e tumorigênese (BRACKEN *et al.*, 2009). Há relatos de expressões disfuncionais de miRNAs característicos nesta patologia (IORIO; CROCE, 2009). No diabetes, alguns miRNAs estão envolvidos na produção de células β pancreáticas deformadas, prejudicando a produção de insulina. Os miRNAs são possíveis alvos para desenvolvimento de fármacos, como nestes casos (OETJEN, 2012).

Os miRNAs podem servir como biomarcadores para exames de diagnóstico. Nas infecções por Epstein-barr crônicas há elevação de miRNAs virais específicos, sendo potenciais biomarcadores da gravidade da doença ou prognóstico (KAWANO *et al.*, 2013). Dois miRNAs (mir-k12-10b, k12-12) provenientes do Herpes vírus causador do sarcoma de Kaposi (KSHV) foram identificados em pacientes que desenvolveram sepse em grandescirurgias (TUDOR *et al.*, 2014). A desregulação destes dois miRNAs podem levar a falência dos órgãos e morte. A identificação precoce destes marcadores poderá detectar pacientes com alto risco de choque séptico (GIZA *et al.*, 2016). Em 2007, pela primeira vez, foi relatado miRNAs disfuncionais no sangue de pacientes com linfoma. Já em 2008 verificou-se a potencialidade para serem utilizados como biomarcadores no câncer. Há vários estudos a este respeito, um destes diz respeito a níveis de expressão dos miRNAs (miR-21, miR-126, miR- 155, miR199a e miR-335), associando às manifestações clínico patológicas do câncer de mama.A possibilidade de utilizar miRNAs como biomarcadores apresenta vantagens por não ser um método invasivo, já que o padrão ouro para diagnóstico na patologia é a biopsia (FERNANDEZ-MERCADO *et al.*, 2015; LAWRIE, 2007; LAWRIE *et al.*, 2008; WANG *et al.*, 2010).

2.3.2 Biogênese dos miRNAs

2.3.2.1 Via Canônica

A via canônica da biogênese de miRNAs é a via dominante no qual os miRNAs são processados pelas RNAases III, Drosha e Dicer, consecutivamente. O miRNAs podem ter origem intragênica (em sua maior parte íntrons) ou intergênica. As unidades de transcrição independentes podem ser transcritas individualmente de forma monocistrônica ou constituindo uma unidade de transcrição policistrônica, sendo transcritos em sequências longas chamadas *clusters*. Os pri-miRNAs (transcrito primário) são transcritos de seus genes pela RNA polimerase II eventualmente regulados pela RNA polimerase III e depois gerados os pré-miRNAs, que possuem aproximadamente 65 nucleotídeos no núcleo celular. Tanto os pri-miRNAs quanto pré-miRNA têm a estrutura secundária em forma de grampo de cabelo (*hairpin)*. Em seguida, os pré-miRNAs são processados em miRNAs maduros no citoplasma (LEE, 2002; TANZER; STADLER, 2004; ZHANG; ZHANG; SU, 2009).

O complexo de microprocessadores, composto por uma proteína de ligação (*DiGeorge Syndrome Critical Region 8* - DGCR8) e uma enzima ribonuclease III (Drosha), realiza a clivagem do pri-miRNA. O DGCR8, também conhecido como Pasha em *Drosophila melanogaster*, se liga em uma estrutura no pri-miRNA, enquanto Drosha realiza a cisão do duplex. Depois que os pré-miRNAs são gerados, eles são transportados para o citoplasma através do complexo exportina 5 (XPO5) / RanGTP, em seguida processados pela endonucelase RNA III Dicer (ALARCÓN *et al*., 2015; DENLI *et al*., 2004; HAN *et al*., 2004; OKADA *et al*., 2009).

No citoplasma os pré-miRNAs são clivados por Dicer, sendo esta constituída por três módulos, dois domínios de RNase III e um domínio PAZ (Piwi, Argonauta e Zwille) (MACRAE *et al*., 2006). A clivagem por Dicer pode ocorrer na extremidade 5' ou 3'. Na extremidade 3', a clivagem ocorre geralmente a uma distância fixa de 21 a 25 nucleotídeos, dependendo da espécie e tipo de Dicer. Na extremidade 5', em mamíferos e moscas, a divisão acontece a 22 nucleotídeos de distância (MACRAE; ZHOU; DOUDNA, 2007; PARK *et al*., 2011). A cadeia 5p surge da extremidade 5' do *hairpin,* enquanto que a cadeia 3p é originária da extremidade 3' do pré-miRNA. Ambos os fios derivados do duplex miRNA maduro podem ser carregados pela família de proteínas Argonauta (AGO), em humanos (AGO1-4) de maneira dependente de ATP (KHVOROVA; REYNOLDS; JAYASENA, 2003; MEIJER; SMITH; BUSHELL, 2014; YODA *et al*., 2010).

2.3.2.2 Via não canônica

A via não canônica de biogênese dos miRNAs foge do processamento sequencial canônico por DGCR8/Drosha e Dicer. Os miRNAs das vias não canônicas podem ser processados por diferentes combinações do processo canônico via DGCR8/Drosha, Dicer, Exportina 5 e Argonauta (AGO2). Nesta via, a biogênese de miRNAs pode ser independente de Drosha ou Dicer (BABIARZ *et al*., 2008; RUBY; JAN; BARTEL, 2007).

Alguns íntrons imitam a característica estrutural em formato de grampo dos pré-miRNAS, para entrar na via de processamento de miRNA sem clivagem por Drosha. Estes íntrons são chamados de mirtrons. Da mesma forma, alguns transcritos curtos com "capeamento" de 7-metilguanosina (m^7G) na extremidade 5' são exportados diretamente para o citoplasma através de Exportina 1 sem a necessidade de clivagem por Drosha. A presença do "capeamento" do miRNA na cadeia 5' impede o carregamento por Argonauta, favorecendo a utilização de miRNA-3p (RUBY; JAN; BARTEL, 2007; XIE *et al.*, 2013).

Alguns miRNAs podem ser processados independentes de Dicer, onde Drosha gera pequenos transcritos de RNA em forma de grampo (shRNA). Estes pré-miRNAs são de comprimento insuficiente para serem processados por Dicer. Dessa forma, são carregados e clivados pelo núcleo catalítico de Argonauta (AGO2) (CHELOUFI *et al.*, 2010; YANG *et al.*, 2010).

2.3.3 Mecanismos de regulação gênica desenvolvidos por miRNAs

Depois que os miRNAs maduros são sintetizados, eles induzem o silenciamento da expressão gênica (em sua maior parte). São formadas estruturas efetoras de ribonucleoproteínas chamado de Complexo de Silenciamento Induzido por miRNAs, miRISC. A formação deste complexo silenciador começa com o recrutamento da família de proteínas GW182, as quais induzem o recrutamento de outras proteínas efetoras (BEHM-ANSMANT *et al.*, 2006). No núcleo de miRISC está a família de proteínas Argonauta (AGO), que é carregada com uma fita guia de miRNA. Os miRNAs pareiam suas bases com uma relação do tipo Watson-Crick com os RNAs alvo. O complexo é direcionado para o pareamento das bases com mRNA resultando em degradação do mRNA ou repressão da tradução (HAMMOND *et al.*, 2000).

Alguns membros da família Argonauta tem a atividade de fatiamento de mRNAs, exigindo grau de complementariedade entre as bases do miRNA e mRNA para tal ação (ELBASHIR; LENDECKEL; TUSCHL, 2001; JO *et al.*, 2015). Quando a clivagem do alvo é inibida devido às limitações no pareamento de bases ou falta da atividade catalítica do Argonauta, a expressão de mRNAs alvo é reprimida via repressão da tradução ou deadenilação seguida pela degradação do mRNA (HUNTZINGER; IZAURRALDE, 2011). Nas células animais, a maioria das interações miRNA-RNA alvo não são totalmente complementares, gerando "*mismatches*". Dessa forma, é impedida a atividade de endonuclease de AGO2, que passa a atuar como mediador da interferência com RNA (JONAS; IZAURRALDE, 2015). A ligação de miRISC com mRNA resulta em quatro vias de reação, clivagem do alvo, ligação transitória com o alvo, ligação estável com o alvo e por fim, ligação da fita guia com o alvo e descarregamento da Argonauta (JO *et al.*, 2015; KAWAMATA; TOMARI, 2010).

A maioria dos estudos relatam inibição da expressão gênica, alguns citam a ativação da tradução mediada por miRNAs. A presença de AGO2 (Argonauta) e outras proteínas como FXRP1 (proteína do retardo mental do X frágil) estão associadas ao complexo miRNA-proteína (microRNPs). Estes elementos estão relacionados a elementos ricos em AU no terminal 3' UTR para ativar a tradução (VASUDEVAN; STEITZ, 2007). Outros exemplos de ativação de genes por miRNAs inclui a ligação ao terminal 5 ′ UTR de mRNAs, que induz a codificação de proteínas ribossômicas durante a privação de aminoácidos (NIELSEN; LUND, 2008).

2.4 miRNAs EM MOLUSCOS E NAS ESPÉCIES *P. canaliculata* E *P. maculata*

Em um estudo em *B. glabrata* utilizando análises *in sílico* e *in vitro* foi possível identificar 89 miRNAs maduros, sendo 5 deles molusco específicos, além de piRNAs identificados no genoma de *B. glabrata* pela primeira vez. Alguns miRNAs se destacaram por apresentar número elevado de leituras; como bgl-bantam, bgl-let-7, bgl-miR-100, bgl-miR-184, bgl-miR-71, bgl-miR-1984, bgl- miR-1a, bgl-miR-315, bgl-miR-375, bgl-miR-750, bgl-miR-8 e bgl-miR-92; sugerindo que estes miRNAs possam ter um papel importante na biologia dos caramujos. Para tanto, bgl-miR-100 e bgl-miR-1a foram os mais expressos, podendo desempenhar papel preponderante na regulação do desenvolvimento e na resposta à infecção de *B. glabrata*. Um número significativo de alvos previstos em *B. glabrata* estão envolvidos na resposta ao estresse, que podem ser determinantes do processo adaptativo dos caramujos à constante colonização por inúmeros microrganismos. Há vários estudos que relatam a susceptibilidade de *B. glabrata* para a infecção com o parasito *Schistosoma mansoni*, alguns dos quais tentam desvendar os mecanismos genéticos que regulam fisiologicamente o molusco. No entanto, nenhum estudo está disponível atualmente que elucida o papel de pequenos RNAs na relação caramujo hospedeiro-parasita (QUEIROZ et al., 2020).

Assim como nos outros metazoários, os miRNAs realizam diversos mecanismos de controle gênico. No bivalve (ostra plana) *Ostrea edulis* após ser exposto a parasitas, revelou expressão diferencial de 63 miRNAs. Desses, 15 miRNAs têm um papel direto nas respostas como proliferação, ativação de macrófagos, inflamação, apoptose e/ou dano oxidativo, conforme relatado para outros organismos. Dessa forma, dentre as diversas funções desempenhadas nos moluscos pelos miRNAs, eles atuam como biomarcadores, que elucidam efeitos imunotóxicos causados por fatores bióticos e abióticos (BURGOS-ACEVES *et al*., 2018; MARTÍN-GÓMEZ *et al*., 2014)

Os miRNAs mostraram-se altamente conservados, principalmente entre os metazoários e também em moluscos. No gastrópode *Littorina littorea* cinco miRNAs precursores foram expressos no músculo do pé (miR-1a-1, -34a, -133a, -125b, -29b e -2a) e três (miR-1a-1, -34a, -29b) no hepatopâncreas, após serem expostos a condições estressantes como hipóxia e gelo. Estes miRNAs mostraram-se conservados em espécies de invertebrados, o que é provável inferir que estariam presentes em todos os moluscos (BIGGAR *et al*., 2012; OWENS; MALHAM, 2015). OmiRNA-124a está relacionado a processos regenerativos do Sistema Nervoso Central (SNC) de vertebrados, quando associado ao metabólito ácido retinóico. Este miRNA foi identificado pela primeira vez no filo Mollusca, no SNC do gastrópode *Lymnaea stagnalis,* com padrões de expressões semelhantes aos vertebrados (WALKER *et al*., 2018), corroborando a alta conservação de miRNAs em moluscos e outras espécies.

Há poucos miRNAs de moluscos identificados no miRBase versão 22 (http://www.mirbase.org/), sendo 245 miRNAs maduros e 154 precursores. Apenas 3 espécies de moluscos já tiveram miRNAs registrados*:* *Lottia gigantea* (64 miRNAs maduros), *Haliotis rufescens* (5 miRNAs maduros) e *Melibe leonina* (176 miRNAs maduros). Já foram catalogados 48.885 miRNAs maduros e 38.589 precursores de 271 espécies identificadas. Há necessidade de maior identificação de miRNAs em moluscos, devido ao extenso número de espécies e poucos registros. Nos artrópodes, filo com maior variedade de espécies, já foram identificados 8.753 miRNAs maduros. Justifica-se assim o estudo de miRNAs em moluscos especificamente nas espécies *P. canaliculata* e *P. maculata*. Estes dados estão disponíveis no *website* do banco de dados miRBase versão 22 (http://www.mirbase.org/) que foi consultado em 16 de maio de 2020.

A regulação da expressão gênica em eucariotos é realizada através de um controle fino, na qual miRNAs e suas vias de silenciamento realizam uma regulação específica da expressão gênica e manutenção da integridade do genoma. Quase todos os processos biológicos, incluindo diferenciação celular, desenvolvimento de tecidos e metabolismo celular podem ser modulado por miRNAs (BARTEL, 2009).

A identificação de miRNAs conservados em moluscos revela diversos mecanismos de modulação nos processos biológicos dentre eles a estivação, que é um fenômeno caracterizado por redução da taxa metabólica para menos de 30% nas condições de repouso. Em um estudo realizado no molusco gastrópode pulmonar terrestre *Otala láctea* (caracol de leite), o fenômeno da estivação é necessário devido às condições ambientais estressantes, com altas temperaturas, comida e água limitadas. Este molusco reduz sua taxa metabólica em um fenômeno parecido com a hibernação dos ursos. Este fenômeno foi verificado também nos caracóis maçã invasores como *P. maculata,* sendo fundamental à sobrevivência da espécie em climas com períodos prolongados de seca (HOYECK; HADJ-MOUSSA, 2019; MUECK et al., 2018).

Os miRNAs são potenciais candidatos desta regulação na estivação, uma vez que são rápidos, reversíveis e com baixa demanda de energia. Neste estudo foi verificada a expressão de 75 miRNA conservados, dos quais 26 miRNAs foram superexpressos durante a estivação no molusco *O. láctea*, sendo seus genes suprimidos durante a dormência. Estes miRNAs superexpressos são implicados nos mecanismos de sobrevivência celular como: i) anti-apoptose; ii) supressão de tumores e; iii) respostas de manutenção muscular. O presente estudo mostrou níveis elevados de cinco miRNAs anti-apoptose (miR-2a-3p, miR-2c-3p, miRNA-124c, miRNA-153) e miRNA-190 no músculo do pé (HOYECK; HADJ-MOUSSA, 2019).

Outro mecanismo de controle gênico é exercido por miRNAs no abalone *Haliotis midae* (molusco gastrópode marinho) cultivado na África do Sul. O animal é considerado uma iguaria na culinária e perdas comerciais ocorrem devido a doenças infecciosas no abalone. Os miRNAs são capazes de modular a respostas imune em resposta ao estresse, causado pelos processos infecciosos. No estudo preliminar pretendia-se identificar possíveis pequenos RNAs moduladores da resposta imune no molusco, para evitar processos infecciosos e perdas econômicas (PICONE; RHODE; ROODT-WILDING, 2017)**.** Os miRNAs silenciam genes alvo através da destruição específica de mRNA. A detecção de genes nos hospedeiros e não nos patógenos envolvidos na infecção podem ser alvos potenciais para o desenvolvimento de fármacos (HONG-GELLER; MICHEVA-VITEVA, 2010).

Em um estudo em *Lymnaea stagnalis* (molusco gastrópode de água doce) foram identificados 264 miRNAs maduros conservados e 219 novos miRNAs (WALKER *et al.*, 2018). Já em *Haliotis discus hannai* (molusco gastrópode marinho) foram identificados 205 miRNAs maduros, dentre estes 15 conservados e 190 novos miRNAs (HUANG *et al.*, 2018). Em animais bivalves (*Mytilus galloprovincialis*) foram identificados 104 miRNAs conservados e 33 novos miRNAs (YU *et al.*, 2020). Os novos miRNAS maduros identificados não correspondem a nenhuma outra espécie conhecida no miRBase versão 22 (http://www.mirbase.org/). Revela-se a importância da pesquisa de novos miRNAs em moluscos (LAGOS-QUINTANA *et al.*, 2001).

A identificação de miRNAs maduros e seus precursores é realizada por estratégias experimentais e computacionais. As abordagens experimentais, apesar de apresentarem evidências da presença de miRNAs, podem excluir moléculas expressas em certos estágios e tecidos (GOMES *et al*., 2011). Assim, análises *in silico* são úteis para espécies com genoma completo sequenciado, permitindo a descoberta de miRNAs usando informações de DNA de genoma inteiro (LI *et al*., 2010).

Os miRNAs let-7a, miR-279-3p e miR-8-3p mostraram-se abundantes em múltiplos tecidos de moluscos, enquanto miR-9a-3p, miR-183, miR-1986, miR-1990-3p e miR-1993-3p foram expressos de uma maneira específica em alguns tecidos. Os miRNAs 9a-3p, miR-1986 e miR-1990-3p foram altamente expressos nos tecidos do manto em comparação com outros tecidos, o que pode indicar um papel vital por parte desses miRNAs no processo de biomineralização em *Pinctada Fucata* (HUANG et al., 2019, 2021).

Não há registros de miRNAs em *P. canaliculata* e *P. maculata* cadastrados no banco de dados miRBase versão 22 (http://www.mirbase.org/). Os miRNAs, precursores e maduros, e os genes envolvidos na via de processamento de pequenos RNAs estão presentes no genoma e transcriptoma de *P. canaliculata* e *P. maculata*. Desta forma, o objetivo deste trabalho foi identificar e caracterizar miRNAs e, genes envolvidos em sua via de processamento, tanto no genoma e quanto no transcriptoma das espécies de moluscos *P. canaliculata* e *P. maculata*.

3 MATERIAIS E MÉTODOS

3.1 BANCO DE DADOS DOS MOLUSCOS *P. canaliculata* E *P. maculata*

As sequências genômicas e transcriptômicas dos moluscos em estudo foram buscadas no banco de dados NCBI (National Centre for Biotechnology Information — http://.ncbi.nlm.nih.gov/) e do GigaDB (http://gigadb.org)(GIGADB), [s.d.]., com a finalidade de identificar e analisar miRNAs e os genes envolvidos na via de processamento de pequenos RNAs.

3.2 PREDIÇÃO DE miRNAs PERCURSORES E MADUROS

A análise *in silico* foi realizada utilizando um algoritmo robusto desenvolvido por GOMES *et al*., 2011 para predição de miRNAs e seus precursores. As análises computacionais foram realizadas no Laboratório de Bioinformática e Análises Moleculares (LBAM), da Universidade Federal de Uberlândia (UFU) no Campus Patos de Minas, seguindo a metodologia estabelecida para predição de miRNAs e precursores a partir do genoma e transcriptoma (GOMES *et al*., 2011). Foram obtidas as sequências do genoma com tendência em formar estruturas de *hairpin* ou semelhança com miRNAs precursores utilizando o programa Einverted (ferramenta EMBOSS) e o programa BLASTn (NCBI). A identificação de miRNAs homólogos foi realizada a partir do miRBase versão 22 (http://www.mirbase.org/). Estas sequências foram submetidas a uma série de filtros para descartar sequências indesejáveis, permanecendo apenas as sequências correspondentes aos prováveis miRNAs. Os filtros utilizados foram baseados em características conservadas de precursores de miRNAs. Estes filtros foram: i) homologia com miRNAs maduros conservados; ii) conteúdo de GC (guanina e citosina) entre 30% a 65%; iii) Energia Mínima Livre (MFE); iv) homologia com regiões repetidas e; v) homologia com RNAs não codificadores de proteínas, exceto miRNAs.

3.3 ALINHAMENTO DOS miRNAs COM ORTÓLOGOS E CONSTRUÇÃO DA ÁRVORE FILOGENÉTICA

Os prováveis precursores de miRNAs foram comparados com seus ortólogos em vários níveis, através do alinhamento múltiplo de sequências e construção de árvores filogenéticas. As sequências de pré-miRNAs e seus respectivos miRNAs foram submetidos ao alinhamento múltiplo, por meio das ferramentas ClustalX 2.1 e RNAalifold (THOMPSON; GIBSON; HIGGINS, 2002). Para a ferramenta ClustalX 2.1 foram utilizados os seguintes parâmetros: abertura de espaço (“*gap opening*”) 22,50 e extensão do espaço (“*gap extension*”) 0,83 (TAKANE *et al.*, 2010). Os prováveis miRNAs precursores escolhidos pela metodologia descrita foram submetidos ao alinhamento entre as espécies mais próximas evolutivamente, bem como aquelas presentes em grupos taxonômicos importantes para comparação. Através do banco de dados miRBase versão 22 (http:// mirbase.org/) foi utilizada a ferramenta BLASTn, na qual foram escolhidos os pré-miRNAs ortólogos. Houve a comparação dos prováveis precursores da espécie estudada com os precursores de miRNAs ortólogos conservados, tanto a nível primário (sequências nucleotídicas) quanto ao nível de estrutura secundária dos precursores.

A análise filogenética dos prováveis pré-miRNAs de *P. canaliculata* e *P. maculata* foi realizada com o auxílio do programa MEGA versão X (TAMURA *et al.*, 2007) utilizando o método Neighbor-joining e aplicando o modelo Kimura dois parâmetros (“*Kimura-twoparameters*”), para estimular a divergência entre as sequências (SAITOU; NEI, 1987). As árvores consenso foram obtidas utilizando “bootstrap” de 5000 réplicas.

3.4 PREDIÇÃO DOS GENES E PROTEÍNAS ENVOLVIDOS NA BIOGÊNESE DA VIA DE miRNAs EM *P. canaliculata* E *P. maculata*

A princípio, para pesquisar as supostas sequências proteicas envolvidas na via miRNA de *P. canaliculata* e *P. maculata*, foi utilizada a ferramenta BLASTp (National Centre for Biotechnology Information - http://www.ncbi.nlm.nih.gov/) usando como consultas asequência de proteínas de referência de espécies animais, como *Drosophila melanogaster* e *C.elegans*, organismos modelos (Tabela 3). As supostas sequências de proteína e os prováveis genes ortólogos de *P. canaliculata* e *P. maculata* foram encontrados e coletados no proteoma predito das espécies no *National Centre for Biotechnology Information* (NCBI - http://www.ncbi.nlm.nih.gov/) e utilizados para análise comparativa de sequências. As sequências de aminoácidos obtidas foram utilizadas para caracterização e validação da maquinaria de processamento de miRNAs em *P. canaliculata* e *P. maculata*. No genoma da espécie foram localizadas as sequências dos genes utilizando informações oriundas dos arquivos de anotação disponível no banco de dados da espécie e um script em PERL, para a busca das posições dos nucleotídeos no genoma (CARDOSO *et al.*, 2020; GOMES *et al.*, 2011).

Tabela 3 - Proteínas ortólogas de organismos modelo utilizado para pesquisa no banco de dados

Organismo modelo	**Proteína**	**Código Acesso NCBI**
D. melanogaster	Exportina-1	NP_001303316.1
D. melanogaster	Drosha	NP_477436.1
D. melanogaster	TSN	NP_001261195.1
D. melanogaster	Argonauta-1	NP_001246314.1
D. melanogaster	Argonauta 2	NP_730054.1
D. melanogaster	Dicer-1	NP_524453.1
D. melanogaster	Argonauta 3	NP_001163498.1
D. melanogaster	Dicer-2	NP_001286540.1
C. elegans	Argonauta	CCD73272.1
C. elegans	Drosha	O01326.2
C. elegans	Dicer	CDK13387.1
C. elegans	Exportina-T	NP_001294153.1
C. elegans	Exportina-1	NP_001368720.1

Fonte: Dados do autor

3.5 ANÁLISE DE DOMÍNIOS CONSERVADOS, ALINHAMENTO MÚLTIPLO DE SEQUÊNCIAS E ANÁLISE FILOGENÉTICA DAS PROTEÍNAS DA VIA DE miRNAs

As sequências preditas de aminoácidos das prováveis proteínas envolvidas na via de processamento de miRNAs em *P. canaliculata* e *P. maculata* foram submetidas ao alinhamento múltiplo de sequência com suas prováveis proteínas ortólogas. Foi utilizada a ferramenta ClustalX 2.1 para analisar alinhamento de estruturas primárias e secundárias das proteínas preditas e seus ortólogos, realizado com parâmetros *default* (LARKIN *et al*., 2007).

A busca por famílias de domínios e motivos conservados das sequências preditas de aminoácidos das prováveis proteínas envolvidas na via de miRNAs de *P. canaliculata* e *P. maculata* foi realizada utilizando o banco de dados de família de proteínas PFAM versão 32.0 (*Protein Family database* - https://pfam.xfam.org/) (EL-GEBALI *et al*., 2019; FINN *et al*., 2010) e o banco de dados de domínios conservados CDD (https://www.ncbi.nlm.nih.gov/Structure/cdd/wrpsb.cgi) (LU *et al*., 2020).

As sequências das proteínas preditas de *P. canaliculata*, *P. maculata e* possíveis ortólogos de referência (RefSeq) foram submetidos a análises filogenéticas comparativas utilizando o programa MEGA versão X (TAMURA *et al*., 2007). As árvores filogenéticas foram construídas utilizando o método Neighbor-joining (NJ) (SAITOU; NEI, 1987) ecalculada com o modelo de substituição JTT. A árvore filogenética consenso foi elaborada utilizando a análise de teste de *bootstrap* para 2000 réplicas representando a história evolutivado grupo de sequências estudadas. A porcentagem de réplicas das árvores filogenéticas foi realizada pelo teste de *boostrop* apresentando no final a árvore consenso. As posições contendoespaços ou dados perdidos de sequências foram subtraídos do conjunto de dados (GOMES *et al*., 2011).

4 RESULTADOS E DISCUSSÃO

4.1 BANCO DE DADOS DOS MOLUSCOS *P. canaliculata* E *P. maculata*.

Foi realizada uma busca em alguns bancos de dados, a fim de identificar informações genômicas e transcriptomicas das espécies *P. canaliculata* e *P. maculata* incluindo o banco de dados do NCBI (National Centre for Biotechnology Information – http://www.ncbi.nlm.nih.gov/), com registros descritos na Tabela 4 abaixo.

Tabela 4: Dados extraídos do banco de dados NCBI de *P. canaliculata* e *P. maculata.*

Espécie	**Genoma**	**Transcriptoma**	**Proteoma**	**Regitro no Genbank**
P. *canaliculata*	BioProject: PRJNA523959	transcriptoma	proteoma predito	sim
P. maculata	BioProject: PRJNA523958	transcriptoma	proteoma predito	sim

Fonte: Dados do autor

4.2 PREDIÇÃO DE miRNAs PERCURSORES E MADUROS

A baixa conservação nucleotídica nos pré-miRNAs restringem as buscas baseadas apenas em métodos de alinhamento de sequências tradicionais (WANG *et al*., 2005). No entanto, para identificação e caracterização destas moléculas são utilizados alguns parâmetros tais como: energia mínima da estrutura secundária, energia livre ajustada, conteúdo de GC dentre outros (Tabelas 5 e 6) (XUE *et al*., 2005). A forma mais exata para estudar os pré-miRNAs é feita através de suas características estruturais e termodinâmicas (ZHANG *et al*., 2009). Para identificação de miRNAs, maduros e precursores, no genoma e transcriptoma de *P. canaliculata* e *P. maculata,* as sequências foram analisadas por suas características estruturais e termodinâmicas e processados computacionalmente de forma a encontrar estruturas secundárias semelhantes aos pré-miRNAs (*hairpins).* As moléculas passaram por diversos filtros como descritos em Materiais e Métodos. Os crivos utilizados foram baseados em características de miRNAs conhecidos (precursores e maduros). Todos os miRNAs identificados obtiveram estrutura secundária estável, ou seja, com energia mínima livre menor que -20 kcal/mol.

Baseado nestas características, neste trabalho foi possível identificar 269 preditos pré-miRNAs em *P. canaliculata* e 299 prováveis precursores em *P. maculata*. O tamanho dos precursores das espécies *Lophotrochozoa* situa-se entre 57 a 153 nucleotídeos, com uma média de 90 nucleotídeos por sequência (GOMES *et al*., 2011). No estudo, o tamanho médio dos precursores de *P. canaliculada* é de 89,55 nucleotídeos e 89,27 nucleotídeos em *P. maculata*. A variação do tamanho dos pré-miRNAs em *P. canaliculata* (63 a 144 nucleotídeos) e *P. maculata* (65 a 100 nucleotídeos) (Tabela 5, coluna 2 e Tabela 6, coluna 2) respectivamente. O tamanho dos pre-miRNAs em espécies animais varia entre 47 a 177 nucleotídeos com um média de 87 ± 29 nucleotídeos (ZHOU *et al*., 2009). Os dados do presente estudo corroboram com o tamanho médio de pré-miRNAs das espécies Lophochotrozoa e metazoários de forma geral.

Os precursores identificados neste trabalho apresentaram estrutura secundária característica estável, com capacidade de dobramento. No formato de grampo de cabelo (*hairpin*), o modelo canônico de pré-miRNAs apresenta duas hastes na configuração de *dúplex* e um contorno na extremidade da sequência. As características termodinâmicas das estruturas secundárias destas moléculas se mostraram dentro dos valores padrões de pré-miRNAS. Conforme observado nas Tabela 5, coluna 4 e Tabela 6, coluna 4 respectivamente, a característica MFE (energia mínima livre) para *P. canaliculata* apresenta um valor médio de -28,70 kcal/mol e -27,55 kcal/ mol para *P. maculata*, sendo valor próximo quando comparado as espécies Lophotrochozoa (-31,27 kcal/mol). Os parâmetros estruturais como a porcentagem de GC (conteúdo) também não apresentaram diferenças estatísticas; *P. canaliculata, P. maculata* e espécies Lophotrochozoa apresentam média de 41,70% , 40,74% e 40,49% respectivamente (GOMES *et al*., 2011). Dessa forma, verifica-se que as características estruturais e termodinâmicas dos pré-miRNAs de *P. canaliculata* e *P. maculata* são conservadas, quando comparados com miRNAs de espécies identificadas corroborando achados relacionados aos pré-miRNAs das espécies em estudo.

Tabela 5: Características estruturais e termodinâmicas dos pré-miRNAs de *P. canaliculata*

miRNA	**Tamanho Nucleotídeos**	**GC (%) Conteúdo**	**MFE kcal/mol**	**MFE-conjunto Kcal/mol**	**Frequência (%)**	**Diversidade (%)**	**AMFE Kcal/mol**	**MFEI**
pcr-bantam-	**70**	**37,14285714**	**-29,4**	**-29,4**	**0,496236**	**1,25**	**-42**	**-1,130769231**
pcr-let-7b	**84**	**36,9047619**	**-18,7**	**-18,7**	**0,092975**	**12,14**	**-22,26190476**	**-0,603225806**
pcr-let-7d	**85**	**43,52941176**	**-35,8**	**-29,3**	**0,0411056**	**14,27**	**-42,11764706**	**-0,967567568**
pcr-let-7f	**98**	**44,89795918**	**-28**	**-20,9**	**0,0575038**	**32,52**	**-28,57142857**	**-0,636363636**
pcr-miR-1	**95**	**26,31578947**	**-18,5**	**-17,2**	**0,0456552**	**8,13**	**-19,47368421**	**-0,74**
pcr-miR-1002	**92**	**35,86956522**	**-21,4**	**-20,5**	**0,0641334**	**16,07**	**-23,26086957**	**-0,648484848**

pcr-miR-10a	95	43,15789474	-36,3	-36,2	0,0249508	6,41	-38,21052632	-0,885365854
pcr-miR-11	76	46,05263158	-23,7	-23,7	0,279042	5,58	-31,18421053	-0,677142857
pcr-miR-1175-	98	44,89795918	-42,7	-38,6	0,0798555	5,62	-43,57142857	-0,970454545
pcr-miR-1187	100	42	-30,8	-25,4	0,0295784	28,62	-30,8	-0,733333333
pcr-miR-12	85	38,82352941	-41,7	-40,3	0,196235	2,83	-49,05882353	-1,263636364
pcr-miR-122b	91	56,04395604	-27,9	-27,4	0,0537648	26,46	-30,65934066	-0,547058824
pcr-miR-1238	100	61	-49,3	-47,6	0,0980696	9,54	-49,3	-0,808196721
pcr-miR-124-5p	74	48,64864865	-33,9	-32,5	0,275153	6,77	-45,81081081	-0,941666667
pcr-miR-124c	73	49,31506849	-19,3	-17,4	0,0833127	8,41	-26,43835616	-0,536111111
pcr-miR-1277-1	92	26,08695652	-19,4	-19,4	0,182954	5,41	-21,08695652	-0,808333333
pcr-miR-1277-2	91	21,97802198	-21,1	-21,1	0,15848	5,1	-23,18681319	-1,055
pcr-miR-1322	72	36,11111111	-19,5	-15,5	0,0305019	11,17	-27,08333333	-0,75
pcr-miR-1328	91	26,37362637	-22,2	-17,7	0,0382424	22,78	-24,3956044	-0,925
pcr-miR-133	102	41,17647059	-42	-38,2	0,118541	3,77	-41,17647059	-1
pcr-miR-133c	75	45,33333333	-30,4	-28,9	0,184949	4,61	-40,53333333	-0,894117647
pcr-miR-137b	84	39,28571429	-33,1	-29,3	0,0390707	10	-39,4047619	-1,003030303
pcr-miR-13b	95	32,63157895	-24,2	-23,8	0,0196353	15,65	-25,47368421	-0,780645161
pcr-miR-1421a	79	53,16455696	-23,9	-23,9	0,0165133	12,42	-30,25316456	-0,569047619
pcr-miR-1421am	96	58,33333333	-31,8	-26,1	0,0477227	21,41	-33,125	-0,567857143
pcr-miR-143	80	48,75	-22,9	-21,7	0,179141	6,12	-28,625	-0,587179487
pcr-miR-153-1	93	32,25806452	-19,2	-19,2	0,260282	8,93	-20,64516129	-0,64
pcr-miR-153-2	87	47,12643678	-37,9	-37,9	0,256161	6,4	-43,56321839	-0,924390244
pcr-miR-1584	78	34,61538462	-20,5	-16	0,0704864	11,15	-26,28205128	-0,759259259
pcr-miR-15c	94	40,42553191	-32	-28,3	0,131794	8,15	-34,04255319	-0,842105263
pcr-miR-1602	96	42,70833333	-25,3	-25,2	0,141519	11,45	-26,35416667	-0,617073171
pcr-miR-1603	86	23,25581395	-20,1	-20,1	0,0339781	10,39	-23,37209302	-1,005
pcr-miR-1632	97	44,32989691	-26	-26	0,016941	14,04	-26,80412371	-0,604651163
pcr-miR-1677	99	51,51515152	-36,2	-35,3	0,0264037	8,83	-36,56565657	-0,709803922
pcr-miR-1744-1	67	43,28358209	-19,2	-19,2	0,273743	3	-28,65671642	-0,662068966

pcr-miR-1744-2	96	44,79166667	-27,6	-27,6	0,347188	6,28	-28,75	-0,641860465
pcr-miR-184	97	50,51546392	-33,8	-33,8	0,131517	7,97	-34,84536082	-0,689795918
pcr-miR-190	82	47,56097561	-32,5	-32,3	0,0800475	9,84	-39,63414634	-0,833333333
pcr-miR-1905c	90	64,44444444	-31,8	-31,7	0,0894559	8,15	-35,33333333	-0,548275862
pcr-miR-1951	94	44,68085106	-26,2	-17,5	0,0795727	16,28	-27,87234043	-0,623809524
pcr-miR-1976	73	64,38356164	-28,7	-28,7	0,157786	6,45	-39,31506849	-0,610638298
pcr-miR-1984	95	60	-47,8	-47,8	0,195263	3,22	-50,31578947	-0,838596491
pcr-miR-1985	63	23,80952381	-27	-27	0,311248	2,46	-42,85714286	-1,8
pcr-miR-1986	76	57,89473684	-33,4	-33,1	0,171582	3,66	-43,94736842	-0,759090909
pcr-miR-1990	72	56,94444444	-30,6	-30,6	0,28064	3,02	-42,5	-0,746341463
pcr-miR-1992	74	47,2972973	-32,7	-31,7	0,194317	3,34	-44,18918919	-0,934285714
pcr-miR-1994	90	55,55555556	-41,1	-41,1	0,501637	4,6	-45,66666667	-0,822
pcr-miR-1994a	72	56,94444444	-36,5	-36,2	0,349274	3,61	-50,69444444	-0,890243902
pcr-miR-1996b	99	37,37373737	-42,2	-42,2	0,0711267	11,72	-42,62626263	-1,140540541
pcr-miR-1a	65	36,92307692	-23,7	-22,1	0,0750244	6,51	-36,46153846	-0,9875
pcr-miR-1c	65	36,92307692	-27,1	-26	0,4687	2,23	-41,69230769	-1,129166667
pcr-miR-2001	76	36,84210526	-36,2	-36,2	0,193282	4,14	-47,63157895	-1,292857143
pcr-miR-2008	96	36,45833333	-24,3	-24,3	0,153115	8,97	-25,3125	-0,694285714
pcr-miR-203b	100	31	-20,1	-18,9	0,0165305	14,54	-20,1	-0,648387097
pcr-miR-204	68	48,52941176	-21,9	-21,9	0,137453	4,4	-32,20588235	-0,663636364
pcr-miR-2047	85	49,41176471	-23,3	-22,2	0,0824169	6,14	-27,41176471	-0,554761905
pcr-miR-2155	86	44,18604651	-20,5	-17,1	0,0594018	25,69	-23,8372093	-0,539473684
pcr-miR-216a	87	33,33333333	-34	-34	0,0973746	3,66	-39,08045977	-1,172413793
pcr-miR-216b-1	93	58,06451613	-34,9	-29	0,0813189	17,93	-37,52688172	-0,646296296
pcr-miR-216b-2	83	51,80722892	-31	-30,7	0,290814	5,92	-37,34939759	-0,720930233
pcr-miR-219-1	77	41,55844156	-27,4	-27,4	0,170426	3,13	-35,58441558	-0,85625
pcr-miR-219-2	67	40,29850746	-21,9	-21,9	0,222542	3,22	-32,68656716	-0,811111111
pcr-miR-2196	88	45,45454545	-21,1	-14,7	0,0688999	11,59	-23,97727273	-0,5275
pcr-miR-2218b	99	37,37373737	-21,1	-19,4	0,0518702	8,22	-21,31313131	-0,57027027

pcr-miR-2284c	**90**	**34,44444444**	**-19,4**	**-19,4**	**0,0416421**	**12,72**	**-21,55555556**	**-0,625806452**
pcr-miR-2284t	**100**	**38**	**-19,3**	**-17,8**	**0,0469214**	**9,84**	**-19,3**	**-0,507894737**
pcr-miR-2324	**91**	**58,24175824**	**-29,7**	**-20,7**	**0,0582309**	**28,39**	**-32,63736264**	**-0,560377358**
pcr-miR-2392	**100**	**56**	**-43,6**	**-43,1**	**0,0445531**	**14,74**	**-43,6**	**-0,778571429**
pcr-miR-2405	**74**	**21,62162162**	**-20,3**	**-20,3**	**0,0607476**	**10,16**	**-27,43243243**	**-1,26875**
pcr-miR-2464	**98**	**29,59183673**	**-27,8**	**-26,8**	**0,142155**	**8,83**	**-28,36734694**	**-0,95862069**
pcr-miR-2470	**67**	**38,80597015**	**-19,9**	**-19,9**	**0,542669**	**2,57**	**-29,70149254**	**-0,765384615**
pcr-miR-2491-1	**95**	**25,26315789**	**-23,2**	**-23,2**	**0,178642**	**4,92**	**-24,42105263**	**-0,966666667**
pcr-miR-2491-2	**86**	**37,20930233**	**-27,25**	**-26,3**	**0,182896**	**6,2**	**-31,68604651**	**-0,8515625**
pcr-miR-2491-3	**94**	**22,34042553**	**-19**	**-15,7**	**0,14487**	**14,83**	**-20,21276596**	**-0,904761905**
pcr-miR-2500	**87**	**35,63218391**	**-24,7**	**-24,1**	**0,0531783**	**5,65**	**-28,3908046**	**-0,796774194**
pcr-miR-252a-	**94**	**51,06382979**	**-37,6**	**-37,6**	**0,0595885**	**5,1**	**-40**	**-0,783333333**
pcr-miR-254	**90**	**38,88888889**	**-21,5**	**-16,1**	**0,0169726**	**23,25**	**-23,88888889**	**-0,614285714**
pcr-miR-279	**94**	**48,93617021**	**-39,9**	**-38,8**	**0,123884**	**5,39**	**-42,44680851**	**-0,867391304**
pcr-miR-279-2	**91**	**49,45054945**	**-39,9**	**-38,8**	**0,126379**	**5,36**	**-43,84615385**	**-0,886666667**
pcr-miR-279b	**88**	**47,72727273**	**-27,8**	**-27,3**	**0,0316856**	**14,17**	**-31,59090909**	**-0,661904762**
pcr-miR-2804	**99**	**35,35353535**	**-25,6**	**-25,4**	**0,00606276**	**12,93**	**-25,85858586**	**-0,731428571**
pcr-miR-281	**88**	**48,86363636**	**-35,2**	**-35,1**	**0,187538**	**3,76**	**-40**	**-0,818604651**
pcr-miR-2962	**94**	**59,57446809**	**-36**	**-36**	**0,402579**	**13,67**	**-38,29787234**	**-0,642857143**
pcr-miR-297	**96**	**28,125**	**-23,6**	**-19,07**	**0,0416441**	**16,8**	**-24,58333333**	**-0,874074074**
pcr-miR-29a	**89**	**49,43820225**	**-36,3**	**-36,3**	**0,263044**	**3,27**	**-40,78651685**	**-0,825**
pcr-miR-29b	**95**	**53,68421053**	**-35,8**	**-30,9**	**0,0431879**	**34,86**	**-37,68421053**	**-0,701960784**
pcr-miR-2a-1	**99**	**44,44444444**	**-40,8**	**-39,5**	**0,13114**	**5,78**	**-41,21212121**	**-0,927272727**
pcr-miR-2a-2	**89**	**43,82022472**	**-39,8**	**-39,4**	**0,329351**	**1,72**	**-44,71910112**	**-1,020512821**
pcr-miR-2a-3	**85**	**47,05882353**	**-40,5**	**-40,2**	**0,12355**	**3,87**	**-47,64705882**	**-1,0125**
pcr-miR-2a-4	**98**	**38,7755102**	**-32,4**	**-30,6**	**0,111894**	**7,44**	**-33,06122449**	**-0,852631579**
pcr-miR-2b-1	**98**	**38,7755102**	**-24,8**	**-24,8**	**0,336462**	**7,7**	**-25,30612245**	**-0,652631579**
pcr-miR-2b-2	**85**	**47,05882353**	**-34,5**	**-34,5**	**0,219013**	**4,37**	**-40,58823529**	**-0,8625**
pcr-miR-2e	**84**	**34,52380952**	**-19,3**	**-19,3**	**0,294993**	**3,58**	**-22,97619048**	**-0,665517241**

pcr-miR-2f	**96**	**47,91666667**	**-28,6**	**-28,6**	**0,0663966**	**6,92**	**-29,79166667**	**-0,62173913**
pcr-miR-3027	**88**	**43,18181818**	**-25,5**	**-23,3**	**0,0225252**	**10,35**	**-28,97727273**	**-0,671052632**
pcr-miR-3050	**89**	**39,3258427**	**-23,6**	**-23,6**	**0,62089**	**5,09**	**-26,51685393**	**-0,674285714**
pcr-miR-3057	**90**	**42,22222222**	**-24,5**	**-23**	**0,146061**	**9,33**	**-27,22222222**	**-0,644736842**
pcr-miR-3071	**96**	**26,04166667**	**-20,1**	**-19,8**	**0,178747**	**6,13**	**-20,9375**	**-0,804**
pcr-miR-308	**74**	**59,45945946**	**-32,3**	**-32,3**	**0,25533**	**2,19**	**-43,64864865**	**-0,734090909**
pcr-miR-3084a	**99**	**38,38383838**	**-27,5**	**-22,4**	**0,0887203**	**12,04**	**-27,77777778**	**-0,723684211**
pcr-miR-31	**98**	**47,95918367**	**-39,3**	**-36,9**	**0,095943**	**6,94**	**-40,10204082**	**-0,836170213**
pcr-miR-315	**78**	**48,71794872**	**-33,4**	**-33,4**	**0,276248**	**3,37**	**-42,82051282**	**-0,878947368**
pcr-miR-317-1	**94**	**40,42553191**	**-40,1**	**-40,1**	**0,224017**	**2,93**	**-42,65957447**	**-1,055263158**
pcr-miR-317-2	**99**	**44,44444444**	**-22,1**	**-22,1**	**0,047361**	**10,2**	**-22,32323232**	**-0,502272727**
pcr-miR-3173	**88**	**46,59090909**	**-29**	**-29**	**0,195168**	**5,51**	**-32,95454545**	**-0,707317073**
pcr-miR-33	**83**	**43,37349398**	**-38,7**	**-37,7**	**0,204831**	**5,09**	**-46,62650602**	**-1,075**
pcr-miR-340	**95**	**31,57894737**	**-21,5**	**-20,6**	**0,0124846**	**17,74**	**-22,63157895**	**-0,716666667**
pcr-miR-345	**98**	**57,14285714**	**-28,8**	**-28,8**	**0,301092**	**13,36**	**-29,3877551**	**-0,514285714**
pcr-miR-3529	**91**	**42,85714286**	**-30,8**	**-29,7**	**0,103281**	**5,67**	**-33,84615385**	**-0,78974359**
pcr-miR-3547	**98**	**44,89795918**	**-22,6**	**-22,6**	**0,156123**	**13,36**	**-23,06122449**	**-0,513636364**
pcr-miR-355	**98**	**30,6122449**	**-22,1**	**-22,1**	**0,0174252**	**13,25**	**-22,55102041**	**-0,736666667**
pcr-miR-3609	**92**	**33,69565217**	**-18,5**	**-17**	**0,0661525**	**14,65**	**-20,10869565**	**-0,596774194**
pcr-miR-3680	**97**	**43,29896907**	**-32,3**	**-32**	**0,0650423**	**7,8**	**-33,29896907**	**-0,769047619**
pcr-miR-36b	**70**	**55,71428571**	**-30,8**	**-30,8**	**0,109108**	**5,78**	**-44**	**-0,78974359**
pcr-miR-3716b	**72**	**36,11111111**	**-20,4**	**-20,4**	**0,0862155**	**4,88**	**-28,33333333**	**-0,784615385**
pcr-miR-3739	**94**	**41,4893617**	**-39,3**	**-38**	**0,332678**	**4,7**	**-41,80851064**	**-1,007692308**
pcr-miR-375-1	**98**	**54,08163265**	**-34,5**	**-34,5**	**0,452928**	**2,76**	**-35,20408163**	**-0,650943396**
pcr-miR-375-2	**70**	**45,71428571**	**-20,3**	**-20,3**	**0,298895**	**4,19**	**-29**	**-0,634375**
pcr-miR-377	**81**	**43,20987654**	**-24**	**-24**	**0,0938628**	**9,2**	**-29,62962963**	**-0,685714286**
pcr-miR-3776	**98**	**36,73469388**	**-25,2**	**-24,8**	**0,0725917**	**27,27**	**-25,71428571**	**-0,7**
pcr-miR-3809	**99**	**37,37373737**	**-24,9**	**-24,7**	**0,0212654**	**13,08**	**-25,15151515**	**-0,672972973**
pcr-miR-3831	**69**	**42,02898551**	**-23,4**	**-23,4**	**0,214068**	**5,17**	**-33,91304348**	**-0,806896552**
pcr-miR-3928	**93**	**38,70967742**	**-21,1**	**-19,7**	**0,138205**	**13,46**	**-22,68817204**	**-0,586111111**

pcr-miR-3957	**78**	**28,20512821**	**-39,7**	**-39,7**	**0,412668**	**1,21**	**-50,8974359**	**-1,804545455**
pcr-miR-3965	**81**	**41,97530864**	**-26,4**	**-26,4**	**0,24589**	**2,99**	**-32,59259259**	**-0,776470588**
pcr-miR-4057	**79**	**62,02531646**	**-33,5**	**-28,4**	**0,21803**	**20,3**	**-42,40506329**	**-0,683673469**
pcr-miR-4066	**100**	**38**	**-29,1**	**-22,3**	**0,120404**	**15,88**	**-29,1**	**-0,765789474**
pcr-miR-4070	**76**	**44,73684211**	**-25,8**	**-25,2**	**0,153161**	**8,62**	**-33,94736842**	**-0,758823529**
pcr-miR-4185	**99**	**34,34343434**	**-24**	**-18,6**	**0,0912733**	**7,49**	**-24,24242424**	**-0,705882353**
pcr-miR-4187	**84**	**28,57142857**	**-21**	**-15,37**	**0,0100236**	**18,73**	**-25**	**-0,875**
pcr-miR-4195	**85**	**32,94117647**	**-22,2**	**-22,2**	**0,0435326**	**6,38**	**-26,11764706**	**-0,792857143**
pcr-miR-4271	**100**	**48**	**-35,9**	**-32,3**	**0,296371**	**32,35**	**-35,9**	**-0,747916667**
pcr-miR-4323	**78**	**55,12820513**	**-28,2**	**-24,9**	**0,122046**	**7,91**	**-36,15384615**	**-0,655813953**
pcr-miR-4472	**79**	**29,11392405**	**-38,6**	**-38,6**	**0,308719**	**10,28**	**-48,86075949**	**-1,67826087**
pcr-miR-449a	**97**	**41,2371134**	**-22,3**	**-19,2**	**0,0202407**	**9,91**	**-22,98969072**	**-0,5575**
pcr-miR-449d	**97**	**44,32989691**	**-25,6**	**-22,8**	**0,0371522**	**9,97**	**-26,39175258**	**-0,595348837**
pcr-miR-4525	**70**	**62,85714286**	**-31**	**-30,8**	**0,214837**	**9,25**	**-44,28571429**	**-0,704545455**
pcr-miR-4617	**85**	**43,52941176**	**-30,3**	**-19,95**	**0,101687**	**17,71**	**-35,64705882**	**-0,818918919**
pcr-miR-4654	**99**	**61,61616162**	**-41,7**	**-40,2**	**0,172557**	**4,86**	**-42,12121212**	**-0,683606557**
pcr-miR-466i-1	**98**	**24,48979592**	**-36,4**	**-36,4**	**0,12872**	**16,86**	**-37,14285714**	**-1,516666667**
pcr-miR-466i-2	**100**	**27**	**-34,8**	**-12,3**	**0,119059**	**30,37**	**-34,8**	**-1,288888889**
pcr-miR-466i-3	**89**	**31,46067416**	**-31,6**	**-16,94**	**0,0208055**	**35,43**	**-35,50561798**	**-1,128571429**
pcr-miR-466i-4	**94**	**24,46808511**	**-36,9**	**-36**	**0,0677417**	**18,68**	**-39,25531915**	**-1,604347826**
pcr-miR-466m	**88**	**23,86363636**	**-26,9**	**-24,4**	**0,0337553**	**13,99**	**-30,56818182**	**-1,280952381**
pcr-miR-466n	**95**	**24,21052632**	**-27,2**	**-25,5**	**0,120489**	**14,18**	**-28,63157895**	**-1,182608696**
pcr-miR-466p	**93**	**29,03225806**	**-19,9**	**-15,6**	**0,0790441**	**7,09**	**-21,39784946**	**-0,737037037**
pcr-miR-466q	**100**	**28**	**-25,7**	**-22,5**	**0,0212462**	**16,27**	**-25,7**	**-0,917857143**
pcr-miR-467f	**80**	**37,5**	**-26,5**	**-26,5**	**0,0594607**	**8,5**	**-33,125**	**-0,883333333**
pcr-miR-4715	**84**	**59,52380952**	**-31,6**	**-31,6**	**0,443187**	**3,44**	**-37,61904762**	**-0,632**
pcr-miR-4757	**93**	**37,6344086**	**-22,4**	**-21,8**	**0,139005**	**15,42**	**-24,08602151**	**-0,64**
pcr-miR-4801	**89**	**42,69662921**	**-19,8**	**-19,8**	**0,127971**	**8,44**	**-22,24719101**	**-0,521052632**

pcr-miR-4860	**100**	**38**	**-21**	**-15**	**0,00932122**	**18,79**	**-21**	**-0,552631579**
pcr-miR-49	**88**	**32,95454545**	**-26**	**-26**	**0,306727**	**8,11**	**-29,54545455**	**-0,896551724**
pcr-miR-4918	**83**	**56,62650602**	**-25,6**	**-24,9**	**0,0316219**	**11,38**	**-30,84337349**	**-0,544680851**
pcr-miR-4968-1	**94**	**39,36170213**	**-30,8**	**-28,6**	**0,0493341**	**13,84**	**-32,76595745**	**-0,832432432**
pcr-miR-4968-2	**96**	**37,5**	**-34,7**	**-33,6**	**0,230058**	**16,55**	**-36,14583333**	**-0,963888889**
pcr-miR-4968-3	**81**	**35,80246914**	**-27,9**	**-24**	**0,0741934**	**12,7**	**-34,44444444**	**-0,962068966**
pcr-miR-4968-4	**96**	**20,83333333**	**-21,1**	**-16,1**	**0,0142023**	**21,15**	**-21,97916667**	**-1,055**
pcr-miR-5317a	**93**	**50,53763441**	**-29,1**	**-29,1**	**0,378556**	**7,01**	**-31,29032258**	**-0,619148936**
pcr-miR-5391	**100**	**33**	**-18,8**	**-15,1**	**0,0106555**	**34,01**	**-18,8**	**-0,56969697**
pcr-miR-5392	**100**	**28**	**-18,9**	**-12,7**	**0,00671688**	**26,01**	**-18,9**	**-0,675**
pcr-miR-5440	**98**	**26,53061224**	**-20,2**	**-18,8**	**0,104707**	**19,27**	**-20,6122449**	**-0,776923077**
pcr-miR-5549	**93**	**26,88172043**	**-19,5**	**-19,3**	**0,0919819**	**11,56**	**-20,96774194**	**-0,78**
pcr-miR-559	**99**	**33,33333333**	**-20,1**	**-16,59**	**0,0392066**	**24,4**	**-20,3030303**	**-0,609090909**
pcr-miR-5594	**99**	**44,44444444**	**-22,2**	**-20**	**0,0158651**	**13,26**	**-22,42424242**	**-0,504545455**
pcr-miR-5595	**100**	**46**	**-26,5**	**-26,5**	**0,0907076**	**10,96**	**-26,5**	**-0,576086957**
pcr-miR-574-1	**140**	**40,71428571**	**-42,8**	**-36,9**	**0,00597424**	**29,56**	**-30,57142857**	**-0,750877193**
pcr-miR-574-2	**67**	**28,35820896**	**-29,2**	**-15,45**	**0,0698327**	**22,63**	**-43,58208955**	**-1,536842105**
pcr-miR-5965-5p	**89**	**42,69662921**	**-20,4**	**-19,7**	**0,0426282**	**9,27**	**-22,92134831**	**-0,536842105**
pcr-miR-6-	**66**	**34,84848485**	**-29**	**-25,9**	**0,187241**	**5,82**	**-43,93939394**	**-1,260869565**
pcr-miR-6013	**82**	**68,29268293**	**-33,6**	**-22,8**	**0,0766232**	**17,71**	**-40,97560976**	**-0,6**
pcr-miR-6037-1	**100**	**35**	**-27,2**	**-27,2**	**0,192392**	**10,82**	**-27,2**	**-0,777142857**
pcr-miR-6037-2	**84**	**38,0952381**	**-27,2**	**-27,2**	**0,62986**	**1,19**	**-32,38095238**	**-0,85**
pcr-miR-6076	**91**	**53,84615385**	**-30,3**	**-29,9**	**0,158352**	**9,81**	**-33,2967033**	**-0,618367347**
pcr-miR-6098	**84**	**41,66666667**	**-22**	**-19,4**	**0,0207902**	**16,09**	**-26,19047619**	**-0,628571429**
pcr-miR-6132-1	**75**	**33,33333333**	**-29,7**	**-28,6**	**0,15428**	**5,96**	**-39,6**	**-1,188**
pcr-miR-6132-2	**84**	**30,95238095**	**-34,2**	**-34,2**	**0,0260952**	**5,82**	**-40,71428571**	**-1,315384615**
pcr-miR-6132-3	**67**	**37,31343284**	**-30,2**	**-21,3**	**0,185778**	**16,57**	**-45,07462687**	**-1,208**
pcr-miR-6335	**92**	**42,39130435**	**-25,4**	**-25,4**	**0,193109**	**5,21**	**-27,60869565**	**-0,651282051**
pcr-miR-6416	**95**	**43,15789474**	**-21,2**	**-20,6**	**0,0525607**	**13,18**	**-22,31578947**	**-0,517073171**

pcr-miR-64c	**83**	**40,96385542**	**-23,4**	**-23,3**	**0,0672748**	**11,61**	**-28,19277108**	**-0,688235294**
pcr-miR-6505	**94**	**35,10638298**	**-21,1**	**-19,17**	**0,0134094**	**12,71**	**-22,44680851**	**-0,639393939**
pcr-miR-6516	**82**	**42,68292683**	**-19,7**	**-19,7**	**0,160405**	**4,33**	**-24,02439024**	**-0,562857143**
pcr-miR-6560	**69**	**44,92753623**	**-19,8**	**-18,8**	**0,0706231**	**8,29**	**-28,69565217**	**-0,638709677**
pcr-miR-6575	**73**	**57,53424658**	**-37,7**	**-37,7**	**0,136478**	**3,17**	**-51,64383562**	**-0,897619048**
pcr-miR-6587	**94**	**25,53191489**	**-20,8**	**-14,3**	**0,00508185**	**23,66**	**-22,12765957**	**-0,866666667**
pcr-miR-6603	**93**	**29,03225806**	**-18,6**	**-16,4**	**0,0336602**	**16,97**	**-20**	**-0,688888889**
pcr-miR-6680	**93**	**45,16129032**	**-24**	**-24**	**0,0375404**	**8,12**	**-25,80645161**	**-0,571428571**
pcr-miR-669f	**73**	**24,65753425**	**-25,6**	**-20,3**	**0,138486**	**10,86**	**-35,06849315**	**-1,422222222**
pcr-miR-67	**73**	**53,42465753**	**-42,5**	**-42,5**	**0,387173**	**1,28**	**-58,21917808**	**-1,08974359**
pcr-miR-6816	**98**	**57,14285714**	**-29,7**	**-29,4**	**0,0358295**	**9,47**	**-30,30612245**	**-0,530357143**
pcr-miR-6852	**98**	**38,7755102**	**-24,8**	**-24,8**	**0,119887**	**4,49**	**-25,30612245**	**-0,652631579**
pcr-miR-6894	**73**	**49,31506849**	**-25,9**	**-25,9**	**0,400909**	**2,89**	**-35,47945205**	**-0,719444444**
pcr-miR-6951	**80**	**41,25**	**-23,5**	**-23,4**	**0,272799**	**2,62**	**-29,375**	**-0,712121212**
pcr-miR-6974	**100**	**58**	**-34,3**	**-27,1**	**0,0158296**	**24,28**	**-34,3**	**-0,59137931**
pcr-miR-6977	**90**	**53,33333333**	**-29,4**	**-28,7**	**0,0366664**	**9,79**	**-32,66666667**	**-0,6125**
pcr-miR-6999	**95**	**46,31578947**	**-27,3**	**-17,3**	**0,0847578**	**25,48**	**-28,73684211**	**-0,620454545**
pcr-miR-7000	**87**	**44,82758621**	**-32,8**	**-32,5**	**0,12354**	**7,83**	**-37,70114943**	**-0,841025641**
pcr-miR-7072	**69**	**50,72463768**	**-22,9**	**-22,9**	**0,435548**	**12,1**	**-33,1884058**	**-0,654285714**
pcr-miR-71-1	**101**	**43,56435644**	**-43,3**	**-43,3**	**0,1969**	**5,13**	**-42,87128713**	**-0,984090909**
pcr-miR-71-2	**95**	**37,89473684**	**-19,9**	**-10,75**	**0,0389519**	**25,08**	**-20,94736842**	**-0,552777778**
pcr-miR-7151	**91**	**37,36263736**	**-33,5**	**-32,8**	**0,126184**	**5,54**	**-36,81318681**	**-0,985294118**
pcr-miR-7307	**97**	**40,20618557**	**-26,8**	**-23,4**	**0,00551393**	**16,02**	**-27,62886598**	**-0,687179487**
pcr-miR-7314	**99**	**50,50505051**	**-32,6**	**-28,7**	**0,0105412**	**12,05**	**-32,92929293**	**-0,652**
pcr-miR-7386e	**98**	**37,75510204**	**-24,7**	**-24,1**	**0,0259444**	**13,11**	**-25,20408163**	**-0,667567568**
pcr-miR-7389	**100**	**48**	**-40,8**	**-39,7**	**0,0839772**	**10,1**	**-40,8**	**-0,85**
pcr-miR-7398h	**93**	**51,61290323**	**-24,3**	**-24,3**	**0,0318795**	**3,28**	**-26,12903226**	**-0,50625**
pcr-miR-7399	**96**	**22,91666667**	**-23,9**	**-23,9**	**0,118362**	**17,65**	**-24,89583333**	**-1,086363636**
pcr-miR-743b	**79**	**48,10126582**	**-26,2**	**-25,6**	**0,0306733**	**10,47**	**-33,16455696**	**-0,689473684**

pcr-miR-745a	**64**	**51,5625**	**-30,5**	**-30,5**	**0,0883802**	**5,12**	**-47,65625**	**-0,924242424**
pcr-miR-745b	**83**	**57,8313253**	**-35,7**	**-31,3**	**0,0253652**	**10,13**	**-43,01204819**	**-0,74375**
pcr-miR-7472	**65**	**46,15384615**	**-20,8**	**-20,8**	**0,342745**	**5,01**	**-32**	**-0,693333333**
pcr-miR-750	**97**	**50,51546392**	**-52,7**	**-51,6**	**0,157091**	**3,48**	**-54,32989691**	**-1,075510204**
pcr-miR-751	**94**	**39,36170213**	**-27,8**	**-22,8**	**0,0515578**	**21,54**	**-29,57446809**	**-0,751351351**
pcr-miR-753d	**94**	**41,4893617**	**-22,8**	**-18,7**	**0,00974595**	**21,51**	**-24,25531915**	**-0,584615385**
pcr-miR-760	**67**	**49,25373134**	**-29,1**	**-28,8**	**0,330018**	**2,01**	**-43,43283582**	**-0,881818182**
pcr-miR-7643	**97**	**28,86597938**	**-20,4**	**-15,3**	**0,0261098**	**14,6**	**-21,03092784**	**-0,728571429**
pcr-miR-7649	**79**	**32,91139241**	**-19,5**	**-18,5**	**0,0657628**	**5,47**	**-24,6835443**	**-0,75**
pcr-miR-76b-1	**90**	**22,22222222**	**-43,4**	**-43,4**	**0,140211**	**4,23**	**-48,22222222**	**-2,17**
pcr-miR-76b-2	**97**	**31,95876289**	**-21**	**-14,9**	**0,0293543**	**18,5**	**-21,64948454**	**-0,677419355**
pcr-miR-770	**77**	**48,05194805**	**-19,8**	**-19,4**	**0,0757209**	**9,14**	**-25,71428571**	**-0,535135135**
pcr-miR-7880k	**76**	**44,73684211**	**-22,8**	**-18,67**	**0,0850987**	**18,19**	**-30**	**-0,670588235**
pcr-miR-7880q	**100**	**33**	**-19,6**	**-19,6**	**0,125227**	**11,54**	**-19,6**	**-0,593939394**
pcr-miR-79	**95**	**28,42105263**	**-18,8**	**-18,8**	**0,0982863**	**9,53**	**-19,78947368**	**-0,696296296**
pcr-miR-7901	**100**	**38**	**-28,4**	**-28**	**0,0588354**	**13,99**	**-28,4**	**-0,747368421**
pcr-miR-7c	**109**	**37,6146789**	**-37,2**	**-34,7**	**0,144483**	**9,32**	**-34,12844037**	**-0,907317073**
pcr-miR-8-1	**86**	**43,02325581**	**-39,6**	**-39,6**	**0,178161**	**3,42**	**-46,04651163**	**-1,07027027**
pcr-miR-8196b	**84**	**23,80952381**	**-19,6**	**-19,4**	**0,0526092**	**9,33**	**-23,33333333**	**-0,98**
pcr-miR-8-2	**94**	**45,74468085**	**-46**	**-46**	**0,192059**	**3,37**	**-48,93617021**	**-1,069767442**
pcr-miR-8296	**83**	**51,80722892**	**-35,3**	**-34,3**	**0,251447**	**8,1**	**-42,53012048**	**-0,820930233**
pcr-miR-8332	**99**	**33,33333333**	**-24,6**	**-24,5**	**0,280827**	**4,23**	**-24,84848485**	**-0,745454545**
pcr-miR-8335-1	**96**	**23,95833333**	**-27,7**	**-27,7**	**0,191059**	**9,75**	**-28,85416667**	**-1,204347826**
pcr-miR-8335-2	**93**	**26,88172043**	**-21**	**-18**	**0,0291252**	**12,45**	**-22,58064516**	**-0,84**
pcr-miR-8364g	**79**	**37,97468354**	**-29**	**-29**	**0,217216**	**5,42**	**-36,70886076**	**-0,966666667**
pcr-miR-8416	**87**	**31,03448276**	**-18,7**	**-18,7**	**0,245232**	**4,5**	**-21,49425287**	**-0,692592593**
pcr-miR-8422	**96**	**25**	**-23,6**	**-23,1**	**0,141217**	**4,11**	**-24,58333333**	**-0,983333333**
pcr-miR-8462-1	**76**	**39,47368421**	**-21,6**	**-18,9**	**0,11733**	**10,16**	**-28,42105263**	**-0,72**
pcr-miR-8462-2	**100**	**30**	**-19,5**	**-18**	**0,0907014**	**10,52**	**-19,5**	**-0,65**

pcr-miR-8485-1	98	20,40816327	-28,9	-25,3	0,0866314	28,22	-29,48979592	-1,445
pcr-miR-8485-2	99	21,21212121	-20,6	-10,9	0,0171759	20,99	-20,80808081	-0,980952381
pcr-miR-87	98	40,81632653	-42	-38,2	0,120168	3,71	-42,85714286	-1,05
pcr-miR-8834a	65	67,69230769	-28,3	-28,3	0,0895969	7	-43,53846154	-0,643181818
pcr-miR-888	78	50	-25,5	-24,5	0,0825903	6,42	-32,69230769	-0,653846154
pcr-miR-8915-1	144	59,72222222	-48,1	-38,5	0,00933589	30,97	-33,40277778	-0,559302326
pcr-miR-8915-2	144	58,33333333	-51,5	-47,1	0,029851	26,76	-35,76388889	-0,613095238
pcr-miR-8915-3	140	60,71428571	-50,2	-44,5	0,00925419	28,93	-35,85714286	-0,590588235
pcr-miR-8915-4	142	60,56338028	-51,2	-50,3	0,0239289	15,77	-36,05633803	-0,595348837
pcr-miR-8915-5	142	53,52112676	-48,4	-48,4	0,094252	13,02	-34,08450704	-0,636842105
pcr-miR-9015	91	51,64835165	-38,6	-36,5	0,0873791	7,6	-42,41758242	-0,821276596
pcr-miR-9-1	88	44,31818182	-27,8	-27,8	0,435416	2,15	-31,59090909	-0,712820513
pcr-miR-9120	93	49,46236559	-26,6	-26,6	0,320862	10,09	-28,60215054	-0,57826087
pcr-miR-9-2	80	38,75	-23,1	-20	0,134445	5,8	-28,875	-0,74516129
pcr-miR-92a-1	90	45,55555556	-31,5	-29,1	0,0366431	11,93	-35	-0,768292683
pcr-miR-92a-2	87	44,82758621	-35,5	-35,5	0,137395	3,64	-40,8045977	-0,91025641
pcr-miR-92b	89	51,68539326	-32	-32	0,0722739	6,85	-35,95505618	-0,695652174
pcr-miR-92c-	99	40,4040404	-26,6	-26,6	0,0552462	15,72	-26,86868687	-0,665
pcr-miR-9-3	93	44,08602151	-34,1	-34	0,28369	3,42	-36,66666667	-0,831707317
pcr-miR-9341-1	88	44,31818182	-31,8	-28,3	0,0187103	14,65	-36,13636364	-0,815384615
pcr-miR-9341-2	95	27,36842105	-29,5	-29,5	0,182004	3,22	-31,05263158	-1,134615385
pcr-miR-9388	88	27,27272727	-19	-17	0,162177	22,34	-21,59090909	-0,791666667
pcr-miR-9-4	93	44,08602151	-45,5	-45,5	0,314517	3,87	-48,92473118	-1,109756098
pcr-miR-1603	86	23,25581395	-20,1	-20,1	0,0339781	10,39	-23,37209302	-1,005
pcr-miR-96	99	25,25252525	-19,7	-17,9	0,0562546	11,21	-19,8989899	-0,788
pcr-miR-122b	91	56,04395604	-27,9	-27,4	0,0537648	26,46	-30,65934066	-0,547058824
pcr-miR-96b	70	38,57142857	-23,4	-23,4	0,19695	2,59	-33,42857143	-0,866666667
pcr-miR-972	98	32,65306122	-23,7	-19,6	0,0440182	24,34	-24,18367347	-0,740625
pcr-miR-981	85	49,41176471	-32,6	-28,1	0,0350449	8	-38,35294118	-0,776190476

pcr-miR-9891	100	65	-37,2	-28,7	0,102885	18,05	-37,2	-0,572307692
pcr-miR-995	91	53,84615385	-25,2	-20,28	0,0730743	27,75	-27,69230769	-0,514285714

AMFE: Energia Mínima Livre ajustada, MFE: Energia mínima livre, MFEI: Índice de energia mínima livre, MFEE: Energia mínima livre do conjunto

Fonte: Dados do autor.

Tabela 6: Características estruturais e termodinâmicas dos pré-miRNAs de *P. maculata*.

miRNA	Tamanho Nucleotídeos	GC (%) Conteúdo	MFE kcal/mol	MFE-conjunto Kcal/mol	Frequência (%)	Diversidade (%)	AMFE Kcal/mol	MFEI
pmc-bantam	70	37,14285714	-29,4	-29,4	0,496236	1,25	-42	-1,130769231
pmc-mir-1	95	26,31578947	-18,5	-17,2	0,0456552	8,13	-19,47368421	-0,74
pmc-mir-1000	74	48,64864865	-28	-27,1	0,0316861	6,94	-37,83783784	-0,777777778
pmc-mir-10031c	100	43	-27,5	-27,5	0,437289	4,9	-27,5	-0,639534884
pmc-mir-10046	98	63,26530612	-37,2	-35,3	0,365188	5,25	-37,95918367	-0,6
pmc-mir-10055	91	35,16483516	-33,1	-29,2	0,0940128	18,78	-36,37362637	-1,034375
pmc-mir-10089	94	36,17021277	-19	-17,9	0,0244499	17,13	-20,21276596	-0,558823529
pmc-mir-10173	75	32	-22,1	-21,7	0,383116	2,01	-29,46666667	-0,920833333
pmc-mir-10249-1	85	37,64705882	-23,1	-18,2	0,173316	22,89	-27,17647059	-0,721875
pmc-mir-10249-2	99	38,38383838	-27,2	-13,3	0,0369505	27,09	-27,47474747	-0,715789474
pmc-mir-10256	100	44	-30,5	-29	0,0749341	11,46	-30,5	-0,693181818
pmc-mir-10267	78	32,05128205	-21,7	-15,8	0,291539	8,02	-27,82051282	-0,868
pmc-mir-10492a	98	25,51020408	-22,8	-11,64	0,0595061	28,54	-23,26530612	-0,912
pmc-mir-10508b	94	28,72340426	-19,8	-16,4	0,0347757	12,65	-21,06382979	-0,733333333
pmc-mir-10509	76	50	-27,4	-24,3	0,170278	7,05	-36,05263158	-0,721052632
pmc-mir-10578	79	54,43037975	-24,8	-24,8	0,0248622	11,14	-31,39240506	-0,576744186
pmc-mir-10611	98	32,65306122	-20,3	-19,5	0,141481	6,16	-20,71428571	-0,634375
pmc-mir-10639	95	41,05263158	-29,6	-26,6	0,0690141	9,88	-31,15789474	-0,758974359
pmc-mir-10770-1	98	29,59183673	-26,4	-25	0,0143851	16,81	-26,93877551	-0,910344828
pmc-mir-10770-2	87	33,33333333	-28,5	-28,5	0,138113	8,95	-32,75862069	-0,982758621
pmc-mir-10892	98	44,89795918	-30,8	-30,8	0,239683	4,62	-31,42857143	-0,7

pmc-mir-10972	**97**	**38,1443299**	**-23,3**	**-23,3**	**0,070908**	**15,83**	**-24,02061856**	**-0,62972973**
pmc-mir-10b-1	**98**	**59,18367347**	**-34**	**-32,9**	**0,060236**	**18,2**	**-34,69387755**	**-0,586206897**
pmc-mir-10b-2	**93**	**41,93548387**	**-40,6**	**-40,6**	**0,262649**	**9,81**	**-43,65591398**	**-1,041025641**
pmc-mir-11054	**100**	**22**	**-21,4**	**-15,8**	**0,0894963**	**35,03**	**-21,4**	**-0,972727273**
pmc-mir-11280	**92**	**31,52173913**	**-21,9**	**-21,2**	**0,135428**	**13,09**	**-23,80434783**	**-0,755172414**
pmc-mir-1175-1	**98**	**44,89795918**	**-42,7**	**-38,6**	**0,0798555**	**5,62**	**-43,57142857**	**-0,970454545**
pmc-mir-1175-2	**98**	**20,40816327**	**-20,9**	**-20**	**0,0792969**	**13,02**	**-21,32653061**	**-1,045**
pmc-mir-1187	**99**	**23,23232323**	**-20,3**	**-19,5**	**0,0641874**	**14,27**	**-20,50505051**	**-0,882608696**
pmc-mir-11922	**79**	**45,56962025**	**-25,9**	**-25,9**	**0,042274**	**4,91**	**-32,78481013**	**-0,719444444**
pmc-mir-12	**85**	**38,82352941**	**-41,7**	**-40,3**	**0,196235**	**2,83**	**-49,05882353**	**-1,263636364**
pmc-mir-12096b	**94**	**42,55319149**	**-32**	**-31,6**	**0,104639**	**7,62**	**-34,04255319**	**-0,8**
pmc-mir-12228	**88**	**55,68181818**	**-33,2**	**-19,6**	**0,0758529**	**14,44**	**-37,72727273**	**-0,67755102**
pmc-mir-1224	**95**	**26,31578947**	**-24,7**	**-21,1**	**0,129321**	**12,41**	**-26**	**-0,988**
pmc-mir-12245	**96**	**68,75**	**-33,6**	**-33,1**	**0,0519779**	**10,23**	**-35**	**-0,509090909**
pmc-mir-12286	**80**	**56,25**	**-28,6**	**-19,19**	**0,08512**	**19,63**	**-35,75**	**-0,635555556**
pmc-mir-12287	**82**	**67,07317073**	**-31,7**	**-15,9**	**0,0997248**	**26,92**	**-38,65853659**	**-0,576363636**
pmc-mir-12292	**86**	**55,81395349**	**-26,4**	**-25,2**	**0,0975496**	**8,86**	**-30,69767442**	**-0,55**
pmc-mir-12293	**100**	**44**	**-39,4**	**-30,9**	**0,181223**	**7,86**	**-39,4**	**-0,895454545**
pmc-mir-12296	**79**	**43,03797468**	**-33,3**	**-33,3**	**0,678283**	**1,48**	**-42,15189873**	**-0,979411765**
pmc-mir-12321	**98**	**40,81632653**	**-38,7**	**-38,7**	**0,150689**	**6,68**	**-39,48979592**	**-0,9675**
pmc-mir-12327	**95**	**25,26315789**	**-29**	**-28,1**	**0,0680169**	**8,35**	**-30,52631579**	**-1,208333333**
pmc-mir-12339	**99**	**36,36363636**	**-43,9**	**-41,47**	**0,0371689**	**11,26**	**-44,34343434**	**-1,219444444**
pmc-mir-12358	**98**	**32,65306122**	**-26,7**	**-26,7**	**0,382757**	**2,63**	**-27,24489796**	**-0,834375**
pmc-mir-12396	**87**	**47,12643678**	**-35,8**	**-35,8**	**0,255939**	**5,41**	**-41,14942529**	**-0,873170732**
pmc-mir-124	**90**	**45,55555556**	**-27,7**	**-19,2**	**0,0731809**	**30,63**	**-30,77777778**	**-0,675609756**
pmc-mir-12410	**96**	**21,875**	**-25,7**	**-23,6**	**0,0448405**	**16,09**	**-26,77083333**	**-1,223809524**
pmc-mir-1273h	**92**	**63,04347826**	**-37,1**	**-12,11**	**0,0897343**	**23,69**	**-40,32608696**	**-0,639655172**
pmc-mir-1277	**66**	**30,3030303**	**-19,3**	**-18,4**	**0,217022**	**13,76**	**-29,24242424**	**-0,965**

pmc-mir-130b	100	26	-19,9	-19,9	0,0421788	8,59	-19,9	-0,765384615
pmc-mir-1322	98	30,6122449	-24,5	-22,2	0,0161323	23,72	-25	-0,816666667
pmc-mir-133c	75	45,33333333	-30,4	-28,9	0,184949	4,61	-40,53333333	-0,894117647
pmc-mir-1376	94	47,87234043	-31,4	-26,8	0,0729514	8,03	-33,40425532	-0,697777778
pmc-mir-137b	86	39,53488372	-33,1	-29,3	0,0221705	10,78	-38,48837209	-0,973529412
pmc-mir-13b	95	32,63157895	-25,4	-18,3	0,0969165	31,5	-26,73684211	-0,819354839
pmc-mir-1421am	96	58,33333333	-31,8	-26,1	0,0477227	21,41	-33,125	-0,567857143
pmc-mir-1422j	99	32,32323232	-22,3	-22,3	0,232835	3,91	-22,52525253	-0,696875
pmc-mir-143	91	48,35164835	-25,3	-24,9	0,120125	12,48	-27,8021978	-0,575
pmc-mir-149	96	37,5	-33	-12,4	0,0174308	29,49	-34,375	-0,916666667
pmc-mir-150	89	47,19101124	-25,1	-25,1	0,0814506	10,2	-28,20224719	-0,597619048
pmc-mir-153	87	47,12643678	-37,9	-37,9	0,256161	6,4	-43,56321839	-0,924390244
pmc-mir-1551	91	38,46153846	-19,4	-19,4	0,255295	14,12	-21,31868132	-0,554285714
pmc-mir-15c	94	41,4893617	-29,6	-28,4	0,0882062	10,15	-31,4893617	-0,758974359
pmc-mir-1603-1	86	23,25581395	-20,1	-20,1	0,0339781	10,39	-23,37209302	-1,005
pmc-mir-1603-2	94	25,53191489	-20,1	-19,5	0,0229654	9,06	-21,38297872	-0,8375
pmc-mir-1632	97	44,32989691	-26	-26	0,016941	14,04	-26,80412371	-0,604651163
pmc-mir-1642	83	42,1686747	-20,1	-14,7	0,16303	24,29	-24,21686747	-0,574285714
pmc-mir-1677	99	51,51515152	-36,2	-35,3	0,0264037	8,83	-36,56565657	-0,709803922
pmc-mir-17	75	61,33333333	-24,1	-19,8	0,261623	4,23	-32,13333333	-0,523913043
pmc-mir-1728	81	45,67901235	-18,9	-18,9	0,107603	12,57	-23,33333333	-0,510810811
pmc-mir-1744	67	43,28358209	-19,2	-19,2	0,273743	3	-28,65671642	-0,662068966
pmc-mir-1775	99	42,42424242	-34,9	-34,9	0,331788	5,38	-35,25252525	-0,830952381
pmc-mir-1781-1	87	35,63218391	-23,1	-23,1	0,071767	5,41	-26,55172414	-0,74516129
pmc-mir-1781-2	87	34,48275862	-23,4	-23,4	0,0797214	4,68	-26,89655172	-0,78
pmc-mir-1896	100	37	-32,2	-31,6	0,027684	15,11	-32,2	-0,87027027
pmc-mir-190	82	47,56097561	-32,5	-32,3	0,0800475	9,84	-39,63414634	-0,833333333
pmc-mir-1951	94	43,61702128	-23,5	-14,8	0,0639105	17,89	-25	-0,573170732

pmc-mir-1955	**99**	**59,5959596**	**-40,3**	**-34,2**	**0,115974**	**33,97**	**-40,70707071**	**-0,683050847**
pmc-mir-196a	**100**	**47**	**-31,3**	**-31,3**	**0,0722099**	**6,07**	**-31,3**	**-0,665957447**
pmc-mir-1984	**73**	**64,38356164**	**-37,9**	**-37,9**	**0,133827**	**2,65**	**-51,91780822**	**-0,806382979**
pmc-mir-1985	**67**	**26,86567164**	**-28,6**	**-28,6**	**0,287364**	**2,77**	**-42,68656716**	**-1,588888889**
pmc-mir-199	**93**	**39,78494624**	**-20,3**	**-17,4**	**0,0562745**	**14,69**	**-21,82795699**	**-0,548648649**
pmc-mir-1990	**95**	**56,84210526**	**-44,2**	**-43,8**	**0,106149**	**4,96**	**-46,52631579**	**-0,818518519**
pmc-mir-1994	**90**	**55,55555556**	**-41,1**	**-41,1**	**0,501637**	**4,6**	**-45,66666667**	**-0,822**
pmc-mir-1994a	**91**	**52,74725275**	**-44,3**	**-44,3**	**0,178398**	**5,12**	**-48,68131868**	**-0,922916667**
pmc-mir-1a	**65**	**36,92307692**	**-23,7**	**-22,1**	**0,0750244**	**6,51**	**-36,46153846**	**-0,9875**
pmc-mir-1b	**92**	**35,86956522**	**-19,8**	**-19,2**	**0,145182**	**8,89**	**-21,52173913**	**-0,6**
pmc-mir-1c	**65**	**36,92307692**	**-27,1**	**-26**	**0,4687**	**2,23**	**-41,69230769**	**-1,129166667**
pmc-mir-2001	**76**	**36,84210526**	**-36,2**	**-36,2**	**0,193282**	**4,14**	**-47,63157895**	**-1,292857143**
pmc-mir-200b	**86**	**39,53488372**	**-21,8**	**-21,6**	**0,126933**	**11,76**	**-25,34883721**	**-0,641176471**
pmc-mir-201	**82**	**59,75609756**	**-25,8**	**-25,8**	**0,163281**	**11,03**	**-31,46341463**	**-0,526530612**
pmc-mir-2017	**70**	**54,28571429**	**-29**	**-29**	**0,16383**	**21,21**	**-41,42857143**	**-0,763157895**
pmc-mir-204	**68**	**48,52941176**	**-21,9**	**-21,9**	**0,137453**	**4,4**	**-32,20588235**	**-0,663636364**
pmc-mir-2064	**78**	**44,87179487**	**-23**	**-22,9**	**0,480687**	**3,28**	**-29,48717949**	**-0,657142857**
pmc-mir-2155	**86**	**44,18604651**	**-20,5**	**-17,1**	**0,0594018**	**25,69**	**-23,8372093**	**-0,539473684**
pmc-mir-216a	**78**	**34,61538462**	**-30,5**	**-30,5**	**0,146188**	**3,64**	**-39,1025641**	**-1,12962963**
pmc-mir-216b-1	**83**	**51,80722892**	**-31**	**-30,7**	**0,290814**	**5,92**	**-37,34939759**	**-0,720930233**
pmc-mir-216b-2	**93**	**59,13978495**	**-34,1**	**-34**	**0,183887**	**24,97**	**-36,66666667**	**-0,62**
pmc-mir-216b-3	**91**	**39,56043956**	**-20,7**	**-20,6**	**0,0564236**	**15,43**	**-22,74725275**	**-0,575**
pmc-mir-2223	**89**	**35,95505618**	**-23,8**	**-18,8**	**0,195676**	**11,23**	**-26,74157303**	**-0,74375**
pmc-mir-2238i	**80**	**61,25**	**-27,6**	**-25,2**	**0,123572**	**5,73**	**-34,5**	**-0,563265306**
pmc-mir-2284c	**96**	**37,5**	**-22,9**	**-19,8**	**0,195896**	**4,07**	**-23,85416667**	**-0,636111111**
pmc-mir-2298	**93**	**53,76344086**	**-44,5**	**-41,7**	**0,259982**	**2,97**	**-47,84946237**	**-0,89**
pmc-mir-2304	**100**	**43**	**-32,8**	**-27,8**	**0,0591964**	**33,54**	**-32,8**	**-0,762790698**
pmc-mir-2361	**100**	**22**	**-25,8**	**-25,8**	**0,0359904**	**8,11**	**-25,8**	**-1,172727273**

pmc-mir-2371	85	22,35294118	-23,4	-20,9	0,161131	8,21	-27,52941176	-1,231578947
pmc-mir-2470	67	38,80597015	-19,9	-19,9	0,542669	2,57	-29,70149254	-0,765384615
pmc-mir-2491-1	94	25,53191489	-25,1	-23,1	0,0665504	7,31	-26,70212766	-1,045833333
pmc-mir-2491-2	86	24,41860465	-24,4	-24,2	0,192125	18,27	-28,37209302	-1,161904762
pmc-mir-2491-3	86	37,20930233	-27,25	-26,3	0,182896	6,2	-31,68604651	-0,8515625
pmc-mir-2493	93	38,70967742	-28,2	-25,4	0,239172	8,2	-30,32258065	-0,783333333
pmc-mir-2505	70	22,85714286	-20	-18,8	0,634526	4,68	-28,57142857	-1,25
pmc-mir-252a	94	51,06382979	-37,6	-37,6	0,0595885	5,1	-40	-0,783333333
pmc-mir-254	90	38,88888889	-21,5	-16,1	0,0169726	23,25	-23,88888889	-0,614285714
pmc-mir-2571	83	50,60240964	-29,2	-29,2	0,309071	7,4	-35,18072289	-0,695238095
pmc-mir-279	91	49,45054945	-39,9	-39,4	0,157559	5,72	-43,84615385	-0,886666667
pmc-mir-279b	88	46,59090909	-25,9	-23	0,0495292	18,81	-29,43181818	-0,631707317
pmc-mir-279c	100	60	-38,8	-38,3	0,103385	8,74	-38,8	-0,646666667
pmc-mir-281	88	40,90909091	-35,2	-35,1	0,187538	3,76	-40	-0,977777778
pmc-mir-282	85	25,88235294	-20,6	-20,6	0,141364	11,91	-24,23529412	-0,936363636
pmc-mir-2953	75	50,66666667	-23,8	-23,8	0,296458	3,88	-31,73333333	-0,626315789
pmc-mir-2a-1	89	42,69662921	-38,6	-38,2	0,276363	1,97	-43,37078652	-1,015789474
pmc-mir-2a-2	98	38,7755102	-34,2	-33,9	0,160179	6,48	-34,89795918	-0,9
pmc-mir-2a-3	91	43,95604396	-40,8	-40,8	0,328315	3,78	-44,83516484	-1,02
pmc-mir-2b-1	98	38,7755102	-24,8	-24,8	0,342782	4,57	-25,30612245	-0,652631579
pmc-mir-2b-2	85	45,88235294	-34,5	-34,5	0,221463	4,35	-40,58823529	-0,884615385
pmc-mir-2f	98	48,97959184	-28,7	-28,7	0,0719843	7,94	-29,28571429	-0,597916667
pmc-mir-300	68	54,41176471	-24,8	-23,9	0,250391	4,52	-36,47058824	-0,67027027
pmc-mir-302a	95	34,73684211	-26,3	-26,3	0,0199855	22,21	-27,68421053	-0,796969697
pmc-mir-302b	93	39,78494624	-25,2	-25,2	0,130818	5,39	-27,09677419	-0,681081081
pmc-mir-3057	86	43,02325581	-24,4	-20,9	0,20764	8,46	-28,37209302	-0,659459459
pmc-mir-3071	96	26,04166667	-20,1	-19,8	0,178747	6,13	-20,9375	-0,804
pmc-mir-308	74	59,45945946	-32,3	-32,3	0,25533	2,19	-43,64864865	-0,734090909

pmc-mir-3084a	99	38,38383838	-27,5	-22,4	0,0887203	12,04	-27,77777778	-0,723684211
pmc-mir-31	100	40	-31,6	-26,5	0,0259012	18,56	-31,6	-0,79
pmc-mir-315	88	34,09090909	-18,6	-18,6	0,0182896	11,85	-21,13636364	-0,62
pmc-mir-317	94	40,42553191	-40,1	-40,1	0,224017	2,93	-42,65957447	-1,055263158
pmc-mir-33	83	43,37349398	-38,7	-37,7	0,204831	5,09	-46,62650602	-1,075
pmc-mir-336	95	37,89473684	-18,9	-14,5	0,0431525	12,58	-19,89473684	-0,525
pmc-mir-33b	72	45,83333333	-23,6	-23,6	0,187831	6,78	-32,77777778	-0,715151515
pmc-mir-345	98	57,14285714	-28,8	-28,8	0,301092	13,36	-29,3877551	-0,514285714
pmc-mir-3529	91	42,85714286	-30,8	-29,7	0,103281	5,67	-33,84615385	-0,78974359
pmc-mir-3532	99	56,56565657	-40,6	-30,6	0,0366184	11,77	-41,01010101	-0,725
pmc-mir-355	98	31,63265306	-22,1	-22,1	0,0174252	13,25	-22,55102041	-0,712903226
pmc-mir-36	93	41,93548387	-25,6	-18,64	0,0514271	15,9	-27,52688172	-0,656410256
pmc-mir-3620	74	51,35135135	-27,3	-24,8	0,167443	4,41	-36,89189189	-0,718421053
pmc-mir-3643	89	47,19101124	-21,4	-13,5	0,040157	24,56	-24,04494382	-0,50952381
pmc-mir-36b	70	55,71428571	-30,6	-27,6	0,171958	5,87	-43,71428571	-0,784615385
pmc-mir-3724	80	46,25	-20,3	-19,1	0,192073	5,28	-25,375	-0,548648649
pmc-mir-375-1	97	55,67010309	-30,9	-30,9	0,454586	4,38	-31,8556701	-0,572222222
pmc-mir-375-2	70	45,71428571	-20,3	-20,3	0,298895	4,19	-29	-0,634375
pmc-mir-3782	66	62,12121212	-27,1	-26,3	0,354302	7,68	-41,06060606	-0,66097561
pmc-mir-3792	66	31,81818182	-19,9	-17,6	0,1503	5,43	-30,15151515	-0,947619048
pmc-mir-3821	93	31,1827957	-28,6	-28,6	0,0721677	9,75	-30,75268817	-0,986206897
pmc-mir-4009c	73	38,35616438	-20,8	-20,7	0,16755	3,74	-28,49315068	-0,742857143
pmc-mir-4013b	99	36,36363636	-30,2	-30,2	0,107048	16,44	-30,50505051	-0,838888889
pmc-mir-4057	79	62,02531646	-33,5	-28,4	0,21803	20,3	-42,40506329	-0,683673469
pmc-mir-4070	81	43,20987654	-27,1	-20,9	0,0822541	16,72	-33,45679012	-0,774285714
pmc-mir-4133	96	30,20833333	-26,4	-26,4	0,0769195	7,8	-27,5	-0,910344828
pmc-mir-4140	88	31,81818182	-18,9	-18,9	0,163592	6,36	-21,47727273	-0,675
pmc-mir-4177	87	27,5862069	-20,5	-19,4	0,131475	8,3	-23,56321839	-0,854166667

pmc-mir-4185	**99**	**35,35353535**	**-24,4**	**-20,7**	**0,0405048**	**9,56**	**-24,64646465**	**-0,697142857**
pmc-mir-4323	**78**	**53,84615385**	**-26,3**	**-25**	**0,219804**	**12,73**	**-33,71794872**	**-0,626190476**
pmc-mir-4472	**73**	**47,94520548**	**-25,5**	**-22,6**	**0,0746092**	**17,51**	**-34,93150685**	**-0,728571429**
pmc-mir-449d	**97**	**43,29896907**	**-25,6**	**-22,8**	**0,0389626**	**9,81**	**-26,39175258**	**-0,60952381**
pmc-mir-4629	**94**	**59,57446809**	**-30,5**	**-25,9**	**0,0163183**	**28,06**	**-32,44680851**	**-0,544642857**
pmc-mir-4654	**99**	**61,61616162**	**-41,7**	**-40,2**	**0,172557**	**4,86**	**-42,12121212**	**-0,683606557**
pmc-mir-466-1	**95**	**21,05263158**	**-24,1**	**-24,1**	**0,0352645**	**12,2**	**-25,36842105**	**-1,205**
pmc-mir-466-2	**99**	**28,28282828**	**-19**	**-10,61**	**0,0240587**	**19,5**	**-19,19191919**	**-0,678571429**
pmc-mir-466-3	**97**	**20,6185567**	**-29,3**	**-29,3**	**0,326455**	**3,38**	**-30,20618557**	**-1,465**
pmc-mir-466h	**79**	**29,11392405**	**-29,5**	**-29,5**	**0,500936**	**3,9**	**-37,34177215**	**-1,282608696**
pmc-mir-466i-1	**86**	**25,58139535**	**-30,8**	**-24,1**	**0,0996183**	**12,29**	**-35,81395349**	**-1,4**
pmc-mir-466i-2	**94**	**21,27659574**	**-20,1**	**-12,9**	**0,0143588**	**21,92**	**-21,38297872**	**-1,005**
pmc-mir-466m	**97**	**25,77319588**	**-27,1**	**-26,2**	**0,171386**	**21,7**	**-27,93814433**	**-1,084**
pmc-mir-466n	**97**	**22,68041237**	**-28,3**	**-28,2**	**0,0494123**	**16,07**	**-29,17525773**	**-1,286363636**
pmc-mir-466q	**100**	**28**	**-33,1**	**-14,84**	**0,0138479**	**33,92**	**-33,1**	**-1,182142857**
pmc-mir-467d	**85**	**37,64705882**	**-25,9**	**-23,2**	**0,0947691**	**13,72**	**-30,47058824**	**-0,809375**
pmc-mir-467f	**82**	**37,80487805**	**-30**	**-30**	**0,050335**	**8,93**	**-36,58536585**	**-0,967741935**
pmc-mir-4715	**84**	**60,71428571**	**-31,6**	**-31,6**	**0,33988**	**8,09**	**-37,61904762**	**-0,619607843**
pmc-mir-4750	**98**	**58,16326531**	**-29,8**	**-19,7**	**0,0669624**	**29,71**	**-30,40816327**	**-0,522807018**
pmc-mir-4757	**93**	**37,6344086**	**-22,4**	**-21,8**	**0,139005**	**15,42**	**-24,08602151**	**-0,64**
pmc-mir-4769	**85**	**54,11764706**	**-34,6**	**-34,5**	**0,204288**	**8,13**	**-40,70588235**	**-0,752173913**
pmc-mir-487a	**80**	**51,25**	**-22,7**	**-22,7**	**0,227369**	**6,98**	**-28,375**	**-0,553658537**
pmc-mir-49	**88**	**32,95454545**	**-26**	**-26**	**0,306727**	**8,11**	**-29,54545455**	**-0,896551724**
pmc-mir-4938	**81**	**22,22222222**	**-23,1**	**-20,9**	**0,193135**	**5,48**	**-28,51851852**	**-1,283333333**
pmc-mir-4949	**89**	**23,59550562**	**-28**	**-28**	**0,194482**	**3,14**	**-31,46067416**	**-1,333333333**
pmc-mir-4968-1	**74**	**31,08108108**	**-35,3**	**-30,4**	**0,160289**	**15,99**	**-47,7027027**	**-1,534782609**
pmc-mir-4968-2	**90**	**41,11111111**	**-33,6**	**-22,2**	**0,144928**	**16,86**	**-37,33333333**	**-0,908108108**
pmc-mir-4968-3	**95**	**31,57894737**	**-24,8**	**-24,1**	**0,103418**	**14,12**	**-26,10526316**	**-0,826666667**

pmc-mir-4968-4	**98**	**40,81632653**	**-38,8**	**-38,8**	**0,480796**	**14,61**	**-39,59183673**	**-0,97**
pmc-mir-4999	**88**	**37,5**	**-20,3**	**-20,2**	**0,0980001**	**21,47**	**-23,06818182**	**-0,615151515**
pmc-mir-501	**100**	**50**	**-28,4**	**-28,4**	**0,207312**	**9,5**	**-28,4**	**-0,568**
pmc-mir-5101-1	**100**	**22**	**-26,2**	**-19,3**	**0,014197**	**18,79**	**-26,2**	**-1,190909091**
pmc-mir-5101-2	**93**	**24,7311828**	**-26,8**	**-26,2**	**0,0651922**	**10,31**	**-28,8172043**	**-1,165217391**
pmc-mir-519f	**97**	**38,1443299**	**-23,4**	**-22,1**	**0,126706**	**9,47**	**-24,12371134**	**-0,632432432**
pmc-mir-5317a	**93**	**60,21505376**	**-33,8**	**-33,8**	**0,197453**	**11,33**	**-36,34408602**	**-0,603571429**
pmc-mir-539	**92**	**36,95652174**	**-20,4**	**-20,2**	**0,064375**	**14,18**	**-22,17391304**	**-0,6**
pmc-mir-5391	**100**	**34**	**-18,8**	**-12,3**	**0,00560855**	**35,95**	**-18,8**	**-0,552941176**
pmc-mir-5396b	**94**	**57,44680851**	**-34,1**	**-26,1**	**0,0205996**	**19,14**	**-36,27659574**	**-0,631481481**
pmc-mir-5549	**93**	**25,80645161**	**-19,5**	**-19,3**	**0,0919819**	**11,56**	**-20,96774194**	**-0,8125**
pmc-mir-5552	**85**	**24,70588235**	**-23**	**-20,4**	**0,0657061**	**32,41**	**-27,05882353**	**-1,095238095**
pmc-mir-5595	**100**	**46**	**-30,6**	**-30,6**	**0,243502**	**5,31**	**-30,6**	**-0,665217391**
pmc-mir-5600	**100**	**33**	**-24,6**	**-24,6**	**0,137757**	**6,7**	**-24,6**	**-0,745454545**
pmc-mir-5612	**91**	**37,36263736**	**-20,8**	**-15,6**	**0,0415629**	**12,19**	**-22,85714286**	**-0,611764706**
pmc-mir-574-1	**99**	**35,35353535**	**-26**	**-18,3**	**0,0198616**	**27,93**	**-26,26262626**	**-0,742857143**
pmc-mir-574-2	**97**	**27,83505155**	**-27,6**	**-27,5**	**0,0326496**	**10,86**	**-28,45360825**	**-1,022222222**
pmc-mir-5918b	**100**	**25**	**-22,4**	**-14**	**0,0952213**	**25,67**	**-22,4**	**-0,896**
pmc-mir-5965	**94**	**43,61702128**	**-24,6**	**-23,9**	**0,0681876**	**8,99**	**-26,17021277**	**-0,6**
pmc-mir-6056	**99**	**43,43434343**	**-50,2**	**-46,8**	**0,12691**	**12,98**	**-50,70707071**	**-1,16744186**
pmc-mir-6076	**97**	**52,57731959**	**-33,4**	**-33,4**	**0,461099**	**5,24**	**-34,43298969**	**-0,654901961**
pmc-mir-6098	**84**	**41,66666667**	**-22**	**-19,4**	**0,0207902**	**16,09**	**-26,19047619**	**-0,628571429**
pmc-mir-623	**88**	**53,40909091**	**-29,8**	**-29,8**	**0,153312**	**7,64**	**-33,86363636**	**-0,634042553**
pmc-mir-626	**96**	**25**	**-22,1**	**-10,23**	**0,0392647**	**24,65**	**-23,02083333**	**-0,920833333**
pmc-mir-6302	**99**	**34,34343434**	**-18,9**	**-17,1**	**0,135467**	**11,6**	**-19,09090909**	**-0,555882353**
pmc-mir-6416	**95**	**43,15789474**	**-21,2**	**-20,6**	**0,0525607**	**13,18**	**-22,31578947**	**-0,517073171**
pmc-mir-6505	**94**	**36,17021277**	**-21,1**	**-19,17**	**0,0121389**	**12,86**	**-22,44680851**	**-0,620588235**
pmc-mir-651	**100**	**29**	**-21,6**	**-21,5**	**0,14346**	**4,59**	**-21,6**	**-0,744827586**

pmc-mir-6516	**82**	**41,46341463**	**-19,7**	**-19,6**	**0,151242**	**5,16**	**-24,02439024**	**-0,579411765**
pmc-mir-6528	**97**	**42,26804124**	**-32,8**	**-27**	**0,0291552**	**15,05**	**-33,81443299**	**-0,8**
pmc-mir-6548	**74**	**55,40540541**	**-31**	**-31**	**0,103929**	**5,12**	**-41,89189189**	**-0,756097561**
pmc-mir-6560	**69**	**44,92753623**	**-19,8**	**-18,8**	**0,0706231**	**8,29**	**-28,69565217**	**-0,638709677**
pmc-mir-6562	**100**	**40**	**-31,7**	**-31,3**	**0,0488026**	**12**	**-31,7**	**-0,7925**
pmc-mir-6568	**77**	**66,23376623**	**-27,4**	**-27**	**0,220859**	**16,73**	**-35,58441558**	**-0,537254902**
pmc-mir-669f	**73**	**24,65753425**	**-25,6**	**-20,3**	**0,138486**	**10,86**	**-35,06849315**	**-1,422222222**
pmc-mir-669j	**82**	**31,70731707**	**-18,5**	**-17,6**	**0,121983**	**11,36**	**-22,56097561**	**-0,711538462**
pmc-mir-67	**96**	**51,04166667**	**-50,4**	**-50,4**	**0,287211**	**2,51**	**-52,5**	**-1,028571429**
pmc-mir-6701	**97**	**35,05154639**	**-19,2**	**-18,9**	**0,066024**	**11,85**	**-19,79381443**	**-0,564705882**
pmc-mir-6876	**95**	**33,68421053**	**-22,5**	**-22**	**0,114242**	**8,28**	**-23,68421053**	**-0,703125**
pmc-mir-6891	**78**	**33,33333333**	**-24,9**	**-24,9**	**0,700959**	**2,36**	**-31,92307692**	**-0,957692308**
pmc-mir-6971	**70**	**55,71428571**	**-24,5**	**-23,2**	**0,750631**	**1,04**	**-35**	**-0,628205128**
pmc-mir-6974	**94**	**59,57446809**	**-35,8**	**-34,3**	**0,187738**	**13,56**	**-38,08510638**	**-0,639285714**
pmc-mir-7000	**87**	**43,67816092**	**-28,9**	**-28,6**	**0,111346**	**9,33**	**-33,2183908**	**-0,760526316**
pmc-mir-7046	**97**	**53,60824742**	**-32,6**	**-28**	**0,0594584**	**25,01**	**-33,60824742**	**-0,626923077**
pmc-mir-71	**77**	**45,45454545**	**-24,6**	**-24,6**	**0,0775404**	**10,03**	**-31,94805195**	**-0,702857143**
pmc-mir-7-1	**95**	**37,89473684**	**-23,3**	**-21,3**	**0,0852991**	**10,39**	**-24,52631579**	**-0,647222222**
pmc-mir-7151	**91**	**36,26373626**	**-37,2**	**-36,5**	**0,178864**	**4,62**	**-40,87912088**	**-1,127272727**
pmc-mir-7-2	**98**	**32,65306122**	**-20,7**	**-16,47**	**0,00683777**	**17,31**	**-21,12244898**	**-0,646875**
pmc-mir-7207	**95**	**29,47368421**	**-20,6**	**-19,9**	**0,070254**	**20,54**	**-21,68421053**	**-0,735714286**
pmc-mir-722	**90**	**33,33333333**	**-20,7**	**-19,6**	**0,0614938**	**7,79**	**-23**	**-0,69**
pmc-mir-7241	**93**	**41,93548387**	**-20,3**	**-17,7**	**0,0293226**	**17,26**	**-21,82795699**	**-0,520512821**
pmc-mir-7314	**99**	**50,50505051**	**-32,6**	**-28,7**	**0,0105412**	**12,05**	**-32,92929293**	**-0,652**
pmc-mir-7323	**88**	**36,36363636**	**-20**	**-20**	**0,0493021**	**14,06**	**-22,72727273**	**-0,625**
pmc-mir-7386e	**98**	**36,73469388**	**-24,7**	**-24,1**	**0,0259444**	**13,11**	**-25,20408163**	**-0,686111111**
pmc-mir-7388c	**80**	**53,75**	**-24,4**	**-24,4**	**0,0842249**	**8,96**	**-30,5**	**-0,56744186**
pmc-mir-7398a	**96**	**36,45833333**	**-21,1**	**-15,38**	**0,110809**	**24,91**	**-21,97916667**	**-0,602857143**

pmc-mir-743b	**79**	**48,10126582**	**-26,2**	**-25,6**	**0,0306733**	**10,47**	**-33,16455696**	**-0,689473684**
pmc-mir-7448	**65**	**40**	**-20,4**	**-19,3**	**0,340793**	**2,64**	**-31,38461538**	**-0,784615385**
pmc-mir-745a	**91**	**53,84615385**	**-46,9**	**-46,9**	**0,0976825**	**4,24**	**-51,53846154**	**-0,957142857**
pmc-mir-745b	**70**	**57,14285714**	**-29**	**-25,7**	**0,0969948**	**8,37**	**-41,42857143**	**-0,725**
pmc-mir-7472	**97**	**49,48453608**	**-30,5**	**-27,9**	**0,0508509**	**8,11**	**-31,44329897**	**-0,635416667**
pmc-mir-750	**97**	**50,51546392**	**-52,7**	**-51,6**	**0,157091**	**3,48**	**-54,32989691**	**-1,075510204**
pmc-mir-751	**94**	**38,29787234**	**-27,8**	**-22,8**	**0,0515578**	**21,54**	**-29,57446809**	**-0,772222222**
pmc-mir-7562	**95**	**28,42105263**	**-20,2**	**-20,2**	**0,122747**	**10,51**	**-21,26315789**	**-0,748148148**
pmc-mir-7575	**75**	**36**	**-19,1**	**-16,7**	**0,188578**	**11,02**	**-25,46666667**	**-0,707407407**
pmc-mir-7643	**97**	**28,86597938**	**-20,4**	**-15,3**	**0,0255881**	**14,82**	**-21,03092784**	**-0,728571429**
pmc-mir-785a	**99**	**21,21212121**	**-19,4**	**-19,4**	**0,222469**	**10,6**	**-19,5959596**	**-0,923809524**
pmc-mir-7880b	**95**	**21,05263158**	**-27,3**	**-24,5**	**0,144234**	**12,84**	**-28,73684211**	**-1,365**
pmc-mir-7880q	**100**	**28**	**-18,6**	**-17**	**0,0486767**	**12,83**	**-18,6**	**-0,664285714**
pmc-mir-79	**95**	**28,42105263**	**-18,8**	**-18,8**	**0,0970813**	**11,62**	**-19,78947368**	**-0,696296296**
pmc-mir-8	**94**	**45,74468085**	**-46**	**-46**	**0,192059**	**3,37**	**-48,93617021**	**-1,069767442**
pmc-mir-8250d	**97**	**45,36082474**	**-26,2**	**-23,38**	**0,012202**	**14,09**	**-27,01030928**	**-0,595454545**
pmc-mir-8265	**96**	**46,875**	**-25,5**	**-25,5**	**0,101749**	**8,96**	**-26,5625**	**-0,566666667**
pmc-mir-8293	**100**	**32**	**-21,5**	**-13,1**	**0,0304044**	**22,6**	**-21,5**	**-0,671875**
pmc-mir-8311	**99**	**42,42424242**	**-28,4**	**-28,4**	**0,262782**	**6,39**	**-28,68686869**	**-0,676190476**
pmc-mir-8335	**96**	**22,91666667**	**-24,4**	**-20,9**	**0,125649**	**19,36**	**-25,41666667**	**-1,109090909**
pmc-mir-8398	**97**	**20,6185567**	**-18,6**	**-15,1**	**0,018215**	**22,4**	**-19,17525773**	**-0,93**
pmc-mir-8422	**96**	**25**	**-23,6**	**-23,1**	**0,141217**	**4,11**	**-24,58333333**	**-0,983333333**
pmc-mir-8443	**92**	**35,86956522**	**-18,7**	**-18,6**	**0,0764442**	**11,42**	**-20,32608696**	**-0,566666667**
pmc-mir-8460	**89**	**35,95505618**	**-23,7**	**-20,9**	**0,049028**	**10,23**	**-26,62921348**	**-0,740625**

AMFE: Energia Mínima Livre ajustada, MFE: Energia mínima livre, MFEI: Índice de energia mínima livre, MFEE: Energia mínima livre do conjunto.

Fonte: Dados do autor

Através de uma abordagem computacional desenvolvida neste trabalho, no genoma e transcriptoma das espécies em estudo, foi possível detectar miRNAs (maduros e precursores).

Foram identificados 296 (*P. canaliculata)* e 316 (*P. maculata)* prováveis miRNAs maduros conservados, semelhantes aos ortólogos das espécies com miRNAs catalogados no miRbase versão 22 (http://www.mirbase.org). O número de miRNAs identificados corrobora com o número de miRNAs conservados referentes ao molusco próximo evolutivamente *Lymnaea stagnalis*, gastrópode de água doce (264 miRNAs maduros) (WALKER *et al*., 2018).

Os miRNA não codificantes de proteínas têm aproximadamente 22 nucleotídeos em sua extensão, variando em média de 17 a 25 nucleotídeos (HA; KIM, 2014; RACHAGANI; KUMAR; BATRA, 2010). Os miRNAs maduros identificados neste trabalho possuem em média 21,86 nucleotídeos (*P. canaliculata)* - Tabela 7, coluna 2 e 21,77 nucleotídeos (*P. maculata)* - Tabela 8, coluna 2; com variação de tamanho entre 17 a 25 nucleotídeos para ambas as espécies, corroborando com dados de espécies animais.

A proteína Ago1 apresenta preferência pelo nucleotídeo uracila na primeira posição da sequência do miRNA na extremidade 5' a ser carregado, para ser inserido no complexo RISC. A proteína Ago 2 também pode ser carregada com o miRNA (uma das fitas do *duplex)*, intermediando a clivagem de alvos de miRNAs (GHILDIYAL *et al*., 2010; SEITZ; TUSHIR; ZAMORE, 2011; ZHANG; STELLWAG; PAN, 2009). Neste trabalho, os miRNAs maduros de *P. canaliculata* e *P. maculata*, respectivamente, apresentaram quantitativo de nucleotídeos na primeira posição de 41,2% e 42,40% de uracila, 20,9% e 17,8% (citosina), 20,4% e 24,5% de adenina e 17,5% e 15,3% de guanina. Os resultados encontrados corroboram os dados da literatura acima descritos.

A sequência madura dos miRNAs tem se mostrado conservada evolutivamente (WIT *et al*., 2009). Esta conservação se acentua na região denominada *seed* (semente), sendo considerada importante para reconhecimento dos alvos de miRNAs (LEWIS; BURGE; BARTEL, 2005). Todos os miRNAs identificados neste trabalho apresentaram 100% de identidade nas regiões *seed* (posição de 2 a 8 nucleotídeos) (LEWIS *et al*., 2003) e no máximo 30% dos nucleotídeos diferentes (*mismathes)* do miRNA maduro, quando comparado com estrutura do seu respectivo ortólogo.

A conservação da região *seed,* segundo Friedman *et al*. (2009) é muito importante evolutivamente para conservação dos alvos de miRNAs silenciados. Como ocorre a correspondência de bases do tipo Watson-Crick, ambos, miRNAs e alvos, necessitam de uma estrutura conservada, apesar de que todas as interações não sejam totalmente complementares (FRIEDMAN *et al*., 2009). Na caracterização dos miRNAs, é possível verificar no alinhamento dos precursores a identificação dos miRNAs, apresentando 100% de identidade nas regiões *seed*. Todos os miRNAs catalogados neste trabalho possuem um ortólogo correspondente no qual a busca foi baseada.

Tabela 7: Prováveis miRNAs maduros conservados identificados em *P. canaliculata,* tamanho dos nucleotídeos (nt), sequência nucleotítica do miRNA maduro, região *seed.*

miRNA	Tamanho (nt)	Sequência maduro (5’a 3’) Região *seed*	miRNA	Tamanho (nt)	Sequência maduro (5’ a 3’) Região *seed*
pcr-miR-bantam-5p	24	CUGGUUUUCACAGUGAUCCAGCAG	pcr-miR-4525-3p	19	GGGGGGAUGUGCAUGCCAG
pcr-miR-bantam-3p	23	UGAGAUCAUUGUAAAAACCAAUU	pcr-miR-4617-3p	22	UGUGGCUGUCACACCUAAACAC
pcr-miR-let-7b-3p	22	UGAGGUAGUGGUUGUAUCGUAU	pcr-miR-4654-5p	23	UGUGGGAUCUGGAGGCAGACUUC
pcr-miR-let-7d-3p	22	CUAUACAACCUGCUGCGGCAGG	pcr-miR-466i-1-3p	20	UGUGUGUGUGUGUGUGUGUG
pcr-miR-let-7f-5p	21	CUGCACAUUCCACCAUCCAUU	pcr-miR-466i-2-5p	20	UGUGUGUGUGUGUGUGUGUG
pcr-miR-1-3p	22	CAUGCUUCUUUAUAGUUUGACC	pcr-miR-466i-3-5p	20	UGUGUGUGUGUGUGUGUGUG
pcr-miR-1002-3p	23	CUAAGUAGUUGAUACAAACAGUA	pcr-miR-466i-4-5p	20	GGUGUGUGUGUGUGUGUGUG
pcr-miR-10a-5p	24	ACCCUGUAGAUCCGAAUUUGUGUA	pcr-miR-466m-3p	23	UGUGUGCAUGUGCAUGUGUGCUU
pcr-miR-11-3p	22	CAUCACAGUCAGAGUUCACUGA	pcr-miR-466n-3p	22	GUGUGUGCGUACAUGUACUCAU
pcr-miR-	23	AGUGGAGAGAGUUUUAUCUCAUC	pcr-miR-	21	UAUGUGUGUGUACAUGUGAAG

1175-5p			466p-3p		
pcr-miR-1175-3p	23	UGAGAUUCAACUCCUCCAACUGC	pcr-miR-466q-3p	19	GUGCACACACACACAUACG
pcr-miR-1187-3p	22	UAUGUGUGUGUGUAUGUGUGUG	pcr-miR-467f-3p	21	AUAUACACACACACACCUUUC
pcr-miR-12-5p	22	UGAGUAUUACAUCAGGUACUGA	pcr-miR-4715-5p	23	GUGCCACCUUAACUGCACGGACC
pcr-miR-12-3p	22	AGUACCUUUUGUGAUAUUCUUA	pcr-miR-4757-3p	22	CAUGACGUCACAGAGGCGAGGU
pcr-miR-122b-5p	20	AGUGUGACACUGGUGUGACA	pcr-miR-4801-3p	17	UACACAAGAAAACCAAA
pcr-miR-1238-5p	20	CUUCCUCGUCUGUCUGAAGC	pcr-miR-4860-3p	24	UGUAGAGAUUGUGUGAACUGCUAG
pcr-miR-124-5p	23	GUGUUCACUGCGUUGGCCUUGGU	pcr-miR-49-3p	22	AAGCACCAUGUGAAGCACUUUU
pcr-miR-124c-5p	21	CAUUCACCGCGUGCCUUAAUA	pcr-miR-4918-5p	24	GUGAGCACGAUGGCCAGCACUGCC
pcr-miR-124-3p	23	UUAAGGCACGCGGUGAAUGCCAA	pcr-miR-4968-1-5p	22	CAGCAACAGCAGCAGCAGCAGA
pcr-miR-1277-1-3p	22	UAUAUAUAUAUAUGUACGUCUA	pcr-miR-4968-2-5p	22	CAGCAACAGCAGCAGCAGCAGC
pcr-miR-1277-2-3p	23	AAAUAUAUAUAUAUAUGUAUGAU	pcr-miR-4968-3-3p	22	CAGCAACAGCAGCAGCAGCAGC
pcr-miR-1322-5p	19	GAUGAUGCUGCUGAUGACA	pcr-miR-4968-4-3p	22	CAGCAACAGCAGCAGCAGCAGC

pcr-miR-1328-5p	18	GAGAGAGAAAUGAGAAAC	pcr-miR-5317a-3p	24	UUUCUUGUCUCUGUGUACCUCUUG
pcr-miR-133-5p	23	AGCUGGUUGAACUCGGGCCAAAU	pcr-miR-5391-5p	20	UGGACAGUUAUGAAGAAGUG
pcr-miR-133-3p	24	UUUGGUCCCCUUCAACCAGCUGUA	pcr-miR-5392-3p	20	UGUUACUUGACAUUUUGAGA
pcr-miR-133c-5p	21	CAGCUGGUUGAAGGGGACCAA	pcr-miR-5440-5p	21	UCAGGAGGACUGAAGGGUGGA
pcr-miR-137b-5p	24	ACGGGUAUUCUUGGGUAAAUAAUA	pcr-miR-5549-3p	19	UCAUGUUGGUUUUUUGAAU
pcr-miR-137b-3p	23	UUAUUGCUUGAGAAUACACGUAA	pcr-miR-559-3p	21	UAAAGUAAAUAAGCACAGACU
pcr-miR-13b-3p	17	UCGUUAAAAUGUUUGUG	pcr-miR-5594-3p	22	AAGAGUACUGUAGUUUUUCCAA
pcr-miR-1421a-3p	22	CUGCAUCCCUUGUGACGGUGGC	pcr-miR-5595-3p	24	UCUCUUUUUUCUCGCAGUCCCGCA
pcr-miR-1421am-3p	22	AGGAGGUCGUAGGUCAUCCUGU	pcr-miR-574-1-5p	24	UGAGUGUGUGUGUGUGAGUGUGUG
pcr-miR-143-3p	23	GUCUGAGAUGAAGCACUCGUUUG	pcr-miR-574-2-3p	24	UGAGUGUGUGUGUGUGAGUGUGUG
pcr-miR-153-1-3p	23	UCAUUUUUGUGAUGUUAUCAAUA	pcr-miR-5965-5p	20	GACAAUUCUGACAUUUUGCA
pcr-miR-153-2-3p	23	UUGCAUAGUCACAAAAGUGAUCG	pcr-miR-6-3p	23	AGGGAACUGCUGCUGCAGCGACU
pcr-miR-	20	CCGGGUGGGGCUGGGCCAUA	pcr-miR-	19	GCUGCUGACGACCGCCUCA

1584-5p			6013-3p		
pcr-miR-15c-3p	22	CAGACCAUUCUGGGCUAUGGGG	pcr-miR-6037-1-3p	21	UAAGCUCUGUGUACUUUAGUG
pcr-miR-1602-5p	22	UGGGCUCUGCAUCACCAGCAGU	pcr-miR-6037-2-3p	21	UAAGCUCUGUGUACUUUAGUG
pcr-miR-1603-3p	20	GUGGUUUGUUUUGUGUUGCG	pcr-miR-6076-3p	21	AGCAUGACAGAGGAGAGGAGA
pcr-miR-1632-3p	23	UGCUUGUUUUUGGAUGAAAUUAU	pcr-miR-6098-5p	20	AUAGUGAUUGUGUGUUUCUA
pcr-miR-1677-3p	23	UUGACUUCAAUAGGAGAGACAGG	pcr-miR-6132-1-5p	19	AGCAGGGCUGGGGAUUCUG
pcr-miR-1744-1-5p	22	ACUUCAACAGGAGCAAGUCAAG	pcr-miR-6132-2-5p	19	AGCAGGGCUGGGGAUUCUG
pcr-miR-1744-2-5p	22	ACUUCAACAGGAGCAAAGUGUA	pcr-miR-6132-3-5p	19	AGCAGGGCUGGGGAUUCUG
pcr-miR-184-5p	22	CCUUGUCACUCCUCCGCCCCGU	pcr-miR-6335-5p	22	CUGCAUACACAGUGAUGCAGGU
pcr-miR-184-3p	24	ACUGGACGGAGAACUGAUAAGGGC	pcr-miR-6416-3p	21	CUCCGUAUCAUCUGCUAGCUA
pcr-miR-190-5p	23	AGAUAUGUUUGAUAUAUUUGGUG	pcr-miR-64c-3p	21	CAUGACACACUGGCUGAACCA
pcr-miR-1905c-3p	25	CACCACCAGCCCCACCACGCGGUAU	pcr-miR-6505-5p	21	UGACUUCUACCUGUUCCCUAA
pcr-miR-1951-3p	22	GUAGUGGAGACUGGUGGCGAAA	pcr-miR-6516-3p	22	AUGCAGUAACAGGUGUCUCCAC

pcr-miR-1976-5p	21	CUCCUGCCCUCCUUGCCCAGA	pcr-miR-6560-5p	23	UGUGUUGCUCCAUACUGCUGAAG
pcr-miR-1984-5p	23	UGCCCUAUCCGUCAGGAACUGUG	pcr-miR-6575-3p	22	GAGUUUUCCCAGGCUGAUGUGU
pcr-miR-1985-5p	24	UGCCAUUUUUAUCAGUCACUGUGU	pcr-miR-6587-3p	23	UGCUGGAAGUGGUGCUAUAUGUC
pcr-miR-1986-3p	22	UGGAUUUCCCAAGAUCCGUGAU	pcr-miR-6603-5p	23	AAGGACAAGAGGAAAUAACUUGA
pcr-miR-1990-5p	22	AGUAAGUUGAUGGGGUCCCAGG	pcr-miR-6680-5p	22	GUUGCUGAUGAAGAUAAAUCGU
pcr-miR-1990-3p	21	CGGGACUACGUCAACGUACUA	pcr-miR-669f-5p	23	CAUAUACAUACACACACACGUGU
pcr-miR-1992-5p	22	AGUCAGUGGUUGACUGCUCGGU	pcr-miR-67-3p	23	UCACAACCUGCAUGAAUGAGGAC
pcr-miR-1992-3p	23	UCAGCAGUUGUACCACUGAUUUG	pcr-miR-6816-3p	18	GAAGGACCUGCACCUUGC
pcr-miR-1994-3p	22	UGAGACAGUGUGUCCUCCCUCU	pcr-miR-6852-5p	17	UGUCCUCUGUUCCUCAU
pcr-miR-1994a-3p	22	UGAGACAGUGUGUCCUCCCUUG	pcr-miR-6894-3p	24	AGGAGGAUGGAGAGCUGUUACAGC
pcr-miR-1996b-3p	19	AUCAAGUGAGGUCAGAAUA	pcr-miR-6951-5p	21	UUGUAUUUGUGUGAUUCCCGA
pcr-miR-1a-5p	24	ACAUACUUCUUUGCUAUCCCAUAU	pcr-miR-6974-3p	22	UCUCCACUCUCUUCUGUGUCGU

pcr-miR-1a-3p	22	UGGAAUGUAAAGAAGUAUGUAC	pcr-miR-6977-5p	25	GUGAGGCGCUGUGGGCAGGUGAGCC
pcr-miR-1c-5p	22	GUACAUACUUCUUUACAUUCCA	pcr-miR-6999-3p	23	AAGGAAGGAGAGUCAGGAGCGUG
pcr-miR-2001-5p	23	UUGUGACCGUUACAAUGGGCAUU	pcr-miR-7000-5p	22	CACCCACCUGCCUGUCACAGUG
pcr-miR-2008-3p	22	GUAUUAACAAUGUGGCUACAGU	pcr-miR-7072-3p	21	CUAGCUUCUCUUCUUCCAGUU
pcr-miR-203b-3p	22	UUGAACUGUUAAGAACAGUAAA	pcr-miR-71-1-5p	23	UGAAAGACAUGGGUAGUGAGAUG
pcr-miR-204-3p	19	UGCAAGGACAGCAAAGGGA	pcr-miR-71-2-3p	22	UGAAAGACACAGGUAGAAUGUA
pcr-miR-2047-3p	22	UUCGCCGAGCAGAUGUGUGAAG	pcr-miR-7151-5p	23	GAUCCAUCUCUGCCUGCUCACCU
pcr-miR-2155-3p	23	GACACUGUUUUACACUCUACUGG	pcr-miR-7307-3p	19	CAAAUGGUCAUUUCUGCGA
pcr-miR-216a-5p	23	UAAUCUCAGCUGGUAAUUCUGAG	pcr-miR-7314-3p	18	UCAGAAACAGUGUCUCUG
pcr-miR-216a-3p	22	CAAGUUACUAGCCGAGAUUACA	pcr-miR-7386e-5p	21	GUAAACACUUUACCCGUUUGA
pcr-miR-216b-1-3p	22	CACAGUGGCUACUGGGUCCACA	pcr-miR-7389-3p	22	GCUGUACUUGAAGCAAGAGAGA
pcr-miR-216b-2-3p	23	UAAUAUCAGCUGGUAAUCCUGAG	pcr-miR-7398h-3p	22	UAACCCUCCCUUCUUUAUGAUA

pcr-miR-219-1-5p	23	UGAUUGUCCAAACGCAAUUCUUG	pcr-miR-7399-5p	22	UACGCAUGUAUAUGUAUAUACA
pcr-miR-219-1-3p	22	AGAACUGUGUGUGGACAUCAGU	pcr-miR-743b-3p	20	UGUUCAGACUGGUGUCUGGU
pcr-miR-219-2-5p	23	UGAUUGUCCAAACGCAAUUCUUG	pcr-miR-745a-3p	23	AGCUGCCUGAUGAAGAGCUGUCC
pcr-miR-219-2-3p	22	AGAACUGUGUGUGGACAUCAGU	pcr-miR-745b-3p	22	GAGCUGCCAAAUGAAGGGCUGU
pcr-miR-2196-5p	24	CCUCUCUGUGCUGCCAGCAAAAAU	pcr-miR-7472-3p	22	AUGUGCUGGAGAUGAAAACUCU
pcr-miR-2218b-3p	22	AAAUUUGUAGUUUGUAUUCUUC	pcr-miR-750-5p	23	AGUUGGAAGAUUGGGUCUUUGGC
pcr-miR-2284c-3p	22	AAAAUCUAAGUGAACUGUGACG	pcr-miR-750-3p	23	CCAGAUCUAACUCUUCCAGCUCA
pcr-miR-2284t-3p	22	AAACUCGAAUGAAUGUAGUUCG	pcr-miR-751-3p	21	CAUGUUUGAAUGGCCACCACU
pcr-miR-2324-5p	23	UGGGGUUGGGGCAGUGACCAGCA	pcr-miR-753d-5p	21	AGAUCAUUAUUCAAGCGAUUG
pcr-miR-2392-3p	22	AUGGAUGGGGGUGAGGGGGAUG	pcr-miR-760-3p	22	CGGCUCUGGGUCUGUGAAGUCA
pcr-miR-2405-3p	18	UGUGUGGUUUGUUUGGUU	pcr-miR-7643-3p	22	GACAUCUGUUUCUCAGUAAUUU
pcr-miR-2464-3p	24	ACUUCUGCUGCAGACCUCAAGGAU	pcr-miR-7649-5p	22	UGACAGCAGUGCUGUAUCUAUG
pcr-miR-	21	UUGUGUGGUUUUGGAUACUUG	pcr-miR-	22	UCUUGUUUUUUUCAACUCCUGG

2470-3p			76b-1-3p		
pcr-miR-2491-1-3p	18	CAACAACAGCAGCAGCAA	pcr-miR-76b-2-3p	22	UCUUGUUUUUUUCAACUGCGUU
pcr-miR-2491-2-3p	18	CAACAACAGCAGCAGCAA	pcr-miR-770-3p	22	AGCACCACGUGUCUGGUAAAUC
pcr-miR-2491-3-5p	18	CAACAACAGCAGCAGCAG	pcr-miR-7880k-3p	26	GGUCAGAAAUGGUUGCUGAGCUCCAC
pcr-miR-2500-3p	17	GGAUUUUGUGUGUGGAC	pcr-miR-7880q-3p	22	UUUGCGACCAAAAUCACUCUCU
pcr-miR-252a-5p	22	CUAAGUACUAGUGCCGCGGGAA	pcr-miR-79-3p	20	GUAAAGCUAAAUUACCAGUG
pcr-miR-254-5p	23	UGCAAAUCUUUUGCAACAGACUU	pcr-miR-7901-3p	21	UACUGUAGUUGAAGCGCUAUU
pcr-miR-279-1-3p	22	UGACUAGAUCCACACUCAUCCA	pcr-miR-7c-5p	23	UGGAAGACUAGUGAUUUUGUUGU
pcr-miR-279-2-3p	22	UGACUAGAUCCACACUCAUCCA	pcr-miR-7c-3p	25	UGGAAGACUAGUGAUUUUGUUGUUC
pcr-miR-279b-5p	22	UGACUAGACUGAUAACCCCUAG	pcr-miR-8-1-5p	22	CAUCUUACCUAACAGCAUUAGA
pcr-miR-2804-3p	22	GUAGUGUAUUACAAUACUGACC	pcr-miR-8-1-3p	23	UAAUACUGUCAGGUAAAGAUGUC
pcr-miR-281-5p	21	AAGGGAGCAUCCGUCGACAGU	pcr-miR-8196b-3p	17	AGUAGUUAAUUUUUCUU
pcr-miR-281-3p	22	UGUCAUGGAGUUGCUCUCUUUA	pcr-miR-8-2-5p	22	CAUCUUACCUAACAGCAUUAGA

pcr-miR-2962-3p	21	CAUCCCCUGCCUCUCUCAUAC	pcr-miR-8-2-3p	22	UAAUACUGUCAGGUAAAGAUGUC
pcr-miR-297-3p	23	AUGUAUGUGUGCAUGUAUGGAGC	pcr-miR-8296-5p	22	UCGAGAUGAUGGAGAGCACACU
pcr-miR-29a-3p	22	UAGCACCAUUUGAAAUCAGUUU	pcr-miR-8332-3p	23	UUCUUUGCUCUUCUCGACAUGCG
pcr-miR-29b-3p	23	UAGCACCAUUUGAAAUCAGUGCU	pcr-miR-8335-1-5p	25	GUUGUUGUUGUUGUUUUGCCAACUG
pcr-miR-2a-1-3p	24	AAUCACAGCCUGCUUUGGUGAGCU			GUUGUUGUUGUUGUUUUGGUUUUUU
pcr-miR-2a-2-3p	24	UAUCACAGCCUGCUUGGAUCAGUA	pcr-miR-8335-2-5p	25	AGAGAGUGUGGAAAAGGGGAGAG
pcr-miR-2a-3-3p	24	UAUCACAGCCAGCUUUGAUGAGCU	pcr-miR-8364g-3p	23	UGAUUGGUCACAUAGUUUA
pcr-miR-2a-4-3p	24	UAUCACAGCCAGCUUUGAUGAGCU	pcr-miR-8416-5p	19	GCAAUUAAAAGUAGCACAA
pcr-miR-2b-1-5p	24	UCAUCAAAGCUGGCUGUGAUAUGA	pcr-miR-8422-5p	19	GAAUGAUCAAAUAAUUGUC
pcr-miR-2b-2-5p	24	UCAUCAAAGCUGGCUGUGAUAUGA	pcr-miR-8462-1-3p	19	UGAUUAAUUUGAUAAGAUGA
pcr-miR-2e-3p	21	UACCAACUUUGACUGAUGAGA	pcr-miR-8462-2-3p	20	CACACACACACACACACGUAU
pcr-miR-2f-3p	23	UAUCACAGCCAUGCUAAUCUCCU	pcr-miR-8485-1-3p	21	CACACACACACACACACGUAC
pcr-miR-	22	CCAGUCUUGCAUUUAUGAAACA	pcr-miR-	21	CUGCCUGAAAUUUAUGCUCAAACCU

3027-5p			8485-2-5p		
pcr-miR-3050-5p	22	UGAGAUCUUGAUAAACAUCAAC	pcr-miR-87-5p	25	UCGAGAUGAUGGAGAGCACACU
pcr-miR-3057-3p	22	UCCCACAGGCCCAGCUGUCACA	pcr-miR-87-3p	22	GUGAGCAAAGUUUCAGGUGUGU
pcr-miR-3071-5p	22	AUCAUCAAAACAAAUGUGCAUA	pcr-miR-8834a-5p	23	UGCCGGGCCUGGAGGCCAGCUGU
pcr-miR-308-5p	22	CGCAGGAUUUGCUUGUUUUGAG	pcr-miR-888-5p	22	GACUGACACCUCUUUGUCCAGC
pcr-miR-3084a-5p	22	UUGAAGGUUCAUUAGCCUUGUG	pcr-miR-8915-1-3p	21	CGGAUGGCCAGCUGCAGGUGA
pcr-miR-31-5p	23	AGGCAAGAUGUUGGCAUAGCUGA	pcr-miR-8915-2-3p	21	CGGAUGGCCAGCUGCAGGUGA
pcr-miR-31-3p	22	AGCUGUGUCGCAUGGUGCCACC	pcr-miR-8915-3-3p	23	CGGAUGGCCAGCUGCAGGUGACG
pcr-miR-315-5p	23	UUUUGAUUGUUGCUCAGAAAGCC	pcr-miR-8915-4-3p	23	CGGAUGGCCAGCUGCAGGUGACG
pcr-miR-317-1-3p	25	UGAACACAGCUGGUGGUAUCUUUUU	pcr-miR-8915-5-3p	23	CGGAUGGCCAGCUGCAGGUGACA
pcr-miR-317-2-5p	22	AGUGAACACAGCUGGUCACAAC	pcr-miR-9015-5p	23	UCCCAGAGGAGACGAGAAAGUUG
pcr-miR-3173-3p	23	GAAGGAGGAAACAGGCUACAGAU	pcr-miR-9-1-5p	23	CCUUUGGUAACCUAGCUUUAUGA
pcr-miR-33-5p	22	GUGCAUUGAGGUUGCAUUGCAU	pcr-miR-9-1-3p	23	UCAUACAGCUAGAUAACCAAAGA

pcr-miR-33-3p	23	GCAAUGCAUCUGCAGUGCAAAUA	pcr-miR-9120-3p	21	UCAGUCUCCCUCUCUCGCUAU
pcr-miR-340-5p	22	UUAUAAAGCAAUGAGAGUUUCC	pcr-miR-9-2-5p	23	CCUUUGGUAACCUAGCUUUAUGA
pcr-miR-345-3p	21	UGCUGACCCCUAGUCCGCCUC	pcr-miR-9-2-3p	23	UCAUACAGCUAGAUAACCAAAGA
pcr-miR-3529-3p	24	AACAACAAAAUCACUAGUCUUCCA	pcr-miR-92a-1-3p	22	GAUUGCACUUGUCCCGGCCUUG
pcr-miR-3547-3p	25	GUGGGAAGAGGGGUGGAGGUGAAGC	pcr-miR-92a-2-3p	23	GAUUGCACUUGUCCCGGCCUUCU
pcr-miR-355-5p	21	UUUGUUUUAGCCUGAGUCGCC	pcr-miR-92b-3p	21	UAUUGCACUCGUCCCGGCCUA
pcr-miR-3609-5p	24	CAAAGUGAUGAGUAAUUAAGGCGC	pcr-miR-92c-5p	24	AGGUUGGGAUGUGGGCUCUGUCGC
pcr-miR-3680-5p	22	GACUCACUCACAGGAUGUGGUG	pcr-miR-9-3-5p	23	CCUUUGGUAACCUAGCUUUAUGA
pcr-miR-36b-3p	23	UCACCGGGUAUACAUUCAUCCGC	pcr-miR-9-3-3p	23	UCAUACAGCUAGAUAACCAAAGA
pcr-miR-3716b-3p	23	CGAUGUUAUUUGAAAUAUCAAAC	pcr-miR-9341-1-3p	21	UCUCGGCCUUUUGGCUAAGAU
pcr-miR-3739-3p	22	UUGGGAGGGGGAGAGAGUGGAA	pcr-miR-9341-2-3p	21	UCUCGGCCUUUUGGCUAAGAU
pcr-miR-375-1-3p	22	UUUGUUCGUUCGGCUCGCGUUA	pcr-miR-9388-5p	23	GUAUGUAUGUAUGUACAUAUAAU
pcr-miR-375-2-3p	20	CUUGUUCGUUCGGCUCGUUC	pcr-miR-9-4-5p	25	UCUUUGGUUAUCUAGCUGUAUGAUU

pcr-miR-377-5p	22	AGAGGUUGCCUUUGGUGAGAGA	pcr-miR-9-4-3p	23	AUAAAGCUAGGUUACCAAAGGCA
pcr-miR-3776-3p	22	AGGAGGGGGGAGAGAGAUUUA	pcr-miR-9543a-3p	18	AAAUAUUGCUUGUUUAUU
pcr-miR-3809-5p	20	GUCGGUCAGUUGGUUAGAUG	pcr-miR-96-5p	22	CAAUCAUGUGCAGUGCAGUUCU
pcr-miR-3831-3p	26	CAAACAUUGCUAUUUUUCAUAUCCAA	pcr-miR-96b-3p	22	AUUAUACACCGGUGCCAAGUCA
pcr-miR-3928-3p	23	UGAAGCUCUAAGGUUCAUCCCAG	pcr-miR-969-3p	21	GAGUUCCACUAAGCAAGUUAU
pcr-miR-3957-3p	22	ACGCACAGCACCUCACACUGAC	pcr-miR-96b-5p	23	AUUUGGCACUUGUGGAAUAAUCG
pcr-miR-3965-5p	19	UGCUUAUCAGCCUGAUUCU	pcr-miR-972-5p	21	UAAAUAUUUUUUUUGUCAACU
pcr-miR-4057-3p	20	GGCUUGGCGACUGUAGUAGG	pcr-miR-981-5p	22	CGGGUUUCGCGGCUUGCGAACA
pcr-miR-4066-5p	19	CAUGCCUUUAUCAUGUGUU	pcr-miR-981-3p	23	UUCGUUGUCGUCGAAACCUGCCC
pcr-miR-4070-3p	19	CACGCGUCACGUGACACCU	pcr-miR-9891-3p	21	UCGGCUUCGUCCUCGUCUUGG
pcr-miR-4185-3p	22	UGUAUUCAUACUGUCUGUUACA	pcr-miR-995-3p	23	GCCCGGAUACUGUGUGUGGCUCA
pcr-miR-4187-3p	20	UUUGGUGUUGUGCUGUUUAA			
pcr-miR-	20	UACAUGUAAUACAUUGUACC			

4195-5p		
pcr-miR-4271-3p	**19**	**GGGGGAAGAAAAGGUGGCA**
pcr-miR-4323-3p	**18**	**CAGCCCCACAGCCUCAGG**
pcr-miR-4472-3p	**18**	**GGUGGGGGGUGUUGUUGA**
pcr-miR-449a-3p	**22**	**ACCAGCUGACAUUCAGUCCAGA**
pcr-miR-449d-3p	**20**	**GAAGGCUGUGUGCUGUAAAG**

Fonte: Dados do autor

Tabela 8: Prováveis miRNAs maduros conservados identificados em *P. maculata,* tamanho dos nucleotídeos (nt), sequência nucleotídica do miRNA maduro, região *seed* em destaque.

miRNA	**Tamanho (nt)**	**Sequência maduro (5'a 3')** **Região *seed***	**miRNA**	**Tamanho (nt)**	**Sequência maduro (5' a 3')** **Região *seed***
pmc-bantam-3p	**23**	**UGAGAUCAUUGUAAAAACCAAUU**	**pmc-miR-3724-3p**	**22**	**AUCGUGGUAAACUUUGUCCUCG**
pmc-bantam-5p	**24**	**CUGGUUUUCACAGUGAUCCAGCAG**	**pmc-miR-375-1-3p**	**22**	**UUUGUUCGUUCGGCUCGCGUUA**
pmc-miR-1000-5p	**22**	**CUGGUGUGAUGGGACAAGACUU**	**pmc-miR-375-2-3p**	**20**	**CUUGUUCGUUCGGCUCGUUC**
pmc-miR-10031c-5p	**22**	**CUCGUCCAUGUCUUCCUCUUCG**	**pmc-miR-3782-3p**	**24**	**CUUACAGAGGCACUUGGGUGGCGC**
pmc-miR-	**19**	**ACUUGUCGCAGUGACGACA**	**pmc-miR-**	**21**	**GACUGAGGAAAGAGAGAUGUU**

10046-3p			**3792-3p**		
pmc-miR-10055-3p	**21**	**GUGGGAGGGAGGGAGGACUUU**	**pmc-miR-3821-3p**	**23**	**AUUUCUAGUGAGUGAGGGUACUU**
pmc-miR-10089-5p	**22**	**UAAUUUGCUGUUUUGGUGAUAU**	**pmc-miR-4009c-3p**	**20**	**UAUUGCACUUUUACUGGUCG**
pmc-miR-10173-3p	**22**	**AGGAAGUGAGGAAGGAAAACAA**	**pmc-miR-4013b-5p**	**20**	**UUACUUGCUUUAACAGCUUA**
pmc-miR-10249-1-3p	**22**	**GUGGUGGCGGUGGUGUUGCCAG**	**pmc-miR-4057-3p**	**20**	**GGCUUGGCGACUGUAGUAGG**
pmc-miR-10249-2-3p	**22**	**GUGGUGGCGGUGGUGUUGCCAG**	**pmc-miR-4070-3p**	**19**	**CACGCGUCACGUGACACCU**
pmc-miR-10256-3p	**20**	**CAUGUCUGCUAUGGCCUUCC**	**pmc-miR-4133-5p**	**19**	**CAUGUUGUGUUUUUGUAUG**
pmc-miR-10267-3p	**20**	**CAGAUCAAACCACCGCCCCC**	**pmc-miR-4140-3p**	**20**	**ACCAGAUGAUUAUGACACAA**
pmc-miR-10492a-3p	**20**	**GUUUAGCUCUCUGAUUUAUG**	**pmc-miR-4177-5p**	**20**	**UAAUUGCUGUUUUAAACAGU**
pmc-miR-10508b-3p	**21**	**AACAGCAUUUCGUGACUUCUU**	**pmc-miR-4185-3p**	**22**	**UGUAUUCAUACUGUCUGUUACA**
pmc-miR-10509-5p	**23**	**UGUGUCACGUACAGUAACACUGA**	**pmc-miR-4323-3p**	**18**	**CAGCCCCACAGCCUCAGG**
pmc-miR-10578-5p	**22**	**AGCAGUGAGUCAGUCUCACGCU**	**pmc-miR-4472-3p**	**18**	**GGUGGGGGGUGUUGUUGA**
pmc-miR-10611-3p	**22**	**UUUCCCUUUUUCACACAGUGGG**	**pmc-miR-449d-3p**	**20**	**GAAGGCUGUGUGCUGUAAAG**

pmc-miR-10639-3p	18	UUGCCAUGAUAGCUUUGG	pmc-miR-4629-3p	20	UGACUGGACGUUCAUCGCCG
pmc-miR-10770-1-3p	22	UUGUUGAUGUUGUUGUUGUUGU	pmc-miR-4654-5p	23	UGUGGGAUCUGGAGGCAGACUUC
pmc-miR-10770-2-5p	22	UUGUUGAUGCUGUUGUUGUUUC	pmc-miR-466-1-3p	23	AUAUAUACACACACACAUAUAUA
pmc-miR-10892-3p	22	UGAAUAAUGGAUGAAUAACAUC	pmc-miR-466-2-5p	23	AUAUAUACACACACACAACCAUG
pmc-miR-10972-3p	22	UUUGCUAUGGUCAUCAUAUAUG	pmc-miR-466-3-3p	23	AUAUAUACACACACACAUAUACA
pmc-miR-10b-1-3p	22	CAUCUCGGUACUCGGGAGGUCA	pmc-miR-466h-3p	19	UACGCACGCACACACACAC
pmc-miR-10b-2-5p	24	UUACCCUGGAGAACCGAGCGUGUG	pmc-miR-466i-1-5p	20	UGUGUGUGUGUGUGUGUGUG
pmc-miR-11054-5p	22	AGUGUUUGUUGCCAGAGAUUUA	pmc-miR-466i-2-5p	20	UGUGUGUGUGUGUGUGUGUG
pmc-miR-11280-3p	22	AUGAGAGAAGAUCCGUGUGUGA	pmc-miR-466m-5p	23	UGUGUGCAUGUGCAUGUGAAUGU
pmc-miR-1175-1-3p	23	UGAGAUUCAACUCCUCCAACUGC	pmc-miR-466n-3p	22	GUGUGUGCGUACAUGUACUCAU
pmc-miR-1175-1-5p	23	AGUGGAGAGAGUUUUAUCUCAUC	pmc-miR-466q-3p	20	GUGCACACACACACAUACAC
pmc-miR-1175-2-5p	22	AAGUGAAGAGAAGAAUGAAGAA	pmc-miR-467d-5p	22	UAAGUGCGCGCAUGUAUUUAUG

pmc-miR-1187-3p	23	UAUGUGUGUGUGUAUGUGUGUAU	pmc-miR-467f-3p	21	AUAUACACACACACACCUUUC
pmc-miR-11922-3p	22	UUCAGGAGAUCAUCGAUAUGUU	pmc-miR-4715-5p	23	GUGCCACCUUAACUGCACGGACC
pmc-miR-12096b-3p	24	UAGGGCAAAAUUAUCACCGAUUAC	pmc-miR-4750-3p	22	CCUGACCCACCCCCUCCCUACU
pmc-miR-12096b-5p	24	AAGCGGUGAUACUUUUGUCUCGCA	pmc-miR-4757-3p	22	CAUGACGUCACAGAGGCGAGGU
pmc-miR-12228-5p	20	UCCCCACUGCUCCCCCAACA	pmc-miR-4769-5p	22	UCUGCCAUCCUCCCUCUCGUCU
pmc-miR-12245-5p	22	CCCCGCCCCGCCCGGCUCAAGG	pmc-miR-487a-5p	22	GUGGCUAUCCCUGCUGUCAUCC
pmc-miR-1224-5p	21	UCACCUCCUCUCUCCUCCCUU	pmc-miR-4938-5p	23	AUCAUCAUGCAUCAUCAUUAUUG
pmc-miR-12286-3p	23	UGGGGCUGCAGGGCUGAGUAAGC	pmc-miR-49-3p	22	AAGCACCAUGUGAAGCACUUUU
pmc-miR-12287-5p	21	UGGGGGUGCAGGUGGGCACAC	pmc-miR-4949-3p	22	AGUGGUGUGCGUGUGUGUGUGU
pmc-miR-12292-5p	19	AUGGGGUCUUGGGGUCAUG	pmc-miR-4968-1-3p	22	CAGCAACAGCAGCAGCAGCAGC
pmc-miR-12293-5p	23	CGCCUCUUGAUCUACUAUAACUA	pmc-miR-4968-2-5p	22	CAGCAACAGCAGCAGCAGCAGC
pmc-miR-12296-3p	18	GGGCAGGGGCAGGGGGCA	pmc-miR-4968-3-3p	22	CAGCAACAGCAGCAGCAGCAAU

pmc-miR-12321-5p	23	GCCCUGCCCUCCCUUCCCCUCUC	pmc-miR-4968-4-3p	22	CAGCAACAGCAGCAGCAGCAGC
pmc-miR-12327-5p	21	UUUCUCUCUUUCCCUCUCUCU	pmc-miR-4999-3p	21	UGCUGUAUUGUCAGGUUUGUA
pmc-miR-12339-3p	22	AGAUUGUUCUGGAUUGCCACUG	pmc-miR-501-5p	21	AAUGCACCCGGGCAAGUCAGA
pmc-miR-12358-3p	22	AUCUAUGAUUUCAUCGGGUAAG	pmc-miR-5101-1-5p	22	UUUGUUUGUUUUGCUGUCUGAC
pmc-miR-12396-5p	23	AGAGAGACACAGACAAGAAAGAU	pmc-miR-5101-2-5p	22	UUUGUUUGUUUUGCUGUGUGCU
pmc-miR-12-3p	22	AGUACCUUUUGUGAUAUUCUUA	pmc-miR-519f-3p	22	AAAGUGCAUCCUUUUAUGGCGA
pmc-miR-12410-3p	22	AAGGAAACUGAGAUCCGGUCAC	pmc-miR-5317a-3p	24	UUUCUUGUCUCUGUGUACCUCUCG
pmc-miR-124-5p	22	CGCUUUCAUCCGUGACGACACC	pmc-miR-5391-5p	20	UGGACAGUUAUGAAGAAGUG
pmc-miR-12-5p	22	UGAGUAUUACAUCAGGUACUGA	pmc-miR-539-5p	22	AUCAUACAAGGACAAUUUGUCA
pmc-miR-1273h-3p	21	CUGCAGACUCGACCUCAGGAU	pmc-miR-5396b-5p	22	CCACACAACCAGCAAGAUUUGC
pmc-miR-1277-3p	22	UAUAUAUAUAUAUGUACUCUAC	pmc-miR-5549-3p	19	UCAUGUUGGUUUUUUGAAU
pmc-miR-130b-3p	23	CAGUGCAAUAAUGAAAGUAUUUU	pmc-miR-5552-3p	23	UGUAGUUUGUAGUCUAAAGUCUG
pmc-miR-1322-5p	18	GAUGCUGCUGCUGAUGGC	pmc-miR-5595-3p	24	UCUCUUUUUUCUCGCAGUCCCGCA

pmc-miR-133c-5p	**21**	**CAGCUGGUUGAAGGGGACCAA**	**pmc-miR-5600-3p**	**22**	**AGUGGAAUGUUUUGUUUCAGUA**
pmc-miR-1376-3p	**21**	**GCUCAGCAGCACUCUUUGAGC**	**pmc-miR-5612-3p**	**22**	**UAUACAUCACCAGUUGACAUGU**
pmc-miR-137b-3p	**23**	**UUAUUGCUUGAGAAUACACGUAA**	**pmc-miR-574-1-3p**	**21**	**UGAGUGUGUGUGUGUGAGUGC**
pmc-miR-137b-5p	**24**	**ACGGGUAUUCUUGGGUAAAUAAUA**	**pmc-miR-574-2-3p**	**24**	**UGAGUGUGUGUGUGUGAGUGUGUG**
pmc-miR-13b-3p	**17**	**UCGUUAAAAUGUUUGUG**	**pmc-miR-5918b-5p**	**22**	**UCGCUCUACAUCGUGUGGUCAG**
pmc-miR-1-3p	**22**	**CAUGCUUCUUUAUAGUUUGACC**	**pmc-miR-5965-5p**	**20**	**GACAAUUCUGACAUUUUGCA**
pmc-miR-1421am-3p	**22**	**AGGAGGUCGUAGGUCAUCCUGU**	**pmc-miR-6056-3p**	**20**	**GAGGGACGAGGAGGGAGGAG**
pmc-miR-1422j-3p	**21**	**AAAACUGCAUCUGGAUUAUGA**	**pmc-miR-6076-3p**	**21**	**AGCAUGACAGAGGAGAGGAGA**
pmc-miR-143-3p	**24**	**GUCUGAGAUGAAGCACUCGUUUGC**	**pmc-miR-6098-5p**	**20**	**AUAGUGAUUGUGUGUUUCUA**
pmc-miR-149-3p	**23**	**GAGGGAGGGACGGGGGAUUCAAG**	**pmc-miR-623-3p**	**23**	**UUCCCUUGCAGGGGCUGAGGUAU**
pmc-miR-150-3p	**21**	**CUGGUACAGGCCUGGGGGAGA**	**pmc-miR-626-5p**	**19**	**AGCUGUCUGAAAAUGUUUG**
pmc-miR-153-3p	**23**	**UUGCAUAGUCACAAAAGUGAUCG**	**pmc-miR-6302-3p**	**22**	**CAAACUCAUCGUCAGCCACUCA**
pmc-miR-	**22**	**UGAGUUUGUGUUGCUGGGACAA**	**pmc-miR-**	**21**	**CUCCGUAUCAUCUGCUAGCUA**

1551-3p			**6416-3p**		
pmc-miR-15c-3p	**22**	CAGACCAUUCUGGGCUAUGGGG	**pmc-miR-6505-5p**	**21**	UGACUUCUACCUGUUCCCUAA
pmc-miR-1603-1-3p	**20**	GUGGUUUGUUUUGUGUUGCG	**pmc-miR-651-3p**	**22**	UUUAGGAUAAGUUUGAACACGU
pmc-miR-1603-2-3p	**20**	UUGGUUUGGUUUGUUUUUUU	**pmc-miR-6516-3p**	**22**	AUGCAGUAACAGGUGUCUCCAC
pmc-miR-1632-3p	**23**	UGCUUGUUUUUGGAUGAAAUUAU	**pmc-miR-6528-5p**	**20**	AGAGGGAAGAGGGAAGAAAG
pmc-miR-1642-3p	**23**	UGAGAGGCUGUCAGUUUUGUAAG	**pmc-miR-6548-3p**	**22**	AGAGGUGCCCCGCUGUCUGUUC
pmc-miR-1677-3p	**23**	UUGACUUCAAUAGGAGAGACAGG	**pmc-miR-6560-5p**	**23**	UGUGUUGCUCCAUACUGCUGAAG
pmc-miR-1728-3p	**23**	UGGUUGAUCCAUUGGAAGAAACA	**pmc-miR-6562-5p**	**22**	AGGGGAAAGGAAUGUGCAGGUA
pmc-miR-17-3p	**23**	ACUGCACUGCAAGCACCGCCACA	**pmc-miR-6568-3p**	**21**	GCGGCCGCCAUCUUGCGGAGC
pmc-miR-1744-5p	**22**	ACUUCAACAGGAGCAAGUCAAG	**pmc-miR-669f-5p**	**23**	CAUAUACAUACACACACACGUGU
pmc-miR-1775-5p	**22**	UCCUGUAGCCAGAAGACAGCGG	**pmc-miR-669j-3p**	**22**	UGCAUAUACUCACAUGUGCACA
pmc-miR-1781-1-5p	**21**	UUUAAAUCAUCCAGCUGCCUU	**pmc-miR-6701-3p**	**21**	AUUAUUUUACAGACAGCAACU

pmc-miR-1781-2-5p	21	UUUAAAUCAUCCAGCUGCCUU	pmc-miR-67-3p	23	UCACAACCUGCAUGAAUGAGGAC
pmc-miR-1896-3p	18	UGGUGGGUGAGGAGGAGG	pmc-miR-6876-5p	24	AGCUGUCUGUGUUUUCCCAGAUGC
pmc-miR-190-5p	23	AGAUAUGUUUGAUAUAUUUGGUG	pmc-miR-6891-5p	21	CCCUCAUCUUCCCCUCCCCUC
pmc-miR-1951-3p	22	GUAGUGGAGACUGGUGGCGAAA	pmc-miR-6971-3p	21	ACAGCCUCUGCUUCUUCAGCU
pmc-miR-1955-5p	21	GAGCAUUGCAUGCUGGACGCG	pmc-miR-6974-3p	22	UCUCCACUCUCUUCUGUGUCGU
pmc-miR-196a-3p	22	ACGGCAACAAGAAACUGUGACU	pmc-miR-7000-5p	22	CACCCACCUGCCUGUCACACUG
pmc-miR-1984-5p	23	UGCCCUAUCCGUCAGGAACUGUG	pmc-miR-7046-5p	24	UGUAGGGUGAGGCUGGUGGGUUAG
pmc-miR-1985-5p	24	UGCCAUUUUUAUCAGUCACUGUGU	pmc-miR-7-1-3p	21	CAACAAAUCGUAGCCUCAGAG
pmc-miR-1990-3p	21	CGGGACUACGUCAACGUACUA	pmc-miR-7151-5p	23	GAUCCAUCUCUGCCUGCUCACCU
pmc-miR-1990-5p	22	AGUAAGUUGAUGGGGUCCCAGG	pmc-miR-71-5p	22	UGAAAGACACAGGUAGAAUGUA
pmc-miR-199-3p	21	ACAGUAGUCUGCACAUGCUAU	pmc-miR-7207-5p	20	AUCUUUUAAAAUACAGGGUG
pmc-miR-1994-3p	22	UGAGACAGUGUGUCCUCCCUCU	pmc-miR-722-5p	22	UUUUGCAGAAACGUUUACAGUU

pmc-miR-1994a-3p	22	UGAGACAGUGUGUCCUCCCUUG	pmc-miR-7-2-3p	22	CAACAAGUCCCAGUCUUGGCGG
pmc-miR-1a-3p	22	UGGAAUGUAAAGAAGUAUGUAC	pmc-miR-7241-3p	23	ACUUGCAUGAGUUAAUGAUUUGU
pmc-miR-1a-5p	24	ACAUACUUCUUUGCUAUCCCAUAU	pmc-miR-7314-3p	18	UCAGAAACAGUGUCUCUG
pmc-miR-1b-3p	17	UCAUACUUCUUUACAUU	pmc-miR-7323-3p	21	UUACAGAAUUGUGUGAUCAAC
pmc-miR-1c-5p	22	GUACAUACUUCUUUACAUUCCA	pmc-miR-7386e-5p	21	GUAAACACUUUACCCGUUUGA
pmc-miR-2001-5p	23	UUGUGACCGUUACAAUGGGCAUU	pmc-miR-7388c-3p	20	UGCACAGACGUGACAUCGUU
pmc-miR-200b-5p	22	CAUCUUACCUGACAGUCAACAG	pmc-miR-7398a-3p	23	UGUGUAGAGGUGAGAAAGUUGUC
pmc-miR-201-3p	22	UGAACAGCGCCUUUCUCACGUG	pmc-miR-743b-3p	20	UGUUCAGACUGGUGUCUGGU
pmc-miR-2017-3p	22	UACCUGUGCACCUGUGCACGUG	pmc-miR-7448-3p	23	AACUGUUGUAGAACAGUUCCGAU
pmc-miR-204-3p	19	UGCAAGGACAGCAAAGGGA	pmc-miR-745a-3p	23	AGCUGCCUGAUGAAGAGCUGUCC
pmc-miR-2064-3p	22	AAGCAGCACUGUGCAAGAUACU	pmc-miR-745b-3p	22	GAGCUGCCAAAUGAAGGGCUGU
pmc-miR-2155-3p	23	GACACUGUUUUACACUCUACUGG	pmc-miR-7472-3p	22	AUGUGCUGGAGAUGAAAACUGU

pmc-miR-216a-3p	22	CAAGUUACUAGCCGAGAUUACA	pmc-miR-750-3p	23	CCAGAUCUAACUCUUCCAGCUCA
pmc-miR-216a-5p	23	UAAUCUCAGCUGGUAAUUCUGAG	pmc-miR-750-5p	23	AGUUGGAAGAUUGGGUCUUUGGC
pmc-miR-216b-1-3p	23	UAAUAUCAGCUGGUAAUCCUGAG	pmc-miR-751-3p	21	CAUGUUUGAAUGGCCACCACU
pmc-miR-216b-2-3p	22	CACAGUGGCUACUGGGUCCACA	pmc-miR-7562-5p	22	CACACACAUUCAUGGACACGCA
pmc-miR-216b-3-3p	22	CACACUUACCUGUAGACUGUUA	pmc-miR-7575-5p	20	GCAUGGUCAUGAUCAUGACA
pmc-miR-216b-3-5p	21	ACAGUUACCUGUAGAUUGUGU	pmc-miR-7643-3p	22	GACAUCUGUUUCUCAGUAAUUC
pmc-miR-2223-3p	21	UUUGUACUCAUGUAAGUAUUU	pmc-miR-785a-5p	21	UAAGUGAAUACUCUGUUAUGU
pmc-miR-2238i-3p	23	CCGUUCACUCAUCUCGGAGCUGA	pmc-miR-7880b-3p	22	UGGUUGGAUCAGUUUGGUUUGG
pmc-miR-2284c-3p	22	AAAAUCUAAGUGAACUGUCAAG	pmc-miR-7880q-3p	22	UUUGCGACCAAAAUCACUCUCU
pmc-miR-2298-3p	20	GACUGAUGUUGUUGUUCAAG	pmc-miR-79-3p	20	GUAAAGCUAAAUUACCAGUG
pmc-miR-2304-3p	20	GGUUGUGUGGUUGUGUGGGC	pmc-miR-8250d-3p	25	UCCUUUAUUUGUCUGGGUGGAGGGU
pmc-miR-2361-3p	20	UUUGUGUUGUUUUGUUUUGC	pmc-miR-8265-3p	24	UCUGAUACUCUUCGCUCCACCUUC
pmc-miR-	20	UAUAGUUUUGUGUGGUGUAU	pmc-miR-	23	AUGAAAGAUAAAAGUGUCUCGUG

2371-5p			**8293-5p**		
pmc-miR-2470-3p	**21**	**UUGUGUGGUUUUGGAUACUUG**	**pmc-miR-8311-3p**	**22**	**UGAUUGUGAGAUUCCCCCACCU**
pmc-miR-2491-1-3p	**18**	**CAACAACAGCAGCAGCAA**	**pmc-miR-8335-3p**	**23**	**GUUGUUGUUGUUGUUUUGUCAGU**
pmc-miR-2491-2-3p	**18**	**CAACAACAGCAGCAGCAA**	**pmc-miR-8398-3p**	**22**	**AGUUCUUUCUUUUUAGCAUGCA**
pmc-miR-2491-3-3p	**18**	**CAACAACAGCAGCAGCAA**	**pmc-miR-8-3p**	**23**	**UAAUACUGUCAGGUAAAGAUGUC**
pmc-miR-2493-3p	**21**	**GCUCACACACACACACAGCCA**	**pmc-miR-8422-5p**	**19**	**GCAAUUAAAAGUAGCACAA**
pmc-miR-2505-3p	**22**	**UCGUGCACACACACACAUACAC**	**pmc-miR-8443-5p**	**19**	**ACUUUGCAUUCAUAUUUGA**
pmc-miR-252a-5p	**22**	**CUAAGUACUAGUGCCGCGGGAA**	**pmc-miR-8460-5p**	**20**	**UAAUUAUAUUAUAUUUGCUU**
pmc-miR-254-5p	**23**	**UGCAAAUCUUUUGCAACAGACUU**	**pmc-miR-8462-3p**	**20**	**UGAUUAAUUGAUAAGAUGA**
pmc-miR-2571-3p	**25**	**GUGAGUGGUGAUGAUGUUUACCACU**	**pmc-miR-8470-3p**	**20**	**CUGAGCAUAAGAAUAUGUUC**
pmc-miR-279-3p	**22**	**UGACUAGAUCCACACUCAUCCA**	**pmc-miR-84f-5p**	**23**	**UGAGGUAGUUUUAAAUGAAGCUU**
pmc-miR-279b-5p	**22**	**UGACUAGACUGAUAACCCCUAG**	**pmc-miR-85-3p**	**24**	**UACAAAGUAUUUGAAACCACAGAG**
pmc-miR-279c-3p	**22**	**CGGGUGUUCGUUUCGAGUACGA**	**pmc-miR-8-5p**	**22**	**CAUCUUACCUAACAGCAUUAGA**

pmc-miR-281-3p	22	UGUCAUGGAGUUGCUCUCUUUA	pmc-miR-8834a-5p	23	UGCCGGGCCUGGAGGCCAGCUGU
pmc-miR-281-5p	21	AAGGGAGCAUCCGUCGACAGU	pmc-miR-888-5p	22	GACUGACACCUCUUUGUCCAGC
pmc-miR-282-3p	22	ACAUAGCCUAAAAGAGGCACCA	pmc-miR-8993-5p	23	CAGGGUCAAGGGUCAAACAUGGU
pmc-miR-2953-3p	21	UACAGAAGUGUUCGUGGGGGA	pmc-miR-9120-5p	21	UCAGUCUCCCUCUCUCACACA
pmc-miR-2a-1-3p	24	UAUCACAGCCUGCUUGGAUCAGUA	pmc-miR-9-1-3p	23	UCAUACAGCUAGAUAACCAAAGA
pmc-miR-2a-2-3p	24	UAUCACAGCCAGCUUUGAUGAGCU	pmc-miR-9-1-5p	23	CCUUUGGUAACCUAGCUUUAUGA
pmc-miR-2a-3-3p	24	AAUCACAGCCUGCUUUGGUGAGCU	pmc-miR-9170-3p	23	UUUGCAUCCUGGCUCUGCUAUCU
pmc-miR-2b-1-5p	24	UCAUCAAAGCUGGCUGUGAUAUGA	pmc-miR-9-2-3p	23	AUAAAGCUAGGUUACCAAAGGCA
pmc-miR-2b-2-5p	24	UCAUCAAAGCUGGCUGUGAUAUGA	pmc-miR-9256b-3p	22	UAAUAAUCAGGCCACUACCUCU
pmc-miR-2f-3p	23	UAUCACAGCCAUGCUAAUCUCCU	pmc-miR-9-2-5p	25	UCUUUGGUUAUCUAGCUGUAUGAUU
pmc-miR-300-3p	22	UAUACAAGGGCAGACUCCAUCC	pmc-miR-92a-1-3p	23	GAUUGCACUUGUCCCGGCCUUCU
pmc-miR-302a-3p	23	AAGUGCUUCCUUUUAUUGGUGAU	pmc-miR-92a-2-3p	22	GAUUGCACUUGUCCCGGCCUUG
pmc-miR-302b-3p	23	AAAGUGCUUCCAUGUUUAAUUCA	pmc-miR-92b-3p	21	UAUUGCACUCGUCCCGGCCUA
pmc-miR-	22	UCCCACAGGCCCAGCUGUCACA	pmc-miR-92c-5p	24	AGGUUGGGAUGUGGGCUCUGUCGC

3057-3p					
pmc-miR-3071-5p	**22**	**AUCAUCAAAACAAAUGUGCAUA**	**pmc-miR-9341-3p**	**21**	**UCUCGGCCUUUUGGCUAAGAU**
pmc-miR-3084a-5p	**22**	**UUGAAGGUUCAUUAGCCUUGUG**	**pmc-miR-9369-5p**	**22**	**AUACAUAUUCUUCUUUCCAUUU**
pmc-miR-308-5p	**22**	**CGCAGGAUUUGCUUGUUUUGAG**	**pmc-miR-9382-5p**	**21**	**AUCACAGUGUGGCUGAAAUAU**
pmc-miR-31-3p	**21**	**UGGCAAGAUGUUGGCACAGCG**	**pmc-miR-9434-3p**	**23**	**UCUGACAGGAAUUCUGAACCCUG**
pmc-miR-315-3p	**23**	**UUUUGAUUGUUGCUCUAGAAAUU**	**pmc-miR-9505-5p**	**21**	**UGAUGUCUGUCAAAAUGUGAU**
pmc-miR-317-3p	**25**	**UGAACACAGCUGGUGGUAUCUUUUU**	**pmc-miR-9542a-3p**	**22**	**CCUGGCACUUUCUAAUUUCUGG**
pmc-miR-33-3p	**23**	**GCAAUGCAUCUGCAGUGCAAAUA**	**pmc-miR-9543a-3p**	**18**	**AAAUAUUGCUUGUUUAUU**
pmc-miR-33-5p	**22**	**GUGCAUUGAGGUUGCAUUGCAU**	**pmc-miR-96-5p**	**22**	**CAAUCAUGUGCAGUGCAGUUCU**
pmc-miR-336-5p	**21**	**UCACCCUUCCAUAUCUUUUGG**	**pmc-miR-969-3p**	**21**	**GAGUUCCACUAAGCAAGUUAU**
pmc-miR-33b-3p	**21**	**CAGUGCCUCUGCAGUGGAUGU**	**pmc-miR-96b-3p**	**22**	**AUUAUACACCGGUGCCAAGUCA**
pmc-miR-345-3p	**21**	**UGCUGACCCCUAGUCCGCCUC**	**pmc-miR-96b-5p**	**23**	**AUUUGGCACUUGUGGAAUAAUCG**
pmc-miR-3529-3p	**24**	**AACAACAAAAUCACUAGUCUUCCA**	**pmc-miR-970-5p**	**25**	**AGCCUUGCAUGAGCUCUAGGUGUCU**
pmc-miR-3532-3p	**22**	**UUGGAGGCUGCAGUGUGACCUC**	**pmc-miR-971-3p**	**22**	**AUGGUGUUCUAGCCUCCAUCGA**

pmc-miR-355-5p	**21**	**UUUGUUUUAGCCUGAGUCGCC**	**pmc-miR-981-3p**	**23**	**UUCGUUGUCGUCGAAACCUGCCC**
pmc-miR-3620-5p	**22**	**GUGGGCUGGGCUGGGCUGGGCU**	**pmc-miR-981-5p**	**22**	**CGGGUUUCGCGGCUUGCGAACA**
pmc-miR-36-3p	**23**	**UCACCGGGUGGAAAUUCGUCUUU**			
pmc-miR-3643-3p	**22**	**GUGAGUUUAUCUAAAUUCAGGA**			
pma-miR-36b-3p	**23**	**UCACCGGGUAUACAUUCAUCCGC**			

Fonte: Dados do autor

A partir do levantamento dos miRNAs das espécies com seus respectivos ortólogos, foram selecionados miRNAs que refletissem a seletividade por alguns táxons, corroborando assim para a ideia de conservação evolucional da estrutural dos precursores e miRNAs. Os táxons que foram utilizados como parâmetro de seleção de miRNAs específicos foram: Bilatéria, Protostômios, Lophotrocozoa e Mollusca.

Dentre estes miRNAs, buscou-se a caracterização de miRNAs bilatéria específicos, protostômio específicos, lophotrochozoa específicos e molusco específicos. As seguintes espécies foram utilizadas para análises; *Lanistes nyassanus* (lny), *Marisa cornuarietis* (mcr), *Pomacea canaliculata* (pcr), *Pomacea maculata* (pmc); animais do clado bilatéria, *Melibe leonina* (mle), *Lottia gigantea* (lgi), miRbase versão 22 (http://www.mirbase.org); todos os moluscos analisados com exceção dos acima citados (HUANG et al., 2021).

4.3 miRNAs BILATERIA ESPECÍFICOS

4.3.1 mir-9-1

A família dos genes de mir-9 é responsável por regular a neurogênese, tendo papel no desenvolvimento cerebral, em linhagens de células neurais e não neurais. A sequência deste miRNA maduro é idêntico em insetos e humanos, revelando sua preservação nucleotídica entre protostômios e deuterostômios. Em deuterostômios há uma preferência para utilização da fita 5p no complexo RISC; no entanto, no molusco marinho gastrópode *Aplysia californica* ambas as fitas são igualmente utilizadas para induzir o silenciamento da expressão gênica (YUVA-AYDEMIR *et al*., 2011). Os precursores de mir-9-1 foram encontrados no genoma das espécies *P. maculata* e *P. canaliculata*, com a presença de dois miRNAs maduros por espécie (Figura 5). Observa-se alta similaridade da estrutura secundária dos precursores de *P. canaliculata, P. maculata, L. nyassanus e M. cornuarietis* (Figura 6). Foi verificado ampla distribuição entre os clados filogenéticos da família mir-9, com a presença de mir-9-1 de *P. canaliculata* e *P. maculata* agrupados com o ampularídeo *M. cornuarietis* no filo dos moluscos (Figura7). Estes dados corroboram para identificação de mir-9-1 nos dois ampularídeos em estudo.

Figura 5: Alinhamento de precursores de miRNA-9-1 com ortólogos.

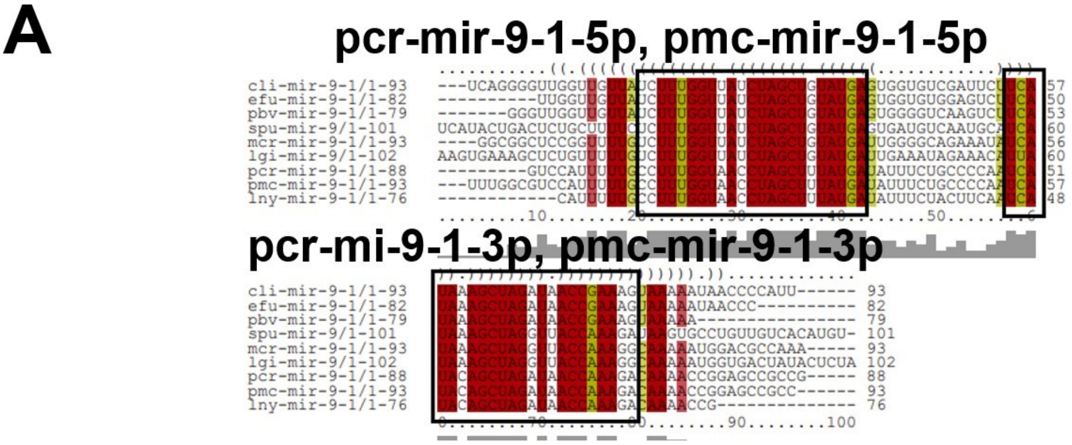


Figura 6: Estrutura secundária de precursores de miRNA-9-1 e ortólogos; spu: *Strongylocentrotus purpuratus*, lgi: *Lottia gigantea*,lny: *Lanistes nyassanus*, mcr: *Marisa cornuarietis*, pcr: *Pomacea canaliculata*, pmc: *Pomacea maculata*

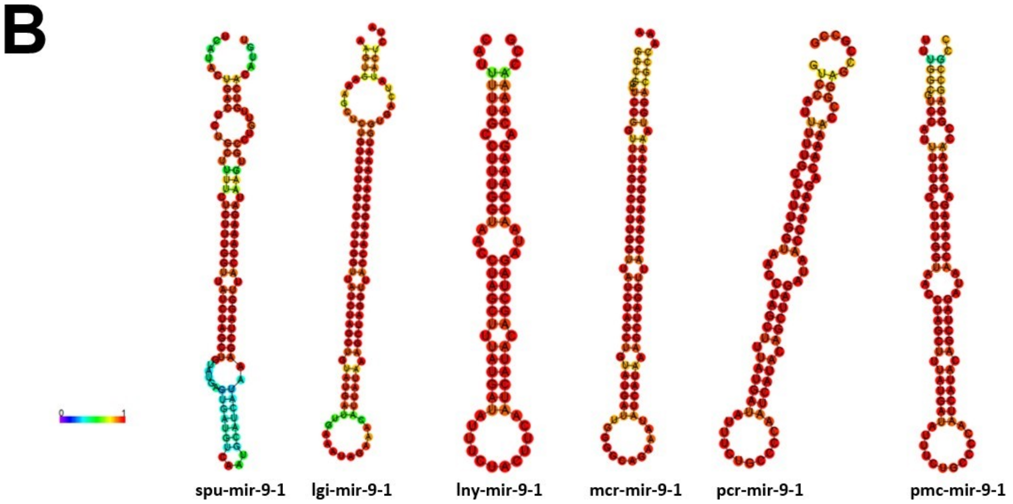
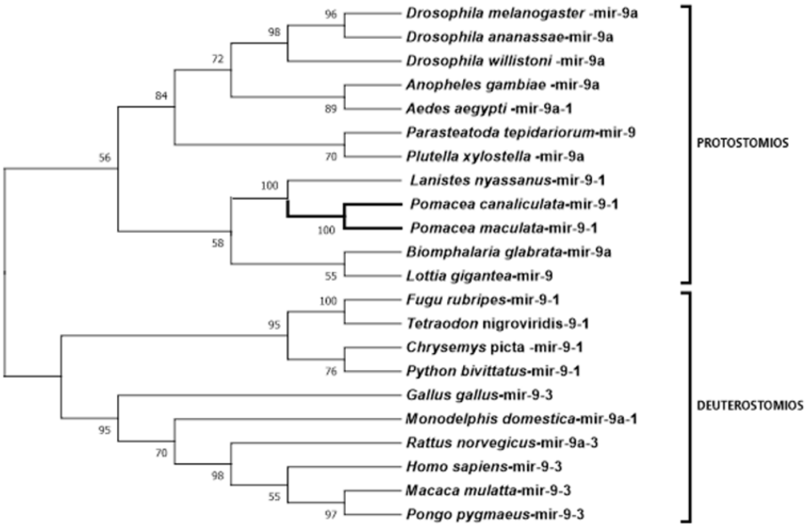


Figura 7: Distribuição filogenética de miR-9-1 (bilatéria-especifico).



Fonte: Dados do autor

4.3.2 mir-216a

A família dos genes de mir-216a está relacionada a processos patológicos em deuterostômios, especificamente em humanos. O mir-216a-3p inibiu a viabilidade, migração, invasão e proliferação de células de câncer de pulmão em humanos (WANG *et al*., 2020). Já o mir-216a-5p foi reconhecido como um oncogene e está envolvido na progressão e metástase de vários subtipos de câncer (CHEN *et al*., 2018). Nos organismos protostômios, a família do mir-216a foi encontrada no genoma de 18 espécies de moluscos, dentre estes 7 gastrópodes. (HUANG et al., 2021). Isso corrobora para a identificação dos precursores de mir-216 em *P. canaliculata* e *P. maculata*, com a presença de dois miRNAs maduros (cadeias 3p e 5p) por espécie (Figura 8). A estrutura secundária dos precursores de *P. canaliculata* e *P. maculata* apresentaram alta similaridade com seus ortólogos, principalmente com *L. nyassanus* e *M. cornuarietis* (Figura 9). Foi verificado ampla distribuição filogenética da família de mir-216a, com mir-216a de *P. canaliculata* e *P. maculata* apresentando-se agrupados (Figura 10).

Figura 8: Alinhamento de precursores de miRNA-216a com ortólogos.

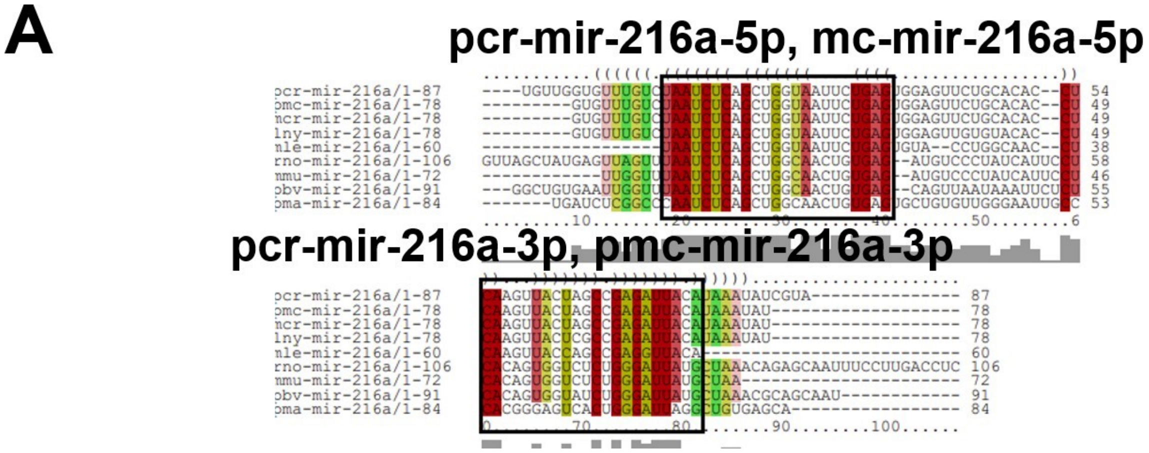


Figura 9: Estrutura secundária de precursores de mir-216a e ortólogos; mle; *Melibe leonina*, lny: *Lanistes nyassanus*, mcr: *Marisa cornuarietis*, pcr: *Pomacea canaliculata*, pmc: *Pomacea maculata,* rno; *Rattus norvegicus.*

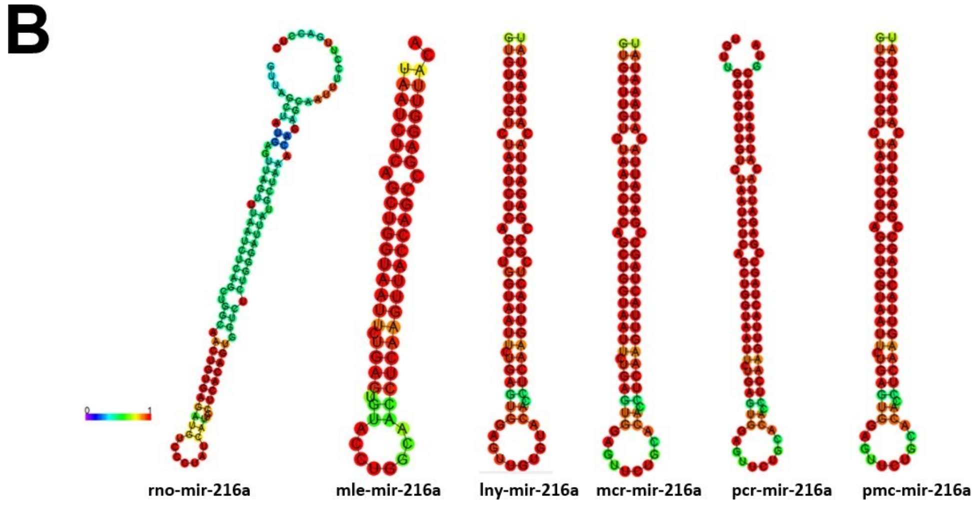
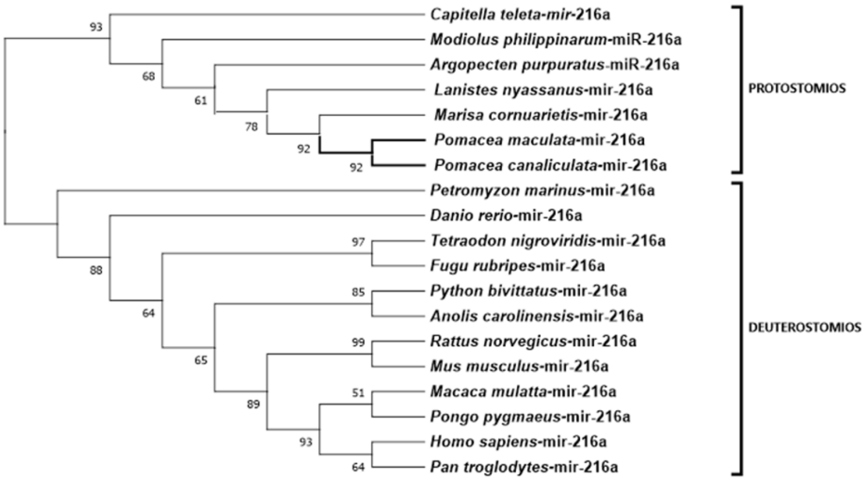


Figura 10: Distribuição filogenética de mir-216a (bilatéria-especifico).



4.3.3 mir-133

A família dos genes mir-133 é expressa em tecido muscular, sendo responsáveis pela remodelação cardíaca em humanos e diferenciação miocárdica em camundongos (LI; ZHOU; TANG, 2018; TAKAYA *et al.*, 2009). Os homólogos de mir-133 foram identificados em diversas espécies, dentre estas os invertebrados como a *Drosophila melanogaster* (CHEN *et al.*, 2006). O mir-133 têm ampla distribuição filogenética entre protostômios e deuterostômios, conforme verificado na Figura 13 deste trabalho. Em moluscos, na ostra *Crassostrea gigas,* o mir-133 mostrou-se ser responsivo ao estímulo de noradrenalina, sendo este miRNA responsável pela modulação do sistema imunológico nestes animais (CHEN *et al*., 2015). A família dos genes de mir-133 foi encontrada no genoma de *P. maculata* na isoforma mir-133c. Já para *P. canaliculata* foi verificado na espécie dois miRNAs maduros no precursor pcr-mir-133 (Figura11). Observa-se extrema similaridade da estrutura secundária dos precursores de mir-133 entre organismos da família Ampullariidae (*L. nyassanus*, *M. cornuarietis*, *P. canaliculata*) e também destes com *L. gigantea* (Figura 12).

Figura 11: Alinhamento de precursores de miRNA-133 e ortólogos, em destaque miRNAs maduros.

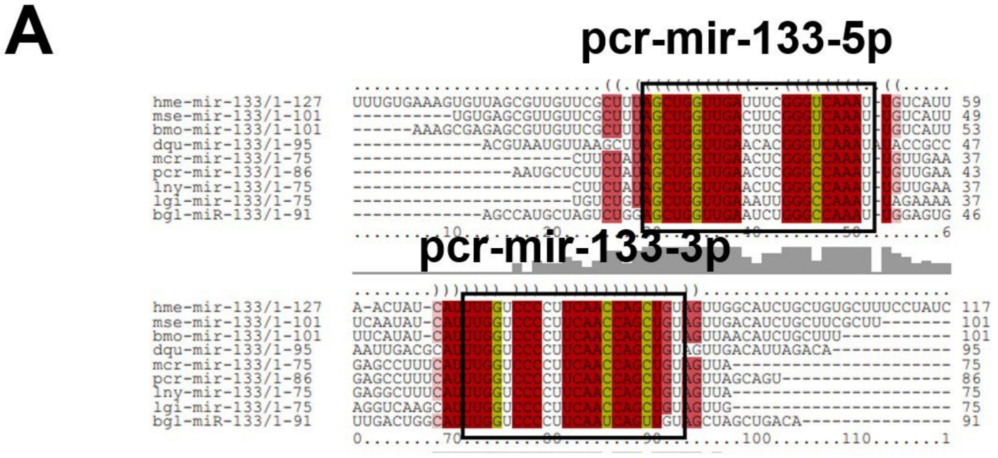


Figura 12: Estrutura secundária de precursores de miRNA-133 e ortólogos; hme: *Heliconius melpomene*, bmo: *Bombyx mori,* lgi: *Lottia gigantea, lny: Lanistes nyassanus, mcr: Marisa cornuarietis, pcr: Pomacea canaliculata.*

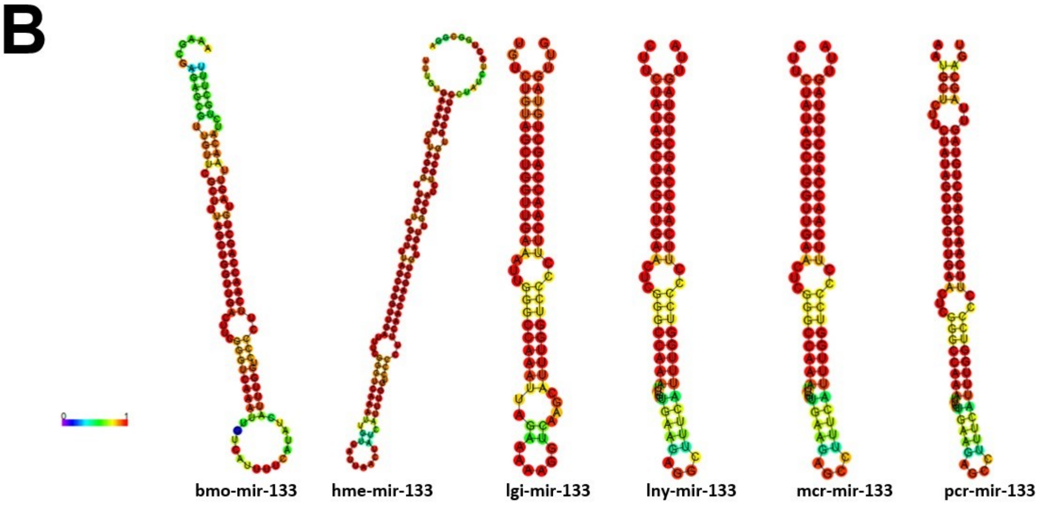
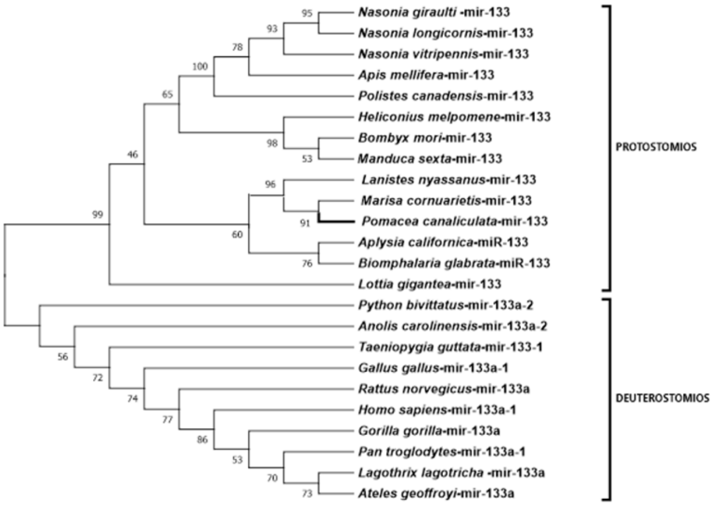


Figura 13: Distribuição filogenética de mir-133 (bilatéria-específico).



Fonte: Dados do autor

4.3.4 mir-33

Os precursores do mir-33 foram encontrados neste estudo: pcr-mir-33, pmc-mir-33 (Figura 14). Os precursores localizados nas 2 espécies apresentaram dois miRNAs maduros cada, 3p e 5p, corroborando estudos observados em moluscos bivalves e *D. melanogaster* (BAO *et al*., 2014; CLERBAUX *et al*., 2021). Foi realizada a predição das estruturas secundárias dos pré-miRNAs e seus ortólogos (Figura 15). O mir-33-5p supostamente regula os genes envolvidos na resposta ao estresse induzido por metais pesados no molusco *Tegillarca granosa* (BAO et al., 2014). Da mesma forma, *P. canaliculata* é capaz de sobreviver em ambientes com metais pesados, onde há alta concentração desses metais nas brânquias (KRUATRACHUE et al., 2011), provavelmente regulados por este micro-RNA. Além disso, miR-33 é um regulador pleiotrópico de processos metabólicos e de desenvolvimento em *D. melanogaster*. Sugere-se que o mir-33 seja um regulador conservado da homeostase lipídica (CLERBAUX et al., 2021). A árvore filogenética mostrou a ocorrência do miRNA em Deuterostômios (peixes e mamíferos), bem como em Protostômios (Ecdysozoa e Lophotrocozoa) (Figura 16).

Figura 14: Alinhamento de precursores de miRNA-33 com ortólogos.

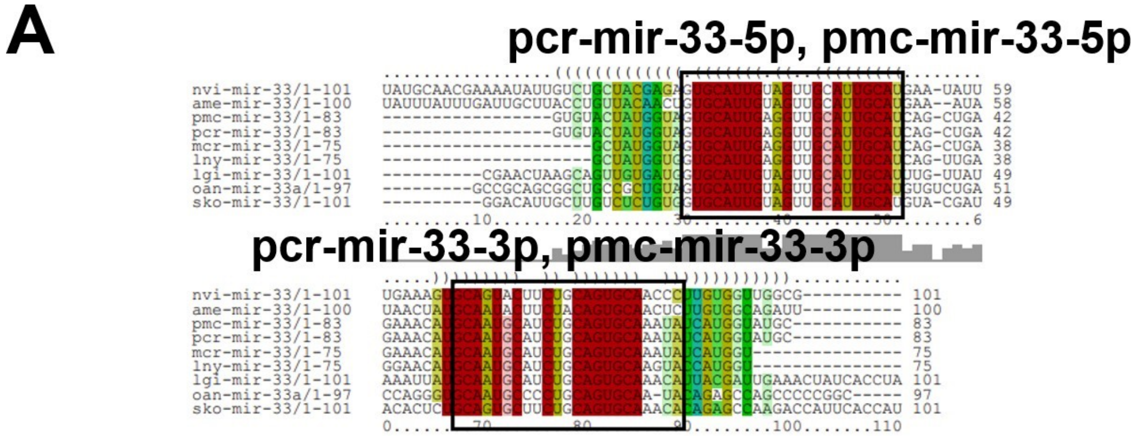


Figura 15: Estrutura secundária de precursores de miRNA-133 e ortólogos; nvi: *Nasonia vitripennis,* lgi: *Lottia gigantea, lny: Lanistes nyassanus, mcr: Marisa cornuarietis, pcr: Pomacea canaliculata,* pmc: *Pomacea maculata*.

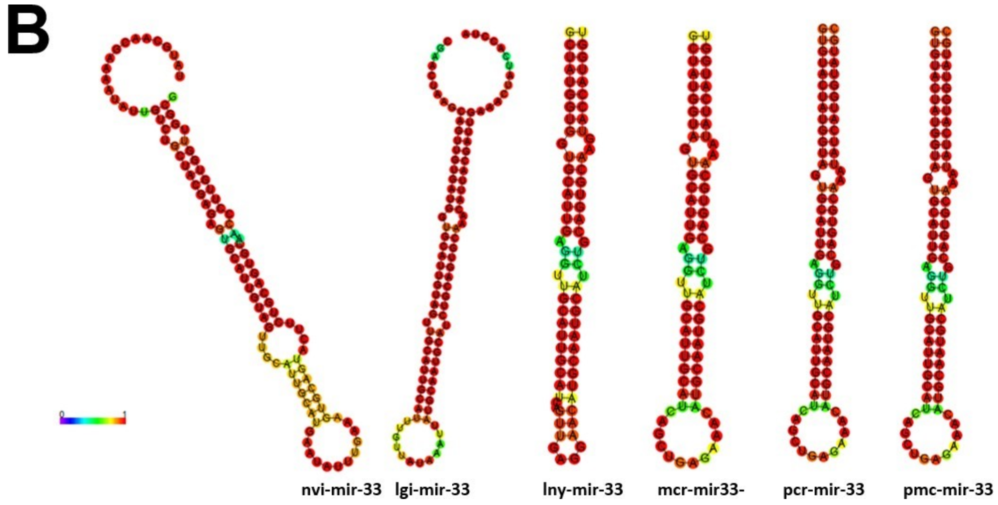
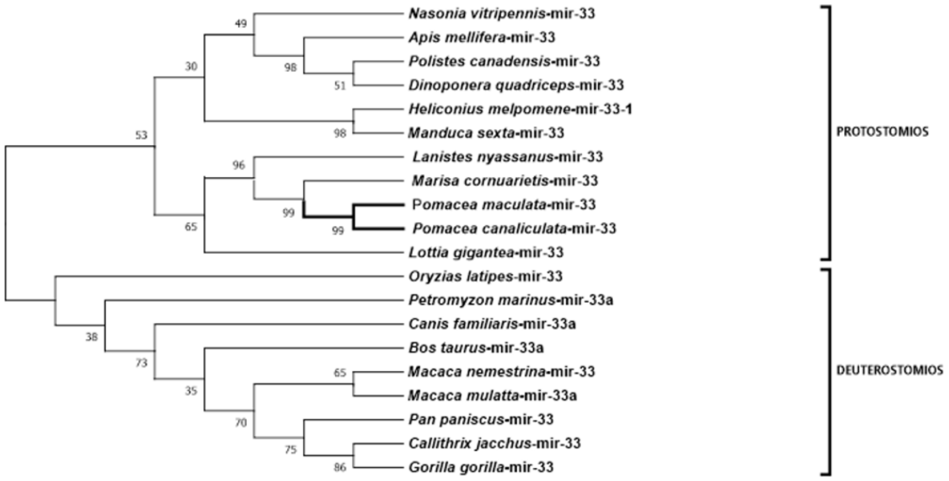


Figura 16: Distribuição filogenética de mir-33 (bilatéria-específico).

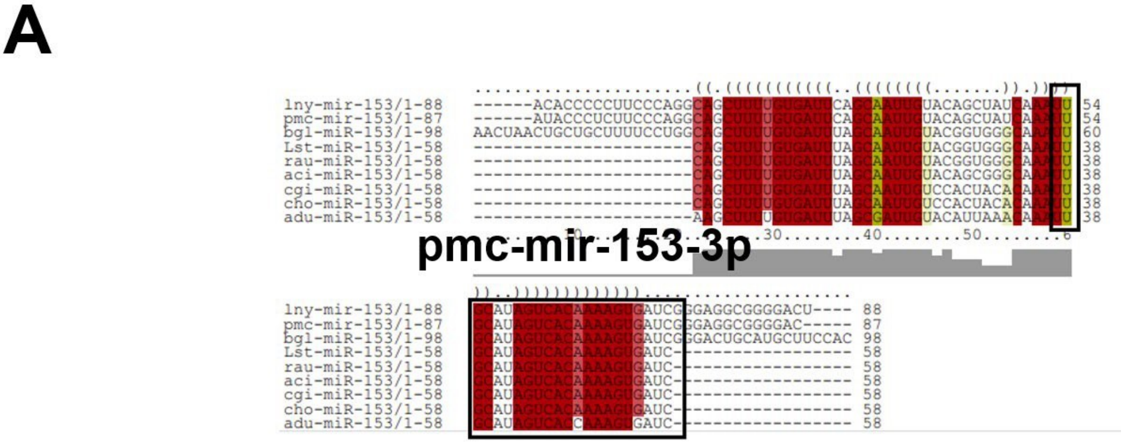


Fonte: Dados do autor

4.3.5 mir-153

A família dos genes de mir-153 foi encontrada no genoma de *P. maculata*. O precursor pmc-mir-153 apresentou o miRNA maduro na extremidade 3' (Figura 17). Observa-se alta preservação nucleotídica na estrutura secundária e primária de *P. maculata* e seus ortólogos, principalmente na região do miRNA maduro (Figuras 17 e 18). Foi verificado ampla distribuição entre Deuterostômios e Protostômios, neste clado com presença do filo molusco sendo representado por bivalves e gastrópodes (Figura 20).

Figura 17: Alinhamento de precursores de miRNA-153 com ortólogos, em destaque miRNA maduro.



```
                      ......................((.((((((((((..((((((((.......)).)))))
lny-mir-153/1-88      ------ACACCCCCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGCUAUCAAAUU 54
pmc-mir-153/1-87      ------AUACCCUCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGCUAUCAAAUU 54
bgl-miR-153/1-98      AACUAACUGCUGCUUUUCCUGGCAGCUUUUGUGAUUUAGCAAUUGUACGGUGGGCAAAUU 60
Lst-miR-153/1-58      ----------------------CAGCUUUUGUGAUUUAGCAAUUGUACGGUGGGCAAAUU 38
rau-miR-153/1-58      ----------------------CAGCUUUUGUGAUUUAGCAAUUGUACGGUGGGCAAAUU 38
aci-miR-153/1-58      ----------------------CAGCUUUUGUGAUUUAGCAAUUGUACAGCGGGCAAAUU 38
cgi-miR-153/1-58      ----------------------CAGCUUUUGUGAUUUAGCAAUUGUCCACUACACAAAUU 38
cho-miR-153/1-58      ----------------------CAGCUUUUGUGAUUUAGCAAUUGUCCACUACACAAAUU 38
adu-miR-153/1-58      ----------------------AAGCUUUUGUGAUUUAGCGAUUGUACAUUAAACAAAUU 38
                                   30........40........50.......6

                      ))..)))))))))))))))...................
lny-mir-153/1-88      GCAUAGUCACAAAAGUGAUCGGGAGGCGGGGACU---- 88
pmc-mir-153/1-87      GCAUAGUCACAAAAGUGAUCGGGAGGCGGGGAC----- 87
bgl-miR-153/1-98      GCAUAGUCACAAAAGUGAUCGGGACUGCAUGCUUCCAC 98
Lst-miR-153/1-58      GCAUAGUCACAAAAGUGAUC------------------ 58
rau-miR-153/1-58      GCAUAGUCACAAAAGUGAUC------------------ 58
aci-miR-153/1-58      GCAUAGUCACAAAAGUGAUC------------------ 58
cgi-miR-153/1-58      GCAUAGUCACAAAAGUGAUC------------------ 58
cho-miR-153/1-58      GCAUAGUCACAAAAGUGAUC------------------ 58
adu-miR-153/1-58      GCAUAGUCACCAAAGUGAUC------------------ 58
```

Figura 18: Estrutura secundária de precursores de miRNA-153 e ortólogos; cgi: *Crassostrea gigas*, cho: *Crassostrea hongkongensis,* bgl: *Biomphalaria glabrata,* rau: *Radix auricularia lny: Lanistes nyassanus,* pmc: *Pomacea maculata.*

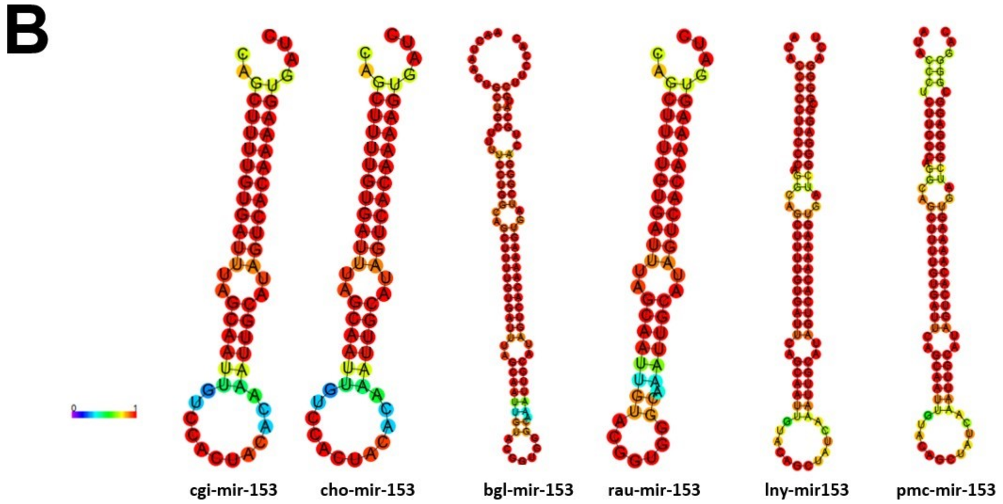
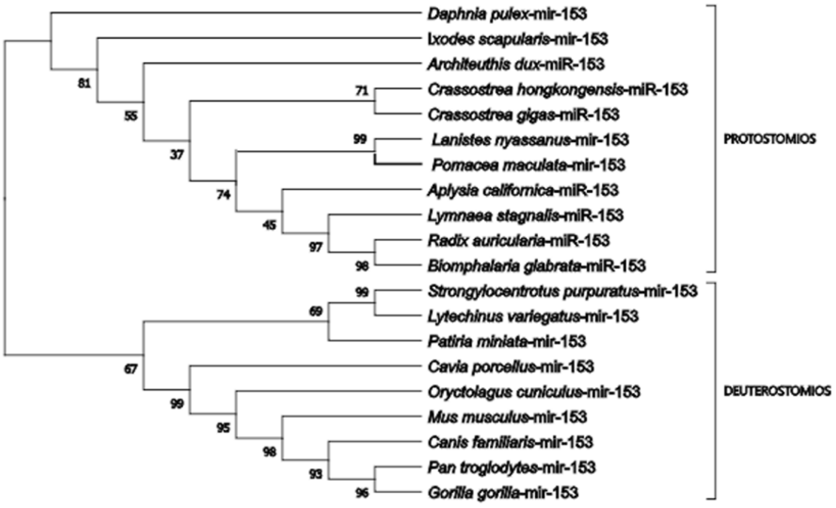


Figura 19: Distribuição filogenética de mir-153 (bilatéria-específico).

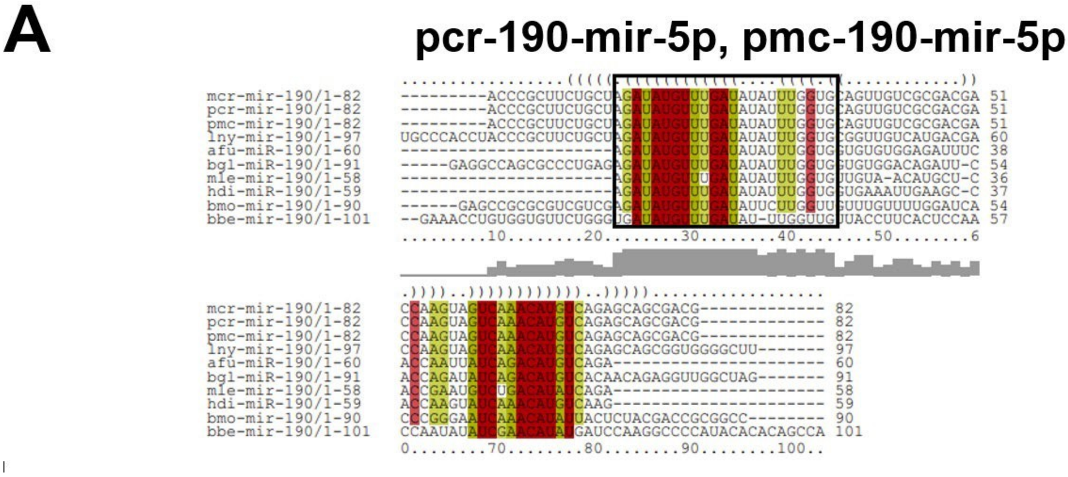


Fonte: Dados do autor

4.3.6 mir-190

A família dos precursores de mir-190 foi encontrada nos genomas de *P. maculata* e *P. canaliculata.* Observou-se um miRNA maduro por espécie na extremidade 5’ (Figura 20). Mais uma vez, verifica-se à similaridade das estruturas secundárias e primarias dos precursores entre os organismos da família Ampullariidae (Figuras 20 e 21). O mir-190 foi encontrado ao longo da cadeia evolutiva sendo classificado como um miRNA bilatéria específico. Desse modo, representantes dos grupos Lophotrocozoa, Ecdysozoa e Deuterostômios estão presentes na árvore filogenética (Figura 22).

Figura 20: Alinhamento de precursores de miRNA-190 com ortólogos.



```
                     .................((((((((((((((((((....((((.(.............))
mcr-mir-190/1-82     ---------ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGA 51
pcr-mir-190/1-82     ---------ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGA 51
pmc-mir-190/1-82     ---------ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGA 51
lny-mir-190/1-97     UGCCCACCUACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCGGUUGUCAUGACGA 60
afu-miR-190/1-60     ----------------------AGAUAUGUUUGAUAUAUUUGGUGGUGUGUGGAGAUUUC 38
bgl-miR-190/1-91     -----GAGGCCAGCGCCCUGAGAGAUAUGUUUGAUAUAUUUGGUGGUGUGGACAGAUU-C 54
mle-mir-190/1-58     ----------------------AGAUAUGUUUGAUAUAUUUGGUGUUGUA-ACAUGCU-C 36
hdi-miR-190/1-59     ----------------------AGAUAUGUUUGAUAUAUUUGGUGGUGAAAUUGAAGC-C 37
bmo-mir-190/1-90     ------GAGCCGCGCGUCGUCGAGAUAUGUUUGAUAUUCUUGGUUGUUUGUUUUGGAUCA 54
bbe-mir-190/1-101    --GAAACCUGUGGUGUUCUGGGUGAUAUGUUUGAUAU-UUGGUUGUUACCUUCACUCCAA 57
                     .........10........20........30........40........50.......6

                     .))))..))))))))))))..)))))..................
mcr-mir-190/1-82     CCAAGUAGUCAAACAUGUCAGAGCAGCGACG------------- 82
pcr-mir-190/1-82     CCAAGUAGUCAAACAUGUCAGAGCAGCGACG------------- 82
pmc-mir-190/1-82     CCAAGUAGUCAAACAUGUCAGAGCAGCGACG------------- 82
lny-mir-190/1-97     CCAAGUAGUCAAACAUGUCAGAGCAGCGGUGGGGCUU------- 97
afu-miR-190/1-60     ACCAAUUAUCAGACAUGUCAGA---------------------- 60
bgl-miR-190/1-91     ACCAGAUAUCAGACAUGUCACAACAGAGGUUGGCUAG------- 91
mle-mir-190/1-58     ACCGAAUGUCUGACAUAUCAGA---------------------- 58
hdi-miR-190/1-59     ACCAAGUAUCAAACAUGUCAAG---------------------- 59
bmo-mir-190/1-90     CCCGGGAAUCAAACAUAUUACUCUACGACCGCGGCC-------- 90
bbe-mir-190/1-101    CCAAUAUAUCGAACAUAUGAUCCAAGGCCCCAUACACACAGCCA 101
                     0........70........80........90........100..
```

Figura 21: Estrutura secundária de precursores de miRNA-190 e ortólogos; hdi: *Haliotis discus hannai*, afu: *Achatina fulica, lny: Lanistes nyassanus, mcr: Marisa cornuarietis, pcr: Pomacea canaliculata,* pmc: *Pomacea maculata,* bgl: *Biomphalaria glabrata*, mle: *Melibe leonina*

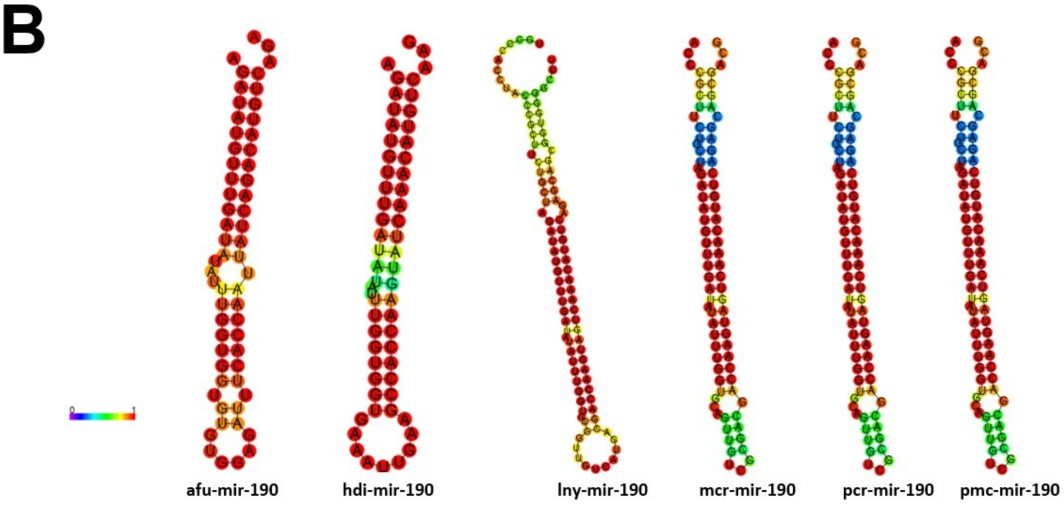
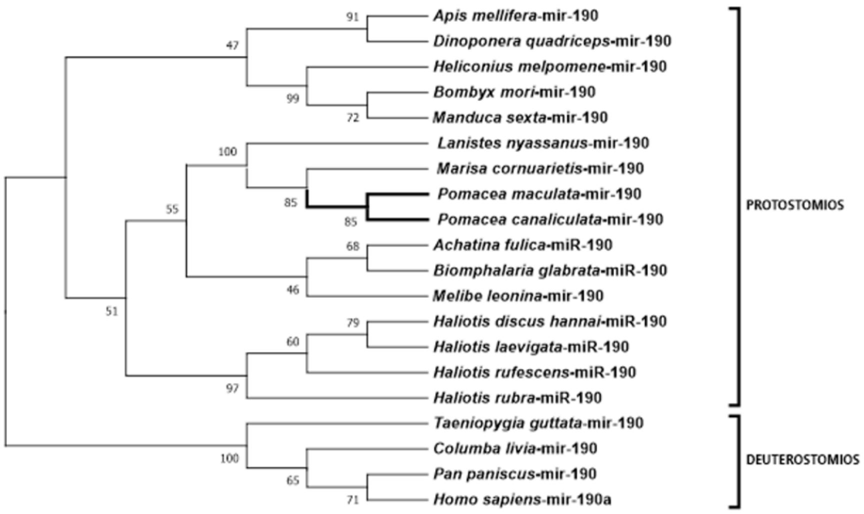


Figura 22: Distribuição filogenética de mir-190 (bilatéria-específico).



Fonte: Dados do autor

4.4 miRNA PROTÔSTOMA ESPECÍFICO

4.4.1 mir-bantam

O mir-bantam está relacionado ao crescimento de células cerebrais e homeostase (LAM *et al*., 2014; LI; PADGETT, 2012). Em *Drosophila*, mir-bantam funciona na manutenção do ritmo circadiano, estimulam a proliferação celular e reduzem a morte celular programada (BRENNECKE *et al*., 2003; KADENER *et al*., 2009). Na esquistossomose este miRNA de trematódeos pode estar envolvido em processos patológicos da doença, sendo mir-bantam detectado na corrente sanguínea de hospedeiros. Há possibilidade de utilizá-lo como possível biomarcador para diagnóstico desta doença (ZHU; LIU; CHENG, 2014). Em um estudo em *B. glabrata* foi possível identificar bgl-bantam, que é altamente conservado em metazoários. Foi sugerido que bgl-bantam atue na regulação do metabolismo de caramujos, uma vez que reconhece alvos com função redox e com ação regulatória sobre o complexo ubiquitina-proteína (QUEIROZ et al., 2020). O alinhamento com os precursores ortólogos de *P. canaliculata e P. maculata* revela alta similaridade com os miRNAs maduros, com 100% de identificação desta espécie com alguns dos seus miRNAs ortólogos (Figura 23). Além disso, há uma similaridade entre as estruturas secundárias dos precursores de miRNAs analisados e seus ortólogos (Figura 24). A árvore filogenética foi representada pelos clados Lophotrocozoa e Ecdysozoa (Figura 25). Estes dados corroboram para identificação de mir-bantam (5p e 3p) nas espécies em estudo.

Figura 23: Alinhamento de precursores miRNA-bantam e ortólogos.

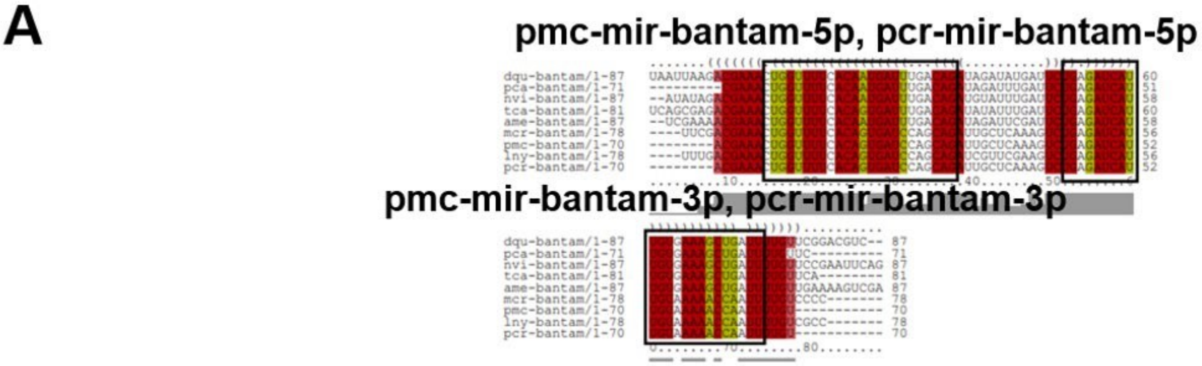


Figura 24: B) Estrutura secundária de precursores de miRNA-bantam e ortólogos; dqu: *Dinoponera quadriceps,* nvi*: Nasonia vitripennis*, *lny: Lanistes nyassanus, mcr: Marisa cornuarietis, pcr: Pomacea canaliculata,* pmc: *Pomacea maculata.*

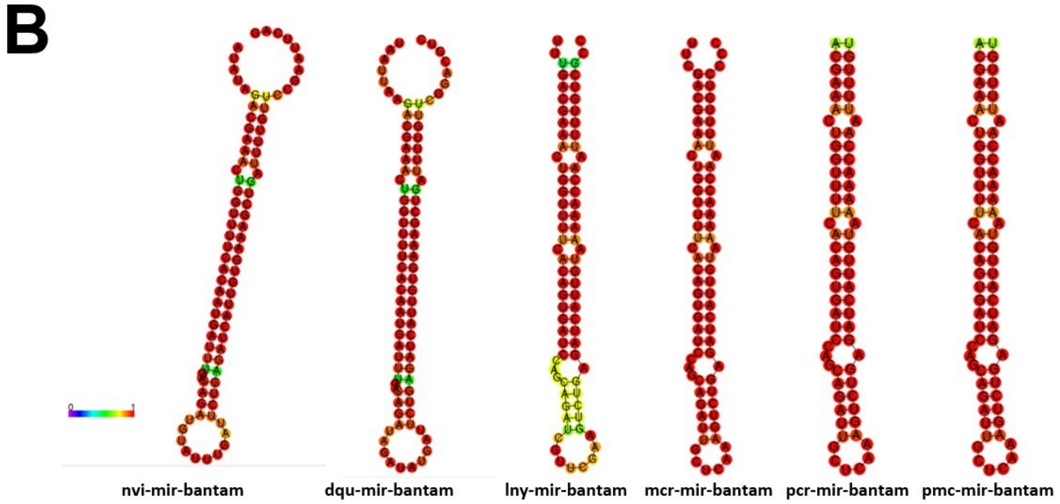
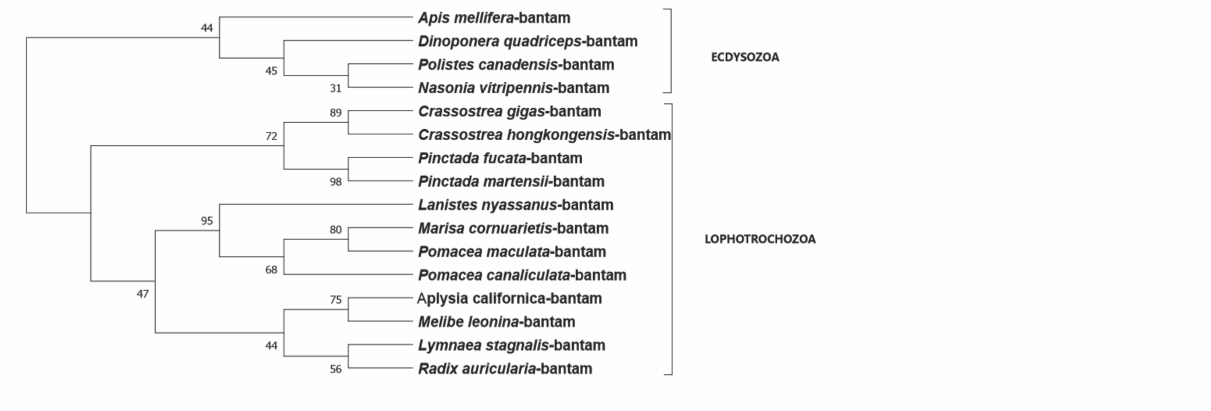


Figura 25: Distribuição filogenética de miR-bantam (protostômio específico).



Fonte: Dados do autor

4.4.2 mir-1175

Há registros de mir-1175 na literatura em mosquitos da ordem insecta como: *Aedes aegypti, Anopheles stephensi, Anopheles gambia*e, *Tinissa amboinensis* (LI *et al*., 2009), conforme observado na figura 28 deste trabalho. Diferentes níveis de expressão de miRNAs são observados em machos e fêmeas da espécie *Anopheles anthropophagus*. No caso do mir- 1175, ocorre uma diminuição de expressão de cerca de 2 vezes menor em machos quando comparado a fêmeas (LIU *et al*., 2014). Em moluscos, em um estudo no gastrópode marinho *Rapana venosa,* analisa os perfis de expressão de miRNAs na metamorfose da espécie. Um total de 195 miRNAs diferentemente expressos foram obtidos neste processo, dentre estes o mir-1175 foi regulado positivamente (SONG *et al*., 2017). Além disso, a família de mir-1175 foi encontrada no genoma de 34 espécies de moluscos (HUANG et al., 2021), dentre estes cefalópodes, bivalves e gastrópodes (Figura 28). Isso corrobora para a identificação do precursor de mir-1175 em *P. canaliculata*, com a presença de dois miRNAs maduros, 5p e 3p (Figura 26). A estrutura secundária do precursor de *P. canaliculata* apresenta alta similaridade com os ortólogos, principalmente com representantes da família Ampullariidae, *L. nyassanus* e *M. cornuarietis* (Figura 27).

Figura 26: Alinhamento de precursores de miRNA-1175 e ortólogos.

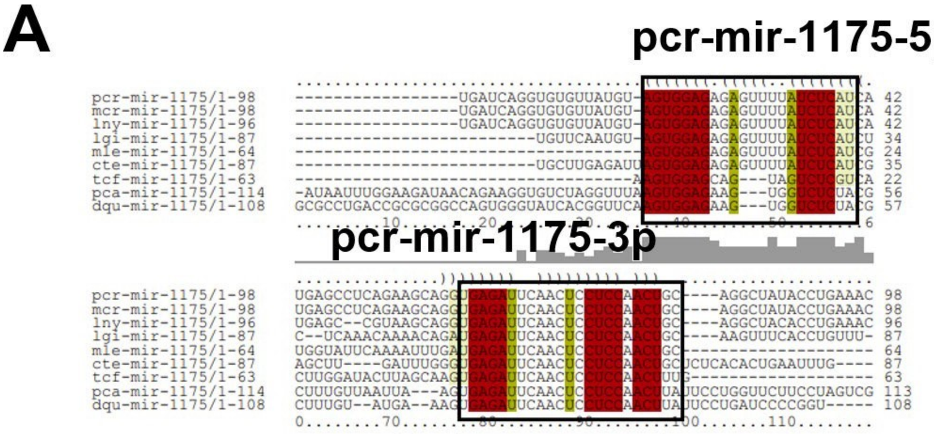


Figura 27: Estrutura secundária de precursores de miRNA-1175 e ortólogos; mle*: Melibe leonina*, cte: *Capitella teleta*, lgi: *Lottia gigantea*, *lny: Lanistes nyassanus, mcr: Marisa cornuarietis, pcr: Pomacea canaliculata.*

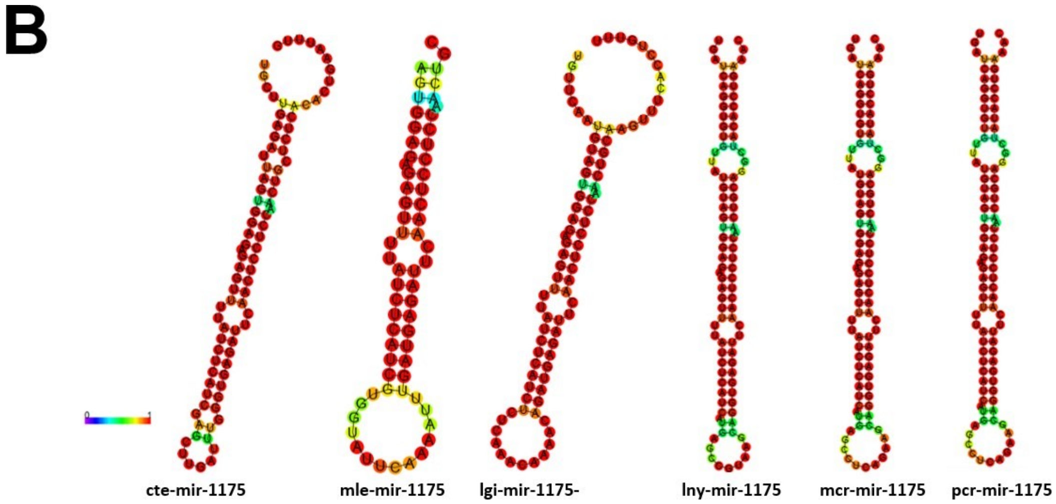
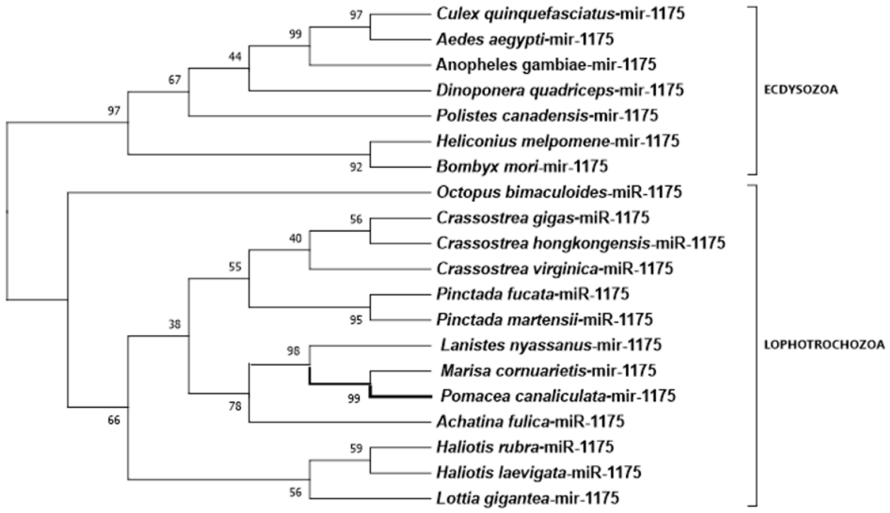


Figura 28: Distribuição filogenética de miR-1175 (protostômio específico).



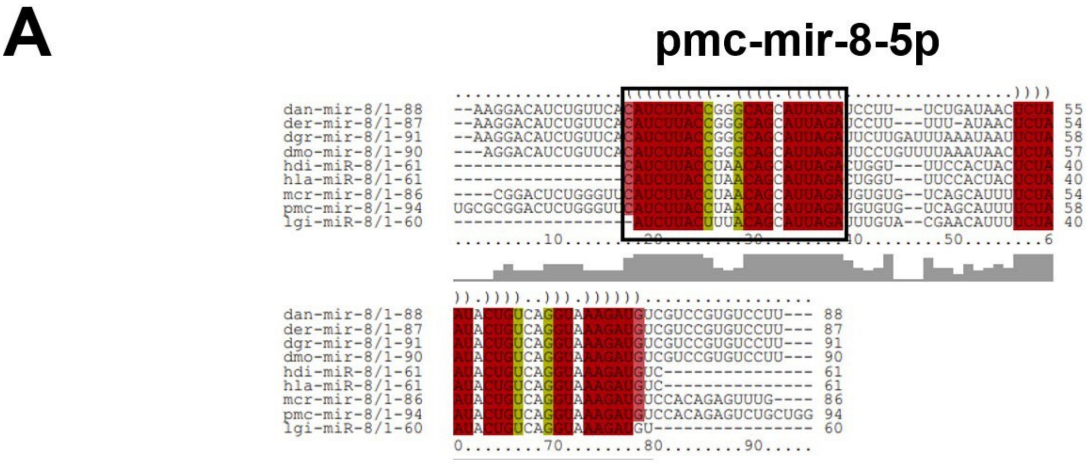
Fonte: Dados do autor

4.4.3 mir-8

O mir-8 assim como o mir-33 está potencialmente associado à regulação gênica no bivalve *T. granosa* quando exposto a níveis tóxicos de cádmio (Cd) (BAO *et al*., 2014). A família de genes de mir-8 foi encontrada no genoma das 2 espécies Ampullariidae analisadas, com a presença das isoformas mir-8-1 e mir-8-2 em *P. canaliculata.* Para *P. maculata* foi observado o mir-8 com a presença de um miRNA maduro, 5p (Figura 29). A estrutura secundária do precursor de *P. maculata* apresentou alta similaridade com o precursor de *M.*

cornuarietis (Figura 30). A árvore filogenética foi representada pelos clados Lophotrocozoa e Ecdysozoa (Figura 31). Estes dados corroboram para identificação de mir-8 (5p) em *P. maculata.*

Figura 29: Alinhamento de precursores de miRNA-8 e ortólogos.



```
               .................((((((((..((((.((((((..................))))
dan-mir-8/1-88 --AAGGACAUCUGUUCACAUCUUACCGGGCAGCAUUAGAUCCUU---UCUGAUAACUCUA 55
der-mir-8/1-87 --AAGGACAUCUGUUCACAUCUUACCGGGCAGCAUUAGAUCCUU---UUU-AUAACUCUA 54
dgr-mir-8/1-91 --AAGGACAUCUGUUCACAUCUUACCGGGCAGCAUUAGAUUCUUGAUUUAAAUAAUUCUA 58
dmo-mir-8/1-90 ---AGGACAUCUGUUCACAUCUUACCGGGCAGCAUUAGAUUCCUGUUUUAAAUAACUCUA 57
hdi-miR-8/1-61 -----------------CAUCUUACCUAACAGCAUUAGACUGGU---UUCCACUACUCUA 40
hla-miR-8/1-61 -----------------CAUCUUACCUAACAGCAUUAGACUGGU---UUCCACUACUCUA 40
mcr-mir-8/1-86 ----CGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUG--UCAGCAUUUUCUA 54
pmc-mir-8/1-94 UGCGCGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUG--UCAGCAUUUUCUA 58
lgi-miR-8/1-60 ------------------AUCUUACUUUACAGCAUUAGAUUUGUA--CGAACAUUUUCUA 40
               .........10.........20.........30.........40.........50.........6

               )).))))..))).)))))).................
dan-mir-8/1-88 AUACUGUCAGGUAAAGAUGUCGUCCGUGUCCUU--- 88
der-mir-8/1-87 AUACUGUCAGGUAAAGAUGUCGUCCGUGUCCUU--- 87
dgr-mir-8/1-91 AUACUGUCAGGUAAAGAUGUCGUCCGUGUCCUU--- 91
dmo-mir-8/1-90 AUACUGUCAGGUAAAGAUGUCGUCCGUGUCCUU--- 90
hdi-miR-8/1-61 AUACUGUCAGGUAAAGAUGUC--------------- 61
hla-miR-8/1-61 AUACUGUCAGGUAAAGAUGUC--------------- 61
mcr-mir-8/1-86 AUACUGUCAGGUAAAGAUGUCCACAGAGUUUG---- 86
pmc-mir-8/1-94 AUACUGUCAGGUAAAGAUGUCCACAGAGUCUGCUGG 94
lgi-miR-8/1-60 AUACUGUCAGGUAAAGAUGU---------------- 60
               0.........70.........80.........90.....
```

Figura 30: Estrutura secundária: precursores de miRNA-8 e ortólogos;dgr: *Drosophila grimshawi*: dan: *Drosophila ananassae,* der*: Drosophila erecta,* hdi: *Haliotis discus hannai, mcr: Marisa cornuarietis,* pmc: *Pomacea maculata.*

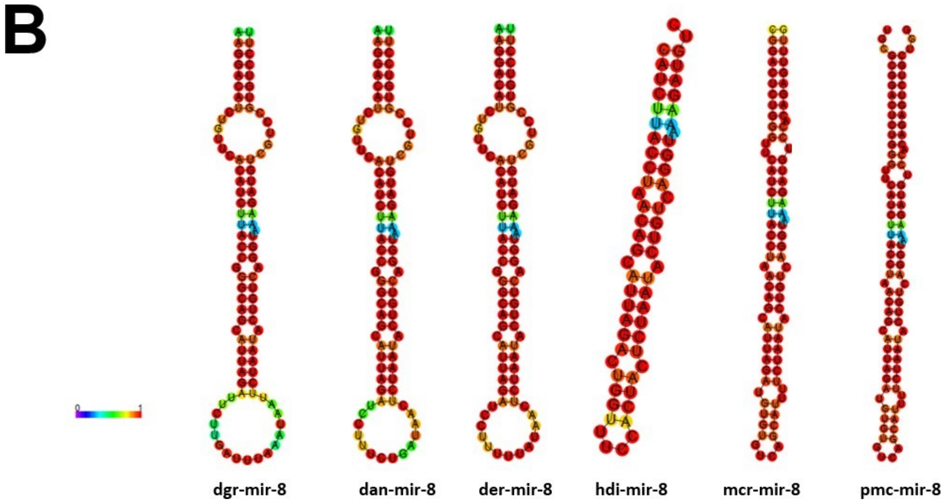
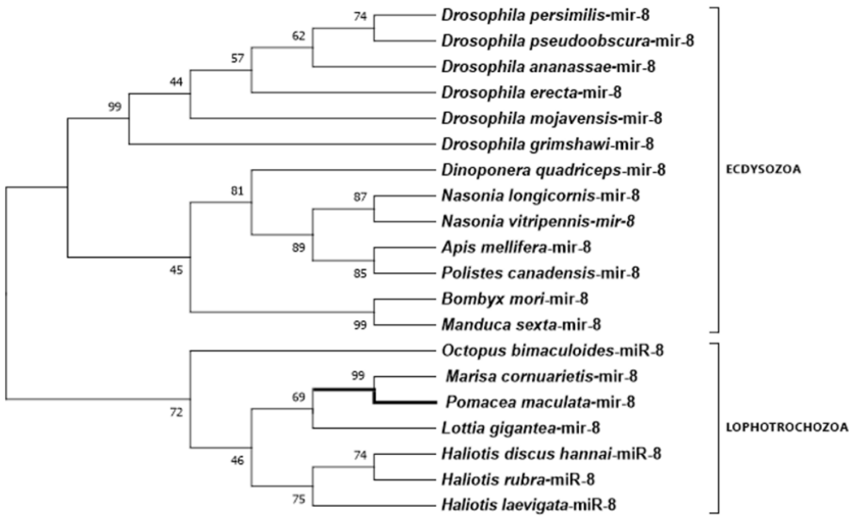


Figura 31: Análise filogenética de miR-8 (protostômio específico).

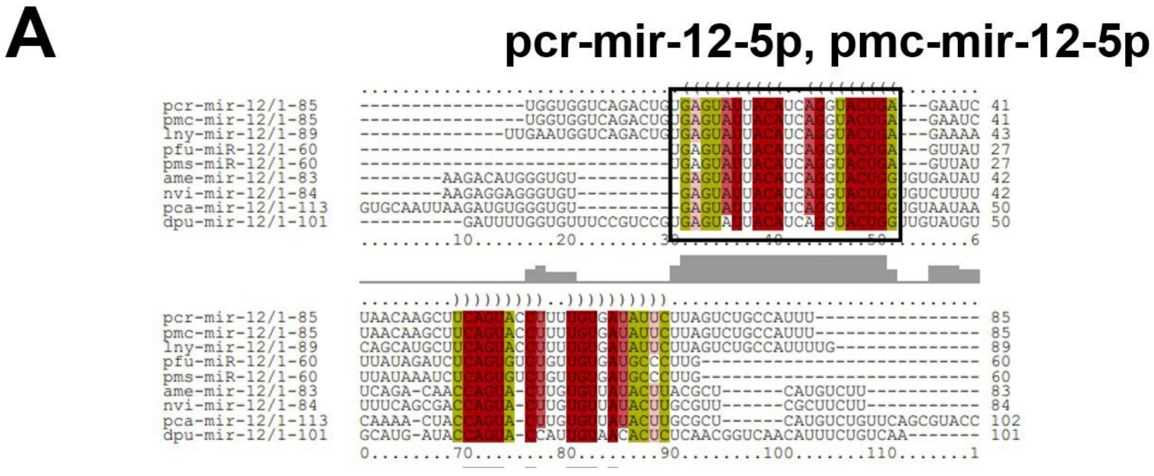


Fonte: Dados do autor

4.4.4 mir-12

Assim como o mir-8, o mir-12 está relacionado potencialmente ao desenvolvimento do crustáceo *Daphnia pulex*. A regulação positiva de mir-8 coincide com mudanças morfológicas e crescimento na espécie (CHEN et al., 2014b). Já em moluscos, a família dos precursores de mir-12 foi identificada no organismo *B. glabrata* (QUEIROZ *et al*., 2020), próximo evolutivamente aos organismos do gênero *Pomacea* analisados. Os precursores de *B. glabrata, P. canaliculata* e *P. maculata* (Figura 32) apresentaram o mir-12-5p para cada espécie. A estrutura secundária dos pré-miRNAs de *P. canaliculata* e *P. maculata* apresentaram formato canônico característico dos precursores, intrinsecamente semelhantes entre si (Figura 33). A distribuição filogenética foi verificada entre os clados Ecdysozoa e Lophotrocozoa, sendo este representado por moluscos das classes cefalópode, bivalve e gastrópode (Figura 34).

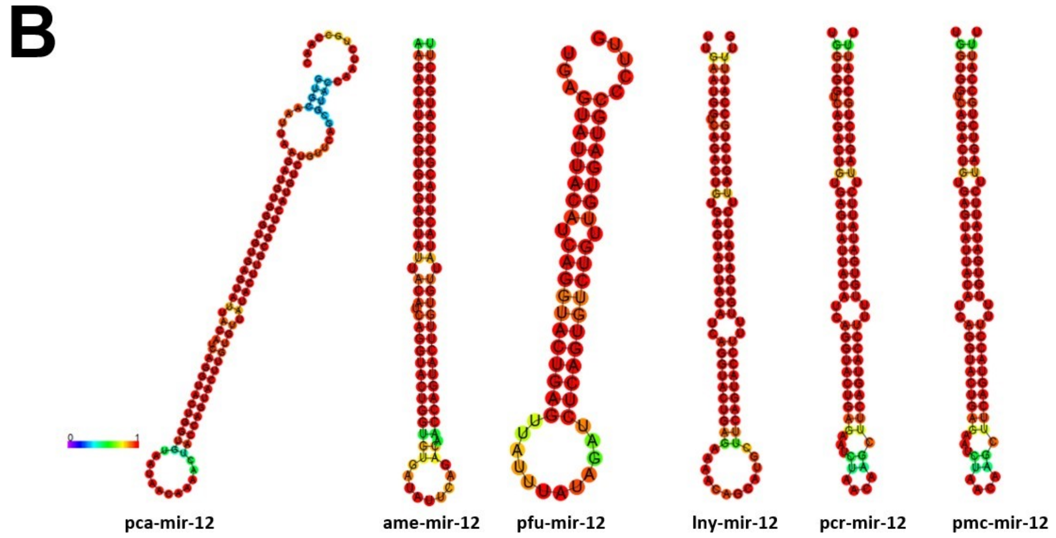
Figura 32: Alinhamento de precursores de miRNA-12 e ortólogos.



```
                     ...............................((((((((((..(((((((((........
pcr-mir-12/1-85      ----------------UGGUGGUCAGACUGUGAGUAUUACAUCAGGUACUGA---GAAUC 41
pmc-mir-12/1-85      ----------------UGGUGGUCAGACUGUGAGUAUUACAUCAGGUACUGA---GAAUC 41
lny-mir-12/1-89      --------------UUGAAUGGUCAGACUGUGAGUAUUACAUCAGGUACUGA---GAAAA 43
pfu-miR-12/1-60      ------------------------------UGAGUAUUACAUCAGGUACUGA---GUUAU 27
pms-miR-12/1-60      ------------------------------UGAGUAUUACAUCAGGUACUGA---GUUAU 27
ame-mir-12/1-83      --------AAGACAUGGGUGU----------GAGUAUUACAUCAGGUACUGGUGUGAUAU 42
nvi-mir-12/1-84      --------AAGAGGAGGGUGU----------GAGUAUUACAUCAGGUACUGGUGUCUUUU 42
pca-mir-12/1-113     GUGCAAUUAAGAUGUGGGUGU----------GAGUAUUACAUCAGGUACUGGUGUAAUAA 50
dpu-mir-12/1-101     ----------GAUUUUGGUGUUUCCGUCCGUGAGUAUUACAUCAGGUACUGGUUGUAUGU 50
                     .........10........20........30........40........50........6

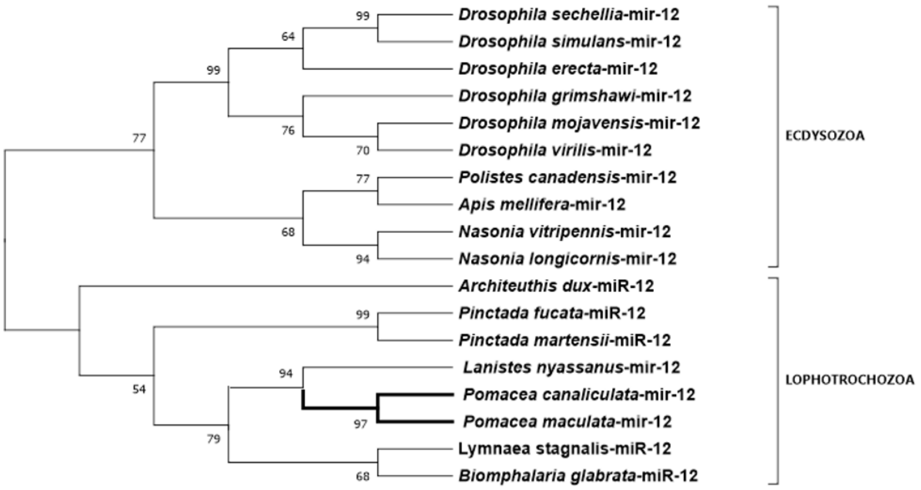
                     .........)))))))))..))))))))))..............................
pcr-mir-12/1-85      UAACAAGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUU---------------- 85
pmc-mir-12/1-85      UAACAAGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUU---------------- 85
lny-mir-12/1-89      CAGCAUGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUUUG-------------- 89
pfu-miR-12/1-60      UUAUAGAUCUCAGUGUCUGUUGUGAUGCCCUUG--------------------------- 60
pms-miR-12/1-60      UUAUAAAUCUCAGUGUCUGUUGUGAUGCCCUUG--------------------------- 60
ame-mir-12/1-83      UCAGA-CAACCAGUA-CUUGUGUUAUACUUACGCU------CAUGUCUU----------- 83
nvi-mir-12/1-84      UUUCAGCGACCAGUA-CUUGUGUUAUACUUGCGUU------CGCUUCUU----------- 84
pca-mir-12/1-113     CAAAA-CUACCAGUA-CUUGUGUUAUACUUGCGCU------CAUGUCUGUUCAGCGUACC 102
dpu-mir-12/1-101     GCAUG-AUACCAGUA-CCAUUGUAACACUCUCAACGGUCAACAUUUCUGUCAA------- 101
                     0........70........80........90........100.......110.......1
```

Figura 33: Estrutura secundária: precursores de miRNA-12 e ortólogos: ame: *Apis melífera,* pca*: Polistes canadensis* pfu*: Pinctada fucata,* lny: *Lanistes nyassanus,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*



pca-mir-12 ame-mir-12 pfu-mir-12 lny-mir-12 pcr-mir-12 pmc-mir-12

Figura 34: Análise filogenética de miR-12 (protostômio específico).



Fonte: dados do autor.

4.4.5 mir-750

A família do mir-750 apresentou o precursor mir-750 nos organismos *P. canaliculata* e *P. maculata* e os miRNAs maduros pcr-mir-750-3p e pmc-miR-750-3p (Figura 35). O alinhamento com ortólogos e estruturas secundárias revelaram alta preservação nucleotídica entre espécies do gênero *Pomacea* e ortólogos (Figuras 35 e 36). Na distribuição filogenética foi observado o clado Ecdysozoa, representado por artrópodes e nematódeo e o clado Lophotrocozoa, constituído exclusivamente por moluscos (Figura 37).

Figura 35: Alinhamento de precursores de miRNA-750 e ortólogos.

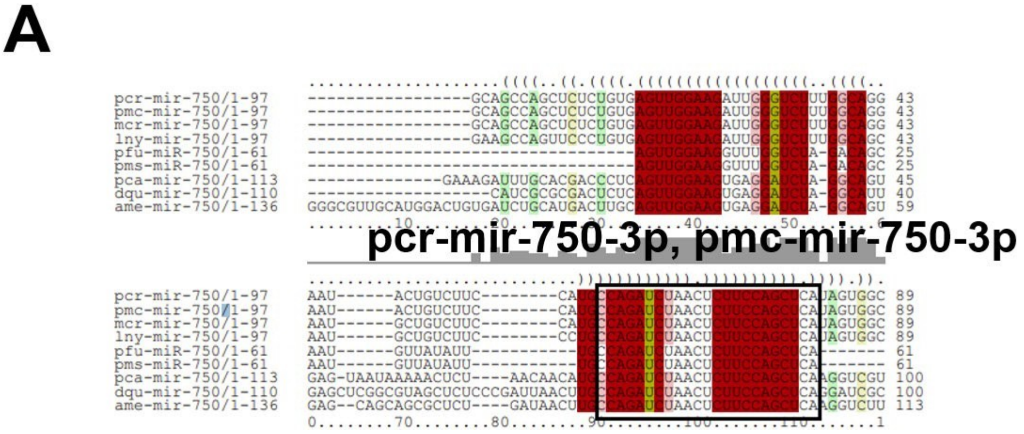


Figura 36: Estrutura secundária: precursores de miRNA-bantam e ortólogos; dqu: *Dinoponera quadriceps,* pca: *Polistes canadensis*, lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*

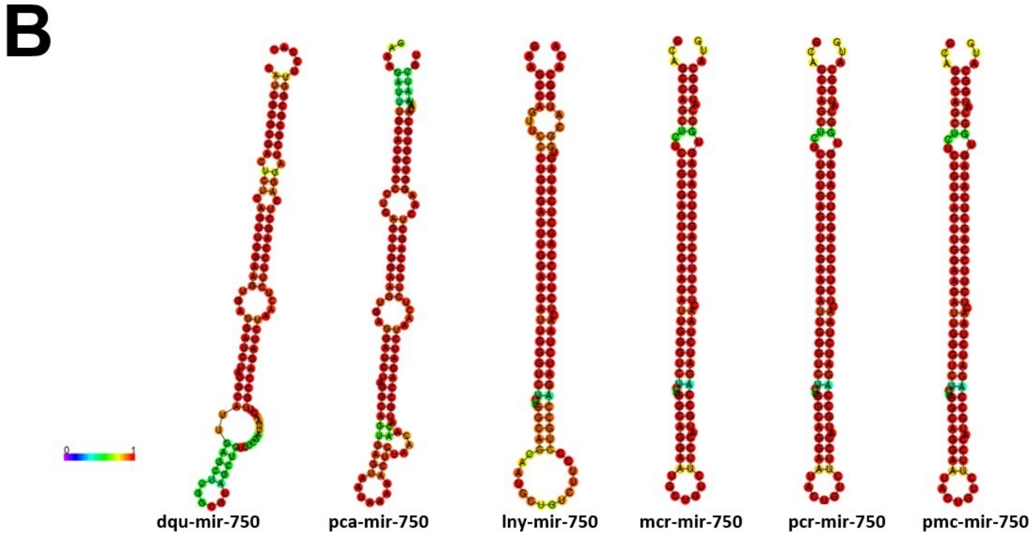
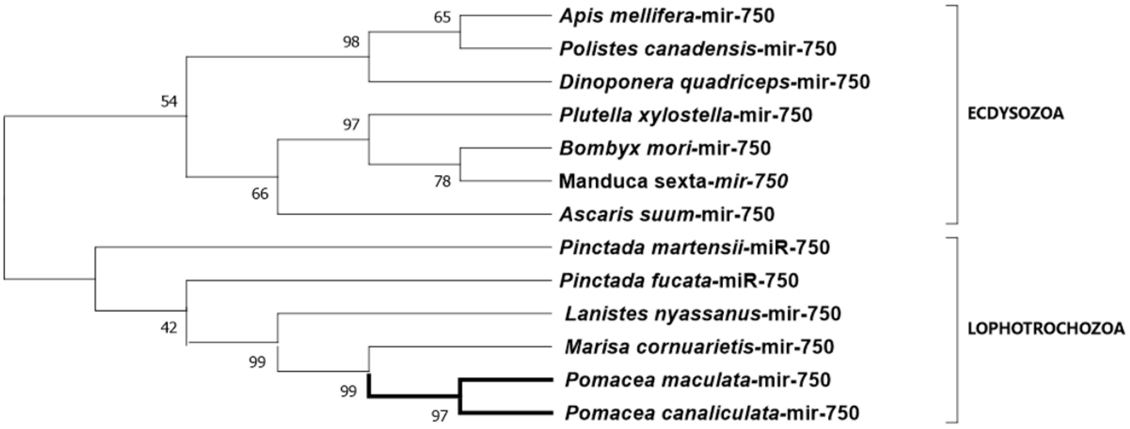


Figura 37: Análise filogenética de miR-750 (protostômio específico).



Fonte: dados do autor.

4.5 miRNA LOPHOTROCOZOA ESPECÍFICO

4.5.1 mir-745a

A família de mir-745a apresentou um miRNA maduro nas sequencias de cada precursor das espécies *P. canaliculata* e *P. maculata*, extremidade 3’ (Figura 38). Frente aos seus ortólogos houve conservação da sequência de nucleotídeos bem como da estrutura secundária (Figuras 38 e 39). Há registros da família mir-745a em 28 espécies de moluscos, dentre estes organismos próximos evolutivamente as espécies analisadas; *B. glabrata, Aplysia californica e*

Lottia gigantea (HUANG et al., 2021). Foi verificado uma distribuição filogenética formada por animais dos filos dos moluscos, platelmintos e anelídeos (Figura 40).

Figura 38: Alinhamento de precursores de miRNA-745a com seus ortólogos.

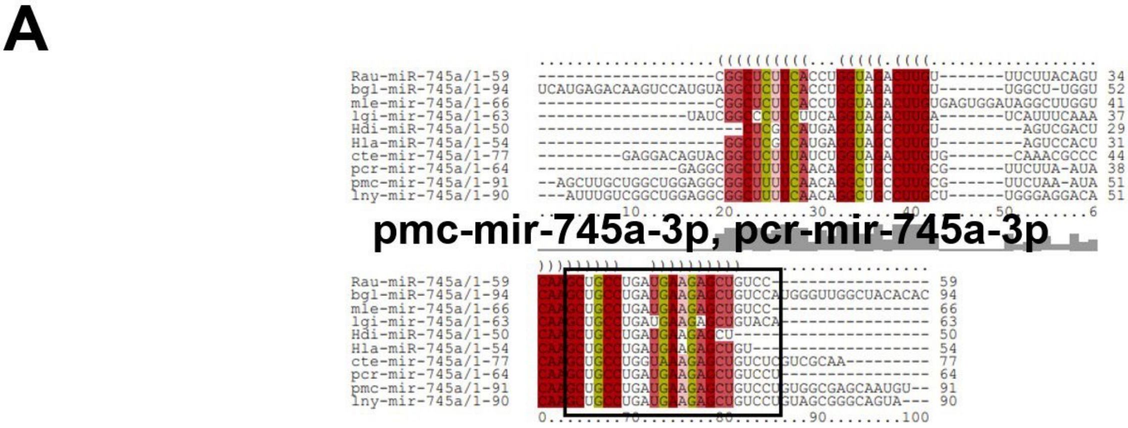


Figura 39: Estrutura secundária dos precursores de miRNA-745a e ortólogos; lgi: *Lottia gigantea*, bgl: *Biomphalaria glabrata*, cte: *Capitella teleta*, *lny: Lanistes nyassanus, pcr: Pomacea canaliculata,* pmc: *Pomacea maculata.*

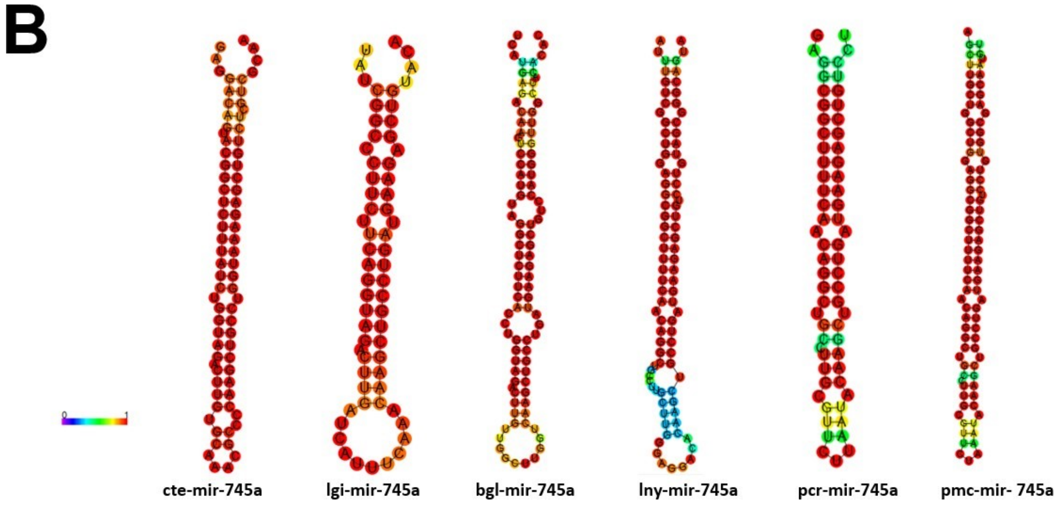
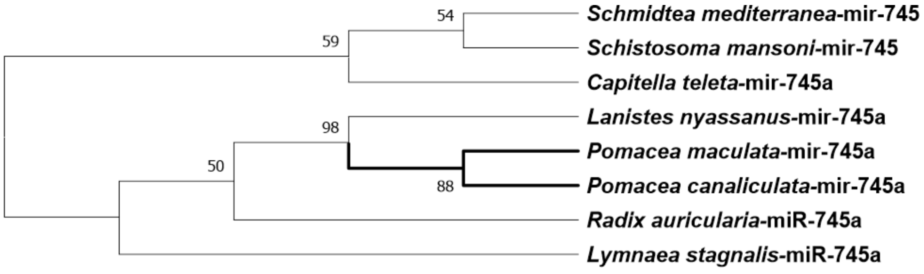


Figura 40: Distribuição filogenética de miR-754a (lophotrochozoa-específico).



Fonte: dados do autor.

4.5.2 mir-1992

A família dos genes de mir-1992 foi identificada apenas no genoma da espécie de *P. canaliculata*, com a presença de dois miRNAs maduros, 3p e 5p (Figura 41). A estrutura secundária do precursor de *P. canaliculata* revelou alta similaridade com seus ortólogos (Figura 42). O mir-1992 revelou expressão diferencial nos testículos e ovários de *Crassostrea hongkongensis*, sendo altamente expressos (WEI *et al*., 2019). Em gastrópodes, o mir-1992 foi identificado em 9 espécies de moluscos (HUANG et al., 2021). Assim, na distribuição filogenética foi representado em sua maior parte por moluscos gastrópodes e também por cefalópodes, além da presença de platelmintos (Figura 42).

Figura 41: Alinhamento de precursores miRNA-1992 com seus ortólogos, em destaque miRNAs maduros

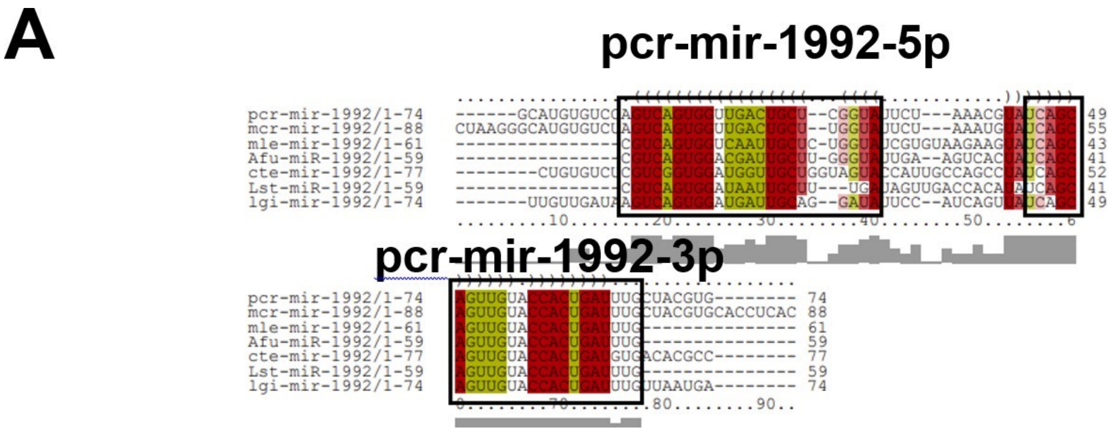


Figura 42: Estrutura secundária dos precursores de miRNA-1992 e ortólogos; lgi: *Lottia gigantea*, mle: *Melibe leonina*, cte: *Capitella teleta*, mcr*: Marisa cornuarietis,* pcr*: Pomacea canaliculata.*

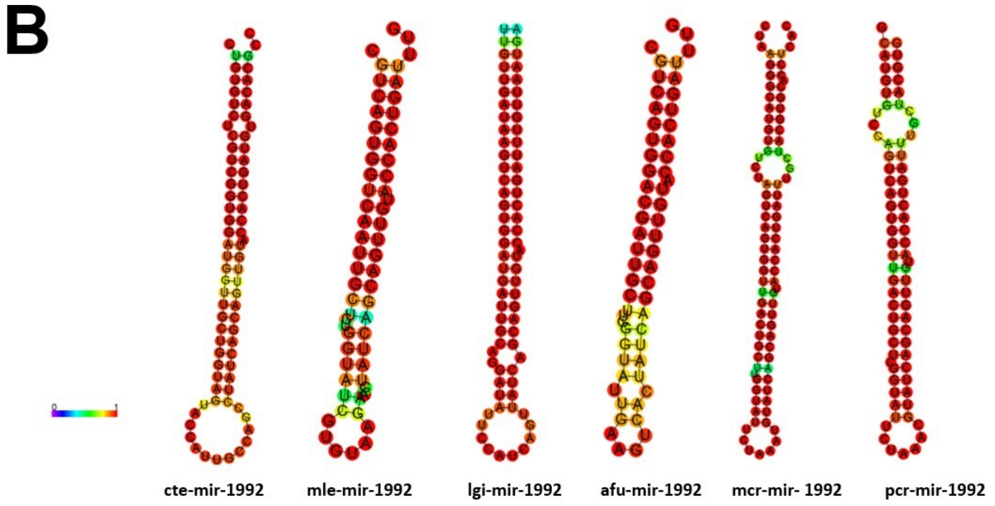
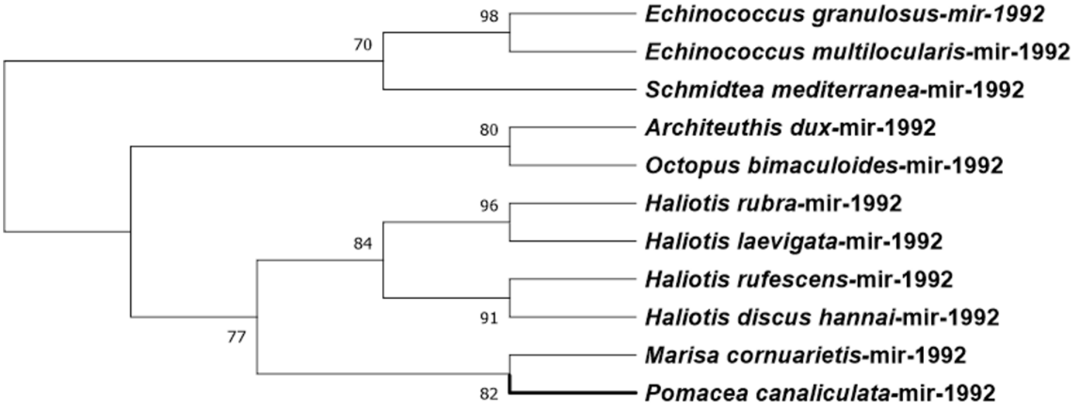


Figura 43: Distribuição filogenética de miR-1992 (lophotrochozoa-específico). Em destaque a espécie: *Pomacea canaliculata.*

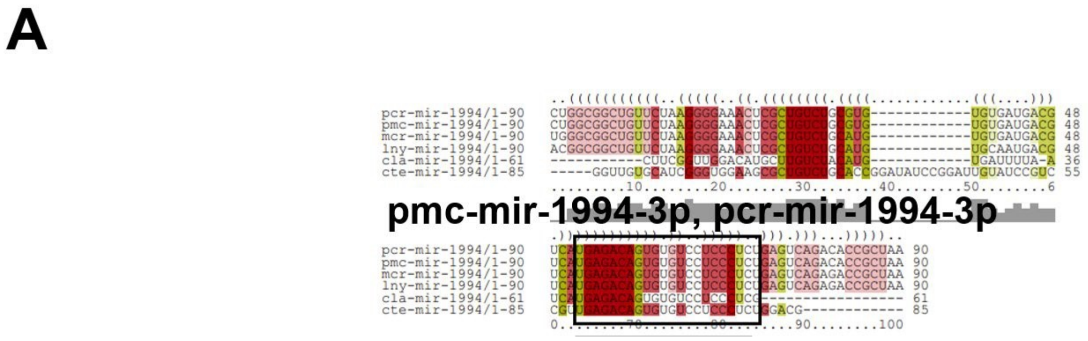


Fonte: dados do autor.

4.5.3 mir-1994

A família do mir-1994 é composta pelos precursores mir-1994 e mir-1994a, para ambas as espécies da família Ampullariidae. *P. canaliculata* e *P. maculata,* apresentam um miRNA maduro (3p) para cada precursor mir-1994 (Figura 44). Comparado aos seus ortólogos as espécies analisadas apresentaram conservação da sequência de nucleotídeos bem como da estrutura secundária (Figuras 44 e 45). A distribuição filogenética observou-se a presença de animais dos filos como Nemertea, Mollusca e Annelida (Figura 46).

Figura 44: Alinhamento de precursores de miRNA-1994 com ortólogos.



```
                      ..((((((((((..(((((..((.((((((((.((((.............(((....)))
pcr-mir-1994/1-90     CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUG------------UGUGAUGACG 48
pmc-mir-1994/1-90     CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUG------------UGUGAUGACG 48
mcr-mir-1994/1-90     UGGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUG------------UGUGAUGACG 48
lny-mir-1994/1-90     ACGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUG------------UGCAAUGACG 48
cla-mir-1994/1-61     -----------CUUCGGUUGGACAUGCUUGUCUACAUG------------UGAUUUUA-A 36
cte-mir-1994/1-85     -----GGUUGUGCAUCGGGUGGAAGCGCUGUCUGCACCGGAUAUCCGGAUUGUAUCCGUC 55
                      .........10........20........30........40........50........6

                      .))))))))))))..)))..)))))).))).)))...)))))))..
pcr-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCGUCUGAGUCAGACACCGCUAA 90
pmc-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCGUCUGAGUCAGACACCGCUAA 90
mcr-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCGUCUGAGUCAGAGACCGCUAA 90
lny-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCGUCUGAGUCAGAGACCGCUAA 90
cla-mir-1994/1-61     UCAUGAGACAGUGUGUCCUCCGUCG----------------- 61
cte-mir-1994/1-85     CGUUGAGACAGUGUGUCCUCCGUCUGGACG------------ 85
                      0.......70........80........90........100
```

Figura 45: Estrutura secundária dos precursores de miRNA1994 com ortólogos; cte: *Capitella teleta*, cla: *Cerebratulus lacteus,* lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*

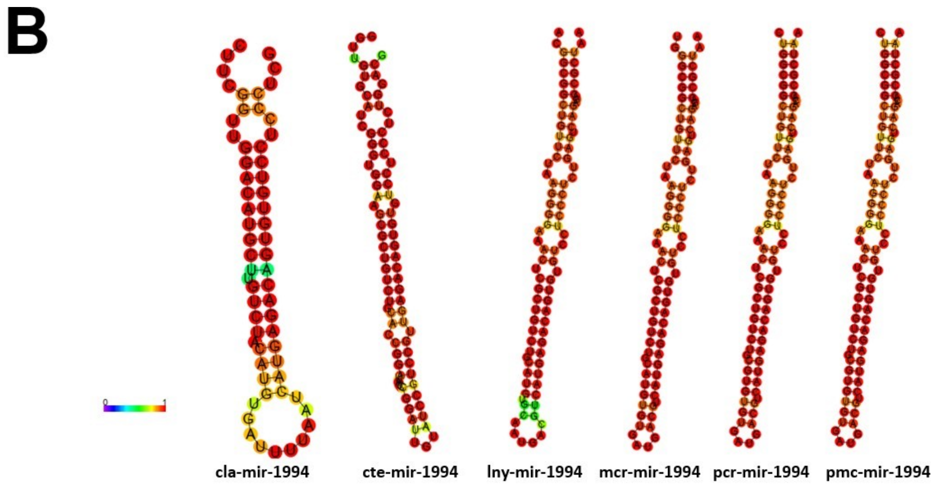
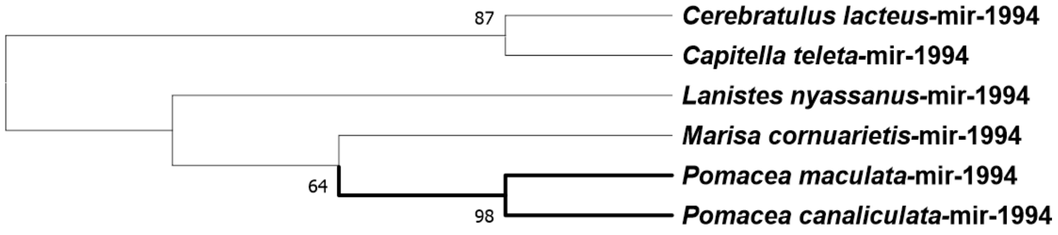


Figura 46: Distribuição filogenética de miR-1994 (lophotrochozoa-específico).



Fonte: dados do autor.

4.5.4 mir-96b

As sequências dos precursores do mir-96b apresentaram dois miRNAs maduros, 3p e 5p, nos genomas das espécies *P. canaliculata* e *P. maculata* (Figura 47). Os precursores de miR-96b para ambas as espécies demonstraram alta similaridade na estrutura secundária com os organismos gastrópodes (Figuras 48). A análise filogenética dos precursores do mir-96b foi verificada a distribuição entre as espécies em três filos como Moluscos, Anelídeos e Platelmintos (Figura 49). O mir-96b regula genes de forma pós-transcricional em Platelmintos e moluscos. No verme de água doce: *Schmidtea mediterrânea* pode ter funções na cicatrização de feridas, proliferação de neoblatos (principais responsáveis pela regeneração em planárias) e diferenciação de blastema (SASIDHARAN *et al.*, 2013). Em moluscos, o mir-96b está envolvido junto com outros miRNAs na pigmentação das conchas de *C. gigas*, induzindo a síntese de melanina, carotenóide ou tetrapirrol (FENG *et al.*, 2020). O mir-96b também foi encontrado no genoma de gastrópodes próximos evolutivamente às 2 espécies Ampullariidae, *B. glabrata* (QUEIROZ *et al.*, 2020) e *A. californica* (HUANG *et al.*, 2021). Achados na literatura científica corroboram os dados deste trabalho.

Figura 47: Alinhamento de precursores de miRNA-96b com seus ortólogos.

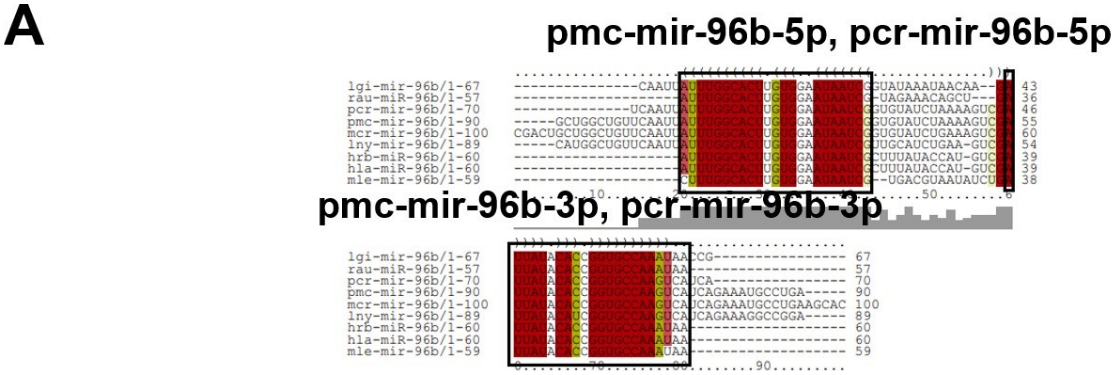


Figura 48: Estrutura secundária de precursores de miRNA-96b com seus ortólogos; mle: *Melibe leonina*, lgi*: Lottia gigantea*, lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*

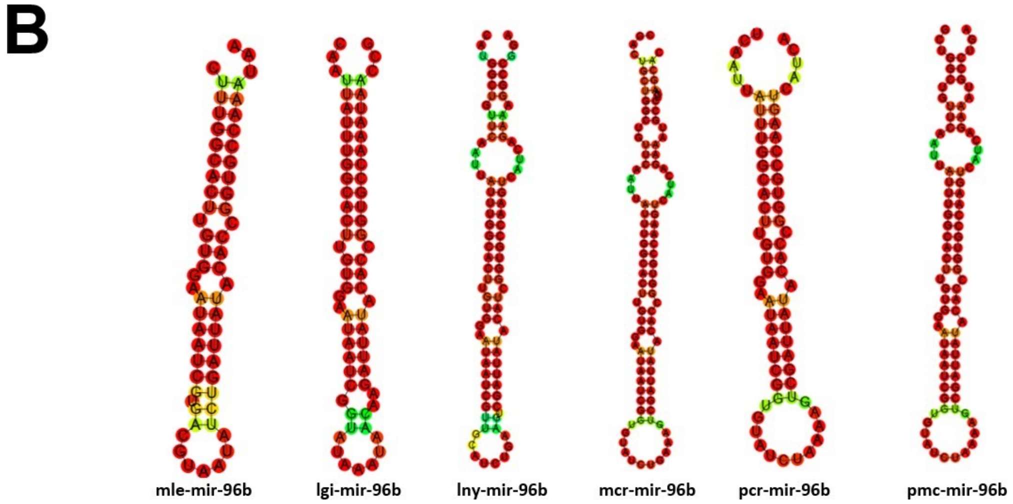
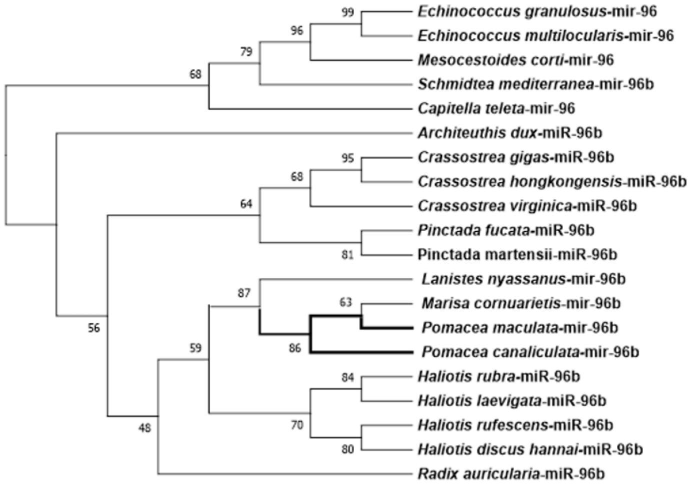


Figura 49: Análise filogenética de miR-96b (lophotrochozoa-específco).



Fonte: Dados do autor.

4.5.5 mir-1990

A família mir-1990 predita para as espécies *P. canaliculata* e *P. maculata* é composta pelos precursores pcr-mir-1990 e pcm-mir-1990 e os miRNAs maduros, 3p e 5p, em cada espécie (Figura 50). Para esta família, observou-se grande conservação da estrutura primária, bem como da estrutura secundária dos precursores de *P. canaliculata*, *P. maculata* e seus ortólogos (Figuras 50 e 51). Verificou-se ampla distribuição de mir-1990 em moluscos principalmente em 15 animais bivalves, 10 gastrópodes e 1 cefalópode. Poucos estudos descreveram o papel dos miRNAs no desenvolvimento de moluscos. O mir-1990-3p pode desempenhar um papel vital no processo de biomineralização da concha em *P. fucata*, visto que é altamente expresso nos tecidos do manto (HUANG *et al.*, 2021). O mir-1990 foi encontrado apenas em moluscos, mas a família mir-1990 é Lophotrochozoa específico pois cte-miR-1990c-3p, cte-miR-1990a e cte-miR-1990b foram identificados em *Capitella teleta* na plataforma miRBase (http://www.mirbase.org /). A ampla distribuição do mir-1990 em moluscos e o provável mecanismo de controle gênico corroboram os dados deste estudo.

Figura 50: Alinhamento de precursores de miRNA-1990 com seus ortólogos.

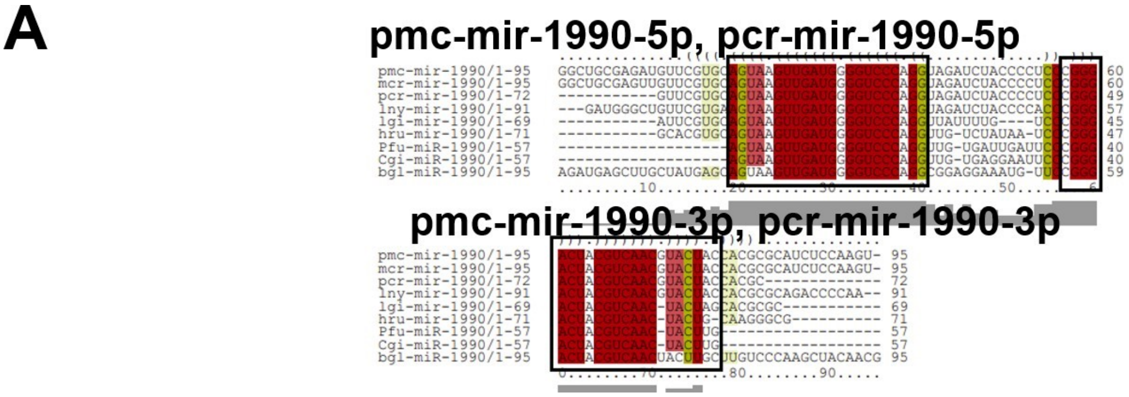
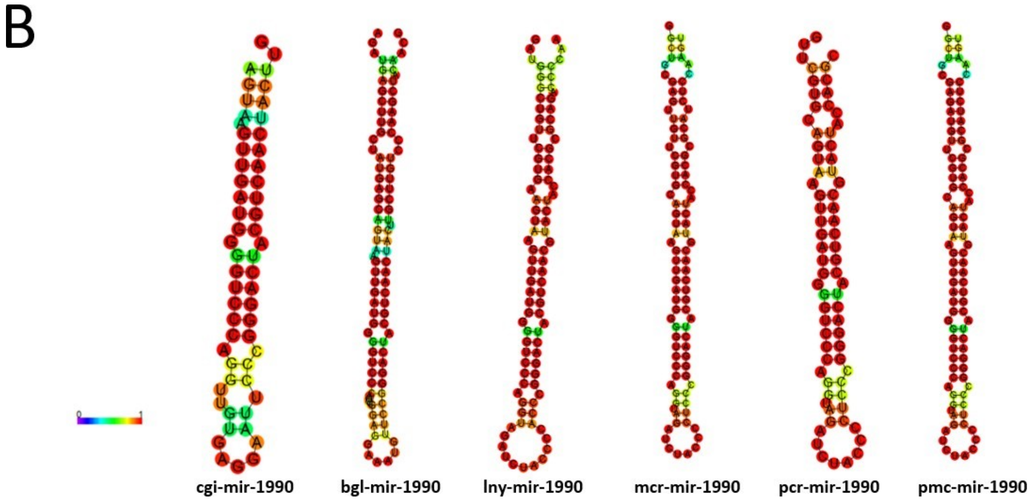


Figura 51: Estrutura secundária dos precursores miRNA-1990 ortólogos; cgi *Crassostrea gigas,* bgl: *Biomphalaria glabrata,* lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*



4.6 miRNA MOLLUSCA ESPECÍFICO

4.6.1 mir-1984

Em *C. gigas*, a expressão de mir-1984 aumentou significativamente em resposta aos estresses: baixa salinidade, calor e infecções bacterianas, sugerindo que este miRNA pode estar implicado em certas funções fisiológicas, como redução da oxidação e metabolismo energético (ZHAO *et al.*, 2016). Já em gastrópodes este miRNA pode estar envolvido no crescimento muscular. Foi verificado que o gene BMP7 é alvo do hdl-miRNA-1984 no molusco gastrópode *Haliotis discus hannai*. A proteína morfogenética óssea 7 (BMP7) induz a formação da cadeia leve quinase de miosina e cadeia pesada de miosina. Desta forma, o mir-1984 pode nortear o crescimento muscular ao regular o gene BMP7 no abalone (HUANG *et al.*, 2018). Achados na literatura científica corroboram os dados deste trabalho, na qual as sequencias dos precursores de *P. canaliculata* e *P. maculata* apresentaram conservação na estrutura primária e secundária em relação aos ortólogos (Figuras 52 e 53).

Figura 52: Alinhamento de precursores de miRNA-1984 com seus ortólogos.

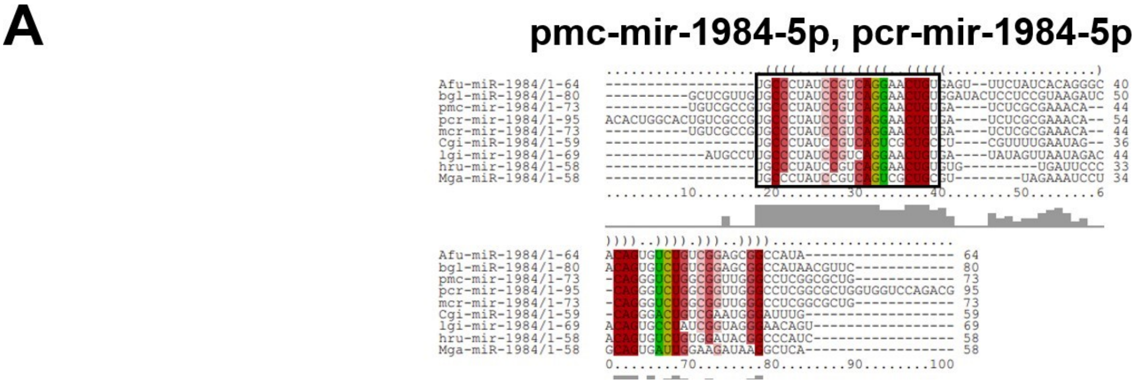
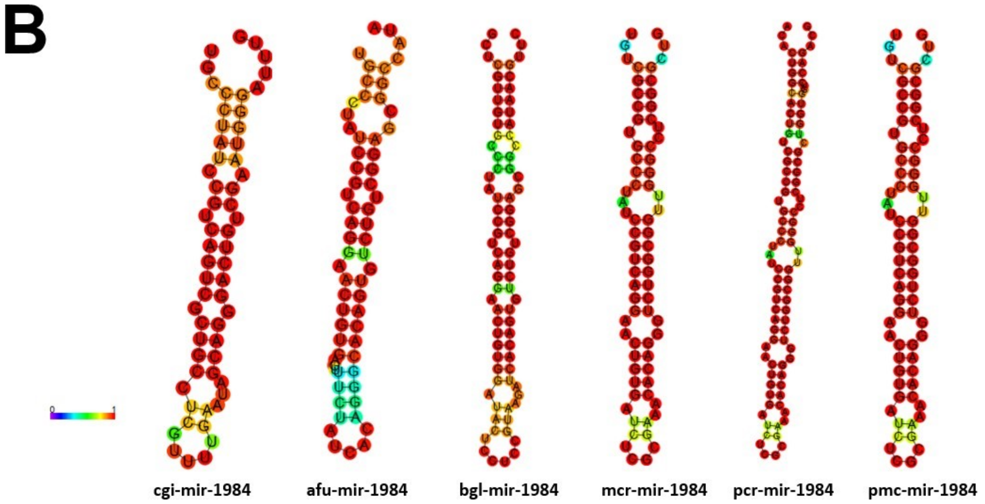


Figura 53: Estrutura secundária dos precursores de miRNA-1984 com seus ortólogos; cgi: *Crassostrea gigas,* afu: *Achatina fulica,* bgl: *Biomphalaria glabrata,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*



4.6.2 mir-1985

A família do mir-1985 predita para as espécies *P. canaliculata* e *P. maculata* é composta pelos precursores pcr-mir-1985 e pmc-mir-1985 e pelos miRNAs maduros pcr-miR-1985-5p e pmc-miR-1985-5p. Estes miRNAs demonstraram 100% de identidade com seus ortólogos (Figura 54). A estrutura secundária também se apresentou conservada entre os moluscos do gênero *Pomacea* e ortólogos (Figura 55).

Figura 54: Alinhamento de precursores de miRNA-1985 com seus ortólogos.

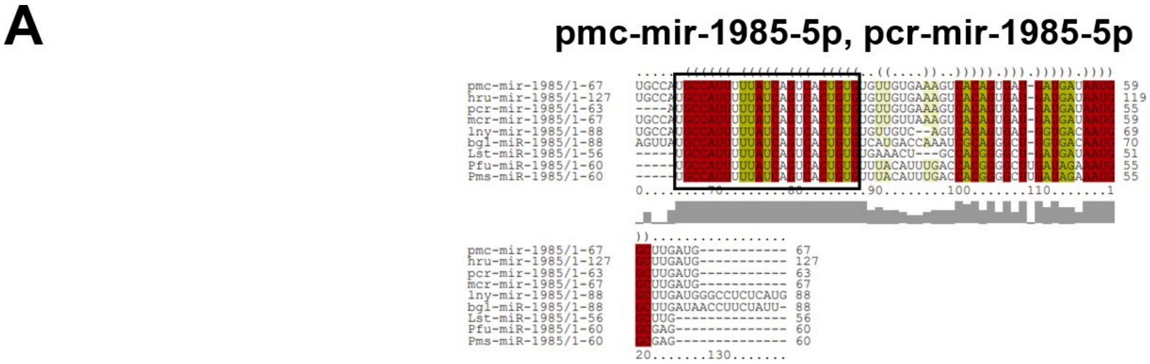
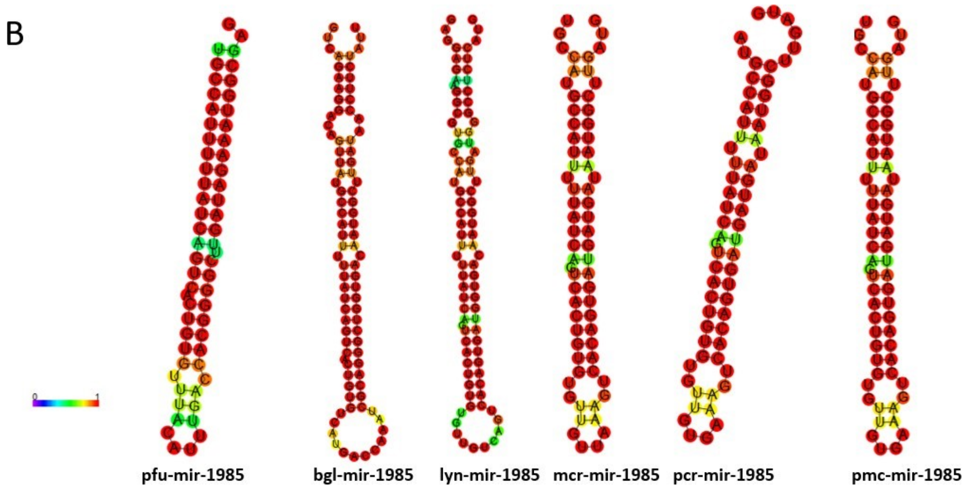


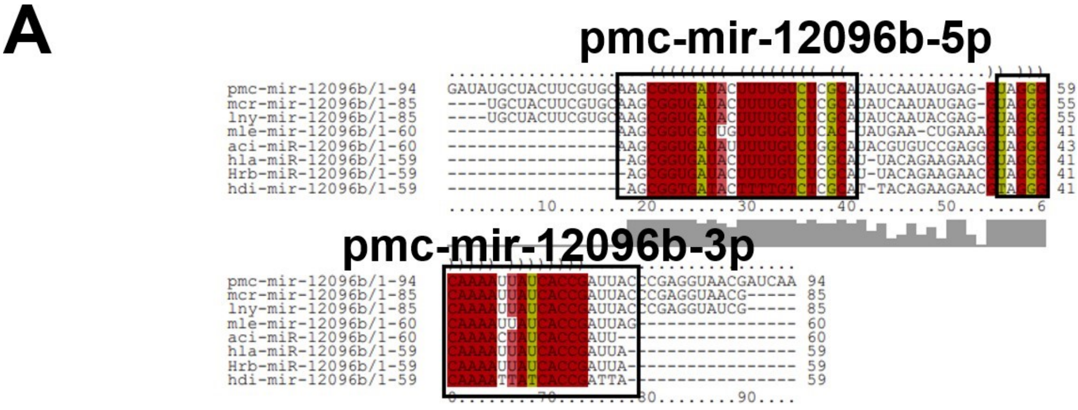
Figura 55: Estrutura secundária dos precursores miRNA-1985 e ortólogos; *pfu*: Pinctada fucata, bgl: *Biomphalaria glabrata,* mcr: *Marisa cornuarietis,* pcr: *Pomacea canaliculata,* pmc: *Pomacea maculata.*



4.6.3 mir-12096b

A família de mir-12096b foi encontrado no genoma de *P. maculata*. O precursor predito pmc-mir-12096b apresentou dois miRNAs maduros, 3p e 5p (Figura 56). Estes miRNAs foram localizados no genoma de *Melibe Leonina* na plataforma miRBase. Além disso, a família mir-12096b foi encontrada no genoma das espécies de moluscos; *A. californica*, *Chlamys farreri*, *Haliotis discus hannai*, *Haliotis laevigata* e *Haliotis rubra* (HUANG *et al.*, 2021). No alinhamento das estruturas primárias de *P. maculata* e ortólogos foi verificado alta conservação de nucleotídeos, sobretudo nas regiões dos miRNAs maduros (Figura 56). A estrutura secundária de pmc-mir 12096b demonstrou alta similaridade estrutural com ortólogos, especialmente entre organismos da família Ampullariidae (Figura 57).

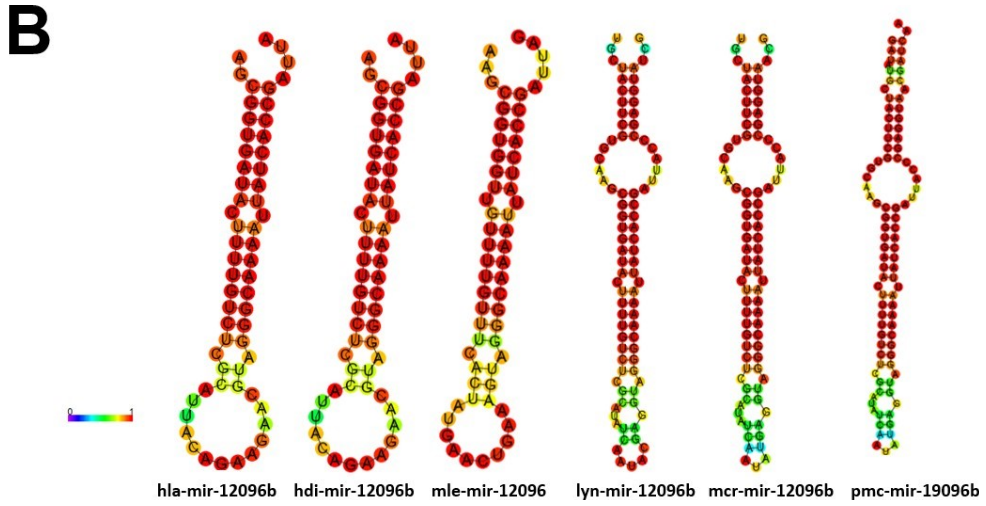
Figura 56: Alinhamento de precursores de miRNA-12096b com seus ortólogos.



```
pmc-mir-12096b/1-94   GAUAUGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAG-CUAGGC 59
mcr-mir-12096b/1-85   ----UGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAG-CUAGGC 55
lny-mir-12096b/1-85   ----UGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUACGAG-CUAGGC 55
mle-mir-12096b/1-60   -----------------AAGCGGUGGUUGUUUUGUUCAC-UAUGAA-CUGAAACUAGGC 41
aci-miR-12096b/1-60   -----------------AAGCGGUGAUAUUUUUGUCUGGCAUACGUGUCCGAGGCUAGGC 43
hla-miR-12096b/1-59   ------------------AGCGGUGAUACUUUUGUCUCGCAU-UACAGAAGAACCUAGGC 41
Hrb-miR-12096b/1-59   ------------------AGCGGUGAUACUUUUGUCUCGCAU-UACAGAAGAACCUAGGC 41
hdi-mir-12096b/1-59   ------------------AGCGGTGATACTTTTGTCTCGCAT-TACAGAAGAACCTAGGC 41
                      .........10........20........30........40........50........6

pmc-mir-12096b/1-94   CAAAAUUAUCACCGAUUACCCGAGGUAACGAUCAA 94
mcr-mir-12096b/1-85   CAAAAUUAUCACCGAUUACCCGAGGUAACG----- 85
lny-mir-12096b/1-85   CAAAAUUAUCACCGAUUACCCGAGGUAUCG----- 85
mle-mir-12096b/1-60   CAAAAUUAUCACCGAUUAG---------------- 60
aci-miR-12096b/1-60   CAAAACUAUCACCGAUU------------------ 60
hla-miR-12096b/1-59   CAAAAUUAUCACCGAUUA----------------- 59
Hrb-miR-12096b/1-59   CAAAAUUAUCACCGAUUA----------------- 59
hdi-mir-12096b/1-59   CAAAATTATCACCGATTA----------------- 59
                      0.........70.........80.........90....
```

Figura 57: Estrutura secundária dos precursores miRNA-12096b e ortólogos; hla: *Haliotis laevigata*, hdi: *Haliotis discus hannai*, mle: *Melibe leonina*, lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pmc: *Pomacea maculata.*



Os prováveis miRNAs maduros de *P. canaliculata* e *P. maculata,* bilatéria-específico, protostômio-específico, lophotrochozoa-específico e mollusca específico, caracterizados neste trabalho apresentaram uma grande conservação na região dos miRNAs maduros, quando comparados com seus respectivos ortólogos. Esta conservação se acentua nas regiões "*seeds*", demonstrando alta preservação nucleotídica, com 100% de similaridade nas posições de 2 a 8 nucleotídeos. As estruturas secundárias dos pré-miRNAs apresentaram formato característico canônico dos precursores de miRNAs e revelaram alta similaridade estrutural entre os ortólogos. Na análise filogenética, foi verificada ampla distribuição entre os clados filogenéticos, verificando uma distribuição evolutiva que corrobora com a árvore da vida.

4.7 IDENTIFICAÇÃO E CARACTERIZAÇÃO DE PROTEÍNAS ENVOLVIDAS NA BIOGÊNESE DA VIA DE miRNAS EM *P. canaliculata* E *P. maculata.*

Os miRNAs são importantes reguladores da expressão gênica que necessitam ser processados no núcleo e citoplasma em pri-miRNA, pré-miRNA e, finalmente, na forma madura do miRNA. Para isso, várias proteínas estruturais e catalíticas executam funções pré-determinadas de forma a obter a forma ativa do regular gênico.

Neste trabalho, as proteínas responsáveis pela biogênese dos miRNAs das espécies *P. canaliculata* e *P. maculata* foram identificadas e recuperadas a partir do proteoma predito das espécies localizados no banco de dados NCBI (http://.ncbi.nlm.nih.gov/). A partir desses dados, através de um script próprio, foi utilizado como ferramenta o BLASTp, usando como sequências de busca (*queries)* as proteínas da via de miRNAs oriundas dos organismos modelo *D. melanogaster* e *C. elegans*.

Foram identificadas e caracterizadas as prováveis proteínas em *P. canaliculata* e *P. maculata* envolvidas na via de miRNA. A detecção foi realizada baseada na homologia, comprimento da proteína, análise de domínios conservados, pesquisa de sítios ativos e filogenia (CARDOSO *et al*., 2020; QUEIROZ *et al*., 2017). As proteínas preditas foram comparadas com suas ortólogas de organismos próximo evolutivamente como *A. californica e B. glabrata* (Tabelas 9 e 10).

Tabela 9: Similaridade das proteínas da via de miRNA de *P. canaliculata* e suas ortólogas.

Proteína ID *P.canaliculata*	Provável proteína	Tamanho (aa)	Proteína ID *A. californica*	E-value	Tamanho (aa)	Proteina ID *B.glabrata*	E-value	Tamanho (aa)
Pca69814_c1_g2	Pca_ Argonauta 2	896	XP_005107588.1	0.0	926	XP_013068463.1	0.0	895
			XP_012943067.1	0.0	930	XP_013068462.1	0.0	899
			XP_005107586.1	0.0	932	XP_013068459.1	0.0	901
Pca61914_c0_g1	Pca_ Argonauta	935	XP_012940110.1	0.0	852	XP_013089248.1	0.0	897
			XP_005096149.1	0.0	918	XP_013068459.1	6,00E-50	901
Pca67531_c1_g2	Pca_Dicer	1984	XP_012942349.1	0.0	2503	XP_013067888.1	0.0	2332
Pca67887_c3_g1	Pca_Drosha	1524	XP_005107264.1	0.0	1528	XP_013069067.1	0.0	1469
Pca59979_c0_g1	Pca_DGCR8	730	XP_005109080.1	5,00E-135	991	XP_013076209.1	1,00E-139	889
			XP_005109077.1	6,00E-135	993			
Pca65193_c2_g1	Pca_Exportina 1	973	XP_005111754.1	0.0	1074	XP_013069551.1	0.0	1076
Pca52466_c2_g3	Pca_Ran	214	XP_005093963.1	3,00E-149	216	XP_013094746.1	8,00E-143	281

Fonte: Dados do autor

Tabela 10: Similaridade das proteínas da via de miRNA de *P. maculata* e suas ortólogas.

Proteína ID P. maculata	Provável proteína	Tamanho (aa)	Proteína ID *A. californica*	E-value	Comprimento (aa)	Proteina ID *B.glabrata*	E-value	Tamanho (aa)
Pma63715_c0_g1_	Pma_ Argonauta 2	890	XP_005107588.1	0.0	926	XP_013068463.1	0.0	895
			XP_012943067.1	0.0	930	XP_013068462.1	0.0	899
			XP_005107586.1	0.0	932	XP_013068459.1	0.0	901
Pma60501_c2_g16	Pma_ Argonauta	936	XP_012940110.1	0.0	852	XP_013089248.1	0.0	897
			XP_005096149.1	0.0	918	XP_013068459.1	3,00E-49	901
Pma95401_c0_g1	Pma_Dicer	288	XP_012942349.1	8,00E-78	2503	XP_013067888.1	1,00E-76	2332
Pma66024_c0_g2	Pma_Dicer	766	XP_005102833.1	0.0	1377	XP_013074821.1	6,00E-180	785

Pma65850_c2_g4	**Pma_Drosha**	**358**	**XP_005107264.1**	**2,00E-180**	**1528**	**XP_013069067.1**	**3,00E-178**	**1469**
Pma51393_c0_g1	**Pma_Drosha**	**580**	**XP_005107264.1**	**0.0**	**1528**	**XP_013069067.1**	**0.0**	**1469**
Pma62419_c0_g1	**PMA_DGCR8**	**708**	**XP_005109080.1**	**3,00E-147**	**991**	**XP_013076209.1**	**2,00E-152**	**889**
			XP_005109077.1	**4,00E-147**	**993**			
Pma59467_c0_g1	**Pma_Exportina 1**	**1069**	**XP_005111754.1**	**0.0**	**1074**	**XP_013069551.1**	**0.0**	**1076**
Pma63331_c0_g1	**Pma_Exportina T**	**965**	**XP_035825468.1**	**0.0**	**978**	**XP_013079973.1**	**0.0**	**963**
Pma54972_c0_g1	**Pma_Ran**	**152**	**XP_005093963.1**	**8,00E-107**	**216**	**XP_013094746.1**	**2,00E-99**	**281**

Fonte: Dados do autor

Os domínios conservados das proteínas foram analisados de forma a caracterizar as proteínas preditas. A partir das sequências de aminoácidos de *P. canaliculata* e *P. maculata* obtidas, foi utilizada a ferramenta Blastp no banco de dados RefSeq do NCBI (http://.ncbi.nlm.nih.gov/), para busca por proteínas ortólogas. Desta forma, os melhores *hits* foram aquelas que tinham maior similaridade e, então, recuperadas para pesquisa de domínios conservados, alinhamento e filogenia. As sequências escolhidas para pesquisa de sítios ativos basearam-se em espécies próximas evolutivamente, de organismos modelos (*D. melanogaster, C. elegans*, *H. sapiens*), além de protostômios e deuterostomios de forma geral. Tendo em vista a relação parasita hospedeiro entre *P. canaliculata* e *A. cantonensis,* procurou-se no banco de dados públicos proteínas do nematoide ortológas ao molusco, não obtendo sucesso na pesquisa.

4.7.1 Análise de domínios conservados, alinhamento global e filogenia das proteínas Argonauta

A proteína Argonauta é caracterizada pela presença dos domínios PIWI e PAZ (PIWI-Argonauta-Zwille) (SONG *et al*., 2004). As proteínas Argonauta de eucariotos podem apresentar além destes os seguintes domínios, N (N-terminal), e MID, junto com dois ligantes de domínio L1 e L2 (Figura 58) (SWARTS *et al*., 2014). O domínio PIWI em Argonauta contém três resíduos catalíticos conservados compostos por dois aspartatos e uma histidina, chamado de 'DDH' (YANG; STEITZ, 1995).

Figura 58: Arquiteturas de domínio das proteínas de eucariotos da superfamília PIWI.

Adaptado de Swarts *et al*. (2014)

Foram identificadas quatro prováveis proteínas Argonauta nas espécies em estudo, sendo duas em *P. canaliculata* (Pca69814_c1_g2, Pca61914_c0_g1) e duas em *P. maculata* (Pma63715_c0_g1_, Pma60501_c2_g16). A sequência proteica Argonauta 2 de *P. canaliculata,* (Pca69814_c1_g2) apresentou os mesmos e-values e ortólogos descritos para *P. maculata,* (Pma63715_c0_g1) para as espécies citadas na tabela 11.

Tabela 11: Similaridade de Argonauta 2 de *P. maculata* e ortólogos (protostômios e deuterostômios).

Proteina ID	**Nome da espécie**	**E-value Blastp**	**Tamanho (aa)**
Pma63715_c0_g1	*Pomacea maculata*	-----------	896
XP_005107586.1	*Aplysia californica*	0.0	932
XP_013068459.1	*Biomphalaria glabrata*	0.0	901
XP_009064000.1	*Lottia gigantea*	0.0	919
XP_008196653.1	*Tribolium castaneum*	0.0	951
XP_029721415.1	*Aedes albopictus*	0.0	929
NP_001367206.1	*Caenorhabditis elegans*	0.0	1023
XP_012792763.2	*Schistosoma haematobium*	0.0	1043
XP_024349713.1	*Echinococcus granulosus*	0.0	1113
XP_032815443.1	*Petromyzon marinus*	0.0	855
XP_041421428.1	*Xenopus laevis*	0.0	861
XP_040210947.1	*Rana temporaria*	0.0	873
XP_016848276.1	*Anolis carolinensis*	0.0	920
XP_007425657.1	*Python bivittatus*	0.0	842
NP_001376478.1	*Gallus gallus*	0.0	800
XP_017602151.1	*Corvus brachyrhynchos*	0.0	864
NP_694818.3	*Mus musculus*	0.0	860
XP_023104240.1	*Felis catus*	0.0	860
NP_036286.2	*Homo sapiens*	0.0	859

Fonte: Dados do autor

As sequências de aminoácidos Pca69814_c1_g2 e Pma63715_c0_g1 apresentaram seis domínios conservados (Tabela 12), enquanto as proteínas Argonauta (Pca61914_c0_g1 e Pma60501_c2_g16) apresentaram apenas os domínios PAZ E PWI (Tabela 13 e Figura 59). O comprimento das sequências para ambas as espécies, além da similaridade com suas ortólogas corroboram para identificação das prováveis proteínas Argonauta 2 e Argonauta (Tabelas 11,12,13 e 14, Figura 60).

Tabela 12: Análise de domínios conservados Argonauta 2, *P. maculata, P. canaliculata* e ortólogos.

ID proteina	espécie	Domínios	ID PFAM	Alinhamento		e-value
				Inicio	**Fim**	
Pma63715_c0_g1	*Pomacea maculata*	ArgoMid	PF16487	461	541	2.8e-33
		ArgoN	PF16486	67	197	2.6e-32
		PAZ	PF02170	271	396	5.8e-25
		ArgoL1	PF08699	207	257	5.8e-23
		ArgoL2	PF16485	405	451	7.3e-17
Pca69814_c1_g2	*Pomacea canaliculata*	Piwi	PF02171	548	854	3.9e-109
		ArgoMid	PF16487	461	541	2.9e-33
		ArgoL2	PF16485	405	451	7.3e-17
		PAZ	PF02170	271	396	5.9e-25
		ArgoL1	PF08699	207	257	5.8e-23
		ArgoN	PF16486	67	197	2.6e-32
XP_005107586.1	*Aplysia californica*	Piwi	PF02171	584	884	1.3e-110
		ArgoMid	PF16487	497	577	5.1e-34
		ArgoN	PF16486	103	233	5.7e-33
		PAZ	PF02170	309	432	1.4e-26
		ArgoL1	PF08699	243	293	3.3e-23
		ArgoL2	PF16485	441	487	3.8e-16
XP_013068459.1	*Biomphalaria glabrata*	Piwi	PF02171	553	853	1.2e-112
		ArgoMid	PF16487	466	546	2.8e-34
		ArgoN	PF16486	72	202	6.1e-32
		PAZ	PF02170	282	401	4.1e-26
		ArgoL1	PF08699	212	262	3.2e-23
		ArgoL2	PF16485	410	456	5.7e-15
XP_009064000.1	*Lottia gigantea*	Piwi	PF02171	577	877	8.7e-113
		ArgoN	PF16486	96	226	9.5e-34
		ArgoMid	PF16487	490	570	1.9e-33
		PAZ	PF02170	300	425	1.9e-26
		ArgoL1	PF08699	236	286	1.3e-23
		ArgoL2	PF16485	434	480	7.5e-13
XP_008196653.1	*Tribolium castaneum*	Piwi	PF02171	609	909	4.2e-114
		ArgoMid	PF16487	520	601	1.2e-33
		ArgoN	PF16486	121	250	2.6e-28
		PAZ	PF02170	324	449	9.6e-26
		ArgoL1	PF08699	260	310	3.5e-23
		ArgoL2	PF16485	458	504	2.7e-17
NP_001367206.1	*Caenorhabditis elegans*	Piwi	PF02171	682	981	9.0e-116
		ArgoMid	PF16487	594	675	4.9e-33
		ArgoN	PF16486	170	300	1.8e-27
		PAZ	PF02170	410	529	2.6e-26
XP_024349713.1	*Echinococcus granulosus*	Piwi	PF02171	791	1091	1.8e-114

		ArgoMid	PF16487	703	784	3.8e-30
		ArgoN	PF16486	308	439	7.3e-27
		PAZ	PF02170	512	639	5.6e-27
XP_032815443.1	*Petromyzon marinus*	Piwi	PF02171	513	813	2.1e-115
		ArgoMid	PF16487	425	506	2.4e-34
		ArgoN	PF16486	32	162	8.8e-31
		PAZ	PF02170	242	361	1.0e-27
XP_016848276.1	*Anolis carolinensis*	Piwi	PF02171	579	878	6.5e-115
		ArgoMid	PF16487	490	571	8.4e-33
		ArgoN	PF16486	97	227	1.3e-29
		PAZ	PF02170	307	426	1.1e-26
NP_694818.3	*Mus musculus*	Piwi	PF02171	519	818	2.2e-115
		ArgoMid	PF16487	430	511	7.6e-33
		ArgoN	PF16486	37	167	7.1e-30
		PAZ	PF02170	247	366	1.0e-26
		ArgoL1	PF08699	177	227	3.0e-21
		ArgoL2	PF16485	375	421	5.8e-13
NP_036286.2	*Homo sapiens*	Piwi	PF02171	518	817	2.1e-115
		ArgoMid	PF16487	429	510	7.6e-33
		ArgoN	PF16486	36	166	7.0e-30
		PAZ	PF02170	246	365	1.0e-26

Fonte: Dados do autor

Tabela 13: Similaridade de Argonauta de *P. maculata* e ortólogos (protostômios e deuterostômios).

Proteina ID	**Nome da espécie**	**E-value Blastp**	**Tamanho (aa)**
Pma60501_c2_g16	*Pomacea maculata*	-----------	936
XP_012940110.1	*Aplysia californica*	0.0	852
XP_013089248.1	*Biomphalaria glabrata*	0.0	897
XP_009064630.1	*Lottia gigantea*	0.0	791
XP_041362450.1	*Gigantopelta aegis*	0.0	943
XP_034311101.1	*Crassostrea gigas*	0.0	938
XP_033734243.1	*Pecten maximus*	0.0	964
XP_021371709.1	*Mizuhopecte nyessoensis*	0.0	878
XP_014773732.1	*Octopus bimaculoides*	0.0	897
XP_015904269.1	*Parasteatoda tepidariorum*	0.0	940
XP_040078863.1	*Ixodes scapularis*	0.0	943
XP_007906012.1	*Callorhinchus milii*	0.0	853
XP_031752134.1	*Xenopus tropicalis*	0.0	869
XP_026572019.1	*Pseudonaja textilis*	0.0	870
XP_007489688.1	*Monodelphis domestica*	0.0	880

XP_032252277.1	*Phoca vitulina*	0.0	861
XP_030856685.1	*Gorilla gorilla gorilla*	0.0	861
NP_004755.2	*Homo sapiens*	0.0	861

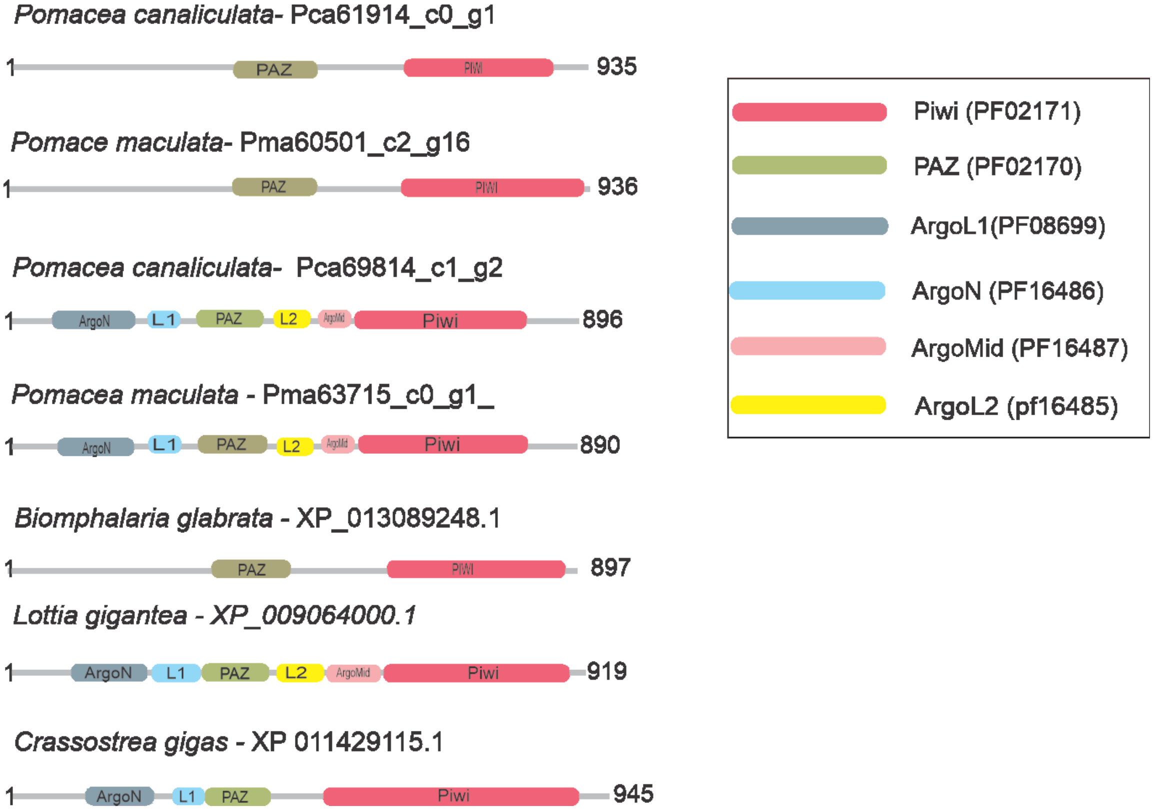
Fonte: Dados do autor

Tabela 14: Análise de domínios conservados Argonauta *P. maculata, P. canaliculata* e ortólogos.

ID proteína	**Espécie**	**Domínios**	**ID PFAM**	**Alinhamento**		**e-value**
				Inicio	**Fim**	
Pma60501_c2_g16	*Pomacea maculata*	Piwi	PF02171	629	921	3.1e-89
		PAZ	PF02170	353	485	5.4e-30
Pca61914_c0_g1	*Pomacea canaliculata*	Piwi	PF02171	628	920	4.3e-89
		PAZ	PF02170	352	484	5.4e-30
XP_012940110.1	*Aplysia californica*	Piwi	PF02171	546	838	1.2e-85
		PAZ	PF02172	274	402	6.4e-31
XP_013089248.1	*Biomphalaria glabrata*	Piwi	PF02173	591	881	4.3e-93
		PAZ	PF02174	310	440	2.4e-32
XP_034311101.1	*Crassostrea gigas*	Piwi	PF02175	633	924	4.7e-87
		PAZ	PF02176	358	489	1.6e-31
		ArgoL1	PF02177	302	348	6.2e-05
NP_492121.1	*Caenorhabditis elegans*	Piwi	PF02178	500	809	7.8e-79
		PAZ	PF02179	230	352	1.3e-24
XP_040078863.1	*Ixodes scapularis*	Piwi	PF02180	636	928	2.2e-87
		PAZ	PF02181	371	491	7.1e-32
		ArgoL1	PF02182	307	352	1.9e-05
NP_001036627.2	*Drosophila melanogaster*	Piwi	PF02183	567	852	1.8e-81
		PAZ	PF02184	297	422	4.5e-28
NP_001352553.1	*Danio rerio*	Piwi	PF02185	741	1031	1.4e-80
		PAZ	PF02186	465	595	1.2e-31
		ArgoL1	PF02187	412	457	8.4e-07
NP_067286.1	*Mus musculus*	Piwi	PF02185	556	848	1.1e-104
		PAZ	PF02186	281	413	2.3e-36
		GAGE	PF02187	1	107	5.4e-21
		ArgoL1	PF02188	228	274	4.6e-05
NP_004755.2	*Homo sapiens*	Piwi	PF02190	555	847	1.4e-106
		PAZ	PF02191	280	412	6.4e-36
		GAGE	PF02192	1	106	4.1e-25
		ArgoL1	PF02193	227	273	5.1e-05

Fonte: Dados do autor

Figura 59: Distribuição dos domínios conservados, prováveis proteínas Argonauta.

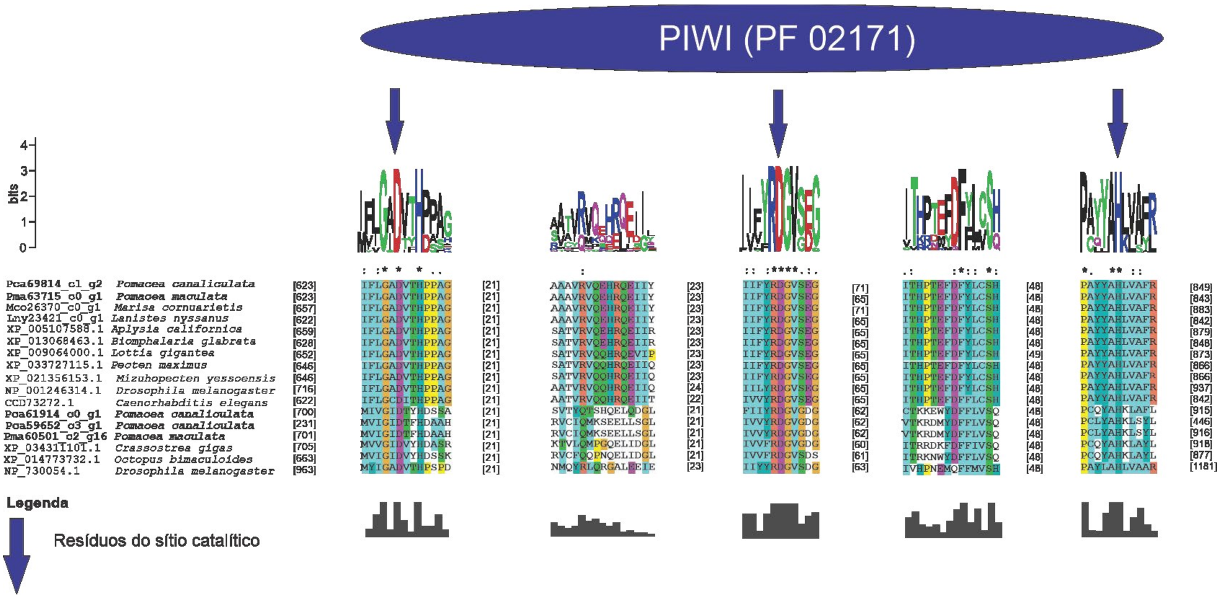


Fonte: Dados do autor

Para *P. canaliculata* e *P. maculata,* foi realizado o alinhamento global de forma a demonstrar a similaridade de sequências peptídicas de organismos diversos das prováveis proteínas da via de miRNAs, além de mostrar os resíduos e as circunvizinhanças dos sítios catalíticos. O alinhamento global foi realizado utilizando o software ClustaX2.1 (Figura 60)

O domínio PIWI das prováveis proteínas de *P. canaliculata* e *P. maculata* apresentaram uma tríade catalítica formada por DDH (ácido aspártico, ácido aspártico e histidina), apresentando alta conservação em relação às proteínas ortólogas de Argonauta, principalmente nas imediações do resíduo catalítico (Figura 60).

Figura 60: Alinhamento global, análise do domínio PIWI das proteínas Argonauta de *P. canaliculata* e *P. maculata.*



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                                                   : :* *  * ..            :                  :: :****.:.          .:     :*:: *:          *.  ** ::
Pca69814_c1_g2   Pomacea canaliculata      [623]   IFLGADVTHPPAG   [21]   AAAVRVQEHRQEIIY   [23]   IIFYRDGVSEG   [71]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [849]
Pma63715_c0_g1   Pomacea maculata          [623]   IFLGADVTHPPAG   [21]   AAAVRVQEHRQEIIY   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [843]
Mco26370_c0_g1   Marisa cornuarietis       [657]   IFLGADVTHPPAG   [21]   AAAVRVQEHRQEIIY   [23]   IIFYRDGVSEG   [71]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [883]
Lny23421_c0_g1   Lanistes nyssanus         [622]   IFLGADVTHPPAG   [21]   AAAVRVQEHRQEIIY   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [842]
XP_005107588.1   Aplysia californica       [659]   IFLGADVTHPPAG   [21]   SATVRVQEHRQEIIR   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [879]
XP_013068463.1   Biomphalaria glabrata     [628]   IFLGADVTHPPAG   [21]   SATVRVQEHRQEIIR   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [848]
XP_009064000.1   Lottia gigantea           [652]   IFLGADVTHPPAG   [21]   SATVRVQQHRQEVIP   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [49]   PAYYAHLVAFR   [873]
XP_033727115.1   Pecten maximus            [646]   IFLGADVTHPPAG   [21]   SATVRVQQHRQEIIQ   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [866]
XP_021356153.1   Mizuhopecten yessoensis   [646]   IFLGADVTHPPAG   [21]   SATVRVQQHRQEIIQ   [23]   IIFYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [866]
NP_001246314.1   Drosophila melanogaster   [716]   IFLGADVTHPPAG   [21]   AATVRVQQHRQEIIQ   [24]   IILYRDGVSEG   [65]   ITHPTEFDFYLCSH   [48]   PAYYAHLVAFR   [937]
CCD73272.1       Caenorhabditis elegans    [622]   IFLGCDITHPPAG   [21]   AATVRVQQHRQEIIT   [22]   IVVYRDGVSEG   [65]   ITHPTEFDFFLCSH   [48]   PAYYAHLVAFR   [842]
Pca61914_c0_g1   Pomacea canaliculata      [700]   MIVGIDTYHDSSA   [21]   SVTYQTSHQELQDGL   [21]   FIIYRDGVGDG   [62]   CTKKEWYDFFLVSQ   [48]   PCQYAHKLAFL   [915]
Pca59652_c3_g1   Pomacea canaliculata      [231]   MVIGIDTFHDAAH   [21]   RVCIQMKSEELLSGL   [21]   IVVYRDGVGDG   [62]   VTKRDMYDFFLVSQ   [48]   PCLYAHKLSYL   [446]
Pma60501_c2_g16  Pomacea maculata          [701]   MVIGIDTFHDAAH   [21]   RVCIQMKSEELLSGL   [21]   IVVYRDGVGDG   [62]   VTKRDMYDFFLVSQ   [48]   PCLYAHKLSYL   [916]
XP_034311101.1   Crassostrea gigas         [705]   MVVGIDVYHDASR   [21]   KTVLQMPGQELIDGL   [21]   IVVFRDGVGDG   [60]   ITRRDKFDFFIVSQ   [48]   PCQYAHKLAYL   [918]
XP_014773732.1   Octopus bimaculoides      [663]   MVIGIDVYHDSSK   [21]   RVCFQQPNQELIDGL   [21]   IVVFRDGVSDS   [61]   ITRKNWYDFFLVSQ   [48]   PCQYAHKLAYL   [877]
NP_730054.1      Drosophila melanogaster   [963]   MYIGADVTHPSPD   [21]   NMQYRLQRGALEEIE   [23]   IIYYRDGVSDG   [63]   IVHPNEMQFFMVSH   [48]   PAYLAHLVAAR   [1181]
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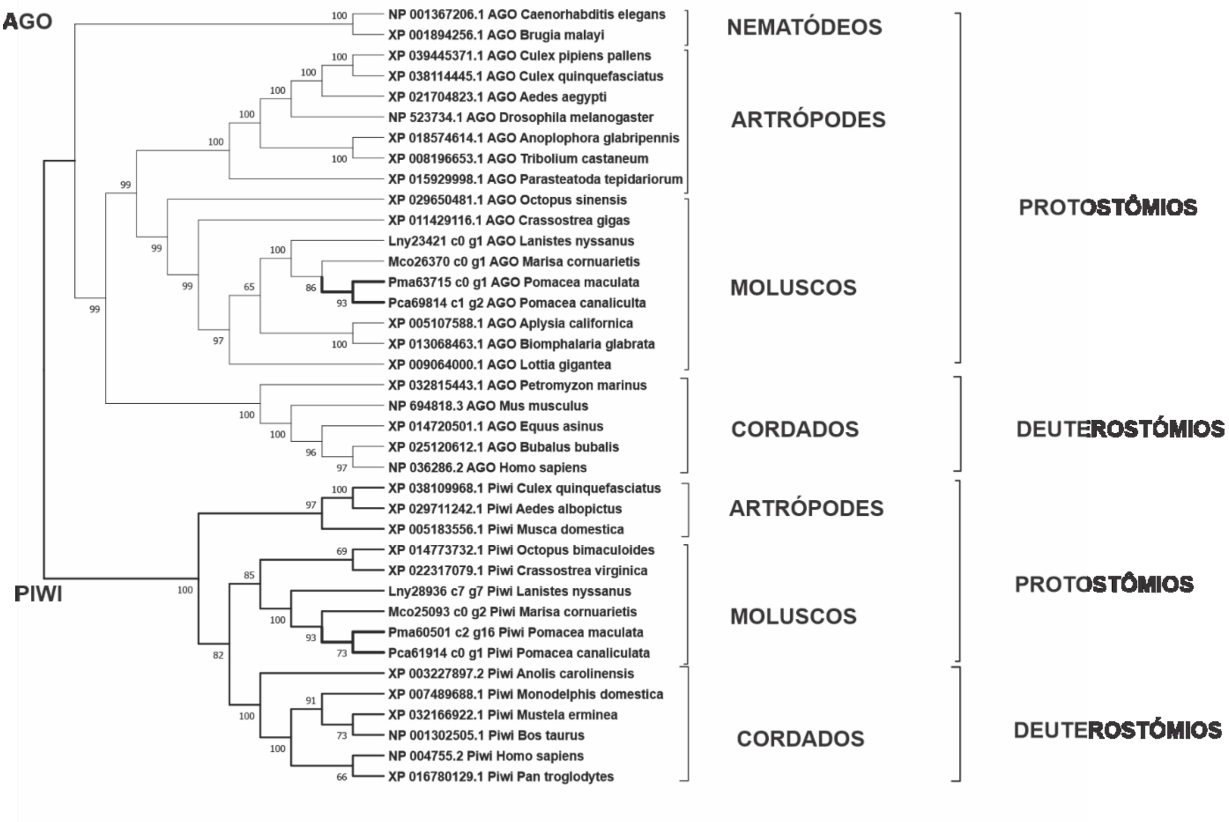
Fonte: Dados do autor

As sequências das proteínas preditas de *P. canaliculata, P. maculata* e ortólogos de referência (RefSeq) (organismos protostômios e deuterostômios) foram submetidas a análises filogenéticas comparativas utilizando o programa MEGA versão X (TAMURA *et al*., 2007). As árvores filogenéticas foram construídas utilizando o método Neighbor-joining (NJ) (SAITOU; NEI, 1987) e calculada com o modelo de substituição JTT. Na análise filogenética foi verificada a ampla distribuição entre os clados filogenéticos, verificando uma distribuição evolutiva que corrobora com a árvore da vida (Figura 61).

Figura 61: Análise filogenética das proteínas Argonauta, de *P. maculata* e *P. canaliculata* e seus ortólogos.



Fonte: Dados do autor

4.7.2 Análise de domínios conservados, alinhamento global e filogenia das proteínas Dicer e Drosha

A ribonuclease III (RNase III) pertence à família das endoribonucleases que apresentam especificidade para RNA de fita dupla (dsRNA). Três classes de enzimas RNase III foram relatadas. As enzimas da classe I são as mais simples, contendo um domínio de endonuclease e um domínio de ligação de dsRNA (dsRBD). As proteínas de classe II contêm dois domínios de endonuclease e um dsRBD. As moléculas de classe III consistem em dois domínios de endonuclease, um dsRBD e um domínio de helicase N-terminal seguido por um domínio PAZ (Figura 62). Funcionalmente, as enzimas de classe 3 produzem produtos de RNA de fita simples (ssRNA) de aproximadamente 22 nucleotídeos de comprimento a partir de substratos de dsRNA longos (BERNSTEIN *et al.*, 2001; BLASZCZYK *et al.*, 2001; FILIPPOV *et al.*, 2000). A via canônica da biogênese de miRNAs é a via dominante no qual os miRNAs são processados pelas RNAases III de classe II (Drosha) e classe III (Dicer), consecutivamente.

Figura 62: Três classes de proteínas da família RNase III representadas por Dicer humano (classe III), Drosha humana (classe II) e RNase III bacteriana (classe I).

Adaptado de Zhang *et al*. (2004)

As RNases III (classes II e II) contém centros catalíticos com aminoácidos importantes para a função. A atividade das enzimas se concentra em domínios RIBOc, que possuem sítios ativos compostos pelos seguintes aminoácidos E (Ácido glutâmico), D (Ácido aspártico), D (Ácido aspártico), E (Ácido glutâmico) (Figura 64) (HAN *et al*., 2004; ZHANG *et al*., 2004)

Tabela 15: Similaridade de Drosha de *P. canaliculata* e ortólogos.

Proteina ID	Nome da espécie	E-value Blastp	Tamanho (aa)
Pca67887_c3_g1	*Pomacea canaliculata*	-----------	1524
XP_033730374.1	*Pecten maximus*	0.0	1443
XP_014774431.1	*Octopus bimaculoides*	0.0	1501
XP_005107264.1	*Aplysia californica*	0.0	1528
XP_014481243.1	*Dinoponera quadriceps*	0.0	1319
XP_040166968.1	*Anopheles arabiensis*	0.0	1498
NP_477436.1	*Drosophila melanogaster*	0.0	1327
XP_012268888.1	*Athalia rosae*	0.0	1553
XP_018652602.1	*Schistosoma mansoni*	0.0	1577
XP_024352870.1	*Echinococcus granulosus*	0.0	1279
XP_035590140.1	*Schistosoma haematobium*	0.0	1650
XP_001901812.1	*Brugia malayi*	0.0	1329
NP_001122460.2	*Caenorhabditis elegans*	0.0	1081
XP_004652583.1	*Jaculus jaculus*	0.0	1336
XP_005248351.1	*Homo sapiens*	0.0	1306
XP_014377706.1	*Alligator sinensis*	0.0	1307
XP_009206471.1	*Papio anubis*	0.0	1337
XP_032193598.1	*Mustela erminea*	0.0	1306
XP_017921117.1	*Capra hircus*	0.0	1337

Fonte: Dados do autor

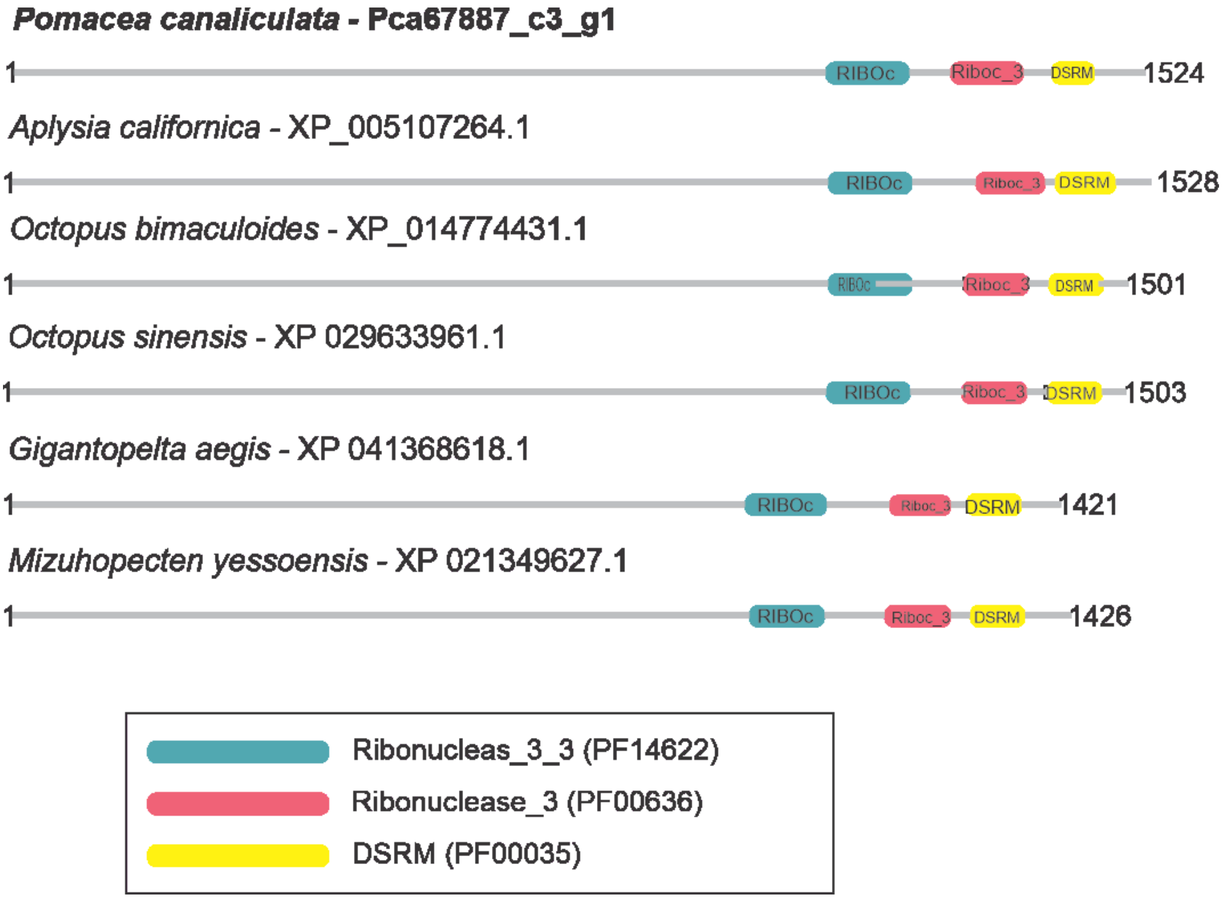
Tabela 16: Análise de domínios conservados de Drosha *P. canaliculata* e ortólogos.

ID Proteina	**Espécie**	**Domínios**	**ID PFAM**	**Alinhamento**		**e-value**
				Inicio	**Fim**	
Pca67887_c3_g1	*Pomacea canaliculata*	Ribonucleas_3_3	PF14622	1095	1201	1.2e-20
		Ribonuclease_3	PF00636	1277	1366	9.2e-21
		dsrm	PF00035	1394	1464	3.4e-11
Pma65850_c2_g4	*Pomacea maculata*	Ribonuclease_3	PF00636	111	200	1.1e-21
		dsrm	PF00035	228	298	4.8e-12
Pma51393_c0_g1	*Pomacea maculata*	Ribonucleas_3_3	PF14622	502	574	2.2e-12
XP_033730374.1	*Pecten maximus*	Ribonucleas_3_3	PF14622	1008	1115	3.2e-21
		Ribonuclease_3	PF00636	1190	1279	1.8e-20
		dsrm	PF00035	1307	1377	2.7e-08
XP_005107264.1	*Aplysia californica*	Ribonucleas_3_3	PF14622	1107	1214	3.9e-22
		Ribonuclease_3	PF00636	1290	1380	2.0e-17
		dsrm	PF00035	1408	1479	9.9e-11
XP_014481243.1	*Dinoponera quadriceps*	Ribonucleas_3_3	PF14622	997	888	3.1e-22
		Ribonuclease_3	PF00636	1156	1070	1.7e-19
		dsrm	PF00035	1256	1184	2.3e-11
NP_477436.1	*Drosophila melanogaster*	Ribonucleas_3_3	PF14622	829	936	5.9e-22
		Ribonuclease_3	PF00636	1012	1098	2.5e-20
		dsrm	PF00035	1126	1197	4.1e-13
XP_018652602.1	*Schistosoma mansoni*	Ribonuclease_3	PF00636	966	1056	1.1e-16
		Ribonuclease_3	PF00636	1145	1236	2.6e-20
		dsrm	PF00035	1265	1334	6.0e-11
NP_001122460.2	*Caenorhabditis elegans*	Ribonuclease_3	PF00636	868	955	1.4e-18
		Ribonucleas_3_3	PF14622	687	789	4.8e-18
		dsrm	PF00035	983	1055	8.7e-11
XP_014774431.1	*Octopus bimaculoides*	Ribonucleas_3_3	PF14622	1096	1205	1.4e-21
		Ribonuclease_3	PF00636	1279	1367	4.9e-21
		dsrm	PF00035	1396	1467	2.1e-12
XP_040166968.1	*Anopheles arabiensis*	Ribonucleas_3_3	PF14622	803	911	2.1e-22
		Ribonuclease_3	PF00636	987	1073	1.0e-19
		dsrm	PF00035	1101	1172	3.2e-13
NP_001103942.1	*Danio rerio*	Ribonucleas_3_3	PF14622	881	989	7.3e-25
		Ribonuclease_3	PF00636	1064	1152	1.0e-20
		dsrm	PF00035	1181	1251	1.9e-14
XP_014377706.1	*Alligator sinensis*	Ribonucleas_3_3	PF14622	894	1003	4.4e-24
		Ribonuclease_3	PF00636	1077	1165	1.1e-20

		dsrm	PF00035	1194	1264	2.0e-14
XP_005248351.1	*Homo sapiens*	Ribonucleas_3_3	PF14622	894	1002	6.2e-24
		Ribonuclease_3	PF00636	1076	1164	8.4e-21
		dsrm	PF00035	1193	1263	1.9e-14
XP_017921117.1	*Capra hircus*	Ribonucleas_3_3	PF14622	925	1033	6.4e-24
		Ribonuclease_3	PF00636	1107	1195	8.7e-21
		dsrm	PF00035	1224	1294	2.0e-14
NP_001123621.1	*Mus musculus*	Ribonucleas_3_3	PF14622	961	1069	6.6e-24
		Ribonuclease_3	PF00636	1143	1231	9.0e-21
		dsrm	PF00035	1260	1330	2.1e-14

Fonte: Dados do autor

Figura 63 : Análise de domínios conservados de Drosha em *P. canaliculata.*



Fonte: Dados do autor

A provável proteína Pca67887_c3_g1 de *P. canaliculata* (Pca_drosha) apresentou comprimento compatível e similaridade com seus ortólogos, principalmente com relação aos clados Mollusca e Arthropoda (Tabela 15 e Figura 63). Na análise de domínios conservados, apresentou os dois domínios de endonuclease e um dsrm (importante para a ligação à dupla fita de RNA), característico das RNAases III de classe 2 (Tabela 16 e Figura 63). Na pesquisa por sítios catalíticos, apresentou apenas o primeiro conjunto composto pelos aminoácidos E-D-D-E no domínio Riboc (figura 64). O comprimento de 1524 aminoácidos é compatível com a proteína de *P. canaliculata* (ID: XP_025111754.1) de 1523 aminoácidos registrada no NCBI (National Centre for Biotechnology Information — http://.ncbi.nlm.nih.gov/), com um percentual de identificação de 99,68% (1522/1524 aminoácidos) e um *gap*. Esses dados corroboram a identificação da provável proteína Drosha em *P. canaliculata*.

Para *P. maculata*, as proteínas Drosha apresentaram tamanho incompatível para os seus ortólogos em deuterostômios e protostômios (Tabela 10). No entanto, revelam indícios de serem prováveis proteínas. A sequências proteica (Pma51393_c0_g1) com 580 aminoácidos tem como ortólogo a proteína de *P. canaliculata* registrada no NCBI (ID: XP_025111754.1). Ao se realizar BLASTp no NCBI do peptídeo Pma51393_c0_g1 de *P. maculata* contra a proteína acima descrita, apresentou percentual de identificação de 99,68% (579/580) dos aminoácidos. Foi verificado parte dos aminoácidos catalíticos (E-D) presentes no primeiro domínio Riboc (Figura 64), apresentando apenas este domínio conservado para a provável proteína Pma51393_c0_g1. Em Drosha, provavelmente devido à falta de cobertura no sequenciamento desta região do genoma de *P. maculata,* não foi possível evidenciar a presençado segundo domínio catalítico Riboc. As regiões no genoma preditas para este gene provavelmente estão com erro de anotação levando ao truncamento das sequências proteicas. Consequentemente, o tamanho das sequências de aminoácidos para estes genes está com tamanho e estrutura subestimadas. Futuras versões do genoma podem resolver este problema (GOMES, 2012).

Tabela 17: Similaridade de Dicer de *P. canaliculata* e ortólogos.

Proteina ID	**Nome da espécie**	**E-value Blastp**	**Tamanho (aa)**
Pca67531_c1_g2	*Pomacea canaliculata*	-----------	1984
XP_009046342.1	*Lottia gigantea*	0.0	1800
XP_033733992.1	*Pecten maximus*	0.0	2132
XP_022331683.1	*Crassostrea virginica*	0.0	1909
XP_011439629.2	*Crassostrea gigas*	0.0	1873
XP_014781473.1	*Octopus bimaculoides*	0.0	2402
XP_021004529.1	*Parasteatoda tepidariorum*	0.0	1998
XP_023219964.1	*Centruroides sculpturatus*	0.0	1950
NP_524453.1	*Drosophila melanogaster*	7,00E-140	2249

XP_018644375.1	*Schistosoma mansoni*	0.0	2319
XP_024350050.1	*Echinococcus granulosus*	0.0	1909
XP_003137813.1	*Loa loa*	0.0	1928
NP_498761.2	*Caenorhabditis elegans*	0.0	1910
XP_024128907.1	*Oryzias melastigma*	0.0	1857
XP_020953825.1	*Sus scrofa*	0.0	1926
XP_018085949.1	*Xenopus laevis*	0.0	1891
XP_008763126.1	*Rattus norvegicus*	0.0	1918
XP_021034238.1	*Mus caroli*	0.0	1917
XP_017170486.1	*Mus musculus*	0.0	1783
NP_001182502.1	*Homo sapiens*	0.0	1829

Fonte: Dados do autor

Tabela 18: Domínios conservados de Dicer de *P. canaliculata* e ortólogos.

ID Proteina	**Espécie**	**Domínios**	**ID PFAM**	**Alinhamento**		**e-value**
				Inicio	**Fim**	
Pca67531_c1_g2	*Pomacea canaliculata*	Ribonucleas_3_3	PF14622	1755	1937	2.0e-33
		PAZ	PF02170	954	1104	9.2e-36
		Dicer_dimer	PF003368	668	758	3.1e-24
		Helicase_C	PF00271	530	591	4.9e-14
Pca66636_c2_g1	*Pomacea canaliculata*	DEAD	PF00270	131	295	5.6e-20
		RIG-I_C-RD	PF11648	708	818	6.9e-20
		Helicase_C	PF00271	521	629	2.2e-19
		RIG-I_C	PF18119	349	496	1.2e-15
Pma66024_c0_g2	*Pomacea maculata*	DEAD	PF00270	55	219	3.2e-20
		RIG-I_C-RD	PF11648	632	742	1.1e-19
		Helicase_C	PF00271	445	553	1.9e-19
		RIG-I_C	PF18119	273	420	2.4e-15
Pma95401_c0_g1	*Pomacea maculata*	Ribonucleas_3_3	PF14622	195	266	1.3e-25
XP_009046342.1	*Lottia gigantea*	Ribonucleas_3_3	PF14622	1289	1473	3.0e-33
		Ribonuclease_3	PF00636	1577	1703	1.1e-22
		PAZ	PF02170	890	1041	1.2e-36
		Dicer_dimer	PF003368	607	698	9.8e-24

		Helicase_C	PF00271	480	536	2.0e-13
XP_033733992.1	*Pecten maximus*	Ribonucleas_3_3	PF14622	1621	1804	9.4e-36
		Ribonuclease_3	PF00636	1914	2035	3.0e-22
		PAZ	PF02170	914	1066	9.3e-37
		Dicer_dimer	PF003368	630	721	1.5e-25
		Helicase_C	PF00271	492	554	7.9e-16
XP_011439629.2	*Crassostrea gigas*	Ribonucleas_3_3	PF14622	1350	1563	2.5e-36
		Ribonuclease_3	PF00636	1656	1775	1.1e-21
		PAZ	PF02170	933	1085	1.5e-37
		Dicer_dimer	PF003368	645	736	5.5e-27
		Helicase_C	PF00271	516	572	2.3e-13
		DEAD	PF00270	31	181	5.3e-06
XP_012942349.1	*Aplysia californica*	Ribonucleas_3_3	PF14622	1972	2157	7.0e-31
		Ribonuclease_3	PF00636	2273	2406	1.3e-21
		PAZ	PF02170	954	1110	1.0e-37
		Dicer_dimer	PF003368	676	766	6.7e-24
		Helicase_C	PF00271	519	583	1.2e-13
XP_013067888.1	*Biomphalaria glabrata*	Ribonucleas_3_3	PF14622	1814	1991	9.9e-34
		Ribonuclease_3	PF00636	2100	2235	1.2e-21
		PAZ	PF02170	946	1097	3.2e-34
		Dicer_dimer	PF003368	662	752	7.1e-25
		Helicase_C	PF00271	493	573	3.9e-12
XP_021004529.1	*Parasteatoda tepidariorum*	Ribonucleas_3_3	PF14622	1519	1688	3.4e-35
		Ribonuclease_3	PF00636	1782	1903	1.0e-22
		PAZ	PF02170	925	1080	4.4e-42
		Dicer_dimer	PF003368	640	730	1.5e-28
		Helicase_C	PF00271	494	565	2.9e-16
		DEAD	PF00270	31	194	1.6e-12
XP_023219964.1	*Centruroides sculpturatus*	Ribonucleas_3_3	PF14622	1468	1636	8.5e-37
		Ribonuclease_3	PF00636	1729	1850	1.0e-22
		PAZ	PF02170	898	1054	5.7e-42
		Dicer_dimer	PF003368	618	710	1.2e-24
		Helicase_C	PF00271	461	545	2.7e-18

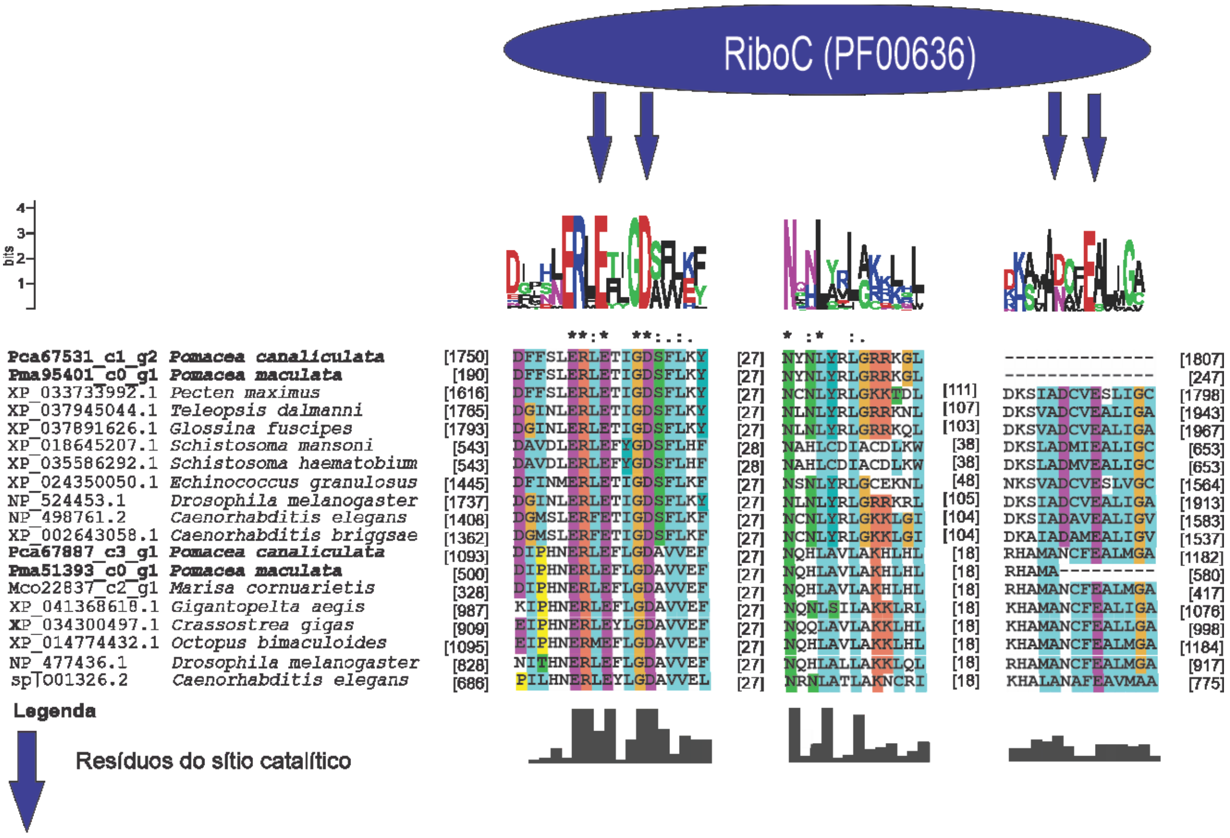
		DEAD	PF00270	31	191	1.6e-17
NP_524453.1	*Drosophila melanogaster*	Ribonucleas_3_3	PF14622	1742	1919	9.3e-36
		Ribonuclease_3	PF00636	2029	2149	8.5e-22
		PAZ	PF02170	1111	1266	3.9e-40
		Dicer_dimer	PF003368	825	916	2.6e-24
		Helicase_C	PF00271	543	604	7.2e-16
XP_018644375.1	*Schistosoma mansoni*	Ribonucleas_3_3	PF14622	1767	1943	6.4e-33
		Ribonuclease_3	PF00636	2053	2208	6.1e-23
		PAZ	PF02170	1255	1428	2.0e-28
		Dicer_dimer	PF003368	814	923	3.5e-16
XP_024350050.1	*Echinococcus granulosus*	Ribonucleas_3_3	PF14622	1450	1570	8.5e-30
		Ribonuclease_3	PF00636	1676	1813	7.0e-20
		PAZ	PF02170	1108	1277	2.8e-20
		Dicer_dimer	PF003368	771	891	7.9e-12
NP_498761.2	*Caenorhabditis elegans*	Ribonucleas_3_3	PF14622	1413	1589	1.1e-35
		Ribonuclease_3	PF00636	1679	1805	6.9e-21
		PAZ	PF02170	856	1025	5.5e-46
		Dicer_dimer	PF003368	571	663	3.5e-24
		Helicase_C	PF00271	381	498	5.2e-17
		ResIII	PF04851	12	182	2.3e-13
NP_524453.1	*Drosophila melanogaster*	Ribonucleas_3_3	PF14622	1742	1919	9.3e-36
		Ribonuclease_3	PF00636	2029	2149	8.5e-22
		PAZ	PF02170	1111	1266	3.9e-40
		Dicer_dimer	PF003368	825	916	2.6e-24
		Helicase_C	PF00271	543	604	7.2e-16
XP_024128907.1	*Oryzias melastigma*	Ribonucleas_3_3	PF14622	1256	1537	1.2e-43
		Ribonuclease_3	PF00636	1637	1759	9.8e-23
		PAZ	PF02170	893	1054	1.9e-39
		Dicer_dimer	PF003368	620	708	7.8e-25
		Helicase_C	PF00271	437	544	2.9e-17
		ResIII	PF04851	22	197	1.4e-10
NP_001154925.1	*Danio rerio*	Ribonucleas_3_3	PF14622	1295	1551	3.7e-45
		Ribonuclease_3	PF00636	1645	1767	1.1e-22

		PAZ	PF02170	889	1050	4.4e-39
		Dicer_dimer	PF003368	616	704	4.3e-24
		Helicase_C	PF00271	428	540	1.9e-17
		DEAD	PF00270	36	177	1.2e-11
XP_020953825.1	*Sus scrofa*	Ribonucleas_3_3	PF14622	1310	1570	1.2e-43
		Ribonuclease_3	PF00636	1694	1816	1.2e-22
		PAZ	PF02170	903	1064	3.2e-39
		Dicer_dimer	PF003368	630	718	1.7e-23
		Helicase_C	PF00271	442	554	1.6e-17
		DEAD	PF00270	46	192	4.6e-12
XP_018085949.1	*Xenopus laevis*	Ribonucleas_3_3	PF14622	1289	1550	1.2e-42
		Ribonuclease_3	PF00636	1671	1793	1.0e-22
		PAZ	PF02170	907	1056	1.7e-38
		Dicer_dimer	PF003368	622	710	4.7e-25
		Helicase_C	PF00271	434	546	2.3e-17
		DEAD	PF00270	36	186	3.3e-11
XP_008763126.1	*Rattus norvegicus*	Ribonucleas_3_3	PF14622	1313	1573	5.4e-43
		Ribonuclease_3	PF00636	1698	1820	1.2e-22
		PAZ	PF02170	903	1064	3.2e-39
		Dicer_dimer	PF003368	630	718	4.2e-23
		Helicase_C	PF00271	442	554	1.4e-17
		ResIII	PF04851	32	207	8.2e-12
XP_017170486.1	*Mus musculus*	Ribonucleas_3_3	PF14622	1303	1561	8.2e-44
		Ribonuclease_3	PF00636	1686	1758	5.0e-15
		PAZ	PF02170	893	1054	2.9e-39
		Dicer_dimer	PF003368	620	708	1.2e-23
		Helicase_C	PF00271	432	544	1.2e-17
		ResIII	PF04851	22	197	1.6e-11
NP_001182502.1	*Homo sapiens*	Ribonucleas_3_3	PF14622	1313	1575	9.3e-44
		Ribonuclease_3	PF00636	1702	1776	4.6e-15
		PAZ	PF02170	903	1064	3.1e-39
		Dicer_dimer	PF003368	630	718	6.0e-24
		Helicase_C	PF00271	442	554	1.5e-17

	ResIII	PF04851	32	207	1.6e-11

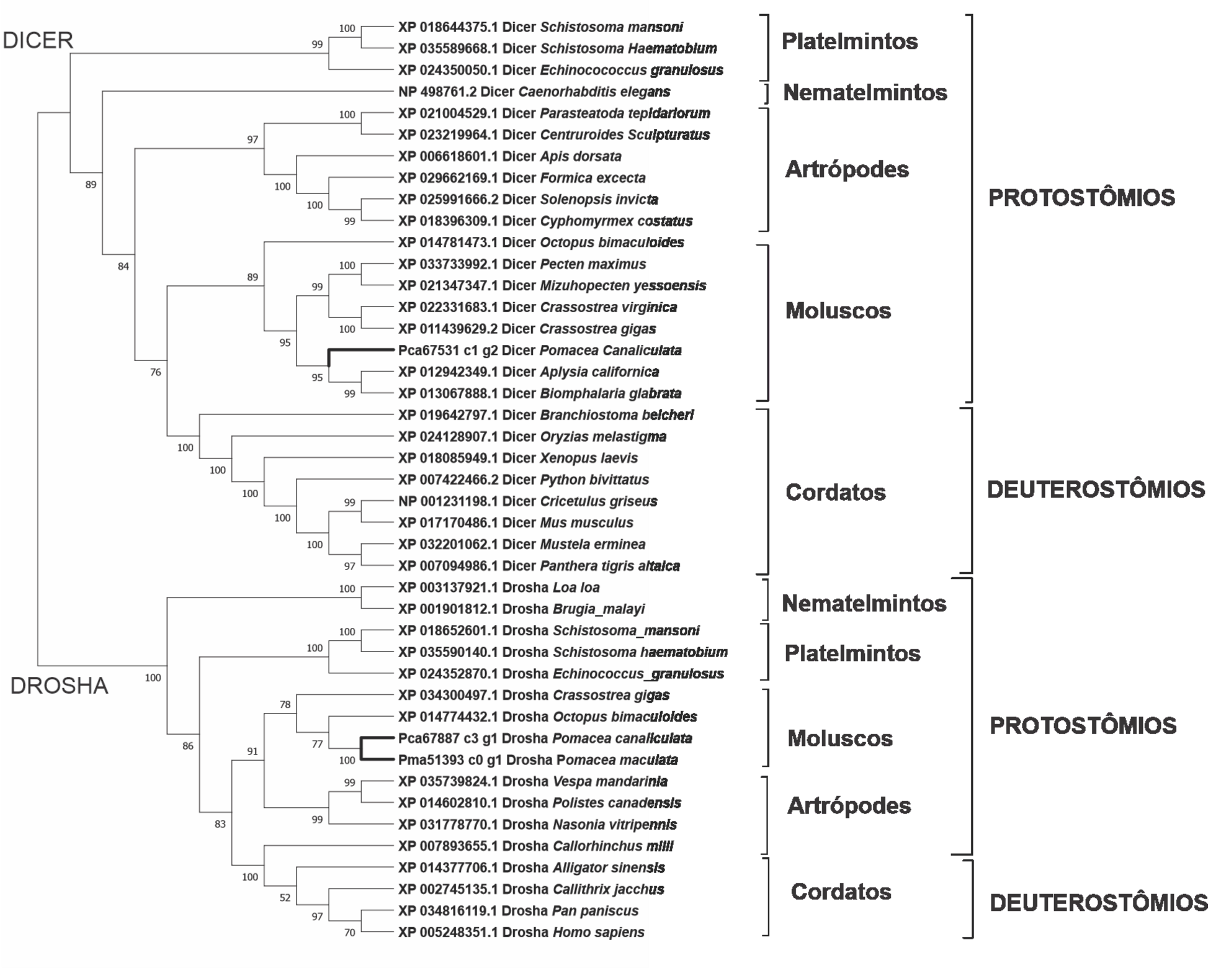
Fonte: Dados do autor

Figura 64: Alinhamento global, análise ribonuclease III (Riboc) das proteínas Dicer (acima representado) e Drosha (abaixo representado) de *P. canaliculata e P. maculata* e seus ortólogos.



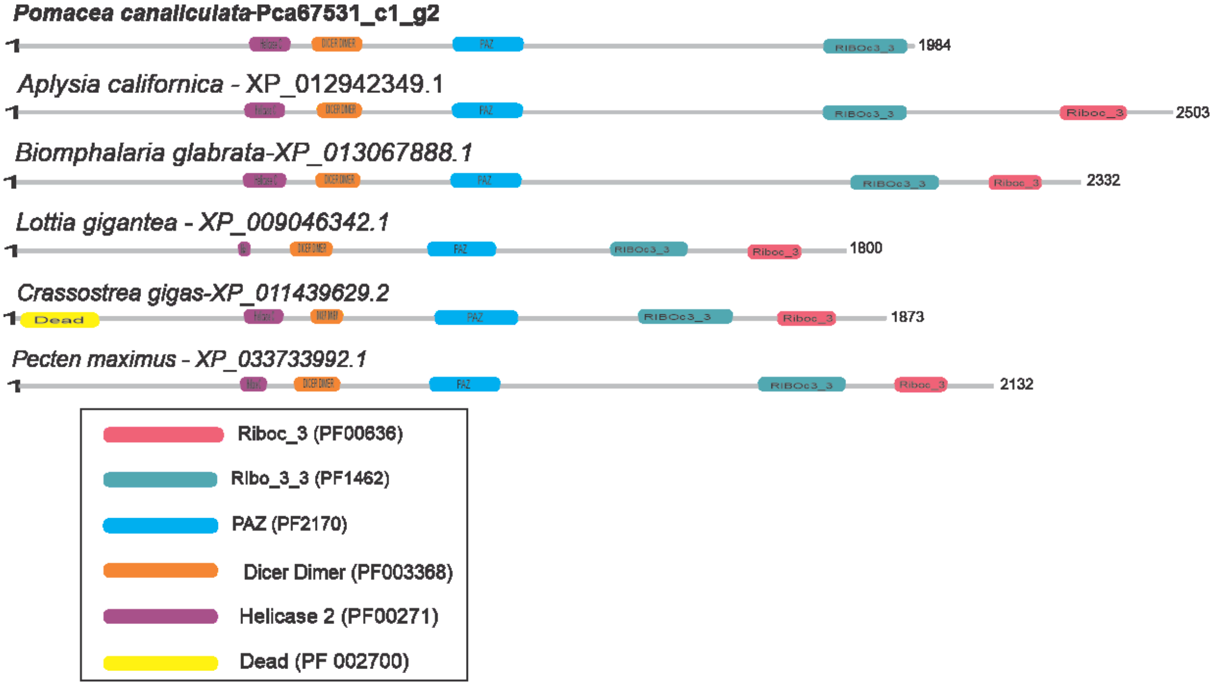
Fonte: Dados do autor

Figura 65: Análise filogenética das proteínas Dicer (acima representado) e Drosha (abaixo representado) de *P. canaliculata* e *P. maculata* e seus ortólogos.



Fonte: Dados do autor

Figura 66: Domínios conservados de Dicer de *P. canaliculata* e ortólogos.



Fonte: Dados do autor

A candidata à sequência proteica Pca67531_c1_g2 de *P. canaliculata* (Pca_Dicer) apresentou comprimento similar (1984 aa) a organismos próximos evolutivamente, *A. californica* (2503 aa) e *B. glabrata* (2332 aa) (Tabelas 9 e 17). Foram identificados apenas os domínios conservados; Ribonucleas_3_3, PAZ, Dicer dimer e Helicase_C; não apresentando o segundo domínio Riboc (Tabela 18 e Figura 66). Ao verificar o posicionamento de início do segundo domínio Riboc de *A. californica* (2273 aa) e *B. glabrata* (2100 aa), corrobora o fato de não identificação da segunda ribonuclease na provável sequência proteica. Em Dicer, provavelmente devido à falta de cobertura no sequenciamento desta região do genoma de *P. maculata,* não foi possível evidenciar a presença do segundo domínio catalítico Riboc. A proteína Dicer de *P. canaliculata* (XP_025104500) identificada no NCBI, comprimento 2249 aminoácidos é ortologa a Pca67531_c1_g2, com uma cobertura da *query* de 99% e porcentagem de identificação de 98,83%. A sequência proteica Pca67531_c1_g2 ao utilizar a ferramenta BLASTp contra o genoma de *P. canaliculata* no banco de dados AmbuBase (Department of Computer Science HKBU – https://www.comp.hkbu.edu.hk/~db/AmpuBase/#&panel1-4) apresentou um percentual de identificação e cobertura de 100% (1984/1984 aminoácidos). Foi observado o primeiro sítio catalítico dos aminoácidos (E-D) na Riboc na proteína candidata citada (Pca67531_c1_g2) (Figura 64). Ao realizar a busca por resíduos catalíticos tendo como query NP_524453.1 Dicer-1 *Drosophila melanogaster,* os moluscos ortólogos apresentaram no primeiro domínio Riboc apenas os aminoácidos catalíticos (E-D), com exceção de Pectem Maximus (E-D-D-E) NCBI (http://.ncbi.nlm.nih.gov/). O alinhamento múltiplo das proteínas mostrou alta similaridade de sequências dos aminoácidos, particularmente em regiões de domínio funcional conservado (Figura 64). Dicer revelou ampla distribuição entre os clados filogenéticos na árvore filogenética, mostrando alta similaridade da proteína catalítica de *P. canaliculata* entre moluscos, artrópodes e cordados sobretudo (Figura 65). Os dados apresentados corroboram para a identificação da provável proteína.

Para *P. maculata*, as prováveis proteínas Dicer, Pma66024_c0_g2 (766 aa) e Pma95401_c0_g1 (288 aa) não apresentaram comprimento compatível com a dimensão média das proteínas ortólogas. As regiões no genoma preditas para este gene provavelmente estão com erro de anotação levando ao truncamento das sequências proteicas. Consequentemente, o tamanho das sequências de aminoácidos para estes genes está com tamanho e estrutura subestimadas. Futuras versões do genoma podem resolver esse problema (GOMES, 2012; QUEIROZ *et al*., 2017). A sequência proteica (Pma66024_c0_g2) não evidenciou os domínios de endonucleases, já a provável proteína (Pma95401_c0_g1) revelou apenas um domínico Riboc com resíduos catalíticos nos aminoácidos E-D (Tabelas 10 e 18, figura 66). A sequência proteica (Pma95401_c0_g1) de *P. maculata* é ortóloga as proteínas *de P. canaliculata* (XP_025104500) identificada no NCBI e Pca67531_c1_g2 (caracterizada neste trabalho), apresentado percentual de identidade de 97,22 % (280/288 aminoácidos) com a proteína de *P. canaliculata* identificada no NCBI.

4.7.3 Análise das proteínas DGCR8, Exportina 1 e T, ran

As proteínas Ras, Exportinas 1 e T, DGCR8 e TDRD1 em *P. canaliculata* e *P. maculata* foram analisadas através da homologia, tamanho da proteína e domínios conservados.

4.7.3.1 DGCR8

A sequência proteica DGCR8 (DiGeorge Syndrome Critical Region 8) faz parte do complexo de microprocessadores que realiza a clivagem do pri-miRNA (transcrito do DNA) em pré-miRNA. O DGCR8 se liga em uma estrutura no pri-miRNA; enquanto, Drosha realiza a cisão do duplex. Esta etapa realizada no núcleo celular é primordial para a biogênese dos miRNAs, reguladores pós-transcricionais da expressão gênica (HAN *et al*., 2004).

Tabela 19: Análise de similaridade de DGCR8 de *P. maculata* e ortólogos.

Proteina ID	Nome da espécie	E-value Blastp	Tamanho (aa)
Pma62419_c0_g1	*Pomacea maculata*	-----------	708
XP_029632956.1	*Octopus sinensis*	0.0	679
XP_021357307.1	*Mizuhopecten yessoensis*	0.0	704
XP_033731508.1	*Pecten maximus*	0.0	790
XP_033731509.1	*Pecten maximus*	0.0	786
XP_033747276.1	*Pecten maximus*	0.0	806
XP_041348914.1	*Gigantopelta aegis*	0.0	778
XP_033747277.1	*Pecten maximus*	0.0	802

Fonte: Dados do autor

Tabela 20: Análise de similaridade de DGCR8 de *P. canaliculata* e ortólogos.

Proteina ID	Nome da espécie	E-value Blastp	Tamanho (aa)
Pca59979_c0_g1	*Pomacea canaliculata*	-----------	730
XP_029632956.1	*Octopus sinensis*	0.0	679
XP_033747276.1	*Pecten maximus*	0.0	806
XP_033747277.1	*Pecten maximus*	0.0	802
XP_021357307.1	*Mizuhopecten yessoensis*	0.0	704
XP_033731508.1	*Pecten maximus*	0.0	790
XP_033731509.1	*Pecten maximus*	0.0	786
XP_041348914.1	*Gigantopelta aegis*	1,00E-175	778
XP_022344386.1	*Crassostrea virginica*	3,00E-171	781
XP_014780090.1	*Octopus bimaculoides*	8,00E-164	646

Fonte: Dados do autor

A proteína identificada no NCBI subunidade DGCR8 (XP_025088948.1) de *P. canaliculata* apresenta o mesmo conjunto de aminoácidos (730/730), 100% de percentual de identificação que a provável sequência proteica predita neste trabalho (Pca59979_c0_g1) de *P. canaliculata*. A sequência de aminoácidos (Pma62419_c0_g1) de *P. maculata* (708 aa) revelou um percentual de identificação de 99,71% com a proteína de *P. canaliculata* acima citada identificada no NCBI. As sequências proteicas preditas de *P. maculata* e *P. canaliculata* (DGCR8) revelam tamanho compatível e similaridade com ortólogos (Tabelas 19 e 20).

4.7.3.2 Exportina 1 e T

Neste trabalho, foram preditas 3 sequências proteicas pertencentes à família de proteínas exportinas (XPO1 e XPOT) no genoma dos caracóis; *P. canaliculata* (Pca65193_c2_g1) e *P. maculata* (Pma59467_c0_g1), Exportina 1; *P. maculata* (Pma63331_c0_g1) exportina T. Intrinsecamente, o transporte de precursores de miRNAs é executado pela proteína XPO5. Ocasionalmente, o transporte de pré-miRNAs pode ser realizado por XPO1 entrando na via de processamento não canônico de miRNAs sem a clivagem por Drosha. O deslocamento de pré-miRNAs também é executado pela exportina T (XPOT) de maneira dependente de Ran-GTP (RUBY; JAN; BARTEL, 2007; XIE *et al*., 2013).

Tabela 21 – Similaridade de XPO-1 de *P. maculata* e ortólogos.

Proteina ID	**Nome da espécie**	**E-value Blastp**	**Tamanho (aa)**
Pma59467_c0_g1	Pomacea maculata	-----------	1069
XP_025103585.1	Pomacea canaliculata	0.0	1069
XP_041355132.1	Gigantopelta aegis	0.0	1073
XP_009046981.1	Lottia gigantea	0.0	1068
XP_021363249.1	Mizuhopecten yessoensis	0.0	1075
XP_033735453.1	Pecten maximus	0.0	1075
XP_022332005.1	Crassostrea virginica	0.0	1076
XP_011451739.1	Crassostrea gigas	0.0	1076
XP_014782256.1	Octopus bimaculoides	0.0	1072
XP_013415269.1	Lingula anatina	0.0	1072
XP_013400816.1	Lingula anatina	0.0	1072

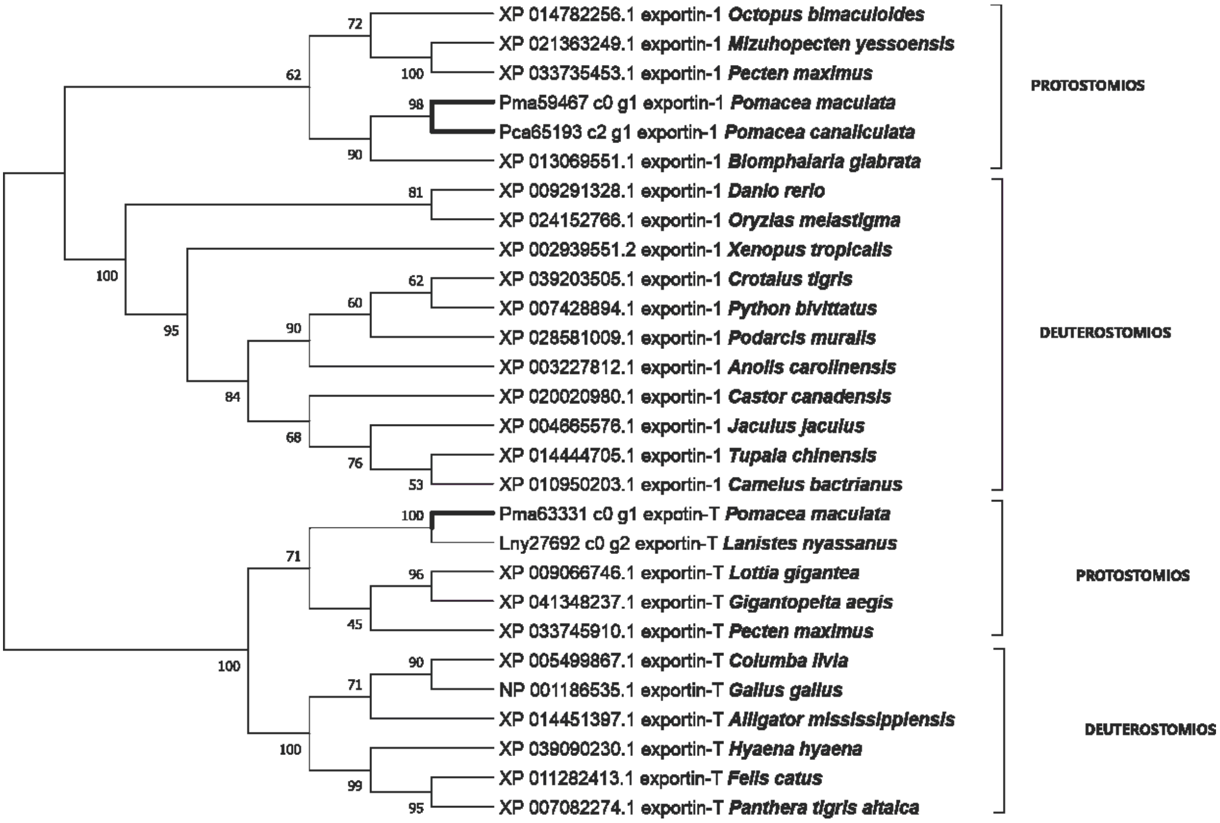
Fonte: Dados do autor

Tabela 22 – Similaridade de XPO-T de *P. maculata* e ortólogos.

Proteina ID	**Nome da espécie**	**E-value Blastp**	**Tamanho (aa)**
Pma63331_c0_g1	*Pomacea maculata*	-----------	965
XP_025106719.1	*Pomacea canaliculata*	0.0	965
XP_041348237.1	*Gigantopelta aegis*	0.0	961
XP_005097615.1	*Aplysia californica*	0.0	968
XP_009066746.1	*Lottia gigantea*	0.0	962
XP_011455074.2	*Crassostrea gigas*	0.0	969
XP_022302836.1	*Crassostrea virginica*	0.0	962
XP_021359684.1	*Mizuhopecten yessoensis*	0.0	963
XP_013079973.1	*Biomphalaria glabrata*	0.0	960

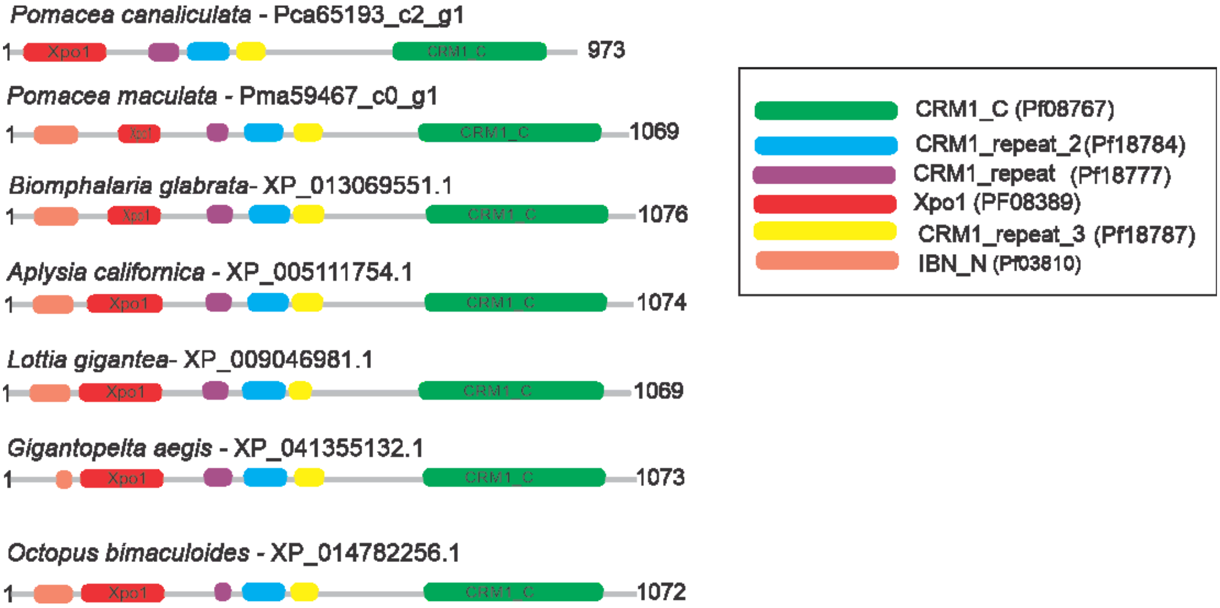
Fonte: Dados do autor

Figura 67: Árvore filogenética XPO-1 e XPO-T *P. canaliculata* e *P. maculata.*



Fonte: Dados do autor

Figura 68: Análise de domínios conservados de *P. maculata* e *P. canaliculata* – XPO-1.



Fonte: Dados do autor

Com o objetivo de enfatizar a conservação destas prováveis proteínas identificados nas espécies em estudo, foi construído uma árvore filogenética e estudos de domínios conservados. A análise filogenética revelou uma distribuição das sequências de aminoácidos (Pca65193_c2_g1 e Pma59467_c0_g1) - XPO-1 e (Pma63331_c0_g1) - XPO-T, agrupadas próximas ao clado Deuterostômio. As prováveis proteínas de *P. maculata* e *P. canaliculata* agregaram-se com as respectivas ortólogas próximas ao filo Mollusca e ao clado Deuterostômio (Figura 67).

A similaridade das Exportinas identificadas do gênero *Pomacea* das espécies em estudo foi verificada junto às suas ortólogas observando nível de preservação dos domínios conservados e comprimento. A proteína predita de *P. maculata* (Pma59467_c0_g1) apresentou os mesmos domínios conservados (XPO-1) de suas ortólogas e comprimento compatível com organismos próximos evolutivamente (Tabela 21 e Figura 68). A sequência proteica de *P. canaliculata* (Pca65193_c2_g1) apresentou um domínio conservado a menos que seus ortólogos, revelando tamanho ligeiramente inferior (Figura 68). A falta de cobertura na predição da proteína prejudicou a predição de domínios conservados, uma vez que, ela se deu na porção N-terminal da proteína. A proteína predita (XPO-T) de *P. maculata* (Pma63331_c0_g1) apresentou comprimento compatível e similaridade com suas ortólogas, organismos próximos evolutivamente (Tabela 22).

4.7.3.3 Ran

A proteína Ran executa importante função na via de miRNAs. Na via canônica da biogênese do miRNAs, após a cisão dos pri-miRNAs em pré-miRNAs no núcleo celular os precursores precisam ser exportados para o citoplasma. Esta etapa é dependente complexo XPO5-Ran, que se ligam aos pré-miRNAs com dispêndio de GTP (YI *et al*., 2003).

Observou-se que a sequência proteica de *P. canaliculata* (Pca52466_c2_g3_Ran) apresentou o mesmo comprimento e 100% de identificação com os aminoácidos da proteína já identificada da espécie (XP_025090191.1) no NCBI com 214 aminoácidos. Da mesma forma, a sequência proteica de *P. maculata* (Pma54972_c0_g) de 152 aminoácidos, provavelmente truncadas no genoma, apresentou percentual de identificação de 100% com a proteína de *P. canaliculata* (XP_025090191.1) *registrada* no NCBI (National Centre for Biotechnology Information — http://.ncbi.nlm.nih.gov/). As regiões no genoma preditas para o gene de Ran de *P. maculata* provavelmente estão com o erro de anotação levando ao truncamento da sequência proteica ocasionando o tamanho relativamente menor da proteína predita comparados com suas ortólogas (Tabela 23).

Tabela 23 – Similaridade de Ran de *P. maculata* e ortólogos.

Nome da espécie	**ID**	**Cobertura Query**	**E value**	**Identidade %**	**Comprimento**
Pomacea maculata	Pma54972_c0_g1	-----------	--------	--------	152
Pomacea canaliculata	XP_025090191.1	99%	2,00E-111	100.00%	214
Aplysia californica	XP_005093963.1	99%	1,00E-105	93.42%	216
Gigantopelta aegis	XP_041348103.1	99%	1,00E-102	92.11%	215
Mizuhopecten yessoensis	XP_021345207.1	98%	4,00E-100	89.40%	222
Crassostrea virginica	XP_022309199.1	99%	2,00E-99	88.16%	213
Crassostrea gigas	XP_011426860.1	99%	3,00E-99	88.16%	213
Octopus sinensis	XP_036369268.1	99%	4,00E-99	88.16%	234
Octopus bimaculoides	XP_014781573.1	99%	4,00E-99	88.16%	213
Biomphalaria glabrata	XP_013094746.1	90%	2,00E-98	95.68%	281
Pecten maximus	XP_033747500.1	98%	5,00E-98	87.42%	216
Lottia gigantea	XP_009051818.1	98%	2,00E-97	88.00%	207
Mizuhopecten yessoensis	XP_021344822.1	98%	3,00E-76	70.86%	214
Pecten maximus	XP_033748005.1	99%	4,00E-74	68.63%	215

Fonte: Dados do autor

Conforme observado nas tabelas descritas acima, as proteínas Ran, Exportinas 1 e T, DGCR8 de *P. canaliculata* e *P. maculata* possuem alta similaridade com seus ortólogos, tamanho compatível com a dimensão média das proteínas ortólogas e domínios conservados característicos de acordo com a função da proteína. Desta forma, foi possível identificar estas prováveis proteínas nas espécies em estudo.

Assim, foi possível identificar 7 prováveis proteínas da via de miRNAs em *P. canaliculata,* Pca69814_c1_g2 (Argonauta 2), Pca61914_c0_g1(Argonauta), Pca67531_c1_g2 (Dicer), Pca67887_c3_g1 (Drosha), Pca59979_c0_g1 (DGRC8), Pca65193_c2_g1 (Exportina 1), Pca52466_c2_g3 (Ran) e 8 prováveis proteínas em *P. maculata,* Pma63715_c0_g1 (Argonauta 2), Pma60501_c2_g16 (Argonauta), Pma62419_c0_g1 (DGRC8), Pma59467_c0_g1 (Exportina 1), Pma63331_c0_g1 (Exportina T), Pma54972_c0_g1 (Ran), Pma51393_c0_g1 (Drosha), Pma95401_c0_g1 (Dicer).

5 CONCLUSÃO

A pesquisa por miRNAs em *P. canaliculata* e *P. maculata* a partir de bancos de dados públicos atendeu as características estruturais e termodinâmicas de miRNAs identificados, para elucidação e predição de miRNAs conservados nos genomas das espécies. Foi possível predizer 296 formas maduras de miRNAs conservados em *P. canaliculata* e 316 miRNAs em *P. maculata*. A submissão de alguns pré-miRNAs ao alinhamento múltiplo de sequências revelou alta similaridade com seus ortólogos.

Como mostrado, os resultados indicam que algumas sequências analisadas de mir-bantam e mir-1990 de *P. canaliculata* e *P. maculata* mostraram 100% de identificação com alguns de seus ortólogos. A fim de evidenciar a conservação de miRNAs preditos e*m P. maculata* e *P. canaliculata* foram construídas árvores filogenéticas, nas quais foram confirmadas a estreita proximidade entre as espécies estudadas e os moluscos da família Ampullariidae, *Lanistes nyassanus* e *Marisa cornuarietis*. A sequência madura de miRNAs se mostrou conservada evolutivamente e esta similaridade aumenta à medida que o grau de parentesco entre as espécies se acentua. Da mesma forma, foi verificada alta similaridade com a estrutura secundária dos precursores de miRNAs preditos e ortólogos, intrinsecamente semelhantes entre moluscos e a família Ampullariidae.

Uma das formas de demonstrar a presença de miRNAs no genoma das espécies: *P. canaliculata* e *P. maculata* é verificar a sua maquinaria de processamento. Dessa forma, foi possível identificar a via de processamento de miRNAs nas espécies: *P. canaliculata* e *P. maculata*, destacando várias prováveis proteínas da via, tais como Argonauta, Drosha, Dicer, DGCR8, XPO1, dentre outras. A caracterização das proteínas da via através de alinhamento múltiplo de sequências, análise de domínios conservados, analogia com similaridade e dimensionamento com ortólogos permitiu confirmar através de análise filogenéticas a conservação e a presença de prováveis proteínas detectas no proteoma das espécies.

As características inerentes de *P. canaliculata* e *P. maculata* referentes ao aspecto invasivo puderam ser ratificadas por mapeamento de características por lócus gênico em estudos anteriores. Estudos de expressão diferencial de miRNAs poderão ser conduzidos na região dos tentáculos cefálicos e palpos labiais, para ratificar estes dados, bem como estabelecer o gene GRL101 como alvo para os miRNAs descobertos neste trabalho. Dessa forma, poderá ser correlacionado o aspecto invasivo das espécies *P. canaliculata* e *P. maculata* a partir da regulação pós-transcricional de miRNAs em genes específicos das espécies.

Por fim, os nossos resultados ampliam o estudo dos miRNAs em moluscos, proporcionando novos desafios para compreensão de processos essenciais relacionados à invasividade das espécies *P. canaliculata* e *P. maculata.*

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CROSSTALK OF CONSERVATION OF SEQUENCES OF miRNAs AND ENZYMATIC MACHINERY OF miRNAs PRODUCTION IN AMPULLARIIDAE

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ABSTRACT:

BACKGROUND: The Ampullariidae family of molluscs is an emerging model for evolutionary studies due to its high diversity, ancient history and wide geographic distribution. miRNAs are essential for the development of the organism, as they help and act in the control of gene expression. The analysis of miRNAs in molluscs, especially in the family Ampullariidae, is especially important due to the low representativeness of analyzed species and the possibility of analyzing the conservation of miRNAs among ampullariids species. **OBJECTIVE:** Identification and characterization of miRNAs (precursors and matures) and their processing pathway genes. **METHODS:** Computational prediction and characterization of miRNAs and genes involved in the miRNA pathway were performed using public database of *Lanistes nyassanus*, *Marisa cornuarietis*, *Pomacea canaliculata* and *Pomacea maculata*. The *in silico* analysis was performed using a robust algorithm to identify and characterize miRNAs and their precursors in genome of ampullariids species. To search for the putative proteins involved in the miRNA biogenesis the putative proteomes from 4 Ampullariids species and Blastp tool were used. Characterization of conserved protein domains was performed using the PFAM and CDD. Phylogenetic analyzes were performed for the ampullariids miRNA precursors and their orthologs and also for the putative ampullariids proteins involved in the miRNA pathway and their orthologs using MEGA program. **FINDINGS:** 141 pre-miRNAs and 162 mature miRNAs were identified in the genome of *L. nyassanus*, 279 pre-miRNAs and 297 mature miRNAs in the genome of *M. cornuarietis*, 269 pre-miRNAs and 296 mature miRNAs in the genome of *P. canaliculata* and 299 pre-miRNAs and 316 mature miRNAs in the genome of *P. maculata*. We identified and characterized 24 putative key proteins involved in the miRNA pathway including Argonaute, DICER, DROSHA and EXPORTIN protein families in the predicted proteome of the 4 ampullariids. The data obtained in this work will support studies of phylogeny, population divergence, speciation and patterns of diversity in the Ampullariidae Family. **MAIN CONCLUSIONS**: The searching for novel miRNAs and their processing pathway genes in 4 species of ampullariids was able to predict new structures expanding the study of miRNAs in molluscs and in Ampullariidae family, as well as open an avenue to study the roles of miRNAs in the organisms.

Keyword: miRNAs. Computational Analysis. Snails. Ampullariidae. Genome

INTRODUCTION

Molluscs of the Ampullariidae family are freshwater snails found in tropical and subtropical regions of Africa, Asia and the Americas. The animals belong to the subclass Caenogastropoda, which comprises about 50,000 species and approximately 60% of Gastropod species (Hayes et al., 2009; Strong et al., 2008). As they are a family that derived from the subclass Caenogastropoda in early evolution (Cowie, 2015) ampullariids are an excellent model to study speciation and adaptation (Hayes et al., 2009).

The scarcity of sequenced molluscs genomes was identified by the Global Invertebrate Genomics Alliance as a problem for species characterization (Voolstra et al., 2017). There are four ampullariids with sequenced genomes including the Old World (*Lanistes nyassanus*) and New World (*Pomacea canaliculata*, *Pomacea maculata* and *Marisa cornuarietis*) clades that can be the object of comparative genomic studies. The division between New and Old World lineages is consistent with the time of separation from Gondwana, around 120 million years ago (Jokat et al., 2003; Sun et al., 2019). The availability of the genome and transcriptome of the cited species makes it promising to study gene regulation, intrinsically the post- transcriptional regulation mediated by miRNAs. Almost all biological processes, including celldifferentiation, tissue development and cell metabolism can be modulated by miRNAs. This regulation of gene expression is carried out through a fine control, where miRNAs and their silencing pathways carry out a specific regulation of gene expression and maintenance of genome integrity (Bartel, 2009). This may help to better understand the biology of the ampullariids studied and their adaptive properties. miRNAs have a low homoplasy, that is, their presence in two different species is more likely to be linked to a common ancestry, thus correlating the distribution of miRNAs in evolutionary relationships of the species (Kenny et al., 2015). This will allow a better understanding of the biology of the Ampullariidae family in molluscs, particularly its medical and economic importance for the humanity of ampullariids species.

P. canaliculata is involved in the transmission of the helminth *Angiostrongylus cantonensis* that causes eosinophilic meningitis in humans. Molluscs are the intermediate hosts when they are contaminated by ingestion of Larva L1. The infecting larva lodges in the Central Nervous System in humans, causing eosinophilic meningitis or ocular angiostrongyliasis in the eye ball (Luessi et al., 2009; Wang et al., 2012, 2008). Despite eosinophilic meningitis usually course in humans with low severity there may be some severe cases of the disease with severe neurological sequelae, coma and death (especially in children) (Chen et al., 2014; Lindo et al., 2002; Tsai et al., 2001). miRNAs are able to modulate the immune system of molluscs in response to stress caused by infectious processes (Picone et al., 2017).

P. canaliculata is used as a source of dietary protein in Southeast Asia. Ingestion of undercooked animals can transmit *A. cantonensis* causing eosinophilic meningitis in humans. In the Nueva Ecija region of the Philippines, the prevalence of *P. canaliculata* contaminated with nematodes was 17.75% (Cawas et al., 2020). The lack of awareness of the population is notorious, being considered a neglected disease and an important public health problem in the region (Dalton et al., 2017).

In mainland China, eosinophilic meningitis caused by *A. cantonensis* is considered an emerging infectious disease (Lv et al., 2008). Epidemiological evidence indicates that *P. canaliculata* is becoming the most prevalent intermediate host of *A. cantonensis* in this country, due to its high susceptibility to the parasite and its wide environmental tolerance (Yang et al., 2013). A major outbreak occurred in Beijing in 2006 with 160 cases of human angiostrongyliasis causing eosinophilic meningitis. The fact occurred due to the consumption of contaminated *P. canaliculata* imported from regions in southern China (Wang et al., 2008). Thus, we note the extreme importance of more epidemiological studies in the country regarding this serious public health problem. The study of the biology and adaptability of *P. canaliculata* has the same degree of importance for better management and control of eosinophilic meningitis.

P. canaliculata is described as an invasive species and was ranked among the top 100 pests worldwide. The species is resistant to elevated temperatures and drought (Matsukura et al., 2009; Yusa et al., 2006) . *P. maculata* has physiological and adaptive characteristics similar to *P. canaliculata*, both species being recognized for their rapid propagation and population increase, constituting a serious threat to the balance of an ecosystem. The economic costs of damage, control and repair can be immense (Pimentel et al., 2005).

P. canaliculata is considered an agricultural pest in many parts of Asia (Wang et al., 2020). In Indonesia, when introduced into rice plantations, it causes a 15% drop in production (Novarino, 2011). The dispersion of *P. canaliculata* in crops planted in humid regions causes serious damage in Southeast Asia (Carlsson et al., 2016) in Japan (Yusa, 1999) and also in China (Yang et al., 2013).

In 2003, China's State Department of the Environment ranked *P. canaliculata* in a list of 16 invasive pests. Since the mollusc was introduced to China around 1980, there is no accurate data on the spread of *P. canaliculata* in agriculture. Over the next twenty years, the molluscs spread widely in the country and as the snails moved into agriculture, control measures against *P. canaliculata* were implemented (Yang et al., 2013). More epidemiological studies are needed regarding the impact of *P. canaliculata* on agriculture in China. Work on the biology of the snail such as adaptability and environmental plasticity is also needed. The study of miRNAs in the species is an important step towards understanding part of the physiology of *P. canaliculata*, for possible mechanisms of population control of the species in agriculture.

The inherent skills of *P. canaliculata* and *P. maculata* referring to the invasive aspect could be confirmed by mapping characteristics by gene locus. The ability of *P. canaliculata* and *P. maculata* species to colonize and occupy new territories is attributed in part to genes that encode the G protein family (GRL101) (Sun et al., 2019), highly expressed in tentacles and cephalic palps (chemoreception in aquatic snails) (Schultz and Adema, 2017) . Still referring to invasiveness, studies have suggested that the voracious appetite of *P. canaliculata* and *P. maculata* species are attributed to the expansion of gene families related to cellulose digestion (Sun et al., 2019).

L. nyassanus and *M. cornuarietis* are snails related to freshwater (Strong et al., 2008) and have divergences regarding their adaptability. While *M. cornuarietis* is better able to resist factors such as low temperature and hypoxia (Matsukura et al., 2016; Mu et al., 2018) *L. nyassanus* has a low adaptability and is endemic to Lake Nyasa, or Malawi, geographically located between Malawi, Tanzania and Mozambique (Cowie, 2015; Dohrn, 1865; Van Bocxlaer, 2017).

Since miRNA-mediated post-transcriptional silencing performs several regulatory aspects in animals, it is necessary to understand the processing mechanisms of the miRNA pathway and their respective genes. The identification of major miRNA biogenesis factors including, AGO2, DGCR8, DICER, DROSHA, RAN and XPO5 was elucidated based on homologous sequence search in several molluscs (Huang et al., 2021) and in *Biomphalaria Glabrata* by researchers from our group (Queiroz et al., 2017).

Sun et al., published in 2019 the sequencing of the total genome of four species of Ampullariidae, *L. nyassanus*, *P. canaliculata*, *P. maculata* and *M. cornuarietis*. From then on, it was putative to carry out the mapping of characteristics by gene locus. Given the relevant regulatory aspect of non-protein coding RNAs in gene expression, specifically miRNAs, it is necessary to study mature and precursor miRNAs, as well as the genes involved in the miRNA processing pathway. In this work, mature and precursor miRNAs and the genes involved in miRNA pathway were identified and characterized using the genome of *L. nyassanus, P. canaliculata*, *P. maculata* and *M. cornuarietis*.

MATERIALS AND METHODS

PREDICTION AND CHARACTERIZATION OF MATURE AND PRECURSOR MIRNAS

The genomes of *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* were retrieved from the NCBI database (https://www.ncbi.nlm.nih.gov/genome). A robust algorithm was used to predict miRNAs, precursor and mature forms. The sequences were processed for identification and characterization of predicted mature miRNAs and their precursors, following the methodology established by Gomes *et al.* (2011).

Genome sequences with potential tendency to formation of hairpin structures or similarity to precursor miRNAs were obtained using the Einverted program (EMBOSS tool) and the BLASTn program (NCBI), for identification of homologous miRNAs from miRBase (http: //www.mirbase.org/). Then, the sequences were submitted through a series of filters, selecting, according to the desired parameters, those that corresponded to putative-putative precursors of miRNAs. These filters were based on conserved characteristics of precursor miRNAs and unwanted sequences were discarded. The filters used were the Minimum Free Energy (MFE) (Gruber et al., 2008; Hofacker, 2009), GC content (Zhou et al., 2009), homology with mature miRNAs already identified. Finally, sequences with high similarity to genes encoding proteins, repetitive elements and non-coding RNAs, except miRNAs, were discarded.

The putative precursors of miRNAs were compared to their orthologs at various levels. The sequences of pre-miRNAs and their respective miRNAs were submitted to multiple alignment, using the tools ClustalX 2.1(Larkin et al., 2007) and RNAalifold (Bernhart et al., 2008). Alignment patterns for ClustalX 2.1 were based on adjusted parameters (gap opening: 22.50; gap extension: 0.83). Secondary structures were obtained using the RNAfold platform (Gruber et al., 2008; Hofacker, 2009). The phylogenetic analysis of the putative pre-miRNAs of *L. nyassanus, M. cornuarietis, P. canaliculata* and *P. maculata* was carried out using MEGA program version X (Tamura et al., 2007) and the Neighbor-joining method applying the Kimura two-parameters model ("Kimura-twoparameters"), to stimulate the divergence between the sequences (N Saitou and Nei, 1987). Consensus trees were obtained using a bootstrap of 5000 replicates.

PREDICTION AND CHARACTERIZATION OF PROTEINS INVOLVED IN MIRNAS BIOGENESIS

In silico identification and characterization of putative miRNA pathway proteins were performed using the amino acid sequences of the proteins from the model organisms *Biomphalaria glabrata, Drosophila melanogaster* and *C. elegans* and the Blastp tool on the ampubase website (https://www.comp.hkbu.edu.hk/~db/AmpuBase/index.php#&panel1-2). Using PFAM (31.0) (Mistry et al., 2021) and conserved Domains CDD (Marchler-Bauer et al., 2017, 2015, 2011; Marchler-Bauer and Bryant, 2004; Shennan Lu et al., 2020) the putative domain architectures of the ampullariids proteins were established. Analysis of the active sites were used for the proteins Argonaut, Dicer and Drosha using Clustal X 2.1 and Weblogo.

Phylogenetic analysis was performed using the MEGA version X program (Kumar et al., 2018), after alignment in default parameters, using the Neighbor-Joining method (Naruya Saitou and Nei, 1987) and the JTT model. The statistical reliability of each branch in the generated tree was evaluated using a 2000-replica bootstrap.

RESULTS

IDENTIFICATION AND CHARACTERIZATION OF PUTATIVE PROTEINS INVOLVED IN MIRNA PATHWAY IN AMPULLARIIDS SPECIES

The predicted proteome of the species *L. nyassanus*. *M. cornuarietis*, *P. canaliculata* and *P. maculata* were retrieved from the NCBI database (National Center for Biotechnology Information - http://.ncbi.nlm.nih.gov/) to identify and characterize the putative proteins responsible for the biogenesis of the miRNAs. The identification was based on conservation, using as parameters protein length, conserved domains, active sites conservation and phylogeny analysis. Lny23421, Lny25187,Lny28936, Lny28171, Lny18581, Lny14710, Lny12572, Lny27692, Lny22624, Lny12572, Lny29643, Lny27624, Lny9931, Mco26370, Mco27042, Mco25093, Mco22837, Mco24030, Mco81614, Mco51286, Mco13761, Mco21592, Mco12258, Pca61914, Pca59652, Pca69814, Pca67531, Pca67887,Pca62316, Pca65193, Pca60969, Pca65193, Pca65324, Pca59979, Pca69254, Pma35144, Pma60501, Pma63715, Pma65850, Pma51393, Pma59467, Pma63331, Pma32975, Pma62419, Pma51302 and Pma61707 putative proteins involved in miRNA pathway were identified in predicted proteome of *L. nyassanus*, *M. cornuarietis*, *P. canaliculate* and *P. maculate*, respectively [Supplementary data I – (Supplementary Tables 1,2,3 and 4)]. The key proteins of the miRNA pathway were selected, Argonaute, DICER, DROSHA and Exportin, and thus used for further analysis.

Ten putative Argonaute proteins were identified in the predicted proteome of the ampullariids species, being 3 sequences for *L. nyassanus* (Lny23421_c0_g1, Lny25187_c0_g1 and Lny28936_c7_g7), 3 sequences for *M.* cornuarietis (Mco25093_c0_g2, Mco26370_c0_g1, Mco27042_c1_g1), 2 sequences for *P. canaliculata* (Pca69814_c1_g2, Pca61914_c0_g1) and 2 sequences for *P. maculata* (Pma63715_c0_g1_, Pma60501_c2_g16). The sequences Lny23421_c0_g1, Mco26370_c0_g, Pca69814_c1_g2 and Pma63715_c0_g1 exhibited six conserved domains, while the protein sequences Lny25187_c0_g1, Lny28936_c7_g7, Mco25093_c0_g2, Mco27042_c1_g1, Pca61914_c0_g1 e Pma60501_c2_g16 displayed only the PAZ and PIWI domains (Figure 1).

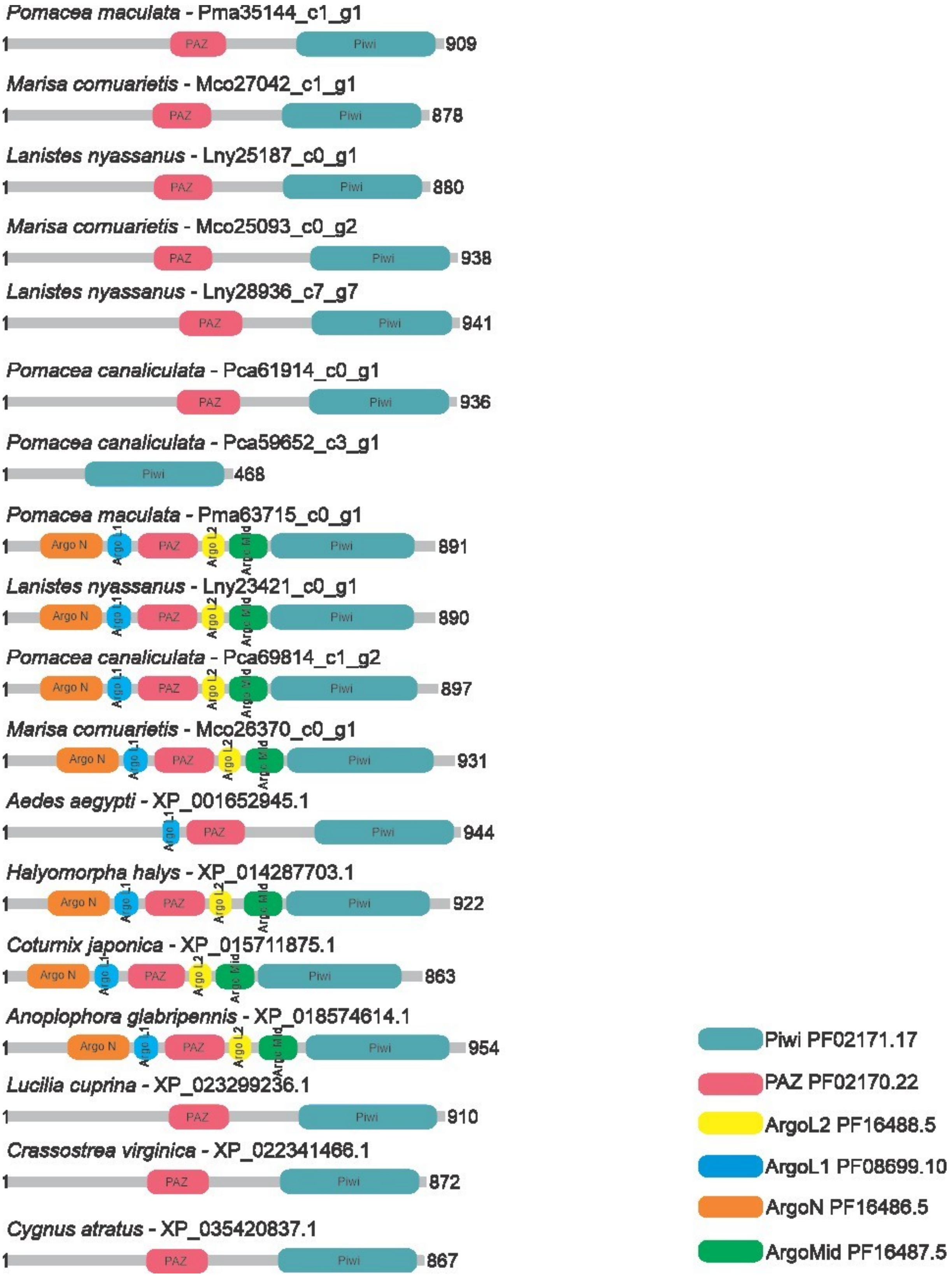


Figure 1: Argonaute's Conserved Domains, *L. nyassanus. M. cornuarietis, P. canaliculata, P. maculata* and orthologs.

The PIWI domain of these proteins presented a catalytic triad formed by DDH (aspartic acid, aspartic acid and histidine). The global alignment of the PIWI domains of the ampullariids

Argonaute proteins and their orthologs showed a conservation of amino acid sequences, being accentuated in the surroundings of the catalytic sites (Figure 2).

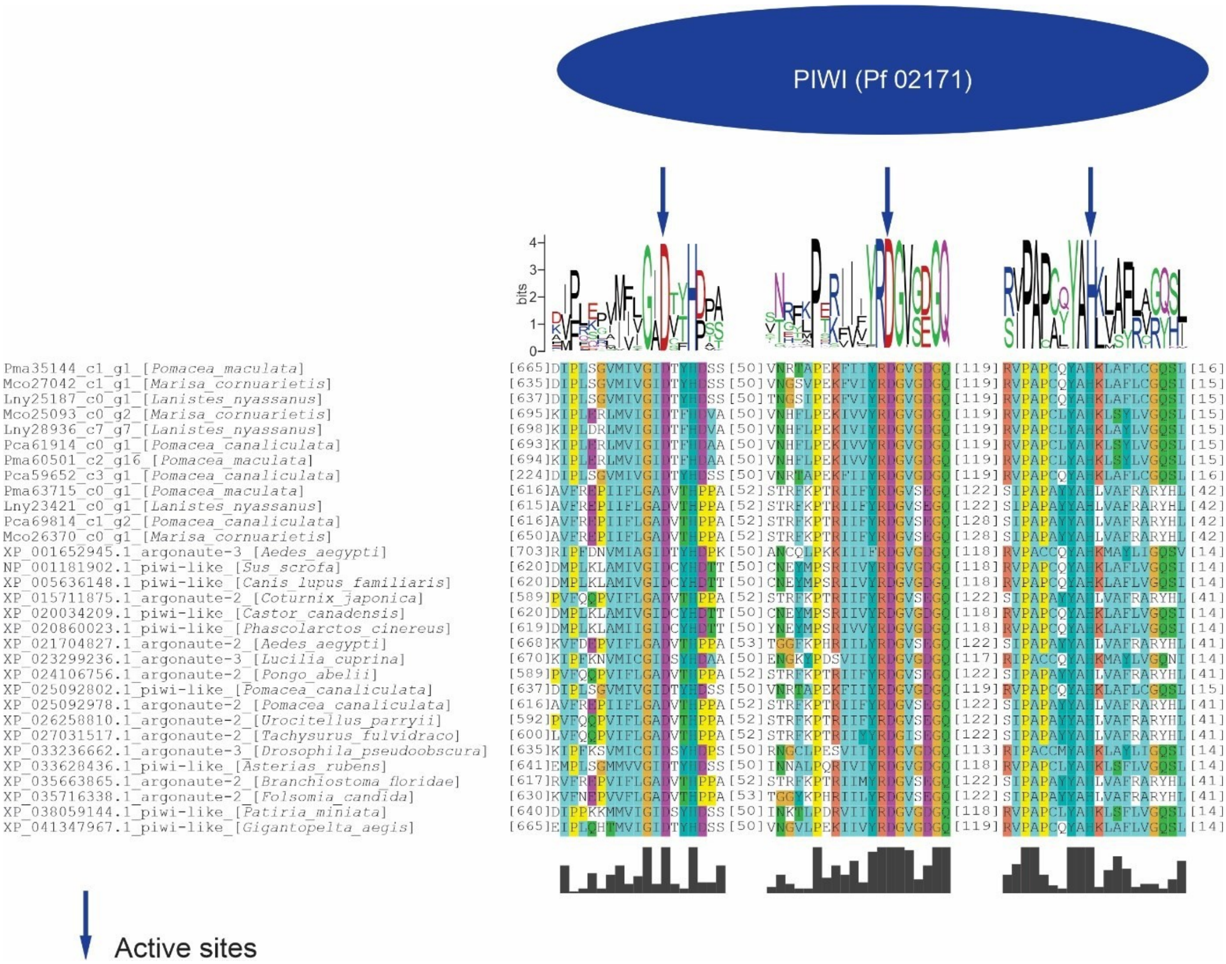


Figure 2: Global alignment analysis of the PIWI domain of Argonaute proteins de *L. nyassanus, M. cornuarietis, P. canaliculata* and *P. maculata*

The two subfamilies, AGO and PIWI compose the AGO protein family. The predicted sequences of *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* were used in the phylogenetic tree and their orthologs, in order to emphasize the conservation of these putative proteins identified in the Ampullariids species and wide distribution among deuterostomes and protostomes species. The distribution of Argonaute and PIWI proteins corroborated the distribution of species in the tree of life. As expected, the ampullariids species grouped together with the phylum Molluscs. The evolutionary proximity of species ampullariids, *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* is represented in the tree (Figure 3).

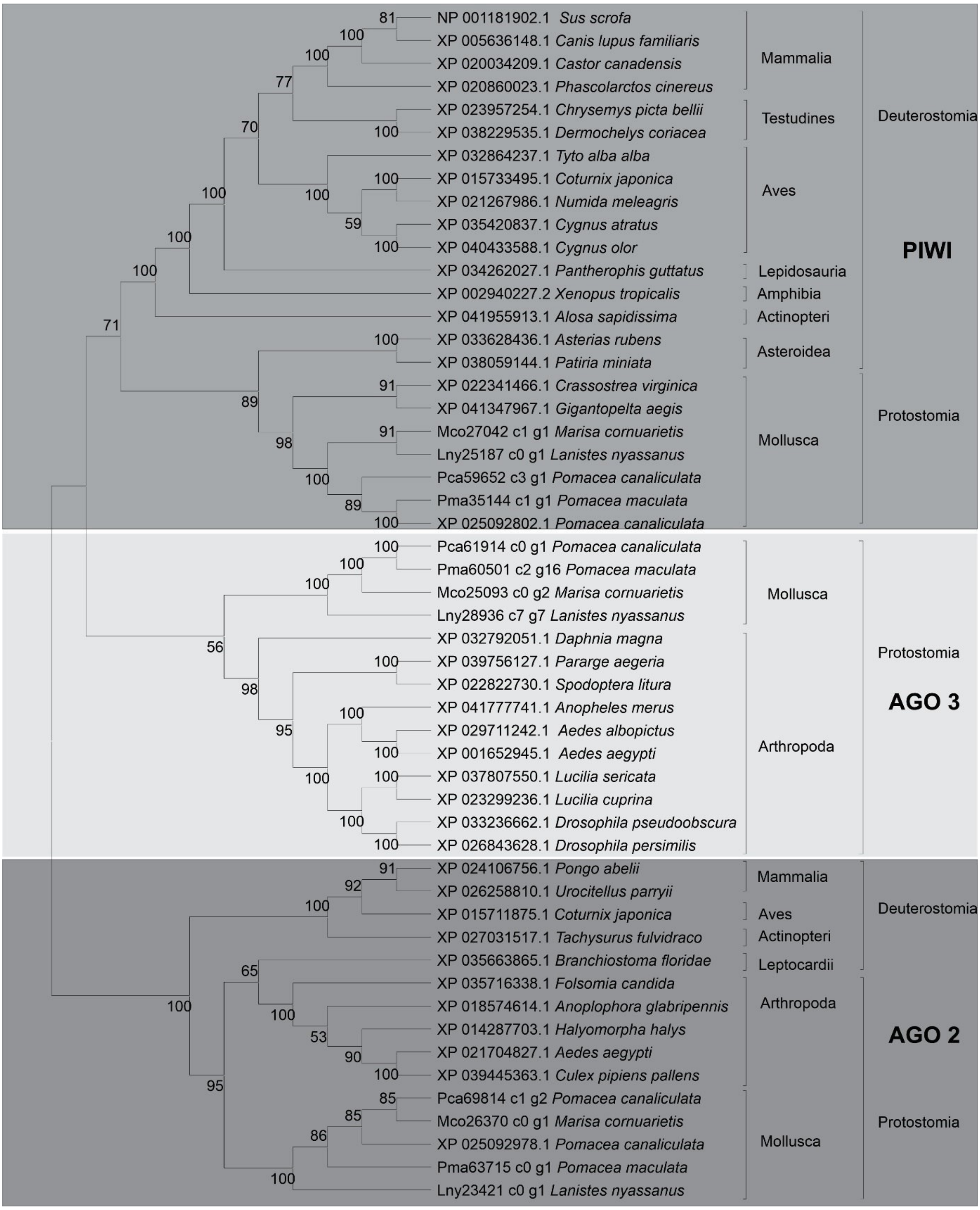


Figure 3: Phylogenetic analysis of AGO proteins, *L. nyassanus, M. cornuarietis, P. canaliculata* e *P. maculata* and their orthologs.

Five putative Drosha and two putative Dicer proteins were identified and characterized in the ampullariids species. For Drosha, 1 sequence for *L. nyassanus* (Lny18581_c0_g2), 1 sequence for *M. cornuarietis* (Mco22837_c2_g1), 1 sequence for *P. canaliculata* (Pca67887_c3_g1) and 2 sequences for *P. maculata* (Pma51393_c0-g1 and Pma65850_c2g4) were identified. The amino acid sequences of Pca67887_c3_g1 and Mco22837_c2_g1 had three conserved domains, Lny18581_c0_g2 and Pma65850_c2_g4 had two conserved domains, while the protein sequence Pma51393_c0_g1 had only one conserved domain. For Dicer 1 amino acid sequence were found for *P. canaliculata* (Pca67531_c1_g2), showing 4 conserved domains and for *L. nyassanus* (Lny28171_c0_g1) showing only 3 conserved domains (Figure 4 and Figure 5).

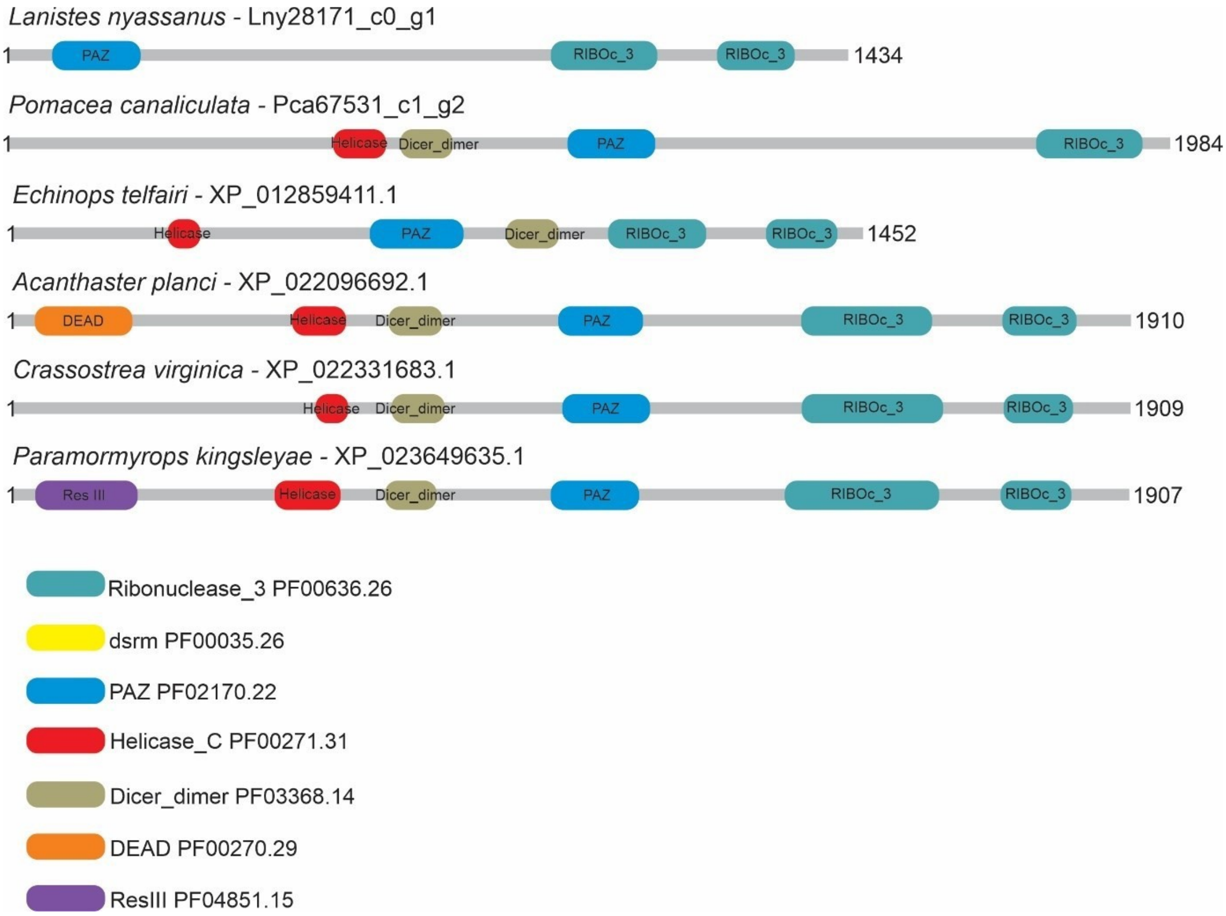


Figure 4: Analysis of conserved domains Dicer for *L. nyassanus, M. cornuarietis, P.* canaliculata, *P. maculata* and their orthologs.

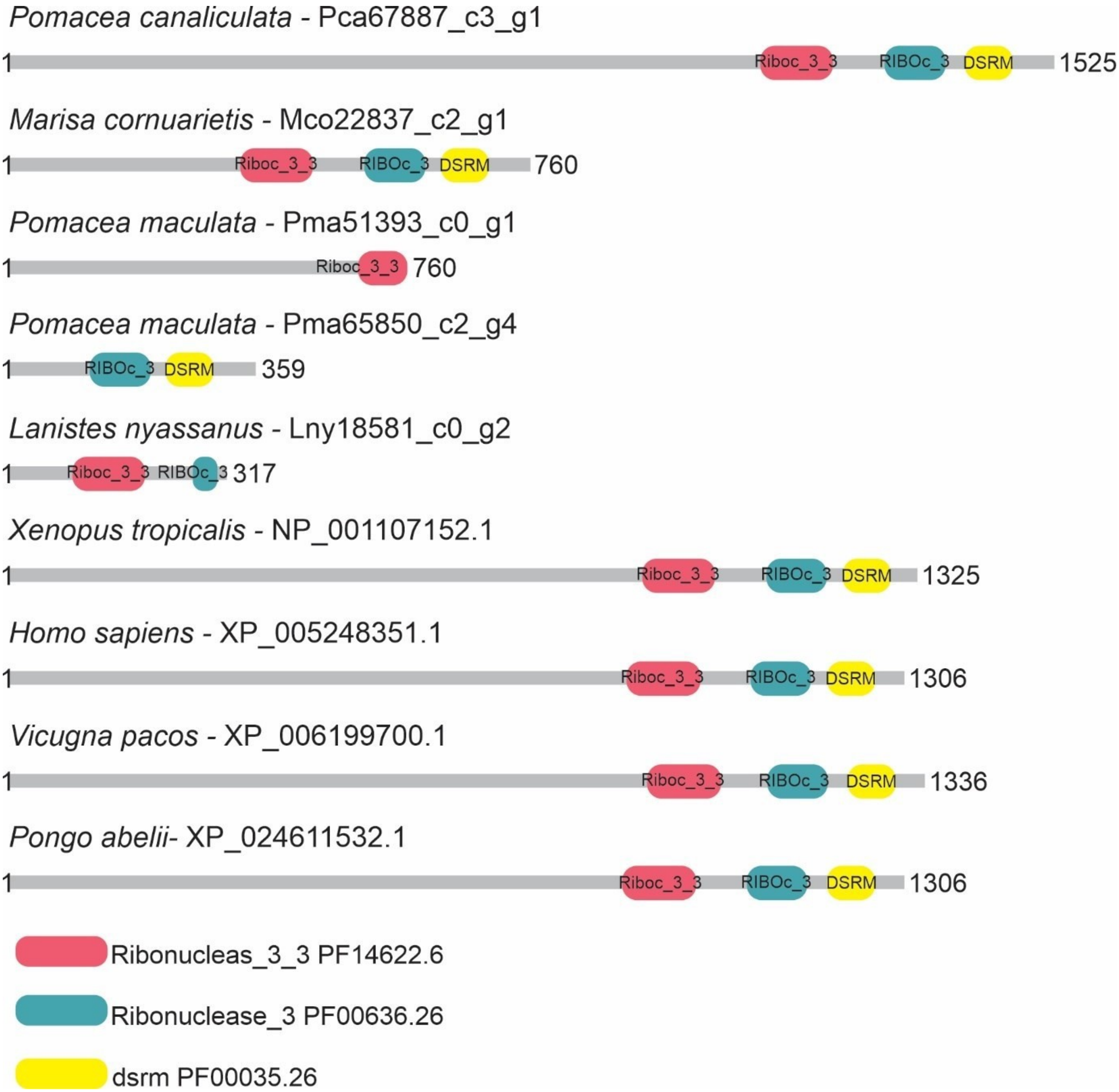


Figure 5: Analysis of conserved domains Drosha for *L. nyassanus*, *M. cornuarietis*, *P. canaliculata*, *P. maculata* and their orthologs.

In the analysis of the Drosha/Dicer proteins, only one conserved domain of Ribonuclease III was identified in the ampullariids studied. The Riboc I domains presented the catalytic amino acids in the conserved site regions (EDDE): glutamic acid (E), aspartate (D), aspartate (D) and glutamic acid (E). This was verified in the sequences Drosha, Lny18581_c0_g2, Mco22837_c2_g1, Pca67887_c3_g1 and Dicer, Lny28171_c0_g1. Note the conservation of amino acids in the active site itself and in the surroundings of the region (Figure 6).

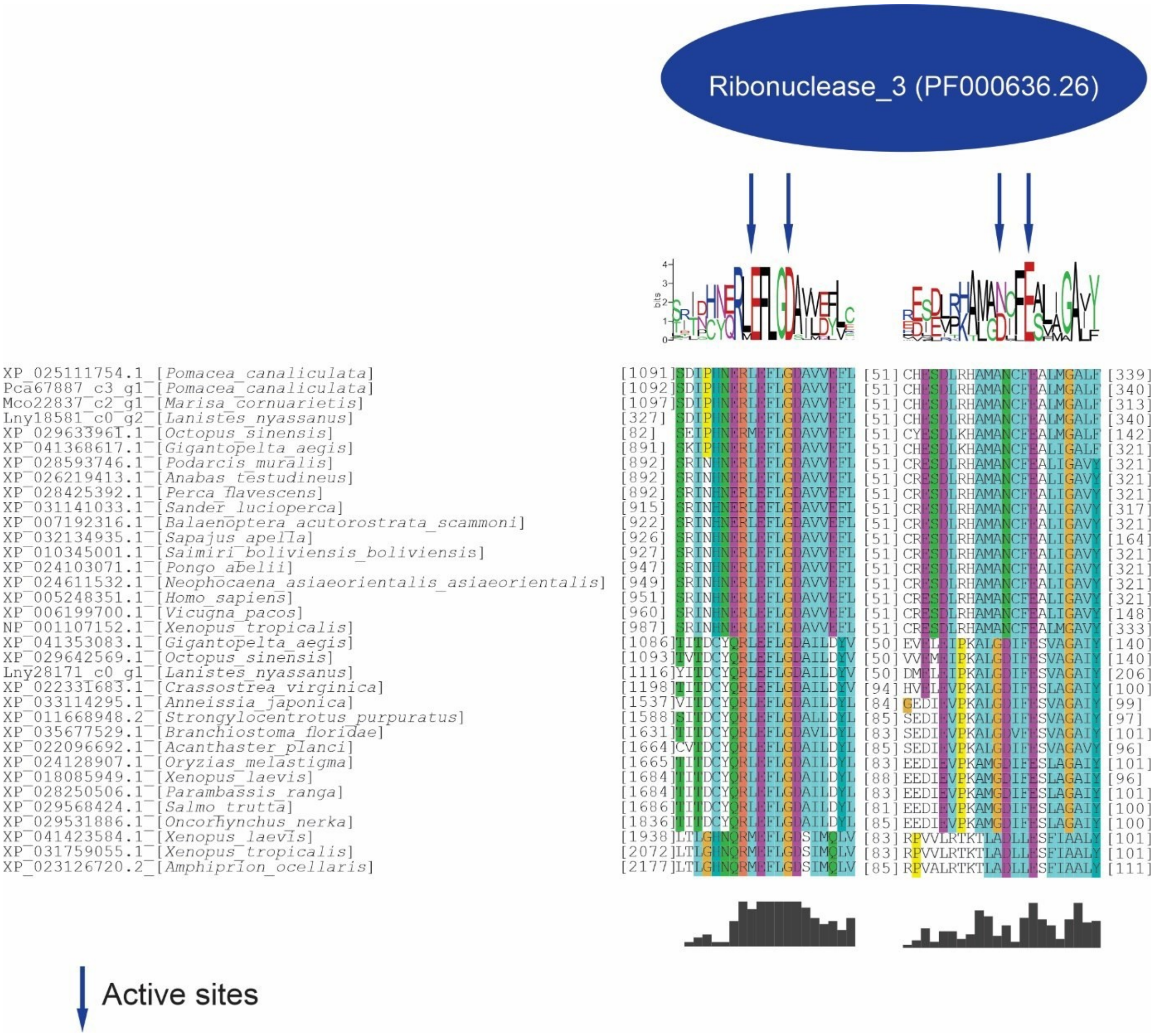


Figure 6: Analysis of the conserved domains of Ribonuclease III (Riboc) of the Drosha and Dicer proteins from ampullariids species.

Phylogenetic analysis of the predicted proteins DROSHA and DICER showed the evolutionary relationship with their orthologs. Proteins from *L. nyassanus*, *M. cornuarietis*, *P. canaliculata*, *P. maculata* grouped in the Molluscs clade, revealing close similarity among the ampullariids studied. The disposition of the orthologs of the putative DICER and DROSHA in the clades in the tree corroborated with the tree of life, with a visible distinction between Deuterostomes and Protostomes for the two classes of RNase (Figure 7).

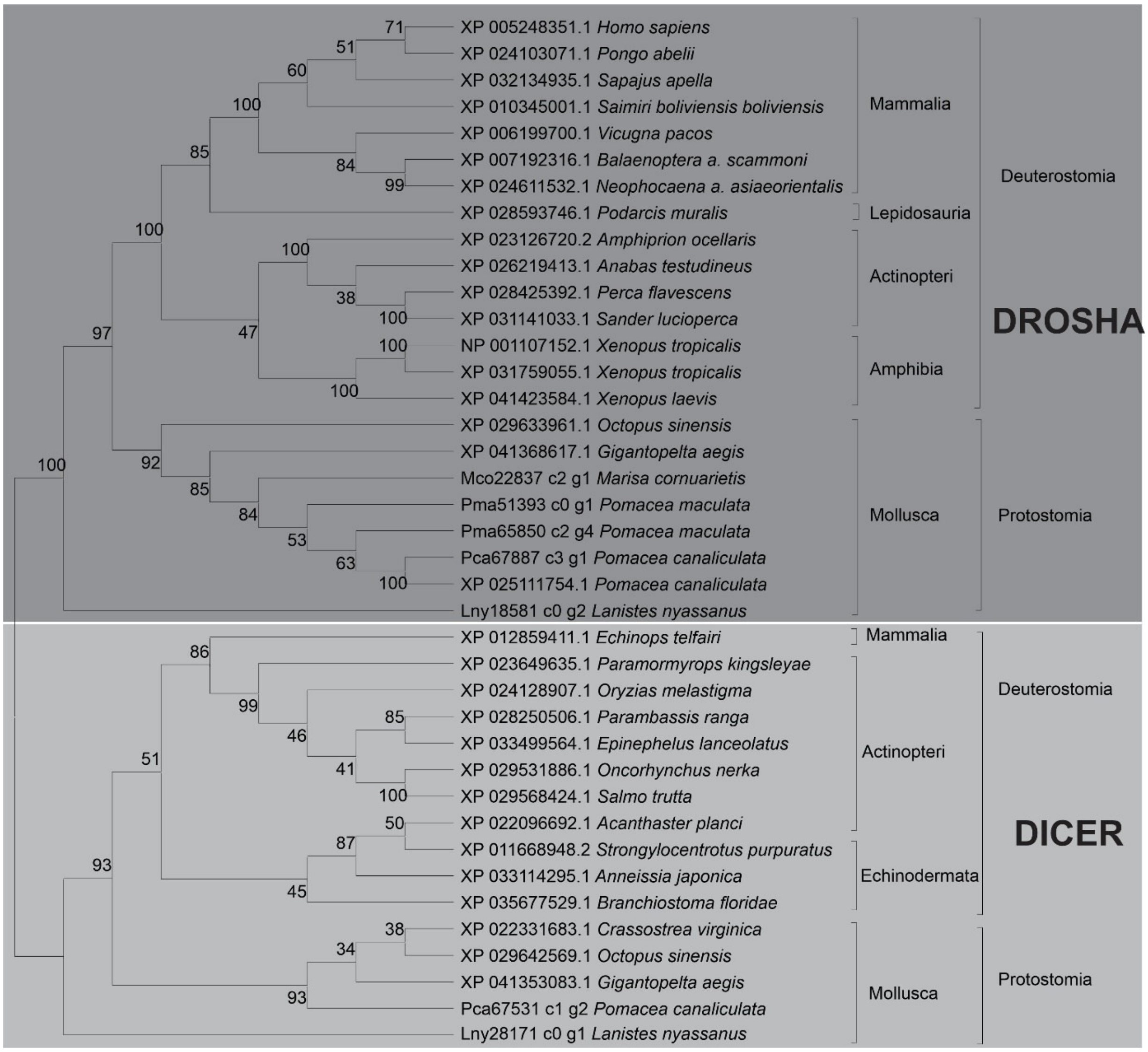


Figure 7: Phylogenetic analysis of DICER and DROSHA proteins from ampullariids and their orthologs.

Similarity of the predicted protein Drosha from *L. nyassanus*, *M. cornuarietis* and *P. canaliculata* with model organism *D. melanogaster* and evolutionarily close organisms. The percent identity, query coverage, length of the predicted protein Drosha from *P. canaliculata* are similar among closely related organisms (Table 1).

Code	**Query coverage**	**E Value**	**Identity %**	**Length of string**
NP_477436.1 Drosha [*D. melanogaster*]	100%	0.0	100,00	1327
XP_013069067.1 (*B. glabrata*)	73%	0.0	54,91	1469
XP_005107264.1 (*Aplysia californica*)	72%	0.0	54,81	1528
XP_041368618.1 (*G. aegis*)	72%	0,0	59,09	1412
Pca67887_c3_g1 (*P.canaliculata)*	66%	0.0	57,75	1525
Mco22837_c2_g1 (*M. cornuarietis*)	56%	0.0	60,96	760

Lny18581_c0_g2 (*L. yassanus*)	27%	$7e^{-150}$	69,57	317

Table 1: Putative DROSHA of *L. nyassanus*, *M. cornuarietis* and *P. canaliculata* cover against NP_477436.1 DROSHA [D. melanogaster], E-Value, % of String Identity and Length.

Comparison of the Dicer protein from the model organism *D. melanogaster* with the putative Dicer protein in *L. nyassanus* and *P. canaliculata* and evolutionarily close organisms. Once again, the predicted protein Dicer of *P. canaliculata* showed greater similarity with its orthologs when compared to the other Ampullariidae (Table 2).

Código	**Cobertura da *query***	**E Value**	**Identidade %**	**Comprimento da sequência**
NP_524453.1 Dicer-1 [*D. melanogaster*]	100%	0.0	100%	2249
XP_012942349.1 (*A. californica*)	77%	$2e^{-141}$	50%	2503
XP_013067888.1 *(B. glabrata*)	74%	$2e^{-148}$	51,90%	2332
XP_021347347.1 (*M. yessoensis*)	72%	$7e^{-152}$	52,87%	2284
XP_041353083.1 *(G. aegis*)	70%	$2e^{-150}$	50,26%	2063
Pca67531_c1_g2 (*P. canaliculata*)	71%	$7e^{-140}$	45.10%	1985
Lny28171_c0_g1 (*L. nyassanus*)	38%	$2e^{-162}$	53,32%	1434

Table 2: Putative Dicer *L. nyassanus* e *P. canaliculata*, coverage against NP_524453.1 Dicer-1 [D. melanogaster], E-Value, % identity and string length

Nine putative protein sequences belonging to the Exportin protein family (XPO-5, XPO-1, XPO-T) were identified and characterized in the predicted proteome of ampullariid species; XPO-5: *L. nyassanus* (Lny14170_c0_g1), *P. canaliculata* (Pca62316_c0_g2); XPO-1: *L. nyassanus* (Lny12572_c0_g1), *M. cornuarietis* (Mco24030_c0_g2), *P. canaliculata* (Pca65193_c2_g1) and *P. maculata* (Pma59467_c0_g1), XPO-T: *L. nyassanus* (Lny27692_c0_g2), *M. cornuarietis* (Mco81614_c0_g2) *P. maculata* (Pma63331_c0_g1). The sequences of XPO-5, Lny14170_c0_g1 and Pca62316_c0_g2, exhibited 2 domains. For the XPO-1 protein sequences, the putative proteins Lny12572_c0_g1, Mco24030_c0_g2 and Pma59467_c0_g1 displayed six conserved domains, except for the Pca65193_c2_g1, which exhibited only five conserved domains. Likewise, XPO-T putative proteins Lny27692_c0_g2, Mco81614_c0_g2 and Pma63331_c0_g1 showed one conserved domain (Figure 8).

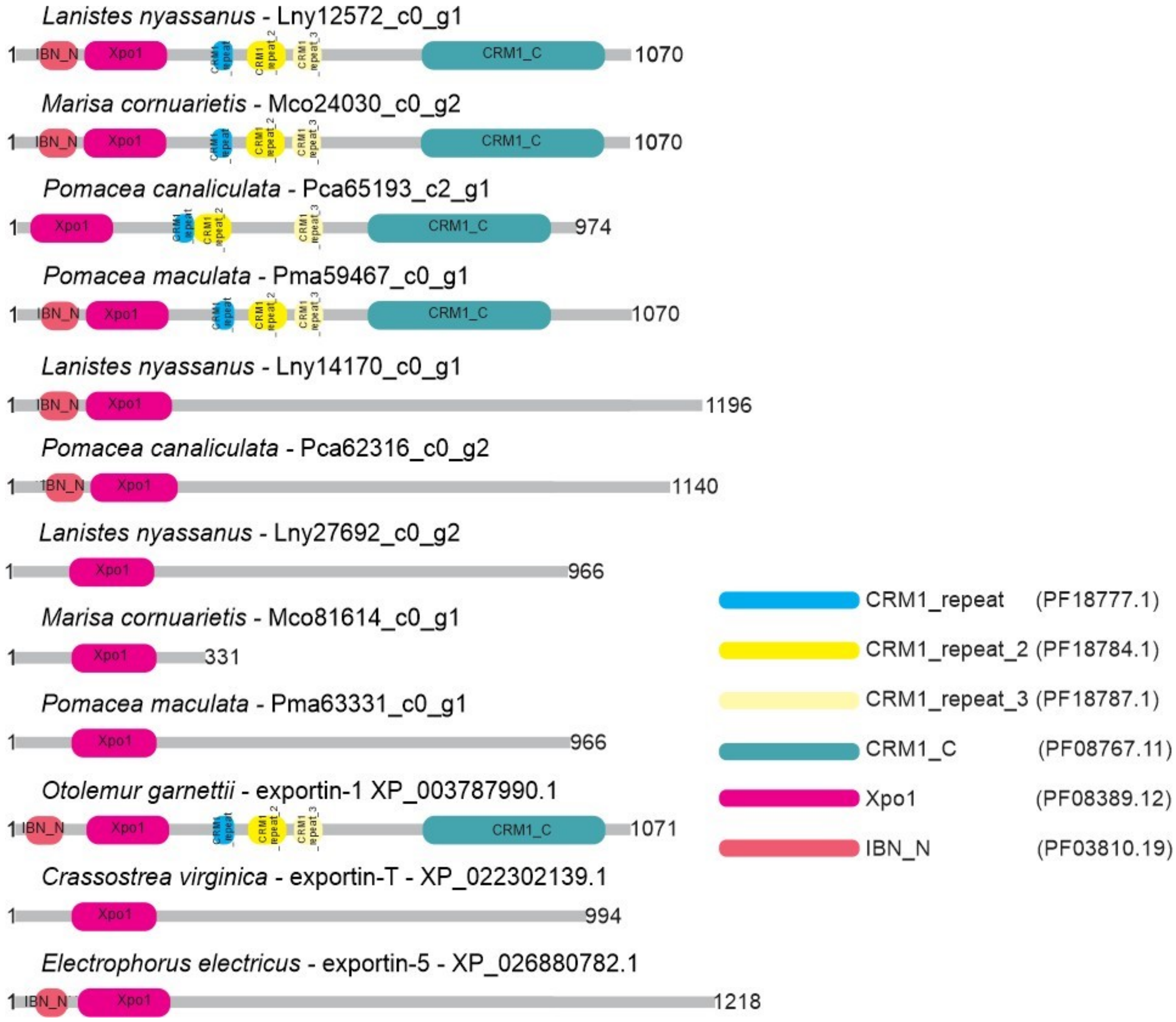


Figure 8: Distribution of conserved domains in putative proteins of the Exportin family in ampullariids species and orthologs.

In the phylogenetic analysis of putative proteins of the Exportin family (XPO-5, XPO-1 and XPO-T) the evolutionary relationship with their orthologs was verified. Protein sequences from *L. nyassanus*. *M. cornuarietis*, *P. canaliculata* and *P. maculata* were grouped in the phylum Molluscs, confirming once again a close proximity between the ampullariids species, as expected by the degree of relatedness of the species. The phylogenetic tree presented three distinct clades (XPO-5, XPO-1 and XPO-T clades), as well as species belonging to the Protostome and Deuterostome groups, corroborating with the literature (Figure 9).

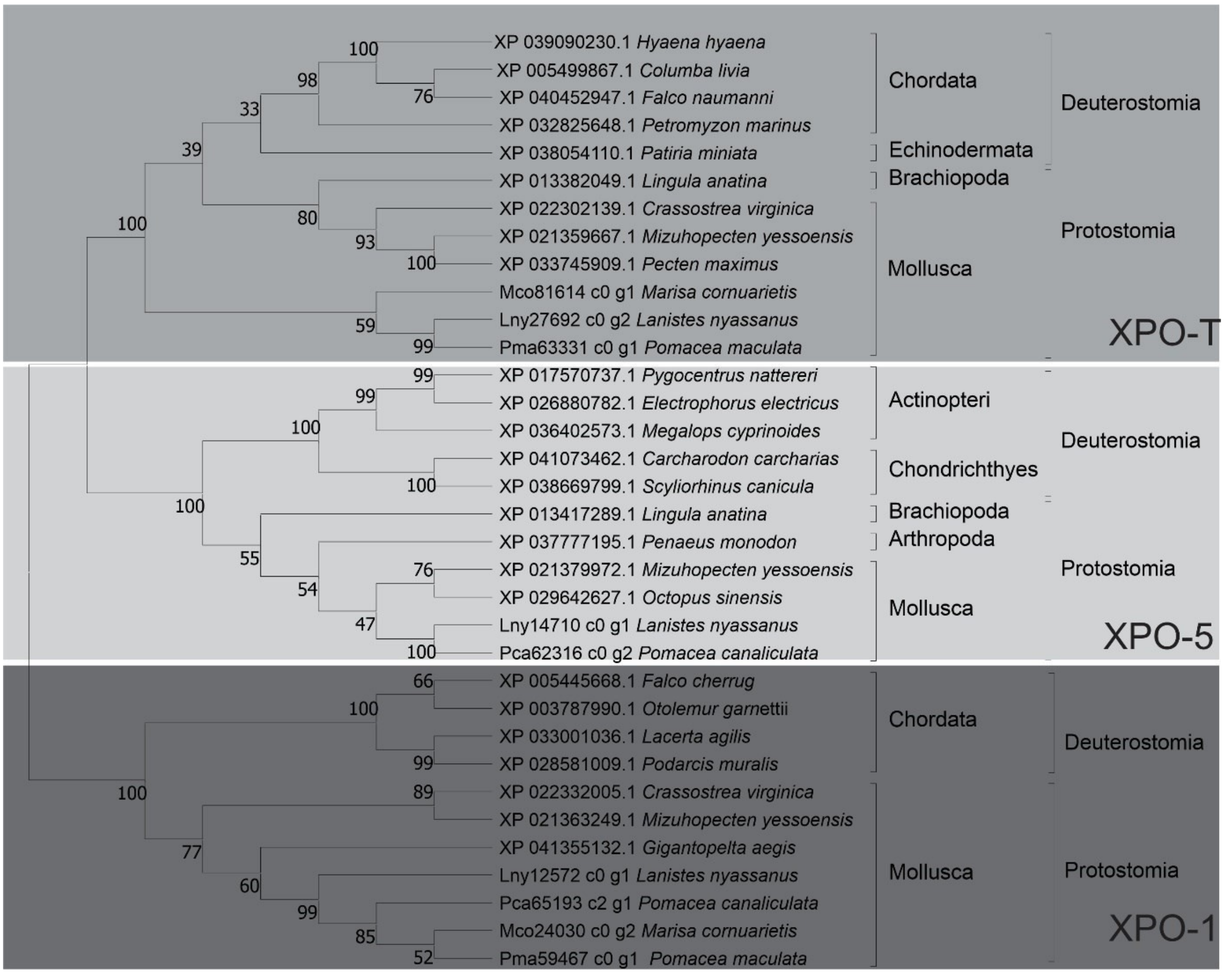


Figure 9: Phylogenetic distribution of putative proteins, XPO-5 XPO-1 and XPO-T identified in *L. nyassanus. M. cornuarietis, P. canaliculata* e *P. maculata* and their orthologs.

PREDICTION AND CHARACTERIZATION OF PERCURSOR AND MATURE MIRNAS.

A robust algorithm was applied to the genome sequences of the four ampullariids species to identify mature miRNAs and their precursors. 141 pre-miRNAs and 162 miRNAs of *L. nyassanus*, 279 pre-miRNAs and 297 miRNAs of *M. cornuarietis*, 269 pre-miRNAs and 296 miRNAs of *P. canaliculata*; 299 pre-miRNAs and 316 miRNAs of *P. maculata* were predicted [Supplementary data II – (Supplementary Tables 5,6,7,8 and 9)].

An accurate and stringent structural and thermodynamic analysis were used to identify and characterize conserved miRNAs(Y. Q. Zhang et al., 2009). All pre-miRNAs identified were analyzed for these particular characteristics [Supplementary data II - (Supplementary Tables 5,6,7 and 8)].The pre-miRNAs for *L. nyassanus* exhibited MFE (minimum free energy) with a mean of -30.30 kcal/mol, with values between -18.50 and -50.90 kcal/mol; for *M. cornuarietis* the mean MFE of -28.05 kcal/mol, with values between -18.50 and -67.10 kcal/mol; for *P. canaliculata* the mean of MFE -28.70 kcal/mol, with values between -18.70 and -48.4 kcal/mol; for *P. maculata* the mean of -27.55 kcal/mol, with values between -18.50 and -52.7 kcal/mol (Table 3).

Species	**Average size of precursors (nucleotides)**	**Precursor size variation (nucleotides)**	**Mean value MFE - Kcal/mol**	**Variation MFE - Kcal/mol**
L. nyassanus	87.04	66 to 100	-30,3	(-50.90 to -18.50)
M. cornuarietis	88.16	65 to 100	-28,05	(-67.10 to -18.50)
P. canaliculata	89,55	63 to 144	-28,7	(-48.40 to -18.70)
P. maculata	89.27	65 to 100	-27,55	(-52.70 to 18.50)

Table 3: Average size and size variation of precursors, average minimum free energy (MFE) and MFE variation of precursors in 4 species Ampullariidae

The precursors showed mean values for the guanine-cytosine (GC) content for *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata*; 41.26%, 41.64%, 41.70%, 40.74%, respectively. The maximum and minimum values were respectively 69.05 and 20.25% for *L. nyassanus*, 67.03% and 20.20% for *M. cornuarietis*, 68.29% and 20.83% for *P. canaliculata*, 68, 75% and 20.40% for *P. maculata*. In this study, the average size of miRNA precursors for *L. nyassanus*, *M. cornuarietis*, *P. canaliculata*, *P. maculata* were respectively; 87.04 nucleotides, 88.16 nucleotides, 89.55 nucleotides and 89.27 nucleotides. Size range of pre-miRNAs for *L. nyassanus* (66 to 100 nucleotides), *M. cornuarietis* (65 to 100 nucleotides), *P. canaliculata* (63 to 144 nucleotides), *P. maculata* (65 to 100 nucleotides).

The sequences of miRNAs have been shown to be evolutionarily conserved (De Wit et al., 2009). This conservation is accentuated in the seed region of the mature miRNAs, being considered important for recognition of their mRNA targets (Lewis et al., 2005). All mature miRNAs identified in this work showed 100% identity in the seed regions (position 2 to 8 nucleotides) with the structure of their respective ortholog deposited in miRBase, on which the homology comparison was based.

The mature miRNAs identified in this work had an average of 22.22 nucleotides in *L. nyassanus*, 21.92 nucleotides in *M. cornuarietis*, *21.86 nucleotides* in *P. canaliculata* and 21.77 nucleotides for *P. maculata*, ranging in size from 17 to 25 nucleotides for all species analyzed.

The Argonaute protein has a preference for the uracil nucleotide in the first position of the miRNA sequence at the 5' end to be loaded, to be inserted into the RISC complex (Seitz et al., 2011). In this work mature miRNAs from L. nyassanus, M. cornuarietis. P. canaliculata and

P. maculata respectively presented uracil as the main nucleotides in the first position; 41.97%, 44.44%, 41.2% and 42.40% (uracil), 17.28%, 17.84%, 20.9% and 17.8% (cytosine), 26.54%, 22.56%, 20.4% and 24.5% (adenine) and 14.19%, 15.15%, 17.5% and 15.3% (guanine) [Supplementary data II - (Supplementary table 9)].

The presence of miRNAs in two different species is more likely to be linked to a common ancestry, thus correlating the distribution of miRNAs in evolutionary relationships of species (Kenny et al., 2015). In order to emphasize the conservation of these putative miRNAs identified in the ampullariid species, phylogenetic trees were constructed. The global alignment and secondary structure characterization of the precursors of miRNAs was performed, to show the similarity of the sequences with their orthologs already identified. The taxa that were used as a parameter for the selection of specific are Bilateria, Protostomes, Lophotrocozoa and Molluscs. The selection of pre-miRNAs based on these taxons was performed having as parameter the miRNAs prevalent simultaneously in the 4 ampullariids species (Table 4). The characterization of miRNAs by taxons was shown in figures 1 to 11 and Supplementary data III - supplementary figures 1 to 34

Tax	Species	Pre-miRNAs
Bilateria	*L. nyassanus*	lny-mir-33, lny-mir-190, lny-mir-252a, lny-mir-281, lny-mir-981
	M. cornuarietis	mcr-mir-33, mcr-mir-190, mcr-mir-252a, mcr-mir-281, mcr-mir-981
	P. canaliculata	pcn-mir-33, pcn-mir-190, pcn -mir-252a, pcn-mir-281, pcn-mir-981
	P. maculata	pmc -mir-33, pmc-mir-190, pmc-mir-252a, pmc-mir-281, pmc-mir-981
Protostomies	*L. nyassanus*	lny-bantam, lny-mir-2a, lny-mir-67, lny-mir-750
	M. cornuarietis	mcr-bantam, mcr-mir-2a, mcr-mir-67, mcr-mir-750
	P. canaliculata	pcn-bantam, pcn-mir-750
	P. maculata	pcr-bantam, pcr-mir-2a, pcr-mir-67, pcr-mir-750
Lophotrocozoa	*L. nyassanus*	lny-mir-1990, lny-mir-745a, lny-mir-96b, lny-mir-1994
	M. cornuarietis	mcr-mir-1990, mcr-mir-96b, mcr-mir-1994
	P. canaliculata	pcn-mir-1990, pcn-mir-745a, pcn-mir-1992, pcn-mir-96b, pcn-mir-1994
	P. maculata	pmc-mir-1990, pmc-mir-745a, pmc-mir-1992, pmc-mir-96b, pmc-mir-1994
Molluscs	*L. nyassanus*	lny-mir-1985, lny-mir-12096b
	M. cornuarietis	mcr-mir-1985, mcr-mir-12096b
	P. canaliculata	pcn-mir-1985, pcn-mir-12096b

	P. maculata	Pmc-mir-1985, Pmc-mir-12096b

Table 4: miRNAs selected by taxon

The precursors of miR-33, Bilateria-specific-miRNA, were found in this study: lny-miR-33, mcr-miR-33, pcn-miR-33, pmc-miR-33. All precursors found the 4 species showed two mature miRNAs, 3p and 5p. The sequences of the precursors showed high conservation in the primary and secondary structures. The similarity of the ampullariids miRNA structures were found to be highly conserved each other (Figure 10 and 11).

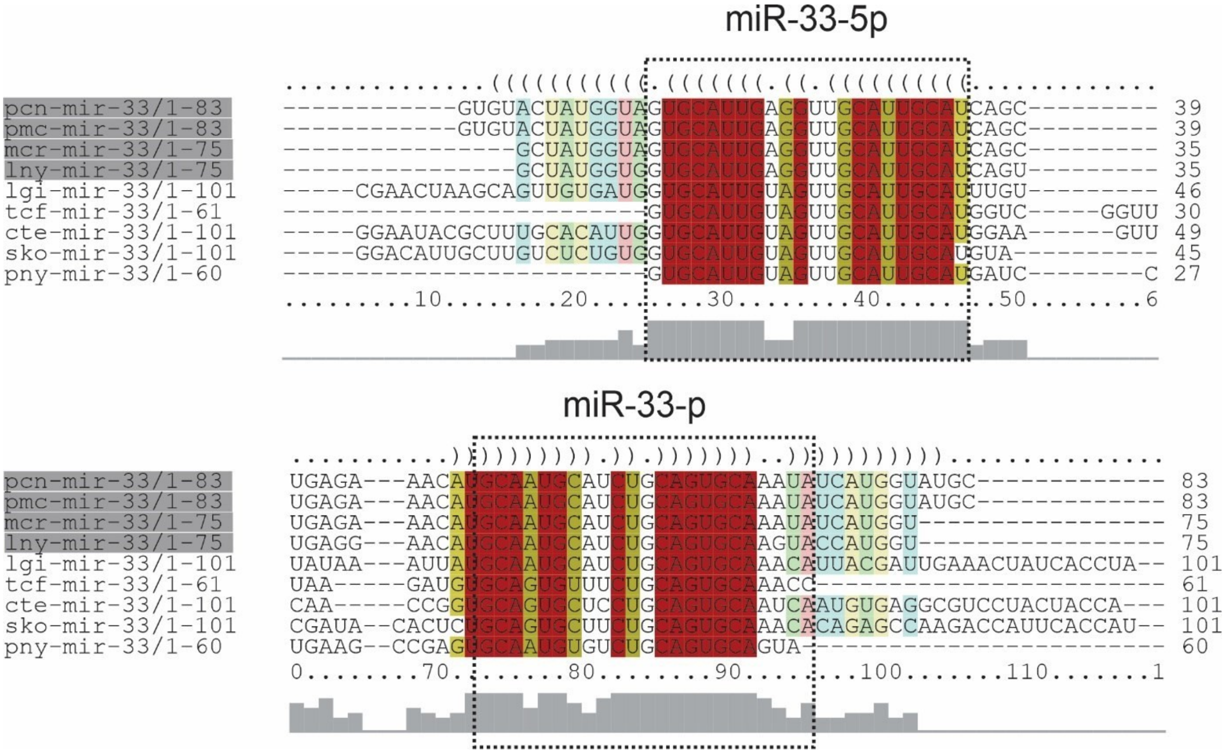


Figure 10: Alignment of mir-33 in *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculata*, mcr - *M. cornuarietis*, lny- *L. nyassanus*, lgi - *L. gigantea*, tcf - *Triops cancriformis*, cte - *Capitella teleta*, sko - *Saccoglossus kowalevskii*, pny - *Pundamilia nyererei*.

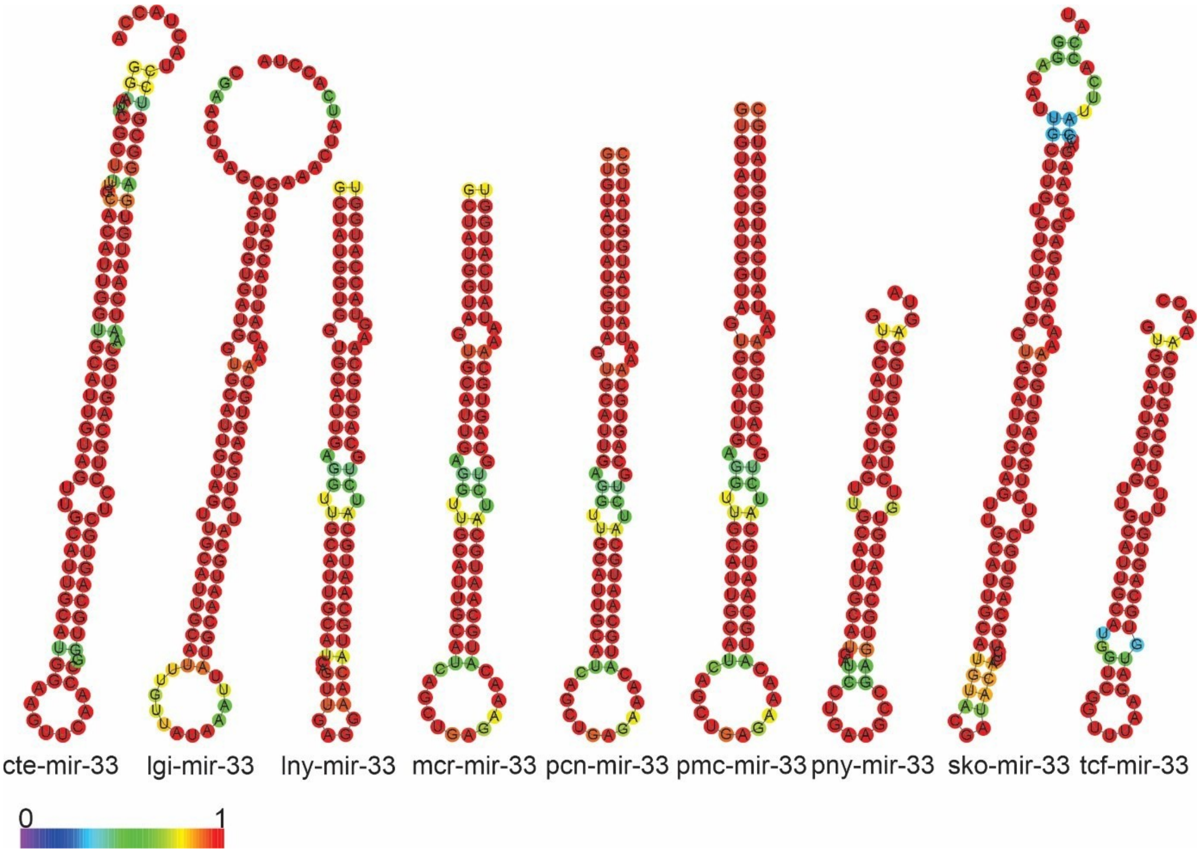


Figure 11: Secondary structure of mir-33 in *L. Nyassanus, M. cornuarietis*, *P. canaliculata*, *P. maculata* and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculata*, mcr - *M. cornuarietis*, lny - *L. nyassanus*, lgi - *L. gigantea*, tcf - *Triops cancriformis*, cte - *C. teleta*, sko - *Saccoglossus kowalevskii*, pny - *Pundamilia nyererei.*

The Protostome and Deuterostome clades were observed in the phylogenetic. The first clade was subdivided into Lophotrochozoa and Ecdysozoa, being represented respectively by molluscs and arthropods. In the phylum Molluscs, the presence of gastropods was observed, where the ampullariids species were grouped. Deuterostome organisms were represented by clades; mammals, birds and amphibians, verifying wide distribution in phylogenetic clades. This distribution corroborated with the tree of life (Figure 12).

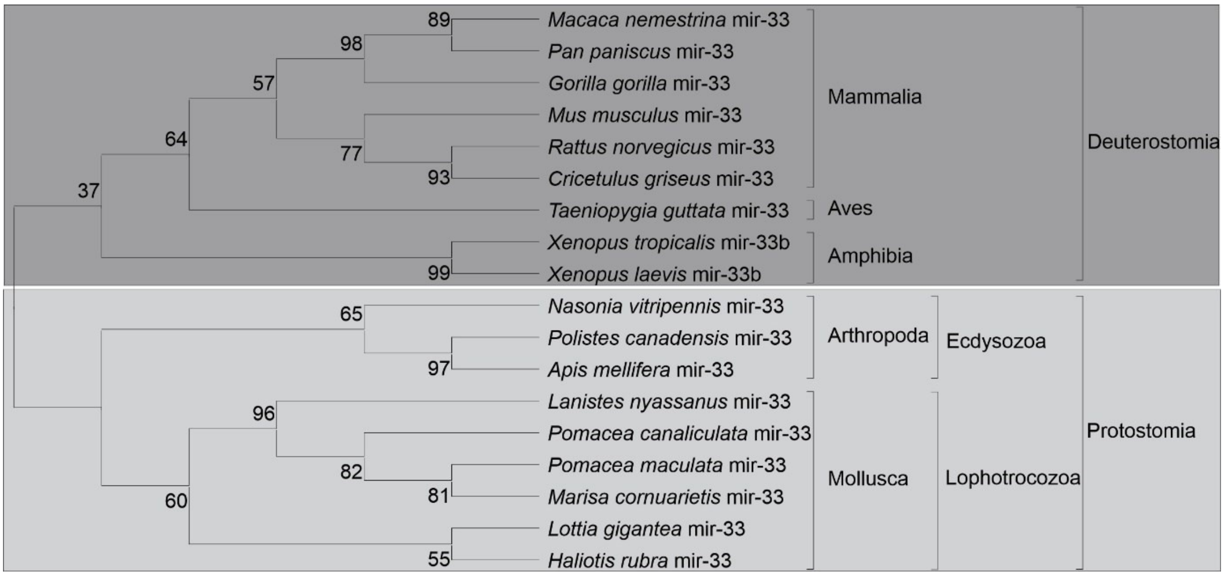
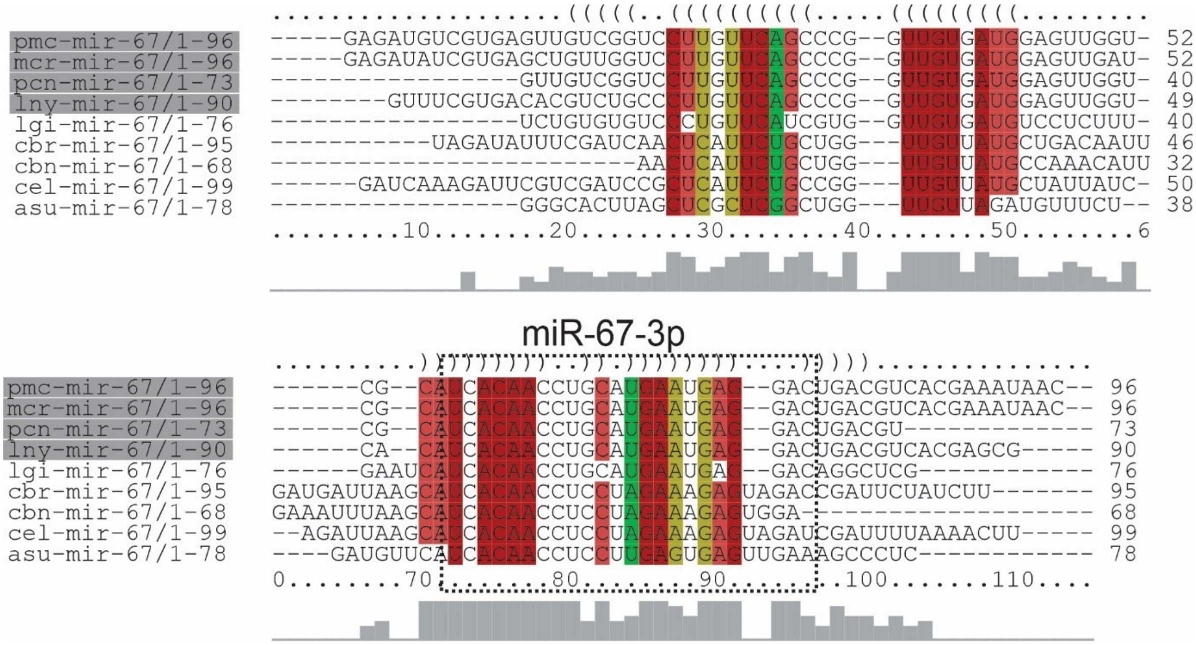


Figure 12: mir-33 phylogenetic distribution for *L. Nyassanus, M. cornuarietis P. canaliculata*, *P. maculata* and their orthologs.

The precursors of miRNAs (protostomes specific) lny-miR-67, mcr-miR-67, pcn-miR-67, pmc-miR-67 and a mature miRNA were identified for each Ampullariidae species analyzed. The primary structures of the mi-67 family in *L. nyassanus*, *M. cornuarietis*. *P. canaliculata* and *P. maculata* were highly conserved between themselves and their orthologs, especially in the region of mature miRNAs. Note the extreme similarity in the secondary structures of the precursors in *M. cornuarietis* and *P. maculata* (Figure 13 andFigure 14).



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                    ....................(((((..((((((((((.....(((((((((.........
pmc-mir-67/1-96     -----GAGAUGUCGUGAGUUGUCGGUCCUUGUUCAGCCCG--GUUGUGAUGGAGUUGGU-  52
mcr-mir-67/1-96     -----GAGAUAUCGUGAGCUGUUGGUCCUUGUUCAGCCCG--GUUGUGAUGGAGUUGAU-  52
pcn-mir-67/1-73     -----------------GUUGUCGGUCCUUGUUCAGCCCG--GUUGUGAUGGAGUUGGU-  40
lny-mir-67/1-90     --------GUUUCGUGACACGUCUGCCCUUGUUCAGCCCG--GUUGUGAUGGAGUUGGU-  49
lgi-mir-67/1-76     -----------------UCUGUGUGUCCCUGUUCAUCGUG--GUUGUGAUGUCCUCUUU-  40
cbr-mir-67/1-95     ------------UAGAUAUUUCGAUCAACUCAUUCUGCUGG---UUGUUAUGCUGACAAUU 46
cbn-mir-67/1-68     -------------------------AACUCAUUCUGCUGG---UUGUUAUGCCAAACAUU  32
cel-mir-67/1-99     ------GAUCAAAGAUUCGUCGAUCCGCUCAUUCUGCCGG---UUGUUAUGCUAUUAUC-  50
asu-mir-67/1-78     -----------------GGGCACUUAGCUCGCUCGGCUGG---UUGUUAGAUGUUUCU--  38
                    ........10........20........30........40........50........6

                                      miR-67-3p
                    ..........)))))))))..)).))))))))....)))))...............
pmc-mir-67/1-96     ------CG--CAUCACAACCUGCAUGAAUGAG--GACUGACGUCACGAAAUAAC--  96
mcr-mir-67/1-96     ------CG--CAUCACAACCUGCAUGAAUGAG--GACUGACGUCACGAAAUAAC--  96
pcn-mir-67/1-73     ------CG--CAUCACAACCUGCAUGAAUGAG--GACUGACGU-------------  73
lny-mir-67/1-90     ------CA--CAUCACAACCUGCAUGAAUGAG--GACUGACGUCACGAGCG-----  90
lgi-mir-67/1-76     ------GAAUCAUCACAACCUGCAUGAAUGAG--GACAGGCUCG------------  76
cbr-mir-67/1-95     GAUGAUUAAGCAUCACAACCUCCUAGAAAGAGUAGACCGAUUCUAUCUU-------  95
cbn-mir-67/1-68     GAAAUUUAAGCAUCACAACCUCCUAGAAAGAGUGGA--------------------  68
cel-mir-67/1-99     --AGAUUAAGCAUCACAACCUCCUAGAAAGAGUAGAUCGAUUUUAAAACUU-----  99
asu-mir-67/1-78     ----GAUGUUCAUCACAACCUCCUUGAGUGAGUUGAAAGCCCUC------------  78
                    0.......70........80........90........100.......110....
```

Figure 13: mir-67 alignment to *L. nyassanus*, *M. cornuarietis*. *P. canaliculata* e *P. maculata* and their orthologs; asu - *Ascaris suum*, cbn - *Caenorhabditis brenneri*, cbr - *Caenorhabditis briggsae*, cel - *C. elegans*, lny - *L. nyassanus*, lgi - *L. gigantea*, mcr - *M. cornuarietis*, pcn - *P. canaliculata*, pmc - *P. maculata*.

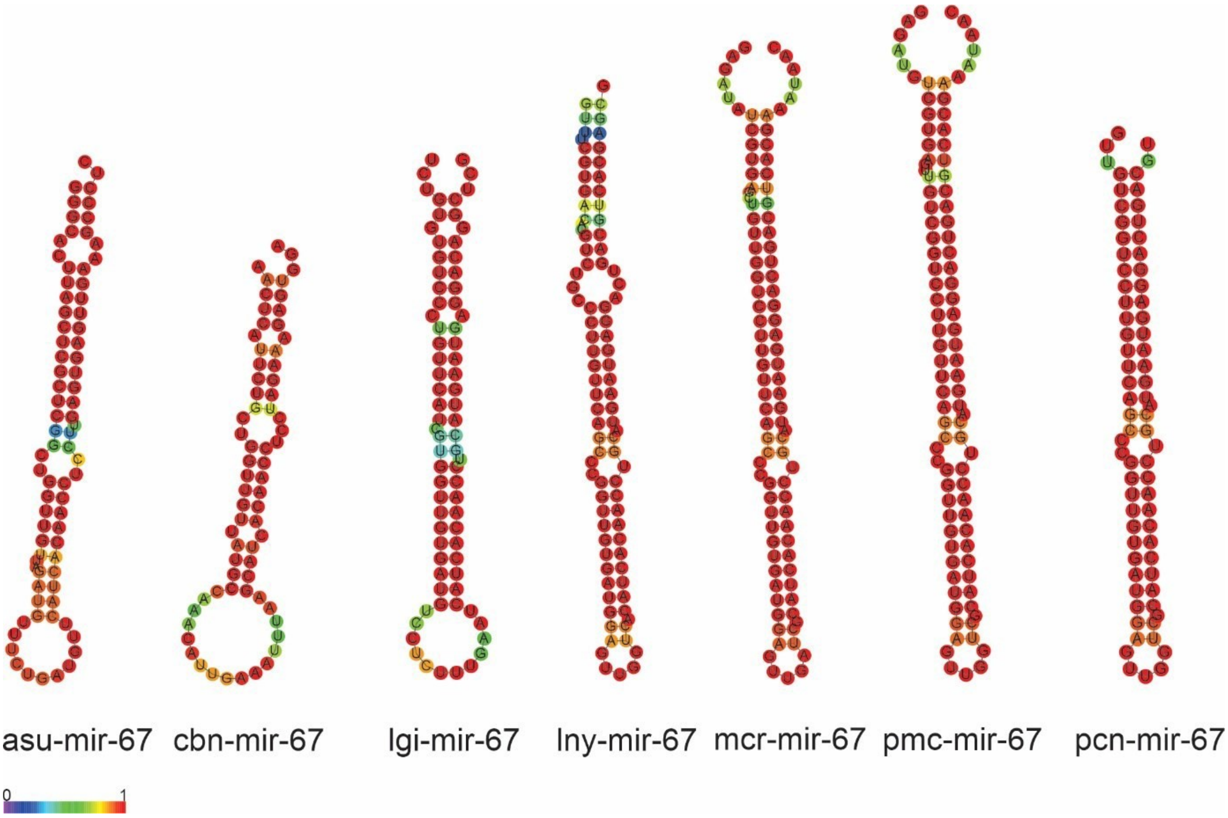


Figure 14: Secondary structure of mir-67 in *L. nyassanus, M. cornuarietis, P. maculata and their orthologs*; asu - *Ascaris suum*, cbn - *Caenorhabditis brenneri*, lny – *L. nyassanus*, lgi - *L. gigantea*, mcr-*M. cornuarietis*, pmc - *P. maculata*.

The Lophotrochozoa clade was subdivided into molluscs, annelids and flatworms. In the phylum Molluscs the presence of bivalves and gastropods was observed. For the first class of molluscs it was represented by the species: *Crassostrea virginica*, *Crassostrea hongkongensis*, *Pinctada martensii*, *Pinctada Fucata*, *Patinopecten yessoensis* and *Argopecten purpuratus*. The gastropod animals were represented exclusively by the analyzed Ampullariidae species: *L. nyassanus*, *M. cornuarietis*. *P. canaliculata* and *P. maculata*. This distribution is consistent with the tree of life, where a wide distribution was found in the phylogenetic clades (Figure 15).

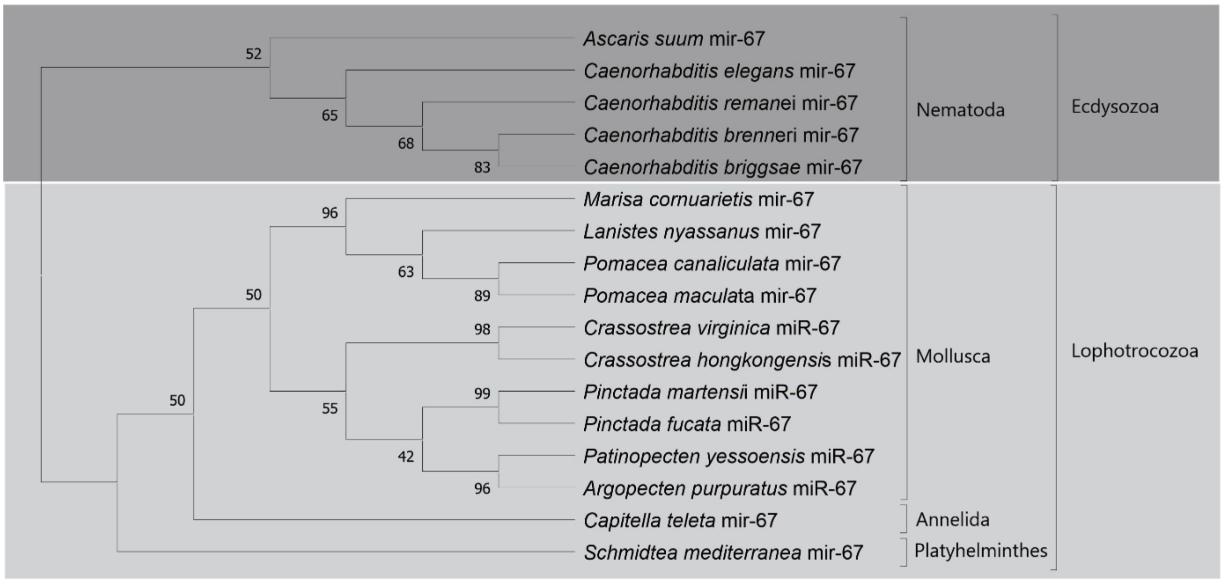


Figure 15: Phylogenetic distribution of mir-67 in *L. nyassanus, M. cornuarietis. P. canaliculata* e *P. maculata* and their orthologs.

Alignment of mir-96b precursors from Ampullariidae species and their orthologs demonstrated the presence of two mature miRNAs (5p and 3p strands) in *L. nyassanus*, *M. cornuarietis*. *P. canaliculata* and *P. maculata*. The 3p strand of mature miRNAs from the analyzed Ampullariidae species showed 100% identity to each other and high similarity with orthologs. The sequence of the precursors of miR-96b, specific Lophochotrozoa, demonstrated high similarity in secondary structure in the gastropod organisms *L.nyassanus*, *M. cornuarietis* and *B.glabrata* (Figure 16 and Figure 17).

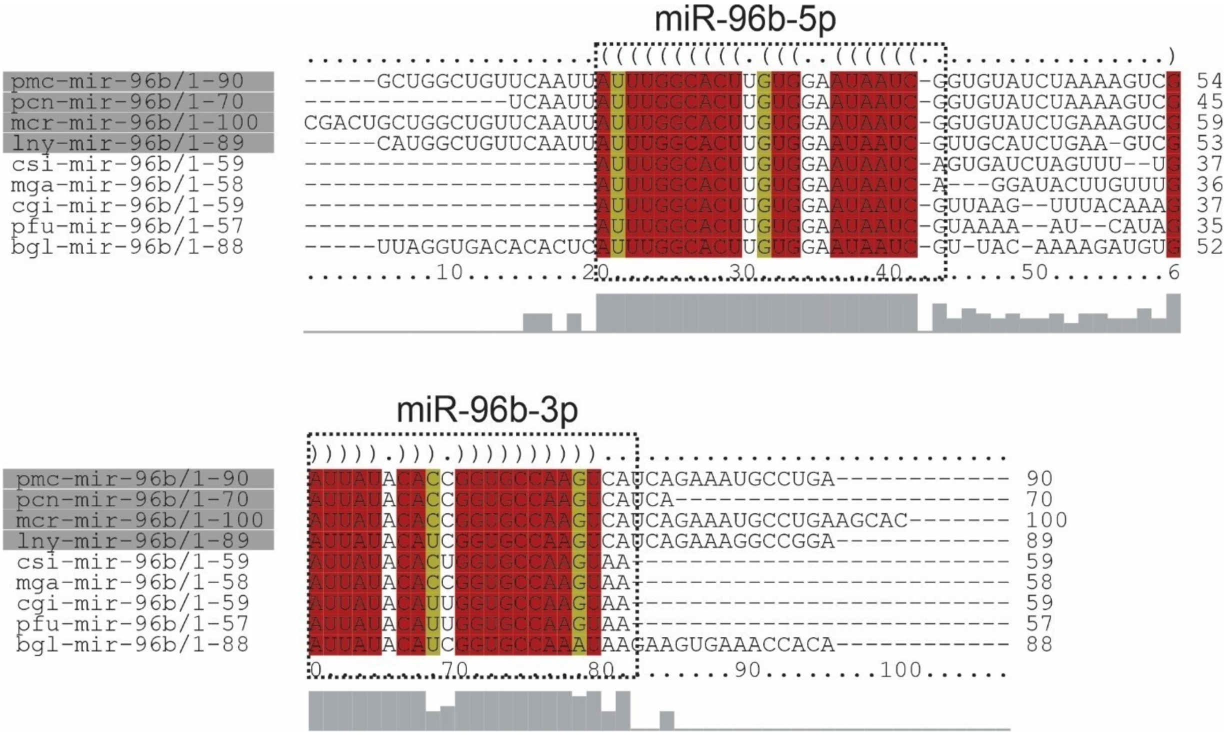


Figure 16: Alignment of miRNA-96b precursors of *L. nyassanus*. *M. cornuarietis, P. canaliculata* e *P. maculata* and their orthologs. bgl: *B. glabrata*, csi: *Cyclina sinensis*, lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pfu: *Pinctada fucata*

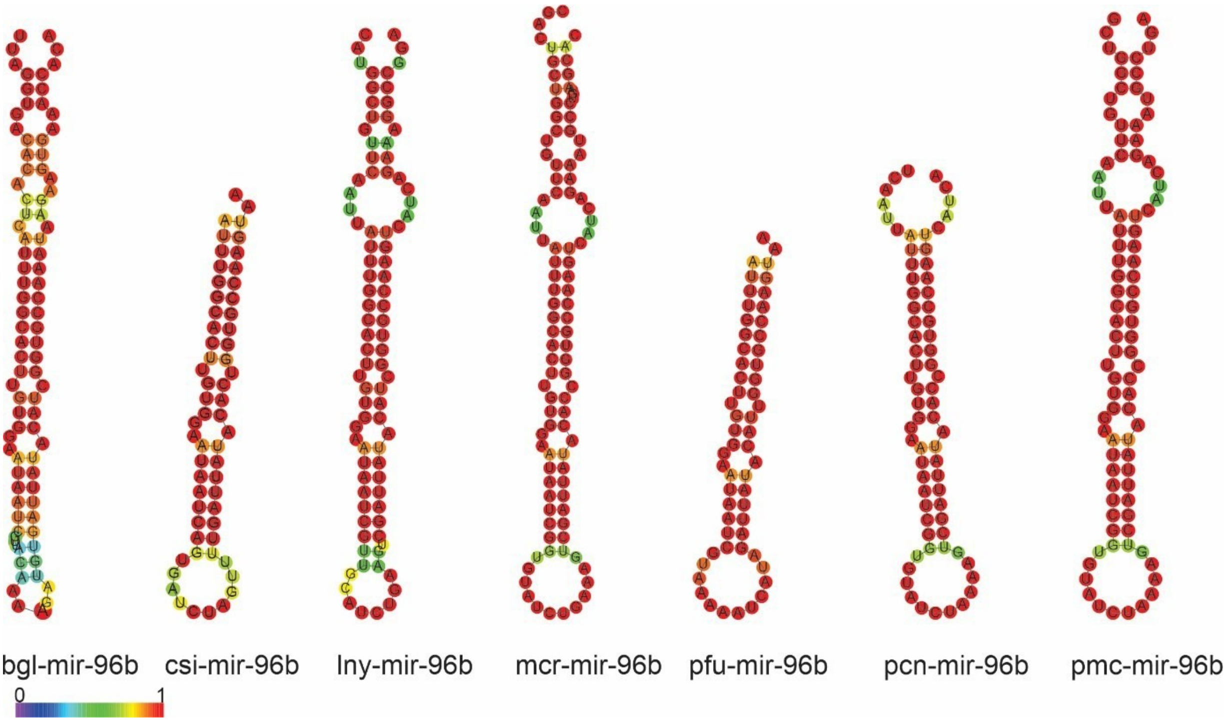


Figure 17: Secondary structure of ampullariids miRNA-96b precursors studied with their orthologs; bgl: *B. glabrata*, csi: *Cyclina sinensis*, lny*: Lanistes nyassanus,* mcr: *Marisa cornuarietis,* pfu: *Pinctada fucata*

The phylogenetic analysis of the precursors of mir-96b (Lophotrochozoa- specific) was verified the distribution among the species in three phyla (Molluscs, Anelideos and

Platelmiltos). A wide distribution was observed in the phylum Molluscs, where the presence of cephalopods, bivalves and gastropods was verified. The first class of molluscs was represented by *Architeuthis dux*. The bivalve class was represented by *Crassostrea gigas*, *C. virginica*, *C. hongkongensis*, *P. Fucata* and *P. martensii*. The presence of snails of the Ampullariidae family analyzed once again appeared grouped together, as expected. In addition, ampullariids appear segregated in the gastropod class with molluscs of the genus *Haliotis* and *Radix auricularia* species. A wide distribution was observed in the Lophotrochozoa clade, being constructed a phylogenetic tree consistent with the tree of life. (Figure 18).

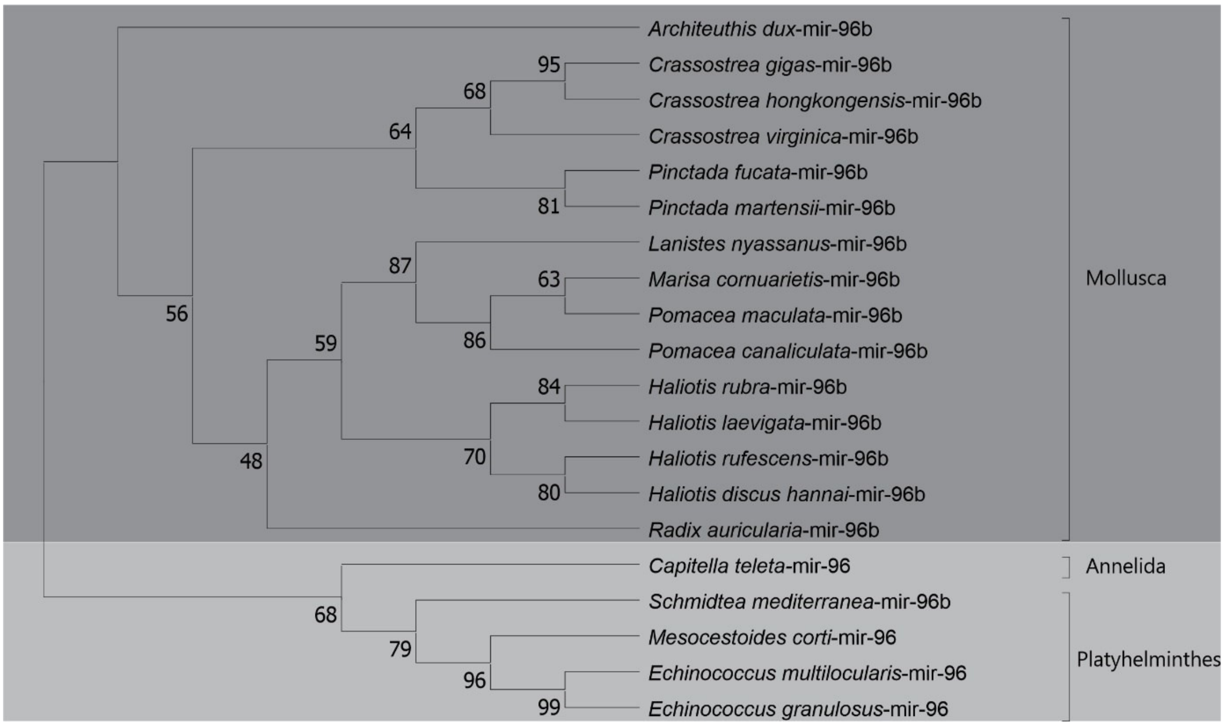
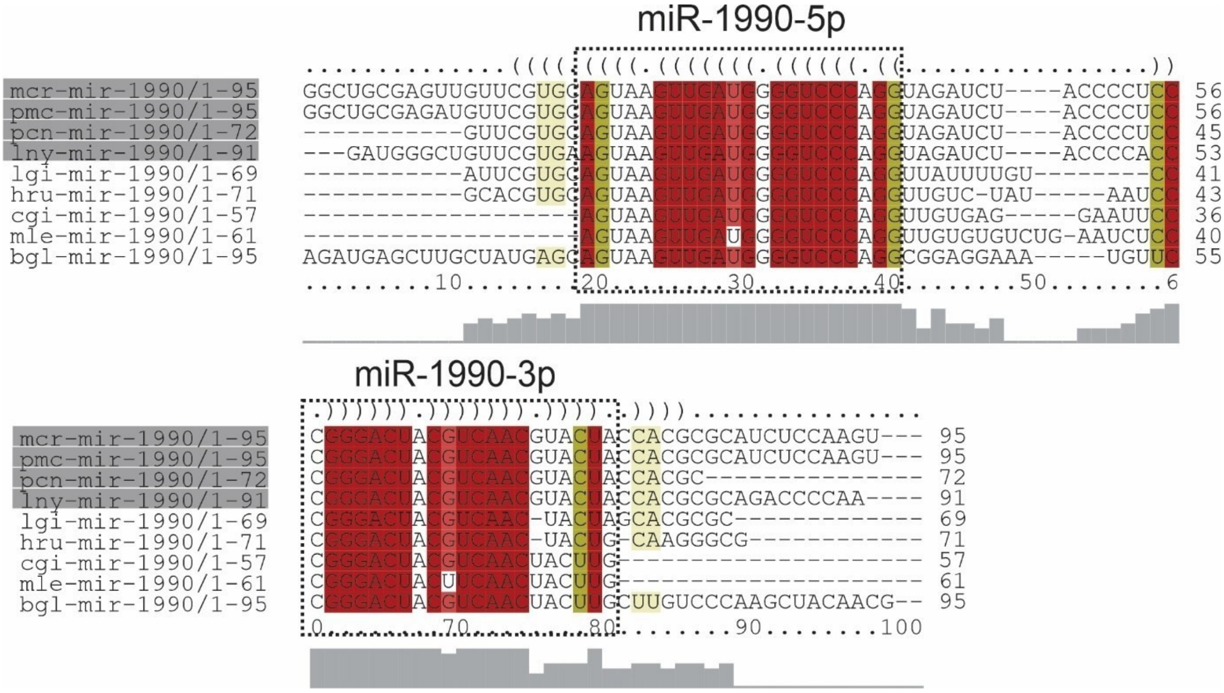


Figure 18: Phylogenetic analysis of miR-96b (Lophotrochozoa-specific) for *L. nyassanus. M. cornuarietis, P. canaliculata* e *P. maculata* and their orthologs.

The mir-1990 family predicted for species *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* is composed of the precursors lny-mir-1990, mcr-mir-1990, pcn-mir-1990 pcm-mir-1990 and the mature miRNAs, 3p and 5p, in each species ampullariids. For this family, the conservation and great similarity of the primary structure was observed, as well as secondary structures demonstrate conservation among orthologs (Figure 19 and Figure 20).



miR-1990-5p

```
                  ..............((((((((.((((((((.((((((.((.................))
mcr-mir-1990/1-95 GGCUGCGAGUUGUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCU----ACCCCUCC 56
pmc-mir-1990/1-95 GGCUGCGAGAUGUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCU----ACCCCUCC 56
pcn-mir-1990/1-72 -----------GUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCU----ACCCCUCC 45
lny-mir-1990/1-91 ---GAUGGGCUGUUCGUGAAGUAAGUUGAUGGGGUCCCAGGUAGAUCU----ACCCCACC 53
lgi-mir-1990/1-69 -----------AUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUUAUUUUGU--------CC 41
hru-mir-1990/1-71 -----------GCACGUGCAGUAAGUUGAUGGGGUCCCAGGUUGUC-UAU-----AAUCC 43
cgi-mir-1990/1-57 -------------------AGUAAGUUGAUGGGGUCCCAGGUUGUGAG-----GAAUUCC 36
mle-mir-1990/1-61 -------------------AGUAAGUUGAUUGGGUCCCAGGUUGUGUGUCUG-AAUCUCC 40
bgl-mir-1990/1-95 AGAUGAGCUUGCUAUGAGCAGUAAGUUGAUGGGGUCCCAGGCGGAGGAAA-----UGUUC 55
                  .........10........20........30........40........50........6
```

miR-1990-3p

```
                  .)))))).)))))))).))))).:.))))...................
mcr-mir-1990/1-95 CGGGACUACGUCAACGUACUACCACGCGCAUCUCCAAGU--- 95
pmc-mir-1990/1-95 CGGGACUACGUCAACGUACUACCACGCGCAUCUCCAAGU--- 95
pcn-mir-1990/1-72 CGGGACUACGUCAACGUACUACCACGC--------------- 72
lny-mir-1990/1-91 CGGGACUACGUCAACGUACUACCACGCGCAGACCCCAA---- 91
lgi-mir-1990/1-69 CGGGACUACGUCAAC-UACUAGCACGCGC------------- 69
hru-mir-1990/1-71 CGGGACUACGUCAAC-UACUG-CAAGGGCG------------ 71
cgi-mir-1990/1-57 CGGGACUACGUCAACUACUUG--------------------- 57
mle-mir-1990/1-61 CGGGACUACUUCAACUACUUG--------------------- 61
bgl-mir-1990/1-95 CGGGACUACGUCAACUACUUGCUUGUCCCAAGCUACAACG-- 95
                  0........70........80........90........100
```

Figure 19: mir-1990 alignment for *L. nyassanus, M. cornuarietis. P. canaliculata* e *P. maculata* and their orthologs; pcn - *P. canaliculata*, lny - *L. nyassanus*, mcr - *M. cornuarietis*, pmc - *P. maculata*, lgi- *L. gigantea*, mle- *M. leonina*, cgi - *C. gigas*, bgl – *B. glabrata*, hru- *H. rufescens*

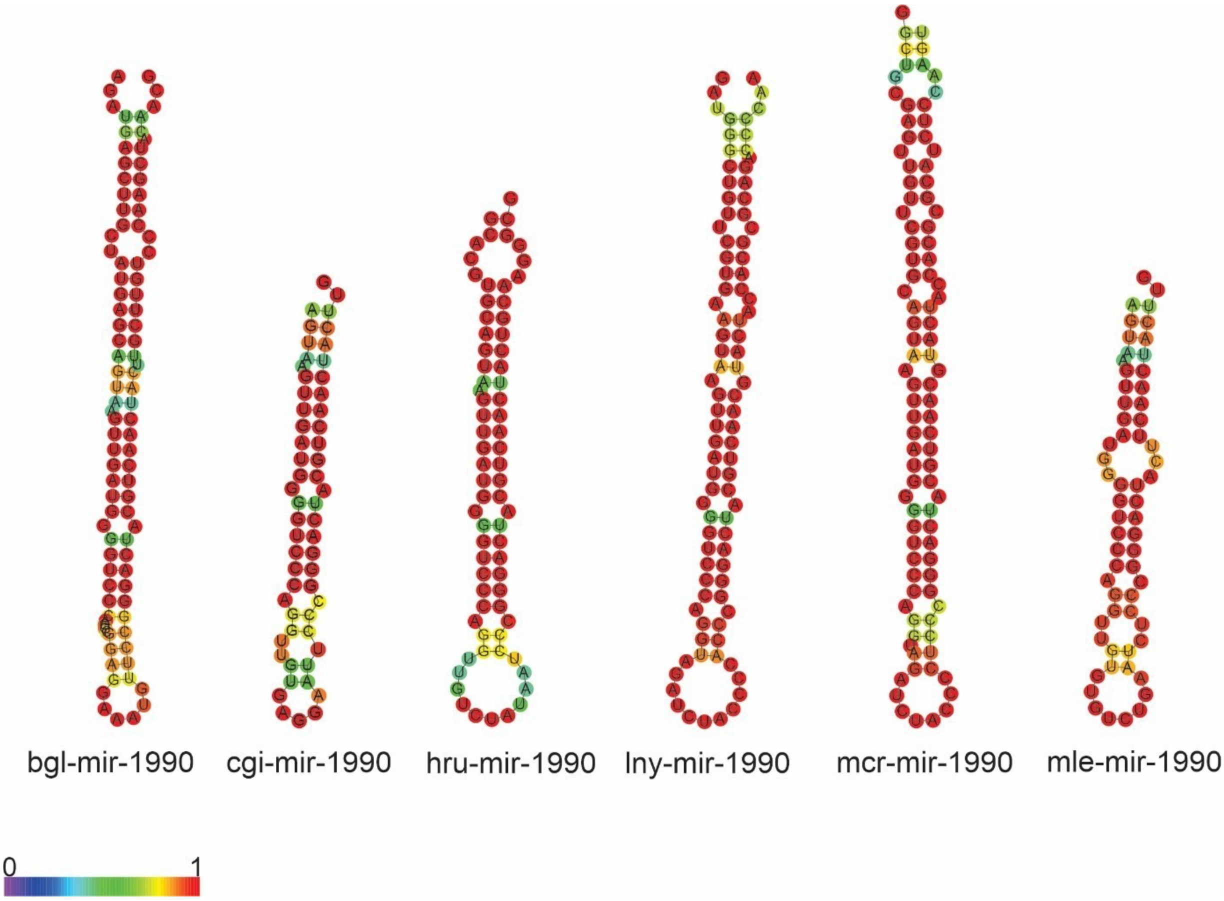


Figure 20: Secondary structure of the mir-1990 in *L. nyassanus, M. cornuarietis. P. canaliculata* e *P. maculata* and their orthologs

DISCUSSION OF RESULTS

In this study, three putative Argonaute proteins were found in *L. nyassanus* (Lny23421_c0_g1, Lny25187_c0_g1 and Lny28936_c7_g7). The same number encountered in *M.* cornuarietis (Mco25093_c0_g2, Mco26370_c0_g1, Mco27042_c1_g1). For each specie of the genus pomacea, two probable piwi protein were found, in orders *P. canaliculata* (Pca69814_c1_g2, Pca61914_c0_g1) and *P. maculata* (Pma63715_c0_g1_, Pma60501_c2_g16). The Argonaute protein is characterized by the presence of the PIWI and PAZ domains (PIWI-Argonaute-Zwille) (Song et al., 2004). The phylogenetic tree presented these two clades in a distinct way (figure 14). The argonaute proteins of eukaryotes can present in addition to these the following domains, N (N-terminal), and MID, together with two domain ligands L1 and L2 (Swarts et al., 2014). The PIWI domain in Argonaute contains three conserved catalytic residues composed of two aspartates and a histidine, called 'DDH' (Yang and Steitz, 1995). It was confirmed the presence of these conserved domains in Argonaute putative proteins identified. The analysis of the amino acid sequence of the PIWI domains confirmed the maintenance of the DDH amino acids (aspartic acid, aspartic acid and histidine) in the proper positions for the activity performed. The PIWI domains have great functional homology with RNase H, which is known for enabling the fragmentation of RNA sequences (Parker et al., 2004). The size of the predicted proteins was compatible with the average size of evolutionarily close organisms.

In this study, Lny28171_c0_g1 and Pca67531_c1_g2 DICER proteins were found in *L. nyassanus* and *P. canaliculata* respectively. Lny18581_c0_g2, Mco22837_c2_g1 and Pca67887_c3_g1 DROSHA proteins were found in *L. nyassanus*, *M. cornuarietis* and *P. canaliculata* respectively. The DROSHA and DICER proteins belong to the family of endoribonucleases, ribonuclease III (RNase III) that have specificity for cleavage of double-stranded RNA (dsRNA). The DROSHA protein is a class II RNAase III containing two endonuclease domains and a dsRBD. Dicer is a class III RNAase III that has two endonuclease domains, a dsRBD, an N-terminal helicase domain and a PAZ domain. Functionally class III enzymes produce single-stranded RNA (ssRNA) products of approximately 22 nucleotides in length from long dsRNA substrates (Bernstein et al., 2001; Blaszczyk et al., 2001; Filippov et al., 2000). The canonical pathway of miRNA biogenesis is the dominant pathway in which miRNAs are processed by class II (Drosha) and class III (Dicer) RNAases consecutively. These RNases III contain two catalytic centers with amino acids important for function. The Riboc I domain is composed of the amino acids; E (Glutamic acid), E (Glutamic acid), D (Aspartic acid) D (Aspartic acid); and the Riboc II domain is formed by E (Glutamic acid), Q (Glutamine), E (Glutamic acid), D (Aspartic acid), D (Aspartic acid), E (Glutamic acid); responsible for the cleavage of diester bonds (Blaszczyk et al., 2001).

The DROSHA protein predicted from *P. canaliculata* (Pca67887_c3_g1) showed compatible length and similarity with its orthologs (Table 1). The analysis of conserved domains showed the two endonuclease domains and one dsrm, characteristic of class II RNAases III. In the search for catalytic sites, he presented the first set composed of amino acids E-E-D-D in the Riboc I domain (figure 16). The 1525 amino acid length was compatible with the 1523 amino acid *P. canaliculata* protein (ID: XP_025111754.1) registered at the NCBI (National Center for Biotechnology Information — http://.ncbi.nlm.nih.gov/). These data corroborated the identification of the putative Drosha protein in *P. canaliculata*. On the other hand, in the DROSHA protein of *M. cornuarietis* (Mco22837 c2 g1), despite having a relatively smaller size than its orthologs (figure 15), the lack of coverage in the prediction of the protein did not affect the prediction of conserved domains, since it occurred, probably in the N-terminal portion of the protein, which, for DROSHA, functional conserved domains have not yet been identified. This predicted Drosha protein had two RIBOc domains and a double-stranded RNA binding domain and the presence of complete catalytic amino acid sequence in the Riboc I domain. These data support the identification for the putative Drosha protein in *M. cornuarietis* (Mco22837 c2 g1). Regarding another DROSHA candidate, *L. nyassanus* (Lny18581_c0_g2) presented a Riboc domain, a fragment of the second Riboc domain and the presence of a complete catalytic amino acid sequence in the Riboc I domain. The regions in the genome predicted for this gene are probably in error of annotation leading to the truncation of the protein sequences, consequently the size of the amino acid sequences for these genes were underestimated in size and structure. Further versions of the genome may solve this problem.

The putative DICER protein of *P. canaliculata* (Pca67531_c1_g2) was shorter in length (1984 aa) than the evolutionarily close organisms *A. californica* (2503 aa) and *B. glabrata* (2332 aa) (Table 2). Only conserved domains were identified; Ribonucleas_3_3, PAZ, Dicer dimer and Helicase_C; not showing the second Riboc domain. When verifying the starting position of the second Riboc domain of closely related organisms, *A. californica* (2273 aa) and *B. glabrata* (2100 aa), corroborated the fact that the second ribonuclease was not identified in the probable putative sequence. In Dicer, probably due to the lack of coverage in the sequencing of this region of the genome of *P. maculata*, it was not putative to evidence the presence of the second catalytic domain Riboc. The *P. canaliculata* Dicer protein (XP_025104500) identified at the NCBI with a length 2249 amino acids was orthologous to Pca67531_c1_g2, with a query coverage of 99% and a percentage of identification of 98.83%. The protein sequence Pca67531_c1_g2 using the BLASTp tool against the genome of *P. canaliculata* in the AmbuBase database (Department of Computer Science HKBU - https://www.comp.hkbu.edu.hk/~db/AmpuBase/#&panel1- 4) presented a percentage of identification and coverage of 100% (1984/1984 amino acids). Part of the first catalytic site of amino acids (ED) was observed in Riboc in the candidate protein cited (Pca67531_c1_g2) When performing the search for catalytic residues having as query NP_524453.1 Dicer-1 *D. melanogaster* the orthologous molluscs presented only the amino acids in the first domain Riboc catalysts (ED), with the exception of *Pectem maximus* (EDDE) NCBI (National Center for Biotechnology Information — http://.ncbi.nlm.nih.gov/). The data presented corroborated the identification of the putative protein. On the other hand, the putative DICER protein from *L. nyassanus* (Lny28171_c0_g1), despite having a relatively smaller size than its orthologs (figure 15), the lack of coverage in the protein prediction did not affect the prediction of the two conserved Riboc domains and a PAZ domain, since it was probably in the N-terminal portion of the protein, which for DICER the catalytic domains lie at the C-terminal end. This predicted DICER protein showed the presence of the complete catalytic amino acid sequence in the Riboc I domain. The data support the identification of the putative DICER protein in *L. nyassanus*. The regions in the genome predicted for this gene are likely to have annotation errorleading to the truncation of the protein sequences. Consequently, the size of the amino acid sequences for these genes are underestimated in size and structure (Queiroz et al., 2017).

In this study, Lny14170_c0_g1, Lny12572_c0_g1, Lny27692_c0_g2, Mco24030_c0_g2, Mco81614_c0_g2, Pca62316_c0_g2, Pca65193_c2_g1, Pma59467_c0_g1, and Pma63331_c0_g1 Exportin protein family (XPO-5, XPO1 and XPO-T) were found *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* respectively. Intrinsically, the transport of miRNA precursors is performed by the XPO-5 protein. Occasionally the transport of pre-miRNAs can be accomplished by XPO-1 entering the non-canonical miRNA processing pathway without Drosha cleavage. Displacement of pre-miRNAs is also performed by exportin T (XPOT) in a Ran-GTP-dependent manner (Ruby et al., 2007). The search for Exportins proteins (XPO-5, XPO-1 and XPO-T) using the methodology of this work, resulted in the putative proteins found.

In order to emphasize the conservation of these putative proteins identified in the ampullariids species, a phylogenetic tree and studies of conserved domains were constructed. The phylogenetic analysis revealed a distribution of the studied amino acid sequences, in the genes of the Exportin protein family (XPO-5, XPO1 and XPO-T), grouped close to the Deuterostomes clade. Putative proteins from *L. nyassanus*. *M. cornuarietis*, *P. canaliculata* and *P. maculata* aggregated with their respective orthologs close to the phylum Molluscs and the clade Deuterostomes. The similarity of the identified Exportins of the Ampullariidae species was verified together with their orthologs, observing the level of preservation of the conserved domains and protein length. For Exportin 5 the putative proteins (Lny14170_c0_g1, Pca62316_c0_g2) presented the two conserved domains (Xpo1 and IBN_N) characteristic of the XPO-5 protein and a size compatible with the average dimension of its orthologs, confirming the identification of the putative Exportin 5 proteins. Exportin 1 putative proteins (Lny12572_c0_g1, Mco24030_c0_g2, Pca65193_c2_g1, Pma59467_c0_g1) showed six conserved domains characteristic of XPO-1 (IBN_N, Xpo1, CRM1_repeat, CRM1_repeat_2, CRM1_repeat_3 and only five domains except for the CRM1_cc1. The Pca65193_c2 did not show the conserved IBN_N domain. The lack of protein sequence prediction in the N-terminal region was already reported in this work for *P. canaliculata*, resulting in a relatively smaller size when compared to the analyzed species. Even the length of the protein sequences analyzed for XPO-1 (Lny12572_c0_g1, Mco24030_c0_g2, Pca65193_c2_g1, Pma59467_c0_g1) were strictly the same (1070 aa). This dimension also matched to the average size of the Exportin 1 protein for orthologous organisms. All these factors corroborated the identification of putative XPO-1 proteins in the analyzed species. Likewise, Exportin T in genes (Lny27692_c0_g2, Mco81614_c0_g2 and Pma63331_c0_g1) showed a characteristic conserved domain for all protein sequences (Xpo1). This time, lack of protein sequence prediction was verified in the N-terminal region for M. cornuarietis (Mco81614_c0_g2), presenting a smaller size (331 aa) than the analyzed species. Interestingly, the protein sequences (Lny27692_c0_g2 and Pma63331_c0_g1) also had the same size (966 aa) and matched the average size of XOP-T proteins from orthologous organisms. They were convincing evidence for the identification of putative Exportin T in the analyzed species.

The genomes of four ampullariids (Old World - Lanistes nyassanus and New World - Pomacea canaliculata, P. maculata, and Marisa cornuarietis) were published by Sun et al., 2019, in order to understand the genomic basis underlying the diversity of the Ampullariidae species and also their behavioral, morphological, and physiological adaptations. In our study we were able to apply a robust analysis to identify and characterize miRNAs and the putative proteins involved in miRNA pathway using the genome and predicted proteome from four Ampullariidae species. Either miRNAs or proteins of their biosynthetic pathway were described in several organisms, including several kingdoms such as plants and animals. In model species such as *D. melanogaster* and *C. elegans* mature and precursor miRNAs and also their pathway proteins have been extensively studied. Even though Mollusca is a phylum containing many species, little has been studied about miRNAs and their processing pathway in this phylum.

Sun et al., 2019, sequenced and analyzed the four genomes of ampullariids species, P. canaliculata, P. maculata, and L. nyassanus. The genomes were assembled in 535.5 Mb million base pairs (Mb) in total length or smaller and produced thousands of scaffolds anchored or not to chromosomes. Automatic annotation analyses of the four genomes displayed around 20 thousand gene models (Sun et al., 2019). So far, no study on miRNAs and their processing pathway in ampullariids species has been reported in the literature. In order to improve and increase the annotation of non-protein coding genes, such as miRNAs, and also of protein-coding genes of small RNA processing pathways, this work proposed an in-depth study of mature and precursory miRNAs and their pathway processing in the genome of the four species of ampullariids.

In our study, we were able to identify conserved miRNAs in four species, being 162 mature miRNAs and 141 miRNA precursors in *L. nyassanus*, 297 mature miRNAs and 279 miRNA precursors in *M. cornuarietis*, 296 mature miRNAs and 269 miRNA precursors in *P. canaliculata* and 316 mature miRNAs and 299 miRNA precursors in *P. maculata*. Two miRNAs found in all species was Mollusca-specific.

The minimum free energy value, -18 kcal/mol, in general, is the value necessary for a miRNA precursor to be stable and generates miRNAs (Gomes et al., 2011; Hofacker, 2009). The sequences of pre-miRNAs presented in this work MFE mean of (minimum free energy) and variation of values (Table 3). The mean value of the studied precursors was similar to the mean value of pre-miRNAs of lophotrochozoan species (-31.27 kcal/mol) (Gomes et al., 2011). Of the pre-miRNAs deposited in miRBase approximately 75% of those had minimum free energy values below -18.50 kcal/mol (Leclercq et al., 2013).

The size of the precursors of the lophotrochozoan species is between 57 and 153 nucleotides, with an average of 90 nucleotides per sequence(Gomes et al., 2011). In this study, the average size of the precursors and the length variation were showed in Table 3. The size of pre-miRNAs in animal species ranges from 47 to 177 nucleotides with a mean of 87 ± 29 nucleotides (Zhou et al., 2009). The data from this study corroborated with the average size of pre-miRNAs and the reference range for the size of lophotrochozoan species and metazoans in general.

GC content is one of the main parameters for the identification of pre-miRNAs and is very important for the stability of the secondary structure of precursors. The percentage of pre-miRNAs present in miRBase that had GC content above 20% and below 65% was 80% (Leclercq et al., 2013). In this work, the variation of GC content and the percentage of miRNAs in the range between 20% and 65% were respectively; *L. nyassanus* (20.25% to 69.05%) and 98.5%, *M. cornuarietis* (20.20% to 67.03%) and 98%, *P. canaliculata* (20.83% and 68.29 %) and 99.25%, *P. maculata* (20.40% and 68.75%) and 98.66%. The precursors presented mean values for the guanine-cytosine (GC) content for *L. nyassanus*, *M. cornuarietis, P. canaliculata* and *P. maculata*; 41.26%, 41.64%, 41.70%, 40.74% respectively. The mean value of the GC content of the studied ampullariids precursors was similar to the mean of the lophotrochozoan species (40.49%) (Gomes et al., 2011). Comparatively the GC content of the miRNA precursors were not observed statistical differences ($p > 0.05$) between the Ampullariidae analyzed and lophotrochozoan species.

Mature miRNAs were identified in the genome of 4 ampullariids species: 162 miRNAs for *L. nyassanus*, 297 miRNAs for *M. cornuarietis*, 296 miRNAs for *P. canaliculata* and 316 miRNAs for *P. maculata*. The number of miRNAs identified in *L. nyassanus* genome was similar to the gastropod species *Conus tribblei* (159 miRNAs) (Huang et al., 2021). The number of miRNAs found in *M. cornuarietis* (297 miRNAs), *P. canaliculata* (296 miRNAs), *P. maculata* (316 miRNAs) corroborated with the number of miRNAs identified for *Lymnaea stagnalis* (freshwater gastropod molluscs), 264 conserved mature miRNAs (Walker et al., 2018). There are plausible justifications for the smaller quantitative number of miRNAs for *L. nyassanus* compared to other ampullariids under study. *L. nyassanus* has fewer adaptive and functional properties when compared to other ampullariids under study (Sun et al., 2019). *L. nyassanus* is endemic to Lake Nyasa in Africa (Cowie, 2015; Dohrn, 1865; Van Bocxlaer, 2017), while *P. canaliculata* and *P. maculata* are geographically distributed on almost every continent, fast growing, high reproduction rate, tolerance and adaptation to environmental stress (Liu et al., 2018; Pimentel et al., 2005). Species of the genus *Pomacea* have the adaptive property of terrestrial egg laying, while *L. nyassanus* does egg laying in the aquatic environment (Sun et al., 2019). In addition, *M. cornuarietis* is better able to resist factors such as low temperature and hypoxia (Matsukura et al., 2016; Mu et al., 2018). As reported in previous studies, there is a significant increase in the number of miRNAs as the morphological complexity of the species increases (Heimberg et al., 2008), thus, justifying a lower number of miRNAs in *L. nyassanus*, when compared to other ampullariids in study.

The data presented corroborated the identification of miRNAs with the literature (Ha and Kim, 2014; Rachagani et al., 2010). The number of nucleotides in the mature miRNAs from ampullariids species had an arithmetic mean of 21.94 nucleotides/miRNA, with a size range from 17 to 25 nucleotides.

The Argonaute protein prefers the uracil nucleotide in the first position of the miRNA sequence, to be inserted in the RISC complex (Seitz et al., 2011). In this work mature miRNAs from *L. nyassanus, M. cornuarietis*. *P. canaliculata* and *P. maculata* presented, respectively, quantitative uracil nucleotides in the first position; 41.97%, 44.44%, 41.2% and 42.40%. Mostly presented this nucleotide in the first position in the miRNA sequence.

The structural and thermodynamic characteristics of pre-miRNAs were more similar among *M. cornuarietis*, *P. canaliculata* and *P. maculata* species. *L. nyassanus* had more dissimilar values, however it is the organism that most diverges with reference to the three species mentioned. The justifications for this probably in phylogeny. The division between the New World (*M. cornuarietis*, *P. canaliculata* and *P. maculata*) and Old World (*L. nyassanus*) lineages may have occurred at the time of the separation from Gondwana, around 120 million years ago (Jokat et al., 2003; Sun et al., 2019).

miR-33, Bilateria-specific, was found in the genomes of *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* and had two mature miRNAs (3p and 5p chains), corroborating studies observed in bivalve molluscs and *D. melanogaster* (Bao et al., 2014; Clerbaux et al., 2021). Mir-33-5p supposedly regulates genes involved in the heavy metal-induced stress response in the mollusc *Tegillarca granosa* (Bao et al., 2014). Likewise, *P. canaliculata* is able to survive in environments with heavy metals, where there is a high concentration of these metals in the gills (Kruatrachue et al., 2011), probably regulated by this micro-RNA. In this study, miR-33-5p showed 100% similarity between the ampullariid and orthologous species (figure 1). Furthermore, miR -33 is a pleiotropic regulator of metabolic and developmental processes in *D. melanogaster*. It is suggested that mir-33 is a conserved regulator of lipid homeostasis (Clerbaux et al., 2021).

The family of precursors of mir-67, protostomes specific, was found in the genome of the 4 species Ampullariidae with the presence of mature miR-67-3p for each species, corroborating the wide distribution of this miRNA in molluscs and *C. elegans* (Huang et al., 2021; Ma et al., 2017). Like miRNA-33, miRNA-67 is potentially associated with gene regulation whenexposed to toxic levels of cadmium (Cd) in the bivalve *T. granosa* (Bao et al., 2014). Regulationof the immune system is also performed by mi-67 in *C. elegans* when exposed to pathogenic bacteria *Pseudomonas aeruginosa*. miRNA-67 inhibits the expression of the SAX-7 protein inthe worm, that induces pathogen avoidance behavior (Ma et al., 2017). Furthermore, mir-67 was identified in 34 species of molluscs (Huang et al., 2021).

The sequences of the precursors of miR-96b presented two mature miRNAs, 3p and 5p, in the genomes of the species: *L. nyassanus*, *M. cornuarietis*. *P. canaliculata* and *P. maculata*. The mir-96b, Lophotrochozoa-specific, regulates genes post-transcriptionally in flatworms and molluscs. In the freshwater worm: *Schmidtea mediterranea* may have functions in wound healing, neoblast proliferation (mainly responsible for regeneration in planarians) and blastema differentiation (Sasidharan et al., 2013). In molluscs, miR-96b is involved together with other miRNAs in the pigmentation of the shells of *C. gigas*, inducing the synthesis of melanin, carotenoid or tetrapyrrole (Feng et al., 2020). The miR-96b was also found in the genome of gastropods evolutionarily close to the 4 species Ampullariidae, *B. glabrata* (Queiroz et al., 2020) and *A. californica* (Huang et al., 2021). Findings in the scientific literature corroborate the data of this work.

The family of mir-1990, Lophotrochozoa-specific miRNA, presented two mature miRNAs, 3p and 5p. A wide distribution of mir-1990 in molluscs was verified, mainly in 15 bivalve animals, 10 gastropods and 1 cephalopod (Huang et al., 2021). Few studies describe the role of miRNAs in molluscs development. However, mir-1990-3p may play a vital role in the shell biomineralization process in *P. fucata*, as it is highly expressed in mantle tissues (Huang et al., 2021). mir-1990 was found only in molluscs, however the mir-1990 family is Lophotrochozoa-specific as cte-miR-1990c-3p, cte-miR-1990a and cte-miR-1990b have been identified in *Capitella teleta* (http: //www.mirbase.org/). The wide distribution of mir-1990 in molluscs and the likely mechanism of gene control in the species corroborated the data in this study.

The functional form of a miRNA is generally 22 nucleotides in length, with a size range from 17 to 25 nucleotides (Ha and Kim, 2014; Rachagani et al., 2010). These small RNAs are generated by two cleavage reactions in which miRNAs are processed by RNAases III Drosha and Dicer consecutively (Lee et al., 2002; Tanzer and Stadler, 2004; Y. F. Zhang et al., 2009), in canonical miRNA processing pathway. The microprocessor complex, composed of a protein (DGCR8) and a ribonuclease III enzyme (Drosha), performs the cleavage of pri-miRNA in pre-miRNA in the cell nucleus. DGCR8 binds to a structure in pri-miRNA while Drosha performs the duplex scission. After the pre-miRNAs are generated, they are transported to the cytoplasm through the exportin 5 (XPO5) / RanGTP complex (Alarcón et al., 2015; Denli et al., 2004; Han et al., 2004; Okada et al., 2009). In the cytoplasm, pre-miRNAs are cleaved by Dicer into mature. The miRNAs can effect their action through the miRISC complex, in which a protein from the AGO family is loaded with a miRNA guide strand that directs the complex to base pairing, which by different mechanisms induces the silencing of gene expression (Ipsaro and

Joshua-Tor, 2015; Jo et al., 2015). The AGO-2 protein has endonucleolytic action and the level of complementarity formed between the miRISC complex and the target mRNA is indicative of the complex's action (Jo et al., 2015). All these factors in our analysis were of great relevance.

CONCLUSION

The search for miRNAs in *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* was able to predict 162 mature forms of miRNAs for *L. nyassanus*, 297 miRNAs for *M. cornuarietis*, 296 miRNAs for *P. canaliculata* and 316 miRNAs for *P. maculata*.

It was possible to identify the biogenesis pathway of miRNAs in species of the Ampullariidae family, highlighting several putative key pathway proteins; Argonaute, DROSHA, DICER, XPOs, among others. The prediction of some proteins of the miRNA pathway was not verified in some species, probably due to annotation error in the part of the sequenced gene specific to each organism.

Our results expand the study of miRNAs in molluscs, bringing new challenges to the understanding of the essential processes related to the invasiveness of *P. canaliculata* and *P. maculata* species and supposed advances in the control of these molluscs considered agricultural pests for humanity.

Thus, the search for miRNAs and their processing pathways in the species *L. nyassanus*, *M. cornuarietis*, *P. canaliculata* and *P. maculata* were able to predict structures that found the established criteria, also complementing previous results from our study group as well, as creating the need to establish putative new target genes for the discovered miRNAs.

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SUPPLEMENTARY DATA I

Supplementary Table 1 Id Protein, putative name, length, ID protein Ortholog, Ortholog species, Ortholog length and E-value of *L. nyassanus* proteins

ID protein	Putative name	Length (AA)	ID protein Ortholog	Ortholog Species	Length (AA)	E-value
Lny23421	Argonaute 1.1	890	NP_493837.1	*Caenorhabditis elegans*	891	0.0
			NP_001163498.1	*Drosophila melanogaster*	866	$7e^{-35}$
			XP_013068463.1	*Biomphalaria glabrata*	895	0.0
Lny25187	Argonaute 1.2	880	NP_493837.1	*Caenorhabditis elegans*	891	$9e^{-44}$
			NP_001163498.1	*Drosophila melanogaster*	866	$8e^{-180}$
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Lny28936	Argonaute 1.3	941	NP_493837.1	*Caenorhabditis elegans*	891	$3e^{-43}$
			NP_001163498.1	*Drosophila melanogaster*	866	0.0
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Lny28171	Dicer	1434	NP_498761.2	*Caenorhabditis elegans*	1910	$9e^{-124}$
			NP_524453.1	*Drosophila melanogaster*	2249	$2e^{-159}$
			XP_013067888.1	*Biomphalaria glabrata*	2332	$2e^{-159}$
Lny18581	Drosha	317	NP_492599.1	*Caenorhabditis elegans*	1086	$4e^{-87}$
			NP_477436.1	*Drosophila melanogaster*	1327	$1e^{-146}$
			XP_013069067.1	*Biomphalaria glabrata*	1469	$2e^{-171}$
Lny14710	Exportin 5	1196	NP_741567.1	*Caenorhabditis elegans*	1080	$1e^{-11}$
			NP_001285453.1	*Drosophila melanogaster*	1241	0.0
			XP_013087907.1	*Biomphalaria glabrata*	1193	0.0
Lny12572	Exportin 1	1070	NP_741567.1	*Caenorhabditis elegans*	1080	0.0
			NP_001303316.1	*Drosophila melanogaster*	1063	0.0
			XP_013069551.1	*Biomphalaria glabrata*	1996	0.0
Lny27692	Exportin T	966	NP_501320.2	*Caenorhabditis elegans*	950	$3e^{-101}$

			XP_013079973.1	*Biomphalaria glabrata*	963	0.0
Lny22624	Tudor - SN	897	NP_494839.1	*Caenorhabditis elegans*	914	0.0
			NP_001261195.1	*Drosophila melanogaster*	926	0.0
Lny29643	Loquacious	338	NP_723813.1	*Drosophila melanogaster*	419	$8e^{-43}$
Lny27624	PASHA	701	NP_001293461.1	*Caenorhabditis elegans*	751	$1e^{-28}$
			NP_651879.1	*Drosophila melanogaster*	642	$1e^{-137}$
Lny9931	VIG	312	NP_001367502.1	*Caenorhabditis elegans*	378	$3e^{-09}$
			NP_723898.1	*Drosophila melanogaster*	490	$1e^{-26}$

Supplementary Table 2 Id Protein, putative name, length, ID protein Ortholog, Ortholog Species, Ortholog Length and E-value of *M. cornuarietis* proteins

ID protein	Putative name	Length (AA)	ID protein Ortholog	Ortholog Species	Length (AA)	E-value
Mco26370	Argonaute 1.1	931	NP_493837.1	*Caenorhabditis elegans*	891	0.0
			NP_001163498.1	*Drosophila melanogaster*	866	$1e^{-32}$
			XP_013068463.1	*Biomphalaria glabrata*	895	0.0
Mco27042	Argonaute 1.2	878	NP_493837.1	*Caenorhabditis elegans*	891	$3e^{-44}$
			NP_001163498.1	*Drosophila melanogaster*	866	$3e^{-178}$
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Mco25093	Argonaute 1.3	938	NP_493837.1	*Caenorhabditis elegans*	891	$1e^{-41}$
			NP_001163498.1	*Drosophila melanogaster*	866	0.0
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Mco22837	Drosha	760	NP_492599.1	*Caenorhabditis elegans*	1086	$6e^{-157}$
			NP_477436.1	*Drosophila melanogaster*	1327	0.0
			XP_013069067.1	*Biomphalaria glabrata*	1469	0.0
Mco24030	Exportin 1	1070	NP_741567.1	*Caenorhabditis elegans*	1080	0.0
			NP_001285453.1	*Drosophila melanogaster*	1241	$3e^{-10}$
			XP_013069551.1	*Biomphalaria glabrata*	1076	0.0
Mco81614	Exportin T	331	NP_001294153.1	*Caenorhabditis elegans*	952	$4e^{-43}$
			NP_001294153.1	*Biomphalaria glabrata*	963	$2e^{-122}$
Mco51286	Tudor - SN	926	NP_494839.1	*Caenorhabditis elegans*	914	0.0
			NP_001261195.1	*Drosophila melanogaster*	926	0.0
Mco13761	Loquacious	316	NP_723813.1	*Drosophila melanogaster*	419	$1e^{-44}$
Mco21592	PASHA	709	NP_001293461.1	*Caenorhabditis elegans*	751	$2e^{-28}$
			NP_651879.1	*Drosophila melanogaster*	642	$8e^{-135}$
Mco12258	VIG	414	NP_001367502.1	*Caenorhabditis elegans*	378	$4e^{-09}$

			NP_723898.1	*Drosophila melanogaster*	490	$2e^{-24}$

Supplementary Table 3 Id Protein, putative name, length, ID protein Ortholog, Ortholog Species, Ortholog Length and E-value of *P. canaliculata* proteins

ID protein	Putative name	Length (AA)	ID protein Ortholog	*Ortholog Species*	Length (AA)	E-value
Pca61914	Argonaute 1.1	936	NP_493837.1	*Caenorhabditis elegans*	891	$7e^{-44}$
			NP_001163498.1	*Drosophila melanogaster*	866	0.0
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Pca59652	Argonaute 1.2	468	NP_493837.1	*Caenorhabditis elegans*	891	$3e^{-36}$
			NP_001163498.1	*Drosophila melanogaster*	866	$6e^{-111}$
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Pca69814	Argonaute 1.3	897	NP_493837.1	*Caenorhabditis elegans*	891	0.0
			NP_001163498.1	*Drosophila melanogaster*	866	$1e^{-32}$
			XP_013068463.1	*Biomphalaria glabrata*	895	0.0
Pca67531	Dicer	1984	NP_498761.2	*Caenorhabditis elegans*	1910	$1e^{-12}$
			NP_524453.1	*Drosophila melanogaster*	2249	$3e^{-48}$
			XP_013067888.1	*Biomphalaria glabrata*	2332	0.0
Pca67887	Drosha	1525	NP_492599.1	*Caenorhabditis elegans*	1086	$5e^{-171}$
			NP_477436.1	*Drosophila melanogaster*	1327	0.0
			XP_013069067.1	*Biomphalaria glabrata*	1469	0.0
Pca62316	Exportin 5	1140	NP_741567.1	*Caenorhabditis elegans*	1080	$1e^{-11}$
			NP_001285453.1	*Drosophila melanogaster*	1241	0.0
			XP_013069067.1	*Biomphalaria glabrata*	1469	0.0
Pca65193	Exportin 1	974	NP_741567.1	*Caenorhabditis elegans*	1080	0.0
			NP_001303316.1	*Drosophila melanogaster*	1063	0.0
			XP_013069067.1	*Biomphalaria glabrata*	1469	0.0
Pca60969	Tudor - SN	898	NP_494839.1	*Caenorhabditis elegans*	914	0.0
			NP_001261195.1	*Drosophila melanogaster*	926	0.0
Pca65324	Loquacious	258	NP_723813.1	*Drosophila melanogaster*	419	$3e^{-25}$

Pca59979	PASHA	731	NP_001293461.1	*Caenorhabditis elegans*	751	$4e^{-28}$
			NP_651879.1	*Drosophila melanogaster*	642	$2e^{-132}$
Pca69254	VIG	390	NP_723898.1	*Drosophila melanogaster*	490	$8e^{-10}$

Supplementary Table 4 Id Protein, putative name, length, ID protein Ortholog, Ortholog Species, Ortholog Length and E-value of *P. maculata* proteins

ID protein	Putative name	Length (AA)	ID protein Ortholog	Ortholog Species	Length (AA)	E-value
Pma35144	Argonaute 1.1	909	NP_493837.1	*Caenorhabditis elegans*	891	$8e^{-41}$
			NP_001163498.1	*Drosophila melanogaster*	866	$2e^{-179}$
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Pma60501	Argonaute 1.2	937	NP_493837.1	*Caenorhabditis elegans*	891	$1e^{-43}$
			NP_001163498.1	*Drosophila melanogaster*	866	0.0
			XP_013089248.1	*Biomphalaria glabrata*	897	0.0
Pma63715	Argonaute 1.3	891	NP_493837.1	*Caenorhabditis elegans*	891	0.0
			NP_001163498.1	*Drosophila melanogaster*	866	$1e^{-34}$
			XP_013068463.1	*Biomphalaria glabrata*	895	0.0
Pma65850	Drosha	359	NP_492599.1	*Caenorhabditis elegans*	1086	$2e^{-96}$
			NP_477436.1	*Drosophila melanogaster*	1327	$2e^{-96}$
			XP_013069067.1	*Biomphalaria glabrata*	1469	$8e^{-178}$
Pma51393	Drosha	580	NP_492599.1	*Caenorhabditis elegans*	1080	$4e^{-77}$
			NP_477436.1	*Drosophila melanogaster*	1241	0.0
			XP_013069067.1	*Biomphalaria glabrata*	1469	0.0
Pma59467	Exportin 1	1070	NP_741567.1	*Caenorhabditis elegans*	1080	0.0
			NP_001303316.1	*Drosophila melanogaster*	1063	0.0
			XP_013069551.1	*Biomphalaria glabrata*	1076	0.0
Pma63331	Exportin T	966	NP_501320.2	*Caenorhabditis elegans*	950	$1e^{-97}$
			XP_013079973.1	*Biomphalaria glabrata*		0.0
Pma32975	Tudor - SN	603	NP_494839.1	*Caenorhabditis elegans*	914	0.0
			NP_001261195.1	*Drosophila melanogaster*	926	0.0
Pma62419	PASHA	709	NP_001293461.1	*Caenorhabditis elegans*	751	$2e^{-28}$

			NP_651879.1	*Drosophila melanogaster*	642	2e^{-135}
Pma51302	VIG	414	NP_001367502.1	*Caenorhabditis elegans*	378	3e^{-10}
			NP_723898.1	*Drosophila melanogaster*	490	8e$_{-25}$
Pma61707	Loquacious	316	NP_723813.1	*Drosophila melanogaster*	419	4e^{-45}

SUPPLEMENTARY DATA II

Supplementary Table 1– Structural and thermodynamic characterization of miRNA precursors in *L. nyassanus*. Size (Tam), G content, A content, C content, U content, GC content, AU content, AU ratio (R-AU), GC ratio (R-GC), Minimum Free Energy (MFE), Minimum Free Energy of the Pool (MFEE), Frequency of the MFE Structure in the Pool (Freq), Diversity of the Pool (Div), Minimum Adjusted Free Energy (AMFE), Minimum Free Energy Index (MFEI).

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-316	UAGUUGUUAAACUGUAGGAAAACGUCUCUUAUUCCUAUCGGCUCAAUAUAUUUAUGAGCCUAGGCAACAAAUAUACUGUCUUUUUCCGCUUACAAAUG	98	15,30612	29,59184	19,38776	35,71429	34,69388	65,30612	1,206897	0,789474	-18,9	-14,1	0,015799	21,7	-19,2857	-0,55588
lny-mir-7965	AUGCAUCGAAGAAACCUCAAAUGACACGUUUUAAUUAGAAAUGGUGAGGGAAUGAAUUUUGUAUGCUAAAACAUAAAAUUGUCAGCAUUCUUGGAUGAUU	100	20	37	12	31	32	68	0,837838	1,666667	-20	-16,2	0,094567	7,12	-20	-0,625
lny-mir-2064	GACCAGUAUGGUCAGAGUUGAUGCUUAAGGCGCCAUUCCGUGGCUUAGCACAGCUUUUGACAAAUAAGCAGCACUGUGCAACUGCAA	87	25,28736	27,58621	22,98851	24,13793	48,27586	51,72414	0,875	1,1	-27,2	-26,8	0,071601	11,7	-31,2644	-0,64762
lny-mir-3484	aauacaaacagCUAAACUAGGAAAAGGAUAUGCUGUGCUUUCCACAUGGUGUUCCUACACAGAUUUUUCUAGUUACcuguuuguggu	87	13,7931	20,68966	14,94253	25,28736	28,73563	45,97701	1,222222	0,923077	-26,7	-26,3	0,091839	12,31	-30,6897	-1,068
lny-mir-10a	GGGGUGAGACCUACCCUGUAGAUCCGAAUUUGUGUGACAUUCGU	82	28,04878	21,95122	23,17073	26,82927	51,21951	48,78049	1,222222	1,210526	-38,9	-38,9	0,039205	2,67	-47,439	-0,92619

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GACGACAAAUUCGUGUUUGCACGGUAAGGCCUCACUGC															
lny-mir-11	GACCCUAUGGCAAUGUGUCGAGGCUCAUCACAGGCAGAGUUCCagaccugucugucugcugacaUCUCUCUCAGACUGCCAUGGCCA	87	18,3908	16,09195	24,13793	17,24138	42,52874	33,33333	1,071429	0,761905	-29,4	-20,8	0,07357	22,9	-33,7931	-0,79459
lny-mir-2b-1	GAUCUGCUCAUCAAAGCUGGCUGUGAUAUGACUUACAAAAACACAUCCCAGCAGCUUUGAUGAGAAGGCC	70	21,42857	31,42857	24,28571	22,85714	45,71429	54,28571	0,727273	0,882353	-27,6	-27,6	0,092458	3,86	-39,4286	-0,8625
lny-mir-745a	AUUUGUCGGCUGGAGGCGGCUUUUCAACAGGCUGCCUUGCUUGGGAGGACACAAGCUGCCUGAUGAAGAGCUGUCCUGUAGCGGGCAGUA	90	34,44444	18,88889	22,22222	24,44444	56,66667	43,33333	1,294118	1,55	-46,8	-40,8	0,132252	8,07	-52	-0,91765
lny-mir-7c	GUGAUCUGGAAGACUAGUGAUUUUGUUGUUCUGAUGCUGACUAACAAUAAAUCACAUUCUUCCCCGAUUGC	71	19,71831	25,35211	19,71831	35,21127	39,43662	60,56338	1,388889	1	-21,1	-18,6	0,131089	9,74	-29,7183	-0,75357
lny-mir-9319	UCUCCUUCCCACUCACAGUCACUAGCAGAGCUAGUGGCCUCACCCCCACUACUUGCACUGGCUGCUUCAGGUGAAGGGGG	80	23,75	18,75	35	22,5	58,75	41,25	1,2	0,678571	-30,4	-30,4	0,219681	2,97	-38	-0,64681
lny-mir-466q-1	AUGGUCUGGUCAUGACAGAcaggcgcgcgcgugcacacacacacauacacacagacgugUAAAUGUCUGCUCGCGCGUCGUCUGACGUAAUACAUACAGA	100	15	16	13	16	28	32	1	1,153846	-34,9	-33,3	0,065729	20,26	-34,9	-1,24643
lny-mir-3297	UAAGCAAGGGUCACAAAACGCCGCUUUCAAGAUAGUGCAGAGAUUCAGCAGAGCACAUCUAGUCAAAA	93	23,65591	33,33333	22,58065	20,43011	46,23656	53,76344	0,612903	1,047619	-22,6	-22	0,137599	8,48	-24,3011	-0,52558

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCAAGGUUUGUGACGAUCUUGCAGC															
lny-mir-7460	AUCAGCAUUCAGCCUGGCACCAUCCUCUUCACAAACAACAUCUGCCUGACUGAGCUCUGCUAGGGCUGUUUUGAGGGCCUGUGCGUGUCGAAUGCUGUC	99	24,24242	19,19192	29,29293	27,27273	53,53535	46,46465	1,421053	0,827586	-28,3	-17,6	0,013885	26,75	-28,5859	-0,53396
lny-mir-96b	CAUGGCUGUUCAAUUAUUUGGCACUUGUGGAAUAAUCGUUGCAUCUGAAGUCGAUUAUACAUCGGUGCCAAGUCAUCAGAAAGGCCGGA	89	24,7191	26,96629	19,10112	29,21348	43,82022	56,17978	1,083333	1,294118	-30,3	-28,8	0,042265	5,05	-34,0449	-0,77692
lny-mir-564	UGAGGUGAACAUUGUACGUGCCAACAAGACUGAAGGCACGGUGGCAGCAGAAGGGCAGGagauagguagguaauuuuuggCAGACAGUUAUUCCACCUUA	100	24	24	17	14	41	38	0,583333	1,411765	-22,3	-14,12	0,009426	22,56	-22,3	-0,5439
lny-mir-6632	CUUAUCGUCAGUGCUCAUCUCAGUCCgauagaaaagagagagaugagaggagUGCUGAACCAUUCC	66	9,090909	10,60606	21,21212	19,69697	30,30303	30,30303	1,857143	0,428571	-18,9	-17,8	0,132348	11,84	-28,6364	-0,945
lny-mir-6504	guuugugguguaacCAUUACAGCACAGCCAUGUUUGCGGUGUAACCAUUACAGCACAGCUAUGUUUAUGGUGUAACCAUUACAGCACAGUU	91	16,48352	24,17582	19,78022	24,17582	36,26374	48,35165	1	0,833333	-26,5	-21	0,034382	27,18	-29,1209	-0,80303
lny-mir-4262	UAAAUGCCCAGCAGCUGGGACAACGUCGACAGGCCGACAUUCAGACUACCUGUCGACGUCGCCUUGAAUGCUGGGAUAAU	80	26,25	26,25	27,5	20	53,75	46,25	0,761905	0,954545	-36,8	-35,1	0,460005	2,87	-46	-0,85581
lny-mir-133c	UAACUACAGCUGGUUGAAGGGGACCAAAUGAAAGCCUCUUCAACA	75	22,66667	33,33333	22,66667	21,33333	45,33333	54,66667	0,64	1	-30,4	-27,8	0,231076	4,17	-40,5333	-0,89412

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AUUUGGCCCGAGUUCAACCAGCUAUAGAAG															
lny-mir-10b	AGAGAGUGUGGGUGGCUUUACCCUGGAGAACCGAGCGUGUgugaucaugacgucacagcucguUUCUGAGGGUCAAGUCGCGAUACUCGCG	91	27,47253	14,28571	15,38462	17,58242	42,85714	31,86813	1,230769	1,785714	-41,7	-41,6	0,288526	9,6	-45,8242	-1,06923
lny-mir-8908i	cucaagaaagaaagaaauuugcaaUCCCCGAUAAACCCAAUGUAAUAGCAACUACCCGGUAAGGUAGUUGCCaagaaugucuuuuuuuuuuacc	94	9,574468	15,95745	14,89362	10,6383	24,46809	26,59574	0,666667	0,642857	-21,2	-18,84	0,113508	8,17	-22,5532	-0,92174
lny-mir-1993	AGGAAGGCCCUGUAAUUUUCGGGAAUAUCGGCAUAAUGCAGUUGACUUGAGAUUCGUAUUAUGCUGCUAUUCACGAGAUGGAAGGGCUUGGA	92	29,34783	26,08696	15,21739	29,34783	44,56522	55,43478	1,125	1,928571	-36,7	-36,7	0,374278	3,58	-39,8913	-0,89512
lny-mir-466m	gugugugugugacugugugugcaugugcaugugugugguuugcaugcaugcacaauaaGGCCCUCCAGUCCCAGACAGGC	79	7,594937	5,063291	12,65823	2,531646	20,25316	7,594937	0,5	0,6	-33,8	-30,8	0,291201	4,92	-42,7848	-2,1125
lny-mir-8279	GAAACAGUUGAAGAACAGAGCUGAGAGAUUCAUGUCAAUCUGACAUUUGCAGUAGGUCUGCUagcuacauguauuuucaagUGUAGA	87	20,68966	25,28736	11,49425	20,68966	32,18391	45,97701	0,818182	1,8	-23,3	-21	0,095311	11,78	-26,7816	-0,83214
lny-mir-7661	CAAGCAGAUGUUGAAGGAAGUUCGUGUuucuacaaaagaaagaaaccuggAUAAACUAGACACUUCCAUCAAAGCGUGCAUC	82	15,85366	24,39024	14,63415	17,07317	30,4878	41,46341	0,7	1,083333	-19,2	-19,2	0,214952	11,41	-23,4146	-0,768
lny-mir-87a	AAAUCUUUGUUAAACACUUGGUAAUUUGCAAAUgaacaugcaaauaaaaaggaacacaugacugGCACAUGGA	99	10,10101	29,29293	11,11111	18,18182	21,21212	47,47475	0,62069	0,909091	-19,3	-19,3	0,407153	3,9	-19,4949	-0,91905

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AAUAAACCCAAGUUAACAACAGAUGA															
lny-mir-466b	CauagagagaauuacauauacauacacacauacuacUCACGCCAGAUAUGUGUCUACAUGUAAUUCUCUCUGUU	74	8,108108	10,81081	13,51351	20,27027	21,62162	31,08108	1,875	0,6	-24,6	-22,9	0,096333	7,47	-33,2432	-1,5375
lny-mir-10349	CACUGAGCACCAAUAAGCAGCAAUAAGCGGCGAUAGCUCACGACCAGUGAUCACGUGGUCGUUAUCAGCGGGGUUAUAUCCCUUGACUGGUAAUCACUG	99	25,25253	27,27273	25,25253	22,22222	50,50505	49,49495	0,814815	1	-28,4	-25,9	0,020752	14,01	-28,6869	-0,568
lny-mir-2049	AACUGUAUGUAGCACAGCAAUGACGUGAAAUUUUGUGCAUGUACGUGgauucaugugcaugcauucauga	70	17,14286	20	10	20	27,14286	40	1	1,714286	-23,5	-23,5	0,409874	4,97	-33,5714	-1,23684
lny-mir-5920	UCAGUGGGUGCCUGUAGCCUGCCGUUUUGUCAUUCGUAUACAAAACAGUAGGCCACUGCCCCACAUA	67	22,38806	22,38806	28,35821	26,86567	50,74627	49,25373	1,2	0,789474	-29,7	-29,7	0,51133	1,3	-44,3284	-0,87353
lny-mir-5600	AGUUGUAGUUAACUAAAUAGGACAUUUUGAAAAGAAGUGUUCUUGCUUCGUAAAUGCACUUAAUACAUGAGUGGaauguuuuguuucaguacuACAGAG	99	18,18182	28,28283	9,090909	25,25253	27,27273	53,53535	0,892857	2	-25,7	-25,7	0,154731	5,98	-25,9596	-0,95185
lny-mir-12096b	UGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUACGAGGUAGGGCAAAAUUAUCACCGAUUACCCGAGGUAUCG	85	23,52941	25,88235	22,35294	28,23529	45,88235	54,11765	1,090909	1,052632	-29	-29	0,105265	6,21	-34,1176	-0,74359
lny-mir-263a-1	ACUUUCAAGAGUUGGCCAUGCUUAAUGGCACUGGUAGAAUAGCACACACCAUAGAGAUUAGUGCUGC	95	25,26316	31,57895	17,89474	25,26316	43,15789	56,84211	0,8	1,411765	-24,2	-20,6	0,027018	17,04	-25,4737	-0,59024

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AGUCAAGAGAGUGGUCAAUAUUGAACGU															
lny-mir-124c	AAAUGAUGGUCUUCCCCUUGGCAUUCACCGCGUGCCUUAAUAGUGUGUCAAUCACCAAGGCCAACGCAgugaacacaaacagguaagCCCGGUCACAC	98	16,32653	19,38776	26,53061	18,36735	42,85714	37,7551	0,947368	0,615385	-26,5	-26,3	0,051306	9,68	-27,0408	-0,63095
lny-mir-4024	GUCUUUUUUACCUGUCCAAAGCCGUUUUUGAAUAGCUGUGCUACCGACUUUACUGUCUGUUAUUUGUAGGAUGAAAAGGUUAUUGGAAGUGAAAAAUUA	99	21,21212	26,26263	14,14141	38,38384	35,35354	64,64646	1,461538	1,5	-18,8	-16,9	0,025084	15,96	-18,9899	-0,53714
lny-mir-216a	guguuugucUAAUCUCAGCUGGUAAUUCUGAGUGGAGUUGUGUACACCUCAAGUUACUCGCCGAGAUUACAUAAAUAU	78	17,94872	25,64103	16,66667	28,20513	34,61538	53,84615	1,1	1,076923	-26,2	-26,2	0,262819	2,61	-33,5897	-0,97037
lny-mir-12	UUGAAUGGUCAGACUGUGAGUAUUACAUCAGGUACUGAGAAAACAGCAUGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUUUG	89	21,34831	25,8427	16,85393	35,95506	38,20225	61,79775	1,391304	1,266667	-42,9	-42,9	0,38245	2,26	-48,2022	-1,26176
lny-mir-8335-1	ucuuguuguuguuguuguuuugguccuuGCAAAAAGUUUGAACCUCUCAACACUUUUUGAGGUACUUCAAAGCAAAUUACACAAACaggc	90	7,777778	25,55556	14,44444	16,66667	22,22222	42,22222	0,652174	0,538462	-24,3	-24,3	0,033164	11,07	-27	-1,215
lny-mir-9-1	CAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUACUUCAAUCAUACAGCUAGAUAACCAAAGACAAAACCG	76	11,84211	32,89474	22,36842	32,89474	34,21053	65,78947	1	0,529412	-24,2	-24,2	0,496783	1,38	-31,8421	-0,93077

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-8335-2	GUACACAAGCAAAAGUACAAUUGACAAAUGUUCCAGAACAGCCCAGCGUCUUUugacacuguuguuguuguuguuuuguuuuguggag	88	10,22727	22,72727	14,77273	12,5	25	35,22727	0,55	0,692308	-21	-18,9	0,011022	19,33	-23,8636	-0,95455
lny-mir-228	AGGCAUCAUGUGAAAAGAGCAUCAAUUGUAUAAUGGCACUGCAUgaauaaaaaauuauguuugCCAGAAAGAAUUGUGUCUUUGCUAAAUGACUGGUU	98	18,36735	26,53061	12,2449	23,46939	30,61224	50	0,884615	1,5	-22,7	-22,5	0,078762	7	-23,1633	-0,75667
lny-mir-2987	CGACCUGAGACUUGUCUGAUUGUUCAGGAAAGUCUGGAUCUCGCACACUACGUUAGCUGCUUGAAGACUUCAGAGCAAGGCUGAGGCUU	89	26,96629	23,59551	22,47191	26,96629	49,4382	50,5618	1,142857	1,2	-24,9	-16,6	0,027121	27,97	-27,9775	-0,56591
lny-mir-252b-1	GAGAGUAAGUACUCUCGACAGGGGGCGCUUCCUCCAACUUGCUGCGCAGAUGGUGUCCUUGGAGGUACGUGUCCCUGCGCAGUCCUACUGGG	92	32,6087	16,30435	27,17391	23,91304	59,78261	40,21739	1,466667	1,2	-34,9	-26,6	0,007771	20,62	-37,9348	-0,63455
lny-mir-9701a-1	GUGACAGCaggcugugugacuguguaccACCAUCAACCCGCGACGUGUGACCACAGUCGUAUUUUACUGGAGGUACAGGACAAAUGUCAGCUGUGUU	97	20,61856	20,61856	19,58763	18,5567	40,20619	39,17526	0,9	1,052632	-30,9	-30,1	0,097108	9,96	-31,8557	-0,79231
lny-mir-281	ACGUGACCUCACGCUGAAGGGAGCAUCCGUCGACAGUCAGAAAUACAGCACUGUCAUGGAGUUGCUCUCUUUACUGGAAAGGUCAAGG	88	27,27273	27,27273	23,86364	21,59091	51,13636	48,86364	0,791667	1,142857	-36,5	-36,1	0,381583	2,16	-41,4773	-0,81111
lny-mir-466i-1	GUUUGACAAGUCUGCuagcugacugugugugugugugagugugugugug	100	11	18	15	12	26	30	0,666667	0,733333	-35,6	-34,2	0,076901	11,8	-35,6	-1,36923

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	ugacgcacAAAGACAUACAAACCACCGUCCCAGUCUGAACUGGUCAGAU															
lny-mir-3128	GAaugguuuuucuuauuucagacUUGGAUUUGAAGAUGAAUGGUCAUGUGAUCCAUGGCACAUCCAUUGCAGUUGAUUUCUGGCAAGUAAAAAACCAUCC	100	18	24	14	23	32	47	0,958333	1,285714	-26,1	-23,4	0,009444	10,94	-26,1	-0,81563
lny-mir-279	GUCUCUUAUCUUUGCGGGUGGCUGUGAGUCUAGUCCAUGUGACAAGGCCAGCUCAUGACUAGAUCCACACUCAUCCACAAGGAUAAGACGG	91	25,27473	24,17582	25,27473	25,27473	50,54945	49,45055	1,045455	1	-40,2	-39,7	0,065907	7,67	-44,1758	-0,87391
lny-mir-36b	cuGGGGUGCGGUGGGUGUACGCCCUGGUUGGUGGGCGUCGGUAACAUCACCGGGUAUACAUUCAUCCGCACCCUGA	76	35,52632	14,47368	25	22,36842	60,52632	36,84211	1,545455	1,421053	-43,9	-41,1	0,167581	3,38	-57,7632	-0,95435
lny-mir-1662	GUAUAUUGUGUUGCAGAAAUUUGACAUCACCAUACUUUAUGUCGUCAGUUUGACAUCAUCAUACUUUAUGUCAUAAGUUUGACAUCACCAUACUU	95	13,68421	28,42105	18,94737	38,94737	32,63158	67,36842	1,37037	0,722222	-19,6	-19,6	0,371187	6,34	-20,6316	-0,63226
lny-mir-9a	ggaagggaaggagacGCAGUCCUUAAGCCCGCAGUUCUGUAACUGUCGAUAAGAAAUAAAGCUAGGCGACUGUCCGUCCUCGUUGCCUAAU	91	18,68132	20,87912	21,97802	21,97802	40,65934	42,85714	1,052632	0,85	-26,4	-17,7	0,034951	9	-29,011	-0,71351
lny-mir-8335-3	UGGCAUCGUUAGAACUUGAUUAAAGCAGGCUUGACucaugcuuuguuguuguuguuguuuugaagccaGAUUAAUCAUCUAAUGUGGCU	92	14,13043	17,3913	9,782609	19,56522	23,91304	36,95652	1,125	1,444444	-20	-18,3	0,054089	28,71	-21,7391	-0,90909

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-2001	GCUGCCAUUUUGUGACCGUUACAAUGGGCauugacagaaagacaaaugcuCCAUGUGAUGUUCACAAGAUGGCGGU	76	21,05263	14,47368	15,78947	21,05263	36,84211	35,52632	1,454545	1,333333	-39,7	-39,7	0,259527	3,56	-52,2368	-1,41786
lny-mir-252b-2	GGGGAUAGUGCCGGCGAUAAGUAGUGGUGCCGCAGGUAUGGCUCGUCAACUACCUGCUCAUCUCUGCUUAAUGUUGCUCUAUCAUU	86	27,90698	18,60465	23,25581	30,23256	51,16279	48,83721	1,625	1,2	-33,5	-33,5	0,133838	3,27	-38,9535	-0,76136
lny-mir-252a	AUUUUCUGGCCCACUACUAAGUACUAGUGCCGCGGGAGGACAGCUUGCCAUGUCCCGUGGUUCUGGUCCUUACUACAGGGCGGAAUUU	88	27,27273	18,18182	26,13636	28,40909	53,40909	46,59091	1,5625	1,043478	-37,2	-36,8	0,059207	5,64	-42,2727	-0,79149
lny-mir-7371d	ACUGUAGGCAGCAGAGAGAACUUCCAGUAAAUGCAGUUCGCAAUCAUGGAAACACCAUUCACCAUAUUUGUACCAGAUGCUCUCUCUGGUGCCCACAGA	99	20,20202	30,30303	26,26263	23,23232	46,46465	53,53535	0,766667	0,769231	-33,9	-33,1	0,261583	12,77	-34,2424	-0,73696
lny-mir-190	UGCCCACCUACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCGGUUGUCAUGACGACCAAGUAGUCAAACAUGUCAGAGCAGCGGUGGGGCUU	97	26,80412	21,64948	22,68041	28,86598	49,48454	50,51546	1,333333	1,181818	-44,3	-44,3	0,312382	10,4	-45,6701	-0,92292
lny-mir-92b-1	GACAGUCAGGGUUGUGUAGACCGGUACAGGGGCAAUGCAGUGAGUCGCUUGUAUUGCACUCGUCCCGGCCUAUACAGUCUGACUCGG	87	32,18391	19,54023	24,13793	24,13793	56,32184	43,67816	1,235294	1,333333	-41,1	-41,1	0,171444	4,85	-47,2414	-0,83878
lny-mir-92a	GCAUUAGCAUUAGGUCUUGAUGGGUGCAAUACUGGUGAGCCUGAUGGCAGAUUGCACUCGUCCCGGCCUUCUGCUAAAAG	80	28,75	21,25	22,5	27,5	51,25	48,75	1,294118	1,277778	-36,4	-36,4	0,391022	2,84	-45,5	-0,8878

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-92b-2	UAGCAUAGGGAGGUUGUGACCUGUGCAAUGAUUGGAGGAAUAUGAGCCAAUUGCACUUGUCCCGGCCUGCCCUGUGUGC	79	31,64557	20,25316	21,51899	26,58228	53,16456	46,83544	1,3125	1,470588	-35,3	-35,1	0,232461	3,35	-44,6835	-0,84048
lny-mir-184b	ACCCACAAGCCCUUAUCAGUUCUCCGUCCAGUGCUUGAGAACAGACGGGGCGGGGGAGUGAUAAGGAAAUGUGAAA	76	30,26316	28,94737	22,36842	18,42105	52,63158	47,36842	0,636364	1,352941	-35	-33,4	0,17411	3,39	-46,0526	-0,875
lny-mir-306	CAGCACCGAGACCGGCCAAUUUUGUCUUUCGAAAGAUCUUCCAGUUAAGAACACCAGCCUUGACACCUGUGACAAAUUUUGGUCAGGUACUGAGUGACU	99	21,21212	27,27273	25,25253	26,26263	46,46465	53,53535	0,962963	0,84	-24,1	-22,8	0,10238	6,82	-24,3434	-0,52391
lny-mir-1a	GCCCGGCACAUACUUCUUUGCUAUCCCAUAUGUUCUUCCAAAGCUAUGGAAUGUAAAGAAGUAUGUACCGUGU	73	19,17808	26,0274	23,28767	31,50685	42,46575	57,53425	1,210526	0,823529	-29,2	-29,2	0,14159	5,1	-40	-0,94194
lny-mir-4690	UCACCUCCCGCUCAUCUCCUGGUCUGACCAGCACCGGCAGUCCGCGAUCGGGUGUGCUGGGGAGCAGGCGAGGCUGGGAGGGCU	84	36,90476	13,09524	32,14286	17,85714	69,04762	30,95238	1,363636	1,148148	-40,6	-39,2	0,073847	8,43	-48,3333	-0,7
lny-mir-8075	gcuUGACUGCUGAUGGCAGAUGUGUGUUGGCUGUUCAAUAAACAGAGCCCCACUGCCACACUCAGAUCAUGU	72	23,61111	23,61111	23,61111	25	47,22222	48,61111	1,058824	1	-19,8	-10,6	0,03638	15,58	-27,5	-0,58235
lny-mir-669k	UUUCUCUCGUACACUGAAGUCCAUAUCUUCAAGAGUGUUUAgaugugcaugucuguuuacuugugugugcauguauggacGUGCAUGUGUAUCAGAGAGG	100	14	15	11	21	25	36	1,4	1,272727	-32,7	-32,3	0,067561	17,27	-32,7	-1,308

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-9708c	GGGAAGUUGAGGGCUAGAAAAAUUUGUGUAGCACCAGACAUUUGugagcacagcuaaacauuuucuCUGCUGAGAUAAUUUUGUGGCGCCUCAACUUCCC	100	21	20	15	22	36	42	1,1	1,4	-39	-31,9	0,079272	20,93	-39	-1,08333
lny-mir-1731	uuauuuacuugacUGCAGGCACUACAGAUUAGUGAUAAUGCUGUUCCAGUCAGUCGAAAUCUGUAUUGAUUCGGUGUAAGUAAACGC	87	20,68966	24,13793	14,94253	25,28736	35,63218	49,42529	1,047619	1,384615	-21,8	-21,7	0,069971	9,58	-25,0575	-0,70323
lny-mir-8066	GauucccgcccccuccccuucucccacacaucGACCUUCCCAUGGUCCGAGGGGCCGAUGCAAUGUGAUCUUUUGGAGaacauggagaugggggaugU	98	16,32653	8,163265	11,22449	13,26531	27,55102	21,42857	1,625	1,454545	-34,7	-29,6	0,27164	13,85	-35,4082	-1,28519
lny-mir-5100	CGGGCACGUGACACCUACGUCAUGGCUGUUGUCGAAUCCCAGCGGUGCCCGAGAGCUGGUUCCAGUAACAGCUCACCUCGUCCUGGAGCCACGUGCGUC	99	29,29293	17,17172	33,33333	20,20202	62,62626	37,37374	1,176471	0,878788	-36,8	-36,7	0,095799	8,39	-37,1717	-0,59355
lny-mir-3906	agGGGCAAUGCUCUUGCAAGUUUUUUAUCCAAUAGUUGGAGCAGGAAGAAAAGCAUUUUGAAUGCAUAAAAAGCAUUCCAUAGACCACUCCUGU	94	20,21277	31,91489	18,08511	27,65957	38,29787	59,57447	0,866667	1,117647	-20,6	-16,8	0,091919	20,31	-21,9149	-0,57222
lny-mir-7062	GAUAAAAAGCUGCACAGCUGAUGGCCAUCCUGUUCGUGGAGGCCAGCUUGUGUCCAGGUUAUUGCA	66	28,78788	22,72727	22,72727	25,75758	51,51515	48,48485	1,133333	1,266667	-20,8	-18,1	0,025013	10,43	-31,5152	-0,61176
lny-mir-8485-1	UUugugguacacacacacacacacacacacacacguacuuaAAAUUUAGACUGCAGGGUGUUCGUGCGUUAAUUUCUAUGUGUGGGGGUACCACCAA	97	16,49485	13,40206	9,278351	20,61856	25,7732	34,02062	1,538462	1,777778	-25,5	-18,6	0,096418	19,61	-26,2887	-1,02

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-6756	ucccCCCACUUCUUGCCCCUCCCUCGCGCAUGGAUAUUACAUAUAUGAAAAUAUUACUUUUUGACUAGGGUGGGGCUGGAGGUGGGGAUA	90	24,44444	21,11111	21,11111	28,88889	45,55556	50	1,368421	1,157895	-39	-37,9	0,064711	9,53	-43,3333	-0,95122
lny-mir-10790	uuggauacAGAAAGCAGACUAACGGACGUCAGCAAGCAGGUGACACUGUCUGGCUgcucucuuuauuucucucucuguucguc	83	16,86747	18,07229	13,25301	8,433735	30,12048	26,50602	0,466667	1,272727	-21,7	-18,1	0,055432	16,72	-26,1446	-0,868
lny-mir-153	ACACCCCCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGCUAUCAAAUUGCAUAGUCACAAAAGUGAUCGGGAGGCGGGGACU	88	23,86364	27,27273	25	23,86364	48,86364	51,13636	0,875	0,954545	-40,2	-40,2	0,334594	4,07	-45,6818	-0,93488
lny-mir-124	GUGUGACCGGGcuuaccuguuugug uucacUGCGUUGGCCUUGGUGAUUGACACACUAUUAAGGCACGCGGUGAAUGCCAAGGGGAAGACCAUCAUUU	98	26,53061	18,36735	16,32653	19,38776	42,85714	37,7551	1,055556	1,625	-40,2	-38,8	0,093576	10,13	-41,0204	-0,95714
lny-mir-133	CUUCUAUAGCUGGUUGAACUCGGGCCAAAUUGUUGAAGAGGCUUUCAUUUGGUCCCCUUCAACCAGCUGUAGUUA	75	22,66667	21,33333	22,66667	33,33333	45,33333	54,66667	1,5625	1	-35,5	-35,1	0,187007	6,7	-47,3333	-1,04412
lny-mir-10365	uuguGCCCGCAGUCCAUUAGAAUGUGGAAUCUAUGCUUUAAUGGUUGUGACAGAUUCUACUUUAAAAGACAUCUGGCAACC	81	19,75309	27,16049	19,75309	28,39506	39,50617	55,55556	1,045455	1	-20,4	-20,4	0,113197	12,46	-25,1852	-0,6375
lny-mir-466h	CCUGCuucgcacacacgcauacgcacgcacacacacacaucuccucGGCGACACGUGAAGAUGCACGUGGGAGGGUGUUCAUGUGUGCGAAUGCCUG	97	22,68041	10,30928	12,37113	12,37113	35,05155	22,68041	1,2	1,833333	-34,7	-33,9	0,086928	14,95	-35,7732	-1,02059

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-981	UUGAUCAGACUUUGUACCGGGUUUCGCGACUGGCGAACAGUCAUUCUGAACUGUGUUCGUUGUCGUCGAAACCUGCCCUGAACUCUGAUGUG	92	26,08696	18,47826	23,91304	31,52174	50	50	1,705882	1,090909	-35,3	-34,1	0,018788	7,48	-38,3696	-0,76739
lny-mir-7398y	AUGAGAUGUCGGAUGCAAAUGUGAUGGAGUGGGUGGUUUAaccuucccuucucucuuucucucucucaacaacUCGAUCUUUGUCAGGCAACAUCUCAA	99	20,20202	17,17172	9,090909	20,20202	29,29293	37,37374	1,176471	2,222222	-24,9	-23,9	0,214701	12,35	-25,1515	-0,85862
lny-bantam	uuugacgAAACUGGUUUUCACAGUGAUCCAGCAGAUCGUUCGAAGUCUGAGAUCAUUGUAAAAACCAAUUUUGUCGCC	78	17,94872	26,92308	19,23077	26,92308	37,17949	53,84615	1	0,933333	-31,9	-31,9	0,266041	5,12	-40,8974	-1,1
lny-mir-7006	GGUCACAGGUGAGUGGGGGUGUUCAAGAccccccaguuuucugugggccagcaucauagGGGUGAUGCACAUCGCCUCUCCCUUGCUG	88	23,86364	10,22727	14,77273	15,90909	38,63636	26,13636	1,555556	1,615385	-44,5	-43,9	0,07812	5,45	-50,5682	-1,30882
lny-mir-2a-1	GUGCCAAAGCUGUGAUGCUGACCAAGUGACUGGGAUGUGUGUUUGUUGUCAUAUCACAGCCUGCUUGGAUC	71	29,57746	19,71831	19,71831	30,98592	49,29577	50,70423	1,571429	1,5	-20,8	-17,8	0,088092	9,24	-29,2958	-0,59429
lny-mir-2b-2	GGCCUUCUCAUCAAAGCUGCUGGGAUGUGUUUUUGUAAGUCAUAUCACAGCCAGCUUUGAUGAGCAGAUC	70	24,28571	22,85714	21,42857	31,42857	45,71429	54,28571	1,375	1,133333	-32,9	-32,9	0,167727	4,14	-47	-1,02813
lny-mir-2d	GUAAGCAUGAAUGGAGGGCCAGCCAAAGUGGCGGUGAUCUGAUGUUCAGAGCAAAUCACAGCCUGCU	92	30,43478	23,91304	20,65217	25	51,08696	48,91304	1,045455	1,473684	-40,6	-40,6	0,14781	5,41	-44,1304	-0,86383

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUGGUGAGCCUUCUGUCAUUCUGAG															
lny-mir-2a-2	AAUUGACCGCAGAGGUCACACCAAAGAGGUUGugacuuugucuuucuguucaaaUCACAGCCUGCUUUGGUGAGCUCUUUGUCGUCAGGU	90	21,11111	16,66667	17,77778	20	38,88889	36,66667	1,2	1,1875	-39,5	-39,1	0,224362	3,21	-43,8889	-1,12857
lny-mir-6868	GUAUUCCUUCUGUUGUCUGUAAACUGCUUucgggaaaaaaacccauuguuCCUCCUUUGAGUUCUCAGGUAACAAGAGGAAAAG	84	15,47619	17,85714	15,47619	26,19048	30,95238	44,04762	1,466667	1	-23	-22,2	0,132737	4,31	-27,381	-0,88462
lny-mir-67	GUUUCGUGACACGUCUGCCCUUGUUCAGCCCGGUUGUGAUGGAGUUGGUCACAUCACAACCUGCAUGAAUGAGGACUGACGUCACGAGCG	90	28,88889	20	25,55556	25,55556	54,44444	45,55556	1,277778	1,130435	-43,9	-40,8	0,079095	4,8	-48,7778	-0,89592
lny-mir-8	UGCGCGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUGUCAGCAUUUUCUAAUACUGUCAGGUAAAGAUGUCCACAGAGUCUGCUGG	94	24,46809	23,40426	21,2766	30,85106	45,74468	54,25532	1,318182	1,15	-46	-46	0,192059	3,37	-48,9362	-1,06977
lny-mir-6544	AAGUUGUAUUUCUUUUCGCAAACGCCUUGCUUGGUGAGGCUGACCGAAACACUAAGCACGUGUAGAUAUAUGCGGcguauuaaaagaaaaacaauug	97	19,58763	19,58763	15,46392	22,68041	35,05155	42,26804	1,157895	1,266667	-29,2	-29,2	0,173343	6,85	-30,1031	-0,85882
lny-mir-359	CGGAAAGAGGGAAGCCAGAUUUUCAUCUGUGUUCCCCCGUCACAAAAUGGAUGGUUCCAGGUGAACGUCAGAACUUGAUCACUGGUUUUCCUCUGUUGCG	100	26	23	23	28	49	51	1,217391	1,130435	-38,9	-38,3	0,123112	6,16	-38,9	-0,79388

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-184	UUUCACAUUUCCUUAUCACUCCCCCGCCCCGUCUGUUCUCAAGCACUGGACGGAGAACUGAUAAGGGCUUGUGGGU	76	22,36842	18,42105	30,26316	28,94737	52,63158	47,36842	1,571429	0,73913	-24,6	-24,3	0,091486	9,76	-32,3684	-0,615
lny-mir-4931	GGCCCAGCUGUCUUCAAACAGUCGUGAUACAUUUCUAACGUUUGCUCAUCGCUGAUUGGUUGAGCAGCCGGAAG	74	25,67568	21,62162	24,32432	28,37838	50	50	1,3125	1,055556	-21,3	-21,3	0,196775	4,36	-28,7838	-0,57568
lny-mir-750	GAAGCCAGUUCCCUGUGAGUUGGAAGAUUGGGUCUUUGGCAGCAAUGCUGUCUUCCCUGCCAGAUCUAACUCUUCCAGCUCAUAGUGGCAUGGCACA	97	25,7732	20,61856	25,7732	27,83505	51,54639	48,45361	1,35	1	-48,9	-47,3	0,207948	5,04	-50,4124	-0,978
lny-mir-1175	UGAUCAGGUGUGUUAUGUAGUGGAGAGAGUUUUAUCUCAUCAUGAGCCGUAAGCAGGUGAGAUUCAACUCCUCCAACUGCAGGCUACACCUGAAAC	96	25	27,08333	20,83333	27,08333	45,83333	54,16667	1	1,2	-44,8	-40,7	0,088251	5,34	-46,6667	-1,01818
lny-mir-137b	CUACCAGUCGGUUACGGGUAUUCUUGGGUAAAUAAUACAUUGAACCGCUGUUAUUGCUUGAGAAUACACGUAACUGACUGGGUG	84	25	26,19048	17,85714	30,95238	42,85714	57,14286	1,181818	1,4	-40	-39,2	0,16289	4,49	-47,619	-1,11111
lny-mir-2580	uguccguaAGUGUACucacgugucuguguguauguuucuaCAUAGACAUAUACAUGUGUACGGCUUCGGUUA	72	12,5	15,27778	9,722222	16,66667	22,22222	31,94444	1,090909	1,285714	-27,3	-27,2	0,115281	4,2	-37,9167	-1,70625
lny-mir-11976	gggcggcggcggcggcgccGGCGUGGGNNNNNNNNNNNNNNNNNNNNNGGGgggggcggcggcggcggcgccGGCGUGG	79	17,72152	0	2,531646	2,531646	20,25316	2,531646	0	7	-38,07	-38,07	0,922324	0,51	-48,1899	-2,37938

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-9-2	CGGUUUUGUCUUUGGUUAUCUAGCUGUAUGAUUGAAGUAGAAAUAUCAUAAAGCUAGGUUACCAAAGGCAAAAAUG	76	22,36842	32,89474	11,84211	32,89474	34,21053	65,78947	1	1,888889	-33,1	-33,1	0,29717	2,88	-43,5526	-1,27308
lny-mir-7290	ACUCUGCAAUCAGAGAUUUCUGGCUCUGUUUGCAGCAAAUCACUAGUCGUUGGCUCUGUUUGCAGCAAAUCACUAGUCGUUGGCUCUGUUUGCAGCAA	98	22,44898	21,42857	23,46939	32,65306	45,91837	54,08163	1,52381	0,956522	-31,6	-28,2	0,065716	6,74	-32,2449	-0,70222
lny-mir-4265	AGCUUCUGAGGAGACCUGUGGGCUCAGCUCUGCAUUUAUAUUAGGAGGCAAAUGUCCCUCAGAAAUA	67	25,37313	26,86567	20,89552	26,86567	46,26866	53,73134	1	1,214286	-18,8	-11,8	0,102899	12,25	-28,0597	-0,60645
lny-mir-9701a-2	CGCCAUAGUAACCACUAGCCGUGAGACACACAACAGGCCGUGUGACUGUGUACCACCAUCAAGGCGUGACGUGUGACCCCAaggugugugacuguguac	99	20,20202	22,22222	26,26263	13,13131	46,46465	35,35354	0,590909	0,769231	-31,4	-31,3	0,319797	5,61	-31,7172	-0,68261
lny-mir-451	GACAGUACCCAGCCCAAAGAACAAACUUUCGUUUGUAGUAAUGGUAAGGGUUUGAAUCUUAGGGUGGGGUGCUAAG	76	28,94737	28,94737	15,78947	26,31579	44,73684	55,26316	0,909091	1,833333	-29	-29	0,334254	2,61	-38,1579	-0,85294
lny-mir-33	GCUAUGGUGGUGCAUUGAGGUUGCAUUGCAUCAGUUGAGGAACAUGCAAUGCAUCUGCAGUGCAAGUACCAUGGU	75	30,66667	24	17,33333	28	48	52	1,166667	1,769231	-38,2	-37,2	0,235986	4,29	-50,9333	-1,06111
lny-mir-653	AAGCUAGGAAAGCCGAGCUGCAGUAGAGAUUGAGACAAUCUCUACUGCAACAACGGGUUCCUAGGUC	67	26,86567	31,34328	22,38806	19,40299	49,25373	50,74627	0,619048	1,2	-35	-33,2	0,416654	2,33	-52,2388	-1,06061

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-8485-2	CAUUGCAAGCAUGGCGCCUGUGUGGCUCUGCGCGCAugcaacgcacacacacacacacacguacaugcacgcacuc	76	15,78947	6,578947	14,47368	10,52632	30,26316	17,10526	1,6	1,090909	-25,8	-25,8	0,527609	3,34	-33,9474	-1,12174
lny-mir-11225	AACUACAGGGCAGUGGACUACAGCAGAUUGUACGUAAAACUGUACGUAAAACUGUUCUUAAGUCAUGGCCUGUACGA	77	23,37662	32,46753	19,48052	24,67532	42,85714	57,14286	0,76	1,2	-23,7	-23,7	0,093776	6,64	-30,7792	-0,71818
lny-mir-466i-2	CACCUCCUGGUUCGCGAAACAUGGCCAUcuauaaauauuauaaauauuauauauacacacacacacacacacaugaccaCUUUCAGCGAAACGGGAGAUG	100	13	12	14	10	27	22	0,833333	0,928571	-20,7	-18,07	0,038561	14,52	-20,7	-0,76667
lny-mir-7195	uauccuccccucucccccccccuccaccacacgAGUCAUACGAUUGACGUCAUGGGAAGUGGGGGAGCGGAAAAGGACAGGUGA	84	25	19,04762	7,142857	9,52381	32,14286	28,57143	0,5	3,5	-29,8	-23,2	0,107685	18,49	-35,4762	-1,1037
lny-mir-29b-1	CAUUGGGGCAGCGGGGCACUGGUCUCGACUGGUGGGUAGAUGGCGCUGUUAGUCUAGCACCAUUUGAAAUCAGUGCUCGCGCAUGUCUCCAGCA	94	32,97872	18,08511	24,46809	24,46809	57,44681	42,55319	1,352941	1,347826	-44,5	-44,5	0,069593	6,31	-47,3404	-0,82407
lny-mir-29a	AGAUGGCGAGAUGGAUGCUGGUCUCUUUUGGUGCUUAGAGUCAUAUCGCCUGUCUAGCACCAUUUGAAAUCAGUUUCCUGACCUGGCCACAG	92	26,08696	20,65217	22,82609	30,43478	48,91304	51,08696	1,473684	1,142857	-34	-34	0,158187	4,7	-36,9565	-0,75556
lny-mir-734	CAGGUGGAACUAUUCUGCAACAUUCAUGCUGAACCUGAGCCAGCCAUUAUGUGGUCAACACCUUGGGGCCAUGAGUUUGCUGACAUUUCGCACAUG	96	23,95833	23,95833	25	27,08333	48,95833	51,04167	1,130435	0,958333	-23,7	-22,5	0,102	11,88	-24,6875	-0,50426

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-9235b	ACGGAGAUAGUUCUAAUGGGACUUGUCUUCUCCGACAAAGGCAUGUUUGAGAAAUUAAUUUUGAUGUGGCUUGAGAACAGUACCAUAAAUUGUCUCCCC	99	22,22222	28,28283	18,18182	31,31313	40,40404	59,59596	1,107143	1,222222	-26,8	-24,1	0,009236	11,43	-27,0707	-0,67
lny-mir-263a-2	CUUAGUUCCCCUGCUAAUCAAUGGCACUGGUAGAAUUCACGGGUGCUUUCAAAAUCCCGUGGUCUUUCAGUGCCAUCUAUGGGCAAAGGGUGACCUGCA	99	24,24242	22,22222	25,25253	28,28283	49,49495	50,50505	1,272727	0,96	-40,8	-39,7	0,053135	7,61	-41,2121	-0,83265
lny-mir-1c	ACACGGUACAUACUUCUUUACAUUCCAUAGCUUUGGAAGAACAUAUGGGAUAGCAAAGAAGUAUGUGCCGGGC	73	23,28767	31,50685	19,17808	26,0274	42,46575	57,53425	0,826087	1,214286	-33,4	-32,3	0,415163	2,78	-45,7534	-1,07742
lny-mir-1990	GAUGGGCUGUUCGUGAAGUAAGUUGAUGGGGUCCCAGGUAGAUCUACCCCACCCGGGACUACGUCAACGUACUACCACGCGCAGACCCCAA	91	27,47253	24,17582	29,67033	18,68132	57,14286	42,85714	0,772727	0,925926	-42,4	-41,8	0,25783	4,43	-46,5934	-0,81538
lny-mir-466q-2	caugugcacacacacacauacaugcuugCACAUGCAGCCUCACGUGCAAGCUUAAGUUUauagugcacaug	71	8,450704	11,26761	12,67606	11,26761	21,12676	22,53521	1	0,666667	-24,9	-24,7	0,237339	3,05	-35,0704	-1,66
lny-mir-1947	AAAUGCUCAGAUAAGCUGUCAGGCGAAUCAAGGAUUUUGAAAACAGUUAACUAGCUGCUUUGUUUUAGCACUGAGCUAGCUCUCUAGCAGUC	92	21,73913	29,34783	19,56522	29,34783	41,30435	58,69565	1	1,111111	-22,3	-21,5	0,091223	6,88	-24,2391	-0,58684
lny-mir-2491	UCCUAUUGGCAUGUAGUCAAAACUGCUCAGUuccuaacaacaacagcagcagcagugaccUCACACCAAUAUAG	74	9,459459	18,91892	14,86486	17,56757	24,32432	36,48649	0,928571	0,636364	-18,7	-17,1	0,17106	10,72	-25,2703	-1,03889

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-1985	GAGGAGAAGGCGUGCCAUGCCAUUUUUAUCAGUCACUGUGUGUUGUCAGUCACAGUGAUGGUGACAAUGGCUUGAUGGGCCUCUCAUG	88	30,68182	20,45455	19,31818	29,54545	50	50	1,444444	1,588235	-38,3	-38,3	0,063273	5,09	-43,5227	-0,87045
lny-mir-1307	AGAUCGCGACUUCGCCACUCGGCGUGGCGUCGGUGCAAAGGUCGAAAUGCGUUCGGGGCGGCGGAGUGGGGAGGACACGAGCC	83	42,16867	18,07229	25,3012	14,45783	67,46988	32,53012	0,8	1,666667	-34,9	-33,3	0,314815	4,28	-42,0482	-0,62321
lny-mir-2944a	GUACAGGAGACAUUUCUCUGUGAAAAAGAGUAUAGGUGAUAUUUUCUCUUGCUGUCAUUUAGGGGGGCGAAGAUUUUCAGAAGGAACUUCUGCUGAUC	98	26,53061	26,53061	14,28571	32,65306	40,81633	59,18367	1,230769	1,857143	-22,4	-11,8	0,008713	30,59	-22,8571	-0,56
lny-mir-5397	agaaaacaaacuacgCAGAAGUUUGCUUCUGuaucauuuucccucccucgGAAUGUUUGCACAUGCGCAGCAAACGUCUCUGACAUUUAUUUCAG	95	13,68421	15,78947	13,68421	21,05263	27,36842	36,84211	1,333333	1	-20,9	-20,9	0,2969	5,73	-22	-0,80385
lny-mir-7045	acucucccccucccccgccccacuuUUCUUCUGCGAGGGACGCCGAUCAGACGACGCCCGGAGAAAGGUggaggcugggugggugg aggga	92	16,30435	10,86957	13,04348	7,608696	29,34783	18,47826	0,7	1,25	-50,9	-50,9	0,357037	6,43	-55,3261	-1,88519
lny-mir-106a	UUUUAUUAUCAGCCCUGCUACUGCAAUGCAAGCACAUUGACUGAAUUUUGCUCAGAGAACGUGCAGUUGGAGCUUGCUUUUCAUUGGCUGAUAAUAGCG	99	22,22222	24,24242	20,20202	33,33333	42,42424	57,57576	1,375	1,1	-30,3	-28,6	0,089339	10,78	-30,6061	-0,72143
lny-mir-3238	AGUGACUGAAAUGAAUCAUGCAUUUGGCAUGUAAACCUGUGGUGAAAUAGUGUguaacauugguguuug	98	18,36735	24,4898	9,183673	27,55102	27,55102	52,04082	1,125	2	-18,5	-18,5	0,036449	18	-18,8776	-0,68519

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	uguaAAUAUUCUGAUAAUUUCUAGUCGUG															
lny-mir-2361	CAUGCUgaguuguguuguuuuuuuuucaauacagcAGAUGAACUAACAAUACUGCAUCCAAAGUGCUAGCAUUAACAAGAAAUAUCAUAGCUUCAGCACA	100	10	29	16	16	26	45	0,551724	0,625	-23	-22,9	0,046036	12,62	-23	-0,88462
lny-mir-2493	AGCCUCUAUCACUCCUCUGAUCCUGUCGUCAGUGUAGUAAGAAAUUUUGGCGAAAGUAgcugcucacacacacacacagacgagagagagagagagaga	99	12,12121	15,15152	13,13131	18,18182	25,25253	33,33333	1,2	0,923077	-26,4	-18,6	0,022709	25,98	-26,6667	-1,056
lny-mir-9032	GUGAUGUUUGUUGCCAGUGAAUUAAUCAAAUUCUGCCAAUAUGCACUGCAGAUUCCGUUGAUGCAAUCCAAUGGUCAUCAUCAUGAG	87	20,68966	27,58621	19,54023	32,18391	40,22989	59,77011	1,166667	1,058824	-19	-19	0,117129	13,25	-21,8391	-0,54286
lny-mir-3963	GUACACACAGUCAAAUAAAGUAUUGUAUCCCACUUCUGACGCACAAUAUUUGUCGUUUGCCACAGAAAAAAUACCUGGGUUGACGGUGUGCAU	93	19,35484	31,1828	21,50538	27,95699	40,86022	59,13978	0,896552	0,9	-21,7	-21	0,161914	20,42	-23,3333	-0,57105
lny-mir-1994	ACGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUGUGCAAUGACGUCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGAGACCGCUAA	90	28,88889	22,22222	25,55556	23,33333	54,44444	45,55556	1,05	1,130435	-38,4	-38,4	0,344812	5,28	-42,6667	-0,78367
lny-mir-51	CUCCCAUCAUGUAUGCGUGCAUGGAAGCAGGUACAGUCACCUGUCCUGACUUAUCCAGCGAACAGUGUGGCCU	73	24,65753	21,91781	28,76712	24,65753	53,42466	46,57534	1,125	0,857143	-21,7	-17,1	0,160663	16,95	-29,726	-0,55641

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
lny-mir-9229d	UAGGGAUAAGGGGGAGGUUAAGAUAAGGAAAGGAUACUGUUAUCUGAGUCUUCCCCUUGAGCCUGU	66	31,81818	27,27273	13,63636	27,27273	45,45455	54,54545	1	2,333333	-25,9	-25,4	0,059825	5,61	-39,2424	-0,86333
lny-mir-4968	GCUCUUCUgcuggguuuguuguuguuguuguuguuaguaaCGAAGCUGUAAUAGUUCCCCGUGGCAGCAGcaauaacagcagcaacagcagcagcagcag	100	10	7	11	10	21	17	1,428571	0,909091	-45,8	-45,8	0,189807	22,44	-45,8	-2,18095
lny-mir-239b	CUCagccaucccccucccccccagaguGGGUCAGAUGGCACUUUUGUGGUGUGCAGGUUGGAUGGCUUGU	69	26,08696	7,246377	10,14493	23,18841	36,23188	30,43478	3,2	2,571429	-25,2	-13,05	0,115483	18,21	-36,5217	-1,008
lny-mir-466i-3	ACUCGCUCAAACACUGUCACUCACUCAAACACAUGGACGCAUUCGCgugcauucugugugugugugugugugugguguguguggcgugc	88	6,818182	15,90909	19,31818	10,22727	26,13636	26,13636	0,642857	0,352941	-35,9	-35,4	0,211217	10,98	-40,7955	-1,56087
lny-mir-277a	UGUUCUUCGGGCAAGGAAACGCGAUCACACGGAgccauuucuuugugugcuuccUUGGCCGCCAAGAUCG	70	21,42857	17,14286	18,57143	12,85714	40	30	0,75	1,153846	-25,5	-25,4	0,03184	11,31	-36,4286	-0,91071
lny-mir-29b-2	UGUAAAGUGAUGUAUCGUAUCCUUUGGGAACGCCAGCUGGUUUCACAUGGACAGAUAACAACACUUUGUC	70	22,85714	27,14286	20	30	42,85714	57,14286	1,105263	1,142857	-19,1	-16,4	0,131874	6,17	-27,2857	-0,63667
lny-mir-466i-4	AUUACACGCUCUCGACCACACCCGGACCcucaaaccaacaaaaaacguUGGugagggaagugugugugugugugugugugug	82	7,317073	8,536585	15,85366	6,097561	23,17073	14,63415	0,714286	0,461538	-32,4	-31,1	0,058041	17,93	-39,5122	-1,70526

Supplementary Table 2 – Structural and thermodynamic characterization of miRNA precursors in *M. cornuarietis*. Size (Tam), G content, A content, C content, U content, GC content, AU content, AU ratio (R-AU), GC ratio (R-GC), Minimum Free Energy (MFE), Minimum Free Energy of the Pool (MFEE), Frequency of the MFE Structure in the Pool (Freq), Diversity of the Pool (Div), Minimum Adjusted Free Energy (AMFE), Minimum Free Energy Index (MFEI).

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-bantam	UUCGACGAAACUGGUUUUCACAGUGAUCCAGCAGAUUGCUCAAAGUCUGAGAUCAUUGUAAAAACCAAUUUUGUCCCC	78	17,94872	29,48718	23,07692	29,48718	41,02564	58,97436	1	0,777778	-33,4	-33,4	0,687059	0,82	-42,8205	-1,04375
mcr-mir-1002	UcguacaccaacaccugucaccucuUGUAGUCUGGUAUCUUUGUCUAAGUAGUUGAUACAUGAGUUGCAGAGGGUAUGGAGUAGUA	86	22,09302	17,44186	5,813953	26,74419	27,90698	44,18605	1,533333	3,8	-20,5	-18,9	0,106705	9,05	-23,8372	-0,85417
mcr-mir-10080	UCUGUCUCGUCUUUUCUCAGGUUGUCUCAAUCUUGCUUCAGUUUUGUCUGAAGACGUACAGACCagugagacaaaaagaaaccGU	85	15,29412	11,76471	18,82353	31,76471	34,11765	43,52941	2,7	0,8125	-19,1	-13,9	0,073241	20,99	-22,4706	-0,65862
mcr-mir-10089	UAUCAGCUCCCGAAGCAAAAUCAUGCCAAACAUAAUACAGGACGGUACUCGUGCAGGGAGAUAAUUUGCUGUUUUGGGAUGCUGCUG	87	25,28736	28,73563	20,68966	25,28736	45,97701	54,02299	0,88	1,222222	-22,8	-16,84	0,043074	18,63	-26,2069	-0,57
mcr-mir-10141	agcccGCAGCCUUGUGCAGUACagaggaaauaaaagaagaaacacgCAUUCGGUCUUCUUCAGUCAGCAUAUGGCUCGGAAA	82	15,85366	13,41463	17,07317	18,29268	32,92683	31,70732	1,363636	0,928571	-23,7	-22,7	0,301246	7,68	-28,9024	-0,87778
mcr-mir-10228	CAUUAGUACUGUGAUAUGUCAGUACACACGACAAUGAGUAUUGUACCAUGUCAGCACAUUAUUAGUC	94	19,14894	28,7234	18,08511	34,04255	37,23404	62,76596	1,185185	1,058824	-27,3	-27,3	0,173696	5,07	-29,0426	-0,78

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUGUACUGUGUGUCAACAUUAGUACUG															
mcr-mir-10237	UCCAUUGUUAGUUUUGAGUGAGCUAGAAAGGGGUUAAAACUUCUUGCAGCAGGGACUGGUUUGUGGCACAUAUUUCAGCCUCucgacuaauaauaau	97	22,68041	19,58763	14,43299	27,83505	37,1134	47,42268	1,421053	1,571429	-23,8	-23,8	0,115043	18,95	-24,5361	-0,66111
mcr-mir-10325	AAAAUUAUCAUCGUCUUCUAUACAAGUGGCAUUCUCCUCGCCAUUAUUAUCAGCAGCAAAGGCGUUUUGCUAACUUGUCAAACAUGUGAUAAUAUA	96	14,58333	31,25	20,83333	33,33333	35,41667	64,58333	1,066667	0,7	-18,7	-18,7	0,080867	8,79	-19,4792	-0,55
mcr-mir-10379	AGUAACACAGUUAUUUUUCAAACAACUCAAACCAGGAAUAACAGACUGUUUUGUUCGUUGAACUAUGAUUGUGUUGCA	78	16,66667	33,33333	16,66667	33,33333	33,33333	66,66667	1	1	-24,4	-24,4	0,273807	6,43	-31,2821	-0,93846
mcr-mir-10456	UUUCUGUCAUACUAGAUGCAGGAUAUUUCUGUCAUACUAGAUGCAGGAUAUUUAUGACAUACUAGAUGCAGGAUAUUUCUGUCAUACUAGAUGCAGGAU	99	20,20202	29,29293	15,15152	35,35354	35,35354	64,64646	1,206897	1,333333	-32,6	-32,6	0,295291	5,75	-32,9293	-0,93143
mcr-mir-10457	AAGCAAAGAGUUUUUGACUACCAaucaaaauaguauuuuaacAAGUUUGCaauuauucuuuguuuguuguguucuACACGACUCUUUGGCA	92	9,782609	16,30435	10,86957	14,13043	20,65217	30,43478	0,866667	0,9	-19,7	-18,9	0,188376	10,01	-21,413	-1,03684
mcr-mir-10698	uuuuaggaGAGGAAUUGCUGAGUUCAUGCAUGGCUUCUUUAGUAA	87	18,3908	22,98851	17,24138	32,18391	35,63218	55,17241	1,4	1,066667	-20,4	-20,4	0,075656	7,96	-23,4483	-0,65806

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CGAGGUUCAUAUUCAUGAUAAGCUGACAUUCAUCUUCUAUCC															
mcr-mir-106b	CAGAGCGGGUAUCCUCUGUGGGUACCCCUUGUCACUGGCAGGACCGCACUGUGGGUACCCCUGUC	65	30,76923	13,84615	32,30769	23,07692	63,07692	36,92308	1,666667	0,952381	-27	-22,5	0,153722	17,97	-41,5385	-0,65854
mcr-mir-10717	GCGACACGGAUGCACACACACCUGCGCGCGCCAAGAGCAGUGGAGGUGAAAUAUUGUUUCGGUUAACAAAGGGCACAGCUCAGUGUGUUGCUGUGUGUU	99	31,31313	24,24242	22,22222	22,22222	53,53535	46,46465	0,916667	1,409091	-36,3	-35	0,209813	4,21	-36,6667	-0,68491
mcr-mir-10738	GUCCCAGUAACUGACCACUCUGACGUCUCUCCGCCCUUCCGGGUGCUGCUGUCUGCUUUGUAACUGAUGACGUCAGUGGGUCACGUGUAGCUGGUGU	97	27,83505	13,40206	28,86598	29,89691	56,70103	43,29897	2,230769	0,964286	-36,5	-35,1	0,181441	6,85	-37,6289	-0,66364
mcr-mir-10761	CUGGUCUGUGCAGACUACUAGUACGGUGACGAUGGUUGAAUUUGUUCUUGUCACCAUUGUGACAGAUUCUGAGCGCACAGUACUAC	86	25,5814	22,09302	20,93023	31,39535	46,51163	53,48837	1,421053	1,222222	-24,9	-19,7	0,050962	23,36	-28,9535	-0,6225
mcr-mir-10863	UCGAUAUACAACCCUACGAACAUCCCCAUGACCAAGUGAUAUCACCUUUGACUUCUUUGUGGGCACGAUGAUGCUUGUAAGAUUGUUUAUAUG	93	18,27957	26,88172	22,58065	32,25806	40,86022	59,13978	1,2	0,809524	-19,7	-17,9	0,1266	11,6	-21,1828	-0,51842
mcr-mir-10903-1	CUGCCACUUGAACGCUCAACAGCUCAUCUUCCGACUCGCUUGUGUCGGCCUCAGACCCACUGGCCUCA	95	24,21053	12,63158	35,78947	27,36842	60	40	2,166667	0,676471	-34,4	-34,4	0,328396	6,79	-36,2105	-0,60351

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CUGCUGUCUGCUGCGUUCCUGUGGUGG															
mcr-mir-10903-2	CUGCCACUUGAACGCCCAACAGCUCAUCUUCCGACUCGCUUGUGUCGGCCUCAGACCCACUGGCCUCACUGCUGUCUGCUGCGUUCCUGUGGUGG	95	24,21053	12,63158	36,84211	26,31579	61,05263	38,94737	2,083333	0,657143	-33,6	-33,6	0,309438	7,34	-35,3684	-0,57931
mcr-mir-10903-3	CUGCCACUUGAACGCCCAACAGCUCAUCUUCCGACUCGCUUGUGUCGGCCUCAGACCCACUAGCCUCACUGCUGUCUGCUGCGUUCCUGUGGUGG	95	23,15789	13,68421	36,84211	26,31579	60	40	1,923077	0,628571	-30,3	-30,2	0,116672	10,54	-31,8947	-0,53158
mcr-mir-10918	GACAGGACAGAGAUUAAGGAguguuaaaauaguuugaagUAUUGGAAACUGGGUGUUCACAUAUAAGCUAUGUUACUCGAUAAUGCUCUUCCUUGC	96	18,75	23,95833	13,54167	23,95833	32,29167	47,91667	1	1,384615	-23,2	-20,8	0,051911	9,44	-24,1667	-0,74839
mcr-mir-10967	AACUGUACACUGCAUACUGUACAAUGUAGCUUGUCGGAUGAACUGUGAACUGCAUACUGUACAAUGUAGCUUGUCGGAUGAACUGUACACUG	92	22,82609	28,26087	19,56522	29,34783	42,3913	57,6087	1,038462	1,166667	-21,6	-20,2	0,048588	15,84	-23,4783	-0,55385
mcr-mir-10a	CGGUGCAAGACCUACCCUGUAGAUCCGaauuuguguaauauuuuaacUACAAAUUCGUGUUUGCACGGUAAGGCCUCGCAUCU	83	18,07229	16,86747	21,68675	19,27711	39,75904	36,14458	1,142857	0,833333	-33	-29,9	0,107106	4,44	-39,759	-1
mcr-mir-10b	AUGAGAGUGUGAGUGGCUUUACCCUGGAGAACCGAGCGUGUGUGAUCAUGACGUCACAGUUCGUUU	93	31,1828	20,43011	19,35484	29,03226	50,53763	49,46237	1,421053	1,611111	-40,6	-40,6	0,254812	9,91	-43,6559	-0,86383

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CUGAGGGUCAAGUCGCGAUACUCUGUU															
mcr-mir-11	GACCUGGGAGAGCCGUGGCCAGCUGGAGACAUCCCUGGAGCUCUGGCUGUGCCGGGUGGAUUUCGUGCCAUGCAGUCAGCCUCUCCAGAUG	91	35,16484	15,38462	28,57143	20,87912	63,73626	36,26374	1,357143	1,230769	-35,2	-21,3	0,010022	23,33	-38,6813	-0,6069
mcr-mir-11591	uuuucGGGUACGUGAAACACCUUUACGAUUCGUUAUCCUUAGGCUGUGCUGUAAGUGUACUUUGAUAACGGUUCGGCAACAGGUUGAUUGUACCCGACAU	100	24	22	19	30	43	52	1,363636	1,263158	-33	-31	0,123962	9,74	-33	-0,76744
mcr-mir-1175	UGAUCAGGUGUGUUAUGUAGUGGAGAGAGUUUUAUCUCAUCAUGAGCCUCAGAAGCAGGUGAGAUUCAACUCCUCCAACUGCAGGCUAUACCUGAAAC	98	24,4898	27,55102	20,40816	27,55102	44,89796	55,10204	1	1,2	-42,7	-38,6	0,079856	5,62	-43,5714	-0,97045
mcr-mir-12	UGUUUCUACAUAUCUGUGGAAUCUUGCAGCAUCAUGUGCAUGUUGGUGGUGGUCAGACUGUGAGUAUUACAUCAGGUACUGAGAAUCU	88	26,13636	22,72727	15,90909	35,22727	42,04545	57,95455	1,55	1,642857	-21,2	-14,6	0,061401	34,13	-24,0909	-0,57297
mcr-mir-12037	UGGUAACUCAUUCUGUCUUCUCUUCCAAAAACCAGCAAAUGUGAAAUUGUGUAUGAGCAGUGGGUAUAUCUUUUUUGGUGGGACAAAUGAGUUAAAA	97	21,64948	29,89691	14,43299	34,02062	36,08247	63,91753	1,137931	1,5	-22,2	-21,7	0,076697	11,37	-22,8866	-0,63429
mcr-mir-12093	UACGGUCAAUAGCUAUCCCCAGGGUaauguuauuuuugcuuuauugCUU	81	14,81481	13,58025	23,45679	22,22222	38,2716	35,80247	1,636364	0,631579	-26,4	-26,4	0,139195	7,63	-32,5926	-0,85161

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CUAACUUCAGCCCCUGGGGUAGCCUUACCUUU															
mcr-mir-12096b	UGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAGGUAGGGCAAAAUUAUCACCGAUUACCCGAGGUAACG	85	23,52941	27,05882	21,17647	28,23529	44,70588	55,29412	1,043478	1,111111	-29,6	-29,2	0,079903	7,54	-34,8235	-0,77895
mcr-mir-12193	UUUAAAGAAUUCAUCUUUAGAUCAUAACUGUUUAAGCAGUACAGACCAGUUUUAAACCGGUCCUUUCUGUCUAAUGUUCUGGAAAAGAUCUUUCUUUGUC	100	15	28	18	39	33	67	1,392857	0,833333	-20	-18,8	0,080489	17,54	-20	-0,60606
mcr-mir-12265	AGAUUCAGCCGUCUUGCAGCUGAAGCCAUACCACGUGAGACUUAGACUGAGGACACGCUGAAGCA	65	26,15385	29,23077	26,15385	18,46154	52,30769	47,69231	0,631579	1	-20	-17,1	0,213126	4,96	-30,7692	-0,58824
mcr-mir-12293	gcugccgUCUCCCUUCUACAUCCGCCUCUUGAUCUACUACAACUACGAGCACGAGGAGAUGUUACAGAGGAGAUCGGCCAU	81	18,51852	23,45679	27,16049	22,22222	45,67901	45,67901	0,947368	0,681818	-27,4	-27,4	0,349688	3,61	-33,8272	-0,74054
mcr-mir-12313	uaggucacgugacauguuAGCAUCUUGACAUAAAGGAAGUCAGUGUGUCUGUAAUGUGUAACAUGUCUCAUUACGAA	77	16,88312	24,67532	11,68831	23,37662	28,57143	48,05195	0,947368	1,444444	-19,1	-18,7	0,051013	5,78	-24,8052	-0,86818
mcr-mir-12322	agaagaaaagagaaguuGAUCAAACAGCCAACUGGGUUAUCCCGUGUGUUGGGACUCGCCUGUUCUCuggucauuucucuuucucac	87	14,94253	10,34483	16,09195	16,09195	31,03448	26,43678	1,555556	0,928571	-27,7	-27,5	0,020633	17,77	-31,8391	-1,02593

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-12352	UUAUUGCUGUUAUGGUGUUAGUUCAGGUGAUUCUUAUGCAUGAGAAGUUAUGAGUUAACACCAUUACCACAacuu	75	20	25,33333	13,33333	36	33,33333	61,33333	1,421053	1,5	-21,8	-21,8	0,14281	5,72	-29,0667	-0,872
mcr-mir-1250	gaugacgacgguGCUGGAUGUGGAUAUGGAAUACGGACAGCUCCGUUUCUGAACAACAUCAGAACCUGUAGGUCCCA	77	22,07792	23,37662	19,48052	19,48052	41,55844	42,85714	0,833333	1,133333	-22,1	-19,8	0,186986	22,97	-28,7013	-0,69062
mcr-mir-1256	cucuggccacugaGGCAUUGACUUCUCUUCAUGGCAUCUCAAAGGACAAGUCAAGUCAUGUGGUCAAAC	69	17,3913	23,18841	18,84058	21,73913	36,23188	44,92754	0,9375	0,923077	-22,5	-22,5	0,137838	3,93	-32,6087	-0,9
mcr-mir-1277	CACGUGCAGAUGUAUUGACAguuucuguguauauauauauauauauguacgauGCAACAGCACAUCUGCACUAC	74	10,81081	17,56757	16,21622	10,81081	27,02703	28,37838	0,615385	0,666667	-24,9	-21,6	0,120471	14,07	-33,6486	-1,245
mcr-mir-1278	AUUAAUCAGGAGCAGGUUUAUUGCAUGAGGUGUAGUAUUGACUGUAGUACUGUGCAUAUCAGCACCACAAAUCAUGCAGGAUGAAGACUGUUCGauugug	100	24	28	15	27	39	55	0,964286	1,6	-29,4	-28,6	0,050476	16,75	-29,4	-0,75385
mcr-mir-1304	CUUCAGGUCGUCACGGAAUGAAUUCUCACUGUAGCCUCGGCUCAGCGUGAUUUCAAACACGUCACAGCCUGCUG	74	22,97297	21,62162	29,72973	25,67568	52,7027	47,2973	1,1875	0,772727	-24,6	-22,7	0,479157	2,33	-33,2432	-0,63077
mcr-mir-130a	AUCCCUCCCUGCUCUGUUCCUUGUAUCUGCUCAUAGUGCAAUGUUAAAAGAGUACCUUUUGAGUUCUCUUGAGGUGACAAUGGAAAAAGCAGGAGGGCU	99	23,23232	24,24242	21,21212	31,31313	44,44444	55,55556	1,291667	1,095238	-31	-29,3	0,028824	19,37	-31,3131	-0,70455

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-133	CUUCUAUAGCUGGUUGAACUCGGGCCAAAUUGUUGAAGAGCCUUUCAUUUGGUCCCCUUCAACCAGCUGUAGUUA	75	21,33333	21,33333	24	33,33333	45,33333	54,66667	1,5625	0,888889	-35,5	-35,1	0,186674	6,73	-47,3333	-1,04412
mcr-mir-133c	UAACUACAGCUGGUUGAAGGGGACCAAAUGAAAGGCUCUUCAACAAUUUGGCCCGAGUUCAACCAGCUAUAGAAG	75	24	33,33333	21,33333	21,33333	45,33333	54,66667	0,64	1,125	-30,4	-28,9	0,184949	4,61	-40,5333	-0,89412
mcr-mir-137b	AGUUCCAGUCGGUUACGGGUAUUCUUGGGUAAAUAAUACAUUGAACAGUUGUUAUUGCUUGAGAAUACACGUAACUACCUGGACGU	86	23,25581	27,90698	16,27907	32,55814	39,53488	60,46512	1,166667	1,428571	-33,1	-33,1	0,033251	6,83	-38,4884	-0,97353
mcr-mir-14	UAUAAAGGGAAGACAAUGAUCUGAGAGGUGCUGGCUGUACGCUGCUGAACUGAUCUCACUCucagucuuuuucucucucuuuuuu	85	20	20	14,11765	17,64706	34,11765	37,64706	0,882353	1,416667	-21,6	-18,47	0,04849	12,49	-25,4118	-0,74483
mcr-mir-1421ai	UUGUAGUCAGCAGGUCAGGUGUGGAAGCACUUUGCCAGGUUUUCCCCAUGGGUCCUGCGACUAGUG	66	31,81818	16,66667	22,72727	28,78788	54,54545	45,45455	1,727273	1,4	-24,4	-23,1	0,219346	4,86	-36,9697	-0,67778
mcr-mir-143	AUGUGUGAGCAUGACAGGAAUGAAAACUGUGCAAACCUCUGUGUGUGCAGUGCUGCAUCUCACAUUA	67	25,37313	28,35821	19,40299	26,86567	44,77612	55,22388	0,947368	1,307692	-20,7	-20,5	0,448845	1,84	-30,8955	-0,69
mcr-mir-146b	UUUAGGAACACGUUCUCAGAUGGAAGUUUCAGACCCUGUAUUCUGGAGGUUGAGAACUGGUUUUGAGCAAGUGCUUCCUUUU	82	25,60976	21,95122	17,07317	35,36585	42,68293	57,31707	1,611111	1,5	-24,2	-24,2	0,193626	6,98	-29,5122	-0,69143

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-153-1	AACCAAAGCCUUAGCGUAGGUCAAGAAGAGUGUUAUAACCAAAUCAUCGUGCUGucauuuuugugauuuugagCUGACAGCUAGAUGCGUUGAAU	95	18,94737	26,31579	15,78947	18,94737	34,73684	45,26316	0,72	1,2	-21	-14,1	0,097127	11,84	-22,1053	-0,63636
mcr-mir-153-2	AUACCCUCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGCUAUCAAAUUGCAUAGUCACAAAAGUGAUCGGGAGGCGGGGAC	87	24,13793	27,58621	22,98851	25,28736	47,12644	52,87356	0,916667	1,05	-37,9	-37,9	0,256161	6,4	-43,5632	-0,92439
mcr-mir-1599	ACAUUACUCGCUCUCUGAGAAUcuggagggaggaaaaaaaaacgcuuAUUUUUUAAGCUCCAGAUGUUGACAGCAGGCGUAACCA	85	12,94118	20	16,47059	21,17647	29,41176	41,17647	1,058824	0,785714	-20,8	-16,6	0,027501	14,66	-24,4706	-0,832
mcr-mir-15a	aaauguuuauccuagcagcacagaauggcAAAAAUAUAGGGGUCCCUUUAAGAAGGAUAUUGGACGCCUGUGUUUUGUGCUGCUAGGGUAAGCAAUU	97	19,58763	19,58763	9,278351	21,64948	28,86598	41,23711	1,105263	2,111111	-47	-47	0,037871	8,59	-48,4536	-1,67857
mcr-mir-1610	ACGAAAAUAAUAUCAGCACUUUGCUGUGGCUUGUGGUGGAACGACAAACCAcuuucaacccccccccccccaauacaUUAAAGUGCCCAUAUAUGUUUCA	100	15	24	14	21	29	45	0,875	1,071429	-21,3	-17,3	0,147086	14,93	-21,3	-0,73448
mcr-mir-1621	GCGGUACGUGCGAGCGGGCUCUCAACCGGCUGCCUCGGUGGUAAGCGGUGUUGGAGGCUGCUGACCUACUGG	72	40,27778	12,5	26,38889	20,83333	66,66667	33,33333	1,666667	1,526316	-32,6	-32,1	0,106716	4,49	-45,2778	-0,67917

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-1628	AAUUAACUUGAAUUAACGCGAAGAAGAUGUCUUUACACUACUUUGUUUCCUUCCUUUUGUAAGAGCUCUUCCUGUUCGUGUGUGAGAAUCACGCUUACAA	100	17	26	20	37	37	63	1,423077	0,85	-19,3	-19,3	0,079518	10,73	-19,3	-0,52162
mcr-mir-1693	ACCUUUAAGUGGCUAUUGAUGUCACCAAGAAAUGGUGCAAAGGAUGAAGCUGUAGCUGUGUUUCCUAAUGGCUGCACAAUAUGUUAUUUAAAAGU	95	23,15789	30,52632	14,73684	31,57895	37,89474	62,10526	1,034483	1,571429	-25,5	-25,5	0,09906	5,04	-26,8421	-0,70833
mcr-mir-1710	GAUCUGUUUCUCAUCUGCUGCAUAACCGUGUGUGACACAGGACACAGCGGUAGCACACAGAUACAAAAGGGA	72	25	30,55556	23,61111	20,83333	48,61111	51,38889	0,681818	1,058824	-21,5	-21,5	0,221235	4,85	-29,8611	-0,61429
mcr-mir-1761	GACCAAGCAUAACAUAACGACGGAGGACAAUGGAGGACAGGGGUCACUUUUUUCCCCUGACCUUCACUUUUUCUUAUGUGAUCUUGUGG	89	23,59551	24,7191	22,47191	29,21348	46,06742	53,93258	1,181818	1,05	-28,5	-22,9	0,114468	11,78	-32,0225	-0,69512
mcr-mir-187	GAAAAUUAGAUGAUGUGCUGGUUGCUACAACACAGGACAAUUCUAAACUGAUUCGUAUUCUGCCUGUGCAAUUGCAAGCCCACAACAGAGAAUUGGU	97	21,64948	31,95876	19,58763	26,80412	41,23711	58,76289	0,83871	1,105263	-22,1	-19	0,108197	9,61	-22,7835	-0,5525
mcr-mir-190	ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGACCAAGUAGUCAAACAUGUCAGAGCAGCGACG	82	25,60976	25,60976	21,95122	26,82927	47,56098	52,43902	1,047619	1,166667	-32,5	-32,3	0,080048	9,84	-39,6341	-0,83333

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-193-1	UUCUAAUAUCAUGGUAUCAGCAUGACUCGUAUGUGAUAUACUGGCCAGCACAAUUAGUGUCUAAUAUCAUGGUAUCACCAUGACUAGUAUGU	92	18,47826	29,34783	18,47826	33,69565	36,95652	63,04348	1,148148	1	-20,8	-20,8	0,129928	10,37	-22,6087	-0,61176
mcr-mir-193-2	GCCAGCACACUUACUGUCUGGUAUCAUGGUAUCAGCAUGACUCGUAUGUGAUAUACUGGCCAGCACAAUUAGUGUCUAAU	80	21,25	26,25	22,5	30	43,75	56,25	1,142857	0,944444	-21,7	-21,7	0,239175	6,47	-27,125	-0,62
mcr-mir-193-3	CUAAUAUCAUGGUAUCAGCAUGACUAGUAUGUGAUAUACUGGCCAGCACAAUUAGUGUCUAAUAUCAUGGUAUCAA	76	18,42105	32,89474	17,10526	31,57895	35,52632	64,47368	0,96	1,076923	-19,8	-19,8	0,11962	11,25	-26,0526	-0,73333
mcr-mir-194	GUUUGGUCAUCUUCAGUUCCAUGCCAGCCCCACCUGGCAGCCAGUGGAGGUGGUGUAGaagguggucacgugaccagcc	78	23,07692	11,53846	20,51282	19,23077	43,58974	30,76923	1,666667	1,125	-32,8	-32,8	0,088541	4,32	-42,0513	-0,96471
mcr-mir-1947	AUCUGAAUGAAGGCUAGUGCACUGAGCUAGCUCUCUGCGUCAUCAACUGCAAUGAAGCAUAGAGGGAGGCAGCGGUGGAGUACAAUUCUUCAUAA	95	27,36842	29,47368	20	23,15789	47,36842	52,63158	0,785714	1,368421	-27,3	-11,2	0,050373	20,57	-28,7368	-0,60667
mcr-mir-194b	GUACUGCAUCAGCCACGCGCUGCGUUGUAACAGCAUCUCCAACUGCGGCGACCACGACGACAGCGACGAGCGCAAAUGUAGGUC	84	27,38095	25	32,14286	15,47619	59,52381	40,47619	0,619048	0,851852	-28,7	-27	0,178572	5,44	-34,1667	-0,574
mcr-mir-196c	GGUGUGUUGCUGACAACAACAUCAAACCACCUGCCGAACAUCUGG	81	24,69136	23,45679	29,62963	22,22222	54,32099	45,67901	0,947368	0,833333	-24,7	-24,7	0,061423	10,88	-30,4938	-0,56136

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCCAGGUGCUGGUGUUUCAUGCUCCAUCAACACGGG															
mcr-mir-1970	UUGUUAUACCCUGUGAUAAACUCUUUGGGUUGGGGGCUUAAAUAUCUUUCUUCUCAGAUUGUGUCACUGGGGAUAAUUA	79	22,78481	21,51899	15,18987	40,50633	37,97468	62,02532	1,882353	1,5	-22,3	-22,1	0,116729	7,91	-28,2278	-0,74333
mcr-mir-1984	UGUCGCCGUGCCCUAUCCGUCAGGAACUGUGAUCUCGCGAAACACAGGGUCUGGCGGUUGGGCCUCGGCGCUG	73	34,24658	13,69863	30,13699	21,91781	64,38356	35,61644	1,6	1,136364	-37,9	-37,9	0,133827	2,65	-51,9178	-0,80638
mcr-mir-1985	UGCCAUGCCAUUUUUAUCAGUCACUGUGUGUUGUUAAAGUcacagugaugaugauaauggcuUGAUG	67	14,92537	13,43284	10,44776	28,35821	25,37313	41,79104	2,111111	1,428571	-28,6	-28,6	0,286882	2,78	-42,6866	-1,68235
mcr-mir-1990	GGCUGCGAGUUGUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCUACCCCUCCCGGGACUACGUCAACGUACUACCACGCGCAUCUCCAAGU	95	27,36842	20	29,47368	23,15789	56,84211	43,15789	1,157895	0,928571	-39,9	-39,5	0,101035	5,08	-42	-0,73889
mcr-mir-1992	CUAAGGGCAUGUGUCUAGUCAGUGGUUGACUGCUUGGUAUUCUAAAUGUAUCAGCAGUUGUACCACUGAUUUGCUACGUGCACCUCAC	88	23,86364	21,59091	21,59091	32,95455	45,45455	54,54545	1,526316	1,105263	-41,8	-41,4	0,125422	3,52	-47,5	-1,045
mcr-mir-1994	UGGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUGUGUGAUGACGUCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGAGACCGCUAA	90	31,11111	20	23,33333	25,55556	54,44444	45,55556	1,277778	1,333333	-40,6	-40,6	0,538688	3,53	-45,1111	-0,82857

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-1994a	CAAAAGCUAUUUCUCAGGGGCGGUCACUCUGAUCUCCAUGUGCUCGCACCAGCAUGAGACAGUGUGUCCUCCCUUGAGGGAUGGGCUUAUA	91	26,37363	20,87912	26,37363	26,37363	52,74725	47,25275	1,263158	1	-44,3	-44,3	0,178398	5,12	-48,6813	-0,92292
mcr-mir-1c	AGUACAUACUUCUUUACAUUCCAUAGCUUUGCAAGAACAUAUGGGAUAGCAAAGAAGUAUGUGCC	65	18,46154	33,84615	18,46154	29,23077	36,92308	63,07692	0,863636	1	-27,1	-26	0,4687	2,23	-41,6923	-1,12917
mcr-mir-2	AAGGCGAAUGAAUGCUCUGUCUGUUGGUAUGCGAUAGGACCCAGUUUUUCAAAAGACUGUGUUGCUAUGUCCCAGACAUGUUCUACUUUCGCUAA	95	23,15789	24,21053	20	32,63158	43,15789	56,84211	1,347826	1,157895	-26,9	-26,9	0,129077	12,83	-28,3158	-0,6561
mcr-mir-2001	AAAGGAGAACUGUGUUGCUCUUGAUCGGAUGCUGCCAUUUUGUGACCGUUACAAUGGGCauugacagaaaaacaaaugcuCCAUG	85	21,17647	16,47059	15,29412	22,35294	36,47059	38,82353	1,357143	1,384615	-20,5	-20,5	0,029997	7,42	-24,1176	-0,66129
mcr-mir-202	CAGAGGCauagggcaugugcggggccuCGACCAUAGCCAUAUUAAUCAAACACGCGGGGCCCUAGGUAGAUGCCUCGUCUGCCUGCCCCUAGGCCUAAG	99	20,20202	19,19192	26,26263	14,14141	46,46465	33,33333	0,736842	0,769231	-39,3	-39,3	0,05055	8,37	-39,697	-0,85435
mcr-mir-204	CCACUCACCAGUCGCUGGCAAUGGACGCUGGGAAGGCAAAGGGAACCUCGCGCGUGCGUUCGagcgaagggagagagu	78	26,92308	16,66667	24,35897	11,53846	51,28205	28,20513	0,692308	1,105263	-30,4	-24,1	0,011827	6,78	-38,9744	-0,76
mcr-mir-2040b	AGAGCAAGAGUCAGGUCAGCUACGACUGUAACACUAACUAUUUAUGAUAAGGUAUCGUGUGUAGCUA	95	24,21053	32,63158	15,78947	27,36842	40	60	0,83871	1,533333	-32,6	-32,6	0,506504	3,85	-34,3158	-0,85789

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GAGUCUUAGCUGUGAUGACUAAUGCAAA															
mcr-mir-216a	GUGUUUGUCUAAUCUCAGCUGGUAAUUCUGAGUGGAGUUCUGCACACCUCAAGUUACUAGCCGAGAUUACAUAAAUAU	78	20,51282	26,92308	19,23077	33,33333	39,74359	60,25641	1,238095	1,066667	-30,5	-30,5	0,146188	3,64	-39,1026	-0,98387
mcr-mir-216b-1	CGUAGGAAGAUGAGCCAGGAUUACAAGAUGGUUGGGCCUGCGGCCCUAAUAUCAGCUGGUAAUCCUGAGUGAGCUUGCCUCAG	83	31,3253	24,09639	21,68675	22,89157	53,01205	46,98795	0,95	1,444444	-31,3	-30	0,152237	7,92	-37,7108	-0,71136
mcr-mir-216b-2	UGUCACUGUGCAUGACACUUACCUAUAGAGAUGUGUAUUGUCACUGUGCAUGGCACCCAGUAUGGUACCUAUAGAGAUGUGUAUUGUCACUGUGCAU	97	23,71134	23,71134	19,58763	32,98969	43,29897	56,70103	1,391304	1,210526	-26,6	-16,37	0,100351	18,69	-27,4227	-0,63333
mcr-mir-216b-3	UACCUAUAGAGAUGUGUAUUGUCACUGUGCAUGACACUUACCUAUAGAGAUGUGUAUUGUCACUGUGCAUGACACUCACCUAUAGAGA	88	20,45455	28,40909	19,31818	31,81818	39,77273	60,22727	1,12	1,058824	-26,2	-26,2	0,289845	6,27	-29,7727	-0,74857
mcr-mir-2238k	CGGCUUGGAUGACAGGUUAGAUGCGAAAUCUUCAAUGACUGAACAGUGCAAUGCAGUUUUCGUGUCUAAACGUGGUUUgcaagugu	86	24,4186	23,25581	15,11628	27,90698	39,53488	51,16279	1,2	1,615385	-27	-26,2	0,149473	4,18	-31,3953	-0,79412
mcr-mir-2253a	GGAUAAACAGCUUGCACGCAUGGCACUGCUGAAGGUUCAUGCAUGUAUAGCAGCAACAUGUGUCAAGUGCCUUUAAGC	78	25,64103	28,20513	21,79487	24,35897	47,4359	52,5641	0,863636	1,176471	-22,7	-22,2	0,18444	5,69	-29,1026	-0,61351

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-2254	ACAUUAUAAUUCUCGGCAGUCUUGCGGUUUGUACUAUGCAAAGUAAGUACUGCCGACCAUAUAUGAAGAA	70	20	31,42857	18,57143	30	38,57143	61,42857	0,954545	1,076923	-21,8	-21,4	0,07388	7,52	-31,1429	-0,80741
mcr-mir-2288	cccaccccaccccacgcCCUCAUCCACCUUCGACUUCUGAGCUCGAUCGUCGUUGCCUACAGCAGguaguaggugugugugugugggggggaguaGG	99	11,11111	8,080808	18,18182	13,13131	29,29293	21,21212	1,625	0,611111	-43,8	-42,9	0,179465	5,79	-44,2424	-1,51034
mcr-mir-22b	AUUAGCCUCAUCAUAGUUUACUGGACUGUGGUGUGGACAGCAGCGUUUGCAGCCUCACCUACAGUUCUUCAACUGGACUGAGCUCGC	87	24,13793	20,68966	26,43678	28,73563	50,57471	49,42529	1,388889	0,913043	-29,4	-28,1	0,059837	13,38	-33,7931	-0,66818
mcr-mir-2304-1	AGAACACAAACGAGAAUCCUAGGUAUAUGUGUUUUGUAUUGUGAAGACACAACACGUGUGGACGUAGUgguugugugguugugugug	87	19,54023	26,43678	11,49425	20,68966	31,03448	47,12644	0,782609	1,7	-24,4	-21	0,020111	11,06	-28,046	-0,9037
mcr-mir-2304-2	AUACGCUgguugugugguugugugugcaauuuuuauuuuuaucaucguGUAAAACUUUGCCAAAAAAAGUCACAUUCCACAAUCCAGCGGUC	93	7,526882	20,43011	15,05376	11,82796	22,58065	32,25806	0,578947	0,5	-30,4	-26,3	0,226556	12,43	-32,6882	-1,44762
mcr-mir-2325c	CUGUACUGUGCAAUuuuuuugggguguuuuuuuucuuuugcuugaagCAGACGAACUUGUAAUAACGAAAAAAACCCAAGAGAAAUAACUACAUAAAU	98	9,183673	31,63265	12,2449	13,26531	21,42857	44,89796	0,419355	0,75	-20,8	-14,6	0,057963	16,12	-21,2245	-0,99048
mcr-mir-234	ACUCCGGCACUUCCGACAUGCAAUAAUUAAAGAUUCUUAAUcaguu	83	14,45783	22,89157	16,86747	21,68675	31,3253	44,57831	0,947368	0,857143	-18,5	-17,5	0,15476	7,04	-22,2892	-0,71154

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	uuuuccuguuuugauUAUUGCUCGAGAAUGACGGCUG															
mcr-mir-2390	aauuuuccugAUUUUUCUACACCUGCAUCCCAACACAAGUGGGAUCUACUAAAUCCUUGGGUCUUGuaguugugugguuuuuguugaaaaacaagaaaaua	100	9	14	15	18	24	32	1,285714	0,6	-24,1	-24,1	0,203585	18,05	-24,1	-1,00417
mcr-mir-242	AAUUUGUUAUGAACAAAUGCUGUAAGUUGAAUCUUGCCUUUUUUCAAGGGCAAGGAUUUGCGUAGGCACUUUGCAUGACAAUGU	84	22,61905	27,38095	14,28571	35,71429	36,90476	63,09524	1,304348	1,583333	-27	-25,4	0,060999	11,38	-32,1429	-0,87097
mcr-mir-2434-1	UUGCCUACAAUGCCCUCAAAUCUGAGaguuuguuggggguuuuuuuugaaaaaauguccGAACAUAUCACGUGAUAUAAGGUGACAUGUUGGACA	94	13,82979	21,2766	13,82979	17,02128	27,65957	38,29787	0,8	1	-26,6	-20,5	0,012933	15,5	-28,2979	-1,02308
mcr-mir-2434-2	GCCUUCAAGGCCUAUGAGGCUCuugcuaaauuuuaaaauaaaagccaguuguuggggguuuuuuuaaucAGCAUGUGCCCCAUGGGCCUUGAAAGU	95	14,73684	10,52632	14,73684	11,57895	29,47368	22,10526	1,1	1	-37,6	-29,5	0,042743	12,24	-39,5789	-1,34286
mcr-mir-2444	UGGUCAAAACUAACUUCAACAACAAUAUAUCGUUGUUUCUUUUACCAUGGCACUAAGCCGugaaggugauuguguuguuuuuuguuuugaaaa	93	8,602151	20,43011	15,05376	20,43011	23,65591	40,86022	1	0,571429	-21,7	-18,9	0,098683	7,42	-23,3333	-0,98636
mcr-mir-247	gagggggaaacagAUAAACAAGUUGUACGGUUUGUCUUCGAGAGGGAGAAAAUCACUAGAGAAAAGUUUCUACCGACAACUUGAAGUUAUUUCCCCCCU	99	17,17172	28,28283	17,17172	24,24242	34,34343	52,52525	0,857143	1	-32,3	-30,6	0,021058	14,61	-32,6263	-0,95

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-2470	GCCAGAAAAACCUAAGCAUGACAAAAUAUCAUGCUUGUGUGGUUUUGGAUACUUGGAUUUUUCUCUC	67	19,40299	28,35821	17,91045	34,32836	37,31343	62,68657	1,210526	1,083333	-18,6	-18,5	0,281925	3,5	-27,7612	-0,744
mcr-mir-2491	GCUGCCaugaaacaacaacagcagcagcaacagcaacagcaagagGGAGCUGCUGAUGUGGAGAUUUUUCUGGCAGA	77	18,18182	7,792208	9,090909	14,28571	27,27273	22,07792	1,833333	2	-27,8	-25	0,10123	15,43	-36,1039	-1,32381
mcr-mir-250	CACUUGUCCCUGUGUACAGCACUUUAGUGAAUCACAGUCACCUGAGUAAAGAUGACUGCGCAGGGGCAACAC	72	23,61111	27,77778	26,38889	22,22222	50	50	0,8	0,894737	-26,4	-26,4	0,33034	11,12	-36,6667	-0,73333
mcr-mir-252	CCCAUAUCUCAGCAGCAACGGGUAGAUUUUUGUGCAACUAUCAAAAUAGUCUCGGUUGUAAAUUCACCUGCUCCCUGCUGCUGCCUUGCGAUAUAUC	97	18,5567	23,71134	26,80412	30,92784	45,36082	54,63918	1,304348	0,692308	-26,9	-26,9	0,313291	8,87	-27,732	-0,61136
mcr-mir-252a	CAUAACUUCUGGCCCAUUACUAAGUACUAGUGCCGCGGGAAGAUAACGUGUCAUGUCCCGUGGUUCUGGUCCUUACUACAGGGCGGAACUUGCA	94	25,53191	22,34043	25,53191	26,59574	51,06383	48,93617	1,190476	1	-37,6	-37,6	0,061669	5,2	-40	-0,78333
mcr-mir-2582a	GUGACAGCCUGUAUUUCACAGUGUUCAUAAUAUUUCAGCUCCGUUUUGCCUUUGUAGCGAUGUCCCUAGCAAGACUGCUGAAACACUGacuuguaaa	97	17,52577	20,61856	21,64948	29,89691	39,17526	50,51546	1,45	0,809524	-19,7	-13,6	0,037527	15,34	-20,3093	-0,51842
mcr-mir-2699	UUUUGAGCUGUUUGUGAUAUUCGGCAUUCUUUACUCccugucucuguuucucucucuccccaucccCCUCACAC	100	14	18	15	23	29	41	1,277778	0,933333	-19,4	-16,9	0,099228	8,03	-19,4	-0,66897

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CGAGAUAUCGAGAAAAAGACUCAGGU															
mcr-mir-277-1	CAACAGACAAAUGUGUCAACCGUAAAUGCAUuuucugcugcauuuuauugACACGAAAUGUCUGGAU	67	13,43284	28,35821	14,92537	14,92537	28,35821	43,28358	0,526316	0,9	-23,1	-23,1	0,480593	2,17	-34,4776	-1,21579
mcr-mir-277-2	cuccccccucccacacacacacucaucugCCCGUUCAGUAAAUGCAUUAUUUGGGGAGGUAAUGCUGGGUGAAGGAAGGGGUUC	84	23,80952	15,47619	8,333333	17,85714	32,14286	33,33333	1,153846	2,857143	-25,5	-13,7	0,072633	12,27	-30,3571	-0,94444
mcr-mir-279	CUCUCUUAUCUUUGCGGGUGGCUGUGAGUCUAGUCCAUGUGACACUGCAAGCUCAUGACUAGAUCCACACUCAUCCACAAGGAUAGGACGU	91	23,07692	23,07692	26,37363	27,47253	49,45055	50,54945	1,190476	0,875	-39,9	-39,4	0,157559	5,72	-43,8462	-0,88667
mcr-mir-281	AAAUGACCUCACGCUGAAGGGAGCAUCCGUCGACAGUCAGAAAUGCAGCACUGUCAUGGAGUUGCUCUCUUUACUGAAAAGGUCAAGA	88	25	30,68182	22,72727	21,59091	47,72727	52,27273	0,703704	1,1	-35,2	-35,1	0,187538	3,76	-40	-0,8381
mcr-mir-283	GCUAAAUCGUGAUCCGUGCUAUAUUUAUCCUUGCUGGAAGAAAGUUAUCGGAAGGGGGCAUCUCCGAUAAAAUAUCAGCUGGUAAACGAAUUGGA	95	25,26316	30,52632	16,84211	27,36842	42,10526	57,89474	0,896552	1,5	-25,4	-24,2	0,095817	6,58	-26,7368	-0,635
mcr-mir-285	UUGUGUACGACGCUCUGUCCAUCAGUCAACUGGCUUCCAAAGGUCUCGAAAGCUCUGUUACUGCUGUAGGGAGGCGUCAUCAUUA	85	24,70588	21,17647	24,70588	29,41176	49,41176	50,58824	1,388889	1	-22,3	-15,9	0,018167	14,62	-26,2353	-0,53095

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-2970-1	cgaAAUGCGGGUGGUGGACAAGGAUGUUCccugcacacagagacacaugAGUCGGUCGACCGAUCAGAUCAUCUCUUGAUCAACCUACAGCAUUCAG	97	20,61856	19,58763	17,52577	18,5567	38,14433	38,14433	0,947368	1,176471	-25,4	-16,4	0,010574	31,19	-26,1856	-0,68649
mcr-mir-2970-2	AGCAGGAGCCGGCCGGAGACGGCAAGCCUGGGUCCCGACAGUCAGCAGUUGGCUCCCCAUGGGUCUCUUGCCGGAUCAACCCGCUCCUACC	91	30,76923	17,58242	36,26374	15,38462	67,03297	32,96703	0,875	0,848485	-41,1	-41,1	0,129828	8,12	-45,1648	-0,67377
mcr-mir-297b	aaauauguaugugugcaugaacAUGCUUUAAAAUGUGUCAGUUGUAAGCUGUACAACUGAACAAGCUUUAAGAACAGAACUAUGAAAAAUCAUAGCA	97	13,40206	31,95876	11,34021	20,61856	24,74227	52,57732	0,645161	1,181818	-23,8	-23,8	0,133853	5,73	-24,5361	-0,99167
mcr-mir-29b	UUAAGAAUCUUGCAACAUAGCACCAUUAGAAAUGUUAGUUGUUGAGUAGAGUAGAUGGCUAAAUAUUUCUUACGGUAUUUUGCGGACUCUCAU	93	20,43011	30,10753	13,97849	35,48387	34,4086	65,5914	1,178571	1,461538	-19,8	-19,8	0,035107	6,69	-21,2903	-0,61875
mcr-mir-2a-1	UGCAAAGCUAUAGUGCUGACCAAGUGACUGGGAUGUGUACCUAAGUGUCAUAUCACAGCCUGCUUGGAUCAGUAUUGUGGUUUUUAA	87	25,28736	25,28736	17,24138	32,18391	42,52874	57,47126	1,272727	1,466667	-38	-37,6	0,220043	3,11	-43,6782	-1,02703
mcr-mir-2a-2	GAAAAGCAAAUGUGCAUGGAGCAAUCAAAGUAGUUGUGAUGUGUUUCUUGGCUUCAUAUCACAGCCAGCUUUGAUGAGCUUCUUCACAAUUUUCUUGCA	99	21,21212	27,27273	18,18182	33,33333	39,39394	60,60606	1,222222	1,166667	-33,6	-31	0,110768	7,68	-33,9394	-0,86154

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-2a-3	UACGACUGCUGAGUCCACAUCAAAGAGGCUGUGACUUUGUGAUGCUGUAUCAAAUCACAGCCUGCUUUGGUGAGCUCUUUGCUGUCCAA	89	23,59551	22,47191	23,59551	30,33708	47,19101	52,80899	1,35	1	-36,2	-35,9	0,18632	8,53	-40,6742	-0,8619
mcr-mir-2b-1	UGCAAGAAAAUUGUGAAGAAGCUCAUCAAAGCUGGCUGUGAUAUGAAGCCAAGAAACACAUCACAACUACUUUGAUUGCUCCAUGCACAUUUGCUUUUC	99	18,18182	33,33333	21,21212	27,27273	39,39394	60,60606	0,818182	0,857143	-24,4	-24,4	0,305458	6,72	-24,6465	-0,62564
mcr-mir-2b-2	GAAGCGAGGCCAGCUCAUCAAAGCUGGCUGUGAUAUGACUGAACUCAACACAUCACAGCAUCUUUGAUGGACAGGUCCUAGCACU	85	23,52941	29,41176	25,88235	21,17647	49,41176	50,58824	0,72	0,909091	-31,5	-30,9	0,181124	5,11	-37,0588	-0,75
mcr-mir-2c	aaaagagaUUCGUCAACAUUGCCUGCUGCUUAUAAGACCUUUUAGCUUCAAUGUUUUAUGACAAGGUGCAAACAUGUUGACUuaagaaucucucuc	96	13,54167	19,79167	15,625	28,125	29,16667	47,91667	1,421053	0,866667	-27,8	-27,8	0,136659	7,8	-28,9583	-0,99286
mcr-mir-3131	UCUUAGCCAAUCAGAAAACAUUCCAUCAAGAAAGUGUGGUGUGGGAUGAGCACACACCUGACUCGAGGACUGGUGGAAUUUUCCAUGAAGUUGGUAACC	99	25,25253	30,30303	20,20202	24,24242	45,45455	54,54545	0,8	1,25	-27,7	-27,5	0,063458	7,98	-27,9798	-0,61556
mcr-mir-315b	UUUGCAGACUGCACUGAUAUAACAAUACCGCCGGAGGUCUCUUUUCUGCUUUGUGGACAUUUUGAUUGUUGUACAGUCCGCUUCUGCUUU	90	21,11111	17,77778	22,22222	38,88889	43,33333	56,66667	2,1875	0,95	-24,6	-24,6	0,280807	8,3	-27,3333	-0,63077

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-317	GGGAAGGAUACCAUUGCUUGUUCACAGUAAAUUGGACAUGUGAACACAGCUGGUGGUAUCUUUUUU	66	25,75758	25,75758	15,15152	33,33333	40,90909	59,09091	1,294118	1,7	-29,1	-29,1	0,162636	2,6	-44,0909	-1,07778
mcr-mir-3223	ugaaaagguuaugggaugaccuuuaccuuuuaccuuuaccuuacagAAGGGUCAACCGGUCAAGCUCAUUCAUGCAUCUUUCCC	84	8,333333	10,71429	14,28571	11,90476	22,61905	22,61905	1,111111	0,583333	-18,5	-18,2	0,011779	13,83	-22,0238	-0,97368
mcr-mir-324	uugAACUCGAUUGUGACGUCUACAUCACAUGGCuucuuuccccacugccccaGGUGUCUUGUGAAGCACAUCGAGUUUGC	80	18,75	16,25	16,25	21,25	35	37,5	1,307692	1,153846	-23	-21,4	0,113698	17,15	-28,75	-0,82143
mcr-mir-3299	ACCUUGGGGGUUUUGGAAGCUUAAGCUCCAGGUUGUUUCUUUCUGCUCUAACUUGCAGGAGAUCGUUCAUUGUUAAAGUCUGCAUAACCCCCAACCU	97	21,64948	20,61856	23,71134	34,02062	45,36082	54,63918	1,65	0,913043	-31,4	-30,2	0,042103	13,35	-32,3711	-0,71364
mcr-mir-33	GCUAUGGUAGUGCAUUGAGGUUGCAUUGCAUCAGCUGAGAAACAUGCAAUGCAUCUGCAGUGCAAAUAUCAUGGU	75	26,66667	28	17,33333	28	44	56	1	1,538462	-33,6	-32,6	0,179681	5,16	-44,8	-1,01818
mcr-mir-33b	AGUGACAUGUACUUAAGUUACUGAAAGUACUCAAAUGUGUUUUAUGCAAUGUACCUGCAGUGCUuucagaaaagcaaaaacuuGUCUAU	89	15,73034	22,47191	12,35955	28,08989	28,08989	50,5618	1,25	1,272727	-19,3	-16,7	0,024573	11,69	-21,6854	-0,772
mcr-mir-34	UGAUCCCGUGGCAGUGUGGUUAGCUGGUUGUGGUCCGGCCGGUUUGUGACAACCACUAUCUGCAGUGCCGCGUGAUUG	78	34,61538	12,82051	23,07692	29,48718	57,69231	42,30769	2,3	1,5	-36,4	-36,4	0,283704	3,17	-46,6667	-0,80889

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-3529	UGUGGCUGCAGCAAUCAGGGAAGAAUGUGAUUUAUUGUUAGUCAGCAUCAGAACAACAAAAUCACUAGUCUUCCAGAUCACGGCAGCCugg	91	21,97802	30,76923	20,87912	23,07692	42,85714	53,84615	0,75	1,052632	-30,8	-29,7	0,103281	5,67	-33,8462	-0,78974
mcr-mir-3530	AGAGACAAGGUAGGCCUCCGGUGAAGAGAACAAUGGUGUGAGCUGGACAUGACUUGAUUAGCGAACGACAUCGAUCUCCCCGGCGCCACCUUUUCGGC	98	29,59184	25,5102	25,5102	19,38776	55,10204	44,89796	0,76	1,16	-31,7	-30,2	0,121532	8,78	-32,3469	-0,58704
mcr-mir-3671	AGUCACAUAGGUCGUAAGUUGAUCAUUACCUGUAACGUUUUUUAAAGUGAUCAAAUAAGGACUAGUGGCA	70	21,42857	32,85714	14,28571	31,42857	35,71429	64,28571	0,956522	1,5	-18,7	-18,5	0,348559	4,37	-26,7143	-0,748
mcr-mir-371b	AGUGUACGGCACGUGCUCCCCGCCGCUAGCGCACUCAAAAGAUGGCGGCAAGGGGGUGACGGGGGAGUCGCACGUUGCCGAACCGU	86	37,2093	19,76744	29,06977	13,95349	66,27907	33,72093	0,705882	1,28	-38,5	-35	0,126533	11,61	-44,7674	-0,67544
mcr-mir-372	GGAUGAUGUUCGAAAGUGCUGCGACAUUAUUACCGACGCUUAUGGUGGCCUCAGGUAUCGUACACGGCUUCUGUGUCGAAAUCCUUUCGUUCAUCAUUA	99	24,24242	21,21212	22,22222	32,32323	46,46465	53,53535	1,52381	1,090909	-32	-29,1	0,23008	11,94	-32,3232	-0,69565
mcr-mir-373	acggaugauAUAAAGGUGAUGUUCAUUAUGCCCAACUUGAUGGGCUUGGGCAGAAGGAUUCUCAAGCAUAAAAGUGCUUCCCUUUUAUUAUCAuu	95	18,94737	25,26316	15,78947	28,42105	34,73684	53,68421	1,125	1,2	-26,2	-25,2	0,010522	20,57	-27,5789	-0,79394

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-375-1	UUUAGCUGCCCAAUGACCCGAGCCGCUCGUAGCAAGGCGUCUUCACAAGUUUUGUUCGUUCGGCUCGCGUUACCAUGGUGGCACUGAC	88	26,13636	17,04545	29,54545	27,27273	55,68182	44,31818	1,6	0,884615	-35,3	-35,3	0,510508	5,41	-40,1136	-0,72041
mcr-mir-375-2	GGCUCUCCGUUUGUUCGUUUUGCUUGCUCAUGGGGUGGGUGGUGUGGGGGGAGAAGGGAGCGAGUAAUAGAGCAAAAUACUAAACUACGGAGACAG	96	37,5	22,91667	14,58333	25	52,08333	47,91667	1,090909	2,571429	-26,5	-26,5	0,439668	2,92	-27,6042	-0,53
mcr-mir-376c	AGGUGCAAACACUGCCGUGUCAUGUUCAACAUAGAGGAAAAUCACCGAGGAAGUUGUGACAGUGCACCUGACACGGUCACACGUUGCAUCG	91	26,37363	29,67033	24,17582	19,78022	50,54945	49,45055	0,666667	1,090909	-27,4	-27,4	0,263715	8,34	-30,1099	-0,59565
mcr-mir-376d	UUUCAGGUGUAGAUUUUCCUUCUACAACUUAAUUGCUUCUCGCCGCAGGUGAAGUGAGGUCGAGGAAGAAGGAAACGUCUCCACCUGUGG	90	26,66667	23,33333	21,11111	28,88889	47,77778	52,22222	1,238095	1,263158	-32,7	-22,97	0,106712	16,3	-36,3333	-0,76047
mcr-mir-3834	UACGUCAGGUCGAUCUCUCUGAUGUUAUUUUCGACAAAUAAGCGAAAAGGCAUCUUGACCUGACUGC	67	20,89552	26,86567	22,38806	29,85075	43,28358	56,71642	1,111111	0,933333	-24,4	-23,6	0,406778	1,27	-36,4179	-0,84138
mcr-mir-3890	UCAGUUCGAACAGUGUUUCUAGACUCUGAAGAGCCGGUUAUGUACGGCUGUUCAAAGGUGGAUAGCUGUCAACCCA	76	26,31579	25	21,05263	27,63158	47,36842	52,63158	1,105263	1,25	-25,4	-24,2	0,10708	10,57	-33,4211	-0,70556
mcr-mir-39	UAAAUGUGAACUUUCACAAUCCCCUAUCAUGUCUUUAGUCACCUGGCUAAAGCUGAUUUCGUCUUGGGAGUUCUCAUAAU	80	16,25	25	22,5	36,25	38,75	61,25	1,45	0,722222	-22,5	-22,5	0,059743	2,98	-28,125	-0,72581

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-4000i	GACAAAGAAAGCUUUUCUUGAAGUUUUUAAGACAGGUUGACACAGUAAGUCUUAAAGUAGGGAAAUCUGAAACUUGCAAGGAACUGCUUUCUUUAUA	97	20,61856	35,05155	13,40206	30,92784	34,02062	65,97938	0,882353	1,538462	-24,9	-21	0,02901	8,86	-25,6701	-0,75455
mcr-mir-4003d	GGAUUUGUUGACACUCGUGACAACUUGCAGAGAAAGGUGCAUAUUAAGUUUACACCUAUUCCGCAGCCUGUCACUGAGAAUGGUAACCAAAAAA	94	21,2766	32,97872	20,21277	25,53191	41,48936	58,51064	0,774194	1,052632	-22,7	-22,7	0,028311	9,4	-24,1489	-0,58205
mcr-mir-4027	CUACCAUCACCUCACCAUUUAACACAUUCUGUGCCUCUGGUGCACUUGAAUUAAAACACUAUGUAUAUUACUUUUAUGUUAAUGAGUGAGACGAAGGUGA	100	16	30	21	33	37	63	1,1	0,761905	-20,8	-18,1	0,009559	16,79	-20,8	-0,56216
mcr-mir-4029	ACCCACACGUGAGGAUCAAUUCCUGUUUGUUUACAUUGCAUGCAUCGCUAGUGUAGUAACUACGAGAAAUAGACACUCACACGUGUGCGC	90	21,11111	27,77778	24,44444	26,66667	45,55556	54,44444	0,96	0,863636	-23	-17,1	0,03183	24,52	-25,5556	-0,56098
mcr-mir-4034	GCCAUUAUUUUUGCAAGGUGAAGGUGAGCAGGGAUCUUUUCUCUGGCACUGAGGACCAAAAUAUCCGUAUUCACACgugcaacugaaaaaaaaauuaa	98	18,36735	20,40816	16,32653	22,44898	34,69388	42,85714	1,1	1,125	-21	-19,9	0,119554	9,47	-21,4286	-0,61765
mcr-mir-4040	AACUAGGGACUAGAUUUCAUGUUUACAAACUGAUGUAAUUUAACCCAUAGCACAAAAUACAACCAGAUCAGAAAGGUGAUAAGCUAGUUUCUACAC	96	15,625	39,58333	18,75	26,04167	34,375	65,625	0,657895	0,833333	-18,9	-18,5	0,175394	6,72	-19,6875	-0,57273

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-4048	UCAUUCCCUUUCCAGCACCUUGGUCACCAUAGUGACCCAUAGUGACCAAGGGGGAUGACGGGAAACC	67	23,8806	25,37313	29,85075	20,89552	53,73134	46,26866	0,823529	0,8	-25,9	-21,3	0,109726	8,22	-38,6567	-0,71944
mcr-mir-41	aagucUUGUCACCGGGUGAAAAACUGACGCCUGCAUAUCAUGGUUGAAUGUGGAGUCUGUGCAGUUGCUUGCGCGUGUGAACAAAGAAAA	90	27,77778	25,55556	16,66667	24,44444	44,44444	50	0,956522	1,666667	-28,1	-28,1	0,033067	9,16	-31,2222	-0,7025
mcr-mir-4127	AGGCAGCUGUGAGAGAGCCCUGUAUAUUAUAAUACCGCUAUAUAAGCCAGCUGUGAUUGAGCCCUGUAUAUUAUAAUACCGCUAUCUAAGCCAGCUGUGA	100	22	29	21	28	43	57	0,965517	1,047619	-22	-15,21	0,013453	23,23	-22	-0,51163
mcr-mir-4130	AUCGAACUCCCUCCGCUGCCCAGACAGCUGCGUGUCUUUAAGAUGGCGGCGCUUCUGCUUGGGUGACGAGAGGAGAGUCUCCCG	84	30,95238	16,66667	29,7619	22,61905	60,71429	39,28571	1,357143	1,04	-36	-35,6	0,091309	4,96	-42,8571	-0,70588
mcr-mir-427	UAGGUGACGGCCAUGUGAucgaaaagaaacagagaaagugCUUUCUGUUACUUCUUCUCAUAUCUGUCGGCACUUG	76	15,78947	10,52632	17,10526	27,63158	32,89474	38,15789	2,625	0,923077	-24	-24	0,200072	3,94	-31,5789	-0,96
mcr-mir-44-1	CGCAGGCGGGAGGUUGAAAUGCCGUUGUCGUGGUCAUCAGAUGACAGGACUGACAGCUGGAUGUGCUCGUUAACGUCGUUCUCCUGCAGCCUAAG	95	32,63158	20	23,15789	24,21053	55,78947	44,21053	1,210526	1,409091	-29,5	-21,4	0,033769	23,95	-31,0526	-0,5566
mcr-mir-44-2	GCUCAGUGCCUGUAUCCCUCAGUGACUGUGGGUUCGCACGACAGU	71	25,35211	23,94366	25,35211	25,35211	50,70423	49,29577	1,058824	1	-19,8	-18,5	0,203176	16,29	-27,8873	-0,55

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GACUAGAUGAUCACAAGACAUUGUCA															
mcr-mir-449a	UCACCUUUGCUUGUCCACUCCUCACAGUUGUAUAAAAUGGAGGUGGCAGUGCGUGUUAGUGAUGGAGAUGGUAAGGCACAGGAGG	85	31,76471	23,52941	17,64706	27,05882	49,41176	50,58824	1,15	1,8	-25,8	-25,6	0,084124	7,48	-30,3529	-0,61429
mcr-mir-4502	AcaaguugcugcugcugaugaugauggugauuggAAGUGCAGUCACAACAGACAGCGGCACACUGAC	67	11,9403	19,40299	14,92537	4,477612	26,86567	23,8806	0,230769	0,8	-25,2	-25,2	0,397491	8,17	-37,6119	-1,4
mcr-mir-452	CGGACCAUAUUAUCAAAUGUGACUGUUCGuuguuuuauuaucuuuugGUCAUAUAAACUGUUUGCAGAGGACAGUCACAUAUGUCCGAUAAGAAGGUCCU	100	18	25	16	23	34	48	0,92	1,125	-32,4	-32,4	0,183463	8,17	-32,4	-0,95294
mcr-mir-4552	CAUCAACUAACCAGCGUGAACAAGAGCGAGACCACCUCCCACUUGAUCACUGACAUUCUGGUAUGACCCGGGGUUAGUuggga	83	20,48193	26,50602	27,71084	19,27711	48,19277	45,78313	0,727273	0,73913	-26,2	-25,6	0,350856	1,8	-31,5663	-0,655
mcr-mir-466h	UcuugaguguuuguguguucaugGCGCCCUUAUGUUAAAGUCGGUUCACGCACUCAGACACguauacgcacgcacacacacacacacac	90	8,888889	10	13,33333	11,11111	22,22222	21,11111	1,111111	0,666667	-24,9	-24,9	0,117884	19,03	-27,6667	-1,245
mcr-mir-466i-1	auacacacacacauacacacgcgcacgcgcgcuaAAUGGCCUGUAUACAAACUCAUCAGAGCCUUUGCGCCAUAUUAUGAUGGUGUGGUC	90	14,44444	15,55556	13,33333	18,88889	27,77778	34,44444	1,214286	1,083333	-23	-17,8	0,062769	25,13	-25,5556	-0,92

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-466i-2	ugugugugugugugugugucucaauUGUUCUCUCCUAGUAGGUCAUCGUCUGAGACAUCUGUAGACAUAUUAAUCACUACAUCACCCU	90	10	17,77778	18,88889	23,33333	28,88889	41,11111	1,3125	0,529412	-23,3	-18,7	0,058678	13,27	-25,8889	-0,89615
mcr-mir-467c	AAAAAUCUGUGUGUCUAAGUGCGUGCAUGUAUGCAGGAACAUUUUGAUCACAUGCAUGCAGGAACACAGAUUGAA	75	24	33,33333	16	26,66667	40	60	0,8	1,5	-31,1	-31,1	0,307267	3,37	-41,4667	-1,03667
mcr-mir-4760	UGAUGAUCUCAUGAUGAUUUCUUAGCAAAGAAUUCAUUUUCUUGUGUCCCUUUUAGAUUGAACAUGAAUGUUUAAAACAGGAUAUGAGGUCAGAU	95	18,94737	30,52632	12,63158	37,89474	31,57895	68,42105	1,241379	1,5	-22,4	-18,9	0,059494	17,85	-23,5789	-0,74667
mcr-mir-4810b	UUCCACAUGUAGGUUCAUGAGUAGACUUUUCAGAGGCUGAAGUGAGUUUAAGCAAUACUCUGAGGGUGUAUGAUGUACAUAAUGCUGCAU	90	25,55556	27,77778	14,44444	32,22222	40	60	1,16	1,769231	-18,6	-16,1	0,008321	19,96	-20,6667	-0,51667
mcr-mir-4865	UACAUGCAAGGCAGGCAACUCACACUACACACGGUAUGUAGAGAGAGUGACGCCGCUUUGUAUCGA	66	25,75758	30,30303	24,24242	19,69697	50	50	0,65	1,0625	-22,8	-22,6	0,613718	1,85	-34,5455	-0,69091
mcr-mir-4968-1	UCAGAGCAGUGCUGCUCAGcagagcagcagcaacagcagcagcagcagcagcacacuCCGCAGGCUGACUCUCAGAGCAACAUGUCUCAGC	91	15,38462	13,18681	18,68132	10,98901	34,06593	24,17582	0,833333	0,823529	-29,8	-28,8	0,303196	26,91	-32,7473	-0,96129
mcr-mir-4968-2	UAGGCAACAGGUCAGCACAacuaagcagcagcagcaacagcagcagcagcagc	82	12,19512	10,97561	12,19512	9,756098	24,39024	20,73171	0,888889	1	-26,6	-18,9	0,141152	31,1	-32,439	-1,33

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	agcagcuguugUAGUGCCACUCUUGCUGG															
mcr-mir-4968-3	CCGGCGUGACGUGCAGAuuaccagcaacaacagcagcagcagccgccgACGCUGACUCGCCGAAGCUGCUUGAUGAUGCAAGUGCCGCCACGCGCC	96	21,875	12,5	22,91667	10,41667	44,79167	22,91667	0,833333	0,954545	-29,2	-26,5	0,033415	18,93	-30,4167	-0,67907
mcr-mir-4975	AUCAGGACUCAGCUGUCAAACCCAAAAUAUGCUUCUUUUUUACUUGGGUUUUCUUUcaggaagaaacaaauauugGUUCUGCUGAgccuuua	92	11,95652	15,21739	15,21739	29,34783	27,17391	44,56522	1,928571	0,785714	-22	-13,18	0,037553	24,75	-23,913	-0,88
mcr-mir-50	CGAGUAAUAUUAGACAGCGGAUUAUUCUUcaauauuauauuuaauaauauguauauauuaucuaUAAUAUAGAGUAACCCGUAGUCUUUUAUAACCUU	98	10,20408	20,40816	10,20408	23,46939	20,40816	43,87755	1,15	1	-24	-24	0,151437	7,98	-24,4898	-1,2
mcr-mir-5009	AGAAGUCAUACUGCAACAGCUCCUUCUCUUCGGUGGCCGAACUUCGGGAGUUGGACUUUUUCAGAUGGACUAGU	74	25,67568	21,62162	22,97297	29,72973	48,64865	51,35135	1,375	1,117647	-20,6	-20,6	0,124	9,33	-27,8378	-0,57222
mcr-mir-5101	GCGAuguugaugacuuuuguuuguuuugcuguauGAGUCUUUGUUGUAGAGCACGUGGAGCUGUUUACAAACUAAUUAGUCCGUcgacauuaa	93	16,12903	13,97849	9,677419	18,27957	25,80645	32,25806	1,307692	1,666667	-27,9	-27,4	0,018271	6,78	-30	-1,1625
mcr-mir-542	AUAAUCUUUACAACUGUGCUUUGAUGAUGUGCUCUCAUAAUAAAAUUGAUGUGACAGAUUGAUAAAGACGCACUUGGGAAAGAUGCA	87	20,68966	34,48276	13,7931	31,03448	34,48276	65,51724	0,9	1,5	-18,8	-18,8	0,137277	8,15	-21,6092	-0,62667

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-5429	ccaggaaccuccgGCAUGACUAUUCCUCAAGGCACAUUUACUACGGCGuugacuacccuccaaggugccuugaggAAUAGUCAUGCUGGAGGUUCCUAG	99	14,14141	15,15152	14,14141	16,16162	28,28283	31,31313	1,066667	1	-67,1	-67,1	0,328258	3,51	-67,7778	-2,39643
mcr-mir-548d	CACUGAGGGGAGUAUACUCCUUCAGUCAACACAGUUUUAUUACCCUgugagaagaacaaaaaaaacugcaguUACUUGAAAAUAAACUACCCUCAUUc	98	9,183673	22,44898	18,36735	22,44898	27,55102	44,89796	1	0,5	-18,9	-17,7	0,01295	10,68	-19,2857	-0,7
mcr-mir-550	CAAGUGCCUGAGGGAGUAGUGCCUUCUACAGGGAAAACAAUGACUGUAUGGAGCACGAUCUCCACCUUCAGAGGACACC	79	26,58228	29,11392	25,31646	18,98734	51,89873	48,10127	0,652174	1,05	-25,3	-22,1	0,053345	9,46	-32,0253	-0,61707
mcr-mir-5552	CUGCAGUCUAUAGAUAUAGCCUGUAGUCUGUUACAUUCUACCAAGUGAUCGCUACAGUUAUGUAGUUUGUAGUCUAUAGAUAUAUCCUGUAGUCUGUUA	99	19,19192	25,25253	17,17172	38,38384	36,36364	63,63636	1,52	1,117647	-28,8	-26,6	0,061307	13,22	-29,0909	-0,8
mcr-mir-5582	auuuguuGUAAUUUAAAACUUUAAGUGUGUCAGACAACUUUUCUUGCAAAAGCUAUGUGACCACAUAAGUUUAUAGUUUAUACAACAGGC	90	14,44444	32,22222	14,44444	31,11111	28,88889	63,33333	0,965517	1	-19	-18,7	0,032948	11,44	-21,1111	-0,73077
mcr-mir-5698	CAUACUGACCGCCCAACUGUGCACUGAAGUGGAUUUUAUUUAUCAGUGCgacacacccccucccccuuucuuuGUCCCAGUGUGGGGGAGUGCAGUGAA	99	22,22222	17,17172	16,16162	20,20202	38,38384	37,37374	1,176471	1,375	-28,3	-21	0,011177	15,74	-28,5859	-0,74474

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-586	CUCCAUGUUGAUGGUAUGCAUAUUGUAUUUUGUACUUGUAGUUCCCUGGGUACAGUCACCAGUACCAAUGAUGCCAACACUUUACAUGCAU	91	18,68132	24,17582	21,97802	35,16484	40,65934	59,34066	1,454545	0,85	-19,1	-15,9	0,059302	17,12	-20,989	-0,51622
mcr-mir-5890c	CUCCAGACUAGUUUCAUUCCCUGUGCAGUUCAGGUCAUUGACCCAGACGCUGCCAUAUAACUGAACUGUCUGUGG	75	21,33333	21,33333	28	29,33333	49,33333	50,66667	1,375	0,761905	-21,4	-18,9	0,044239	9,08	-28,5333	-0,57838
mcr-mir-592	CCUGACGGUGAACAAGAACUCGGUCCUCGCAUAGCGACGUGACGUCAUCACGUGGUGACGUGUUCUACGCGCCAUCUAGGUCUUUUUGCCCGUCGUC	97	26,80412	17,52577	29,89691	25,7732	56,70103	43,29897	1,470588	0,896552	-31	-28,5	0,039975	20,27	-31,9588	-0,56364
mcr-mir-6013	UUGUCCACGGCUGCUGACGACCGCCUCAAACGUCAGAAGUUGAAGGAAAACAGAGAGAGCUUGAGCGAGUCGCUGGCGCAGUGGAAGC	88	32,95455	27,27273	23,86364	15,90909	56,81818	43,18182	0,583333	1,380952	-27,3	-26,6	0,063669	7,38	-31,0227	-0,546
mcr-mir-6058	GGAGAAGAACUGGACAUUACUCAGCUGAUGACAAUGGUGAUGAAUCAGAUGGGUUACAAUAUGUAGUUGUCCUCUGCUGAUUUAUGGACGUCUUCCAU	98	25,5102	27,55102	16,32653	30,61224	41,83673	58,16327	1,111111	1,5625	-26,2	-26,2	0,10919	8,67	-26,7347	-0,63902
mcr-mir-6340	GCGGUGCUGGGUUUGCUCAGCUGGUGGAAGAUCAACAGACGUCAGCAGCAGCUUCGAUACCCAACACGAC	70	30	24,28571	27,14286	18,57143	57,14286	42,85714	0,764706	1,105263	-22,1	-22,1	0,286043	7,87	-31,5714	-0,5525

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-6552	CCCAUCAUUCGACUUCAGCAGGGAAGACUUUCGCGAUCCUUUUCUAAAGCAACAAGAAGUGGACACGGUCUAUUUCCUGCAGGUGUCUGAAAUGAUUCA	99	21,21212	27,27273	24,24242	27,27273	45,45455	54,54545	1	0,875	-28,3	-23,2	0,012686	10,87	-28,5859	-0,62889
mcr-mir-6560	CGCUGUGUUGCUCCAUACUGCUGAAGAAGUGUCUGCACUUCUGUUUCAGUCAUACUGAGCAGCUCAUCA	69	21,73913	20,28986	26,08696	31,88406	47,82609	52,17391	1,571429	0,833333	-24,8	-24,8	0,250515	3,95	-35,942	-0,75152
mcr-mir-669f	CCUGCGAACCAUUAUGCUGGaauucucucucuauauauauauauauacacacacacacacauaagaauUUCAUUUAAUGGUUUCGCGUG	89	11,23596	7,865169	10,11236	16,85393	21,34831	24,7191	2,142857	1,111111	-19,4	-18,5	0,056046	7,7	-21,7978	-1,02105
mcr-mir-67	GAGAUAUCGUGAGCUGUUGGUCCUUGUUCAGCCCGGUUGUGAUGGAGUUGAUCGCAUCACAACCUGCAUGAAUGAGGACUGACGUCAcgaaauaac	96	27,08333	19,79167	18,75	25	45,83333	44,79167	1,263158	1,444444	-47,7	-47,7	0,225173	3,14	-49,6875	-1,08409
mcr-mir-676	CUCUUCAACUUCAGGAUCGUUAAGACAAAAUCAUAGUUGCUGAUAUAGCAGAGCCUGGAuauguccuugucuuggccuugaCGAUGCUGAAUUUAAAGUG	100	17	25	14	22	31	47	0,88	1,214286	-30,9	-30,9	0,08456	5,13	-30,9	-0,99677
mcr-mir-6769b	UGAUGGUGGGUGAGGAGGAGAAAGCAAUUAGCAAUAGUUUUCGAAAGACUUUCUGGAAACUCGAUCGAuuauuucacucacucacucacucacucacuc	99	21,21212	22,22222	8,080808	17,17172	29,29293	39,39394	0,772727	2,625	-29,4	-23,1	0,04736	29,05	-29,697	-1,01379

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-6780a	AAGCUCGUGUAGAAAAGCUCcuccucuguuuucuuuucaaaaugcaUGUCAGGUGCAACAUAGACGUGUAGUAGGAUGAAUCUUGACACACGAGGCC	97	20,61856	22,68041	14,43299	15,46392	35,05155	38,14433	0,681818	1,428571	-23,6	-23,3	0,05752	17,22	-24,3299	-0,69412
mcr-mir-6796	GGAGUACAAAGUUUCCGUCCUGAUUUUCUUGUGGGGUUGGAGAGUAUGGCUCCAUGCAAAGUCUAGAAACGUaaacuuggacaau	85	24,70588	20	14,11765	25,88235	38,82353	45,88235	1,294118	1,75	-24,9	-23,7	0,22456	3,8	-29,2941	-0,75455
mcr-mir-6800	CUGUAGUAGGUGACAGUCAGGCCUGGUCUCUUCUUGUCACCAAGGCCACAUUAUAGGGCUAAGAGGAGGCACUAGUAGUUCACUACUAUAU	91	25,27473	25,27473	21,97802	27,47253	47,25275	52,74725	1,086957	1,15	-26,7	-19	0,021509	23,54	-29,3407	-0,62093
mcr-mir-6803	CUACACGCACCGCUGCCAGCACCGCCACGCGCCGAGGGCAGGUGUAAACGGAGCUGUCACGUGACCccgagggugggggugggggcugggAGAGUGGCU	100	24	16	26	9	50	25	0,5625	0,923077	-43,7	-40,7	0,072188	21,19	-43,7	-0,874
mcr-mir-6826	CACCUUACUGAUUCGUUUCACAACAGGACUGGUAUGUUCUGUGUCAAUAGGAAAGAGGUCUGUGAUUUGUCCUGUUUGUGAAUUGAUCAUACAGUAC	97	22,68041	24,74227	17,52577	35,05155	40,20619	59,79381	1,416667	1,294118	-25,9	-25,1	0,012492	20,76	-26,701	-0,6641
mcr-mir-6830	AGGAACAGGUGCGAUACGAUCGAGAGAGACAAGCGGGUCCUCAGAAAAUCCUCUUGAAAugcuuucuugucuuucuucucucccccaucuguuuuu	96	17,70833	21,875	12,5	9,375	30,20833	31,25	0,428571	1,416667	-29,9	-29,5	0,041352	6,82	-31,1458	-1,03103

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-6864	GCAGUCGGAGGGUUAAAGUGAAAACAAAGGCAGUACUUGAAGGGACAAGUCAGACUUUAAUGACGCCAUGGGUAUUUCCUGUUACCCCGACAAU	94	26,59574	31,91489	19,14894	22,34043	45,74468	54,25532	0,7	1,388889	-23,6	-23,2	0,154259	11,86	-25,1064	-0,54884
mcr-mir-6873	CCUUGCAUGUGUCACAUGAGCAAAGAUUCAGAUAUAUGCACACACUUGCAUauguucucucugucuuucucucaugugcacUUGCAUGA	89	12,35955	20,22472	14,60674	19,10112	26,96629	39,32584	0,944444	0,846154	-35,2	-34,8	0,118812	5,2	-39,5506	-1,46667
mcr-mir-6925	ACAACACCUGAAGACUuuucuucaaggcaccuuggaggguagucgaugcUCAGUUGCAAGAGGAAGAAAUGGCUUACAAAUGUcag	86	12,7907	23,25581	10,46512	11,62791	23,25581	34,88372	0,5	1,222222	-20,5	-19,9	0,218392	9,52	-23,8372	-1,025
mcr-mir-6955	GCCCACAAGAGACAGCAAGUAGGAGGCACAUCCAGACACGAGUGCCUGGCAUUCUACACCUGUCUCCUUUGUCGUGUCCUUUGCUU	86	23,25581	22,09302	30,23256	24,4186	53,48837	46,51163	1,105263	0,769231	-24	-23,1	0,092411	15,34	-27,907	-0,52174
mcr-mir-6975	AUCGCGAGGGGAAAGGCUGAGCACCGCUCAGCAUACUUUAGUUGCUGUUGGCUCUCUCCUUUCUCCUCCGCCUC	74	24,32432	14,86486	32,43243	28,37838	56,75676	43,24324	1,909091	0,75	-33,1	-33,1	0,158058	5,02	-44,7297	-0,7881
mcr-mir-6988	gggugggguggagagcUGAGUUACACGCAGUGAGCACUGCUCAUCCUCAGGCACUGCCUACAACUACGCUCCGCCUCAUCG	81	16,04938	17,28395	29,62963	17,28395	45,67901	34,5679	1	0,541667	-33,4	-33,4	0,234622	9,97	-41,2346	-0,9027
mcr-mir-7	AAUGCAAAUACUAGGGACAGGGUUUUCUUUUACACAGACGGGCUGAAUCGCUGACUGGUUACAAAA	91	25,27473	29,67033	21,97802	23,07692	47,25275	52,74725	0,777778	1,15	-22,6	-19,7	0,032478	10,33	-24,8352	-0,52558

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GCCAACAAGUCCCAGUCUUGGCGGA															
mcr-mir-7042	AAGGGAGAAUUCAGUGGGAUCAUGUGGGGCUUGGAAGGGGAAGUAAUUUCAUGUUGAAGCCACCUAGUGACAGAUGUcccuuuguuuucuccaca	95	28,42105	23,15789	9,473684	20	37,89474	43,15789	0,863636	3	-36,4	-36,4	0,334669	3,58	-38,3158	-1,01111
mcr-mir-7051	AUGUUUAAAAGUUAUGUAGCUAAGUAUAAUGUUCACCAGGAGGAGGUGUGCACGUCCUAacuaaucauuauuauuauugcauaacAUUGUAAAGCG	96	18,75	22,91667	9,375	21,875	28,125	44,79167	0,954545	2	-21,7	-19,9	0,057538	7,51	-22,6042	-0,8037
mcr-mir-71	aaauuccuuuUGGUGUGAAAGACAUGGGUAGUGAGAUGUCGGUCCUGAAGAUUCACCUUAUUACCCUGUCUUUUGCAUCAAGGGGAAACC	90	24,44444	22,22222	16,66667	25,55556	41,11111	47,77778	1,15	1,466667	-44,2	-44,2	0,187606	4,57	-49,1111	-1,19459
mcr-mir-7186	UGACAGUAGAAUAGCGAGUUGGCGUGUGCAUUAAGAGCUGUGGUUCCUGUAUGUUCGGGAAGCACAGUCACGCUCAGCCGAUGCAUUUCACUGGAU	96	30,20833	22,91667	19,79167	27,08333	50	50	1,181818	1,526316	-38	-38	0,237347	14,88	-39,5833	-0,79167
mcr-mir-7234	ACUGUCAGAGUUUCGGGCUUUCUGUGGUCCCCUAAAGACAUGUUUUAAUCGUGUAGCUUGUUUUCUCCAAAGAACACCAUAAACUUCGACUGA	93	19,35484	24,73118	22,58065	33,33333	41,93548	58,06452	1,347826	0,857143	-21,5	-21,5	0,443504	4,24	-23,1183	-0,55128
mcr-mir-7241	GUGUUUGUUAGGGUAGUAGAUACUCAUGGUGAGUAACCCAAAAC	94	21,2766	20,21277	11,70213	20,21277	32,97872	40,42553	1	1,818182	-28,1	-27,8	0,130969	8,02	-29,8936	-0,90645

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCGGGGUUACGGAUGUaucugcuucauaacuacgaccccagAGCAAAUUC															
mcr-mir-7260	CGCAGCUGUUUUAAAGCCAGUAAUCAAGUCAUCCAGUACAAGGAGAAACAGUGCUGACAGUGCCUUUUACCGACUUUAGUGAUGGCUAAAACAACUGAC	99	21,21212	32,32323	22,22222	24,24242	43,43434	56,56566	0,75	0,954545	-22,8	-16,37	0,04421	16,66	-23,0303	-0,53023
mcr-mir-7371b	UCGAGCAACUUGAUGAUAUCGCUUAUUAUCUUCUCUUGAGUUGUAGAGAUGGACGGAGGAUAAGAUGCUUUGUAUCAGAUGCUUGC	86	25,5814	24,4186	15,11628	34,88372	40,69767	59,30233	1,428571	1,692308	-22,4	-20,5	0,034701	13,15	-26,0465	-0,64
mcr-mir-742	auuaucuGUGAUCUAGAGAGAAAGCCACCAUGCUGUUAUAUCACGUCUAUUGUUGAAUCGACUUCUUCGGUGGUGUCUUUUUCUACAUAACUACAUCAG	99	18,18182	23,23232	19,19192	32,32323	37,37374	55,55556	1,391304	0,947368	-19,2	-11,69	0,033442	29,11	-19,3939	-0,51892
mcr-mir-7456	UGCUGUUCGCCUGGCGAUCGCAUGGUAGCGUCUGGUUCAGGGAGAUGUACCUGCUGCCAGCUGAAACAACG	71	32,39437	18,30986	25,35211	23,94366	57,74648	42,25352	1,307692	1,277778	-24,5	-21,8	0,048336	5,34	-34,507	-0,59756
mcr-mir-7461	ACAUGCUCUUUGAUGACCAGCAUGGCUGGAAUGUGGCUGAGCAACAGUUCCCGGACGUUCUGGUUGAAUCAGAAGGCAAGC	81	29,62963	24,69136	22,22222	23,45679	51,85185	48,14815	0,95	1,333333	-26,8	-26,5	0,04965	8,31	-33,0864	-0,6381
mcr-mir-7462	UAUUAUGGCCUGCUCCAGGUACUGCGGUGUGUGAUGAGCGAUGAUAGGAUGCAACAGAAUAGCCACU	98	24,4898	28,57143	23,46939	23,46939	47,95918	52,04082	0,821429	1,043478	-23,7	-17	0,032162	19,67	-24,1837	-0,50426

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GACACAACCUCAUUUCUAGCAGACCAUACAG															
mcr-mir-7465	CACcaggaauuauuuuuaaacacucaGUUACAAAUCAGCUCAAAUCUGGUAAAACUGCUUCAGAUAGCUGAUACUCAUUUUUAGAACAAUAUUUCUGCAG	100	11	26	16	24	27	50	0,923077	0,6875	-19,7	-19,7	0,176123	6,74	-19,7	-0,72963
mcr-mir-750	GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUGGCAGGAAUGCUGUCUUCCAUGCCAGAUCUAACUCUUCCAGCUCAUAGUGGCAUGGCAUG	97	27,83505	19,58763	23,71134	28,86598	51,54639	48,45361	1,473684	1,173913	-52,7	-51,6	0,157091	3,48	-54,3299	-1,054
mcr-mir-7589	AUGUGCUUAAUUGAGCCCCCUUCGUCAGUUUCUCAUUCUGGAUGGAUUUCUGGAAAUGGAGAAAUCAGCCGAUGCGGAGGCUAACAGAGCAGUG	94	27,65957	24,46809	20,21277	27,65957	47,87234	52,12766	1,130435	1,368421	-29,8	-24,4	0,024214	13,36	-31,7021	-0,66222
mcr-mir-765	UGGUGCAGAAGGGAGUUCAGCUUACCAAAUACUGAGGACAUGGAGGAGAAGGAAGGUGUGGUCAGCUGAUCCUCACUUCCUGACAUUC	88	30,68182	27,27273	19,31818	22,72727	50	50	0,833333	1,588235	-24,1	-24,1	0,047094	9,07	-27,3864	-0,54773
mcr-mir-7683	ACUUGGAAAGGUGGAACACUAACCAUACUGGCACUGUGGACCUUGGAAACCUGGAGGACAGGCGCAGGAUGGAUAGUAGCCUCCUCUGCACUG	93	30,10753	26,88172	23,65591	19,35484	53,76344	46,23656	0,72	1,272727	-28,7	-24,8	0,044924	9,43	-30,8602	-0,574
mcr-mir-770	GGUCACAUUCAAUCACGUUACAGGUGCAGAUCGAACCUCGUGCCAG	79	22,78481	24,05063	27,8481	25,31646	50,63291	49,36709	1,052632	0,818182	-22,2	-22,2	0,116359	8,08	-28,1013	-0,555

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UCACGAUCAGCACCACGUGUCUGGAAUCUGUUU															
mcr-mir-7789	CUAUGAGCGAGUAGACUGCUGUGUUCCUcugagaagaggaagaagaugaaacGAUUCAGCAGAGCAGACGUCCAGCAGUCGAUGAUCGCUCACUU	95	20	16,84211	18,94737	18,94737	38,94737	35,78947	1,125	1,055556	-35,1	-32,4	0,057006	10,22	-36,9474	-0,94865
mcr-mir-7795	AGGAGACACGUGAUGCUUACAAUAUGACACCAGCAUCUAAUCUCUUAUCGGUAUAGGUGUGAAGUGUCAGAUGAUCGGUCUUGA	84	25	28,57143	17,85714	28,57143	42,85714	57,14286	1	1,4	-19,9	-19,9	0,054929	9,78	-23,6905	-0,55278
mcr-mir-789b	CCAUCCUGGUACACCACCAGCACAUCUCUCCAUUCCAGGUAUGAUGGCGUCGUGCUUCCGCUGCUGGAGAUGGCGGUCGCAGAUGCAGCAGGAAUC	96	27,08333	19,79167	31,25	21,875	58,33333	41,66667	1,105263	0,866667	-29,6	-23,8	0,110276	11,46	-30,8333	-0,52857
mcr-mir-7950b	UUUGGCAGGCCGGCGAAAGAGUUGCGCCAGACUCACCCAUCAAAGCAACCCGCAGGUCUGGCGCCUGCCUCCCUCG	76	27,63158	19,73684	36,84211	15,78947	64,47368	35,52632	0,8	0,75	-28	-24,8	0,1728	20,48	-36,8421	-0,57143
mcr-mir-7964a	CGCUCUAGGAUGCAGUGCGUAGUGCAAUCAAGCGGAGCGGAUGGGGAUGGUCAACCACUCGCUGCAUAUAGAUAA	75	32	26,66667	21,33333	20	53,33333	46,66667	0,75	1,5	-20,8	-17,3	0,208513	17,37	-27,7333	-0,52
mcr-mir-8	CGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUGUCAGCAUUUUCUAAUACUGUCAGGUAAAGAUGUCCACAGAGUUUG	86	22,09302	25,5814	19,76744	32,55814	41,86047	58,13953	1,272727	1,117647	-38,5	-38,5	0,175975	3,55	-44,7674	-1,06944

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-802	uuaccuuuguaCCCAUUCCAUAGUUUGUCAGUAACAAAGAUUCUGUUACACAAGUGACUUGGACCUUGGUGACAAGGAUU	80	17,5	25	17,5	26,25	35	51,25	1,05	1	-20,1	-20,1	0,088615	9,87	-25,125	-0,71786
mcr-mir-81b	CCUGCAAAAACUCAUGGGCCCUAUUUCGGCCCUAUAUGGGCUUAAAAUAGGCCCUGCAUGGUGAAACGAUCAUGGGCCUAUGUGUGUUUUGCCCA	95	24,21053	23,15789	25,26316	27,36842	49,47368	50,52632	1,181818	0,958333	-38,4	-35,6	0,107415	11,5	-40,4211	-0,81702
mcr-mir-8234	GAACAGGAUAUUGGCAUGGAGAAGCAACACCACUCUUUGCCUUCUCCAUCCCAUCUUCUCCUGCCU	66	16,66667	22,72727	33,33333	27,27273	50	50	1,2	0,5	-23,7	-23,7	0,285672	2,31	-35,9091	-0,71818
mcr-mir-8293	AGGAGUGAUAGGUAGCUGAUUUAUCAUGAAAGAUAAAAGUGGAUGAAUACAUUCCCUGUCCUAGACAUGAUAAACCACUCACAGUUCACUGua	93	20,43011	34,4086	17,2043	25,80645	37,63441	60,21505	0,75	1,1875	-21,3	-20,8	0,146917	10,11	-22,9032	-0,60857
mcr-mir-83	AUGUAGCACCAUAUAAAUUCCUGCUGCUGAGCUGCUGCUAACUAAUGCAUUGAUAGUUUUAAGGAAUGUGGAGCAGCAUAGUUACUGUGUUGCUAUAC	98	22,44898	27,55102	17,34694	32,65306	39,79592	60,20408	1,185185	1,294118	-26,9	-26,9	0,117633	12,41	-27,449	-0,68974
mcr-mir-8319	AACAAUGCAGCAAGAGGCUCAUUGCCAUagauuuugguuguuuuuuucugagcuGUUCAAAUUAGCAAGUGAGGUCAGGCUGCCUUUAG	89	17,97753	21,34831	14,60674	16,85393	32,58427	38,20225	0,789474	1,230769	-21	-10,6	0,048695	25,91	-23,5955	-0,72414

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-8335-1	uuguuguuguuguuguuuugcucaUGGCUGGUCAUUAAGACCAAACUGACAAAACUGCUUACAGACAAUUU	71	11,26761	23,94366	14,08451	16,90141	25,35211	40,84507	0,705882	0,8	-21,2	-18,8	0,146315	6,81	-29,8592	-1,17778
mcr-mir-8335-2	GCGCUGCAGAGguguauauauaauguuugccauguuguuguuguuguuuuuuuuuuuuuagcuuacaGACGAUUAUUAUAUUGAUACACUUGUGAGGCC	99	12,12121	11,11111	8,080808	12,12121	20,20202	23,23232	1,090909	1,5	-23,4	-20,9	0,130829	15,27	-23,6364	-1,17
mcr-mir-8364j	UCGGGGCGACUGCGUGAUCUCACGUGugagcaugcaugugugucuuCAUUUCUCACGUGAAUGCGCGCGcccgag	75	21,33333	9,333333	18,66667	16	40	25,33333	1,714286	1,142857	-28,6	-24,2	0,014357	24,88	-38,1333	-0,95333
mcr-mir-8485-1	cacacacacacacacacaaaagucacAAUGUAACUGCAGUGUGUUUUUACACAUGGUCAGCAGUGACAGUGUGAUAUAUUGAAGUGAGAGAGUGUUUU	98	20,40816	20,40816	7,142857	25,5102	27,55102	45,91837	1,25	2,857143	-25,6	-20,8	0,086901	11,94	-26,1224	-0,94815
mcr-mir-8485-2	gaguuugcacacacacacacacacguaugauACAUUUCGUACUCCGCAAGCUCAGGUGAAUAUGGCAGGAGUGUGUGUUUCAUGAUGCUGGCAAAAAU	98	18,36735	18,36735	12,2449	19,38776	30,61224	37,7551	1,055556	1,5	-24,8	-19,4	0,065813	22,98	-25,3061	-0,82667
mcr-mir-8485-3	cacacacacacacacacacacgucacauuCAAGAGGGGAGCAAUCUAGAUCCCUGACAGAUUAGGUAUAUGCAugugaugugugaauguuuuuuguaAG	99	13,13131	16,16162	8,080808	9,090909	21,21212	25,25253	0,5625	1,625	-26,8	-24,6	0,07266	13,36	-27,0707	-1,27619
mcr-mir-8881	CUGGUUUUUAAACCAAGAAAGGAAGAUAAAUUAUACAGAAUAUAAAUGCAGCCGUUGCUUUGUUUU	92	15,21739	30,43478	16,30435	38,04348	31,52174	68,47826	1,25	0,933333	-20,4	-20,4	0,106037	7,86	-22,1739	-0,70345

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CUCUGGUUCUUCUCUUUCAAAACUUU															
mcr-mir-8956	GCAGAUUCUGUUGACACAGCAUCCAACACAUCAGUGUGAAGGUGUGUUUCUUUGCCUGCAAAGCCCCUACAUUGACUUGGAUCUUUGCAACGUAUCGGU	99	22,22222	23,23232	24,24242	30,30303	46,46465	53,53535	1,304348	0,916667	-25,5	-24,6	0,178773	11,53	-25,7576	-0,55435
mcr-mir-8993	GUCAUGACCUUGACCUUUGACCAAGUGCAGCGCCUCUCGUGCUGACAUGAAUGAAGCAAAGGCCAGACUUGCUCCUGGCCAAGUGGCAUUGG	92	27,17391	22,82609	27,17391	22,82609	54,34783	45,65217	1	1	-28,9	-27	0,19811	10,01	-31,413	-0,578
mcr-mir-9077	UGUCUCAAAUUAUCUGAGGGCUUUUGCACUGCUGCUGAAAAUUUCUGCUGAUAUGCAGAAGUCAUAAAUAAAUGaguga	79	18,98734	27,8481	15,18987	31,64557	34,17722	59,49367	1,136364	1,25	-18,7	-18,3	0,09631	8,26	-23,6709	-0,69259
mcr-mir-9093	GCUGUCACGUAGAACCUGAUGGAUGUCACCUACGACUGUAGUCCAUGUCCCGGACCAGUGUGUGUCACAAUAGUUUCAAGUUCUUACAGGUGACUUU	97	23,71134	22,68041	23,71134	29,89691	47,42268	52,57732	1,318182	1	-25,6	-25,3	0,077213	8,56	-26,3918	-0,55652
mcr-mir-9-1	GGCGGCUCCGGUUUUGUCUUUGGUUAUCUAGCUGUAUGAUUGGGGCAGAAAUAUCAUAAAGCUAGGUUACCAAAGGCAAAAAUGGACGCCAAA	93	26,88172	29,03226	17,2043	26,88172	44,08602	55,91398	0,925926	1,5625	-45,5	-45,5	0,314517	3,87	-48,9247	-1,10976
mcr-mir-9186o	UGACUAAGAGGUGGCAUGGCUGGUGUUGGCAACAUUCCCUGACCUCUCUGAAAGCCUUGUGGUAUCCC	98	25,5102	18,36735	29,59184	26,53061	55,10204	44,89796	1,444444	0,862069	-29,1	-25,24	0,012852	15,39	-29,6939	-0,53889

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCCUGACCCAGGCAUGCCUUCUUCUAGCGA															
mcr-mir-9-2	uugaacagcacaggugacagGGGACAACCUUAACGUAUGCUUACUGUAaugcaaaaguaagcuccucUUUGGUUAUCCAGUAGUAUUGUGCUUGUUGAG	99	15,15152	13,13131	9,090909	23,23232	24,24242	36,36364	1,769231	1,666667	-27,4	-24,7	0,019343	8,03	-27,6768	-1,14167
mcr-mir-92a-1	cUGGCAUUAGCAGUAGGUCUUGAUAAGUGCAAUUCUGGUGAAUUAGAUAACAGAUUGCACUUGUCCCGGCCUUCUGCUAAUAGCUAA	87	22,98851	26,43678	18,3908	31,03448	41,37931	57,47126	1,173913	1,25	-43,2	-43,2	0,272477	2,43	-49,6552	-1,2
mcr-mir-92a-2	AUUUGUGUGCUCAGGAAAGGUUGUGACUUGGGCAAUUCUGUGAUGUCCUGUCAGAUUGCACUUGUCCCGGCCUUGUCUUGGCUUACACUU	90	26,66667	15,55556	21,11111	36,66667	47,77778	52,22222	2,357143	1,263158	-31,5	-29,1	0,036643	11,93	-35	-0,73256
mcr-mir-92b	CACCGAUCAGGGUUGUGUAGACCGGAACAGGGGCAAUGCAUUUUGAGUCGUCUGUAUUGCACUCGUCCCGGCCUAUCCAAUCUGACUCGUAG	92	27,17391	20,65217	26,08696	26,08696	53,26087	46,73913	1,263158	1,041667	-33,5	-31,7	0,067381	6,55	-36,413	-0,68367
mcr-mir-92e	ACCAAGCAUUUCAAAGAUCcacguggguguguggcgugcgugugcgugggcggugugggugggugcaugGCACACACACGUGGAUCUUUGAACUGCUUGAU	100	9	16	13	13	22	29	0,8125	0,692308	-55,5	-54,6	0,151057	12,27	-55,5	-2,52273
mcr-mir-9-3	UUUGGCGUCCAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUGCCCCAAUCAUACAGCUAGA	93	17,2043	26,88172	26,88172	29,03226	44,08602	55,91398	1,08	0,64	-34,1	-34	0,28369	3,42	-36,6667	-0,83171

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UAACCAAAGACAAAACCGGAGCCGCC															
mcr-mir-9358	UGAGGUACCCCGAGACAACUUUUUCGCUUUGACUAUUCGUUUGACAUGCUUGUUCCUGCAUUUUUUCUUCAAACGAGAACGAUGGCCCUGGGUACCUGU	99	21,21212	19,19192	24,24242	35,35354	45,45455	54,54545	1,842105	0,875	-26	-26	0,127101	8,99	-26,2626	-0,57778
mcr-mir-936	CAGCUCGCUGGAcaggagagggaggaaggggacUGGAACCUUCCUUUCCUCAACAUCUCUCCAGUUGAGGCC	72	13,88889	12,5	25	19,44444	38,88889	31,94444	1,555556	0,555556	-31,6	-29,2	0,090386	19,4	-43,8889	-1,12857
mcr-mir-9392	AAACACUGGCUUUCCUUCAUAUUAUGACUAGAUGAAAuucauuaaauguuuuucuagUCACAUUAAUUCACUGAAAGAAGUGGCA	85	11,76471	28,23529	14,11765	22,35294	25,88235	50,58824	0,791667	0,833333	-18,6	-18,6	0,301505	7,56	-21,8824	-0,84545
mcr-mir-943	ACUACAAGACAGGGAGAGAAUGUCAGACGACAAUGAAAUAUCUUUCGAAAUGUCUGCCAGUGAUUCUUUCUGACUGUUGCUGUCCGUUUUUUGUCUG	97	22,68041	25,7732	18,5567	32,98969	41,23711	58,76289	1,28	1,222222	-21,5	-12,9	0,033141	29,31	-22,1649	-0,5375
mcr-mir-9438	AGCCUGGCUACACAUCAUACUACACAAGUGUCUAGUGUGGCUACAAACAUUGCAAGCACCCCCCACUUGUCUACUCUGGGUGCAGUGCAGGGG	93	23,65591	24,73118	29,03226	22,58065	52,68817	47,31183	0,913043	0,814815	-28,8	-28,3	0,192015	6,09	-30,9677	-0,58776
mcr-mir-9505	UGGAUGAUGUCUGUCAAAAUCCAAUGGAGACAUAAGAGGCCAGCAAGCACACUGCCUGCUGACCUGCU	99	23,23232	30,30303	23,23232	23,23232	46,46465	53,53535	0,766667	1	-27,7	-27,5	0,050009	11,15	-27,9798	-0,60217

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CUUGGACUACAUGAUUACAGACACUAAUGUG															
mcr-mir-9542a	CGACUGACAAAACGGCUCACGACAUGGCCUGGCACUUUCUAAUUUCUGGCCAAGUUAGGCGCCUGUUUGUCGAGGGC	77	27,27273	20,77922	27,27273	24,67532	54,54545	45,45455	1,1875	1	-23,6	-19,5	0,032476	12,94	-30,6494	-0,5619
mcr-mir-9573	agaaaaaauaucaaaacuaGGAUUGUGAGGCUGUUGGUCCAGCCCUGCCAUCAUGGUGUUGGUUUUUUACC	71	22,53521	8,450704	15,49296	26,76056	38,02817	35,21127	3,166667	1,454545	-19,6	-17,5	0,079399	19,85	-27,6056	-0,72593
mcr-mir-9592	GGAUUGCUUUGCUGUUUGGGGCUGAUGCAAGGUUCCUUGCAGUGACGACCUUAUUCAGACAAAGAAACGG	70	30	22,85714	18,57143	28,57143	48,57143	51,42857	1,25	1,615385	-20,9	-20,4	0,264055	3,01	-29,8571	-0,61471
mcr-mir-9598	GAUAUUAUUGAUAUAAAACUGUCAGUGCUUAUUAGUACUAGGAUUUUGAUUAGCUGUUGUAAUUAGUGCUACAGCUUAUAUCAAGGUAAUCGU	93	19,35484	30,10753	10,75269	39,78495	30,10753	69,89247	1,321429	1,8	-21,6	-20,9	0,026556	30,6	-23,2258	-0,77143
mcr-mir-9640	UUCACAUGUCUAAGCAUGCUUUCGUGUUUCAGAUGGCAGAGCUGGACAAGGUGUCGCACCCUGAGAAGCCCUACCCGUACAGCAUGGCUGACAUGUUCA	99	25,25253	23,23232	26,26263	25,25253	51,51515	48,48485	1,086957	0,961538	-27,2	-22,7	0,044766	14,41	-27,4747	-0,53333
mcr-mir-965	GGGGGAGAAGUUAUUGCUGCGAAGCCUCAGCUUCUGCAAGCAGGAguuggucuucagcaugucacGAAUGUGCUCGCAGCUAGCGUCUCCAAC	93	24,73118	18,27957	19,35484	16,12903	44,08602	34,4086	0,882353	1,277778	-38	-36,4	0,037571	7,54	-40,8602	-0,92683

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-96b	CGACUGCUGGCUGUUCAAUUAUUUGGCACUUGUGGAAUAAUCGGUGUAUCUGAAAGUCGAUUAUACACCGGUGCCAAGUCAUCAGAAAUGCCUGAAGCAC	100	24	27	21	28	45	55	1,037037	1,142857	-33,2	-33,2	0,110762	3,59	-33,2	-0,73778
mcr-mir-9771g	CAAAACAGAGAACUACAUAUUUUACAUGCAAUGCUUCUAUUGCACUGGGcugucggaaaagucaugacgcAUUUUUGCAAUAUAUGGUGCUCGUUCAU	98	13,26531	23,46939	14,28571	27,55102	27,55102	51,02041	1,173913	0,928571	-19,7	-19,7	0,07496	11,01	-20,102	-0,72963
mcr-mir-981	GCUAUCAGACUUUGUUCCGGGUUUCGCGGCUGGCGAACAGUCAUUUUGAGCUGUGUUCGUUGUCGUCGAAACCUGCCCUGAAAUCUGAUGUG	92	28,26087	16,30435	22,82609	32,6087	51,08696	48,91304	2	1,238095	-35,5	-31	0,014511	9,61	-38,587	-0,75532
mcr-mir-9896	GACCUGAUGGAACAGACUAUAGAGCGAGUGCUAAUGAAGGCCAUUACAAUAAUCGGACACACCGCCAGGCCACUCGCACUGCACGUGCUCAAUCAGUCC	99	23,23232	30,30303	29,29293	17,17172	52,52525	47,47475	0,566667	0,793103	-26,4	-26,4	0,216116	8,18	-26,6667	-0,50769
mcr-mir-99	CUGGUGGGUCUUCACCCUCCUCAUCCUCUCCGUCUACACGGCCAAGCUCGCCUCUGUGCUGACGGUGGAGGUGCAGGGCGCGAAGAUCGACUCU	94	28,7234	13,82979	34,04255	23,40426	62,76596	37,23404	1,692308	0,84375	-40,8	-40,8	0,362029	6,15	-43,4043	-0,69153
mcr-mir-9934	UUGCUUAUCAGCGAUGCAUCUGAUGGUCUUGAUAUGUCCAUUUCUAUAUCAGAUCCUUAGGCUUCGUCUGGUUAAGGGA	79	22,78481	20,25316	18,98734	37,97468	41,77215	58,22785	1,875	1,2	-24,6	-22	0,079099	4,85	-31,1392	-0,74545

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
mcr-mir-994	UUGUUCAGUAAUGCCAUAAUuucuaagaaagaaaaaguaaauucuUGUAGCUACAUAAUCACAGUUGCUGUUUCUUAGUUUGUUACUGGUACUGAAUAU	99	13,13131	19,19192	11,11111	31,31313	24,24242	50,50505	1,631579	1,181818	-23	-20,7	0,065496	12,27	-23,2323	-0,95833
mcr-mir-9945	cccccucccccaguugACCACCUGUCCACCUGAGCCCAUAGACAUAGAGGGUUGGACAAGGUCAGGUAGCAAUGGAGAGGAAA	83	24,09639	26,50602	18,07229	12,04819	42,16867	38,55422	0,454545	1,333333	-27,9	-21,9	0,019816	14,79	-33,6145	-0,79714
mcr-mir-9b	UUGUGUUAUGAGUUAACAUAAAGGCUGUUAUGCUGUAAGAACUGCGACUAAAGCUGAAGCAACUAUUGUUAAGUCUUAACAUAU	84	20,2381	33,33333	13,09524	33,33333	33,33333	66,66667	1	1,545455	-19,9	-17,3	0,08065	9,04	-23,6905	-0,71071

Supplementary Table 3 – Structural and thermodynamic characterization of miRNA precursors in *P. canaliculata*. Size (Tam), G content, A content, C content, U content, GC content, AU content, AU ratio (R-AU), GC ratio (R-GC), Minimum Free Energy (MFE), Minimum Free Energy of the Pool (MFEE), Frequency of the MFE Structure in the Pool (Freq), Diversity of the Pool (Div), Minimum Adjusted Free Energy (AMFE), Minimum Free Energy Index (MFEI).

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-bantam	ACGAAACUGGUUUUCACAGUGAUCCAGCAGAUUGCUCAAAGUCUGAGAUCAUUGUAAAAACCAAUUUUGU	70	18,57143	32,85714	18,57143	30	37,14286	62,85714	0,913043	1	-29,4	-29,4	0,496236	1,25	-42	-1,13077
pcn-let-7b	UCAUACUGUGACUAGAUCAUAUCUCAUUGAGGUAGUGGUUGUAUCGUAUUACUCUCUCUGUAAUCAGUUGUCAGUCACAUAAAG	84	19,04762	26,19048	17,85714	36,90476	36,90476	63,09524	1,409091	1,066667	-18,7	-18,7	0,092975	12,14	-22,2619	-0,60323
pcn-let-7d	cugcugcugcugcugcuguaugUGCCCAGAGAAGACGUCUGCCUCAGACGUUGUGGCUGCACUAUACAACCUGCUGCGGCAGGCA	85	21,17647	16,47059	22,35294	14,11765	43,52941	30,58824	0,857143	0,947368	-35,8	-29,3	0,041106	14,27	-42,1176	-0,96757
pcn-let-7f	GGUCUGCACAUUCCACCAUCCAUUACUGUAGGUAAUAUUGUGUCCUCUUUGAGAGCCUAUAUCAGCAAAGCUACGGUAAUUGCUGGUAGUGGCAGUCA	98	22,44898	24,4898	22,44898	30,61224	44,89796	55,10204	1,25	1	-28	-20,9	0,057504	32,52	-28,5714	-0,63636
pcn-mir-1	AUUAUUACAUAUCUGGUAUGUUUGUGGAGUUGAUGUGUUCAUGCUUCUUUAUAGUUUGACCUUUUCCCUCacacaacauaaagaugagaaaUUCU	95	13,68421	12,63158	12,63158	38,94737	26,31579	51,57895	3,083333	1,083333	-18,5	-17,2	0,045655	8,13	-19,4737	-0,74
pcn-mir-1002	CACUGCAGGUGUAAAGGUUAUGUACAGGUCAGUGCCGAGACUGAGAAUAUUGUACAGCUAAGUAGU	92	22,82609	29,34783	13,04348	21,73913	35,86957	51,08696	0,740741	1,75	-21,4	-20,5	0,064133	16,07	-23,2609	-0,64848

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UGAUACAAACAGUAacaccagcaaaa															
pcn-mir-10a	CAUGUCGGUGCAAGACCUACCCUGUAGAUCCGAAUUUGUGuaauauuauuaacaacaaauuCGUGUUUGCACGGUAAGGCCUCGCACGUGACGCU	95	22,10526	14,73684	21,05263	20	43,15789	34,73684	1,357143	1,05	-36,3	-36,2	0,024951	6,41	-38,2105	-0,88537
pcn-mir-11	CAAACACCAUUCAGCAUCAUGUUCAUCACAGUCAGAGUUCACUGACUAACUGUGAGCAAAGCUCUGCUGGUGUCGA	76	19,73684	28,94737	26,31579	25	46,05263	53,94737	0,863636	0,75	-23,7	-23,7	0,279042	5,58	-31,1842	-0,67714
pcn-mir-1175	UGAUCAGGUGUGUUAUGUAGUGGAGAGAGUUUUAUCUCAUCAUGAGCCUCAGAAGCAGGUGAGAUUCAACUCCUCCAACUGCAGGCUAUACCUGAAAC	98	24,4898	27,55102	20,40816	27,55102	44,89796	55,10204	1	1,2	-42,7	-38,6	0,079856	5,62	-43,5714	-0,97045
pcn-mir-1187	GCGACUACAACACCACCUUAGUCAACACACUGCACCAGCCCCGGGGUGGCUGGGGGACGUAGGGUGggaguuuauauauauguguguguguaugugugug	100	21	15	21	9	42	24	0,6	1	-30,8	-25,4	0,029578	28,62	-30,8	-0,73333
pcn-mir-12	UGGUGGUCAGACUGUGAGUAUUACAUCAGGUACUGAGAAUCUAACAAGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUU	85	21,17647	24,70588	17,64706	36,47059	38,82353	61,17647	1,47619	1,2	-41,7	-40,3	0,196235	2,83	-49,0588	-1,26364
pcn-mir-122b	GGUCAACAGUAGUGUGACACUGGUGUGACAGUCGUGCCUGUCGUCACACAGGCCAGGGCCUAUACCC	91	26,37363	21,97802	29,67033	21,97802	56,04396	43,95604	1	0,888889	-27,9	-27,4	0,053765	26,46	-30,6593	-0,54706

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UGUCAUCACACAAUCCGCUUGCAG															
pcn-mir-1238	GACAGGGCGCGAGGGCCAUCGACUGGGUGUCUUCCUCGUCUGUCUGAAGCAGUGAUGCUGCGGGGAAGUCUGGUCAGUCGGAGGCUCUCUCUGGCCUUUA	100	36	14	25	25	61	39	1,785714	1,44	-49,3	-47,6	0,09807	9,54	-49,3	-0,8082
pcn-mir-124	UACCUGUUUGUGUUCACUGCGUUGGCCUUGGUGAUUUACACACUAUUAAGGCACGCGGUGAAUGCCAAGGGGAA	74	28,37838	21,62162	20,27027	29,72973	48,64865	51,35135	1,375	1,4	-33,9	-32,5	0,275153	6,77	-45,8108	-0,94167
pcn-mir-124c	UCCCCUUGGCAUUCACCGCGUGCCUUAAUAGUGUGUAAAUCACCAAGGCCAACGCAGUGAACACAAACAGGUA	73	20,54795	30,13699	28,76712	20,54795	49,31507	50,68493	0,681818	0,714286	-19,3	-17,4	0,083313	8,41	-26,4384	-0,53611
pcn-mir-1277-1	AAGACUCUAUGGUUAAACUAcgaugguauauauauauauguacguCUAUACGUUUCUACAAUGUAUAUUUGCCUUGUUCAUCCAUGCGUGAC	92	10,86957	19,56522	15,21739	27,17391	26,08696	46,73913	1,388889	0,714286	-19,4	-19,4	0,182954	5,41	-21,087	-0,80833
pcn-mir-1277-2	UGUAUAGAUAGUGUAUUUAUCUGUAUUUCUGCGUGUGUUCUUGAGUCUUUGCCUCUUUUaggcaaaaaaaauauauauauauauguaugau	91	13,18681	8,791209	8,791209	34,06593	21,97802	42,85714	3,875	1,5	-21,1	-21,1	0,15848	5,1	-23,1868	-1,055
pcn-mir-1322	UCUCUUACCUGcgaugaugcugcugaugacaaGGGGUAGAAUGGCAGCAUUCAUCAUCCGCAUGUACAGCUC	72	16,66667	15,27778	19,44444	19,44444	36,11111	34,72222	1,272727	0,857143	-19,5	-15,5	0,030502	11,17	-27,0833	-0,75

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-1328	gagagagagagaaaugagaaacaaauauuucgCUAGAGCUACAACAGUUUUACGAGUGCUGCUAUUUGCUGUUCACAUUGUCUCCUCUUAU	91	10,98901	13,18681	15,38462	25,27473	26,37363	38,46154	1,916667	0,714286	-22,2	-17,7	0,038242	22,78	-24,3956	-0,925
pcn-mir-133	AAUGCUCUUCUAUAGCUGGUUGAACUCGGGCCAAAUUGUUGAAGAGCCUUUCAUUUGGUCCCCUUCAACCAGCUGUAGUUAGCAGU	86	22,09302	22,09302	23,25581	32,55814	45,34884	54,65116	1,473684	0,95	-39,7	-39,3	0,1295	7,69	-46,1628	-1,01795
pcn-mir-133c	UAACUACAGCUGGUUGAAGGGGACCAAAUGAAAGGCUCUUCAACAAUUUGGCCCGAGUUCAACCAGCUAUAGAAG	75	24	33,33333	21,33333	21,33333	45,33333	54,66667	0,64	1,125	-30,4	-28,9	0,184949	4,61	-40,5333	-0,89412
pcn-mir-137b	AGUUCCAGUCGGUUACGGGUAUUCUUGGGUAAAUAAUACAUUGAGUAGUUGUUAUUGCUUGAGAAUACACGUAACUACCUGGAC	84	23,80952	27,38095	15,47619	33,33333	39,28571	60,71429	1,217391	1,538462	-33,1	-29,3	0,039071	10	-39,4048	-1,00303
pcn-mir-13b	uaaaauacacuuuaacUGACGUGUGGCUUCUGAUGUCUCUGAAGGGUGCGAGAGUUUGUUGUUCAGACUCAUCAAGCCUcguuaaaauguuugug	95	20	11,57895	12,63158	22,10526	32,63158	33,68421	1,909091	1,583333	-24,2	-23,8	0,019635	15,65	-25,4737	-0,78065
pcn-mir-1421a	CUCCUUCAGCAGUGAUUCAGUCUCACGCUGCAUCCCUUGUGACGGUGGCUGCGAGACUGAGACACAUGGCAUAAAGACA	79	25,31646	24,05063	27,8481	22,78481	53,16456	46,83544	0,947368	0,909091	-23,9	-23,9	0,016513	12,42	-30,2532	-0,56905
pcn-mir-1421am	GGUGCGUCAAGGCGGUCAGUAAGACGUCCGCCACCUUAGGCGUUCAAACGGAUUUCGAGGAGGUCGU	96	34,375	19,79167	23,95833	21,875	58,33333	41,66667	1,105263	1,434783	-31,8	-26,1	0,047723	21,41	-33,125	-0,56786

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AGGUCAUCCUGUAGACGACUUGGCGCUGG															
pcn-mir-143	ACGACAGAGGGCCUCAUCAGUCUGGCUGAACACAAGAAGUAGAAAGUGUUGCACACAGUCUGAGAUGAAGCACUCGUUUG	80	27,5	31,25	21,25	20	48,75	51,25	0,64	1,294118	-22,9	-21,7	0,179141	6,12	-28,625	-0,58718
pcn-mir-153-1	CUGUUGAUCACAACACCAGAUAAAUUACAGGAAACAGAACAUGUAACACGAGUAACAUGUUUACGCAUAUCAUUUUUGUGAUGUUAUCAAUAA	93	15,05376	38,70968	17,2043	29,03226	32,25806	67,74194	0,75	0,875	-19,2	-19,2	0,260282	8,93	-20,6452	-0,64
pcn-mir-153-2	AUACCCUCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGCUAUCAAAUUGCAUAGUCACAAAAGUGAUCGGGAGGCGGGGAC	87	24,13793	27,58621	22,98851	25,28736	47,12644	52,87356	0,916667	1,05	-37,9	-37,9	0,256161	6,4	-43,5632	-0,92439
pcn-mir-1584	GAGUUACAAUCAUAACCGGGUGGGGCUGGGCCAUAUAAGgguagaucacgugaccacucGGUGAUUAGUUAGUAAGCA	78	24,35897	21,79487	10,25641	17,94872	34,61538	39,74359	0,823529	2,375	-20,5	-16	0,070486	11,15	-26,2821	-0,75926
pcn-mir-15c	UUGAAGAAUAUCUCUUAUCCCAUAGGACAGAUAUUUCAAGGAAUCAUGUGACCAGCAGACCAUUCUGGGCUAUGGGGUCAGAGCUAUUCUUUUU	94	21,2766	27,65957	19,14894	31,91489	40,42553	59,57447	1,153846	1,111111	-32	-28,3	0,131794	8,15	-34,0426	-0,84211
pcn-mir-1602	AUCCUAUAUCCAUGUAUCUAUCCUGUAAUGGGCUCUGCAUCACCAGCAGUCUAAUCUGUCUUCAGUA	96	18,75	27,08333	23,95833	30,20833	42,70833	57,29167	1,115385	0,782609	-25,3	-25,2	0,141519	11,45	-26,3542	-0,61707

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CAUGGCAGGUUAGACAUGAAUCUGAGCAA															
pcn-mir-1603	AUAGCAGAAAUCAAAACAGCCCACAUUUUAUGUCAAACUUAUUACACAGAGAUAUUAAAGCAAuggugguuguuuuguguugcgCA	86	8,139535	34,88372	15,11628	17,44186	23,25581	52,32558	0,5	0,538462	-20,1	-20,1	0,033978	10,39	-23,3721	-1,005
pcn-mir-1632	GAUGGCUGUUGACACACUCGAGUGACAAGACAAAGGUCUGAUUUCCUUGCUUGUUUUUGGAUGAAAUUAUUUUGCGACGCAGCUGUUCACAGCCUCU	97	23,71134	22,68041	20,61856	32,98969	44,3299	55,6701	1,454545	1,15	-26	-26	0,016941	14,04	-26,8041	-0,60465
pcn-mir-1677	UGACCAUCACCUUUAUCUCUCUGCCGUCGAGGUAGCGCGACACGAACUCCACUCCAUUCUGUGCGUUGACUUCAAUAGGAGAGACAGGAGUGAUGGUGA	99	25,25253	23,23232	26,26263	25,25253	51,51515	48,48485	1,086957	0,961538	-36,2	-35,3	0,026404	8,83	-36,5657	-0,7098
pcn-mir-1744-1	UUUGAGAAGAGGUACUUCAACAGGAGCAAGUCAAGGCAUGCUUCUUUUUAACACCAGUCUUCUCUGC	67	20,89552	26,86567	22,38806	29,85075	43,28358	56,71642	1,111111	0,933333	-19,2	-19,2	0,273743	3	-28,6567	-0,66207
pcn-mir-1744-2	CCACACACUUCAACAGGAGCAAAGUGUAAGAUGAUAAUGCUGUGCCUACAUGUUAGCCUCUUACAUCAUGUGGGCUAUCUGAUACUAAGUGUGGCC	96	21,875	28,125	22,91667	27,08333	44,79167	55,20833	0,962963	0,954545	-27,6	-27,6	0,347188	6,28	-28,75	-0,64186
pcn-mir-184	CAGUUCACCUUUUUCACGUUUCCUUGUCACUCCUCCGCCCCGUCUGUGAACAAGCACUGGACGGAGAAC	97	23,71134	20,61856	26,80412	28,86598	50,51546	49,48454	1,4	0,884615	-33,8	-33,8	0,131517	7,97	-34,8454	-0,6898

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UGAUAAGGGCUUGUGGGUGAUUUGACAA															
pcn-mir-190	ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGACCAAGUAGUCAAACAUGUCAGAGCAGCGACG	82	25,60976	25,60976	21,95122	26,82927	47,56098	52,43902	1,047619	1,166667	-32,5	-32,3	0,080048	9,84	-39,6341	-0,83333
pcn-mir-1905c	UGUAGGUGUGUGUCUGUUGCUGGAUGCGCUUGAUGGUCGCCCCCUUGGGUCCCACCACCAGCCCCACCACGCGGUAUGGCACAGCCACCC	90	28,88889	13,33333	35,55556	22,22222	64,44444	35,55556	1,666667	0,8125	-31,8	-31,7	0,089456	8,15	-35,3333	-0,54828
pcn-mir-1951	UUAGUUUUGCGCAUCAAUCACCAUCAUUUGUGUAAACGUCUUCCCUGACUUAUUUGGAGCCCCCAAAUGUAGUGGAGACUGGUGGCGAAACCUU	94	21,2766	23,40426	23,40426	31,91489	44,68085	55,31915	1,363636	0,909091	-26,2	-17,5	0,079573	16,28	-27,8723	-0,62381
pcn-mir-1976	ACUCUCCUGCCCUCCUUGCCCAGACAGGGGCAGCGUGUCCAUGACCUGUCUGUGCGAUCAGCUGCACGAGGGC	73	28,76712	15,06849	35,61644	20,54795	64,38356	35,61644	1,363636	0,807692	-28,7	-28,7	0,157786	6,45	-39,3151	-0,61064
pcn-mir-1984	ACACUGGCACUGUCGCCGUGCCCUAUCCGUCAGGAACUGUGAUCUCGCGAAACACAGGGUCUGGCGGUUGGGCCUCGGCGCUGGUGGUCcagacg	95	31,57895	13,68421	28,42105	20	60	33,68421	1,461538	1,111111	-47,8	-47,8	0,195263	3,22	-50,3158	-0,8386
pcn-mir-1985	AUGCCAUUUUUAUCAGUCACUGUGUGUUGUGAAAGucacagugaugaugauaauggcuUGAUG	63	15,87302	14,28571	7,936508	25,39683	23,80952	39,68254	1,777778	2	-27	-27	0,311248	2,46	-42,8571	-1,8

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-1986	CGUUUGUGGCCGCGGGUCAUGGGAGCGCCACGUACAAUCAAAGCGGUGGAUUUCCCAAGAUCCGUGAUCGCACUAC	76	30,26316	21,05263	27,63158	21,05263	57,89474	42,10526	1	1,095238	-33,4	-33,1	0,171582	3,66	-43,9474	-0,75909
pcn-mir-1990	GUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCUACCCCUCCCGGGACUACGUCAACGUACUACCACGC	72	26,38889	20,83333	30,55556	22,22222	56,94444	43,05556	1,066667	0,863636	-30,6	-30,6	0,28064	3,02	-42,5	-0,74634
pcn-mir-1992	GCAUGUGUCCAGUCAGUGGUUGACUGCUCGGUAUUCUAAACGUAUCAGCAGUUGUACCACUGAUUUGCUACGUG	74	25,67568	20,27027	21,62162	32,43243	47,2973	52,7027	1,6	1,1875	-32,7	-31,7	0,194317	3,34	-44,1892	-0,93429
pcn-mir-1994	CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUGUGUGAUGACGUCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGACACCGCUAA	90	30	18,88889	25,55556	25,55556	55,55556	44,44444	1,352941	1,173913	-41,1	-41,1	0,501637	4,6	-45,6667	-0,822
pcn-mir-1994a	UUUCUCAGGGGCGGUCACUCUGAUCUCCAUGUGCUCGCACCAGCAUGAGACAGUGUGUCCUCCCUUGAGGGA	72	27,77778	16,66667	29,16667	26,38889	56,94444	43,05556	1,583333	0,952381	-36,5	-36,2	0,349274	3,61	-50,6944	-0,89024
pcn-mir-1996b	UAUUCUGACCUUAUUUGAUAAUGACCAGUAUUUCCUUUCGUCGUCCUCUUGUAGACUGAUGCUGAUGCUUAACUGUCAUUAUCAAGUGAGGUCAGAAUA	99	18,18182	24,24242	19,19192	38,38384	37,37374	62,62626	1,583333	0,947368	-42,2	-42,2	0,071127	11,72	-42,6263	-1,14054

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-1a	GGCACAUACUUCUUUGCUAUCCCAUAUGUUCUUGCAAAGCUAUGGAAUGUAAAGAAGUAUGUACU	65	18,46154	29,23077	18,46154	33,84615	36,92308	63,07692	1,157895	1	-23,7	-22,1	0,075024	6,51	-36,4615	-0,9875
pcn-mir-1c	AGUACAUACUUCUUUACAUUCCAUAGCUUUGCAAGAACAUAUGGGAUAGCAAAGAAGUAUGUGCC	65	18,46154	33,84615	18,46154	29,23077	36,92308	63,07692	0,863636	1	-27,1	-26	0,4687	2,23	-41,6923	-1,12917
pcn-mir-2001	GCUGCCAUUUUGUGACCGUUACAAUGGGcauugacagaaaaacaaaugcuCCAUGUGGUGUUCACAAGAUGGCGGU	76	22,36842	13,15789	14,47368	21,05263	36,84211	34,21053	1,6	1,545455	-36,2	-36,2	0,193282	4,14	-47,6316	-1,29286
pcn-mir-2008	AGGUUAAUUAUAGUUUGUGUCCUCACUUGAAGGCACACUUCCAGUGUGAGAGUAUUAACAAUGUGGCUACAGUGAUGUGACAAAUAUUGUUAAUAC	96	21,875	30,20833	14,58333	33,33333	36,45833	63,54167	1,103448	1,5	-24,3	-24,3	0,153115	8,97	-25,3125	-0,69429
pcn-mir-203b	ACUUGUGAAGUUACCACCCAACAAAUAUUAGUUUGGGUCAAAGAAUUGAAUAUUUGAACUGUUAAGAACAGUAAAUUAGUUGCUAAUGGAACUUCCAUUU	100	17	36	14	33	31	69	0,916667	1,214286	-20,1	-18,9	0,016531	14,54	-20,1	-0,64839
pcn-mir-204	GUUCUUUGUGUCACCUCACUUCCACUUCCGGUGAAAUCCGGAUUGGUAAUGCAAGGACAGCAAAGGGA	68	25	25	23,52941	26,47059	48,52941	51,47059	1,058824	1,0625	-21,9	-21,9	0,137453	4,4	-32,2059	-0,66364
pcn-mir-2047	CCAAAGCCUAUGUCACGUCUCUCAGAGAUAAAGGACGUCAGCAGUGUAUUACGCCCUUUCGCCGAGCAGAUGUGUGAAGGUUAAG	85	25,88235	27,05882	23,52941	23,52941	49,41176	50,58824	0,869565	1,1	-23,3	-22,2	0,082417	6,14	-27,4118	-0,55476

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-2155	GUGUGCUCUGGUGCUCUGUGAAUGUAAGUUCUUUGUGGUAGAAUGAAGUGACACUGUUUUACACUCUACUGGAGCCUCAAGCAAAG	86	26,74419	23,25581	17,44186	32,55814	44,18605	55,81395	1,4	1,533333	-20,5	-17,1	0,059402	25,69	-23,8372	-0,53947
pcn-mir-216a	uguuggguguuugucUAAUCUCAGCUGGUAAUUCUGAGUGGAGUUCUGCACACCUCAAGUUACUAGCCGAGAUUACAUAAAUAUCGUA	87	16,09195	25,28736	17,24138	25,28736	33,33333	50,57471	1	0,933333	-34	-34	0,097375	3,66	-39,0805	-1,17241
pcn-mir-216b-1	GCGGGACCACUUGCCCUGUGGAGCGAGGAUGCUCUGGAGGACGGCUGGACCUUUAUUAAAACUCGCCUCUACACAGUGGCUACUGGGUCCACA	93	30,10753	20,43011	27,95699	21,50538	58,06452	41,93548	1,052632	1,076923	-34,9	-29	0,081319	17,93	-37,5269	-0,6463
pcn-mir-216b-2	CGUAGGAAGAUGAGCCAGGAUUACAAGAUUGUUGGGCCUGCGGCCCUAAUAUCAGCUGGUAAUCCUGAGUGAGCUUGCCUCAG	83	30,12048	24,09639	21,68675	24,09639	51,80723	48,19277	1	1,388889	-31	-30,7	0,290814	5,92	-37,3494	-0,72093
pcn-mir-219-1	GUUGUCUGGCUGUGAUUGUCCAAACGCAAUUCUUGUGAAGUUUAAAUCAAGAACUGUGUGUGGACAUCAGUGCUUGA	77	25,97403	24,67532	15,58442	33,76623	41,55844	58,44156	1,368421	1,666667	-27,4	-27,4	0,170426	3,13	-35,5844	-0,85625
pcn-mir-219-2	GUGAUUGUCCAAACGCAAUUCUUGUGAAGUUUAAAUCAAGAACUGUGUGUGGACAUCAGUGCUUGAG	67	25,37313	28,35821	14,92537	31,34328	40,29851	59,70149	1,105263	1,7	-21,9	-21,9	0,222542	3,22	-32,6866	-0,81111
pcn-mir-2196	CAAGGUCAACUGCAAGAAUUUGCCUCUCUGUGCUGCCAGCAAAAAU	88	20,45455	32,95455	25	21,59091	45,45455	54,54545	0,655172	0,818182	-21,1	-14,7	0,0689	11,59	-23,9773	-0,5275

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UCUGAGCGCUCUGCAAGACAAAUACAAGCAUGUUGAACCAAG															
pcn-mir-2218b	aaaaauaauaauaaaguacauaUUUGGGUGGAGGCAUGGUCUAGCGCCUUGGGUUGGCGUGGGAACAUUUGGGAAAAAUUUGUAGUUUGUAUUCUUCGG	99	28,28283	13,13131	9,090909	27,27273	37,37374	40,40404	2,076923	3,111111	-21,1	-19,4	0,05187	8,22	-21,3131	-0,57027
pcn-mir-2284c	uaaugauaagCUGUACAUGAAACAGCUAAAAAUCUAAGUGAACUGUGACGGUGGAAAAUUUUACAUGUUGCACAGUCAUCUUGCUCAGCA	90	17,77778	30	16,66667	24,44444	34,44444	54,44444	0,814815	1,066667	-19,4	-19,4	0,041642	12,72	-21,5556	-0,62581
pcn-mir-2284t	UCUCUUUGAUUGAAAUCCCUCUUGAUCCAUGUCAAUAAAACUCGAAUGAAUGUAGUUCGUAGGACAGGCUCACUGUCAAAGAAAGUUACAAUCUAAGACC	100	17	33	21	29	38	62	0,878788	0,809524	-19,3	-17,8	0,046921	9,84	-19,3	-0,50789
pcn-mir-2324	AAACACGUGACUAACGUCGUUGACCUGGGGUUGGGGCAGUGACCAGCAGGUGUCGUCUGCUAGGCUUCAAAGCCGUUAGACCGACGUGGGC	91	34,06593	20,87912	24,17582	20,87912	58,24176	41,75824	1	1,409091	-29,7	-20,7	0,058231	28,39	-32,6374	-0,56038
pcn-mir-2392	AUCCCCUCUCACUCCCGGCCCUCGUUACGCCUUAUGUUAGCAUCCUGUCUCCUCUUUGCCUACAGGACUUAGUGAACGAUGGAUGGGGGUGAGGGGGAUG	100	27	16	29	28	56	44	1,75	0,931034	-43,6	-43,1	0,044553	14,74	-43,6	-0,77857
pcn-mir-2405	UUGCAGAAAACUGCAACAACACACAAGGCUUGUAUAUAUAUGGggu	74	10,81081	22,97297	10,81081	13,51351	21,62162	36,48649	0,588235	1	-20,3	-20,3	0,060748	10,16	-27,4324	-1,26875

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	agauugugugugugguuuguuuggu u															
pcn-mir-2464	GAAUGUUGUCUGGCGUCCucgacaucaacaucaacaucaacaucagcuUCAUCAGCUAUUGACUUCUGCUGCAGACCUCAAGGAUGCcagaaacaaga	98	14,28571	11,22449	15,30612	17,34694	29,59184	28,57143	1,545455	0,933333	-27,8	-26,8	0,142155	8,83	-28,3673	-0,95862
pcn-mir-2470	GCCAGAAAAACCUAAGCAUGACAAAAUAUCAUGCUUGUGUGGUUUUGGAUACUUGGAUUUUUCUCCC	67	19,40299	28,35821	19,40299	32,83582	38,80597	61,19403	1,157895	1	-19,9	-19,9	0,542669	2,57	-29,7015	-0,76538
pcn-mir-2491-1	GCAGUUGUCUUCUUUUGCAUGUUCUGUGUUCGUAAAAUCGCUUAUCAAUGAUGUAGUCGGUagccaacaacagcagcagcaacaacaacaacggc	95	14,73684	11,57895	10,52632	27,36842	25,26316	38,94737	2,363636	1,4	-23,2	-23,2	0,178642	4,92	-24,4211	-0,96667
pcn-mir-2491-2	GAUGGCGGCGAAGAAGUCCACGCcugaagaagaacaacaacagcagcagcaacaacaaggGUGGCGAGGGCUGCUUCGUCACCUGU	86	22,09302	9,302326	15,11628	10,46512	37,2093	19,76744	1,125	1,461538	-27,25	-26,3	0,182896	6,2	-31,686	-0,85156
pcn-mir-2491-3	agcagcagcagcagcagcaacaacaacagcagcagcagaacaGGCAAGGUAAUGGUGAUUGCUCUGCAUUUUUGUGUACUUCUGAUCUCCUAAA	94	12,76596	11,70213	9,574468	21,2766	22,34043	32,97872	1,818182	1,333333	-19	-15,7	0,14487	14,83	-20,2128	-0,90476
pcn-mir-2500	GAUCAUACAUUAAAGUGUUCAUAUGUAAUGUAAAGGGAAAAGGUGUAAUGUCUUCCCUUAACAGACAAGAGGAUUUUGUGUGUGGAC	87	24,13793	33,33333	11,49425	31,03448	35,63218	64,36782	0,931034	2,1	-24,7	-24,1	0,053178	5,65	-28,3908	-0,79677
pcn-mir-252a	CCUAACUUCUGGCCCAUUACUAAGUACUAGUGCCGCGGGAAGAUAACUUGUCAUGUCCCGUGGUUCU	94	24,46809	21,2766	26,59574	27,65957	51,06383	48,93617	1,3	0,92	-37,6	-37,6	0,059589	5,1	-40	-0,78333

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GGUCCUUACUACAGGGCGGAACUUGCA															
pcn-mir-254	GUGUGUGCAAAUCUUUUGCAACAGACUUUUAAACUAUGGGUGUUUGGUGUUUGAGUGUGCAGAGUUGUUUGCAAACUCUUUGCUACAGAA	90	25,55556	23,33333	13,33333	37,77778	38,88889	61,11111	1,619048	1,916667	-21,5	-16,1	0,016973	23,25	-23,8889	-0,61429
pcn-mir-279-1	UCUCUUAUCUUUGCGGGUGGCUGUGAGUCUAGUCCAUGUGACACGUCAAGCUCAUGACUAGAUCCACACUCAUCCACAAGGAUAGGACGUUCAC	94	22,34043	23,40426	26,59574	27,65957	48,93617	51,06383	1,181818	0,84	-39,9	-38,8	0,123884	5,39	-42,4468	-0,86739
pcn-mir-279-2	CUCUCUUAUCUUUGCGGGUGGCUGUGAGUCUAGUCCAUGUGACACGUCAAGCUCAUGACUAGAUCCACACUCAUCCACAAGGAUAGGACGU	91	23,07692	23,07692	26,37363	27,47253	49,45055	50,54945	1,190476	0,875	-39,9	-38,8	0,126379	5,36	-43,8462	-0,88667
pcn-mir-279b	AAGGGGACCAACCACUAGAGUGACUAGACUGAUAACCCCUAGAGAUGUUGUGCUUCAGUUAUCACUAACCAUGGUGGUGGAUCCCUGA	88	25	28,40909	22,72727	23,86364	47,72727	52,27273	0,84	1,1	-27,8	-27,3	0,031686	14,17	-31,5909	-0,6619
pcn-mir-2804	UUUUUAAACCACAGUAACAGAUGACAUGGUGAACCAUGAUGAAGGGUAGUGUAUUACAAUACUGACCAAGUUUUCAGCUGUUUGCUGUGUUUGUAAAUA	99	21,21212	31,31313	14,14141	33,33333	35,35354	64,64646	1,064516	1,5	-25,6	-25,4	0,006063	12,93	-25,8586	-0,73143
pcn-mir-281	AAAUGACCUCACGCUGAAGGGAGCAUCCGUCGACAGUCAGAAAUGC	88	26,13636	29,54545	22,72727	21,59091	48,86364	51,13636	0,730769	1,15	-35,2	-35,1	0,187538	3,76	-40	-0,8186

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AGCACUGUCAUGGAGUUGCUCUCUUUACUGAAAAGGUCAAGG															
pcn-mir-2962	UCGUGCAGUGAGUCAGGUGCUCGUGACAGGGAUGCUGUGGCAGGUGUUGUGGGGCGUGACACAGGUCACUGUGCAUCCCCUGCCUCUCUCAUAC	94	34,04255	14,89362	25,53191	25,53191	59,57447	40,42553	1,714286	1,333333	-36	-36	0,402579	13,67	-38,2979	-0,64286
pcn-mir-297	AUGAACGAAUGAAUGUCUACAGUACUGUguacguguauguaugugugcauguauggaGCCUGGCAGGUAGAGAGAUGUAUAGAUGCAUUCAUUUUG	96	18,75	20,83333	9,375	20,83333	28,125	41,66667	1	2	-23,6	-19,07	0,041644	16,8	-24,5833	-0,87407
pcn-mir-29a	GACUGGGCCGGGAUGGAUGCUGGUCUCUUUUGGUGCUUAGAGUCAUUCCCUGUCUAGCACCAUUUGAAAUCAGUUUCUUGAACUGGCCA	89	26,96629	17,97753	22,47191	32,58427	49,4382	50,5618	1,8125	1,2	-36,3	-36,3	0,263044	3,27	-40,7865	-0,825
pcn-mir-29b	GGGAGGAGAGUGGGAGGCAGCGGGACACUGGUCUCGACUGGUGGAUAGAUGGCUGCUAUUCUAGCACCAUUUGAAAUCAGUGCUCUCGCUUGUUU	95	34,73684	20	18,94737	26,31579	53,68421	46,31579	1,315789	1,833333	-35,8	-30,9	0,043188	34,86	-37,6842	-0,70196
pcn-mir-2a-1	CAUCAGUUAUGACUGCUGAGUUCACAUCAAAGAGGCUGUGACUUUGUGAUGCUGUUUCAAAUCACAGCCUGCUUUGGUGAGCUCUUUGCUGUCAGAUUC	99	23,23232	21,21212	21,21212	34,34343	44,44444	55,55556	1,619048	1,095238	-40,8	-39,5	0,13114	5,78	-41,2121	-0,92727
pcn-mir-2a-2	AGGCAAAGCUAUGGUGCUGACCAAGUGACUGGGAUGUGUACCUUA	89	25,8427	25,8427	17,97753	30,33708	43,82022	56,17978	1,173913	1,4375	-39,8	-39,4	0,329351	1,72	-44,7191	-1,02051

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AUGUCAUAUCACAGCCUGCUUGGAUCAGUAUUAUGGUUUUGAAC															
pcn-mir-2a-3	AGUGCUAGGAUCUGCCCAUCAAAGAUGCUGUGAUGUGUUGAGUUCAGUCAUAUCACAGCCAGCUUUGAUGAGCUGGUCUAGCUUC	85	25,88235	22,35294	21,17647	30,58824	47,05882	52,94118	1,368421	1,222222	-40,5	-40,2	0,12355	3,87	-47,6471	-1,0125
pcn-mir-2a-4	ACAAAGCAAAUGUGCAUGGAGCAAUCAAAGUAGUUGUGAUGUGUUUGUUGACUUCAUAUCACAGCCAGCUUUGAUGAGCUUUUUCACAGUUUCUUGCA	98	21,42857	27,55102	17,34694	33,67347	38,77551	61,22449	1,222222	1,235294	-32,4	-30,6	0,111894	7,44	-33,0612	-0,85263
pcn-mir-2b-1	UGCAAGAAACUGUGAAAAAGCUCAUCAAAGCUGGCUGUGAUAUGAAGUCAACAAACACAUCACAACUACUUUGAUUGCUCCAUGCACAUUUGCUUUGU	98	17,34694	33,67347	21,42857	27,55102	38,77551	61,22449	0,818182	0,809524	-24,8	-24,8	0,336462	7,7	-25,3061	-0,65263
pcn-mir-2b-2	GAAGCUAGACCAGCUCAUCAAAGCUGGCUGUGAUAUGACUGAACUCAACACAUCACAGCAUCUUUGAUGGGCAGAUCCUAGCACU	85	21,17647	30,58824	25,88235	22,35294	47,05882	52,94118	0,730769	0,818182	-34,5	-34,5	0,219013	4,37	-40,5882	-0,8625
pcn-mir-2e	CUACUGGUAUAACUCAAUUAUCAAUCUCUUGAAUUCAGAGAUACCAACUUUGACUGAUGAGAUCUGAUACUGAAUAGCCAGAAA	84	15,47619	35,71429	19,04762	29,7619	34,52381	65,47619	0,833333	0,8125	-19,3	-19,3	0,294993	3,58	-22,9762	-0,66552
pcn-mir-2f	UCGGGUAGAUAAGCCAGCGAUGUGAGACUCAGACGAUGGUGUGACUGAGAGUUAGCAGACCAGUGU	96	27,08333	27,08333	20,83333	25	47,91667	52,08333	0,923077	1,3	-28,6	-28,6	0,066397	6,92	-29,7917	-0,62174

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AUGUAUAUCACAGCCAUGCUAAUCUCCUCU															
pcn-mir-3027	UCCAGGCCCCAGUCUUGCAUUUAUGAAACAUCAUUAGGAUUAUGCCUGGGUUAGUGGUGGAAUAUGAAAUGCAAGCUUUGUGCCUAAG	88	25	26,13636	18,18182	30,68182	43,18182	56,81818	1,173913	1,375	-25,5	-23,3	0,022525	10,35	-28,9773	-0,67105
pcn-mir-3050	AAUGAUCAGAUAUGAGAUCUUGAUAAACAUCAACUGACUGCUAAUUGGUCAUCAGGUCGAUGUAUGACCCAGUCCAAGAUGUGAUCUCU	89	20,22472	31,46067	19,10112	29,21348	39,32584	60,67416	0,928571	1,058824	-23,6	-23,6	0,62089	5,09	-26,5169	-0,67429
pcn-mir-3057	agagugacUGGCUGUUCUUGAGAGAUGUUAGCAAGUUAGUUCAAAAUGUUAUGGCGCGAACCAACAUCCCACAGGCCCAGCUGUCACAAA	90	21,11111	26,66667	21,11111	22,22222	42,22222	48,88889	0,833333	1	-24,5	-23	0,146061	9,33	-27,2222	-0,64474
pcn-mir-3071	AuaucaucaaaacaaauguGCAUAGUGUUACACAGAUACCAGGAUCAUGGAUUCAGCUGAUUUAGUGACUGaaacacuuucauuucuugAUGAAGC	96	15,625	19,79167	10,41667	16,66667	26,04167	36,45833	0,842105	1,5	-20,1	-19,8	0,178747	6,13	-20,9375	-0,804
pcn-mir-308	CGAGUGCGCAUGACAGCGCGCGCAGGAUUUGCUUGUUUUGAGCAACUCGGGUGGCGAUGUCCAUGACGCACAGC	74	33,78378	18,91892	25,67568	21,62162	59,45946	40,54054	1,142857	1,315789	-32,3	-32,3	0,25533	2,19	-43,6486	-0,73409
pcn-mir-3084a	UAUAUGUAUUGAAGGUUCAUUAGCCUUGUGGUCAGAGCACUCCAAUUUGGAGCAAAAGGUUACUGGUUCAAGGUCUGCUUGAACCUAUGACUAAAUaau	99	22,22222	27,27273	16,16162	31,31313	38,38384	58,58586	1,148148	1,375	-27,5	-22,4	0,08872	12,04	-27,7778	-0,72368

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-31	UCAAGGUGUGAAUCCAGGUUAGGCAAGAUGUUGGCAUAGCUGAGUGAACUCUAACAUCAGCUGUGUCGCAUGGUGCCACCAUUUGGGUUCUUGUGAAC	98	28,57143	23,46939	19,38776	28,57143	47,95918	52,04082	1,217391	1,473684	-39,3	-36,9	0,095943	6,94	-40,102	-0,83617
pcn-mir-315	UUGGCUCAUUUUUUGAUUGUUGCUCAGAAAGCCGGUGACUCAGCUGCGGCACUCGAGUAACAAUCAAAGCGUGACGCU	78	25,64103	23,07692	23,07692	28,20513	48,71795	51,28205	1,222222	1,111111	-33,4	-33,4	0,276248	3,37	-42,8205	-0,87895
pcn-mir-317-1	AAUAUGUGCACUGUGGGAAGGAUACCAUUGCUUGUUCACAGUGAAUUGGACAUGUGAACACAGCUGGUGGUAUCUUUUUUAAUGUGAACAUGAC	94	25,53191	27,65957	14,89362	31,91489	40,42553	59,57447	1,153846	1,714286	-40,1	-40,1	0,224017	2,93	-42,6596	-1,05526
pcn-mir-317-2	UCACUUAUUGGCAUGUACAGUGAACACAGCUGGUCACAACAUCCUCACAUACAGUUGUACAGUGAACACAGCUGGUCACAACAUCCUCACAUACAGUUG	99	17,17172	31,31313	27,27273	24,24242	44,44444	55,55556	0,774194	0,62963	-22,1	-22,1	0,047361	10,2	-22,3232	-0,50227
pcn-mir-3173	ACAUGUACUGUUUCUAGCUGAGCACAGAUACCUUUAAGCCAGAGUGCUCGGAGUGUUCUGAUGCUGAAGGAGGAAACAGGCUACAGAU	88	27,27273	28,40909	19,31818	25	46,59091	53,40909	0,88	1,411765	-29	-29	0,195168	5,51	-32,9545	-0,70732
pcn-mir-33	GUGUACUAUGGUAGUGCAUUGAGGUUGCAUUGCAUCAGCUGAGAAACAUGCAAUGCAUCUGCAGUGCAAAUAUCAUGGUAUGC	83	26,50602	27,71084	16,86747	28,91566	43,37349	56,62651	1,043478	1,571429	-38,7	-37,7	0,204831	5,09	-46,6265	-1,075

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-340	GAAUAAACAUAUAAAACGUCUGUGGGAUUAUAAAGCAAUGAGAGUUUCCGUUGUGUGUUGUUUAGAAAAUCCCACAUGUUGCAGuuuguuuaaau	95	20	29,47368	11,57895	27,36842	31,57895	56,84211	0,928571	1,727273	-21,5	-20,6	0,012485	17,74	-22,6316	-0,71667
pcn-mir-345	UCAGCGGCUCGUGGUGCAACAUUUGUUACCUGCACCUUUCUGCACUCGGCGGAGAUGAAGAACACGUGACGAGAACGUGCUGACCCCUAGUCCGCCUC	98	26,53061	20,40816	30,61224	22,44898	57,14286	42,85714	1,1	0,866667	-28,8	-28,8	0,301092	13,36	-29,3878	-0,51429
pcn-mir-3529	UGUGGCUGCAGCAAUCAGGGAAGAAUGUGAUUUAUUGUUAGUCAGCAUCAGAACAACAAAAUCACUAGUCUUCCAGAUCACGGCAGCCugg	91	21,97802	30,76923	20,87912	23,07692	42,85714	53,84615	0,75	1,052632	-30,8	-29,7	0,103281	5,67	-33,8462	-0,78974
pcn-mir-3547	ACAAUUAAUAGAAGCUAGACAAUGAUAGCGGGUGGCCAAGUGGGAAGAGGGGUGGAGGUGAAGCCACCGCAUACACACCCUGUCaacuuuuuaauauc	98	27,55102	27,55102	17,34694	13,26531	44,89796	40,81633	0,481481	1,588235	-22,6	-22,6	0,156123	13,36	-23,0612	-0,51364
pcn-mir-355	UGAcugauuuaucuguuuauuuguuuuagccUGAGUCGCCAGAGGAAAUGGAUGAUGAAUCUGAACAACCUAAAACAGCUGGCAAGAUAAUUCAGGCA	98	18,36735	27,55102	12,2449	13,26531	30,61224	40,81633	0,481481	1,5	-22,1	-22,1	0,017425	13,25	-22,551	-0,73667
pcn-mir-3609	CAAAGUGAUGAGUAAUUAAGGCGCAUUUGAAUUAUGGCUACCUCCAAUGAGAUUUAUAACACUAUA	92	16,30435	34,78261	17,3913	31,52174	33,69565	66,30435	0,90625	0,9375	-18,5	-17	0,066153	14,65	-20,1087	-0,59677

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AAUGCCAUAAAUUGUGCAUCUCUCAU															
pcn-mir-3680	GAAUGUCUUGCUCCUGGUGUGGGUGACUCACUCACAGGAUGUGGUGAGGACCUCGGGCAGCUAGCUUCUGCCgaucacacacaccaacaacacaaaa	97	24,74227	11,34021	18,5567	19,58763	43,29897	30,92784	1,727273	1,333333	-32,3	-32	0,065042	7,8	-33,299	-0,76905
pcn-mir-36b	UGAUGCGGUGAGUGUAAACCCUGGUCGGUGCUCACCAGCAACAUCACCGGGUAUACAUUCAUCCGCACCC	70	24,28571	22,85714	31,42857	21,42857	55,71429	44,28571	0,9375	0,772727	-30,8	-30,8	0,109108	5,78	-44	-0,78974
pcn-mir-3716b	CUCAUCAGUCGGCUAUUCGAUUCGAUGUUAUUUGAAAUAUCAAACAUUCGAUUUGAAGUGUCGCUGUAUAUC	72	18,05556	26,38889	18,05556	37,5	36,11111	63,88889	1,421053	1	-20,4	-20,4	0,086216	4,88	-28,3333	-0,78462
pcn-mir-3739	CAGUCCCCCUUUCCAUGUCGACCUCUGCAAGUUCAUUGCCAUGAAGGCAACGAAAGGUGcuugggagggggagagaguggAAAGGUGAGGAGGG	94	22,34043	20,21277	19,14894	15,95745	41,48936	36,17021	0,789474	1,166667	-39,3	-38	0,332678	4,7	-41,8085	-1,00769
pcn-mir-375-1	ACAGAACAUUUCGCUGCCCAAUGACCCGAGCCGCUCGUAGCAAGGCAUCUUCACAAGUUUUGUUCGUUCGGCUCGCGUUACCAUGGUGGCACUUGCAG	98	24,4898	20,40816	29,59184	25,5102	54,08163	45,91837	1,25	0,827586	-34,5	-34,5	0,452928	2,76	-35,2041	-0,65094
pcn-mir-375-2	GGGACUUAAUUCAUCGCGAGCAGAAAAGAACGCAACAUCUUGUUCGUUCGGCUCGUUCUGAAAAAGUGAC	70	24,28571	30	21,42857	24,28571	45,71429	54,28571	0,809524	1,133333	-20,3	-20,3	0,298895	4,19	-29	-0,63438

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-377	UUUCCAGAUUAACUAGAGGUUGCCUUUGGUGAGAGAAGUCAUAGCCGAUGUCUUCAUCCUAGGAAUUAGCUAGUCUGGUAG	81	25,92593	24,69136	17,28395	32,09877	43,20988	56,79012	1,3	1,5	-24	-24	0,093863	9,2	-29,6296	-0,68571
pcn-mir-3776	UCGAGCACGCUUCACCUUUGCUUUCUCUAUGCCUGAAAAGGAGGGGGGGAGAGAGauuuauaaacacauauauaagcagauaugagugAGCGUCUGUG	98	22,44898	13,26531	14,28571	16,32653	36,73469	29,59184	1,230769	1,571429	-25,2	-24,8	0,072592	27,27	-25,7143	-0,7
pcn-mir-3809	UUACUACUUCAACUUUUCGGUUAGUCGGUCGGUCAGUUGGUUAGAUGACUUUUUGUCAUUUAGCAACUCGGCACAGAUAAAGAGAUAGAUGAAUAGAUA	99	22,22222	28,28283	15,15152	34,34343	37,37374	62,62626	1,214286	1,466667	-24,9	-24,7	0,021265	13,08	-25,1515	-0,67297
pcn-mir-3831	CAGCUUGUGUGUCUCAGAGCAAACAUUGCUAUUUUUCAUAUCCAAUGUGGCUCCGAGAUGCAAAAGGUA	69	21,73913	27,53623	20,28986	30,43478	42,02899	57,97101	1,105263	1,071429	-23,4	-23,4	0,214068	5,17	-33,913	-0,8069
pcn-mir-3928	ACCAAGCAACUUUGUCUCAGACCUAUCAUUGAGAUUCACUAUGAAAUCAGUAUAUGAAGCUCUAAGGUUCAUCCCAGACAAGGCUGCAUUUUA	93	16,12903	32,25806	22,58065	29,03226	38,70968	61,29032	0,9	0,714286	-21,1	-19,7	0,138205	13,46	-22,6882	-0,58611
pcn-mir-3957	guugguguugguguuggugcugguguugguguugguccCAUAUCCAGAAGACGCACAGCACCUCACACUGACGCCGUU	78	8,974359	15,38462	19,23077	7,692308	28,20513	23,07692	0,5	0,466667	-39,7	-39,7	0,412668	1,21	-50,8974	-1,80455

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-3965	UGCUUAUCAGCCUGAUUCUGCUAACGCUGCAUCGUUGAAGUCAGCUAGCUGUGAUAGAAGGAAUAACAGGCUUGAUAAAAA	81	23,45679	30,8642	18,51852	27,16049	41,97531	58,02469	0,88	1,266667	-26,4	-26,4	0,24589	2,99	-32,5926	-0,77647
pcn-mir-4057	GGUCGGCAACUGACCUCCUGCAUCUCCAGGGUGUAGGAGAUGGGGGUGGCUUGGCGACUGUAGUAGGGCAGUUCCGCUG	79	39,24051	15,18987	22,78481	22,78481	62,02532	37,97468	1,5	1,722222	-33,5	-28,4	0,21803	20,3	-42,4051	-0,68367
pcn-mir-4066	aacccaucauGCAGAACAUGCCUUUAUCAUGUGUUAGCUAAACACUUGGUUAUCGUGCUCACACGAAGACAGUGUUAAACCAGUGUUUUUCAUGUGGGGA	100	21	24	17	28	38	52	1,166667	1,235294	-29,1	-22,3	0,120404	15,88	-29,1	-0,76579
pcn-mir-4070	GCAAGCAGGUUGUGAGCGACCACCacgcgucacgugacaccuaGUGACAGCUGUGUUGACGUCACAGGCAGCUGUC	76	25	15,78947	19,73684	14,47368	44,73684	30,26316	0,916667	1,266667	-25,8	-25,2	0,153161	8,62	-33,9474	-0,75882
pcn-mir-4185	CAUAAAGUAUCUAUGUUUAAUGCGCUAUUGUGUGAGUCUCCUUCUUUGUAUUCAUACUGUCUGUUACAAGCAAGCGGAUUAUUCAAAGAAUACUUUGCA	99	17,17172	27,27273	17,17172	38,38384	34,34343	65,65657	1,407407	1	-24	-18,6	0,091273	7,49	-24,2424	-0,70588
pcn-mir-4187	CUAGUGUGGUCAAUUAACAACUGUUUUGGUGUUGUGCUGUUUAACUUCACGUCGAAGUAAACAguauauuuucacacacacauc	84	16,66667	19,04762	11,90476	27,38095	28,57143	46,42857	1,4375	1,4	-21	-15,37	0,010024	18,73	-25	-0,875
pcn-mir-4195	ACCCUAGUUACAUGUAAUACAUUGUACCUUGGUUACAUGUAAUAC	85	12,94118	32,94118	20	34,11765	32,94118	67,05882	1,035714	0,647059	-22,2	-22,2	0,043533	6,38	-26,1176	-0,79286

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AUUGUACCCUAGUUACAUGUAAUACAUUGUACCCUAGAAA															
pcn-mir-4271	UACAGGUCACCUAACAUCAUUUGCCACAGUUUUUCUCUUAGAACUGUGGGGAGAGGUGGUAGGGGGAAGAAAAGGUGGCAGGUGUGGCAUGUGUCCUAAA	100	32	26	16	26	48	52	1	2	-35,9	-32,3	0,296371	32,35	-35,9	-0,74792
pcn-mir-4323	AUCUGCGGCUUCCUGCACUUAGAGGGGAGCGAUUUGCUUUUGACAGCCCCACAGCCUCAGGAGCAGAAAGCUGCAUAG	78	28,20513	23,07692	26,92308	21,79487	55,12821	44,87179	0,944444	1,047619	-28,2	-24,9	0,122046	7,91	-36,1538	-0,65581
pcn-mir-4472	UGCUCUGCAGCAACAAUCGACUAUUCCCACCCUCCUUUUUuacugggggggggugggggguguuGUUGAUCAGAAAU	79	8,860759	16,4557	20,25316	21,51899	29,11392	37,97468	1,307692	0,4375	-38,6	-38,6	0,308719	10,28	-48,8608	-1,67826
pcn-mir-449a	ACAACUGUACAGACUUUAGUCCAAUCAUCUAUACUGGCUUGUAUGUUUGACACCAGCUGACAUUCAGUCCAGAUGAUAAGCAGAUCUUGACAGUGUC	97	18,5567	28,86598	22,68041	29,89691	41,23711	58,76289	1,035714	0,818182	-22,3	-19,2	0,020241	9,91	-22,9897	-0,5575
pcn-mir-449d	CCUAUCAACCAUUGUCAGCAGUGAAACUAAUGGUAUUCAUUUGUCUUGAAGGCUGUGUGCUGUAAAGAUAAUGCUCACUGCUGCCCUCGUGGAUCUG	97	22,68041	23,71134	21,64948	31,95876	44,3299	55,6701	1,347826	1,047619	-25,6	-22,8	0,037152	9,97	-26,3918	-0,59535
pcn-mir-4525	AGUGCAUGCCAGAUAUCCCACCACGUCAUGCGCAGGUGUGCCUG	70	35,71429	18,57143	27,14286	18,57143	62,85714	37,14286	1	1,315789	-31	-30,8	0,214837	9,25	-44,2857	-0,70455

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GACCUGGGGGGGAUGUGCAUGCCAG															
pcn-mir-4617	CCAGGUGUAGACGGGUGUGACAAGUCcucaacacacagacacuggUUAGUGGGGUCUUCUGUGGCUGUCACACCUAAACACCCGC	85	23,52941	15,29412	20	18,82353	43,52941	34,11765	1,230769	1,176471	-30,3	-19,95	0,101687	17,71	-35,6471	-0,81892
pcn-mir-4654	GGCUGAUACGCCAGGUGAUGUGGGAUCUGGAGGCAGACUUCGGCUUCCGCAUCGGCCCGUGGAACCAGGCCUACAUGUACGACACCCUGCCGAUCAGUC	99	31,31313	19,19192	30,30303	19,19192	61,61616	38,38384	1	1,033333	-41,7	-40,2	0,172557	4,86	-42,1212	-0,68361
pcn-mir-466i-1	CAUAACCACAAGCUACACACGACUAUUCAGAUAUGCAUUGACACCUAUCUACACUUGUGUcaguauguguaucugugugugugugugugugugugugugugu…	98	7,142857	20,40816	17,34694	16,32653	24,4898	36,73469	0,8	0,411765	-36,4	-36,4	0,12872	16,86	-37,1429	-1,51667
pcn-mir-466i-2	gugugugugugugugugugugugugugugugugugugugauguuuaucACUCACCAUCUCAAUCUUUUAUGUGCACAAACACACCCCCAGCACACACAGAACAAGC	100	5	21	22	10	27	31	0,47619	0,227273	-34,8	-12,3	0,119059	30,37	-34,8	-1,28889
pcn-mir-466i-3	GUGUgcauagcugugugugugugugugugugcgugcgcgcgcccCACGUCUCGUGAUGAGCCAGUACACACCACAGAACUCGUGCAGCG	89	14,60674	13,48315	16,85393	10,11236	31,46067	23,59551	0,75	0,866667	-31,6	-16,94	0,020806	35,43	-35,5056	-1,12857
pcn-mir-466i-4	ggugugugugugugugugugugagaagugUCCUCUUAAUUCAGACAGAAGAG	94	13,82979	12,76596	10,6383	12,76596	24,46809	25,53191	1	1,3	-36,9	-36	0,067742	18,68	-39,2553	-1,60435

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUGUGCUCGUGAGGACAGACUGCuaaacacacaucacacacuaU															
pcn-mir-466m	CAUAGUAUGUUCuucucggugugugugugugcaugugcaugugugcuugcauguaaGUGAACAAGCAACCGAGUCAGCAAAUGCUCGG	88	12,5	15,90909	11,36364	10,22727	23,86364	26,13636	0,642857	1,1	-26,9	-24,4	0,033755	13,99	-30,5682	-1,28095
pcn-mir-466n	uuuuauauuACUUCAUGCUCAcgaaugugugcacacacacacacaugggugCAUAGGAGACAGGUGUGUGCGUACAUGUACUCAUGUAUAUACAU	95	12,63158	16,84211	11,57895	17,89474	24,21053	34,73684	1,0625	1,090909	-27,2	-25,5	0,120489	14,18	-28,6316	-1,18261
pcn-mir-466p	UGCUGUUUAUCCCUUCACUAUCUUCCACACUUCCUAUCUCACUGAAUGAGUGAAuuauaauaugugugugua caugugaagAGAGAAUCACUC	93	8,602151	18,27957	20,43011	23,65591	29,03226	41,93548	1,294118	0,421053	-19,9	-15,6	0,079044	7,09	-21,3978	-0,73704
pcn-mir-466q	UCGAUGAGGUAGUGGUGCACAAAUGAGAGAAUUAUUUUUGAGAGAAGAAUUCAGAUUUUGAGGCAGUAAAUACUGCaucaugugcacacacacacauacg	100	21	26	7	22	28	48	0,846154	3	-25,7	-22,5	0,021246	16,27	-25,7	-0,91786
pcn-mir-467f	UAUGCCAACGUCUCCCUCCCGCUGAaaaauuuauauacacacacacaccuuucaaGUGGGAGGAGAUGGACAAUGGCCCA	80	18,75	13,75	18,75	11,25	37,5	25	0,818182	1	-26,5	-26,5	0,059461	8,5	-33,125	-0,88333
pcn-mir-4715	CGCCACGCUGAUCACGUGCCAACGUGCCACCUUAACUGCACGGACCGCGGAAGAUGGUUUGGCCGUGGAAAGUGAGAGGUGAGU	84	33,33333	22,61905	26,19048	17,85714	59,52381	40,47619	0,789474	1,272727	-31,6	-31,6	0,443187	3,44	-37,619	-0,632

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-4757	aaaaaaaaccaaaacucaAGCGUCUCAGUAAAGGGUCUACAUUUUACACCUGUCAUUUCCAUGACGUCACAGAGGCGAGGUUUGGCUUUUGGA	93	20,43011	18,27957	17,2043	24,73118	37,63441	43,01075	1,352941	1,1875	-22,4	-21,8	0,139005	15,42	-24,086	-0,64
pcn-mir-4801	AAAGGUCUUCCUCUGUGUGUCUCCAGGAACCGGUCAAUAUUUCCAGCUGAUCAACACCGUUCUGAACGAAAUACACAAGAAAACCAAA	89	16,85393	33,70787	25,8427	23,59551	42,69663	57,30337	0,7	0,652174	-19,8	-19,8	0,127971	8,44	-22,2472	-0,52105
pcn-mir-4860	GGUGCACAUGCUUGUUUAGAAGUUACAAGCUUUGCUAGAAAAUGAUUGUAAGUUAUGUUGUAGAGAUUGUGUGAACUGCUAGACACAUCACUGAUGCUCU	100	24	28	14	34	38	62	1,214286	1,714286	-21	-15	0,009321	18,79	-21	-0,55263
pcn-mir-49	aacaaaagaaaccaaaaaggGCAUCUUCCCAUGAUGCAAAUAGACUAAGGGAAGCACCAUGUGAAGCACUUUUCUGGUUUCUUUUCCa	88	14,77273	19,31818	18,18182	23,86364	32,95455	43,18182	1,235294	0,8125	-26	-26	0,306727	8,11	-29,5455	-0,89655
pcn-mir-4918	AGCAACGAACCGAUGGGAGCUGUGAGCACGAUGGCCAGCACUGCCAGUGUCAGAACCUGCCCACAGUCACCGUUUGCGuucaa	83	27,71084	22,89157	28,91566	14,45783	56,62651	37,3494	0,631579	0,958333	-25,6	-24,9	0,031622	11,38	-30,8434	-0,54468
pcn-mir-4968-1	agcaucagcaucagcaacagcagcagcagcagaggaCCAGCCCAGAAGAUUCUGGUAGCCAGACCGCUGCACGCGCUGGUCCUGCGCAGAUCGA	94	18,08511	12,76596	21,2766	9,574468	39,3617	22,34043	0,75	0,85	-30,8	-28,6	0,049334	13,84	-32,766	-0,83243
pcn-mir-4968-2	CGACAGCGcacuggcagcagcaacagcagcagcagcagcagcagcacucgGCGGA	96	21,875	6,25	15,625	12,5	37,5	18,75	2	1,4	-34,7	-33,6	0,230058	16,55	-36,1458	-0,96389

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UGUGCAGCUUCCCUCGGUGGUUGCUGGCAGGGCCACUGGUU															
pcn-mir-4968-3	ACUUGCGCAUGCUUCGAGGUGUGGCUGCGCCCUCUCUUAGACAAGGUUGAGcgcacagcaacagcagcagcagcagcagca	81	19,75309	9,876543	16,04938	17,28395	35,80247	27,16049	1,75	1,230769	-27,9	-24	0,074193	12,7	-34,4444	-0,96207
pcn-mir-4968-4	uugugugugugugugagacuggcAUCAUUAUGCUAAGUACUCAUCAGCCUCGACUGUAGCCACUAAGGAUguacagcaacagcagcagcagcagcg	96	8,333333	14,58333	12,5	13,54167	20,83333	28,125	0,928571	0,666667	-21,1	-16,1	0,014202	21,15	-21,9792	-1,055
pcn-mir-5317a	GGGCCAGGGCCACAUCCCUAGCUACGGUccguuucuugucucuguguaCCUCUUGUGCUGCCAGACCGAGUGGUGAUCGGUGACCGCUGGAAC	93	25,80645	12,90323	24,73118	15,05376	50,53763	27,95699	1,166667	1,043478	-29,1	-29,1	0,378556	7,01	-31,2903	-0,61915
pcn-mir-5391	uuccugcaaGGUACCUUAUGAGGUAACUUGUUUGGACAGUUAUGAAGAAGUGGAAUUUAAUGUCCAACAGUUUAGAAUCAAACAGGCUAUCAUUCAGACc	100	20	30	13	27	33	57	0,9	1,538462	-18,8	-15,1	0,010656	34,01	-18,8	-0,5697
pcn-mir-5392	AAGUGAUUUUGCUCCAUGUGUGCAAUAAACUGGGAAAGAACGAUCUUUUUAGGUUUUAUUUGGCACAAUAUGUGUUACUugacauuuugagaaaacagUG	100	18	22	10	31	28	53	1,409091	1,8	-18,9	-12,7	0,006717	26,01	-18,9	-0,675
pcn-mir-5440	aaaaaauuugcgAAUGCUUUUUAGUUUAUCAGGAGGACUGAAGGGUGGAUGUGAAUACCACAUCCuuaac	98	15,30612	19,38776	11,22449	21,42857	26,53061	40,81633	1,105263	1,363636	-20,2	-18,8	0,104707	19,27	-20,6122	-0,77692

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	aaacuaaaaauaaccUCACUAAUUUGAC															
pcn-mir-5549	CCccaaaauaucaaaauagaaUAAUAAGAGGAGGGUGGGACUUCUUUAACAUUACACAGAUAUCCUAUUAUUCUUCAUGUUGGUUUUUUGAAU	93	15,05376	21,50538	11,82796	31,1828	26,88172	52,68817	1,45	1,272727	-19,5	-19,3	0,091982	11,56	-20,9677	-0,78
pcn-mir-559	AUCUGUCAAAGUACACAAACUAAUGAUGCUCAAAAUGUUAGCCAUUUGCGUUUAUAUGAUAAAGUAAAUAAGCACAGACUAUGUGUGGUUGUGACAGUU	99	19,19192	35,35354	14,14141	31,31313	33,33333	66,66667	0,885714	1,357143	-20,1	-16,59	0,039207	24,4	-20,303	-0,60909
pcn-mir-5594	GCAGUAAAGAAGGAAGACACGGUAUUUGCUGUUACAGACCUCGCAUAGCUAAGAGUACUGUAGUUUUUCCAAAGACCGUUCUCGCUCUCCGUUUACUCA	99	21,21212	27,27273	23,23232	28,28283	44,44444	55,55556	1,037037	0,913043	-22,2	-20	0,015865	13,26	-22,4242	-0,50455
pcn-mir-5595	UAGUGUCACGUGCUUUAAAAAAGGUGCAGGCUCUCUUUUUUCUCGCAGUCCCGCACUGACCUGAGAGAGUCACCAUCUUAUGUCAAACACAUGGACAAGA	100	21	27	25	27	46	54	1	0,84	-26,5	-26,5	0,090708	10,96	-26,5	-0,57609
pcn-mir-574-1	UCUAGACCUCAGUGCACGCGcggcgugagugugugugugagugugugugcgugugugugugcagagcuGAACGCAGGAGCACGAGCGGAGCUCCCCCCUCAACACCCCAGUGCUGUCGUCAUGGACCAUGCUACCUCC	140	15,71429	13,57143	25	10	40,71429	23,57143	0,736842	0,628571	-42,8	-36,9	0,005974	29,56	-30,5714	-0,75088

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-574-2	AAAGCGCCACACUCUAGACCUCAGUGCACGCGcggcgugagugugugugugugagugugugugcgug	67	10,44776	13,43284	17,91045	5,970149	28,35821	19,40299	0,444444	0,583333	-29,2	-15,45	0,069833	22,63	-43,5821	-1,53684
pcn-mir-5965	AUCUUGGGUGAUGACAAUUCUGACAUUUUGCAAGGUUUCAUCCUUGUCUGUUUCAUUCUUCAAACUGCGAAGUCGCGCAUCACCAAUGG	89	20,22472	22,47191	22,47191	34,83146	42,69663	57,30337	1,55	0,9	-20,4	-19,7	0,042628	9,27	-22,9213	-0,53684
pcn-mir-6	CCGCCAGUCGGACUAAAGCAGUCCGUUUUGUUGGCAGGAGggaacugcugcugcagcgacugggcc	66	19,69697	12,12121	15,15152	13,63636	34,84848	25,75758	1,125	1,3	-29	-25,9	0,187241	5,82	-43,9394	-1,26087
pcn-mir-6013	GGCGCCCUGGCCUACGAGCGGAGGAACAGCAUGGCGACCCUGUCCUCGGCUGCUGACGACCGCCUCAAACGCCAGAGGCUGA	82	32,92683	19,5122	35,36585	12,19512	68,29268	31,70732	0,625	0,931034	-33,6	-22,8	0,076623	17,71	-40,9756	-0,6
pcn-mir-6037-1	GGAAUCUUAUACACUAUACUGUGUGUACUUUAGUGUAACAUGGAAUCUUAUGCACUAAGCUCUGUGUACUUUAGUGUAACAUAGUAUGUGGACAGAUAUG	100	21	29	14	36	35	65	1,241379	1,5	-27,2	-27,2	0,192392	10,82	-27,2	-0,77714
pcn-mir-6037-2	AACUCUGUGUACUUUAGUGUAACAUAGUCAUGGAGUCUUAUACACUAAGCUCUGUGUACUUUAGUGUAACACAGUCACGGAAUG	84	20,2381	28,57143	17,85714	33,33333	38,09524	61,90476	1,166667	1,133333	-27,2	-27,2	0,62986	1,19	-32,381	-0,85
pcn-mir-6076	GCUGACACCCUUCCCUCAGGUCGUCCUCCUCUAAGAAGCAAACAGAAGCAUGACAGAGGAGAGGAGAAAACCCAAAGGGAGGGCUCUCUGU	91	26,37363	30,76923	27,47253	15,38462	53,84615	46,15385	0,5	0,96	-30,3	-29,9	0,158352	9,81	-33,2967	-0,61837

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-6098	GUGCUCGUCAUAGUGAUUGUGUGUUUCUAGAAGACAAGACCGUGUUCUGUUCAGAACACAAACAACAUGCUGAUUGUCGAGUAA	84	23,80952	28,57143	17,85714	29,7619	41,66667	58,33333	1,041667	1,333333	-22	-19,4	0,02079	16,09	-26,1905	-0,62857
pcn-mir-6132-1	gaaagcagggcuggggauucuggcauggcaccguuCCCCCAUCGGCCAGGUGCCAGAGCACCAAACCUGACUGAa	75	12	13,33333	21,33333	5,333333	33,33333	18,66667	0,4	0,5625	-29,7	-28,6	0,15428	5,96	-39,6	-1,188
pcn-mir-6132-2	CCCUGCAagagcagggcuggggauucuggcaaugGCGCCUCCUCCCUUCGCCAUGGUCCAGAGCcaaccacccuggcugcaaga	84	9,52381	4,761905	21,42857	8,333333	30,95238	13,09524	1,75	0,444444	-34,2	-34,2	0,026095	5,82	-40,7143	-1,31538
pcn-mir-6132-3	ggaagcagggcuggggauucuggcaugggcacGCGCCCCGCCACGUCCAGAGCUACACCCUGCUGGA	67	13,43284	8,955224	23,8806	5,970149	37,31343	14,92537	0,666667	0,5625	-30,2	-21,3	0,185778	16,57	-45,0746	-1,208
pcn-mir-6335	AUAGACAUUCAAGCCAAUCACUGCAUACACAGUGAUGCAGGUCAUUCGACAACUUUACUGCACCACUGUUUGAGCAUAUGUCUUGUGUCAGU	92	18,47826	28,26087	23,91304	29,34783	42,3913	57,6087	1,038462	0,772727	-25,4	-25,4	0,193109	5,21	-27,6087	-0,65128
pcn-mir-6416	AGCAAAUUGCUUGAUCAUAGUGAGCGUAUCAUGGCAGCACAUCCUCAUGGGUUUGCGUCUCCGUAUCAUCUGCUAGCUACUGAGAACAAUUUUUU	95	21,05263	24,21053	22,10526	32,63158	43,15789	56,84211	1,347826	0,952381	-21,2	-20,6	0,052561	13,18	-22,3158	-0,51707
pcn-mir-64c	uaauuaguccagugUUGAUGUCACACCAGUGUUAAUUGUCUCCAUGUCCAGCUGCUCGUUGGCAUGACACACUGGCUGAACCA	83	18,07229	18,07229	22,89157	24,09639	40,96386	42,16867	1,333333	0,789474	-23,4	-23,3	0,067275	11,61	-28,1928	-0,68824

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-6505	UUUAGCUCCAUCUGACUUCUACCUGUUCCCUAAACUCAAAUCCCACCUAGUUGGUCAccauuuuggaaacaaugauGAAGUCAUAUGAGCUGUG	94	12,76596	19,14894	22,34043	25,53191	35,10638	44,68085	1,333333	0,571429	-21,1	-19,17	0,013409	12,71	-22,4468	-0,63939
pcn-mir-6516	CGGUCUUAUUCUAACUGGUUUCAUGAAUGCAGUAACAGGUGUCUCCACCUCUGCAUUGCUCAAACCACAUUACAUCAGAAAG	82	17,07317	28,04878	25,60976	29,26829	42,68293	57,31707	1,043478	0,666667	-19,7	-19,7	0,160405	4,33	-24,0244	-0,56286
pcn-mir-6560	CGCUGUGUUGCUCCAUACUGCUGAAGUAGUGUCUGUACUUCUGUUUCAGUCAUAAUGAGCAGCUCAUCA	69	21,73913	20,28986	23,18841	34,78261	44,92754	55,07246	1,714286	0,9375	-19,8	-18,8	0,070623	8,29	-28,6957	-0,63871
pcn-mir-6575	GAACUCCAAACGGUGGGCCUGGGAAGAGGACAGGAGCAUCUGUCGAGUUUUCCCAGGCUGAUGUGUGGAGCAG	73	36,9863	23,28767	20,54795	19,17808	57,53425	42,46575	0,823529	1,8	-37,7	-37,7	0,136478	3,17	-51,6438	-0,89762
pcn-mir-6587	guuuuuuuuugguuuuuuguuuuuuguuuauugcuuugcUGGAAGUGGUGCUAUAUGUCGCUUCUUCGUAGAAACCAUAGGACCAAAGAAACUG	94	14,89362	18,08511	10,6383	14,89362	25,53191	32,97872	0,823529	1,4	-20,8	-14,3	0,005082	23,66	-22,1277	-0,86667
pcn-mir-6603	acacacagugagauCAAGUUAAAGGACAAGAGGAAAUAACUUGAGCAGCUUUGUCAUUUUCAGCUCGUAUUUUAUUUUGAUCCAUCAGUGAUU	93	16,12903	25,80645	12,90323	30,10753	29,03226	55,91398	1,166667	1,25	-18,6	-16,4	0,03366	16,97	-20	-0,68889
pcn-mir-6680	CCACAAAACACACUAAAGCGGAUGUUGCUGAUGAAGAUAAAUCGUGGACCUAAGGUAUCAGCGGACAU	93	22,58065	31,1828	22,58065	23,65591	45,16129	54,83871	0,758621	1	-24	-24	0,03754	8,12	-25,8065	-0,57143

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCAAGCUCUAGUAUUGUCUCUGGUC															
pcn-mir-669f	acacacauauacauacacacacacguguuguUUCACAAGUAGUACAACAGGCGUGUGAGAUGUUUAUGUGAGG	73	17,80822	16,43836	6,849315	16,43836	24,65753	32,87671	1	2,6	-25,6	-20,3	0,138486	10,86	-35,0685	-1,42222
pcn-mir-67	GUUGUCGGUCCUUGUUCAGCCCGGUUGUGAUGGAGUUGGUCGCAUCACAACCUGCAUGAAUGAGGACUGACGU	73	31,50685	17,80822	21,91781	28,76712	53,42466	46,57534	1,615385	1,4375	-42,5	-42,5	0,387173	1,28	-58,2192	-1,08974
pcn-mir-6816	UCCUGCACGUCGCUCCCGUCGUUGAUCAGGGCGUUCCCAAAUUGAAGGACCUGCACCUUGCAUGGAACGCCAUAUCCAGCCUCGAGGACAAUGCACUC	98	23,46939	21,42857	33,67347	21,42857	57,14286	42,85714	1	0,69697	-29,7	-29,4	0,03583	9,47	-30,3061	-0,53036
pcn-mir-6852	CUAAAGAUUUGUCCUCUGUUCCUCAUCGUUCGUGAUCACAAAGCCAAACGCUUUGCUUUCAGUGAACAAAAUUGUCAAAACAGUUGGAUAAUCUUCAC	98	15,30612	29,59184	23,46939	31,63265	38,77551	61,22449	1,068966	0,652174	-24,8	-24,8	0,119887	4,49	-25,3061	-0,65263
pcn-mir-6894	CCGCUUGGUGGUAAUGUCAAGGAGGAUGGAGAGCUGUUACAGCAUCUUCGCCAUCUUACAUCCUAGCAAGUAG	73	27,39726	24,65753	21,91781	26,0274	49,31507	50,68493	1,055556	1,25	-25,9	-25,9	0,400909	2,89	-35,4795	-0,71944
pcn-mir-6951	UUGUUGUAUUUGUGUGAUUCCCGACACUGUUCCUUGUACUGUAUAUACAGCGAACAGGACCUUUAUCACACCUACAAUGG	80	18,75	23,75	22,5	35	41,25	58,75	1,473684	0,833333	-23,5	-23,4	0,272799	2,62	-29,375	-0,71212

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-6974	UCAACCACAACGACGACGGUGGACAAGGGGUGGACCUCGAUGCCAGCAUGGUGUCCACCAGCUACAUUGCGCUCUUCUCCACUCUCUUCUGUGUCGUGGG	100	27	19	31	23	58	42	1,210526	0,870968	-34,3	-27,1	0,01583	24,28	-34,3	-0,59138
pcn-mir-6977	CCUCGUGCGUGGAGAAAACAUUGAUUGUGAGGCGCUGUGGGCAGGUGAGCCAGACUGUCACUAAUGUAUGACUUCUGCUCACUCACGGCU	90	30	21,11111	23,33333	25,55556	53,33333	46,66667	1,210526	1,285714	-29,4	-28,7	0,036666	9,79	-32,6667	-0,6125
pcn-mir-6999	CACACGUGCACUGCACGCCAUCUCGCCUCCAUCCCAUCAUCUcaggagaaggaaggagagucAGGAGCGUGAAUGUCUUGCAGAUGCCCCGUCAC	95	15,78947	15,78947	30,52632	16,84211	46,31579	32,63158	1,066667	0,517241	-27,3	-17,3	0,084758	25,48	-28,7368	-0,62045
pcn-mir-7000	CAAACAGUGCCCACCCACCUGCCUGUCACAGUgugauuuauuuacauuguaaCUGGAUGCUCAUGGCUGGUGGCCCAGCACUGUAUA	87	18,3908	16,09195	26,43678	16,09195	44,82759	32,18391	1	0,695652	-32,8	-32,5	0,12354	7,83	-37,7011	-0,84103
pcn-mir-7072	UGAGGAUACAAAGGAGGGGCAGGUCUAGCUUCUCUUCUUCCAGUUCCUGCUGGUCUCUUUGUCUCCAGA	69	26,08696	17,3913	24,63768	31,88406	50,72464	49,27536	1,833333	1,058824	-22,9	-22,9	0,435548	12,1	-33,1884	-0,65429
pcn-mir-71-1	AAGAAAUCCUUUUGGUGUGAAAGACAUGGGUAGUGAGAUGUCGGUCCUUCAGAUUCACCUUAUUACCCUGUCUUUUGCAUCAAGGGGAAACCAUGCGUCAA	101	23,76238	26,73267	19,80198	29,70297	43,56436	56,43564	1,111111	1,2	-43,3	-43,3	0,1969	5,13	-42,8713	-0,98409

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-71-2	gcacagacacaucUGCAUUCACAUUACACUUACUUGCACCCGUACCCUAGUCUGGUGAAAGACACAGGUAGAAUGUAAUUCGUGUUGUUCUGCUG	95	17,89474	21,05263	20	27,36842	37,89474	48,42105	1,3	0,894737	-19,9	-10,75	0,038952	25,08	-20,9474	-0,55278
pcn-mir-7151	UUGUUGACACUGAGAAAAGUGACAGGAUCCAUCUCUGCCUGCUCACCUUCACUUUUGUGGUGAGCAGcugucauuucucucuuugCAAGAU	91	18,68132	19,78022	18,68132	23,07692	37,36264	42,85714	1,166667	1	-33,5	-32,8	0,126184	5,54	-36,8132	-0,98529
pcn-mir-7307	AGUCAGGAGAAUGCCAUUGAGUUAUUAACUUGGUGUGGUGGACAUUGUUUCAGUCACAUCAGCGAUGAAGAUGAUAAUCAAAUGGUCAUUUCUGCGA	97	25,7732	28,86598	14,43299	30,92784	40,20619	59,79381	1,071429	1,785714	-26,8	-23,4	0,005514	16,02	-27,6289	-0,68718
pcn-mir-7314	GUCACAGGGACCCUGGUUGGCCUGCACAAGCCCCAGUAAUACUGGUGUCAAGGAUAGACGAAACUCCUUGAAGAUUGUCAGAAACAGUGUCUCUGUGCa	99	26,26263	26,26263	24,24242	22,22222	50,50505	48,48485	0,846154	1,083333	-32,6	-28,7	0,010541	12,05	-32,9293	-0,652
pcn-mir-7386e	UGUAAAGUAGUAAACACUUUACCCGUUUGACUGUUUGCCCUUGUUCUUGAACAGUGUGGGUGAACCUCUUGACUGGUGUAAAGUUGUAUAACUUUUUA	98	21,42857	23,46939	16,32653	38,77551	37,7551	62,2449	1,652174	1,3125	-24,7	-24,1	0,025944	13,11	-25,2041	-0,66757
pcn-mir-7389	GUUGGGAAGAAGCUGCGGUCACAGCUGUUGGUGAGGGUGAGCCCUUGCUGUACUUGAAGCAAGAGAGACCUCCUGACUGUUACCUCCAgcuucucccccu	100	28	19	20	21	48	40	1,105263	1,4	-40,8	-39,7	0,083977	10,1	-40,8	-0,85

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-7398h	CGUACAGGUACACGGGACUCAACAGUCACCUUGUCGUAACCCUCCCUUCUUUAUGAUAGCCCUUCAAGAUGCUUUACCGCAGUGACCUGUGGG	93	21,50538	21,50538	30,10753	26,88172	51,6129	48,3871	1,25	0,714286	-24,3	-24,3	0,03188	3,28	-26,129	-0,50625
pcn-mir-7399	AGGACUUCAAUGUAGCACUUAUacgcauguauauguauauacaugaaUGUAUUUAUGAAUGUACACAUGUAUCUCUCAUAGGUACUUUGAAGUAUU	96	12,5	21,875	10,41667	29,16667	22,91667	51,04167	1,333333	1,2	-23,9	-23,9	0,118362	17,65	-24,8958	-1,08636
pcn-mir-743b	AGUAGACACUUCUGAACGAGCUUCUUGUACUAACCUUGCUGUAAACCCCAGGACGAAGCUGUUCAGACUGGUGUCUGGU	79	24,05063	24,05063	24,05063	27,8481	48,10127	51,89873	1,157895	1	-26,2	-25,6	0,030673	10,47	-33,1646	-0,68947
pcn-mir-745a	GAGGCGGCUUUUCAACAGGCUGCCUUGCGUUCUUAAUACAAGCUGCCUGAUGAAGAGCUGUCCU	64	26,5625	20,3125	25	28,125	51,5625	48,4375	1,384615	1,0625	-30,5	-30,5	0,08838	5,12	-47,6563	-0,92424
pcn-mir-745b	AGCCUAUUUCCCUCCGCUCUUCCUUUGGACAGCUAUCGUGCCGACAAGGAGAGCUGCCAAAUGAAGGGCUGUGGUGGACGGGC	83	30,12048	19,27711	27,71084	22,89157	57,83133	42,16867	1,1875	1,086957	-35,7	-31,3	0,025365	10,13	-43,012	-0,74375
pcn-mir-7472	GCAUUCCGGGGAGAACAUUUCAUAUGAGAUGUGCUGGAGAUGAAAACUCUUCCUCCCCAGAACAU	65	23,07692	29,23077	23,07692	24,61538	46,15385	53,84615	0,842105	1	-20,8	-20,8	0,342745	5,01	-32	-0,69333
pcn-mir-750	GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUGGCAGGAAUACUGUCUUCCAUGCCAGAUCU	97	26,80412	20,61856	23,71134	28,86598	50,51546	49,48454	1,4	1,130435	-52,7	-51,6	0,157091	3,48	-54,3299	-1,07551

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AACUCUUCCAGCUCAUAGUGGCAUGGCAUG															
pcn-mir-751	CUUGGCUCUGUGUGAUUGGUUUCUGUUCUCCAAGauuaaaacauguuugaauGGCCACCACUUUGGCACAGUAAACAUCAUCCAUAGAGCCCUU	94	17,02128	15,95745	22,34043	25,53191	39,3617	41,48936	1,6	0,761905	-27,8	-22,8	0,051558	21,54	-29,5745	-0,75135
pcn-mir-753d	AGAAGAUCAUUAUUCAAGCGAUUGCAGAGAGAAAAACUAUUCGCAUGCAUGAAUGCAUAGGUAUGAGUGUGCACGUGCCUGCAUAUGAUCUCAC	94	23,40426	32,97872	18,08511	25,53191	41,48936	58,51064	0,774194	1,294118	-22,8	-18,7	0,009746	21,51	-24,2553	-0,58462
pcn-mir-760	GAGCAAGUGACUGACACAGAAAAGCUGAUCAUGAAGAUCGGCUCUGGGUCUGUGAAGUCAGUUGGCA	67	31,34328	29,85075	17,91045	20,89552	49,25373	50,74627	0,7	1,75	-29,1	-28,8	0,330018	2,01	-43,4328	-0,88182
pcn-mir-7643	aguuuguuuuucaguGAACAUUGAAUACUGUCGACAUCUGUUUCUCAGUAAUUUUUACUUAGUAUCACAGAUUCAACUGAACACUGAUACAGAACGA	97	12,37113	28,86598	16,49485	26,80412	28,86598	55,6701	0,928571	0,75	-20,4	-15,3	0,02611	14,6	-21,0309	-0,72857
pcn-mir-7649	GAGAUCAUAGUAAUGACAGCAGUGCUGUAUCUAUGUAUGCCAACAAAGUGAAUCAUUGUUGUACUUUAUUUAUGAUUUG	79	20,25316	30,37975	12,65823	36,70886	32,91139	67,08861	1,208333	1,6	-19,5	-18,5	0,065763	5,47	-24,6835	-0,75
pcn-mir-76b-1	ACAUCCAAGCCUCACCCAUAGUUGAGAAAAGCCAUAGUUGUGGUAAcaaacugucuuguuuuuuucaacuccugggugaggcuuagaaca	90	10	17,77778	12,22222	11,11111	22,22222	28,88889	0,625	0,818182	-43,4	-43,4	0,140211	4,23	-48,2222	-2,17

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-76b-2	UACACUGCUGCCAAAUUGUUGAUAGAAAUGGUAGUUCCGCAAUUGCAGgugucuuguuuuuuucaacUGCGUUUGCACUACAAUUUUGAACAGUUGU	97	17,52577	21,64948	14,43299	26,80412	31,95876	48,45361	1,238095	1,214286	-21	-14,9	0,029354	18,5	-21,6495	-0,67742
pcn-mir-770	UGUUGAUUGGACUGUACAUGUAUGUACCAGCAGUUGGGGCAGCACCACGUGUCUGGUAAAUCUGAGUGUCAACAUGG	77	29,87013	23,37662	18,18182	28,57143	48,05195	51,94805	1,222222	1,642857	-19,8	-19,4	0,075721	9,14	-25,7143	-0,53514
pcn-mir-7880k	AAACGUCACAGUCCUGAGCUCUGCCAAACCCgaggucacgaggucagaaAUGGUUGCUGAGCUCCACGUGACGCCG	76	18,42105	17,10526	26,31579	14,47368	44,73684	31,57895	0,846154	0,7	-22,8	-18,67	0,085099	18,19	-30	-0,67059
pcn-mir-7880q	UCAAUGACAUUUACAUACUCUUGGAAGAGGGUCAUGCCUACCAUGCAGUUUGCGACCAaaaucacucucucuuucaaguACUAUUGCUUACAUGCAUUCA	100	14	22	19	24	33	46	1,090909	0,736842	-19,6	-19,6	0,125227	11,54	-19,6	-0,59394
pcn-mir-79	GGUUGGUAUUAUAUcuguauuauauuauauuagUCAGUUGCUGUAGCCACUGUAACUCUGUAGCUCUUUUAUAAUGUAAAGCUAAAUUACCAGUG	95	15,78947	21,05263	12,63158	30,52632	28,42105	51,57895	1,45	1,25	-18,8	-18,8	0,098286	9,53	-19,7895	-0,6963
pcn-mir-7901	CGUCGUCAUUUCUGAAAGUGAUUGUUUGUCUUUAUGAUUACUGUAGUUGAAGCGCUAUUGGUUCAACUUGAAAAACAGUGACUUCUCUGUUCUGACGACU	100	21	23	17	39	38	62	1,695652	1,235294	-28,4	-28	0,058835	13,99	-28,4	-0,74737

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-7c	aaaaaaaaacuaacccaGGCUGCCGUGAUCUGGAAGACUAGUGAUUUUGUUGUUCUGAUGCUGACUAACAAUAAAUCACAUUCUUCCCUGAUUGCUGCAGCCACAUGCU	109	18,34862	19,26606	19,26606	27,52294	37,61468	46,78899	1,428571	0,952381	-37,2	-34,7	0,144483	9,32	-34,1284	-0,90732
pcn-mir-8-1	GCGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUGUCAGCAUUUUCUAAUACUGUCAGGUAAAGAUGUCCACAGAGUCU	86	22,09302	25,5814	20,93023	31,39535	43,02326	56,97674	1,227273	1,055556	-39,6	-39,6	0,178161	3,42	-46,0465	-1,07027
pcn-mir-8196b	UGCAAGAAAUUUACGaugcuguuuuaaaauguaacggGUUACAAGUCCGUUCUGGCAUUUUGACACCAGuaguuaauuuuucuu	84	11,90476	15,47619	11,90476	16,66667	23,80952	32,14286	1,076923	1	-19,6	-19,4	0,052609	9,33	-23,3333	-0,98
pcn-mir-8-2	UGCGCGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUGUCAGCAUUUUCUAAUACUGUCAGGUAAAGAUGUCCACAGAGUCUGCUGG	94	24,46809	23,40426	21,2766	30,85106	45,74468	54,25532	1,318182	1,15	-46	-46	0,192059	3,37	-48,9362	-1,06977
pcn-mir-8296	UGAGACCUCGAGAUGAUGGAGAGCACACUUGACUUACUCUGGCUGAGAGUCCGUGAUGUGUCUCUCGUCUUCUCCUGGUCAGA	83	27,71084	19,27711	24,09639	28,91566	51,80723	48,19277	1,5	1,15	-35,3	-34,3	0,251447	8,1	-42,5301	-0,82093
pcn-mir-8332	UCCAUUUAUCUGCACAUCUCUUGAUGAAGUCAGUAAAGACCACAGAUCCUCCGggauguuucuuucuuugcucuucUCGACAUGCGUGCACAAAAUAAG	99	13,13131	24,24242	20,20202	19,19192	33,33333	43,43434	0,791667	0,65	-24,6	-24,5	0,280827	4,23	-24,8485	-0,74545

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-8335-1	uuuuguuguuguuguuguuuugccaACUGGAUUGUAGAUGCGGCACGCAGAGGCAGUAAAUAUGCUAACCACugguuaaauaaaauaacaacagac	96	13,54167	15,625	10,41667	9,375	23,95833	25	0,6	1,3	-27,7	-27,7	0,191059	9,75	-28,8542	-1,20435
pcn-mir-8335-2	uaccuguuguuguuguuguuguuuugguuuuuuuugcugCAAGGCUGUUAGCUUAUCUUCACCCGCAAGCACAAGCAUUACAACGAACAGACA	93	9,677419	20,43011	17,2043	10,75269	26,88172	31,1828	0,526316	0,5625	-21	-18	0,029125	12,45	-22,5806	-0,84
pcn-mir-8364g	GCCCACCUUCCCUUUCUGCUUACCAAACGGAAAAAGGAUGCUACGCAGAGAGUGUGgaaaaggggagagggaggugcAG	79	17,72152	20,25316	20,25316	15,18987	37,97468	35,44304	0,75	0,875	-29	-29	0,217216	5,42	-36,7089	-0,96667
pcn-mir-8416	uaugauugGUCACAUAGUUUAUCAGAUGAUAAAUGGAGUGAACAUAGUAUUUACUUCCCCUGCUGAUAAGUUAAGUAUCCAUUCUUG	87	16,09195	27,58621	14,94253	32,18391	31,03448	59,77011	1,166667	1,076923	-18,7	-18,7	0,245232	4,5	-21,4943	-0,69259
pcn-mir-8422	AGGAAAAGAUCAUCAGAACUUGCAAUUAAAAGUAgcacaaacaagaaagcaagaCCAAGUGUUAGUAAAAUGCAUGUUGAGGAUGAUUCUUUUUCA	96	15,625	30,20833	9,375	23,95833	25	54,16667	0,793103	1,666667	-23,6	-23,1	0,141217	4,11	-24,5833	-0,98333
pcn-mir-8462-1	AAUCCCAGGUGCAGAUAGGCAAAAAUCAUAAGAAUGAUCAAAUAAUUGUCCUUGUGAUUUGACACUCACUGGGGUG	76	22,36842	34,21053	17,10526	26,31579	39,47368	60,52632	0,769231	1,307692	-21,6	-18,9	0,11733	10,16	-28,4211	-0,72
pcn-mir-8462-2	aaacugucaagcAUCUUUCUUUUGGAAACAAGUAAAUCAUAUGCCACCUGUCAGUGACAUGAUGAUUAAU	100	16	30	14	28	30	58	0,933333	1,142857	-19,5	-18	0,090701	10,52	-19,5	-0,65

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUGAUAAGAUGAAAUGGUCUUCAACAGCAG															
pcn-mir-8485-1	UGUACAGGCAGCCCAUACCUGUCAUUUCUGUUUGGUAGAguguacgcgcgcgcacacacacacacacacguauucaUAGAGUAuguuuugcgugugug	98	11,22449	11,22449	9,183673	15,30612	20,40816	26,53061	1,363636	1,222222	-28,9	-25,3	0,086631	28,22	-29,4898	-1,445
pcn-mir-8485-2	acacacacacacacacacguacucacacacacacauguucauuCAGGCUCGCGCCACAGGAAACAUCAGGUGAGAGACAugucgcugcuguugcugugg	99	11,11111	12,12121	10,10101	3,030303	21,21212	15,15152	0,25	1,1	-20,6	-10,9	0,017176	20,99	-20,8081	-0,98095
pcn-mir-87	UUUUAAAAGUGCGGAACUGGCCUGCCUGAAAUUUAUGCUCAAACCUAUCUUGUGAUAAGGUGAGCAAAGUUUCAGGUGUGUUAGAUCUGUACCUGAUC	98	23,46939	26,53061	17,34694	32,65306	40,81633	59,18367	1,230769	1,352941	-42	-38,2	0,120168	3,71	-42,8571	-1,05
pcn-mir-8834a	UUGCUGCCGGGCCUGGAGGCCAGCUGUACCACACAUCUGCUGCCCCUCCACACCUGCGGCAGGUG	65	29,23077	13,84615	38,46154	18,46154	67,69231	32,30769	1,333333	0,76	-28,3	-28,3	0,089597	7	-43,5385	-0,64318
pcn-mir-888	UCAAAGGACUGACACCUCUUUGUCCAGCUCAUGACAGGCUGCACAGAUGAGCCGGACAAGUGUUGUUAGUGGCUUCAU	78	25,64103	24,35897	24,35897	25,64103	50	50	1,052632	1,052632	-25,5	-24,5	0,08259	6,42	-32,6923	-0,65385
pcn-mir-8915-1	UAUUUGCCACCGCUUGGCGGCUUCUGGGUUUUCUUCGGGAGCAGGACGGCCUGGAUGUUGGGCAGCACACCACCUUGCGCGAUGGUGACGCCAGACAGAAGCUUGUUCAACU	144	32,63889	15,97222	27,08333	24,30556	59,72222	40,27778	1,521739	1,205128	-48,1	-38,5	0,009336	30,97	-33,4028	-0,5593

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	CCUCGUCGUUGCGGAUGGCCAGCUGCAGGUGA															
pcn-mir-8915-2	GCUUAUUUGCCGCCGCUUGGCGGCUUCUGGAUUUUCUUGGGGAGGACGGCUUGGAUGUUGGGCAGCACACCACCGUGUGCGAUGGUGACGCCAGACAGAAGCUUGUUCAACUCCUUGUCGUUGCGGAUGGCCAGCUGCAGGUGA	144	34,02778	15,27778	24,30556	26,38889	58,33333	41,66667	1,727273	1,4	-51,5	-47,1	0,029851	26,76	-35,7639	-0,6131
pcn-mir-8915-3	ACGGCCUGGAUGUUGGGCAGCACACCACCUUGCGCGAUGGUGACGCCGGACAGAAGCUUGUUCAACUCCUCGUCGUUGCGGAUGGCCAGCUGCAGGUGACGGGGGAUGAUUCUCGUCUUCUUGUUGUCGCGAGCGGCGUU	140	34,28571	15	26,42857	24,28571	60,71429	39,28571	1,619048	1,297297	-50,2	-44,5	0,009254	28,93	-35,8571	-0,59059
pcn-mir-8915-4	GACGGCCUGGAUGUUGGGCAGCACACCACCUUGCGCGAUGGUGACGCCAGACAGAAGCUUGUUCAACUCCUCGUCGUUGCGGAUGGCCAGCUGCAGGUGACGGGGGAUGAUUCUCGUCUUCUUGUUGUCGCGAGCGGCGUUG	142	34,50704	15,49296	26,05634	23,94366	60,56338	39,43662	1,545455	1,324324	-51,2	-50,3	0,023929	15,77	-36,0563	-0,59535
pcn-mir-8915-5	GACGGCUUGGAUGUUGGGCAGCACACCACCGUGUGCGAUGGUGACGCCAGACAGAAGCUUGUUCAACUCCUUGUCGUUGCGGAUGGCCAGCUGCAGGUGACAGGUUACUGCAAACCCUAGAAAGGAUUUGAGAUUUGUAGCU	142	30,98592	22,53521	22,53521	23,94366	53,52113	46,47887	1,0625	1,375	-48,4	-48,4	0,094252	13,02	-34,0845	-0,63684

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-9015	CGAGGAGUGUCACGAUGGUCCCAGAGGAGACGAGAAAGUUGUUUGGCUUCCAUCACUCCGAUCUCCUAAUCGACUGUUGUGACAUUCCGCU	91	26,37363	21,97802	25,27473	26,37363	51,64835	48,35165	1,2	1,043478	-38,6	-36,5	0,087379	7,6	-42,4176	-0,82128
pcn-mir-9-1	GUCCAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUGCCCCAAUCAUACAGCUAGAUAACCAAAGACAAAACCGGAGCCGCCG	88	17,04545	28,40909	27,27273	27,27273	44,31818	55,68182	0,96	0,625	-27,8	-27,8	0,435416	2,15	-31,5909	-0,71282
pcn-mir-9120	CCUCUUCUACACAACAAUCUCACUGGGGAGAGGUACACUCGUGACUCAGUCUCCCUCUCUCGCUAUCUCGCGCACAGAGAUGGUUAGAAgcac	93	19,35484	22,58065	30,10753	23,65591	49,46237	46,23656	1,047619	0,642857	-26,6	-26,6	0,320862	10,09	-28,6022	-0,57826
pcn-mir-9-2	UUGGCGUCCAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUGCCCCAAUCAUACAGCUAGAUAACCAAAGACAAA	80	15	28,75	23,75	32,5	38,75	61,25	1,130435	0,631579	-23,1	-20	0,134445	5,8	-28,875	-0,74516
pcn-mir-92a-1	AUUUGUGUGCUCAGGAAAGGUUGUGACUUGGGCAAUUCUGUGAUGUCCUGUCAGAUUGCACUUGUCCCGGCCUUGUCUUGGCUUAcacuu	90	26,66667	14,44444	18,88889	34,44444	45,55556	48,88889	2,384615	1,411765	-31,5	-29,1	0,036643	11,93	-35	-0,76829
pcn-mir-92a-2	CUGGCAUUAGCAGUAGGUCUUGAUGGGUGCAAUUCUGAUGCAUUAAAUAACAGAUUGCACUUGUCCCGGCCUUCUGCCAAUAGCUAA	87	22,98851	25,28736	21,83908	29,88506	44,82759	55,17241	1,181818	1,052632	-35,5	-35,5	0,137395	3,64	-40,8046	-0,91026

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pcn-mir-92b	AGCGAUCAGGGUUGUGUAGACCGGAACAGGGGCAAUGCAUUUGAGUCGUCUGUAUUGCACUCGUCCCGGCCUAUCCAAUCUGACUCAUA	89	26,96629	22,47191	24,7191	25,8427	51,68539	48,31461	1,15	1,090909	-32	-32	0,072274	6,85	-35,9551	-0,69565
pcn-mir-92c	AUUAUGUGUUAUUGUAAAAGGUUGGGAUGUGGGCUCUGUCGCGCAAUAGAUAACUUUCUUUGACAAGCAUUCGCCCACACCUUAUUACAACACAAUCAC	99	19,19192	28,28283	21,21212	31,31313	40,40404	59,59596	1,107143	0,904762	-26,6	-26,6	0,055246	15,72	-26,8687	-0,665
pcn-mir-9-3	UUUGGCGUCCAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUGCCCCAAUCAUACAGCUAGAUAACCAAAGACAAAACCGGAGCCGCC	93	17,2043	26,88172	26,88172	29,03226	44,08602	55,91398	1,08	0,64	-34,1	-34	0,28369	3,42	-36,6667	-0,83171
pcn-mir-9341-1	ACCUGAUUGUUAGUUAAAAGGGCCGAGGAUAGUCAGGCAUAUCGACAAGUUUGUGUAUCGCUUCUCGGCCUUUUGGCUAAGAUCAAAG	88	26,13636	26,13636	18,18182	29,54545	44,31818	55,68182	1,130435	1,4375	-31,8	-28,3	0,01871	14,65	-36,1364	-0,81538
pcn-mir-9341-2	caauucugacauuuuuacugugCCAUGGAUGAGACUUGUUGUCAAAAUGGACAUCGCUUCUCGGCCUUUUGGCUAAGAucaaagugugagaaaaa	95	14,73684	13,68421	12,63158	17,89474	27,36842	31,57895	1,307692	1,166667	-29,5	-29,5	0,182004	3,22	-31,0526	-1,13462
pcn-mir-9388	UUAUGUGCacguauguauguauguacauauaauaUUUAUCAGUGGGUGUGAGCACCAAAACAUUUAUGAAUGCAUGAUAAGCACAGUC	88	15,90909	21,59091	11,36364	21,59091	27,27273	43,18182	1	1,4	-19	-17	0,162177	22,34	-21,5909	-0,79167
pcn-mir-9-4	GGCGGCUCCGGUUUUGUCUUUGGUUAUCUAGCUGUAUGAUUGGGGCAGAAAUAUCAUAAAGCUAGG	93	26,88172	29,03226	17,2043	26,88172	44,08602	55,91398	0,925926	1,5625	-45,5	-45,5	0,314517	3,87	-48,9247	-1,10976

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUACCAAAGGCAAAAAUGGACGCCAAA															
pcn-mir-9543a	GAGAAACUAGCAAGUUCAUUAUAUGUCAGUAUGCAGAUUGCAAUGGUUUACUUUCCAUUGCACUGCACUGGAGUAAAUAUUgcuuguuuauu	92	17,3913	27,17391	14,13043	29,34783	31,52174	56,52174	1,08	1,230769	-28	-26,4	0,033319	14,41	-30,4348	-0,96552
pcn-mir-96	ACAUgguauuuauuuacaaucauguGCAGUGCAGUUCUUUAUAUAACUAAUCAUACUGCAAAAAGUACAUGACUGCAAUAUGCAAAAAACAUAUCACAC	99	9,090909	33,33333	16,16162	20,20202	25,25253	53,53535	0,606061	0,5625	-19,7	-17,9	0,056255	11,21	-19,899	-0,788
pcn-mir-969	AAGAAGCGCGAAUCUUCUUCUCCUGGAucaggaaaauaaauuauuuaagaGUUCCACUAAGCAAGUUAUCUUGCAGGAGCUAGAAAAUCGCCCUUAGA	98	16,32653	22,44898	18,36735	19,38776	34,69388	41,83673	0,863636	0,888889	-22,7	-22,7	0,05101	17,67	-23,1633	-0,66765
pcn-mir-96b	UCAAUUAUUUGGCACUUGUGGAAUAAUCGGUGUAUCUAAAAGUCGAUUAUACACCGGUGCCAAGUCAUCA	70	20	30	18,57143	31,42857	38,57143	61,42857	1,047619	1,076923	-23,4	-23,4	0,19695	2,59	-33,4286	-0,86667
pcn-mir-972	CUCUGGGAUGUAGGGGGAAAACUGUCaguaauguaaauauuuuuuuugucaacuGAUAUAUGAGAAUUAUGUGGCUACUUUAACCCCUUCACCCACAC	98	16,32653	19,38776	16,32653	19,38776	32,65306	38,77551	1	1	-23,7	-19,6	0,044018	24,34	-24,1837	-0,74063
pcn-mir-981	AUCAGACUUUGUUCCGGGUUUCGCGGCUUGCGAACAGUCAUUUU	85	25,88235	17,64706	23,52941	32,94118	49,41176	50,58824	1,866667	1,1	-32,6	-28,1	0,035045	8	-38,3529	-0,77619

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GAGCUGUGUUCGUUGUCGUCGAAACCUGCCCUGAAAUCUGA															
pcn-mir-9891	CUCCACCUGGCGGCCGACCGAUGGUUCGGGAUGCGCGUUGACCUCAUCGGCUUCGUCCUCGUCUUGGCCACGACACUCGGCUCGGCACUUCAUGGUGCAC	100	29	12	36	23	65	35	1,916667	0,805556	-37,2	-28,7	0,102885	18,05	-37,2	-0,57231
pcn-mir-995	CUGACCGCCAGGAGCAGACGAGAACUAACCAGAGCCCGGAUACUGUGUGUGGCUCAUGAUUGACUGUCACACCUUCUCGCUGGAUGGUAAA	91	27,47253	25,27473	26,37363	20,87912	53,84615	46,15385	0,826087	1,041667	-25,2	-20,28	0,073074	27,75	-27,6923	-0,51429

Supplementary Table 4– Structural and thermodynamic characterization of miRNA precursors in P. maculata. Size (Tam), G content, A content, C content, U content, GC content, AU content, AU ratio (R-AU), GC ratio (R-GC), Minimum Free Energy (MFE), Minimum Free Energy of the Pool (MFEE), Frequency of the MFE Structure in the Pool (Freq), Diversity of the Pool (Div), Minimum Adjusted Free Energy (AMFE), Minimum Free Energy Index (MFEI).

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-bantam	ACGAAACUGGUUUUCACAGUGAUCCAGCAGAUUGCUCAAAGUCUGAGAUCAUUGUAAAAACCAAUUUUGU	70	18,57143	32,85714	18,57143	30	37,14286	62,85714	0,913043	1	-29,4	-29,4	0,496236	1,25	-42	-1,13077
pmc-mir-1	AUUAUUACAUAUCUGGUAUGUUUGUGGAGUUGAUGUGUUCAUGCUUCUUUAUAGUUUGACCUUUUCCCUCacacaacauaaagaugagaaaUUCU	95	13,68421	12,63158	12,63158	38,94737	26,31579	51,57895	3,083333	1,083333	-18,5	-17,2	0,045655	8,13	-19,4737	-0,74
pmc-mir-1000	ACUACAGAUGUGGACUGGCUGGUGUGAUGGGACAAGACUUCAUCACUCUCUCUGCCGGUUUCAUCAUCUGUCAA	74	24,32432	21,62162	24,32432	29,72973	48,64865	51,35135	1,375	1	-28	-27,1	0,031686	6,94	-37,8378	-0,77778
pmc-mir-10031c	UUACUGAUGUUAUUACUCGUCCAUGUCUUCCUCUUCGUCCUUAUGAUCCCUAACAUAGGUAUGUGCACACCUGACACUGUGAGGAGUUUUAACGCAGGUA	100	19	22	24	35	43	57	1,590909	0,791667	-27,5	-27,5	0,437289	4,9	-27,5	-0,63953
pmc-mir-10046	CGCCGUCCUGCGAGAUCUCACCUGUCUAACGCGCAUGCGCCUCUCAUCACCACGCGCAACGGUUCCGCCUGACAGGGCGACUUGUCGCAGUGACGACA	98	25,5102	18,36735	37,7551	18,36735	63,26531	36,73469	1	0,675676	-37,2	-35,3	0,365188	5,25	-37,9592	-0,6

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-10055	UUGCUCCUGCAUCUGCAAGCCUGGGCCAGGGUACCccgggugggagggagggaggacuuuuuuuuuccucagguAGCGAUGACUGGAGACG	91	18,68132	10,98901	16,48352	10,98901	35,16484	21,97802	1	1,133333	-33,1	-29,2	0,094013	18,78	-36,3736	-1,03437
pmc-mir-10089	UCCAGCAUAAUUUGCUGUUUUGGUGAUAUACUCGUGUACAAUGAAAAUGAGCAAGACUAUGAUAUCAUCAUGAAAGCGAGAAAUGUUAGCUCAG	94	21,2766	34,04255	14,89362	29,78723	36,17021	63,82979	0,875	1,428571	-19	-17,9	0,02445	17,13	-20,2128	-0,55882
pmc-mir-10173	UAGAACUUUUUUCUUCCCACCAUUCCUACACGCCUACUUCUUGCGGAAGGAAGugaggaaggaaaacaaauuaaa	75	10,66667	16	21,33333	22,66667	32	38,66667	1,416667	0,5	-22,1	-21,7	0,383116	2,01	-29,4667	-0,92083
pmc-mir-10249-1	UGAAGAAGUGACUAACGCUGAUAACAGUCGAUGGACGAGGUAGAguugguggugcgguggugguugCCAGCGGAGCAGUUCUGAC	85	23,52941	23,52941	14,11765	12,94118	37,64706	36,47059	0,55	1,666667	-23,1	-18,2	0,173316	22,89	-27,1765	-0,72188
pmc-mir-10249-2	AGAAGUGAACAAGAAGUGACUAACGCUGAUAACAGUUGGUGGACGAGGUGGAguugguggugcgguggguugCCAGCGGAGCAGUUCUGACACUUGU	99	25,25253	24,24242	13,13131	15,15152	38,38384	39,39394	0,625	1,923077	-27,2	-13,3	0,036951	27,09	-27,4747	-0,71579
pmc-mir-10256	UAGCCAGUAUAGAACAGUAGCCAGGCAUGUGAGCACCACAAGCUAUGUAUAUAAAACUUAUGGUACCCCUAUCACAUGUCUGCUAUGGCCUUCCUGGUAU	100	20	29	24	27	44	56	0,931034	0,833333	-30,5	-29	0,074934	11,46	-30,5	-0,69318

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-10267	AGAGCUUUGUCGGUUUUUGGCUGUGAGGGAGACCAGAUCAAACCAccgcccccacacacaccaaaaaagcAAGGCAAU	78	20,51282	19,23077	11,53846	16,66667	32,05128	35,89744	0,866667	1,777778	-21,7	-15,8	0,291539	8,02	-27,8205	-0,868
pmc-mir-10492a	gucauugucauugucaucgcugucgucaucgucgucguuuaGCUCUCUGAUUUAUGUAGCAGACGAGAGGCAUGAGAGACGAAUUGGACAGAAAUUCU	98	16,32653	18,36735	9,183673	14,28571	25,5102	32,65306	0,777778	1,777778	-22,8	-11,64	0,059506	28,54	-23,2653	-0,912
pmc-mir-10508b	agagagagauauguaaAAUGUCAGGAAAGGUAAACAGGUGUAAUCAUUCAAUACCUCUGAACAGCAUUUCGUGACUUCUUUAAAUAUCUUCAGU	94	13,82979	27,65957	14,89362	26,59574	28,7234	54,25532	0,961538	0,928571	-19,8	-16,4	0,034776	12,65	-21,0638	-0,73333
pmc-mir-10509	CUGUGUCACGUACAGUAACACUGAUCGUCACGACUCCACCGUGACAUGACGAUGUACAGUGAGUGACGUGACAUGA	76	25	27,63158	25	22,36842	50	50	0,809524	1	-27,4	-24,3	0,170278	7,05	-36,0526	-0,72105
pmc-mir-10578	CUCCUUCAGCAGUGAGUCAGUCUCACGCUGCAUCCCUUGUGACGGUGGCUGCGAGACUGAGACACAUGGCAUAAAGACA	79	26,58228	24,05063	27,8481	21,51899	54,43038	45,56962	0,894737	0,954545	-24,8	-24,8	0,024862	11,14	-31,3924	-0,57674
pmc-mir-10611	GUCAUGAUGAUUAGCUUUCACUGAUUUCUAAGGGUUCUAAUAGCAUCACCACUCUUGAUUUCCCUUUUUCACACAGUGGGaaauuuaaucuucauuuu	98	14,28571	18,36735	18,36735	30,61224	32,65306	48,97959	1,666667	0,777778	-20,3	-19,5	0,141481	6,16	-20,7143	-0,63438

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-10639	GAUAAGCUUAUCAUACGUAACCCAUACAAAGGUUAUGGAAGUGAGGAGAAGAACAAGAAGUUCUCUCCCAUGGGUAUUUGCCAUGAUAGCUUUGG	95	24,21053	32,63158	16,84211	26,31579	41,05263	58,94737	0,806452	1,4375	-29,6	-26,6	0,069014	9,88	-31,1579	-0,75897
pmc-mir-10770-1	GCGAAACCACAGAACACGACUGACUGAUACGAUCUCAAAGGGCUUUCUGCUAAAAAGCAUCCUUgaguuguugauguuguuguuguuguuuuguu	98	12,2449	22,44898	17,34694	13,26531	29,59184	35,71429	0,590909	0,705882	-26,4	-25	0,014385	16,81	-26,9388	-0,91034
pmc-mir-10770-2	cuuguugaugcuguuguuguuucccGCAGCCUCAAAGAUGCGGACCUUGUCAAGUAAGGGGGAGACAAAAAUACAGUAUCUAACCUC	87	17,24138	25,28736	16,09195	12,64368	33,33333	37,93103	0,5	1,071429	-28,5	-28,5	0,138113	8,95	-32,7586	-0,98276
pmc-mir-10892	GCCACGAAAAAAGACCAGUGGACAACCAUCGGUGAAUAAUGGAUGAAUAACAUCCUGUUUCUAUCAUCCGAUGGGCGAGCUGUGUUUGUUGUCGUCUU	98	24,4898	27,55102	20,40816	27,55102	44,89796	55,10204	1	1,2	-30,8	-30,8	0,239683	4,62	-31,4286	-0,7
pmc-mir-10972	CCGGUAGUGAUGACAACGAUGAGAACAAUGAUGAUUCAUCAACGAUGAUUUGCUAUGGUCAUCAUAUAUGUGUCUCAUAUAGUUUGCACAAUACAGG	97	21,64948	31,95876	16,49485	29,89691	38,14433	61,85567	0,935484	1,3125	-23,3	-23,3	0,070908	15,83	-24,0206	-0,62973
pmc-mir-10b-1	UGCCAAGUCGACCACGGCACCCGGUGUGUCCCUCCCACUGACCAGUCAUCUCGGUACUCGGGAGGUCAAAUCGACUGAGGUCAGGGGUCGAAUUGACU	98	28,57143	20,40816	30,61224	20,40816	59,18367	40,81633	1	0,933333	-34	-32,9	0,060236	18,2	-34,6939	-0,58621

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-10b-2	GUGAGAGUGUGAGUGGCUUUACCCUGGAGAACCGAGCGUGUGUgaucaugacgucacaguucGUUUCUGAGGGUCAAGUCGCGAUACUCUGUU	93	27,95699	13,97849	13,97849	23,65591	41,93548	37,63441	1,692308	2	-40,6	-40,6	0,262649	9,81	-43,6559	-1,04103
pmc-mir-11054	uguuuguaguguuuguUGCCAGAGAUUUAGCAGAUAUGAUGGAAAcugcauuuucuuguuuucacacaUUAGAAUCUAAGAGAAACAGCACUAUCAACCA	100	12	26	10	13	22	39	0,5	1,2	-21,4	-15,8	0,089496	35,03	-21,4	-0,97273
pmc-mir-11280	AGAUACAGAUGUGUGACAUACaugucuaacccuaacccugaaugAGAGAAGAUCCGUGUGUGAGGUUGGAUGUAGGUGUAAACAUCUUACAC	92	21,73913	23,91304	9,782609	19,56522	31,52174	43,47826	0,818182	2,222222	-21,9	-21,2	0,135428	13,09	-23,8043	-0,75517
pmc-mir-1175-1	UGAUCAGGUGUGUUAUGUAGUGGAGAGAGUUUUAUCUCAUCAUGAGCCUCAGAAGCAGGUGAGAUUCAACUCCUCCAACUGCAGGCUAUACCUGAAAC	98	24,4898	27,55102	20,40816	27,55102	44,89796	55,10204	1	1,2	-42,7	-38,6	0,079856	5,62	-43,5714	-0,97045
pmc-mir-1175-2	CACACGGAAGUGAAGAGAAGAAUGAAGAAGGAGGCUAUAGCGAAAUUAUagguauuucuuuuuuaucaucaucaucaucaucucaucaucgucgucgu	98	15,30612	22,44898	5,102041	7,142857	20,40816	29,59184	0,318182	3	-20,9	-20	0,079297	13,02	-21,3265	-1,045
pmc-mir-1187	UGACUCAGGUAUCAAAUGuucuuuuauguguguauguguguguguauguguguauacguguguugCUAGCGGCUAAAAGCAAUAUGUGAGAGCGAGUGU	99	15,15152	17,17172	8,080808	12,12121	23,23232	29,29293	0,705882	1,875	-20,3	-19,5	0,064187	14,27	-20,5051	-0,88261

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-11922	CGUUUCGACCUGACAGAUGCAGUUGGAUCGUUGAUCUUUUCAGGAGAUCAUCGAUAUGUUCAUCGUUCAGGGUGAAGGA	79	27,8481	22,78481	17,72152	31,64557	45,56962	54,43038	1,388889	1,571429	-25,9	-25,9	0,042274	4,91	-32,7848	-0,71944
pmc-mir-12	UGGUGGUCAGACUGUGAGUAUUACAUCAGGUACUGAGAAUCUAACAAGCUUCAGUACCUUUUGUGAUAUUCUUAGUCUGCCAUUU	85	21,17647	24,70588	17,64706	36,47059	38,82353	61,17647	1,47619	1,2	-41,7	-40,3	0,196235	2,83	-49,0588	-1,26364
pmc-mir-12096b	GAUAUGCUACUUCGUGCAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAGGUAGGGCAAAAUUAUCACCGAUUACCCGAGGUAACGAUCAA	94	22,34043	29,78723	20,21277	27,65957	42,55319	57,44681	0,928571	1,105263	-32	-31,6	0,104639	7,62	-34,0426	-0,8
pmc-mir-12228	UCCCCACUGCUCCCCCAACAUCCCCCCACCAGCGCCGgcuaccccacccacccugcUGCGGGGGACGAGCUCGGAGGAGUCGUGGUCU	88	22,72727	11,36364	32,95455	11,36364	55,68182	22,72727	1	0,689655	-33,2	-19,6	0,075853	14,44	-37,7273	-0,67755
pmc-mir-1224	uugaucuucaccuccucucuccucccuucaaCUAAUAUGUCUGAGGUGUCAAUCAUGGGCUGUUUGAAGUUAGCGAAAGAGUUGACGAAGAUAUU	95	18,94737	20	7,368421	21,05263	26,31579	41,05263	1,052632	2,571429	-24,7	-21,1	0,129321	12,41	-26	-0,988
pmc-mir-12245	CCUUCCCCCGCCCCGCCCGGCUCAAGGUCAGCGCGCCAGAGAGCAAUUCGUCCAGGUAGACCAGGUGCUUGAACCCGAGAGGCGCGGUGGCGACAG	96	32,29167	18,75	36,45833	12,5	68,75	31,25	0,666667	0,885714	-33,6	-33,1	0,051978	10,23	-35	-0,50909
pmc-mir-12286	ACGCUGCUCAGCACGUCACUCUAUCAUUUCACAGUCUCGUGGGGC	80	31,25	18,75	25	25	56,25	43,75	1,333333	1,25	-28,6	-19,19	0,08512	19,63	-35,75	-0,63556

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UGCAGGGCUGAGUAAGCUGAGUGGCUGAGUAGGCU															
pmc-mir-12287	GGCUGUGGGGGUGCAGGUGGGCACACAUUCAUGGGAGGGGGCCCCCCCAUCAUAGGAGGCCAAGGUGCCGGCCCCAUCAUAG	82	39,02439	18,29268	28,04878	14,63415	67,07317	32,92683	0,8	1,391304	-31,7	-15,9	0,099725	26,92	-38,6585	-0,57636
pmc-mir-12292	GUUUCUGACGAUAUGGGGUCUUGGGGUCAUGCAGGAAGAGAUCUACGCGCUCGUGAACGACCCCAACCACCGCGAGCAUCAGAUUC	86	29,06977	23,25581	26,74419	20,93023	55,81395	44,18605	0,9	1,086957	-26,4	-25,2	0,09755	8,86	-30,6977	-0,55
pmc-mir-12293	gcugcuggugcugccGUCUCCUUUCUACAUCCGCCUCUUGAUCUACUAUAACUACGAGCACGAGGAGAUGAUAGAGAGGAGAUCGGCCAUCUCCCGCCUG	100	19	20	25	21	44	41	1,05	0,76	-39,4	-30,9	0,181223	7,86	-39,4	-0,89545
pmc-mir-12296	ACUGCAGUGCCCUGUGUGGUGUGGGGAGUgaggggcaggggcaggggcagggggCACACGACAUUACGGGCACAGCGGG	79	26,58228	12,65823	16,4557	12,65823	43,03797	25,31646	1	1,615385	-33,3	-33,3	0,678283	1,48	-42,1519	-0,97941
pmc-mir-12321	AAUGCGCCCUgcccucccuuccccucuccuccacccucaCACCGUUACUACCGAUUGUGAGAACGGGACCACAAGGAGGAACAACGGUAGGGCGCGAC	98	21,42857	20,40816	19,38776	9,183673	40,81633	29,59184	0,45	1,105263	-38,7	-38,7	0,150689	6,68	-39,4898	-0,9675
pmc-mir-12327	caacuucuuuucucucuuucccucucucuuugcUUUAGUGUUCAAAUGUACACAAGGUAUAAAGAAAGAGGAUUGGUGAGCGAGCAGGGAAGcaa	95	20	23,15789	5,263158	13,68421	25,26316	36,84211	0,590909	3,8	-29	-28,1	0,068017	8,35	-30,5263	-1,20833

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-12339	uuccaucuuuuucugcCAUGUUUAUCUUCACUGCUGGAAAGAUUGUUCUGGAUUGCCACUGGAGUGCAGUGCAGAGAAACAUGGCAUAGAAAGAUGGAA	99	23,23232	25,25253	13,13131	22,22222	36,36364	47,47475	0,88	1,769231	-43,9	-41,47	0,037169	11,26	-44,3434	-1,21944
pmc-mir-12358	CCCCAUCUCCUCCCUUCUACCUAAUCAUCUUAAAAUCAUCUCACUGUCAUCCGCUUUAUCUAUGAUUUCAUCGGGUAagguaaagggagguaauguaa	98	6,122449	17,34694	26,53061	28,57143	32,65306	45,91837	1,647059	0,230769	-26,7	-26,7	0,382757	2,63	-27,2449	-0,83438
pmc-mir-12396	CCGCGCAGUGUGAGUGACUGAAAAGAGAGACACAGACAAGAAAGAUAUUCUUUAAAACCUCUCCUUUCGUCAGCUCGCACUGCGAAG	87	22,98851	32,18391	24,13793	20,68966	47,12644	52,87356	0,642857	0,952381	-35,8	-35,8	0,255939	5,41	-41,1494	-0,87317
pmc-mir-124	cuaUCGCUUUCAUCCGUGACGACACCGUGUCCUCCCGGCAAGUGACGUUCCCGcgacuguugcugcugucgcCAGGAGUCAAGGCGAACA	90	20	15,55556	25,55556	14,44444	45,55556	30	0,928571	0,782609	-27,7	-19,2	0,073181	30,63	-30,7778	-0,67561
pmc-mir-12410	GUCCAUCGUAAAcgguugcuguuuuuuuuuucuucguggaACUAAAGAAACCcgggaagaaggaagaaaggaaacuGAGAUCCGGUCACGUGACG	96	10,41667	15,625	11,45833	7,291667	21,875	22,91667	0,466667	0,909091	-25,7	-23,6	0,044841	16,09	-26,7708	-1,22381
pmc-mir-1273h	GUGCUGGGUGAGUACUCGGUGCGCGUGCUGUGGGACACAGGUGUAGAGAGCACGUGCAGAGCAGGGCGGGACUGCAGACUCGACCUCAGGAU	92	41,30435	19,56522	21,73913	17,3913	63,04348	36,95652	0,888889	1,9	-37,1	-12,11	0,089734	23,69	-40,3261	-0,63966

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-1277	GACGUACUGCAAGGAucccuguauauauauauauguacucuaCAAGGGAGUCGAUGUACGUACAGA	66	19,69697	19,69697	10,60606	9,090909	30,30303	28,78788	0,461538	1,857143	-19,3	-18,4	0,217022	13,76	-29,2424	-0,965
pmc-mir-130b	AAUUCUGAUAAGCGAAACAUAUGUUUCAUCUAUGUUGGUAUAACCGAUUACUAAACAUAUGCAAAACAGUGCaauaaugaaaguauuuuuguucaGACGA	100	13	29	13	22	26	51	0,758621	1	-19,9	-19,9	0,042179	8,59	-19,9	-0,76538
pmc-mir-1322	GGAAUAUCUGGUCGUAGCUCAgagcugaugcugcugcugauggcuaUCAAGUAAAUGAUGCUGGUCAACAGAUGAAAUAUGGCACACCAAGAUAUCUG	98	17,34694	25,5102	13,26531	18,36735	30,61224	43,87755	0,72	1,307692	-24,5	-22,2	0,016132	23,72	-25	-0,81667
pmc-mir-133c	UAACUACAGCUGGUUGAAGGGGACCAAAUGAAAGGCUCUUCAACAAUUUGGCCCGAGUUCAACCAGCUAUAGAAG	75	24	33,33333	21,33333	21,33333	45,33333	54,66667	0,64	1,125	-30,4	-28,9	0,184949	4,61	-40,5333	-0,89412
pmc-mir-1376	CUCAGCUGCAACAGACUCUGACAUUAGCAGAUCUCAAGCUCAGCAGCACUCUUUGAGCUUGGAGGAUGUUAGUCAUGAUGCUUUGCAGACUCUG	94	23,40426	24,46809	24,46809	27,65957	47,87234	52,12766	1,130435	0,956522	-31,4	-26,8	0,072951	8,03	-33,4043	-0,69778
pmc-mir-137b	AGUUCCAGUCGGUUACGGGUAUUCUUGGGUAAAUAAUACAUUGAGUAGUUGUUAUUGCUUGAGAAUACACGUAACUACCUGGACGU	86	24,4186	26,74419	15,11628	33,72093	39,53488	60,46512	1,26087	1,615385	-33,1	-29,3	0,022171	10,78	-38,4884	-0,97353
pmc-mir-13b	uaaaauacacuuuaacUGACGUCUGGCUUCUGAUGUCUCUGAAGGGU	95	18,94737	11,57895	13,68421	22,10526	32,63158	33,68421	1,909091	1,384615	-25,4	-18,3	0,096917	31,5	-26,7368	-0,81935

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GCGAGAGUUUGUUGUUCAGACUCAUCAAGCCUcguuaaaauguuugug															
pmc-mir-1421am	GGUGCGUCAAGGCGGUCAGUAAGACGUCCGCCACCUUAGGCGUUCAAACGGAUUUCGAGGAGGUCGUAGGUCAUCCUGUAGACGACUUGGCGCUGG	96	34,375	19,79167	23,95833	21,875	58,33333	41,66667	1,105263	1,434783	-31,8	-26,1	0,047723	21,41	-33,125	-0,56786
pmc-mir-1422j	UGGGUGUUCUACUGUUUCAUGAUUUAUCAUAGGGAGAUGACACAUAAUCGAAAACUGCAUCUGGAUUAUGAUAUCAAGUGAAAUAUAUCAGAAAACUAA	99	19,19192	36,36364	13,13131	31,31313	32,32323	67,67677	0,861111	1,461538	-22,3	-22,3	0,232835	3,91	-22,5253	-0,69688
pmc-mir-143	GCACAUGACAGAGGGCCUCAUCGGUCUGGCUGAACACAAGAAGUAGAAAGUGUUGCACACAGUCUGAGAUGAAGCACUCGUUUGCAUGAUA	91	27,47253	30,76923	20,87912	20,87912	48,35165	51,64835	0,678571	1,315789	-25,3	-24,9	0,120125	12,48	-27,8022	-0,575
pmc-mir-149	acaugaaucgucuccccccccccccaccaccgcAAUACAUGGCGCAGUGCAAAGGUCACACCGCUAGUGCUGAGAGGGAGGGACGGGGGAUUCAAGA	96	25	19,79167	12,5	9,375	37,5	29,16667	0,473684	2	-33	-12,4	0,017431	29,49	-34,375	-0,91667
pmc-mir-150	ACAUGUCACCACAUGCGUGUGCACGCACAUAAAUAUGUCAGCUUAUCUCACAGAUACAUGUUGCUGGUACAGGCCUGGGGGAGACAAAU	89	23,59551	29,21348	23,59551	23,59551	47,19101	52,80899	0,807692	1	-25,1	-25,1	0,081451	10,2	-28,2022	-0,59762
pmc-mir-153	AUACCCUCUUCCCAGGCAGCUUUUGUGAUUCAGCAAUUGUACAGC	87	24,13793	27,58621	22,98851	25,28736	47,12644	52,87356	0,916667	1,05	-37,9	-37,9	0,256161	6,4	-43,5632	-0,92439

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UAUCAAAUUGCAUAGUCACAAAAGUGAUCGGGAGGCGGGGAC															
pmc-mir-1551	CUUUUGAUCUUAAUGUGGUAAAACGCAUUUCAGGUUUAUGAGUUUGUGUUGCUGGGACAAUGACUGUUUUAGGCCAUCUGUACAUCAACCA	91	21,97802	24,17582	16,48352	37,36264	38,46154	61,53846	1,545455	1,333333	-19,4	-19,4	0,255295	14,12	-21,3187	-0,55429
pmc-mir-15c	UUGAAGAAUAUCUCUUAUCCCAUAGGACAGCUAUUUCAAGGAAUCAUGUGACCAGCAGACCAUUCUGGGCUAUGGGGUCAGAGCUAUUCUUUUU	94	21,2766	26,59574	20,21277	31,91489	41,48936	58,51064	1,2	1,052632	-29,6	-28,4	0,088206	10,15	-31,4894	-0,75897
pmc-mir-1603-1	AUAGCAGAAAUCAAAACAGCCCACAUUUUAUGUCAAACUUAUUACACAGAGAUAUUAAAGCaaugugguuuguuuuguguugcgCA	86	8,139535	32,55814	15,11628	17,44186	23,25581	50	0,535714	0,538462	-20,1	-20,1	0,033978	10,39	-23,3721	-1,005
pmc-mir-1603-2	GGGUUAGUGUUUGACAGAGGAUUGCAGCUUGCAauguugguuugguuuguuuuuuuucuaaacacagCGACAAUAUCUUGUGAAAUAGUAAGUU	94	18,08511	18,08511	7,446809	20,21277	25,53191	38,29787	1,117647	2,428571	-20,1	-19,5	0,022965	9,06	-21,383	-0,8375
pmc-mir-1632	GAUGGCUGUUGACACACUCGAGUGACAAGACAAAGGUCUGAUUUCCUUGCUUGUUUUUGGAUGAAAUUAUUUUGCGACGCAGCUGUUCACAGCCUCU	97	23,71134	22,68041	20,61856	32,98969	44,3299	55,6701	1,454545	1,15	-26	-26	0,016941	14,04	-26,8041	-0,60465
pmc-mir-1642	GUGAACACUUAAAUAACUGCUUGUCCCCUCUACCCUGAUAGAGGG	83	25,3012	27,71084	16,86747	30,12048	42,16867	57,83133	1,086957	1,5	-20,1	-14,7	0,16303	24,29	-24,2169	-0,57429

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AAAGUGGUGAGAGGCUGUCAGUUUUGUAAGAUAUUUAG															
pmc-mir-1677	UGACCAUCACCUUUAUCUCUCUGCCGUCGAGGUAGCGCGACACGAACUCCACUCCAUUCUGUGCGUUGACUUCAAUAGGAGAGACAGGAGUGAUGGUGA	99	25,25253	23,23232	26,26263	25,25253	51,51515	48,48485	1,086957	0,961538	-36,2	-35,3	0,026404	8,83	-36,5657	-0,7098
pmc-mir-17	CACGGCGAUGACUGGAGUCAUUAGGGAGGCGCACGCGCGGCACUAGACCUUAACUGCACUGCAAGCACCGCCACA	75	29,33333	25,33333	32	13,33333	61,33333	38,66667	0,526316	0,916667	-24,1	-19,8	0,261623	4,23	-32,1333	-0,52391
pmc-mir-1728	UGUGACUGUUCUGCAGUGACUGUAUGCGUCCCAUGAAUCAUUGGUUGAUCCAUUGGAAGAAACAUGACUGGGUACAUCCUG	81	25,92593	23,45679	19,75309	30,8642	45,67901	54,32099	1,315789	1,3125	-18,9	-18,9	0,107603	12,57	-23,3333	-0,51081
pmc-mir-1744	UUUGAGAAGAGGUACUUCAACAGGAGCAAGUCAAGGCAUGCUUCUUUUUAACACCAGUCUUCUCUGC	67	20,89552	26,86567	22,38806	29,85075	43,28358	56,71642	1,111111	0,933333	-19,2	-19,2	0,273743	3	-28,6567	-0,66207
pmc-mir-1775	guaaaaauuaaggCAAAGAAAUCCUGUAGCCAGAAGACAGCGGAGGACCAGAGUUGUCUGUGCAGCCUGGCUACAUGUUUCUCUGUCCAAGAUUUUGAG	99	23,23232	23,23232	19,19192	21,21212	42,42424	44,44444	0,913043	1,210526	-34,9	-34,9	0,331788	5,38	-35,2525	-0,83095
pmc-mir-1781-1	GUUUGAAAGGAUUUAAAUCAUCCAGCUGCCUUUUUGACUGUUUAGCAGUCUCCAACAGGAGUGGAUUAAUUUGAGUAAUUUUCAUUC	87	19,54023	26,43678	16,09195	37,93103	35,63218	64,36782	1,434783	1,214286	-23,1	-23,1	0,071767	5,41	-26,5517	-0,74516

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-1781-2	GUUUGAAAAGAUUUAAAUCAUCCAGCUGCCUUUUUGACUGUUUAGCAGUCUCCAACAGGAGUGGAUUAAUUUGAGUAAUUUUCAUUC	87	18,3908	27,58621	16,09195	37,93103	34,48276	65,51724	1,375	1,142857	-23,4	-23,4	0,079721	4,68	-26,8966	-0,78
pmc-mir-1896	CAUUCUUCCUCAAACCAAGUGUGUAUUAGAGAUAAAUGGGGUCUCAAAGGACAUAGACACCGAGUGUAUCCACUCAUACGGUGGugggugaggaggagga	100	19	26	18	21	37	47	0,807692	1,055556	-32,2	-31,6	0,027684	15,11	-32,2	-0,87027
pmc-mir-190	ACCCGCUUCUGCUAGAUAUGUUUGAUAUAUUUGGUGCAGUUGUCGCGACGACCAAGUAGUCAAACAUGUCAGAGCAGCGACG	82	25,60976	25,60976	21,95122	26,82927	47,56098	52,43902	1,047619	1,166667	-32,5	-32,3	0,080048	9,84	-39,6341	-0,83333
pmc-mir-1951	UUAGUUUUGUGCAUCAAUCACCAUCAUUUGUGUAAACGUCUUCCCUGACUUAUUUGGAGCCCCCAAAUGUAGUGGAGACUGGUGGCGAAACCUU	94	21,2766	23,40426	22,34043	32,97872	43,61702	56,38298	1,409091	0,952381	-23,5	-14,8	0,063911	17,89	-25	-0,57317
pmc-mir-1955	GGAGGAGCAUUGCAUGCUGGACGCGCGUGAUCAGGUCCUGGCGGUGUCAGUGACAGUCAUUGCCAUGACAGCAGCGUGCACAGGUCAUGUGACUCCACC	99	33,33333	20,20202	26,26263	20,20202	59,59596	40,40404	1	1,269231	-40,3	-34,2	0,115974	33,97	-40,7071	-0,68305
pmc-mir-196a	AAACAUCCUUUUUGCACACGACAAUGACCUGGUGGCCGACGGCAACAAGAAACUGUGACUGAGCCACAGUGUAUUGAUAUUGAAUGCAGACAGGAUGCCA	100	24	32	23	21	47	53	0,65625	1,043478	-31,3	-31,3	0,07221	6,07	-31,3	-0,66596

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-1984	UGUCGCCGUGCCCUAUCCGUCAGGAACUGUGAUCUCGCGAAACACAGGGUCUGGCGGUUGGGCCUCGGCGCUG	73	34,24658	13,69863	30,13699	21,91781	64,38356	35,61644	1,6	1,136364	-37,9	-37,9	0,133827	2,65	-51,9178	-0,80638
pmc-mir-1985	UGCCAUGCCAUUUUUAUCAGUCACUGUGUGUUGUGAAAGUcacagugaugaugauaauggcuUGAUG	67	16,41791	13,43284	10,44776	26,86567	26,86567	40,29851	2	1,571429	-28,6	-28,6	0,287364	2,77	-42,6866	-1,58889
pmc-mir-199	CAUUGCAUGUCAAAACUAAGGGAUGUUUACCUAUAUUUGUAGAGACACACAUGACAGUAGUCUGCACAUGCUAUAGAUUUUGGGGAUGCAGCA	93	22,58065	31,1828	17,2043	29,03226	39,78495	60,21505	0,931034	1,3125	-20,3	-17,4	0,056275	14,69	-21,828	-0,54865
pmc-mir-1990	GGCUGCGAGAUGUUCGUGCAGUAAGUUGAUGGGGUCCCAGGUAGAUCUACCCCUCCCGGGACUACGUCAACGUACUACCACGCGCAUCUCCAAGU	95	27,36842	21,05263	29,47368	22,10526	56,84211	43,15789	1,05	0,928571	-44,2	-43,8	0,106149	4,96	-46,5263	-0,81852
pmc-mir-1994	CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUGUGUGAUGACGUCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGACACCGCUAA	90	30	18,88889	25,55556	25,55556	55,55556	44,44444	1,352941	1,173913	-41,1	-41,1	0,501637	4,6	-45,6667	-0,822
pmc-mir-1994a	CAAAAGCUAUUUCUCAGGGGCGGUCACUCUGAUCUCCAUGUGCUCGCACCAGCAUGAGACAGUGUGUCCUCCCUUGAGGGAUGGGCUUAUA	91	26,37363	20,87912	26,37363	26,37363	52,74725	47,25275	1,263158	1	-44,3	-44,3	0,178398	5,12	-48,6813	-0,92292

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-1a	GGCACAUACUUCUUUGCUAUCCCAUAUGUUCUUGCAAAGCUAUGGAAUGUAAAGAAGUAUGUACU	65	18,46154	29,23077	18,46154	33,84615	36,92308	63,07692	1,157895	1	-23,7	-22,1	0,075024	6,51	-36,4615	-0,9875
pmc-mir-1b	CCAGUAGAGACAAGUGAUGUUACACAGGUUAUGCGUAUAAACAAAGGUAUGGUCGCAUUAUAUUACGUGUCCUAAUCAUACUUCUUUACAUU	92	18,47826	31,52174	17,3913	32,6087	35,86957	64,13043	1,034483	1,0625	-19,8	-19,2	0,145182	8,89	-21,5217	-0,6
pmc-mir-1c	AGUACAUACUUCUUUACAUUCCAUAGCUUUGCAAGAACAUAUGGGAUAGCAAAGAAGUAUGUGCC	65	18,46154	33,84615	18,46154	29,23077	36,92308	63,07692	0,863636	1	-27,1	-26	0,4687	2,23	-41,6923	-1,12917
pmc-mir-2001	GCUGCCAUUUUGUGACCGUUACAAUGGGCauugacagaaaaacaaaugcucCAUGUGGUGUUCACAAGAUGGCGGU	76	22,36842	13,15789	14,47368	21,05263	36,84211	34,21053	1,6	1,545455	-36,2	-36,2	0,193282	4,14	-47,6316	-1,29286
pmc-mir-200b	AAUUUCAUACGUCAUCUUACCUGACAGUCAACAGCAAACAAAGUUGCUCCUUUGGUCUUAUUGACACAGGUGAUGAGAUUGAAGGU	86	19,76744	30,23256	19,76744	30,23256	39,53488	60,46512	1	1	-21,8	-21,6	0,126933	11,76	-25,3488	-0,64118
pmc-mir-201	AGUUGCUGGAAGAGCUGCAGGCCACUUCCUAUGAACAGCGCCUUUCUCACGUGCGGUGUCCUCAGCACCUCGCCCAGCACGA	82	25,60976	19,5122	34,14634	20,73171	59,7561	40,2439	1,0625	0,75	-25,8	-25,8	0,163281	11,03	-31,4634	-0,52653
pmc-mir-2017	GAAGUGCACGUGGUGUUGACAGUUACCUGUGCACCUGUGCACGUGGUGUUGACAAUGACCUGUGCACGUG	70	32,85714	18,57143	21,42857	27,14286	54,28571	45,71429	1,461538	1,533333	-29	-29	0,16383	21,21	-41,4286	-0,76316

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-204	GUUCUUUGUGUCACCUCACUUCCACUUCCGGUGAAAUCCGGAUUGGUAAUGCAAGGACAGCAAAGGGA	68	25	25	23,52941	26,47059	48,52941	51,47059	1,058824	1,0625	-21,9	-21,9	0,137453	4,4	-32,2059	-0,66364
pmc-mir-2064	AUGAGAUUCAGAAACAUGUCCAUCACAAGCUGCUGAAAGAAGCAGCACUGUGCAAGAUACUUUUUCCCCGAAUCUGGG	78	21,79487	32,05128	23,07692	23,07692	44,87179	55,12821	0,72	0,944444	-23	-22,9	0,480687	3,28	-29,4872	-0,65714
pmc-mir-2155	GUGUGCUCUGGUGCUCUGUGAAUGUAAGUUCUUUGUGGUAGAAUGAAGUGACACUGUUUUACACUCUACUGGAGCCUCAAGCAAAG	86	26,74419	23,25581	17,44186	32,55814	44,18605	55,81395	1,4	1,533333	-20,5	-17,1	0,059402	25,69	-23,8372	-0,53947
pmc-mir-216a	guguuugucUAAUCUCAGCUGGUAAUUCUGAGUGGAGUUCUGCACACCUCAAGUUACUAGCCGAGAUUACAUAAAUAU	78	16,66667	26,92308	17,94872	26,92308	34,61538	53,84615	1	0,928571	-30,5	-30,5	0,146188	3,64	-39,1026	-1,12963
pmc-mir-216b-1	CGUAGGAAGAUGAGCCAGGAUUACAAGAUUGUUGGGCCUGCGGCCCUAAUAUCAGCUGGUAAUCCUGAGUGAGCUUGCCUCAG	83	30,12048	24,09639	21,68675	24,09639	51,80723	48,19277	1	1,388889	-31	-30,7	0,290814	5,92	-37,3494	-0,72093
pmc-mir-216b-2	GCGGGACCACUUGCCCUGUGGAGCGAGGAUGCUCUGGAGGACGGCUGGACCUUUAUUAAAACUCGCCGCUACACAGUGGCUACUGGGUCCACA	93	31,1828	20,43011	27,95699	20,43011	59,13978	40,86022	1	1,115385	-34,1	-34	0,183887	24,97	-36,6667	-0,62
pmc-mir-216b-3	ACACAGUUACCUGUAGAUUGUGUCACUGACUAGUAACACAGUUACCUGUAGAUUGUCACUGACUAGU	91	17,58242	29,67033	21,97802	30,76923	39,56044	60,43956	1,037037	0,8	-20,7	-20,6	0,056424	15,43	-22,7473	-0,575

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AACACACUUACCUGUAGACUGUUA															
pmc-mir-2223	AAGUGUUUGUAACAAACAUCCUGUGUCUGUACUGUACACAUGACGUGCUACAGUAUUUGUACUCAUGUAAGUAUUUGUAACAAACAUCC	89	16,85393	30,33708	19,10112	33,70787	35,95506	64,04494	1,111111	0,882353	-23,8	-18,8	0,195676	11,23	-26,7416	-0,74375
pmc-mir-2238i	GUACCCGCACGUGGUCAUCACCAAGUUCCGGUCCAAGGAAGGCCGUUCACUCAUCUCGGAGCUGAUCAUCGAGCGGGGCC	80	28,75	20	32,5	18,75	61,25	38,75	0,9375	0,884615	-27,6	-25,2	0,123572	5,73	-34,5	-0,56327
pmc-mir-2284c	AUAUGAUGAUAAGGUGUACAUGAAACAGCUAAAAAUCUAAGUGAACUGUCAAGGUGGAAAAUUUUACAUGUUGCACAGUCACCUUGCUCAGCAGGA	96	21,875	36,45833	15,625	26,04167	37,5	62,5	0,714286	1,4	-22,9	-19,8	0,195896	4,07	-23,8542	-0,63611
pmc-mir-2298	CUACCUGCUGGGUUGGUCAGGGUAGUGGGAUACAGAACGAUGACUGAUGUUGUUGUUCAAGAACCAAACCCUGACCAUGGAGCGAGCAGGCGC	93	32,25806	24,73118	21,50538	21,50538	53,76344	46,23656	0,869565	1,5	-44,5	-41,7	0,259982	2,97	-47,8495	-0,89
pmc-mir-2304	AGUUCAGCGGUUGGUAGUGGAAGUGGUGUGUAGGUGugguugugugguuguguggGCGGUCAGUGUGCUCCUGCACUUCACACGACAAACACGUUGACAA	100	27	18	16	20	43	38	1,111111	1,6875	-32,8	-27,8	0,059196	33,54	-32,8	-0,76279
pmc-mir-2361	CAUUCAAUGGAAGGUCGUUAUUCAUUAUCGUAAACAAAACACAUGGAAGacaguuuguguuguuuuguuuu	100	12	30	10	18	22	48	0,6	1,2	-25,8	-25,8	0,03599	8,11	-25,8	-1,17273

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	gcauaguaACAAAAUGAAUACAUUGAAUG															
pmc-mir-2371	AUGUUAUGUAUAGUUUUGUGUGGUGUAUAAGUuguauaaauuuuacagaaacaaCUUGUCCACCACUAAAAAAAGAUACUAAUGA	85	14,11765	24,70588	8,235294	27,05882	22,35294	51,76471	1,095238	1,714286	-23,4	-20,9	0,161131	8,21	-27,5294	-1,23158
pmc-mir-2470	GCCAGAAAAACCUAAGCAUGACAAAAUAUCAUGCUUGUGUGGUUUUGGAUACUUGGAUUUUUCUCCC	67	19,40299	28,35821	19,40299	32,83582	38,80597	61,19403	1,157895	1	-19,9	-19,9	0,542669	2,57	-29,7015	-0,76538
pmc-mir-2491-1	CAGUGUUGCAAAGUUUACCUGCUGCUAUUGUGACUCAGACAUCUCAAGGGCaccugcaacaacaacagcagcagcaacaacaacagcaacaaca	94	12,76596	12,76596	12,76596	15,95745	25,53191	28,7234	1,25	1	-25,1	-23,1	0,06655	7,31	-26,7021	-1,04583
pmc-mir-2491-2	GAAUGUUAAUGACGAUGACUGCUGUUGUUCCCUUGAAAUAGGCUAAGGUCUacuuaacaacaacagcagcagcaacaacaacaaca	86	15,11628	15,11628	9,302326	19,76744	24,4186	34,88372	1,307692	1,625	-24,4	-24,2	0,192125	18,27	-28,3721	-1,1619
pmc-mir-2491-3	GAUGGCGGCGAAGAAGUCCACGCcugaagaagaacaacaacagcagcagcaacaacaaggGUGGCGAGGGCUGCUUCGUCACCUGU	86	22,09302	9,302326	15,11628	10,46512	37,2093	19,76744	1,125	1,461538	-27,25	-26,3	0,182896	6,2	-31,686	-0,85156
pmc-mir-2493	AUCGCUGUGUGGGACUCGUGUUGAUCAUCGCGGUGCACUAAAUGUCAUCCGGCCAGACAUCAGACCAucgugcucacacacacacagccag	93	19,35484	16,12903	19,35484	17,2043	38,70968	33,33333	1,066667	1	-28,2	-25,4	0,239172	8,2	-30,3226	-0,78333

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-2505	GACCGUGUAUUCUCGcucucgugcacacacacacauacacacgcgcgcgcgcgagcaGACAUACCCGCUG	70	10	7,142857	12,85714	10	22,85714	17,14286	1,4	0,777778	-20	-18,8	0,634526	4,68	-28,5714	-1,25
pmc-mir-252a	CCUAACUUCUGGCCCAUUACUAAGUACUAGUGCCGCGGGAAGAUAACUUGUCAUGUCCCGUGGUUCUGGUCCUUACUACAGGGCGGAACUUGCA	94	24,46809	21,2766	26,59574	27,65957	51,06383	48,93617	1,3	0,92	-37,6	-37,6	0,059589	5,1	-40	-0,78333
pmc-mir-254	GUGUGUGCAAAUCUUUUGCAACAGACUUUUAAACUAUGGGUGUUUGGUGUUUGAGUGUGCAGAGUUGUUUGCAAACUCUUUGCUACAGAA	90	25,55556	23,33333	13,33333	37,77778	38,88889	61,11111	1,619048	1,916667	-21,5	-16,1	0,016973	23,25	-23,8889	-0,61429
pmc-mir-2571	UCCUUCUGCAUGGCCACUUCGCUGGUGUGAGUGGUGAUGAUGUUUACCACUCAGGACGAAGUCAAAGAGGCGAUCAGCAACAA	83	27,71084	25,3012	22,89157	24,09639	50,60241	49,39759	0,952381	1,210526	-29,2	-29,2	0,309071	7,4	-35,1807	-0,69524
pmc-mir-279	CUCUCUUAUCUUUGCGGGUGGCUGUGAGUCUAGUCCAUGUGACACUGCAAGCUCAUGACUAGAUCCACACUCAUCCACAAGGAUAGGACGU	91	23,07692	23,07692	26,37363	27,47253	49,45055	50,54945	1,190476	0,875	-39,9	-39,4	0,157559	5,72	-43,8462	-0,88667
pmc-mir-279b	AAGGGGACCAACCACUAGAGUGACUAGACUGAUAACCCCUAGAGAUGUUGUGCUUCAGUUAUCACUAACCAUUGUGGUGGAUCCCUGA	88	23,86364	28,40909	22,72727	25	46,59091	53,40909	0,88	1,05	-25,9	-23	0,049529	18,81	-29,4318	-0,63171
pmc-mir-279c	GUUUCUGCCGGAACAUGACCAGCAUCGUUCUGGAAGGAGGCACUCGGCUGGCGGGUGUUCGUUUCGA	100	36	18	24	22	60	40	1,222222	1,5	-38,8	-38,3	0,103385	8,74	-38,8	-0,64667

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUACGAGGGCGGGAUCAUCAGCUUCGGCGACAG															
pmc-mir-281	AAAUGACCUCACGCUGAAGGGAGCAUCCGUCGACAGUCAGAAAUGCAGCACUGUCAUGGAGUUGCUCUCUUUAcugaaaaggucaagg	88	20,45455	22,72727	20,45455	19,31818	40,90909	42,04545	0,85	1	-35,2	-35,1	0,187538	3,76	-40	-0,97778
pmc-mir-282	uaauauguuuuuugugguuguuuuuggCAGUCCUGUAGACAUCAAUACUAGACAUAGCCUAAAAGAGGCACCAAGAAUGAUAaca	85	11,76471	27,05882	14,11765	11,76471	25,88235	38,82353	0,434783	0,833333	-20,6	-20,6	0,141364	11,91	-24,2353	-0,93636
pmc-mir-2953	AGACCAUGCCUCAGACACUGUGCUGCUCCUGAACUUGUUCCUUCAUGCUACAGAAGUGUUCGUGGGGGAUGGAAA	75	26,66667	22,66667	24	26,66667	50,66667	49,33333	1,176471	1,111111	-23,8	-23,8	0,296458	3,88	-31,7333	-0,62632
pmc-mir-2a-1	AGGCAAAGCUAUGGUGCUGACCAAGUGACUGGGAUGUGUACCUAAAUAUCAUAUCACAGCCUGCUUGGAUCAGUAUUAUGGUUUUGAAC	89	24,7191	28,08989	17,97753	29,21348	42,69663	57,30337	1,04	1,375	-38,6	-38,2	0,276363	1,97	-43,3708	-1,01579
pmc-mir-2a-2	AAAAAGCAAAUGUGCAUGGAGCAAUCAAAGUAGUUGUGAUGUGUUUGUUGACUUCAUAUCACAGCCAGCUUUGAUGAGCUUCUUCACAGUUUCUUGCA	98	21,42857	28,57143	17,34694	32,65306	38,77551	61,22449	1,142857	1,235294	-34,2	-33,9	0,160179	6,48	-34,898	-0,9
pmc-mir-2a-3	UUAUGACUGCUGAGUUCACAUCAAAGAGGCUGUGACUUUGUGAUGCUGUUUCAAAUCACAGCCUGCUUUGGUGAGCUCUUUGCUGUCAGAU	91	24,17582	20,87912	19,78022	35,16484	43,95604	56,04396	1,684211	1,222222	-40,8	-40,8	0,328315	3,78	-44,8352	-1,02

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-2b-1	UGCAAGAAACUGUGAAGAAGCUCAUCAAAGCUGGCUGUGAUAUGAAGUCAACAAACACAUCACAACUACUUUGAUUGCUCCAUGCACAUUUGCUUUUU	98	17,34694	32,65306	21,42857	28,57143	38,77551	61,22449	0,875	0,809524	-24,8	-24,8	0,342782	4,57	-25,3061	-0,65263
pmc-mir-2b-2	GAAGCUAGACCAGCUCAUCAAAGCUGGCUGUGAUAUGACUGAACUCAACACAUCACAGCAUCUUUGAUGGGCAGAUCCUAGCAAU	85	21,17647	31,76471	24,70588	22,35294	45,88235	54,11765	0,703704	0,857143	-34,5	-34,5	0,221463	4,35	-40,5882	-0,88462
pmc-mir-2f	UCGGGUAGAUAAGCCAGCGAUGUGAGACUCCAGACGAUGGUGUGACUGAGGAGUUAGCAGACCAGUGUAUGUAUAUCACAGCCAUGCUAAUCUCCUCU	98	27,55102	26,53061	21,42857	24,4898	48,97959	51,02041	0,923077	1,285714	-28,7	-28,7	0,071984	7,94	-29,2857	-0,59792
pmc-mir-300	AGCGCUCGGUGGAUUAAGCGUGUCUUGCCCAUGUAUCUAUACAAGGGCAGACUCCAUCCCCAAGCCAC	68	23,52941	23,52941	30,88235	22,05882	54,41176	45,58824	0,9375	0,761905	-24,8	-23,9	0,250391	4,52	-36,4706	-0,67027
pmc-mir-302a	CUACACCCGUGGACCACUCCAUUUAUUUGAUaacaggaaagugcuuccuuUUAUUGGUGAUUUCACACUAUUAUUGAGAGGAACACCGGGUGGUG	95	17,89474	18,94737	16,84211	26,31579	34,73684	45,26316	1,388889	1,0625	-26,3	-26,3	0,019986	22,21	-27,6842	-0,79697
pmc-mir-302b	GAAACUAGCUUGACAGCCAAUGCAUAGCCAUCAGACAAAGUGCUUCCAUGUUUAAUUCAGUCCAUGGCUUUGUAUUAGCCUUAAAGCUGUAUA	93	18,27957	30,10753	21,50538	30,10753	39,78495	60,21505	1	0,85	-25,2	-25,2	0,130818	5,39	-27,0968	-0,68108

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-3057	agagugacUGGCUGUUCUUGAGAGAUGUUAGCAAGUUCAAAAUGUUAUGGCGCGAACCAACAUCCCACAGGCCCAGCUGUCACAAA	86	20,93023	26,74419	22,09302	20,93023	43,02326	47,67442	0,782609	0,947368	-24,4	-20,9	0,20764	8,46	-28,3721	-0,65946
pmc-mir-3071	AuaucaucaaaacaaauguGCAUAGUGUUACACAGAUACCAGGAUCAUGGAUUCAGCUGAUUUAGUGACUGaaacacuuucauuucuugauGAAGC	96	15,625	18,75	10,41667	15,625	26,04167	34,375	0,833333	1,5	-20,1	-19,8	0,178747	6,13	-20,9375	-0,804
pmc-mir-308	CGAGUGCGCAUGACAGCGCGCGCAGGAUUUGCUUGUUUUGAGCAACUCGGGUGGCGAUGUCCAUGACGCACAGC	74	33,78378	18,91892	25,67568	21,62162	59,45946	40,54054	1,142857	1,315789	-32,3	-32,3	0,25533	2,19	-43,6486	-0,73409
pmc-mir-3084a	UAUAUGUAUUGAAGGUUCAUUAGCCUUGUGGUCAGAGCACUCCAAUUUGGAGCAAAAGGUUACUGGUUCAAGGUCUGCUUGAACCUAUGACUAAAUaau	99	22,22222	27,27273	16,16162	31,31313	38,38384	58,58586	1,148148	1,375	-27,5	-22,4	0,08872	12,04	-27,7778	-0,72368
pmc-mir-31	GUCUGUGCUGGUCuacauuuuguaucuguuugaaCUGUCUGUUGGCAAGAUGUUGGCACAGCGUAGUGAGAUUGACAGAAAGUGAAAACCGGCCACAGAA	100	25	21	15	18	40	39	0,857143	1,666667	-31,6	-26,5	0,025901	18,56	-31,6	-0,79
pmc-mir-315	UGAUGUGAAGUUCUUAGCAGAGGGGUCAAUGUAAGUUAAUUUUGAUUGUUGCUCUAGAAAUUAGAGCUCUUUGUAAAAGUUUACAGAG	88	25	29,54545	9,090909	36,36364	34,09091	65,90909	1,230769	2,75	-18,6	-18,6	0,01829	11,85	-21,1364	-0,62
pmc-mir-317	AAUAUGUGCACUGUGGGAAGGAUACCAUUGCUUGUUCACAGUGAAUUGGACAUGUGAACACAGCUG	94	25,53191	27,65957	14,89362	31,91489	40,42553	59,57447	1,153846	1,714286	-40,1	-40,1	0,224017	2,93	-42,6596	-1,05526

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUGGUAUCUUUUUUAAUGUGAACAUGAC															
pmc-mir-33	GUGUACUAUGGUAGUGCAUUGAGGUUGCAUUGCAUCAGCUGAGAAACAUGCAAUGCAUCUGCAGUGCAAAUAUCAUGGUAUGC	83	26,50602	27,71084	16,86747	28,91566	43,37349	56,62651	1,043478	1,571429	-38,7	-37,7	0,204831	5,09	-46,6265	-1,075
pmc-mir-336	UGACAGAGUACUUGAUUUGCUAUGUCACCCUUCCAUAUCUUUUGGUUUACUUUGUAUGAUGUGAAGUGUAUGUGAGCAAAGCUGAACAUCUGGAA	95	22,10526	25,26316	15,78947	36,84211	37,89474	62,10526	1,458333	1,4	-18,9	-14,5	0,043153	12,58	-19,8947	-0,525
pmc-mir-33b	CAUUGACUACUAUAAUCCAUCUGUCAAGCAGACACGUUUCAGUGCCUCUGCAGUGGAUGUAUGGUGGCAUCU	72	22,22222	23,61111	23,61111	30,55556	45,83333	54,16667	1,294118	0,941176	-23,6	-23,6	0,187831	6,78	-32,7778	-0,71515
pmc-mir-345	UCAGCGGCUCGUGGUGCAACAUUUGUUACCUGCACCUUUCUGCACUCGGCGGAGAUGAAGAACACGUGACGAGAACGUGCUGACCCCUAGUCCGCCUC	98	26,53061	20,40816	30,61224	22,44898	57,14286	42,85714	1,1	0,866667	-28,8	-28,8	0,301092	13,36	-29,3878	-0,51429
pmc-mir-3529	UGUGGCUGCAGCAAUCAGGGAAGAAUGUGAUUUAUUGUUAGUCAGCAUCAGAACAACAAAAUCACUAGUCUUCCAGAUCACGGCAGCCugg	91	21,97802	30,76923	20,87912	23,07692	42,85714	53,84615	0,75	1,052632	-30,8	-29,7	0,103281	5,67	-33,8462	-0,78974
pmc-mir-3532	CACGCUCGACGGACUUCACGAGAGCUGUUUUACACUCAUACACAAGCUCGGGUCUUGGAGGCUGCAGU	99	27,27273	19,19192	29,29293	24,24242	56,56566	43,43434	1,263158	0,931034	-40,6	-30,6	0,036618	11,77	-41,0101	-0,725

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUGACCUCAGCCUCGUGUCUGUCGCAGCAGU															
pmc-mir-355	UGACugauuuaucuguuuauuuguuuuagccUGAGUCGCCAGAGGAAAUGGAUGAUGAAUCUGAACAACCUAAAACAGCUGGCAAGAUAAUUCAGGCA	98	18,36735	27,55102	13,26531	13,26531	31,63265	40,81633	0,481481	1,384615	-22,1	-22,1	0,017425	13,25	-22,551	-0,7129
pmc-mir-36	UGAGACUCUUGUCAAUAAAGUACGUGUCGUUCUUGUCACCGGGUGGAAAUUCGUCUUUAAGACCUCCUUCAGUAUUUUGUGAGAAGGGUCAUA	93	23,65591	23,65591	18,27957	34,4086	41,93548	58,06452	1,454545	1,294118	-25,6	-18,64	0,051427	15,9	-27,5269	-0,65641
pmc-mir-3620	GGGUGGGCUGGGCUGGGCUGGGCUGAAAUAGGAAAAGAAUUAAUCCUCUUUCUGCUCCACAGUACAGCACAUCA	74	29,72973	25,67568	21,62162	22,97297	51,35135	48,64865	0,894737	1,375	-27,3	-24,8	0,167443	4,41	-36,8919	-0,71842
pmc-mir-3643	GGUCGAGAAUCACGGCAACUGGCUUGAGAUGUGAGUUUAUCUAAAUUCAGGAAAUGAGGAGCAGACAAGCUCGCUGGGUGAUUUCGUUG	89	31,46067	26,96629	15,73034	25,8427	47,19101	52,80899	0,958333	2	-21,4	-13,5	0,040157	24,56	-24,0449	-0,50952
pmc-mir-36b	UGAUGCGGUGAGUGUAAACCCUGGUCGGUGCACACCAGCAACAUCACCGGGUAUACAUUCAUCCGCACCC	70	24,28571	24,28571	31,42857	20	55,71429	44,28571	0,823529	0,772727	-30,6	-27,6	0,171958	5,87	-43,7143	-0,78462
pmc-mir-3724	AACUGCAUUGUAGUAAUUGGCAGGAAUCCCCCAGAAUCGUGGUAAACUUUGUCCUCGCCCUACAUUCUUACAGUGCACUC	80	18,75	25	27,5	28,75	46,25	53,75	1,15	0,681818	-20,3	-19,1	0,192073	5,28	-25,375	-0,54865

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-375-1	CGCCGCCAGACGACAUCACAGAACAUGUAGCUGCCCAAUGACCCGAGCCGCUCGUAGCAAGGCAUCUUCACAAGUUUUGUUCGUUCGGCUCGCGUUA	97	23,71134	22,68041	31,95876	21,64948	55,6701	44,3299	0,954545	0,741935	-30,9	-30,9	0,454586	4,38	-31,8557	-0,57222
pmc-mir-375-2	GGGACUUAAUUCAUCGCGAGCAGAAAAGAACGCAACAUCUUGUUCGUUCGGCUCGUUCUGAAAAAGUGAC	70	24,28571	30	21,42857	24,28571	45,71429	54,28571	0,809524	1,133333	-20,3	-20,3	0,298895	4,19	-29	-0,63438
pmc-mir-3782	AGGAGAGGAUCGGGGCCAGCCAAGGAGGCUUACAGAGGCACUUGGGUGGCGCGCCAUUCCACUUGA	66	37,87879	22,72727	24,24242	15,15152	62,12121	37,87879	0,666667	1,5625	-27,1	-26,3	0,354302	7,68	-41,0606	-0,66098
pmc-mir-3792	CUUAUCUUUCUGGGCCUCGUGUUAUGUAAGUCAUCAGACACUAACGACugaggaaagagagauguu	66	13,63636	16,66667	18,18182	24,24242	31,81818	40,90909	1,454545	0,75	-19,9	-17,6	0,1503	5,43	-30,1515	-0,94762
pmc-mir-3821	AAGCUUACUAUUGAGAGGGUCAguacuuuuuucauuugaguUCAUUUCUAGUGAGUGAGGGUACUUUGUAGCUGAUGUUCUCAAUGUUAGUAG	93	21,50538	19,35484	9,677419	29,03226	31,1828	48,3871	1,5	2,222222	-28,6	-28,6	0,072168	9,75	-30,7527	-0,98621
pmc-mir-4009c	CUUGCAUgcagcacagacacagauagUGCAGCCUCUCAUUAUUGCACUUUUACUGGUCGUGGCUCCAGGCUCC	73	15,06849	9,589041	23,28767	26,0274	38,35616	35,61644	2,714286	0,647059	-20,8	-20,7	0,16755	3,74	-28,4932	-0,74286
pmc-mir-4013b	auuugauuuugcuuACUUGCUUUAACAGCUUACUUACAGGUACAAAGUAGGCUUUUAACCUGUGUACCUG	99	17,17172	25,25253	19,19192	24,24242	36,36364	49,49495	0,96	0,894737	-30,2	-30,2	0,107048	16,44	-30,5051	-0,83889

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUAGAGACGGGUACACACACAAGUCACGU															
pmc-mir-4057	GGUCGGCAACUGACCUCCUGCAUCUCCAGGGUGUAGGAGAUGGGGGUGGCUUGGCGACUGUAGUAGGGCAGUUCCGCUG	79	39,24051	15,18987	22,78481	22,78481	62,02532	37,97468	1,5	1,722222	-33,5	-28,4	0,21803	20,3	-42,4051	-0,68367
pmc-mir-4070	CUCACAAGCAGGUUGUGAGCGACCACCacgcgucacgugacaccuagUGACAGCUGUGUUGACGUCACAGGCAGCUGUCUC	81	20,98765	16,04938	22,22222	16,04938	43,20988	32,09877	1	0,944444	-27,1	-20,9	0,082254	16,72	-33,4568	-0,77429
pmc-mir-4133	aaaccauguuguguuuuuguaugcAGGAUUAGCUGCAAGUGACAAGGAAAAUUACUGUCACAAGAUAAUCCUUCCAUCAGGAAACUCAAAUGGGAC	96	15,625	29,16667	14,58333	15,625	30,20833	44,79167	0,535714	1,071429	-26,4	-26,4	0,07692	7,8	-27,5	-0,91034
pmc-mir-4140	UCAAUUAUAUAAAAUCAACUGGUGCAAGAUGCUUUGCUGGAGAUUUCAUGGUAAUACUUCAGUAACCAGAUGAUUAUGACACAAUAAA	88	17,04545	37,5	14,77273	30,68182	31,81818	68,18182	0,818182	1,153846	-18,9	-18,9	0,163592	6,36	-21,4773	-0,675
pmc-mir-4177	UUACAGGGUGUUUACAUAAUUGcuguuuuaaacaguuuucaggUUAUAUACCAGGAUCAAACAGACUCUAGUCAUAAUACCCUGGGU	87	13,7931	24,13793	13,7931	24,13793	27,58621	48,27586	1	1	-20,5	-19,4	0,131475	8,3	-23,5632	-0,85417
pmc-mir-4185	CAUAAAGUAUCUAUGUUUAAUGCGCUGUUGUGUGAGUCUCCUUCUUUGUAUUCAUACUGUCUGUUACAAGCAAGCGGAUUAUUCAAAGAAUACUUUGCA	99	18,18182	26,26263	17,17172	38,38384	35,35354	64,64646	1,461538	1,058824	-24,4	-20,7	0,040505	9,56	-24,6465	-0,69714

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-4323	AUCUGCGGCUUCCUGCACUUAGAGGGGAGAGAUUUGCUUUUGACAGCCCCACAGCCUCAGGAGCAGAAAGCUGCAUAG	78	28,20513	24,35897	25,64103	21,79487	53,84615	46,15385	0,894737	1,1	-26,3	-25	0,219804	12,73	-33,7179	-0,62619
pmc-mir-4472	UGCUCUGCAGCAACAAUCGACUAUUCCCACCCUCCUUUUUUUACUGGGGUGGGGGGUGUUGUUGAUCAGAAAU	73	24,65753	19,17808	23,28767	32,87671	47,94521	52,05479	1,714286	1,058824	-25,5	-22,6	0,074609	17,51	-34,9315	-0,72857
pmc-mir-449d	CCUAUCAACCAUUGUCAGCAGUGAAACUAAUGGUAUUCAUUUGUCUUGAAGGCUGUGUGCUGUAAAGAUAAUGCUCACUGCUGCCCUCGUGGAUCUA	97	21,64948	24,74227	21,64948	31,95876	43,29897	56,70103	1,291667	1	-25,6	-22,8	0,038963	9,81	-26,3918	-0,60952
pmc-mir-4629	AACGAGGCUCACGUGUGUGAUGACGUGGCAGUGUCACGGCGAGGAUCCUGUCCCAAUGACUGGACGUUCAUCGCCGACAGCGAUGGCUCUCCCU	94	30,85106	19,14894	28,7234	21,2766	59,57447	40,42553	1,111111	1,074074	-30,5	-25,9	0,016318	28,06	-32,4468	-0,54464
pmc-mir-4654	GGCUGAUACGCCAGGUGAUGUGGGAUCUGGAGGCAGACUUCGGCUUCCGCAUCGGCCCGUGGAACCAGGCCUACAUGUACGACACCCUGCCGAUCAGUC	99	31,31313	19,19192	30,30303	19,19192	61,61616	38,38384	1	1,033333	-41,7	-40,2	0,172557	4,86	-42,1212	-0,68361
pmc-mir-466-1	AUACAGUCUGUGGCGUGUUUUGUAUGauguauaauauauacacacacacauauauauauauacacacauacacauacaaggCCUAGAGUCUGCGG	95	13,68421	6,315789	7,368421	14,73684	21,05263	21,05263	2,333333	1,857143	-24,1	-24,1	0,035265	12,2	-25,3684	-1,205

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-466-2	uauauauauacacacacacaaccauggAGUCUCCACAUGAGUGAGACAAGUGACCCUGAAAUGAGUGACUCAGCACCCAAGUUUUUGUguauaaauagc	99	14,14141	18,18182	14,14141	15,15152	28,28283	33,33333	0,833333	1	-19	-10,61	0,024059	19,5	-19,1919	-0,67857
pmc-mir-466-3	CAGGUGAUACAGAGCGAACUGUAAUAUUcauacuguguauauauauacacacacacauauacauauaaugaaCAUUAUGUGUGCACUGUAUUACUAG	97	12,37113	16,49485	8,247423	17,52577	20,61856	34,02062	1,0625	1,5	-29,3	-29,3	0,326455	3,38	-30,2062	-1,465
pmc-mir-466h	AGCUGAUCGGCCAGGAUUCUCGCGCGUGcgcguacgcacgcacacacacacacacgcacgcacgccugGCCAAACAACa	79	13,92405	11,39241	15,18987	7,594937	29,11392	18,98734	0,666667	0,916667	-29,5	-29,5	0,500936	3,9	-37,3418	-1,28261
pmc-mir-466i-1	gugugugugugugugugugugugugugugugaucauaaCAUAUGCCAUCUCCUGAUCACAGGCAUUCACACUAAACAACACCAGGA	86	6,976744	19,76744	18,60465	10,46512	25,5814	30,23256	0,529412	0,375	-30,8	-24,1	0,099618	12,29	-35,814	-1,4
pmc-mir-466i-2	ugugugugugugugugugugugucauucuAGUcuauccucugugugugugugugccaucaAGAAGCCUCUCAAUGAACCACCAGAACAGCACGGG	94	9,574468	14,89362	11,70213	4,255319	21,2766	19,14894	0,285714	0,818182	-20,1	-12,9	0,014359	21,92	-21,383	-1,005
pmc-mir-466m	aaauugugugcaugugcaugugaaugugugugcaugcuuacCUGCUUUAGAGAUAUCAGAGCAGAGAACAUCUACCAUGCAUGACCAUGCCAAACCU	97	10,30928	19,58763	15,46392	12,37113	25,7732	31,95876	0,631579	0,666667	-27,1	-26,2	0,171386	21,7	-27,9381	-1,084
pmc-mir-466n	uuuuguauUACUUCAUGCUCAcgaaugugugcacacacacacacaugggugCA	97	12,37113	11,34021	10,30928	13,40206	22,68041	24,74227	1,181818	1,2	-28,3	-28,2	0,049412	16,07	-29,1753	-1,28636

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UAGGGGACAGGUGUGUGCGUACAUGUACUCAuguauauacauag															
pmc-mir-466q	CCGCGAGCUGUaacgugcacacagacacacagacuguugCGUGCUGuaacgugugcacacacacacauacacugguGCGUGCUGUAACGUACUGCCGCGU	100	15	4	13	10	28	14	2,5	1,153846	-33,1	-14,84	0,013848	33,92	-33,1	-1,18214
pmc-mir-467d	GAGGGUUGUAAGUGCGCGCAUGUAUUUAUGUUGCAUUCGUAUGUAGUUAUGCAUGUAUAAAUACAUGCACUCGAUAAAAACCAGA	85	23,52941	30,58824	14,11765	31,76471	37,64706	62,35294	1,038462	1,666667	-25,9	-23,2	0,094769	13,72	-30,4706	-0,80938
pmc-mir-467f	GUAGGCCAACGUCUCCCUCCCGCUGAaaaauuuauauacacacacacaccuuucaagUGGGAGGAGAUGGACAAUGGCCCAU	82	19,5122	13,41463	18,29268	10,97561	37,80488	24,39024	0,818182	1,066667	-30	-30	0,050335	8,93	-36,5854	-0,96774
pmc-mir-4715	CGCCACGCUGAUCACGUGCCAACGUGCCACCUUAACUGCACGGACCGCGGAAGAUGGUUUGGCCGUGGAAAGUGAGCGGUGAGU	84	33,33333	21,42857	27,38095	17,85714	60,71429	39,28571	0,833333	1,217391	-31,6	-31,6	0,33988	8,09	-37,619	-0,61961
pmc-mir-4750	CCUUCAAUGUCUCACGUGCACGAUGGGGGGAAGGGGCUGUGAACACAUGGACUGACACCUGACCCACCCCCUCCCUACUCUGACCGUGACAGUGACAA	98	25,5102	23,46939	32,65306	18,36735	58,16327	41,83673	0,782609	0,78125	-29,8	-19,7	0,066962	29,71	-30,4082	-0,52281
pmc-mir-4757	aaaaaaaaccaaaacucaAGCGUCUCAGUAAAGGGUCUACAUUUUACACCUGUCAUUUCCAUGACGUCACAGAGGCGAGGUUUGGCUUUUGGA	93	20,43011	18,27957	17,2043	24,73118	37,63441	43,01075	1,352941	1,1875	-22,4	-21,8	0,139005	15,42	-24,086	-0,64

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-4769	AUUGCUGGCUUCACUCUUCUGCCAUCCUCCCUCUCGUCUGUCGACAUGGCUGACCCCAAGAUGGUCAGAAGAGUGAACAAGCGAG	85	23,52941	21,17647	30,58824	24,70588	54,11765	45,88235	1,166667	0,769231	-34,6	-34,5	0,204288	8,13	-40,7059	-0,75217
pmc-mir-487a	GGGAUUGUGCACUAGUGGCUAUCCCUGCUGUCAUCCCACCGAUAUCACGGCCAUAGCACGGUCACCACUAUUACAAUAGU	80	21,25	23,75	30	25	51,25	48,75	1,052632	0,708333	-22,7	-22,7	0,227369	6,98	-28,375	-0,55366
pmc-mir-49	aacaaaagaaaccaaaaaggGCAUCUUCCCAUGAUGCAAAUAGACUAAGGGAAGCACCAUGUGAAGCACUUUUCUGGUUUCUUUUCCa	88	14,77273	19,31818	18,18182	23,86364	32,95455	43,18182	1,235294	0,8125	-26	-26	0,306727	8,11	-29,5455	-0,89655
pmc-mir-4938	UUUCUgcauauuauuauaaucaucaugcaucaucauuauugaCCAUGCAGGUGCAUGAUCGAUCCUUAAUUGUAUGAGGAU	81	12,34568	12,34568	9,876543	19,75309	22,22222	32,09877	1,6	1,25	-23,1	-20,9	0,193135	5,48	-28,5185	-1,28333
pmc-mir-4949	AUUAGCAUUCAUGAUACGGAUUUUAAGGAaguggugugcgugugugugugugcccacuUAUGACAAAGUCCACAUCAUGUAACGCUGUU	89	12,35955	22,47191	11,23596	21,34831	23,59551	43,82022	0,95	1,1	-28	-28	0,194482	3,14	-31,4607	-1,33333
pmc-mir-4968-1	gaCGUUGCUGCCGAGCUCGGCUGUGCGUCGUGCUGgacggcagcagcagcaacagcagcagcagcagcaacauu	74	17,56757	1,351351	13,51351	12,16216	31,08108	13,51351	9	1,3	-35,3	-30,4	0,160289	15,99	-47,7027	-1,53478
pmc-mir-4968-2	CGACAGCGCAcuggcagcaacagcagcagcagcagcagcacucgGCGGAUGUGCAGCUUCCCUCGGUGGUUGCUGGCAGGGCCACUGGUU	90	23,33333	7,777778	17,77778	13,33333	41,11111	21,11111	1,714286	1,3125	-33,6	-22,2	0,144928	16,86	-37,3333	-0,90811

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-4968-3	ACACGACAGCCCGCAUCUGAUGCACUGCUUCCUCAGUUUCauccagcaacaacagcagcagcaacagcagcagcagcaaugccGAGAUGCCGGGA	95	13,68421	11,57895	17,89474	11,57895	31,57895	23,15789	1	0,764706	-24,8	-24,1	0,103418	14,12	-26,1053	-0,82667
pmc-mir-4968-4	CUACACCCUGGGCUGCGGCGGUUGUGUCCAGcaucacagcagcaacagcagcagcagcagcagcagccucaaCCAGCGCAGCAGCUGCAGCGGUGCGA	98	21,42857	8,163265	19,38776	9,183673	40,81633	17,34694	1,125	1,105263	-38,8	-38,8	0,480796	14,61	-39,5918	-0,97
pmc-mir-4999	GACGCAUUGACCAACAAUAUGAUACACUGAUGGCUUUCCUAUUGCUGUAUUGUCAGGUUUGUAGAUAUUGUAAUAGGUUAACUGCCAU	88	20,45455	27,27273	17,04545	35,22727	37,5	62,5	1,291667	1,2	-20,3	-20,2	0,098	21,47	-23,0682	-0,61515
pmc-mir-501	AAUGCACCCGGGCAAGUCAGAGGACACAUUAAGAGCCCCAUUAGAGUAAAACGGGAAUGGAGUUCCGUUAACUGACGUCAUUUGUUUCCAGGGGUGCCAG	100	28	29	22	21	50	50	0,724138	1,272727	-28,4	-28,4	0,207312	9,5	-28,4	-0,568
pmc-mir-5101-1	uuuguuuguuuguuuguuuguuuguuuugcugucugACGUUUGCGGAAAACCACAGCUCAGUCUGUCAGCAGACAACCAgaugaaaugaaagauaacaug	100	9	14	13	7	22	21	0,5	0,692308	-26,2	-19,3	0,014197	18,79	-26,2	-1,19091
pmc-mir-5101-2	UCGACCGAAGGAUAGUGACGGuccuauuuguuuguuuugcugugugcUUAUUUGUGUUAGGUUUGGUCUUAGGACUGUUUUaccuuugguuuu	93	18,27957	10,75269	6,451613	23,65591	24,73118	34,4086	2,2	2,833333	-26,8	-26,2	0,065192	10,31	-28,8172	-1,16522
pmc-mir-519f	GUGCGCAUAUUUCUUGCUGAAAAUUGUCAUGUUUAAUUUGGAGGAUAAAGUGCAUCCUUUUAUGGC	97	24,74227	27,83505	13,40206	34,02062	38,14433	61,85567	1,222222	1,846154	-23,4	-22,1	0,126706	9,47	-24,1237	-0,63243

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GAAACAUCAAUUCGGAAUGUAAUGUGCGGGA															
pmc-mir-5317a	GGGCCAGGGCCACAUCCCUAGCUCUGGUCCGUUUCUUGUCUCUGUGUACCUCUCGUGCUGCCAGACCAAGUGGUGAUCGGUGACCGCUGGAAC	93	29,03226	13,97849	31,1828	25,80645	60,21505	39,78495	1,846154	0,931034	-33,8	-33,8	0,197453	11,33	-36,3441	-0,60357
pmc-mir-539	AUGGUGAUGACUCAUCAUACAAGGACAAUUUGUCACCAGCGAAGAUGUUAUUUAUACCAUGACAGAAUGUGUCUUGUAUUUGUUCACACGAA	92	19,56522	31,52174	17,3913	31,52174	36,95652	63,04348	1	1,125	-20,4	-20,2	0,064375	14,18	-22,1739	-0,6
pmc-mir-5391	uuccugcaagGUACCUUAUGAGGUAACUUGUUUGGACAGUUAUGAAGAAGUGGAAUCUAAUGUCCAACAGUUUAGAAUCAAACAGGCUAUCAUUCAGACC	100	19	30	15	26	34	56	0,866667	1,266667	-18,8	-12,3	0,005609	35,95	-18,8	-0,55294
pmc-mir-5396b	GAAGAGGAAUCCCCCACACAACCAGCAAGAUUUGCUGCUGCGGAAGGAACAUCCCGGCAUUCGUUGACUGCCUCUGGUGGGUGGAGUCCUCGUG	94	29,78723	22,34043	27,65957	20,21277	57,44681	42,55319	0,904762	1,076923	-34,1	-26,1	0,0206	19,14	-36,2766	-0,63148
pmc-mir-5549	CcccaaaauaucaaaauagaaUAAUAAGAGGAGGGUGGGACUUCUUUAACAUUACACAGAUAUCCUAUUAUUCUUCAUGUUGGUUUUUUGAAU	93	15,05376	21,50538	10,75269	31,1828	25,80645	52,68817	1,45	1,4	-19,5	-19,3	0,091982	11,56	-20,9677	-0,8125
pmc-mir-5552	gucuguagucuauagucuguagucuguagucuaUAGUCUGUGGUCUGUAGU	85	15,29412	12,94118	9,411765	23,52941	24,70588	36,47059	1,818182	1,625	-23	-20,4	0,065706	32,41	-27,0588	-1,09524

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	UUGUAGUCUAAAGUCUGCAGCAAUGAGUUCAUCU															
pmc-mir-5595	UAGUGUCACGUGCUUUAAAAAAGGUGCAGGCUCUCUUUUUUCUCGCAGUCCCGCACUGACCUGAGAGAGUCACCAUCUUAUGUCAAAGACAUGGACAAGA	100	22	27	24	27	46	54	1	0,916667	-30,6	-30,6	0,243502	5,31	-30,6	-0,66522
pmc-mir-5600	UGAGGUCUUUGACAGUAAGUACAGAGUUUGCUCAGACAAGUAAGGAAUCAGuggaauguuuuguuucaguaGUUGUCUAUCUUCUGUACCAAUGACCAUC	100	18	23	15	24	33	47	1,043478	1,2	-24,6	-24,6	0,137757	6,7	-24,6	-0,74545
pmc-mir-5612	ACAGGUGUCUAUAUGCUACAUCACCUGAAUAAUAGCUAUUGUGUCUAUACAUCACCAGUUGACAUGUUAGGUAUGGUAUUUAUACACCACC	91	16,48352	29,67033	20,87912	32,96703	37,36264	62,63736	1,111111	0,789474	-20,8	-15,6	0,041563	12,19	-22,8571	-0,61176
pmc-mir-574-1	CAGCUCACAGAAACCAUCAGCCACAGGCUUCAAGAAAAAUACUCUAAUGACAGCUCGCGCCAUUGAGAGUCGUggaggugagugugugugugagugc	99	14,14141	25,25253	21,21212	13,13131	35,35354	38,38384	0,52	0,666667	-26	-18,3	0,019862	27,93	-26,2626	-0,74286
pmc-mir-574-2	AUUUUCCGCCAUUACUCAUCUUCCACCCAGCAUCACGUUAGUUAAUAGUCAGGcggagugugagugugugugugagugugugugaaggGGGAAUAC	97	10,30928	16,49485	17,52577	18,5567	27,83505	35,05155	1,125	0,588235	-27,6	-27,5	0,03265	10,86	-28,4536	-1,02222
pmc-mir-5918b	UUCUCAUGGUUGUGUGUUAUUAUGUUCGCUCUACAUCGUGUGGUCAGAGAUggugagagagggaagagaga	100	15	10	10	24	25	34	2,4	1,5	-22,4	-14	0,095221	25,67	-22,4	-0,896

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	gugagagaaagaaugacaagaCAUGAUGC															
pmc-mir-5965	CAACCUUGGGUGAUGACAAUUCUGACAUUUUGCAAGGUUUCAUCCUUGUCUGUUUCAUUCUUCAAACUGCGAAGUCGCGCAUCACCAAUGGGUA	94	20,21277	23,40426	23,40426	32,97872	43,61702	56,38298	1,409091	0,863636	-24,6	-23,9	0,068188	8,99	-26,1702	-0,6
pmc-mir-6056	UCUCUGUCGACUUCAUUUCCUCCUCGGCCCAGUCUUCCUCUCUCUGGUGAUGAUAGGGGUGAGGUGAGGGacgaggagggaggaggaagacgAGAGAGC	99	23,23232	11,11111	20,20202	23,23232	43,43434	34,34343	2,090909	1,15	-50,2	-46,8	0,12691	12,98	-50,7071	-1,16744
pmc-mir-6076	AAGGCUGACACCCUUCCCUCAGGUCGUCCUCCUCUAAGAAGCAAACAGAAGCAUGACAGAGGAGAGGAGAAAACCCAAAGGGAAGGCUCUCUGCAUG	97	25,7732	32,98969	26,80412	14,43299	52,57732	47,42268	0,4375	0,961538	-33,4	-33,4	0,461099	5,24	-34,433	-0,6549
pmc-mir-6098	GUGCUCGUCAUAGUGAUUGUGUGUUUCUAGAAGACAAGACCGUGUUCUGUUCAGAACACAAACAACAUGCUGAUUGUCGAGUAA	84	23,80952	28,57143	17,85714	29,7619	41,66667	58,33333	1,041667	1,333333	-22	-19,4	0,02079	16,09	-26,1905	-0,62857
pmc-mir-623	UAACAUACCUGUUCCGCCAUGGUGAACGAGUGAGGAAAAGACCGGAACGGGGUUCACACGUUUCCCUUGCAGGGGCUGAGGUAUGAGA	88	31,81818	26,13636	21,59091	20,45455	53,40909	46,59091	0,782609	1,473684	-29,8	-29,8	0,153312	7,64	-33,8636	-0,63404
pmc-mir-626	UGACACUGUGUCACACCAGCAAGcugucugaaaauguuugcaugaaaacau	96	14,58333	10,41667	10,41667	6,25	25	16,66667	0,6	1,4	-22,1	-10,23	0,039265	24,65	-23,0208	-0,92083

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	gaagacauuaaaacauuuauaaaaagugCGGGUGGGGAAGUGCAC															
pmc-mir-6302	GGAUGAGUCACUCgagauauuugauauuuuucaucaaUGAAACCAUGUUCCUGUGAUGAAGAUAGAUAAUGCACCAAACUCAUCGUCAGCCACUCACGA	99	15,15152	24,24242	19,19192	17,17172	34,34343	41,41414	0,708333	0,789474	-18,9	-17,1	0,135467	11,6	-19,0909	-0,55588
pmc-mir-6416	AGCAAAUUGCUUGAUCAUAGUGAGCGUAUCAUGGCAGCACAUCCUCAUGGGUUUGCGUCUCCGUAUCAUCUGCUAGCUACUGAGAACAAUUUUUU	95	21,05263	24,21053	22,10526	32,63158	43,15789	56,84211	1,347826	0,952381	-21,2	-20,6	0,052561	13,18	-22,3158	-0,51707
pmc-mir-6505	CUUAGCUCCAUCUGACUUCUACCUGUUCCCUAAACUCAAAUCCCACCUAGUUGGUCAccauuuuggaaacaaugauGAAGUCAUAUGAGCUGUG	94	12,76596	19,14894	23,40426	24,46809	36,17021	43,61702	1,277778	0,545455	-21,1	-19,17	0,012139	12,86	-22,4468	-0,62059
pmc-mir-651	CAUGAUAUUUUAAGUGAUAAAGGCUUUUACCUGCAAAAGAUAUGGAGUUUUAUGCUCGAUAUAAAAUUUUAGGAUAAGUUUGAACACGUAAAGCAUCAAU	100	18	37	11	34	29	71	0,918919	1,636364	-21,6	-21,5	0,14346	4,59	-21,6	-0,74483
pmc-mir-6516	GAGUCUUAUUCUAACUGGUUUCAUGAAUGCAGUAACAGGUGUCUCCACCUCUGCAUUGCUCAAACCACAUUACAUCAGAAAG	82	17,07317	29,26829	24,39024	29,26829	41,46341	58,53659	1	0,7	-19,7	-19,6	0,151242	5,16	-24,0244	-0,57941
pmc-mir-6528	GGUGGGuaggagagagggaagagggaagAAAGCAAGAUGAAAGUGUCGCCUCUGGGAGAGCUUUCGGUGGU	97	23,71134	14,43299	18,5567	20,61856	42,26804	35,05155	1,428571	1,277778	-32,8	-27	0,029155	15,05	-33,8144	-0,8

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	AUUCUCCUCCCUGUCACGUAGCCUCU															
pmc-mir-6548	CCGUUCACACAGAGAUUUGAGAGGGGGAGGGGCAGAAAAGAGAGGUGCCCCGCUGUCUGUUCUCUGGUGAAAAG	74	36,48649	25,67568	18,91892	18,91892	55,40541	44,59459	0,736842	1,928571	-31	-31	0,103929	5,12	-41,8919	-0,7561
pmc-mir-6560	CGCUGUGUUGCUCCAUACUGCUGAAGUAGUGUCUGUACUUCUGUUUCAGUCAUAAUGAGCAGCUCAUCA	69	21,73913	20,28986	23,18841	34,78261	44,92754	55,07246	1,714286	0,9375	-19,8	-18,8	0,070623	8,29	-28,6957	-0,63871
pmc-mir-6562	AGGCAUAUGAGGGGAAAGGAAUGUGCAGGUAAUGGUACUUGUAGUUUCCUAGUUUGUAAACAUUAACACUGGACACUUGUCUCCCUCGGUUCAuaugcaa	100	24	25	16	28	40	53	1,12	1,5	-31,7	-31,3	0,048803	12	-31,7	-0,7925
pmc-mir-6568	CUGAGCCUGACGAUGCCGGGCGCCAUAAUGGCCACGAGCUAACUGUCGCACGGCGGCCGCCAUCUUGCGGAGCUGUG	77	33,76623	16,88312	32,46753	16,88312	66,23377	33,76623	1	1,04	-27,4	-27	0,220859	16,73	-35,5844	-0,53725
pmc-mir-669f	acacacauauacauacacacacacguguuguUUCACAAGUAGUACAACAGGCGUGUGAGAUGUUUAUGUGAGG	73	17,80822	16,43836	6,849315	16,43836	24,65753	32,87671	1	2,6	-25,6	-20,3	0,138486	10,86	-35,0685	-1,42222
pmc-mir-669j	CUUUGACAUGACAGUUUUUGAUUUGUGGAUCguauaaaagacaaauacaucCACAGGCAUGCAUAUACUCACAUGUGCACAC	82	14,63415	19,5122	17,07317	24,39024	31,70732	43,90244	1,25	0,857143	-18,5	-17,6	0,121983	11,36	-22,561	-0,71154

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-67	GAGAUGUCGUGAGUUGUCGGUCCUUGUUCAGCCCGGUUGUGAUGGAGUUGGUCGCAUCACAACCUGCAUGAAUGAGGACUGACGUCACGAAAUAAC	96	30,20833	22,91667	20,83333	26,04167	51,04167	48,95833	1,136364	1,45	-50,4	-50,4	0,287211	2,51	-52,5	-1,02857
pmc-mir-6701	CCUAUCAUUAUGGAGUUGCUUUGUUGUUAUCUGACAGCCCAGGAAAAAUAUUCUGAUUAUUUUACAGACAGCAACUAUGAAUCCAAAACUGGAUGAU	97	17,52577	31,95876	17,52577	32,98969	35,05155	64,94845	1,032258	1	-19,2	-18,9	0,066024	11,85	-19,7938	-0,56471
pmc-mir-6876	CUUUCACUUCAGCUGUCUGUGUUUUCCCAGAUGCAGAAAUAAGUAUAUUGUGGUUUAUUGUGUUCUAGUCUGUAAACAAUGCAGUGaagcgaugu	95	18,94737	21,05263	14,73684	35,78947	33,68421	56,84211	1,7	1,285714	-22,5	-22	0,114242	8,28	-23,6842	-0,70312
pmc-mir-6891	CACCCUcaucuuccccuccccucuccaucccacccccaccucGGCGAGCGGACUGCGAAACGGAGAGGAAAUGGGAAG	78	21,79487	16,66667	11,53846	3,846154	33,33333	20,51282	0,230769	1,888889	-24,9	-24,9	0,700959	2,36	-31,9231	-0,95769
pmc-mir-6971	CGCCCGUUUCCCUAGAGAAGAAGCGUGUCAUCUGCGACAGCCUCUGCUUCUUCAGCUAGUGUCACGGAAG	70	25,71429	20	30	24,28571	55,71429	44,28571	1,214286	0,857143	-24,5	-23,2	0,750631	1,04	-35	-0,62821
pmc-mir-6974	ACAACGACGACGGUGGACAGGGGGUGGACCUCGAUGCCAGCAUGGUGUCCACCAGCUACAUUGCGCUCUUCUCCACUCUCUUCUGUGUCGUGGG	94	29,78723	17,02128	29,78723	23,40426	59,57447	40,42553	1,375	1	-35,8	-34,3	0,187738	13,56	-38,0851	-0,63929

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-7000	CAAACAGUGCCCACCCACCUGCCUGUCACAcugugauuuauuuacauuguaaCUGGAUGCUCAUGGCUGGUGGCCCAGCACUGUAUA	87	17,24138	16,09195	26,43678	14,94253	43,67816	31,03448	0,928571	0,652174	-28,9	-28,6	0,111346	9,33	-33,2184	-0,76053
pmc-mir-7046	GGUGUAGGGUGAGGCUGGUGGGUUAGUUGGGUGGCUGGUCGGGUGAAGAAAGGGAAGUAAGUCAAAUUUAAACGUGACACCACCACCCACCUACCCU	97	35,05155	24,74227	18,5567	21,64948	53,60825	46,39175	0,875	1,888889	-32,6	-28	0,059458	25,01	-33,6082	-0,62692
pmc-mir-71	CACCCGUACCCUAGUCUGGUGAAAGACACAGGUAGAAUGUAAUUCUACUGUUCACUUUUAAACGAGGGAUGCGGACU	77	23,37662	28,57143	22,07792	25,97403	45,45455	54,54545	0,909091	1,058824	-24,6	-24,6	0,07754	10,03	-31,9481	-0,70286
pmc-mir-7-1	UACUGUUCUCCAGAUUGACUUCCUCUAAGUACUGAUAGUUGUUGUUACCAACAAAUCGUAGCCUCAGAGGUAAGGCAGGUCAUUGaacaacacau	95	18,94737	23,15789	18,94737	28,42105	37,89474	51,57895	1,227273	1	-23,3	-21,3	0,085299	10,39	-24,5263	-0,64722
pmc-mir-7151	UUGUUGACACUGAGAAAAGUGACAGGAUCCAUCUCUGCCUGCUCACCUUCACGUUUGUGGUGAGCAgcugucauuuuucucuuugcAAGAU	91	18,68132	19,78022	17,58242	21,97802	36,26374	41,75824	1,111111	1,0625	-37,2	-36,5	0,178864	4,62	-40,8791	-1,12727
pmc-mir-7-2	AAUGCAAAUACUAGGGACAggguucuuuuuaaaaaaaaaaugcagacggCUGAAUCGCUGACUGGUUACAAAAGCCAACAAGUCCCAGUCUUGGCGGA	98	16,32653	23,46939	16,32653	13,26531	32,65306	36,73469	0,565217	1	-20,7	-16,47	0,006838	17,31	-21,1224	-0,64688

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-7207	AGUUGAGGAAAUGAAAGuugcuuucaucuuuuaaaauacaggGUGUUGACAGUCAUUUCACCAGAUGAAUAAACACGUGGCUCAAUUCCUCAUUC	95	15,78947	24,21053	13,68421	20	29,47368	44,21053	0,826087	1,153846	-20,6	-19,9	0,070254	20,54	-21,6842	-0,73571
pmc-mir-722	UAUAAGGUAGAAUUUUGCAGAAACGUUUACAGUUUACUACUCUUUGGAGUUGGCUGGUGUAUAUGUUACAAGUAUAUUUCCUACCUUUUG	90	20	25,55556	13,33333	41,11111	33,33333	66,66667	1,608696	1,5	-20,7	-19,6	0,061494	7,79	-23	-0,69
pmc-mir-7241	CACCUGUUGUGAAAAUGCUUUGAACUUAGGUGUAUGCAUGUUGGAUCCUGACUCCUACUUGCAUGAGUUAAUGAUUUGUCAGGUCAACAGAGG	93	24,73118	24,73118	17,2043	33,33333	41,93548	58,06452	1,347826	1,4375	-20,3	-17,7	0,029323	17,26	-21,828	-0,52051
pmc-mir-7314	GUCACAGGGACCCUGGUUGGCCUGCACAAGCCCCAGUAAUACUGGUGUCAAGGAUAGACGAAACUCCUUGAAGAUUGUCAGAAACAGUGUCUCUGUGCa	99	26,26263	26,26263	24,24242	22,22222	50,50505	48,48485	0,846154	1,083333	-32,6	-28,7	0,010541	12,05	-32,9293	-0,652
pmc-mir-7323	UUUUUCCGAAAUAGCUUUGUGCUUUGACAAGUGCACUCUGCAAGUUACAGAAUUGUGUGAUCAACGUCAAUUCUAAUUUCCGGAAUAU	88	18,18182	27,27273	18,18182	36,36364	36,36364	63,63636	1,333333	1	-20	-20	0,049302	14,06	-22,7273	-0,625
pmc-mir-7386e	UGUAAAGUAGUAAACACUUUACCCGUUUGACUGUUUGCCCUUGUUCUUGAACAGUGUGGGUGAACCUCUUGACUGGUGUAAAGUUGUAUAacuuuuua	98	21,42857	21,42857	15,30612	33,67347	36,73469	55,10204	1,571429	1,4	-24,7	-24,1	0,025944	13,11	-25,2041	-0,68611

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-7388c	UGGUCCUUCAAGUGUGGCGUCAGUUACCUGCCCCUGUUCUACCUGCACAAGGUGCACAGACGUGACAUCGUUGAGGAAGU	80	27,5	20	26,25	26,25	53,75	46,25	1,3125	1,047619	-24,4	-24,4	0,084225	8,96	-30,5	-0,56744
pmc-mir-7398a	aaagagaacuUCCGUAACCUGUAACAUUCACAUGAUGAUUUGUUAAUUUGGAGCUUAUGAGCUCACCGUAUGUGUAGAGGUGAGAAAGUUGUCGCU	96	21,875	22,91667	14,58333	30,20833	36,45833	53,125	1,318182	1,5	-21,1	-15,38	0,110809	24,91	-21,9792	-0,60286
pmc-mir-743b	AGUAGACACUUCUGAACGAGCUUCUUGUACUAACCUUGCUGUAAACCCCAGGACGAAGCUGUUCAGACUGGUGUCUGGU	79	24,05063	24,05063	24,05063	27,8481	48,10127	51,89873	1,157895	1	-26,2	-25,6	0,030673	10,47	-33,1646	-0,68947
pmc-mir-7448	ACAACUUGAGUCUCGAAGAAGCGAACUGUUGUAGAACAGUUCCGAUCAAUCGAUAAUCGAGUAUU	65	21,53846	33,84615	18,46154	26,15385	40	60	0,772727	1,166667	-20,4	-19,3	0,340793	2,64	-31,3846	-0,78462
pmc-mir-745a	AGCUUGCUGGCUGGAGGCGGCUUUUCAACAGGCUGCCUUGCGUUCUAAAUACAAGCUGCCUGAUGAAGAGCUGUCCUGUGGCGAGCAAUGU	91	30,76923	19,78022	23,07692	26,37363	53,84615	46,15385	1,333333	1,333333	-46,9	-46,9	0,097683	4,24	-51,5385	-0,95714
pmc-mir-745b	UUCCCUCCGCUCUUCCUUUGGACAGCUAUCGUGCCGACAAGGAGAGCUGCCAAAUGAAGGGCUGUGGUGG	70	30	18,57143	27,14286	24,28571	57,14286	42,85714	1,307692	1,105263	-29	-25,7	0,096995	8,37	-41,4286	-0,725
pmc-mir-7472	CCUGUGCCUGCUGAUUGUGCAUUCCGGGGAGAACAUUUCAUAUGAGAUGUGCUGGAGAUGAAAACU	97	24,74227	24,74227	24,74227	25,7732	49,48454	50,51546	1,041667	1	-30,5	-27,9	0,050851	8,11	-31,4433	-0,63542

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUUCCUCCCCAGAACAUAAAUCUGGCACCGC															
pmc-mir-750	GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUGGCAGGAAUACUGUCUUCCAUGCCAGAUCUAACUCUUCCAGCUCAUAGUGGCAUGGCAUG	97	26,80412	20,61856	23,71134	28,86598	50,51546	49,48454	1,4	1,130435	-52,7	-51,6	0,157091	3,48	-54,3299	-1,07551
pmc-mir-751	CUUGGCUCUGUGUGAUUGGUUUCUGUUCUCCAAGauuaaaacauguuugaaugGCCACCACUUUGGCACAGUAAACAUCAUCCAUAGAGCCCUU	94	15,95745	15,95745	22,34043	25,53191	38,29787	41,48936	1,6	0,714286	-27,8	-22,8	0,051558	21,54	-29,5745	-0,77222
pmc-mir-7562	cacacaccccgacacacacacacacauucauggaCACGCAUGCCUGCACUAAAAGUUUGGUUGGGAAUUGUGAGAGAUAUGUCGCGCAUGUUUA	95	17,89474	15,78947	10,52632	18,94737	28,42105	34,73684	1,2	1,7	-20,2	-20,2	0,122747	10,51	-21,2632	-0,74815
pmc-mir-7575	AGCACAAAAAGUUAGCAUGGUCAUGAUCAUGACACAAUGGGACAAUUAUGCACCAUAUGGCUGAUUAUUUGUUUA	75	20	34,66667	16	29,33333	36	64	0,846154	1,25	-19,1	-16,7	0,188578	11,02	-25,4667	-0,70741
pmc-mir-7643	aguuuguuuuucagugaACAUUGAAUACUGUCGACAUCUGUUUCUCAGUAAUUCUUACUUAGUAUCACAGAUUCAACUGAACACUGAUACAGAACGA	97	11,34021	27,83505	17,52577	25,7732	28,86598	53,60825	0,925926	0,647059	-20,4	-15,3	0,025588	14,82	-21,0309	-0,72857
pmc-mir-785a	AAUCUCGGACUUAUGAAAAUAAGUGAAUACUCUGUUAUGUCGUUAUUGUCCAgguguuuugaaaaagaa	99	12,12121	17,17172	9,090909	20,20202	21,21212	37,37374	1,176471	1,333333	-19,4	-19,4	0,222469	10,6	-19,596	-0,92381

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	uuucauuuauuguucuuuuguccUAGCGG															
pmc-mir-7880b	uggcagacagacaacugucaaccuauacacguccaugguuggAUGGUUGGUUAUUUUGGUUUAUAGUUGGUUGGAUCAGUUUGGUUUGGCUGGUA	95	18,94737	7,368421	2,105263	27,36842	21,05263	34,73684	3,714286	9	-27,3	-24,5	0,144234	12,84	-28,7368	-1,365
pmc-mir-7880q	UCAAUGACAUUUACAUACUCUUGGAAGAGGGUCAUGCCUACCAUGCAGUUUGCGACCAaaaucacucucucuuucaaguACUAUUGguuacaugcauaca	100	13	18	15	19	28	37	1,055556	0,866667	-18,6	-17	0,048677	12,83	-18,6	-0,66429
pmc-mir-79	GGUUGGUAUUAUAAcuguauuauauuauauuagUCAGUUGCUGUAGCCACUGUAACUCUGUAGCUCUUUUAUAAUGUAAAGCUAAAUUACCAGUG	95	15,78947	22,10526	12,63158	29,47368	28,42105	51,57895	1,333333	1,25	-18,8	-18,8	0,097081	11,62	-19,7895	-0,6963
pmc-mir-8	UGCGCGGACUCUGGGUUCAUCUUACCUAACAGCAUUAGAUGUGUGUCAGCAUUUUCUAAUACUGUCAGGUAAAGAUGUCCACAGAGUCUGCUGG	94	24,46809	23,40426	21,2766	30,85106	45,74468	54,25532	1,318182	1,15	-46	-46	0,192059	3,37	-48,9362	-1,06977
pmc-mir-8250d	CAGGCUGCAAACGUUCCAAUCCACACAAGUCCAGUCAUAGAACAGAUCUUAUAGACAGAAGUCCUUUAUUUGUCUGGGUGGAGGGUUUGGCUGCUUA	97	23,71134	26,80412	21,64948	27,83505	45,36082	54,63918	1,038462	1,095238	-26,2	-23,38	0,012202	14,09	-27,0103	-0,59545
pmc-mir-8265	CAUGAAGCUCAGCGAACAGCACCUAGCAUUAUCUACAUGCCAUCAGUUAAUGAACUGUGGGCUGUAGU	96	18,75	23,95833	28,125	29,16667	46,875	53,125	1,217391	0,666667	-25,5	-25,5	0,101749	8,96	-26,5625	-0,56667

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUCUGAUACUCUUCGCUCCACCUUCCUU															
pmc-mir-8293	AUCUUAGAUGAAAGAUAAAAGUGUCUCGUGAGUGCUUGUUGCAGUUUCGCAUCUGUGGUGCAGAUAAGUAAGCAgcaucuguuuauuuuccucCUAAUAC	100	20	24	12	25	32	49	1,041667	1,666667	-21,5	-13,1	0,030404	22,6	-21,5	-0,67188
pmc-mir-8311	UGUUUACUAGAAGAGAGAACUCCUACAAGGGAAAAACCACCAUCAGUGAUUGUGAGAUUCCCCCACCUAGCCACUAGAAGUUCUCUUUCGUAGUAAAGU	99	19,19192	32,32323	23,23232	25,25253	42,42424	57,57576	0,78125	0,826087	-28,4	-28,4	0,262782	6,39	-28,6869	-0,67619
pmc-mir-8335	UCCGACAUAACAACCACGACUACAGUCAACAGAUGGCCACUgaacucucauucucucuccaCUGcuccauguuguuguuguuguuguuuugucagu	96	7,291667	15,625	15,625	7,291667	22,91667	22,91667	0,466667	0,466667	-24,4	-20,9	0,125649	19,36	-25,4167	-1,10909
pmc-mir-8398	UGCUUGCACUGAUCUagaagacagaagugaaaaCACUUUUCUAAAUGAAGUGAAUCUGAACAAAUGAUcacaguucuuucuuuuuagcauGCAAAGU	97	10,30928	19,58763	10,30928	18,5567	20,61856	38,14433	0,947368	1	-18,6	-15,1	0,018215	22,4	-19,1753	-0,93
pmc-mir-8422	AGGAAAAGAUCAUCAGAACUUGCAAUUAAAAGUAgcacaaacaagaaagcaagaCCAAGUGUUAGUAAAAUGCAUGUUGAGGAUGAUUCUUUUUCA	96	15,625	30,20833	9,375	23,95833	25	54,16667	0,793103	1,666667	-23,6	-23,1	0,141217	4,11	-24,5833	-0,98333
pmc-mir-8443	ACCAGAUUGGUGGUACUUUGCAUUCAUAUUUGAGUUUCGUAACAUUCUUUUCAGCUGGGUGAUAUG	92	21,73913	28,26087	14,13043	35,86957	35,86957	64,13043	1,269231	1,538462	-18,7	-18,6	0,076444	11,42	-20,3261	-0,56667

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
	GUAGGUAAUAGUACUACAAAACUAAG															
pmc-mir-8460	AAGGUCAGCUAUAGUUUCCUUGUCACAcuaauuauauuauauuugcuUUCUAGGGAAUUAUAGGACGAGGCAGUCACUAAGCUGACGGG	89	21,34831	21,34831	14,60674	20,22472	35,95506	41,57303	0,947368	1,461538	-23,7	-20,9	0,049028	10,23	-26,6292	-0,74062
pmc-mir-8462	aaacugucaagcAUCUUUCUUUUGGAAACAAGUAAAUCAUAUGCCACCUGUCAGUGACAUUAUGAUUAAUUUGAUAAGAUGAAAUGGUCUUCAACAGCAG	100	15	30	14	29	29	59	0,966667	1,071429	-19,5	-18	0,088648	10,6	-19,5	-0,67241
pmc-mir-8470	AAAUUAAAACUAUAUAUUGUGAUGCAAUAGCUAGGAUGGCAAAAUUUUGUGCCUGAAAGCUGAGCAUAAGAAUAUGUucuuuaacau	87	18,3908	34,48276	9,195402	26,43678	27,58621	60,91954	0,766667	2	-19,4	-18,4	0,195062	6,64	-22,2989	-0,80833
pmc-mir-84f	uuguauugucaUGUUUCACAAGAGAUGAGGUAGUUUUAAAUGAAGCUUGAUAUUUAAAGCACCAUCUGGUGCUGAAAAUGGAUAGAUAGUU	91	20,87912	29,67033	8,791209	28,57143	29,67033	58,24176	0,962963	2,375	-21,2	-20,2	0,110814	22,46	-23,2967	-0,78519
pmc-mir-85	AGGUGACAAGGGAUCUGGGUGUCAGUGGGUGGGUGACCUCAGGACCUCAUCAAGUCCCGGGUACAAAGUAUUUGAAACCACAGAGUCGCUAGUGCAAGA	99	32,32323	27,27273	20,20202	20,20202	52,52525	47,47475	0,740741	1,6	-26,6	-24,3	0,014883	18,45	-26,8687	-0,51154
pmc-mir-8834a	UUGCUGCCGGGCCUGGAGGCCAGCUGUACCACACAUCUGCUGCCCCUCCACACCUGCGGCAGGUG	65	29,23077	13,84615	38,46154	18,46154	67,69231	32,30769	1,333333	0,76	-28,3	-28,3	0,089597	7	-43,5385	-0,64318

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-888	UCAAAGGACUGACACCUCUUUGUCCAGCUCAUGACAGGCUGCACAGAUGAGCCGGACAAGUGUUGUUAGUGGCUUCAU	78	25,64103	24,35897	24,35897	25,64103	50	50	1,052632	1,052632	-25,5	-24,5	0,08259	6,42	-32,6923	-0,65385
pmc-mir-8993	aucgCAGAACGUGACCUCUAUUacagggucaagggucaaaCAUGGUUAUGUCCGCCAUCUUGCUGGUGACCCUGUGGAGAAUUUACGugaucugcuuu	98	16,32653	13,26531	16,32653	20,40816	32,65306	33,67347	1,538462	1	-39,4	-39,4	0,233529	5,01	-40,2041	-1,23125
pmc-mir-9-1	UUUGGCGUCCAUUUUUGCCUUUGGUAACCUAGCUUUAUGAUAUUUCUGCCCCAAUCAUACAGCUAGAUAACCAAAGACAAAACCGGAGCCGCC	93	17,2043	26,88172	26,88172	29,03226	44,08602	55,91398	1,08	0,64	-34,1	-34	0,28369	3,42	-36,6667	-0,83171
pmc-mir-9120	CACUUACAUUCAUAUAUACACUCUUGUUcagucucccucucucacacacacaagcagcagCAAAGAGGUUGUAACACGAGAGAAGAUGAGAAAGUAAGGA	100	14	27	10	17	24	44	0,62963	1,4	-19,1	-18,3	0,095566	17,68	-19,1	-0,79583
pmc-mir-9170	AUAGCUCUUAAGAGCCUGAAUAGCCGGCGAGUCACCAGAGUCAAGUGUUGAAGGUGAUGUUUGCAUCCUGGCUCUGCUAUCUACAGUAUUAAGUGCUAU	99	25,25253	25,25253	20,20202	29,29293	45,45455	54,54545	1,16	1,25	-25,6	-23,2	0,013327	17,12	-25,8586	-0,56889
pmc-mir-9-2	GGCGGCUCCGGUUUUGUCUUUGGUUAUCUAGCUGUAUGAUUGGGGCAGAAAUAUCAUAAAGCUAGGUUACCAAAGGCAAAAAUGGACGCCAAA	93	26,88172	29,03226	17,2043	26,88172	44,08602	55,91398	0,925926	1,5625	-45,5	-45,5	0,314517	3,87	-48,9247	-1,10976

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-9256b	GGGGUGGGCGGCUACCCGCGGUGAGGUCUCACGAGGUGUAAUAAUCAGGCCACUACCUCUUACCGAGACCUCACAGGGGGCCGCUUCACAUU	92	31,52174	19,56522	29,34783	19,56522	60,86957	39,13043	1	1,074074	-43,8	-42,6	0,14803	4,74	-47,6087	-0,78214
pmc-mir-92a-1	CUGGCAUUAGCAGUAGGUCUUGAUGGGUGCAAUUCUGAUGAAUUAAAUAACAGAUUGCACUUGUCCCGGCCUUCUGCCAAUAGCUAA	87	22,98851	26,43678	20,68966	29,88506	43,67816	56,32184	1,130435	1,111111	-35,5	-35,5	0,154162	3,54	-40,8046	-0,93421
pmc-mir-92a-2	AUUUGUGUGCUCAGGAAAGGUUGUGACUUGGGCAAUUCUGUGAUGUCCUGUCAGAUUGCACUUGUCCCGGCCUUGUCUUGGCUUAcacuu	90	26,66667	14,44444	18,88889	34,44444	45,55556	48,88889	2,384615	1,411765	-31,5	-29,1	0,036643	11,93	-35	-0,76829
pmc-mir-92b	AGCGAUCAGAGUUGUGUAGACCGGAACAGGGGCAAUGCAUUUGAGUCGUCUGUAUUGCACUCGUCCCGGCCUAUCCAAUCUGACUCAUA	89	25,8427	23,59551	24,7191	25,8427	50,5618	49,4382	1,095238	1,045455	-32,6	-32,6	0,172648	4,96	-36,6292	-0,72444
pmc-mir-92c	AUUAUGUGUUAUUGUAAAAGGUUGGGAUGUGGGCUCUGUCGCGCAAUAGAUAACUUUCUUUGACAAGCAUUCGCCCACACCUUAUUACAACACAAUCAC	99	19,19192	28,28283	21,21212	31,31313	40,40404	59,59596	1,107143	0,904762	-26,6	-26,6	0,055246	15,72	-26,8687	-0,665
pmc-mir-9341	ACCUGAUUGUUAGUUAAAAGGGCCGAGGAUAGUCAGGCAUAUCGACAAGCUGUAGUAUCGCUUCUCGGCCUUUUGGCUAAGAUCAAAG	88	26,13636	27,27273	19,31818	27,27273	45,45455	54,54545	1	1,352941	-31,1	-21,81	0,019761	18,12	-35,3409	-0,7775

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-9369	CCAACAGGCCUGACCAGAAUACAUAUUCUUCUUUCCAUUUCCUUACAGUGUUGCUGAUUGUUUGAUAAGAUUUGUCAGUCUUAGUGCCUGUCAU	94	17,02128	22,34043	22,34043	38,29787	39,3617	60,6383	1,714286	0,761905	-18,9	-18,8	0,083295	15,62	-20,1064	-0,51081
pmc-mir-9382	GAUGUUGACCAUCGGAAUAUCACAGUGUGGCUGAAAUAUGCUGAGAUGGAAAUGAAGUAaguauuaauaaauuuucugugaUAUUCAUGCUAAACCAA	98	18,36735	27,55102	11,22449	20,40816	29,59184	47,95918	0,740741	1,636364	-19,3	-15,05	0,237583	13,37	-19,6939	-0,66552
pmc-mir-9434	UAGUCAGUGAGCAGACUUUCUGUCGUUGGUUAUGUAACGGCUCGCAGCUCUGACAGGAAUUCUGAACCCUGAUGU	75	26,66667	21,33333	21,33333	30,66667	48	52	1,4375	1,25	-23	-19,2	0,186112	9,2	-30,6667	-0,63889
pmc-mir-9505	UCUUGAAGUUAGGUUUAUGAUGUCUGUCAAAAUGUGAUCUCUCACGAAGGCAGACACCGUGACCCUUUCUUCAUGC	76	21,05263	23,68421	22,36842	32,89474	43,42105	56,57895	1,388889	0,941176	-29,4	-29,4	0,503167	1,22	-38,6842	-0,89091
pmc-mir-9542a	CACCGACUGACAGAACGCCUCACGACAUGGCCUGGCACUUUCUAAUUUCUGGCCAAGUUAGGCGCCUGUUUGUCGAGG	78	25,64103	20,51282	29,48718	24,35897	55,12821	44,87179	1,1875	0,869565	-29,2	-29,2	0,234782	2,71	-37,4359	-0,67907
pmc-mir-9543a	GAGAAACUAGCAAGUUCAUUAUAUGUCAGUAUGCAGAUUGCAAUGGUUUACUUUCCAUUGCACUGCACUGGAGUAAAUAUugcuuguuuauu	92	17,3913	27,17391	14,13043	28,26087	31,52174	55,43478	1,04	1,230769	-28	-26,4	0,033319	14,41	-30,4348	-0,96552

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-96	UACAUaguauuuauuuacaaucaugUGCAGUGCAGUUCUUUAUAUAACUAAUCAUACUGCAAAAAGUACAUGACUGCAAUAUGCAAAAAACAUAUCAC	98	9,183673	32,65306	15,30612	22,44898	24,4898	55,10204	0,6875	0,6	-19,5	-19,1	0,124316	9,09	-19,898	-0,8125
pmc-mir-969	AAGAAGCGCGAAUCUUCUUCUCCUGGaucaggaaaauaaauuauuuaagaGUUCCACUAAGCAAGUUAUCUUGCAGGAGCUAGAAAAUCGCCCUUAGA	98	16,32653	21,42857	18,36735	19,38776	34,69388	40,81633	0,904762	0,888889	-22,7	-22,7	0,05101	17,67	-23,1633	-0,66765
pmc-mir-96b	GCUGGCUGUUCAAUUAUUUGGCACUUGUGGAAUAAUCGGUGUAUCUAAAAGUCGAUUAUACACCGGUGCCAAGUCAUCAGAAAUGCCUGA	90	23,33333	27,77778	18,88889	30	42,22222	57,77778	1,08	1,235294	-30,3	-30,3	0,149015	2,78	-33,6667	-0,79737
pmc-mir-970	AAGAGAAGCCUUGCAUGAGCUCUAGGUgucuccagauguuuguacagaguauucaGCAGGAACAGCUUGGAUCAUCUACACCUCUCCCAGCUCUAUA	97	15,46392	19,58763	19,58763	16,49485	35,05155	36,08247	0,842105	0,789474	-20,4	-15,2	0,030264	26,39	-21,0309	-0,6
pmc-mir-971	UGCUGAUGAAAGUGAAAUGAUCUCGAUGGUGUUCUAGCCUCCAUCGACAUAGAAAAAACACCAUCAUGCAGAUACAUGCCUUUAUCAUGU	90	18,88889	32,22222	21,11111	27,77778	40	60	0,862069	0,894737	-21,2	-20,1	0,039445	9,59	-23,5556	-0,58889
pmc-mir-981	GCUAUCAGACUUUGUUCCGGGUUUCGCGGCUUGCGAACAGUCAUUUUGAGCUGUGUUCGUUGUCGUCGAAACCUGCCCUGAAAUCUGAUGUG	92	27,17391	16,30435	22,82609	33,69565	50	50	2,066667	1,190476	-35,2	-30,7	0,023194	9,17	-38,2609	-0,76522

Pre-miRNA	Sequence	Length (AA)	G (%)	A (%)	C (%)	U (%)	GC (%)	AU (%)	R-AU	R-GC	MFE (kcal/mol)	MFEE(kcal/mol)	Freq	Div	AMFE (kcal/mol)	MFEI (kcal/mol)
pmc-mir-9851	AUGACAACAGCAGCUCAACUGUUACUGUGGAUGGCACCAGCACUGGCACUGCCAUGGAUCAGAGCAGUGAUGUCAGUUGUUUU	83	26,50602	25,3012	22,89157	25,3012	49,39759	50,60241	1	1,157895	-26,3	-24,6	0,066358	12,72	-31,6867	-0,64146

Supplementary Table 5: – Structural characterization of miRNAs in Apullariidae species, lny - *L. nyassanus*, mcr - *M. cornuarietis*, pcn - *P. canaliculata*, pmc - *P. maculata*.

Mature miRNA	**Mature miRNA sequence**	**miRNA length**	**Mature miRNA**	**Mature miRNA sequence**	**miRNA length**	**Mature miRNA**	**Mature miRNA sequence**	**miRNA length**	**Mature miRNA**	**Mature miRNA sequence**	**miRNA length**
pmc-bantam-3p	UGAGAUCAUUGUAAAAACCAAUU	23	pcn-bantam-3p	UGAGAUCAUUGUAAAAACCAAUU	23	mcr-bantam-3p	UGAGAUCAUUGUAAAAACCAAUU	23	lny-bantam-3p	UGAGAUCAUUGUAAAAACCAAUU	23
pmc-bantam-5p	CUGGUUUUCACAGUGAUCCAGCAG	24	pcn-bantam-5p	CUGGUUUUCACAGUGAUCCAGCAG	24	mcr-bantam-5p	CUGGUUUUCACAGUGAUCCAGCAG	24	lny-bantam-5p	CUGGUUUUCACAGUGAUCCAGCAG	24
pmc-miR-1000-5p	CUGGUGUGAUGGGACAAGACUU	22	pcn-let-7b-3p	UGAGGUAGUGGUUGUAUCGUAU	22	mcr-miR-1002-3p	CUAAGUAGUUGAUACAUGAGUUG	23	lny-miR-10349-5p	UGAGCACCAAUAAGCAGCAAUAAG	24
pmc-miR-10031c-5p	CUCGUCCAUGUCUUCCUCUUCG	22	pcn-let-7d-3p	CUAUACAACCUGCUGCGGCAGG	22	mcr-miR-10080-5p	UUUCUCAGGUUGUCUCAAUC	20	lny-miR-10365-5p	AUUAGAAUGUGGAAUCUAUGCUU	23
pmc-miR-10046-3p	ACUUGUCGCAGUGACGACA	19	pcn-let-7f-5p	CUGCACAUUCCACCAUCCAUU	21	mcr-miR-10089-3p	UAAUUUGCUGUUUUGGGAUGCU	22	lny-miR-106a-5p	UACUGCAAUGCAAGCACAUUGA	22
pmc-miR-10055-3p	GUGGGAGGGAGGGAGGACUUU	21	pcn-miR-1002-3p	CUAAGUAGUUGAUACAAACAGUA	23	mcr-miR-10141-3p	AGAAGAAACACGCAUUCGGUCU	22	lny-miR-10790-3p	UUUAUUUCUCUCUCUGUUCGUC	22
pmc-miR-10089-5p	UAAUUUGCUGUUUUGGUGAUAU	22	pcn-miR-10a-5p	ACCCUGUAGAUCCGAAUUUGUGUA	24	mcr-miR-10228-3p	UGAGUAUUGUACCAUGUCAGCA	22	lny-miR-10a-5p	UACCCUGUAGAUCCGAAUUUGUGU	24
pmc-miR-10173-3p	AGGAAGUGAGGAAGGAAAACAA	22	pcn-miR-11-3p	CAUCACAGUCAGAGUUCACUGA	22	mcr-miR-10237-3p	AGCAGGGACUGGUUUGUGGCAC	22	lny-miR-10b-5p	UUACCCUGGAGAACCGAGCGUGUG	24
pmc-miR-10249-1-3p	GUGGUGGCGGUGGUGUUGCCAG	22	pcn-miR-1175-3p	UGAGAUUCAACUCCUCCAACUGC	23	mcr-miR-10325-5p	UAUCAUCGUCUUCUAUACAAGUGG	24	lny-miR-11225-3p	AAAACUGUUCUUAAGUCAUGGC	22
pmc-miR-10249-2-3p	GUGGUGGCGGUGGUGUUGCCAG	22	pcn-miR-1175-5p	AGUGGAGAGAGUUUUAUCUCAUC	23	mcr-miR-10379-3p	ACUGUUUUGUUCGUUGAACUAU	22	lny-miR-11-3p	CAUCACAGGCAGAGUUCCAGA	21
pmc-miR-10256-3p	CAUGUCUGCUAUGGCCUUCC	20	pcn-miR-1187-3p	UAUGUGUGUGUGUAUGUGUGUG	22	mcr-miR-10456-3p	GAUGCAGGAUAUUUAUGACA	20	lny-miR-11-5p	CAUCACAGGCAGAGUUCCAGAC	22
pmc-miR-10267-3p	CAGAUCAAACCACCGCCCCC	20	pcn-miR-122b-5p	AGUGUGACACUGGUGUGACA	20	mcr-miR-10457-3p	UUUGUUUGUUGUGUGUCUAC	20	lny-miR-1175-3p	UGAGAUUCAACUCCUCCAACUGC	23

pmc-miR-10492c-3p	GUUUAGCUCUCUGAUUUAUG	20	pcn-miR-1238-5p	CUUCCUCGUCUGUCUGAAGC	20	mcr-miR-10698-5p	UCAUGCAUGGCUUCUUUAGUA	21	lny-miR-1175-5p	AGUGGAGAGAGUUUUAUCUCAUC	23
pmc-miR-10508b-3p	AACAGCAUUUCGUGACUUCUU	21	pcn-miR-12-3p	AGUACCUUUUGUGAUAUUCUUA	22	mcr-miR-106b-3p	GACCGCACUGUGGGUACCCCUG	22	lny-miR-11976-5p	GGCGGCGGCGGCGCCGGCGUG	21
pmc-miR-10509-5p	UGUGUCACGUACAGUAACACUGA	23	pcn-miR-124-3p	UUAAGGCACGCGGUGAAUGCCAA	23	mcr-miR-10717-5p	CACGGAUGCACACACACCUGCG	22	lny-miR-12096b-3p	UAGGGCAAAAUUAUCACCGAUUAC	24
pmc-miR-10578-5p	AGCAGUGAGUCAGUCUCACGCU	22	pcn-miR-124-5p	GUGUUCACUGCGUUGGCCUUGGU	23	mcr-miR-10738-3p	CGGGUGCUGCUGUCUGCUUUG	21	lny-miR-12096b-5p	AAGCGGUGAUACUUUUGUCUCGCA	24
pmc-miR-10611-3p	UUUCCCUUUUUCACACAGUGGG	22	pcn-miR-124c-5p	CAUUCACCGCGUGCCUUAAUA	21	mcr-miR-10761-3p	UUGAAUUUGUUCUUGUCACCAU	22	lny-miR-12-3p	AGUACCUUUUGUGAUAUUCUUA	22
pmc-miR-10639-3p	UUGCCAUGAUAGCUUUGG	18	pcn-miR-12-5p	UGAGUAUUACAUCAGGUACUGA	22	mcr-miR-10863-3p	UGACUUCUUUGUGGGCACGAUG	22	lny-miR-124-3p	UUAAGGCACGCGGUGAAUGCCAA	23
pmc-miR-10770-1-3p	UUGUUGAUGUUGUUGUUGUUGU	22	pcn-miR-1277-1-3p	UAUAUAUAUAUAUGUACGUCUA	22	mcr-miR-10903-1-3p	GACUCGCUUGUGUCGGCCUC	20	lny-miR-124-5p	GUGUUCACUGCGUUGGCCUUGGU	23
pmc-miR-10770-2-5p	UUGUUGAUGCUGUUGUUGUUUC	22	pcn-miR-1277-2-3p	AAAUAUAUAUAUAUAUGUAUGAU	23	mcr-miR-10903-2-3p	GACUCGCUUGUGUCGGCCUC	20	lny-miR-124c-5p	CAUUCACCGCGUGCCUUAAUA	21
pmc-miR-10892-3p	UGAAUAAUGGAUGAAUAACAUC	22	pcn-miR-1322-5p	GAUGAUGCUGCUGAUGACA	19	mcr-miR-10903-3-3p	GACUCGCUUGUGUCGGCCUC	20	lny-miR-12-5p	UGAGUAUUACAUCAGGUACUGA	22
pmc-miR-10972-3p	UUUGCUAUGGUCAUCAUAUAUG	22	pcn-miR-1328-5p	GAGAGAGAAAUGAGAAAC	18	mcr-miR-10918-3p	CAUAUAAGCUAUGUUACUCGAU	22	lny-miR-1307-5p	ACUCGGCGUGGCGUCGGUGCAAA	23
pmc-miR-10b-1-3p	CAUCUCGGUACUCGGGAGGUCA	22	pcn-miR-133-3p	UUUGGUCCCCUUCAACCAGCUGUA	24	mcr-miR-10967-3p	UGAACUGUGAACUGCAUACUG	21	lny-miR-133-3p	UUUGGUCCCCUUCAACCAGCUGUA	24
pmc-miR-10b-2-5p	UUACCCUGGAGAACCGAGCGUGUG	24	pcn-miR-133-5p	AGCUGGUUGAACUCGGGCCAAAU	23	mcr-miR-10a-5p	ACCCUGUAGAUCCGAAUUUGUGUA	24	lny-miR-133-5p	AGCUGGUUGAACUCGGGCCAAAU	23
pmc-miR-11054-5p	AGUGUUUGUUGCCAGAGAUUUA	22	pcn-miR-133c-5p	CAGCUGGUUGAAGGGGACCAA	21	mcr-miR-10b-5p	UUACCCUGGAGAACCGAGCGUGUG	24	lny-miR-133c-5p	CAGCUGGUUGAAGGGGACCAA	21
pmc-miR-11280-3p	AUGAGAGAAGAUCCGUGUGUGA	22	pcn-miR-137b-3p	UUAUUGCUUGAGAAUACACGUAA	23	mcr-miR-11-3p	GGAGCUCUGGCUGUGCCGGGUG	22	lny-miR-137b-3p	UUAUUGCUUGAGAAUACACGUAA	23
pmc-miR-1175-1-3p	UGAGAUUCAACUCCUCCAACUGC	23	pcn-miR-137b-5p	ACGGGUAUUCUUGGGUAAAUAAUA	24	mcr-miR-11591-3p	GGCUGUGCUGUAAGUGUACUUU	22	lny-miR-137b-5p	ACGGGUAUUCUUGGGUAAAUAAUA	24
pmc-miR-1175-1-5p	AGUGGAGAGAGUUUUAUCUCAUC	23	pcn-miR-13b-3p	UCGUUAAAAUGUUUGUG	17	mcr-miR-1175-3p	UGAGAUUCAACUCCUCCAACUGC	23	lny-miR-153-3p	UUGCAUAGUCACAAAAGUGAUCG	23
pmc-miR-1175-2-5p	AAGUGAAGAGAAGAAUGAAGAA	22	pcn-miR-1-3p	CAUGCUUCUUUAUAGUUUGACC	22	mcr-miR-1175-5p	AGUGGAGAGAGUUUUAUCUCAUC	23	lny-miR-1662-3p	UUGACAUCAUCAUACUUUAUGU	22
pmc-miR-1187-3p	UAUGUGUGUGUGUAUGUGUGUAU	23	pcn-miR-1421a-3p	CUGCAUCCCUUGUGACGGUGGC	22	mcr-miR-12037-5p	CAUUCUGUCUUCUCUUCCAA	20	lny-miR-1731-5p	ACUUGACUGCAGGCACUACAGAU	23
pmc-miR-11922-3p	UUCAGGAGAUCAUCGAUAUGUU	22	pcn-miR-1421am-3p	AGGAGGUCGUAGGUCAUCCUGU	22	mcr-miR-12093-3p	UCAGCCCCUGGGGUAGCCUUAC	22	lny-miR-184-3p	ACUGGACGGAGAACUGAUAAGGGC	24
pmc-miR-12096b-3p	UAGGGCAAAAUUAUCACCGAUUAC	24	pcn-miR-143-3p	GUCUGAGAUGAAGCACUCGUUUG	23	mcr-miR-12096b-3p	UAGGGCAAAAUUAUCACCGAUUAC	24	lny-miR-184-5p	CCUUAUCACUCCCCCGCCCCGU	22
pmc-miR-12096b-5p	AAGCGGUGAUACUUUUGUCUCGCA	24	pcn-miR-153-1-3p	UCAUUUUUGUGAUGUUAUCAAUA	23	mcr-miR-12096b-5p	AAGCGGUGAUACUUUUGUCUCGCA	24	lny-miR-184b-5p	CCUUAUCAGUUCUCCGUCCAGU	22
pmc-miR-12228-5p	UCCCCACUGCUCCCCCAACA	20	pcn-miR-153-2-3p	UUGCAUAGUCACAAAAGUGAUCG	23	mcr-miR-12193-3p	ACCGGUCCUUUCUGUCUAAUG	21	lny-miR-190-5p	AGAUAUGUUUGAUAUAUUUGGUG	23

pmc-miR-12245-5p	CCCCGCCCCGCCCGGCUCAAGG	22	pcn-miR-1584-5p	CCGGGUGGGGCUGGGCCAUA	20	mcr-miR-12265-5p	UCAGCCGUCUUGCAGCUGAAG	21	lny-miR-1947-3p	GCACUGAGCUAGCUCUCUAGCA	22
pmc-miR-1224-5p	UCACCUCCUCUCUCCUCCCUU	21	pcn-miR-15c-3p	CAGACCAUUCUGGGCUAUGGGG	22	mcr-miR-12293-5p	CGCCUCUUGAUCUACUACAACUA	23	lny-miR-1985-5p	UGCCAUUUUUAUCAGUCACUGUGU	24
pmc-miR-12286-3p	UGGGGCUGCAGGGCUGAGUAAGC	23	pcn-miR-1602-5p	UGGGCUCUGCAUCACCAGCAGU	22	mcr-miR-12313-5p	ACAUGUUAGCAUCUUGACAUAA	22	lny-miR-1990-3p	CGGGACUACGUCAACGUACUA	21
pmc-miR-12287-5p	UGGGGGUGCAGGUGGGCACAC	21	pcn-miR-1603-3p	GUGGUUUGUUUUGUGUUGCG	20	mcr-miR-12322-3p	UCUGGUCAUUUCUCUUUCUCAC	22	lny-miR-1990-5p	AGUAAGUUGAUGGGGUCCCAGG	22
pmc-miR-12292-5p	AUGGGGUCUUGGGGUCAUG	19	pcn-miR-1632-3p	UGCUUGUUUUUGGAUGAAAUUAU	23	mcr-miR-12352-5p	UAUUGCUGUUAUGGUGUUAGUU	22	lny-miR-1993-3p	UAUUAUGCUGCUAUUCACGAGA	22
pmc-miR-12293-5p	CGCCUCUUGAUCUACUAUAACUA	23	pcn-miR-1677-3p	UUGACUUCAAUAGGAGAGACAGG	23	mcr-miR-12-3p	UGAGUAUUACAUCAGGUACUGA	22	lny-miR-1993-5p	UCGGGAAUAUCGGCAUAAUGCA	22
pmc-miR-12296-3p	GGGCAGGGGCAGGGGGCA	18	pcn-miR-1744-1-5p	ACUUCAACAGGAGCAAGUCAAG	22	mcr-miR-1250-5p	ACGGUGCUGGAUGUGGAUAU	20	lny-miR-1994-3p	UGAGACAGUGUGUCCUCCCUCU	22
pmc-miR-12321-5p	GCCCUGCCCUCCCUUCCCCUCUC	23	pcn-miR-1744-2-5p	ACUUCAACAGGAGCAAAGUGUA	22	mcr-miR-1256-5p	AGGCAUUGACUUCUCUUCAUGG	22	lny-miR-1a-3p	UGGAAUGUAAAGAAGUAUGUAC	22
pmc-miR-12327-5p	UUUCUCUCUUUCCCUCUCUCU	21	pcn-miR-184-3p	ACUGGACGGAGAACUGAUAAGGGC	24	mcr-miR-1277-3p	UAUAUAUAUAUAUGUACGAUGC	22	lny-miR-1a-5p	ACAUACUUCUUUGCUAUCCCAUAU	24
pmc-miR-12339-3p	AGAUUGUUCUGGAUUGCCACUG	22	pcn-miR-184-5p	CCUUGUCACUCCUCCGCCCCGU	22	mcr-miR-1278-3p	UAGUACUGUGCAUAUCAGCAC	21	lny-miR-1c-5p	GUACAUACUUCUUUACAUUCCA	22
pmc-miR-12358-3p	AUCUAUGAUUUCAUCGGGUAAG	22	pcn-miR-1905c-3p	CACCACCAGCCCCACCACGCGGUAU	25	mcr-miR-1304-3p	UCUCACUGUAGCCUCGGCUCAG	22	lny-miR-2001-5p	UUGUGACCGUUACAAUGGGCAUU	23
pmc-miR-12396-5p	AGAGAGACACAGACAAGAAAGAU	23	pcn-miR-190-5p	AGAUAUGUUUGAUAUAUUUGGUG	23	mcr-miR-130a-3p	UAGUGCAAUGUUAAAAGAGUAC	22	lny-miR-2049-3p	GAUUCAUGUGCAUGCAUUCAUG	22
pmc-miR-12-3p	AGUACCUUUUGUGAUAUUCUUA	22	pcn-miR-1951-3p	GUAGUGGAGACUGGUGGCGAAA	22	mcr-miR-133-3p	UUUGGUCCCCUUCAACCAGCUGUA	24	lny-miR-2064-3p	AAGCAGCACUGUGCAACUGCAA	22
pmc-miR-12410-3p	AAGGAAACUGAGAUCCGGUCAC	22	pcn-miR-1976-5p	CUCCUGCCCUCCUUGCCCAGA	21	mcr-miR-133-5p	AGCUGGUUGAACUCGGGCCAAAU	23	lny-miR-216a-3p	CAAGUUACUCGCCGAGAUUACA	22
pmc-miR-124-5p	CGCUUUCAUCCGUGACGACACC	22	pcn-miR-1984-5p	UGCCCUAUCCGUCAGGAACUGUG	23	mcr-miR-133c-5p	CAGCUGGUUGAAGGGGACCAA	21	lny-miR-216a-5p	UAAUCUCAGCUGGUAAUUCUGAG	23
pmc-miR-12-5p	UGAGUAUUACAUCAGGUACUGA	22	pcn-miR-1985-5p	UGCCAUUUUUAUCAGUCACUGUGU	24	mcr-miR-137b-3p	UUAUUGCUUGAGAAUACACGUAA	23	lny-miR-228-5p	AAUGGCACUGCAUGAAUAAAAAA	23
pmc-miR-1273h-3p	CUGCAGACUCGACCUCAGGAU	21	pcn-miR-1986-3p	UGGAUUUCCCAAGAUCCGUGAU	22	mcr-miR-137b-5p	ACGGGUAUUCUUGGGUAAAUAAUA	24	lny-miR-2361-5p	GUUGUGUUGUUUUUUUUUCA	20
pmc-miR-1277-3p	UAUAUAUAUAUAUGUACUCUAC	22	pcn-miR-1990-3p	CGGGACUACGUCAACGUACUA	21	mcr-miR-1421ai-5p	AGCAGGUCAGGUGUGGAAGCAC	22	lny-miR-239b-3p	GCACUUUUGUGGUGUGCAGGUU	22
pmc-miR-130b-3p	CAGUGCAAUAAUGAAAGUAUUUU	23	pcn-miR-1990-5p	AGUAAGUUGAUGGGGUCCCAGG	22	mcr-miR-143-3p	UGUGCAGUGCUGCAUCUCACA	21	lny-miR-2491-3p	CAACAACAGCAGCAGCAG	18
pmc-miR-1322-5p	GAUGCUGCUGCUGAUGGC	18	pcn-miR-1992-3p	UCAGCAGUUGUACCACUGAUUUG	23	mcr-miR-14-3p	UCAGUCUUUUUCUCUCUCUUUU	22	lny-miR-2493-3p	GCUCACACACACACACAGACG	21
pmc-miR-133c-5p	CAGCUGGUUGAAGGGGACCAA	21	pcn-miR-1992-5p	AGUCAGUGGUUGACUGCUCGGU	22	mcr-miR-146b-3p	UGAGAACUGGUUUUGAGCAAGU	22	lny-miR-252a-5p	CUAAGUACUAGUGCCGCGGGAG	22
pmc-miR-1376-3p	GCUCAGCAGCACUCUUUGAGC	21	pcn-miR-1994-3p	UGAGACAGUGUGUCCUCCCUCU	22	mcr-miR-153-1-3p	GUCAUUUUUGUGAUUUUGAGCU	22	lny-miR-252b-1-3p	CCCUGCGCAGUCCUACUGGG	20

pmc-miR-137b-3p	UUAUUGCUUGAGAAUACACGUAA	23	pcn-miR-1994a-3p	UGAGACAGUGUGUCCUCCCUUG	22	mcr-miR-153-2-3p	UUGCAUAGUCACAAAAGUGAUCG	23	lny-miR-252b-2-5p	AUAAGUAGUGGUGCCGCAGGUA	22
pmc-miR-137b-5p	ACGGGUAUUCUUGGGUAAAUAAUA	24	pcn-miR-1996b-3p	AUCAAGUGAGGUCAGAAUA	19	mcr-miR-1599-5p	GGAGGGAGGAAAAAAAAACG	20	lny-miR-2580-3p	CGUGUCUGUGUGUAUGUUUCUA	22
pmc-miR-13b-3p	UCGUUAAAAUGUUUGUG	17	pcn-miR-1a-3p	UGGAAUGUAAAGAAGUAUGUAC	22	mcr-miR-15a-5p	UAGCAGCACAGAAUGGCAAAA	21	lny-miR-263a-1-5p	CUUAAUGGCACUGGUAGAAUAGCA	24
pmc-miR-1-3p	CAUGCUUCUUUAUAGUUUGACC	22	pcn-miR-1a-5p	ACAUACUUCUUUGCUAUCCCAUAU	24	mcr-miR-1610-5p	UGGCUUGUGGUGGAACGACAA	21	lny-miR-263a-2-5p	AAUGGCACUGGUAGAAUUCACGG	23
pmc-miR-1421am-3p	AGGAGGUCGUAGGUCAUCCUGU	22	pcn-miR-1c-5p	GUACAUACUUCUUUACAUUCCA	22	mcr-miR-1621-3p	ACCGGCUGCCUCGGUGGUAA	20	lny-miR-277a-3p	GAGCCAUUUCUUUGUGUGCU	20
pmc-miR-1422j-3p	AAAACUGCAUCUGGAUUAUGA	21	pcn-miR-2001-5p	UUGUGACCGUUACAAUGGGCAUU	23	mcr-miR-1628-3p	AAGAGCUCUUCCUGUUCGUGUG	22	lny-miR-279-3p	UGACUAGAUCCACACUCAUCCA	22
pmc-miR-143-3p	GUCUGAGAUGAAGCACUCGUUUGC	24	pcn-miR-2008-3p	GUAUUAACAAUGUGGCUACAGU	22	mcr-miR-1693-3p	GCAAAGGAUGAAGCUGUAGCU	21	lny-miR-281-3p	UGUCAUGGAGUUGCUCUCUUUA	22
pmc-miR-149-3p	GAGGGAGGGACGGGGGAUUCAAG	23	pcn-miR-203b-3p	UUGAACUGUUAAGAACAGUAAA	22	mcr-miR-1710-5p	UCAUCUGCUGCAUAACCGUGUG	22	lny-miR-281-5p	AAGGGAGCAUCCGUCGACAGU	21
pmc-miR-150-3p	CUGGUACAGGCCUGGGGGAGA	21	pcn-miR-204-3p	UGCAAGGACAGCAAAGGGA	19	mcr-miR-1761-3p	CAGGGGUCACUUUUUUCCCC	20	lny-miR-2944a-3p	GAAGGAACUUCUGCUGAUC	19
pmc-miR-153-3p	UUGCAUAGUCACAAAAGUGAUCG	23	pcn-miR-2047-3p	UUCGCCGAGCAGAUGUGUGAAG	22	mcr-miR-187-5p	GCUACAACACAGGACAAUUCUA	22	lny-miR-2987-3p	AGAGCAAGGCUGAGGCUU	18
pmc-miR-1551-3p	UGAGUUUGUGUUGCUGGGACAA	22	pcn-miR-2155-3p	GACACUGUUUUACACUCUACUGG	23	mcr-miR-190-5p	AGAUAUGUUUGAUAUAUUUGGUG	23	lny-miR-29a-3p	UAGCACCAUUUGAAAUCAGUUU	22
pmc-miR-15c-3p	CAGACCAUUCUGGGCUAUGGGG	22	pcn-miR-216a-3p	CAAGUUACUAGCCGAGAUUACA	22	mcr-miR-193-1-3p	UACUGGCCAGCACAAUUAGUGU	22	lny-miR-29b-1-3p	UAGCACCAUUUGAAAUCAGUGCU	23
pmc-miR-1603-1-3p	GUGGUUUGUUUUGUGUUGCG	20	pcn-miR-216a-5p	UAAUCUCAGCUGGUAAUUCUGAG	23	mcr-miR-193-2-3p	UACUGGCCAGCACAAUUAGUGU	22	lny-miR-29b-2-3p	AGCUGGUUUCACAUGGACAGAUAAC	25
pmc-miR-1603-2-3p	UUGGUUUGGUUUGUUUUUUU	20	pcn-miR-216b-1-3p	CACAGUGGCUACUGGGUCCACA	22	mcr-miR-193-3-3p	UACUGGCCAGCACAAUUAGUGU	22	lny-miR-2a-1-3p	UAUCACAGCCUGCUUGGAUC	20
pmc-miR-1632-3p	UGCUUGUUUUUGGAUGAAAUUAU	23	pcn-miR-216b-2-3p	UAAUAUCAGCUGGUAAUCCUGAG	23	mcr-miR-194-3p	CCAGUGGAGGUGGUGUAGAAGU	22	lny-miR-2a-2-3p	AAUCACAGCCUGCUUUGGUGAGCU	24
pmc-miR-1642-3p	UGAGAGGCUGUCAGUUUUGUAAG	23	pcn-miR-219-1-3p	AGAACUGUGUGUGGACAUCAGU	22	mcr-miR-1947-5p	GCACUGAGCUAGCUCUCUGCGU	22	lny-miR-2b-1-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24
pmc-miR-1677-3p	UUGACUUCAAUAGGAGAGACAGG	23	pcn-miR-219-1-5p	UGAUUGUCCAAACGCAAUUCUUG	23	mcr-miR-194b-3p	UGUAACAGCAUCUCCAACUGCGG	23	lny-miR-2b-2-3p	UAUCACAGCCAGCUUUGAUGAGC	23
pmc-miR-1728-3p	UGGUUGAUCCAUUGGAAGAAACA	23	pcn-miR-219-2-3p	AGAACUGUGUGUGGACAUCAGU	22	mcr-miR-196c-5p	ACAACAACAUCAAACCACCUGC	22	lny-miR-2d-3p	AAUCACAGCCUGCUUUGGUGAG	22
pmc-miR-17-3p	ACUGCACUGCAAGCACCGCCACA	23	pcn-miR-219-2-5p	UGAUUGUCCAAACGCAAUUCUUG	23	mcr-miR-1970-3p	UGUGUCACUGGGGAUAAUUA	20	lny-miR-306-3p	UCAGGUACUGAGUGACU	17
pmc-miR-1744-5p	ACUUCAACAGGAGCAAGUCAAG	22	pcn-miR-2196-5p	CCUCUCUGUGCUGCCAGCAAAAAU	24	mcr-miR-1984-5p	UGCCCUAUCCGUCAGGAACUGUG	23	lny-miR-3128-3p	UCUGGCAAGUAAAAACCAUCC	22
pmc-miR-1775-5p	UCCUGUAGCCAGAAGACAGCGG	22	pcn-miR-2218b-3p	AAAUUUGUAGUUUGUAUUCUUC	22	mcr-miR-1985-5p	UGCCAUUUUUAUCAGUCACUGUGU	24	lny-miR-316-3p	UGUCUUUUUCCGCUUACAAAUG	22
pmc-miR-1781-1-5p	UUUAAAUCAUCCAGCUGCCUU	21	pcn-miR-2284c-3p	AAAAUCUAAGUGAACUGUGACG	22	mcr-miR-1990-3p	CGGGACUACGUCAACGUACUA	21	lny-miR-3238-5p	AAAUGAAUCAUGCAUUUGGCAUGUA	25

pmc-miR-1781-2-5p	UUUAAAUCAUCCAGCUGCCUU	21	pcn-miR-2284t-3p	AAACUCGAAUGAAUGUAGUUCG	22	mcr-miR-1990-5p	AGUAAGUUGAUGGGGUCCCAGG	22	lny-miR-3297-3p	GCACAUCUAGUCAAAACCAAGGUUU	25
pmc-miR-1896-3p	UGGUGGGUGAGGAGGAGG	18	pcn-miR-2324-5p	UGGGGUUGGGGCAGUGACCAGCA	23	mcr-miR-1992-3p	UCAGCAGUUGUACCACUGAUUUG	23	lny-miR-33-3p	GCAAUGCAUCUGCAGUGCAAGUA	23
pmc-miR-190-5p	AGAUAUGUUUGAUAUAUUUGGUG	23	pcn-miR-2392-3p	AUGGAUGGGGGUGAGGGGGAUG	22	mcr-miR-1994-3p	UGAGACAGUGUGUCCUCCCUCU	22	lny-miR-33-5p	GUGCAUUGAGGUUGCAUUGCAU	22
pmc-miR-1951-3p	GUAGUGGAGACUGGUGGCGAAA	22	pcn-miR-2405-3p	UGUGUGGUUUGUUUGGUU	18	mcr-miR-1994a-3p	UGAGACAGUGUGUCCUCCCUUG	22	lny-miR-3484-5p	AUACAAACAGCUAAACUAGG	20
pmc-miR-1955-5p	GAGCAUUGCAUGCUGGACGCG	21	pcn-miR-2464-3p	ACUUCUGCUGCAGACCUCAAGGAU	24	mcr-miR-1c-5p	GUACAUACUUCUUUACAUUCCA	22	lny-miR-359-3p	UCACUGGUUUCCUCUGUUGCG	22
pmc-miR-196a-3p	ACGGCAACAAGAAACUGUGACU	22	pcn-miR-2470-3p	UUGUGUGGUUUUGGAUACUUG	21	mcr-miR-2001-3p	UUGUGACCGUUACAAUGGGCAUU	23	lny-miR-36b-3p	UCACCGGGUAUACAUUCAUCCGC	23
pmc-miR-1984-5p	UGCCCUAUCCGUCAGGAACUGUG	23	pcn-miR-2491-1-3p	CAACAACAGCAGCAGCAA	18	mcr-miR-202-5p	AGAGGCAUAGGGCAUGUGCGGG	22	lny-miR-3906-3p	AAAAGCAUUUUGAAUGCAUAAAA	23
pmc-miR-1985-5p	UGCCAUUUUUAUCAGUCACUGUGU	24	pcn-miR-2491-2-3p	CAACAACAGCAGCAGCAA	18	mcr-miR-2040b-5p	UCAGGUCAGCUACGACUGUAACA	23	lny-miR-3963-5p	UGUAUCCCACUUCUGACGC	19
pmc-miR-1990-3p	CGGGACUACGUCAACGUACUA	21	pcn-miR-2491-3-5p	CAACAACAGCAGCAGCAG	18	mcr-miR-204-3p	GCUGGGAAGGCAAAGGGAACC	21	lny-miR-4024-3p	UUUGUAGGAUGAAAAGGUU	19
pmc-miR-1990-5p	AGUAAGUUGAUGGGGUCCCAGG	22	pcn-miR-2500-3p	GGAUUUUGUGUGUGGAC	17	mcr-miR-216a-3p	CAAGUUACUAGCCGAGAUUACA	22	lny-miR-4262-3p	GACAUUCAGACUACCUG	17
pmc-miR-199-3p	ACAGUAGUCUGCACAUGCUAU	21	pcn-miR-252a-5p	CUAAGUACUAGUGCCGCGGGAA	22	mcr-miR-216a-5p	UAAUCUCAGCUGGUAAUUCUGAG	23	lny-miR-4265-5p	CUGUGGGCUCAGCUCUGCA	19
pmc-miR-1994-3p	UGAGACAGUGUGUCCUCCCUCU	22	pcn-miR-254-5p	UGCAAAUCUUUUGCAACAGACUU	23	mcr-miR-216b-1-3p	UAAUAUCAGCUGGUAAUCCUGAG	23	lny-miR-451-3p	UAGUAAUGGUAAGGGUUUG	19
pmc-miR-1994a-3p	UGAGACAGUGUGUCCUCCCUUG	22	pcn-miR-279-1-3p	UGACUAGAUCCACACUCAUCCA	22	mcr-miR-216b-2-5p	ACACUUACCUAUAGAGAUGUGU	22	lny-miR-466b-5p	AUAUACAUACACACAUACUAC	21
pmc-miR-1a-3p	UGGAAUGUAAAGAAGUAUGUAC	22	pcn-miR-279-2-3p	UGACUAGAUCCACACUCAUCCA	22	mcr-miR-216b-3-3p	ACACUUACCUAUAGAGAUGUGU	22	lny-miR-466h-5p	UACGCACGCACACACACAC	19
pmc-miR-1a-5p	ACAUACUUCUUUGCUAUCCCAUAU	24	pcn-miR-279b-5p	UGACUAGACUGAUAACCCCUAG	22	mcr-miR-2238k-3p	AAUGACUGAACAGUGCAAUGCA	22	lny-miR-466i-1-5p	UGUGUGUGUGUGUGAGUG	20
pmc-miR-1b-3p	UCAUACUUCUUUACAUU	17	pcn-miR-2804-3p	GUAGUGUAUUACAAUACUGACC	22	mcr-miR-2253a-5p	CAUGGCACUGCUGAAGGUUCA	21	lny-miR-466i-2-3p	AUACACACACACACACACACAU	22
pmc-miR-1c-5p	GUACAUACUUCUUUACAUUCCA	22	pcn-miR-281-3p	UGUCAUGGAGUUGCUCUCUUUA	22	mcr-miR-2254-3p	UUUGUACUAUGCAAAGUAAGUAC	23	lny-miR-466i-3-3p	UGUGUGUGUGUGUGUGUG	20
pmc-miR-2001-5p	UUGUGACCGUUACAAUGGGCAUU	23	pcn-miR-281-5p	AAGGGAGCAUCCGUCGACAGU	21	mcr-miR-2288-3p	AGGUAGUAGGUGUGUGUGUG	20	lny-miR-466i-4-3p	UGUGUGUGUGUGUGUGUG	20
pmc-miR-200b-5p	CAUCUUACCUGACAGUCAACAG	22	pcn-miR-2962-3p	CAUCCCCUGCCUCUCUCAUAC	21	mcr-miR-22b-3p	ACAGUUCUUCAACUGGACUGAG	22	lny-miR-466m-5p	UGUGUGCAUGUGCAUGUGUGUGU	23
pmc-miR-201-3p	UGAACAGCGCCUUUCUCACGUG	22	pcn-miR-297-3p	AUGUAUGUGUGCAUGUAUGGAGC	23	mcr-miR-2304-1-3p	GGUUGUGUGGUUGUGUGUG	19	lny-miR-466q-1-5p	GUGCACACACACACAUACAC	20
pmc-miR-2017-3p	UACCUGUGCACCUGUGCACGUG	22	pcn-miR-29a-3p	UAGCACCAUUUGAAAUCAGUUU	22	mcr-miR-2304-2-5p	GGUUGUGUGGUUGUGUGUGG	20	lny-miR-466q-2-5p	GUGCACACACACACAUACAU	20
pmc-miR-204-3p	UGCAAGGACAGCAAAGGGA	19	pcn-miR-29b-3p	UAGCACCAUUUGAAAUCAGUGCU	23	mcr-miR-2325c-5p	GGUUGUUUUUUUUCUUUUGC	20	lny-miR-4690-3p	GAGCAGGCGAGGCUGGGAGGGC	22

pmc-miR-2064-3p	AAGCAGCACUGUGCAAGAUACU	22	pcn-miR-2a-1-3p	AAUCACAGCCUGCUUUGGUGAGCU	24	mcr-miR-234-3p	AUUAUUGCUCGAGAAUGACGGC	22	lny-miR-4931-3p	UCGCUGAUUGGUUGAGCAGC	20
pmc-miR-2155-3p	GACACUGUUUUACACUCUACUGG	23	pcn-miR-2a-2-3p	UAUCACAGCCUGCUUGGAUCAGUA	24	mcr-miR-2390-3p	GUUGUGUGUUUUUGUUGAAA	20	lny-miR-4968-3p	CAGCAACAGCAGCAGCAGCAG	21
pmc-miR-216a-3p	CAAGUUACUAGCCGAGAUUACA	22	pcn-miR-2a-3-3p	UAUCACAGCCAGCUUUGAUGAGCU	24	mcr-miR-2-3p	UUCAAAAGACUGUGUUGCUAUG	22	lny-miR-5100-5p	UCGAAUCCCAGCGGUGCCCGA	21
pmc-miR-216a-5p	UAAUCUCAGCUGGUAAUUCUGAG	23	pcn-miR-2a-4-3p	UAUCACAGCCAGCUUUGAUGAGCU	24	mcr-miR-242-3p	UUGCGUAGGCACUUUGCAUGAC	22	lny-miR-51-5p	CAUGGAAGCAGGUACAGUCACC	22
pmc-miR-216b-1-3p	UAAUAUCAGCUGGUAAUCCUGAG	23	pcn-miR-2b-1-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	mcr-miR-2434-1-5p	UUGUUGGGGUUUUUUUUGAA	20	lny-miR-5397-5p	UACGCAGAAGUUUGCUUCUGUA	22
pmc-miR-216b-2-3p	CACAGUGGCUACUGGGUCCACA	22	pcn-miR-2b-2-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	mcr-miR-2434-2-3p	UUGUUGGGGUUUUUUUAAUC	20	lny-miR-5600-3p	AGUGGAAUGUUUUGUUUCAGUA	22
pmc-miR-216b-3-3p	CACACUUACCUGUAGACUGUUA	22	pcn-miR-2e-3p	UACCAACUUUGACUGAUGAGA	21	mcr-miR-2444-3p	UUUGUGUUGUUUUUUGUUUU	20	lny-miR-564-5p	AGGCACGGUGGCAGCAGAA	19
pmc-miR-216b-3-5p	ACAGUUACCUGUAGAUUGUGU	21	pcn-miR-2f-3p	UAUCACAGCCAUGCUAAUCUCCU	23	mcr-miR-2470-3p	UUGUGUGGUUUUGGAUACUUG	21	lny-miR-5920-3p	GUUUUGUCAUUCGUAUACAAAACA	24
pmc-miR-2223-3p	UUUGUACUCAUGUAAGUAUUU	21	pcn-miR-3027-5p	CCAGUCUUGCAUUUAUGAAACA	22	mcr-miR-247-3p	UAGAGAAAAGUUUCUACCGACA	22	lny-miR-6504-5p	CAUUACAGCACAGCCAUGUU	20
pmc-miR-2238i-3p	CCGUUCACUCAUCUCGGAGCUGA	23	pcn-miR-3050-5p	UGAGAUCUUGAUAAACAUCAAC	22	mcr-miR-2491-5p	CAACAACAGCAGCAGCAA	18	lny-miR-653-3p	UUGAGACAAUCUCUACUGCAAC	22
pmc-miR-2284c-3p	AAAAUCUAAGUGAACUGUCAAG	22	pcn-miR-3057-3p	UCCCACAGGCCCAGCUGUCACA	22	mcr-miR-250-3p	AAUCACAGUCACCUGAGUAA	20	lny-miR-6544-5p	AAGUUGUAUUUCUUUUCGCAAACG	24
pmc-miR-2298-3p	GACUGAUGUUGUUGUUCAAG	20	pcn-miR-3071-5p	AUCAUCAAAACAAAUGUGCAUA	22	mcr-miR-252-3p	ACCUGCUCCCUGCUGCUGCCUU	22	lny-miR-6632-3p	AGAGAUGAGAGGAGUGCUGAACCAU	25
pmc-miR-2304-3p	GGUUGUGUGGUUGUGUGGGC	20	pcn-miR-3084a-5p	UUGAAGGUUCAUUAGCCUUGUG	22	mcr-miR-252a-5p	CUAAGUACUAGUGCCGCGGGAA	22	lny-miR-669k-3p	UGUGCAUGUCUGUUUACUUGUGUGU	25
pmc-miR-2361-3p	UUUGUGUUGUUUUGUUUUGC	20	pcn-miR-308-5p	CGCAGGAUUUGCUUGUUUUGAG	22	mcr-miR-2582a-5p	UUCACAGUGUUCAUAAUAUUUCAG	24	lny-miR-67-3p	UCACAACCUGCAUGAAUGAGGAC	23
pmc-miR-2371-5p	UAUAGUUUUGUGUGGUGUAU	20	pcn-miR-31-3p	AGCUGUGUCGCAUGGUGCCACC	22	mcr-miR-2699-5p	GUUUGUGAUAUUCGGCAUUCUU	22	lny-miR-6756-3p	AGGGUGGGGCUGGAGGUGGGGAU	23
pmc-miR-2470-3p	UUGUGUGGUUUUGGAUACUUG	21	pcn-miR-315-5p	UUUUGAUUGUUGCUCAGAAAGCC	23	mcr-miR-277-1-3p	GUAAAUGCAUUUUCUGCUGCA	21	lny-miR-6868-5p	UUCCUUCUGUUGUCUGUAAAC	21
pmc-miR-2491-1-3p	CAACAACAGCAGCAGCAA	18	pcn-miR-31-5p	AGGCAAGAUGUUGGCAUAGCUGA	23	mcr-miR-277-2-3p	UAAAUGCAUUAUUUGGGGAGGUA	23	lny-miR-7006-5p	UGGGGGUGUUCAAGACCCCCCAGUU	25
pmc-miR-2491-2-3p	CAACAACAGCAGCAGCAA	18	pcn-miR-317-1-3p	UGAACACAGCUGGUGGUAUCUUUUU	25	mcr-miR-279-3p	UGACUAGAUCCACACUCAUCCA	22	lny-miR-7045-5p	UCUCCCCCCUCCCCCGCCCCA	21
pmc-miR-2491-3-3p	CAACAACAGCAGCAGCAA	18	pcn-miR-317-2-5p	AGUGAACACAGCUGGUCACAAC	22	mcr-miR-281-3p	UGUCAUGGAGUUGCUCUCUUUA	22	lny-miR-7062-3p	UGGAGGCCAGCUUGUGUCCAGGUU	24
pmc-miR-2493-3p	GCUCACACACACACACAGCCA	21	pcn-miR-3173-3p	GAAGGAGGAAACAGGCUACAGAU	23	mcr-miR-281-5p	AAGGGAGCAUCCGUCGACAGU	21	lny-miR-7195-5p	UAUCCUCCCCUCUCCCCCCCCCU	23
pmc-miR-2505-3p	UCGUGCACACACACACAUACAC	22	pcn-miR-33-3p	GCAAUGCAUCUGCAGUGCAAAUA	23	mcr-miR-283-3p	AAAUAUCAGCUGGUAAACGAAU	22	lny-miR-7290-5p	UCUGGCUCUGUUUGCAGCAA	20
pmc-miR-252c-5p	CUAAGUACUAGUGCCGCGGGAA	22	pcn-miR-33-5p	GUGCAUUGAGGUUGCAUUGCAU	22	mcr-miR-285-3p	ACUGGCUUCCAAAGGUCUCGAAA	23	lny-miR-734-5p	GAACUAUUCUGCAACAUUCAUG	22

pmc-miR-254-5p	UGCAAAUCUUUUGCAACAGACUU	23	pcn-miR-340-5p	UUAUAAAGCAAUGAGAGUUUCC	22	mcr-miR-2970-1-3p	UCAGAUCAUCUCUUGAUCAAC	21	lny-miR-7371d-3p	UAUUUGUACCAGAUGCUCUCUCUGG	25
pmc-miR-2571-3p	GUGAGUGGUGAUGAUGUUUACCACU	25	pcn-miR-345-3p	UGCUGACCCCUAGUCCGCCUC	21	mcr-miR-2970-2-3p	GACAGUCAGCAGUUGGCUCCCC	22	lny-miR-7398y-3p	UAACCUUCCCUUCUCUCUUUCU	22
pmc-miR-279-3p	UGACUAGAUCCACACUCAUCCA	22	pcn-miR-3529-3p	AACAACAAAAUCACUAGUCUUCCA	24	mcr-miR-297b-5p	AUGUAUGUGUGCAUGAACAUGC	22	lny-miR-745a-3p	AGCUGCCUGAUGAAGAGCUGUCC	23
pmc-miR-279b-5p	UGACUAGACUGAUAACCCCUAG	22	pcn-miR-3547-3p	GUGGGAAGAGGGGUGGAGGUGAAGC	25	mcr-miR-29b-5p	UAGCACCAUUAGAAAUGUUAGU	22	lny-miR-7460-3p	CCUGACUGAGCUCUGCUAGGGC	22
pmc-miR-279c-3p	CGGGUGUUCGUUUCGAGUACGA	22	pcn-miR-355-5p	UUUGUUUUAGCCUGAGUCGCC	21	mcr-miR-2a-1-3p	UAUCACAGCCUGCUUGGAUCAGUA	24	lny-miR-750-3p	CCAGAUCUAACUCUUCCAGCUCA	23
pmc-miR-281-3p	UGUCAUGGAGUUGCUCUCUUUA	22	pcn-miR-3609-5p	CAAAGUGAUGAGUAAUUAAGGCGC	24	mcr-miR-2a-2-3p	UAUCACAGCCAGCUUUGAUGAGCU	24	lny-miR-750-5p	AGUUGGAAGAUUGGGUCUUUGGC	23
pmc-miR-281-5p	AAGGGAGCAUCCGUCGACAGU	21	pcn-miR-3680-5p	GACUCACUCACAGGAUGUGGUG	22	mcr-miR-2a-3-3p	AAUCACAGCCUGCUUUGGUGAGCU	24	lny-miR-7661-3p	AAGAAAGAAACCUGGAUAAACUAGA	25
pmc-miR-282-3p	ACAUAGCCUAAAAGAGGCACCA	22	pcn-miR-36b-3p	UCACCGGGUAUACAUUCAUCCGC	23	mcr-miR-2b-1-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	lny-miR-7965-3p	UUUUGUAUGCUAAAACAUAAAAU	23
pmc-miR-2953-3p	UACAGAAGUGUUCGUGGGGGA	21	pcn-miR-3716b-3p	CGAUGUUAUUUGAAAUAUCAAAC	23	mcr-miR-2b-2-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	lny-miR-7c-5p	UGGAAGACUAGUGAUUUUGUUGUUC	25
pmc-miR-2c-1-3p	UAUCACAGCCUGCUUGGAUCAGUA	24	pcn-miR-3739-3p	UUGGGAGGGGGAGAGAGUGGAA	22	mcr-miR-2c-5p	UCGUCAACAUUGCCUGCUGCUU	22	lny-miR-8066-3p	CAAUGUGAUCUUUUGGAGAAC	21
pmc-miR-2c-2-3p	UAUCACAGCCAGCUUUGAUGAGCU	24	pcn-miR-375-1-3p	UUUGUUCGUUCGGCUCGCGUUA	22	mcr-miR-3131-3p	UCGAGGACUGGUGGAAUUUUCCA	23	lny-miR-8075-5p	UGCUGAUGGCAGAUGUGUGUUGGC	24
pmc-miR-2c-3-3p	AAUCACAGCCUGCUUUGGUGAGCU	24	pcn-miR-375-2-3p	CUUGUUCGUUCGGCUCGUUC	20	mcr-miR-315b-3p	UUUUGAUUGUUGUACAGUCCGC	22	lny-miR-8279-5p	AGAUUCAUGUCAAUCUGACAUU	22
pmc-miR-2b-1-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	pcn-miR-377-5p	AGAGGUUGCCUUUGGUGAGAGA	22	mcr-miR-317-3p	UGAACACAGCUGGUGGUAUCUUUUU	25	lny-miR-8335-1-5p	GUUGUUGUUGUUGUUUUGGUCCUUG	25
pmc-miR-2b-2-5p	UCAUCAAAGCUGGCUGUGAUAUGA	24	pcn-miR-3776-3p	AGGAGGGGGGGAGAGAGAUUUA	22	mcr-miR-3223-5p	UACCUUUUACCUUUACCUUACA	22	lny-miR-8335-2-3p	GUUGUUGUUGUUGUUUUGUUUUGUG	25
pmc-miR-2f-3p	UAUCACAGCCAUGCUAAUCUCCU	23	pcn-miR-3809-5p	GUCGGUCAGUUGGUUAGAUG	20	mcr-miR-324-3p	CCCACUGCCCCAGGUGUCUUGUG	23	lny-miR-8335-3-3p	GUUGUUGUUGUUGUUUUGAAGCCAG	25
pmc-miR-300-3p	UAUACAAGGGCAGACUCCAUCC	22	pcn-miR-3831-3p	CAAACAUUGCUAUUUUUCAUAUCCAA	26	mcr-miR-3299-3p	UCAUUGUUAAAGUCUGCAUAACC	23	lny-miR-8-3p	UAAUACUGUCAGGUAAAGAUGUC	23
pmc-miR-302a-3p	AAGUGCUUCCUUUUAUUGGUGAU	23	pcn-miR-3928-3p	UGAAGCUCUAAGGUUCAUCCCAG	23	mcr-miR-33-3p	GCAAUGCAUCUGCAGUGCAAAUA	23	lny-miR-8485-1-5p	CACACACACACACACACGUAC	21
pmc-miR-302b-3p	AAAGUGCUUCCAUGUUUAAUUCA	23	pcn-miR-3957-3p	ACGCACAGCACCUCACACUGAC	22	mcr-miR-33-5p	GUGCAUUGAGGUUGCAUUGCAU	22	lny-miR-8485-2-3p	CACACACACACACACACGUAC	21
pmc-miR-3057-3p	UCCCACAGGCCCAGCUGUCACA	22	pcn-miR-3965-5p	UGCUUAUCAGCCUGAUUCU	19	mcr-miR-33b-3p	CAAUGUACCUGCAGUGCUUUC	21	lny-miR-8-5p	CAUCUUACCUAACAGCAUUAGA	22
pmc-miR-3071-5p	AUCAUCAAAACAAAUGUGCAUA	22	pcn-miR-4057-3p	GGCUUGGCGACUGUAGUAGG	20	mcr-miR-34-3p	CAACCACUAUCUGCAGUGCCGCG	23	lny-miR-87a-5p	AACACUUGGUAAUUUGCAAAUGAA	24
pmc-miR-3084a-5p	UUGAAGGUUCAUUAGCCUUGUG	22	pcn-miR-4066-5p	CAUGCCUUUAUCAUGUGUU	19	mcr-miR-34-5p	UGGCAGUGUGGUUAGCUGGUUGUG	24	lny-miR-8908i-3p	UACCCGGUAAGGUAGUUGCCAAGAA	25
pmc-miR-308-5p	CGCAGGAUUUGCUUGUUUUGAG	22	pcn-miR-4070-3p	CACGCGUCACGUGACACCU	19	mcr-miR-3529-3p	AACAACAAAAUCACUAGUCUUCCA	24	lny-miR-9032-3p	UGCAGAUUCCGUUGAUGCAAUCCAA	25

pmc-miR-31-3p	UGGCAAGAUGUUGGCACAGCG	21	pcn-miR-4185-3p	UGUAUUCAUACUGUCUGUUACA	22	mcr-miR-3530-5p	CAAUGGUGUGAGCUGGACAUG	21	lny-miR-9-1-3p	UCAUACAGCUAGAUAACCAAAGA	23
pmc-miR-315-3p	UUUUGAUUGUUGCUCUAGAAAUU	23	pcn-miR-4187-3p	UUUGGUGUUGUGCUGUUUAA	20	mcr-miR-3671-3p	AUCAAAUAAGGACUAGUGGCA	21	lny-miR-9-1-5p	CCUUUGGUAACCUAGCUUUAUGA	23
pmc-miR-317-3p	UGAACACAGCUGGUGGUAUCUUUUU	25	pcn-miR-4195-5p	UACAUGUAAUACAUUGUACC	20	mcr-miR-371b-3p	ACUCAAAAGAUGGCGGCAAGG	21	lny-miR-9229d-3p	UGAGUCUUCCCCUUGAG	17
pmc-miR-33-3p	GCAAUGCAUCUGCAGUGCAAAUA	23	pcn-miR-4271-3p	GGGGGAAGAAAAGGUGGCA	19	mcr-miR-372-5p	AAAGUGCUGCGACAUUAUUACCG	23	lny-miR-9235b-3p	UAAUUUUGAUGUGGCUUGAGAACA	24
pmc-miR-33-5p	GUGCAUUGAGGUUGCAUUGCAU	22	pcn-miR-4323-3p	CAGCCCCACAGCCUCAGG	18	mcr-miR-373-3p	AAAGUGCUUCCCUUUUAUUAUC	22	lny-miR-9-2-3p	AUAAAGCUAGGUUACCAAAGGCA	23
pmc-miR-336-5p	UCACCCUUCCAUAUCUUUUGG	21	pcn-miR-4472-3p	GGUGGGGGGUGUUGUUGA	18	mcr-miR-375-1-3p	UUUGUUCGUUCGGCUCGCGUUA	22	lny-miR-9-2-5p	UCUUUGGUUAUCUAGCUGUAUGAUU	25
pmc-miR-33b-3p	CAGUGCCUCUGCAGUGGAUGU	21	pcn-miR-449a-3p	ACCAGCUGACAUUCAGUCCAGA	22	mcr-miR-375-2-5p	UUUGUUCGUUUUGCUUGCUCAUG	23	lny-miR-92a-3p	GAUUGCACUCGUCCCGGCCUUC	22
pmc-miR-345-3p	UGCUGACCCCUAGUCCGCCUC	21	pcn-miR-449d-3p	GAAGGCUGUGUGCUGUAAAG	20	mcr-miR-376c-3p	AACAUAGAGGAAAAUCACCGA	21	lny-miR-92b-1-3p	UAUUGCACUCGUCCCGGCCUA	21
pmc-miR-3529-3p	AACAACAAAAUCACUAGUCUUCCA	24	pcn-miR-4525-3p	GGGGGGAUGUGCAUGCCAG	19	mcr-miR-376c-5p	AACAUAGAGGAAAAUCACCGA	21	lny-miR-92b-2-3p	AAUUGCACUUGUCCCGGCCUGC	22
pmc-miR-3532-3p	UUGGAGGCUGCAGUGUGACCUC	22	pcn-miR-4617-3p	UGUGGCUGUCACACCUAAACAC	22	mcr-miR-376d-5p	GUAGAUUUUCCUUCUACAACUU	22	lny-miR-9319-3p	AGCAGAGCUAGUGGCCUCACCCC	23
pmc-miR-355-5p	UUUGUUUUAGCCUGAGUCGCC	21	pcn-miR-4654-5p	UGUGGGAUCUGGAGGCAGACUUC	23	mcr-miR-3834-3p	UGAUGUUAUUUUCGACAAAU	20	lny-miR-96b-3p	AUUAUACAUCGGUGCCAAGUCA	22
pmc-miR-3620-5p	GUGGGCUGGGCUGGGCUGGGCU	22	pcn-miR-466i-1-3p	UGUGUGUGUGUGUGUGUG	20	mcr-miR-3890-3p	AGCCGGUUAUGUACGGCUG	19	lny-miR-96b-5p	AUUUGGCACUUGUGGAAUAAUCG	23
pmc-miR-36-3p	UCACCGGGUGGAAAUUCGUCUUU	23	pcn-miR-466i-2-5p	UGUGUGUGUGUGUGUGUG	20	mcr-miR-39-3p	AGCUGAUUUCGUCUUGGGAGUU	22	lny-miR-9701a-1-5p	CUGUGUACCACCAUCAACCCGCG	23
pmc-miR-3643-3p	GUGAGUUUAUCUAAAUUCAGGA	22	pcn-miR-466i-3-5p	UGUGUGUGUGUGUGUGUG	20	mcr-miR-4000i-3p	UGAAACUUGCAAGGAACUGC	20	lny-miR-9701a-2-3p	CUGUGUACCACCAUCAAGGCGUG	23
pmc-miR-36b-3p	UCACCGGGUAUACAUUCAUCCGC	23	pcn-miR-466i-4-5p	GGUGUGUGUGUGUGUGUG	20	mcr-miR-4003d-3p	UGAGAAUGGUAACCAAAAAA	20	lny-miR-9708c-3p	UGAGCACAGCUAAACAUUUUCU	22
pmc-miR-3724-3p	AUCGUGGUAAACUUUGUCCUCG	22	pcn-miR-466m-3p	UGUGUGCAUGUGCAUGUGUGCUU	23	mcr-miR-4027-3p	UAUAUUACUUUUAUGUUAAU	20	lny-miR-981-3p	UUCGUUGUCGUCGAAACCUGCCC	23
pmc-miR-375-1-3p	UUUGUUCGUUCGGCUCGCGUUA	22	pcn-miR-466n-3p	GUGUGUGCGUACAUGUACUCAU	22	mcr-miR-4029-5p	GUUUACAUUGCAUGCAUCGC	20	lny-miR-981-5p	CGGGUUUCGCGACUGGCGAACA	22
pmc-miR-375-2-3p	CUUGUUCGUUCGGCUCGUUC	20	pcn-miR-466p-3p	UAUGUGUGUGUACAUGUGAAG	21	mcr-miR-4034-3p	CUUUUCUCUGGCACUGAGGA	20	lny-miR-9a-3p	AUAAAGCUAGGCGACUGUCCGU	22
pmc-miR-3782-3p	CUUACAGAGGCACUUGGGUGGCGC	24	pcn-miR-466q-3p	GUGCACACACACACAUACG	19	mcr-miR-4040-3p	CAACCAGAUCAGAAAGGUGA	20			
pmc-miR-3792-3p	GACUGAGGAAAGAGAGAUGUU	21	pcn-miR-467f-3p	AUAUACACACACACACCUUUC	21	mcr-miR-4048-5p	GCACCUUGGUCACCAUAGUGAC	22			
pmc-miR-3821-3p	AUUUCUAGUGAGUGAGGGUACUU	23	pcn-miR-4715-5p	GUGCCACCUUAACUGCACGGACC	23	mcr-miR-4127-5p	AGGCAGCUGUGAGAGAGCCCU	21			
pmc-miR-4009c-3p	UAUUGCACUUUUACUGGUCG	20	pcn-miR-4757-3p	CAUGACGUCACAGAGGCGAGGU	22	mcr-miR-4130-5p	CAGACAGCUGCGUGUCUUUA	20			

pmc-miR-4013b-5p	UUACUUGCUUUAACAGCUUA	20	pcn-miR-4801-3p	UACACAAGAAAACCAAA	17	mcr-miR-41-5p	UCACCGGGUGAAAAACUGAC	20			
pmc-miR-4057-3p	GGCUUGGCGACUGUAGUAGG	20	pcn-miR-4860-3p	UGUAGAGAUUGUGUGAACUGCUAG	24	mcr-miR-427-3p	GAAAGUGCUUUCUGUUACUUCUU	23			
pmc-miR-4070-3p	CACGCGUCACGUGACACCU	19	pcn-miR-4918-5p	GUGAGCACGAUGGCCAGCACUGCC	24	mcr-miR-44-1-3p	CUGGAUGUGCUCGUUAACGUC	21			
pmc-miR-4133-5p	CAUGUUGUGUUUUUGUAUG	19	pcn-miR-49-3p	AAGCACCAUGUGAAGCACUUUU	22	mcr-miR-44-2-3p	UGACUAGAUGAUCACAAGACAU	22			
pmc-miR-4140-3p	ACCAGAUGAUUAUGACACAA	20	pcn-miR-4968-1-5p	CAGCAACAGCAGCAGCAGCAGA	22	mcr-miR-449a-3p	UGGCAGUGCGUGUUAGUGAUG	21			
pmc-miR-4177-5p	UAAUUGCUGUUUUAAACAGU	20	pcn-miR-4968-2-5p	CAGCAACAGCAGCAGCAGCAGC	22	mcr-miR-4502-5p	GCUGAUGAUGAUGGUGAUUGGA	22			
pmc-miR-4185-3p	UGUAUUCAUACUGUCUGUUACA	22	pcn-miR-4968-3-3p	CAGCAACAGCAGCAGCAGCAGC	22	mcr-miR-452-3p	AACUGUUUGCAGAGGACAGUCACA	24			
pmc-miR-4323-3p	CAGCCCCACAGCCUCAGG	18	pcn-miR-4968-4-3p	CAGCAACAGCAGCAGCAGCAGC	22	mcr-miR-4552-3p	UCCCACUUGAUCACUGACAUUCU	23			
pmc-miR-4472-3p	GGUGGGGGGUGUUGUUGA	18	pcn-miR-5317a-3p	UUUCUUGUCUCUGUGUACCUCUUG	24	mcr-miR-466h-3p	UACGCACGCACACACACAC	19			
pmc-miR-449d-3p	GAAGGCUGUGUGCUGUAAAG	20	pcn-miR-5391-5p	UGGACAGUUAUGAAGAAGUG	20	mcr-miR-466i-1-5p	AUACACACACACAUACACACGC	22			
pmc-miR-4629-3p	UGACUGGACGUUCAUCGCCG	20	pcn-miR-5392-3p	UGUUACUUGACAUUUUGAGA	20	mcr-miR-466i-2-5p	UGUGUGUGUGUGUGUGUG	20			
pmc-miR-4654-5p	UGUGGGAUCUGGAGGCAGACUUC	23	pcn-miR-5440-5p	UCAGGAGGACUGAAGGGUGGA	21	mcr-miR-467c-5p	UAAGUGCGUGCAUGUAUGCAGG	22			
pmc-miR-466-1-3p	AUAUAUACACACACACAUAUAUA	23	pcn-miR-5549-3p	UCAUGUUGGUUUUUUGAAU	19	mcr-miR-4760-3p	UUUAGAUUGAACAUGAAUGUUU	22			
pmc-miR-466-2-5p	AUAUAUACACACACACAACCAUG	23	pcn-miR-559-3p	UAAAGUAAAUAAGCACAGACU	21	mcr-miR-4810b-5p	GUAGGUUCAUGAGUAGACUUUU	22			
pmc-miR-466-3-3p	AUAUAUACACACACACAUAUACA	23	pcn-miR-5594-3p	AAGAGUACUGUAGUUUUUCCAA	22	mcr-miR-4865-3p	AUGUAGAGAGAGUGACGCCGCUU	23			
pmc-miR-466h-3p	UACGCACGCACACACACAC	19	pcn-miR-5595-3p	UCUCUUUUUUCUCGCAGUCCCGCA	24	mcr-miR-4968-1-3p	CAGCAACAGCAGCAGCAGCAGC	22			
pmc-miR-466i-1-5p	UGUGUGUGUGUGUGUGUG	20	pcn-miR-574-1-5p	UGAGUGUGUGUGUGUGAGUGUGUG	24	mcr-miR-4968-2-3p	CAGCAACAGCAGCAGCAGCAGC	22			
pmc-miR-466i-2-5p	UGUGUGUGUGUGUGUGUG	20	pcn-miR-574-2-3p	UGAGUGUGUGUGUGUGAGUGUGUG	24	mcr-miR-4968-3-5p	CAGCAACAACAGCAGCAGCAGC	22			
pmc-miR-466m-5p	UGUGUGCAUGUGCAUGUGAAUGU	23	pcn-miR-5965-5p	GACAAUUCUGACAUUUUGCA	20	mcr-miR-4975-3p	UUUACUUGGGUUUUCUUUCAG	21			
pmc-miR-466n-3p	GUGUGUGCGUACAUGUACUCAU	22	pcn-miR-6013-3p	GCUGCUGACGACCGCCUCA	19	mcr-miR-5009-3p	UUGGACUUUUUCAGAUGGACUAGU	24			
pmc-miR-466q-3p	GUGCACACACACACAUACAC	20	pcn-miR-6037-1-3p	UAAGCUCUGUGUACUUUAGUG	21	mcr-miR-50-5p	CGAGUAAUAUUAGACAGCGGAUU	23			
pmc-miR-467d-5p	UAAGUGCGCGCAUGUAUUUAUG	22	pcn-miR-6037-2-3p	UAAGCUCUGUGUACUUUAGUG	21	mcr-miR-5101-5p	UUUGUUUGUUUUGCUGUAUGAG	22			

pmc-miR-467f-3p	AUAUACACACACACACCUUUC	21	pcn-miR-6076-3p	AGCAUGACAGAGGAGAGGAGA	21	mcr-miR-542-3p	UGUGACAGAUUGAUAAAGACGC	22			
pmc-miR-4715-5p	GUGCCACCUUAACUGCACGGACC	23	pcn-miR-6098-5p	AUAGUGAUUGUGUGUUUCUA	20	mcr-miR-5429-3p	AUAGUCAUGCUGGAGGUUCCUA	22			
pmc-miR-4750-3p	CCUGACCCACCCCCUCCCUACU	22	pcn-miR-6132-1-5p	AGCAGGGCUGGGGAUUCUG	19	mcr-miR-548d-3p	AAAAAACUGCAGUUACUUGAA	21			
pmc-miR-4757-3p	CAUGACGUCACAGAGGCGAGGU	22	pcn-miR-6132-2-5p	AGCAGGGCUGGGGAUUCUG	19	mcr-miR-550-5p	AGUGCCUGAGGGAGUAGUGCCUU	23			
pmc-miR-4769-5p	UCUGCCAUCCUCCCUCUCGUCU	22	pcn-miR-6132-3-5p	AGCAGGGCUGGGGAUUCUG	19	mcr-miR-5552-3p	UGUAGUUUGUAGUCUAUAGAUAU	23			
pmc-miR-487c-5p	GUGGCUAUCCCUGCUGUCAUCC	22	pcn-miR-6335-5p	CUGCAUACACAGUGAUGCAGGU	22	mcr-miR-5582-5p	UAAAACUUUAAGUGUGUCAGAC	22			
pmc-miR-4938-5p	AUCAUCAUGCAUCAUCAUUAUUG	23	pcn-miR-6-3p	AGGGAACUGCUGCUGCAGCGACU	23	mcr-miR-5698-3p	UGGGGGAGUGCAGUGAA	17			
pmc-miR-49-3p	AAGCACCAUGUGAAGCACUUUU	22	pcn-miR-6416-3p	CUCCGUAUCAUCUGCUAGCUA	21	mcr-miR-586-5p	UAUGCAUAUUGUAUUUUGUACUUG	24			
pmc-miR-4949-3p	AGUGGUGUGCGUGUGUGUGUGU	22	pcn-miR-64c-3p	CAUGACACACUGGCUGAACCA	21	mcr-miR-5890c-3p	CAUAUAACUGAACUGUCUGUG	21			
pmc-miR-4968-1-3p	CAGCAACAGCAGCAGCAGCAGC	22	pcn-miR-6505-5p	UGACUUCUACCUGUUCCCUAA	21	mcr-miR-592-3p	UCAUCACGUGGUGACGUGUUC	21			
pmc-miR-4968-2-5p	CAGCAACAGCAGCAGCAGCAGC	22	pcn-miR-6516-3p	AUGCAGUAACAGGUGUCUCCAC	22	mcr-miR-6013-5p	GCUGCUGACGACCGCCUCA	19			
pmc-miR-4968-3-3p	CAGCAACAGCAGCAGCAGCAAU	22	pcn-miR-6560-5p	UGUGUUGCUCCAUACUGCUGAAG	23	mcr-miR-6058-5p	AUGACAAUGGUGAUGAAUCAG	21			
pmc-miR-4968-4-3p	CAGCAACAGCAGCAGCAGCAGC	22	pcn-miR-6575-3p	GAGUUUUCCCAGGCUGAUGUGU	22	mcr-miR-6340-3p	GUCAGCAGCAGCUUCGAUACCCA	23			
pmc-miR-4999-3p	UGCUGUAUUGUCAGGUUUGUA	21	pcn-miR-6587-3p	UGCUGGAAGUGGUGCUAUAUGUC	23	mcr-miR-6552-3p	CAACAAGAAGUGGACACGGUC	21			
pmc-miR-501-5p	AAUGCACCCGGGCAAGUCAGA	21	pcn-miR-6603-5p	AAGGACAAGAGGAAAUAACUUGA	23	mcr-miR-6560-5p	UGUGUUGCUCCAUACUGCUGAAG	23			
pmc-miR-5101-1-5p	UUUGUUUGUUUUGCUGUCUGAC	22	pcn-miR-6680-5p	GUUGCUGAUGAAGAUAAAUCGU	22	mcr-miR-669f-3p	UAUAUACACACACACACACAUAA	23			
pmc-miR-5101-2-5p	UUUGUUUGUUUUGCUGUGUGCU	22	pcn-miR-669f-5p	CAUAUACAUACACACACACGUGU	23	mcr-miR-67-3p	UCACAACCUGCAUGAAUGAGGAC	23			
pmc-miR-519f-3p	AAAGUGCAUCCUUUUAUGGCGA	22	pcn-miR-67-3p	UCACAACCUGCAUGAAUGAGGAC	23	mcr-miR-676-5p	CUCUUCAACUUCAGGAUCGUUA	22			
pmc-miR-5317a-3p	UUUCUUGUCUCUGUGUACCUCUCG	24	pcn-miR-6816-3p	GAAGGACCUGCACCUUGC	18	mcr-miR-6769b-5p	UGGUGGGUGAGGAGGAGAAAGCA	23			
pmc-miR-5391-5p	UGGACAGUUAUGAAGAAGUG	20	pcn-miR-6852-5p	UGUCCUCUGUUCCUCAU	17	mcr-miR-6780a-5p	CUCCUCUGUUUUCUUUUCAAA	21			
pmc-miR-539-5p	AUCAUACAAGGACAAUUUGUCA	22	pcn-miR-6894-3p	AGGAGGAUGGAGAGCUGUUACAGC	24	mcr-miR-6796-3p	UUGUGGGGUUGGAGAGUAUGGCU	23			
pmc-miR-5396b-5p	CCACACAACCAGCAAGAUUUGC	22	pcn-miR-6951-5p	UUGUAUUUGUGUGAUUCCCGA	21	mcr-miR-6800-5p	GUAGGUGACAGUCAGGCCUGG	21			

pmc-miR-5549-3p	UCAUGUUGGUUUUUUGAAU	19	pcn-miR-6974-3p	UCUCCACUCUCUUCUGUGUCGU	22	mcr-miR-6803-3p	GUGGGGGUGGGGGGCUGGGAGA	22			
pmc-miR-5552-3p	UGUAGUUUGUAGUCUAAAGUCUG	23	pcn-miR-6977-5p	GUGAGGCGCUGUGGGCAGGUGAGCC	25	mcr-miR-6826-3p	UCAAUAGGAAAGAGGUCUGUGAU	23			
pmc-miR-5595-3p	UCUCUUUUUUCUCGCAGUCCCGCA	24	pcn-miR-6999-3p	AAGGAAGGAGAGUCAGGAGCGUG	23	mcr-miR-6830-3p	UGUCUUUCUUCUCUCCCCCAUCU	23			
pmc-miR-5600-3p	AGUGGAAUGUUUUGUUUCAGUA	22	pcn-miR-7000-5p	CACCCACCUGCCUGUCACAGUG	22	mcr-miR-6864-3p	UUGAAGGGACAAGUCAGACUUUAA	24			
pmc-miR-5612-3p	UAUACAUCACCAGUUGACAUGU	22	pcn-miR-7072-3p	CUAGCUUCUCUUCUUCCAGUU	21	mcr-miR-6873-3p	UUCUCUCUGUCUUUCUCUCAUGU	23			
pmc-miR-574-1-3p	UGAGUGUGUGUGUGAGUGC	21	pcn-miR-71-1-5p	UGAAAGACAUGGGUAGUGAGAUG	23	mcr-miR-6925-3p	AGAGGAAGAAAUGGCUUACAAA	22			
pmc-miR-574-2-3p	UGAGUGUGUGUGUGAGUGUGUG	24	pcn-miR-71-2-3p	UGAAAGACACAGGUAGAAUGUA	22	mcr-miR-6955-3p	ACACCUGUCUCCUUUGUCGUGU	22			
pmc-miR-5918b-5p	UCGCUCUACAUCGUGUGGUCAG	22	pcn-miR-7151-5p	GAUCCAUCUCUGCCUGCUCACCU	23	mcr-miR-6975-3p	UCUCUCCUUUCUCCUCCGCC	20			
pmc-miR-5965-5p	GACAAUUCUGACAUUUUGCA	20	pcn-miR-7307-3p	CAAAUGGUCAUUUCUGCGA	19	mcr-miR-6988-5p	GGGGUGGAGAGCUGAGUUACA	21			
pmc-miR-6056-3p	GAGGGACGAGGAGGGAGGAG	20	pcn-miR-7314-3p	UCAGAAACAGUGUCUCUG	18	mcr-miR-7042-3p	UGUCCCUUUGUUUUCUCCACA	21			
pmc-miR-6076-3p	AGCAUGACAGAGGAGAGGAGA	21	pcn-miR-7386e-5p	GUAAACACUUUACCCGUUUGA	21	mcr-miR-7051-5p	UCACCAGGAGGAGGUGUGCACG	22			
pmc-miR-6098-5p	AUAGUGAUUGUGUGUUUCUA	20	pcn-miR-7389-3p	GCUGUACUUGAAGCAAGAGAGA	22	mcr-miR-71-5p	UGAAAGACAUGGGUAGUGAGAUG	23			
pmc-miR-623-3p	UUCCCUUGCAGGGGCUGAGGUAU	23	pcn-miR-7398h-3p	UAACCCUCCCUUCUUUAUGAUA	22	mcr-miR-7186-3p	CUGUGGUUCCUGUAUGUUCGGG	22			
pmc-miR-626-5p	AGCUGUCUGAAAAUGUUUG	19	pcn-miR-7399-5p	UACGCAUGUAUAUGUAUAUACA	22	mcr-miR-7234-3p	UUGUUUUCUCCAAAGAACACCAU	23			
pmc-miR-6302-3p	CAAACUCAUCGUCAGCCACUCA	22	pcn-miR-743b-3p	UGUUCAGACUGGUGUCUGGU	20	mcr-miR-7241-5p	UAGUAGAUACUCAUGGUGAGUA	22			
pmc-miR-6416-3p	CUCCGUAUCAUCUGCUAGCUA	21	pcn-miR-745a-3p	AGCUGCCUGAUGAAGAGCUGUCC	23	mcr-miR-7260-3p	AUGGCUAAAACAACUGAC	18			
pmc-miR-6505-5p	UGACUUCUACCUGUUCCCUAA	21	pcn-miR-745b-3p	GAGCUGCCAAAUGAAGGGCUGU	22	mcr-miR-7371b-3p	UUUGUAUCAGAUGCUUGC	18			
pmc-miR-651-3p	UUUAGGAUAAGUUUGAACACGU	22	pcn-miR-7472-3p	AUGUGCUGGAGAUGAAAACUCU	22	mcr-miR-7-3p	CAACAAGUCCCAGUCUUGGCGG	22			
pmc-miR-6516-3p	AUGCAGUAACAGGUGUCUCCAC	22	pcn-miR-750-3p	CCAGAUCUAACUCUUCCAGCUCA	23	mcr-miR-742-5p	GAAAGCCACCAUGCUGUUAUAU	22			
pmc-miR-6528-5p	AGAGGGAAGAGGGAAGAAAG	20	pcn-miR-750-5p	AGUUGGAAGAUUGGGUCUUUGGC	23	mcr-miR-7456-3p	CAGGGAGAUGUACCUGCUGCCA	22			
pmc-miR-6548-3p	AGAGGUGCCCCGCUGUCUGUUC	22	pcn-miR-751-3p	CAUGUUUGAAUGGCCACCACU	21	mcr-miR-7461-5p	CAUGGCUGGAAUGUGGCUGAGCAAC	25			
pmc-miR-6560-5p	UGUGUUGCUCCAUACUGCUGAAG	23	pcn-miR-753d-5p	AGAUCAUUAUUCAAGCGAUUG	21	mcr-miR-7462-3p	CAACCUCAUUUCUAGCAGACCA	22			

pmc-miR-6562-5p	AGGGGAAAGGAAUGUGCAGGUA	22	pcn-miR-760-3p	CGGCUCUGGGUCUGUGAAGUCA	22	mcr-miR-7465-3p	UCAGAUAGCUGAUACUCAUU	20			
pmc-miR-6568-3p	GCGGCCGCCAUCUUGCGGAGC	21	pcn-miR-7643-3p	GACAUCUGUUUCUCAGUAAUUU	22	mcr-miR-750-3p	CCAGAUCUAACUCUUCCAGCUCA	23			
pmc-miR-669f-5p	CAUAUACAUACACACACACGUGU	23	pcn-miR-7649-5p	UGACAGCAGUGCUGUAUCUAUG	22	mcr-miR-750-5p	AGUUGGAAGAUUGGGUCUUUGGC	23			
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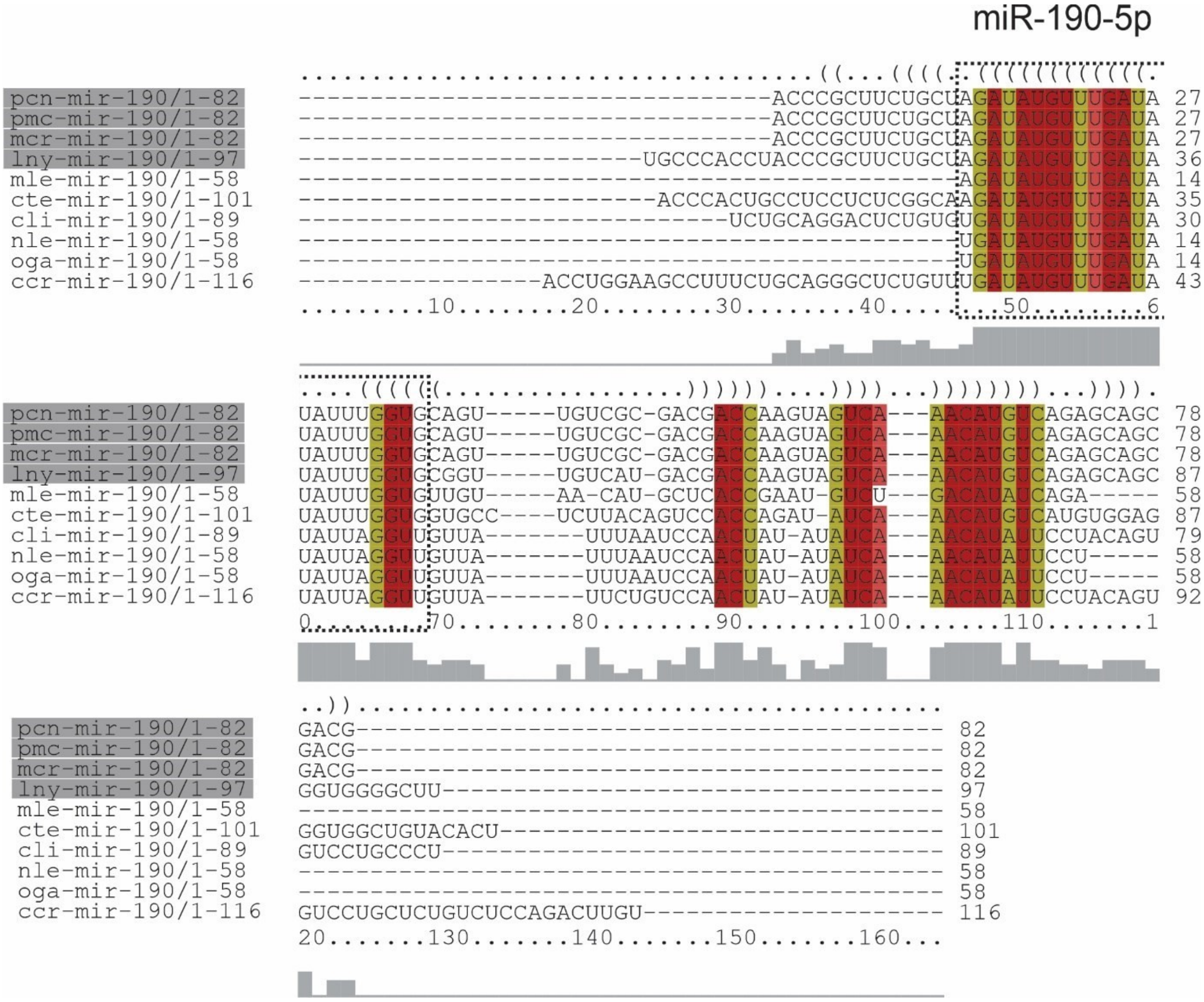
SUPPLEMENTARY DATA III

Bilaterial specific miRNAs

mir-190



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ccr-mir-190/1-116   -----------------ACCUGGAAGCCUUUCUGCAGGGCUCUGUUUGAUAUGUUUGAUA 43
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oga-mir-190/1-58    UAUUAGGUUGUUA-------UUUAAUCCAACUAU-AUAUCA---AACAUAUUCCU----- 58
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mcr-mir-190/1-82    GACG--------------------------------------- 82
lny-mir-190/1-97    GGUGGGGCUU--------------------------------- 97
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ccr-mir-190/1-116   GUCCUGCUCUGUCUCCAGACUUGU------------------- 116
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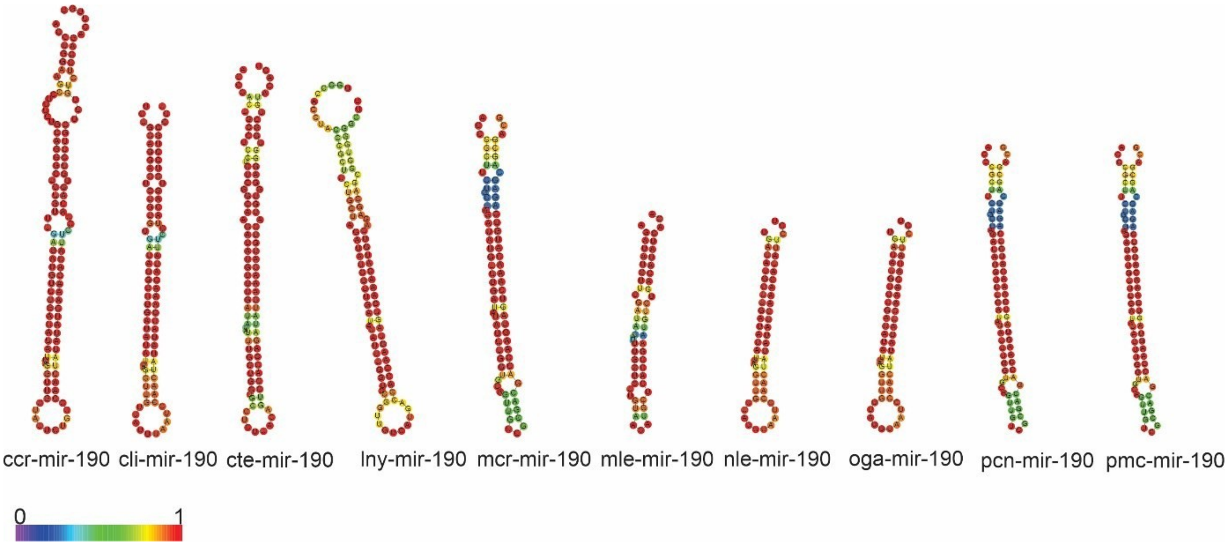
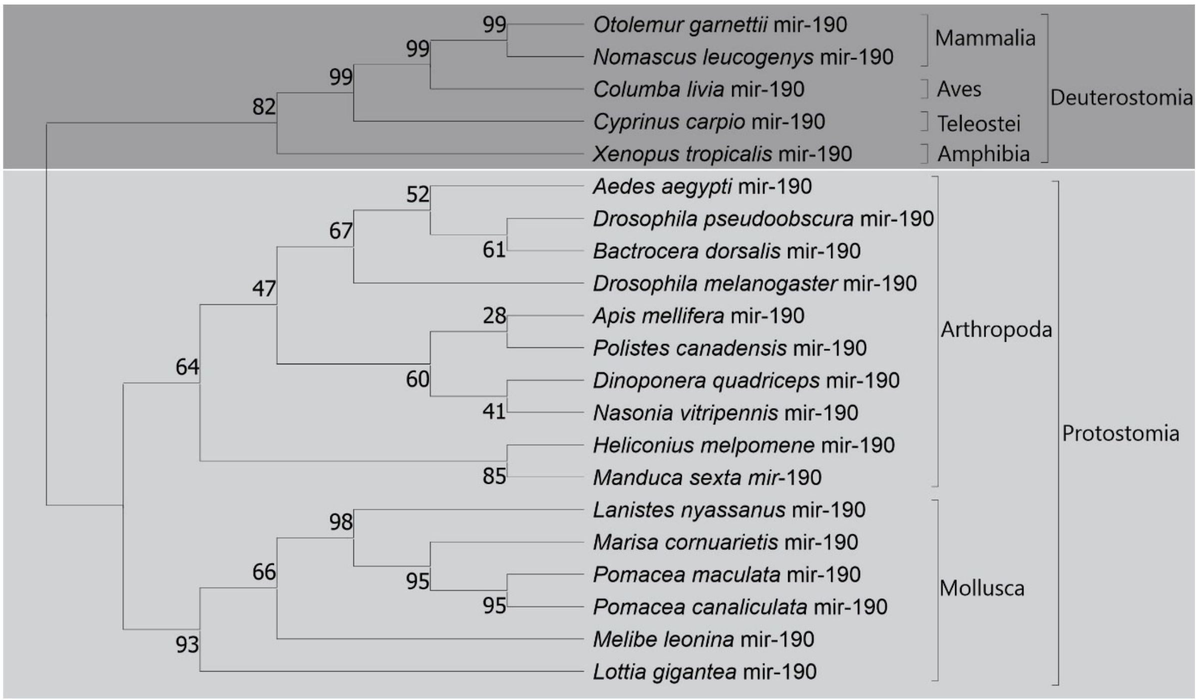
Supplementary figure 1: Alignment of mi-190 in Ampullariidae species and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculala*, mcr - *M. cornuarietis*, lny - *L. nyassanus*, mle – *M. leonina*, cte - C. teleta, cli - *Columba livia*, nle - *Nomascus leucogenys*, oga - *Otolemur garnettii*, ccr - *Cyprinus carpio*.

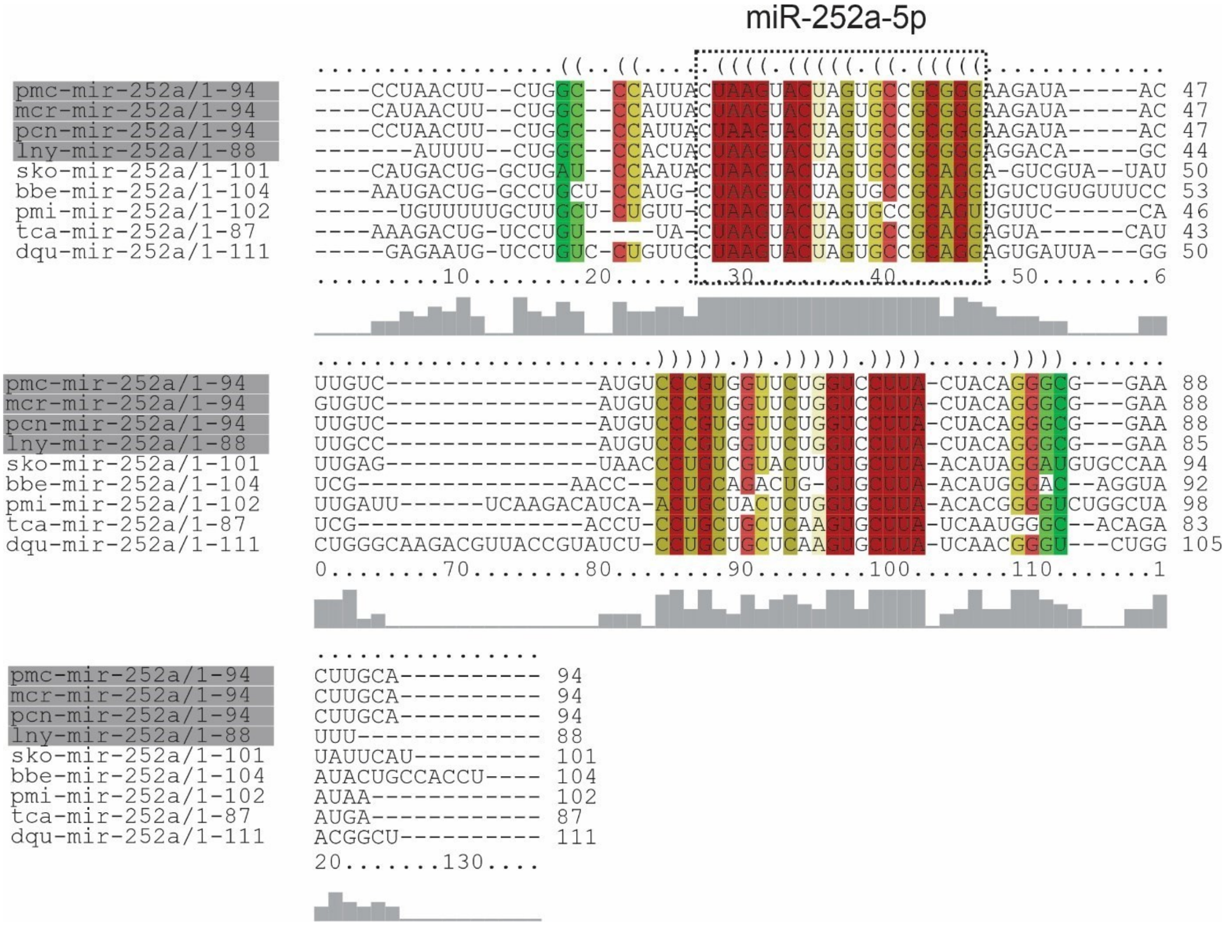


Supplementary figure 2: Secondary structure of mir-190 in Ampullariidae species and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculata*, mcr - *M. cornuaritis*, lny - *L. nyassanus*, mle - *M. leonina*, cte - *C. teleta*, cli - *Columba livia*, nle - *Nomascus leucogenys*, oga - *Otolemur garnettii*, ccr - *Cyprinus carpio*.



Supplementary figure 3: Phylogenetic distribution of mir- in Ampullariidae species and their orthologs.

mir-252a



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bbe-mir-252a/1-104      ----AAUGACUG-GCCUGCU-CCAUG-CUAAGUACUAGUGCCGCAGGUGUCUGUGUUUCC 53
pmi-mir-252a/1-102      ------UGUUUUUGCUUGCU-CUGUU-CUAAGUACUAGUGCCGCAGUUGUUC------CA 46
tca-mir-252a/1-87       ----AAAGACUG-UCCUGU-----UA-CUAAGUACUAGUGCCGCAGGAGUA------CAU 43
dqu-mir-252a/1-111      -----GAGAAUG-UCCUGUC-CUGUUCCUAAGUACUAGUGCCGCAGGAGUGAUUA---GG 50
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lny-mir-252a/1-88       UUGCC---------------AUGUCCCGUGGUUCUGGUCCUUA-CUACAGGGCG---GAA 85
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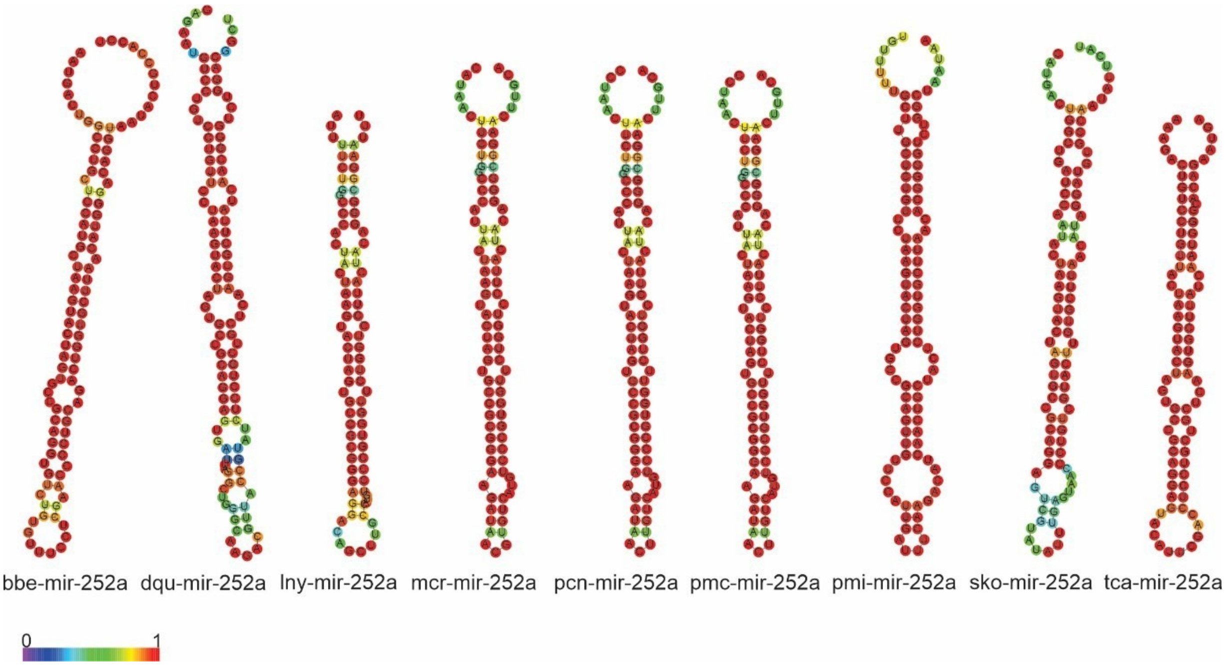
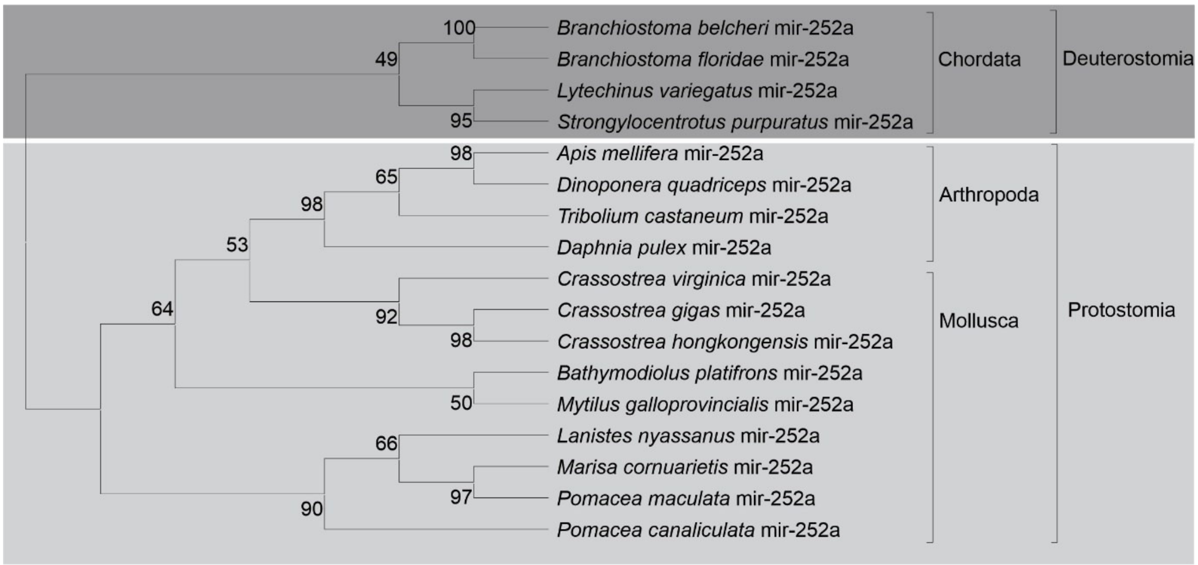
bbe-mir-252a/1-104      UCG---------------AACC--CCUGCAGACUG-GUGCUUA-ACAUGGGAC--AGGUA 92
pmi-mir-252a/1-102      UUGAUU------UCAAGACAUCA-ACUGCUACUCUGGUGCUUA-ACACGGGGUCUGGCUA 98
tca-mir-252a/1-87       UCG----------------ACCU-CCUGCUGCUCAAGUGCUUA-UCAAUGGGC--ACAGA 83
dqu-mir-252a/1-111      CUGGGCAAGACGUUACCGUAUCU-CCUGCUGCUCAAGUGCUUA-UCAACGGGU---CUGG 105
                        0........70........80........90........100.......110.......1

                        ................
pmc-mir-252a/1-94       CUUGCA---------- 94
mcr-mir-252a/1-94       CUUGCA---------- 94
pcn-mir-252a/1-94       CUUGCA---------- 94
lny-mir-252a/1-88       UUU------------- 88
sko-mir-252a/1-101      UAUUCAU--------- 101
bbe-mir-252a/1-104      AUACUGCCACCU---- 104
pmi-mir-252a/1-102      AUAA------------ 102
tca-mir-252a/1-87       AUGA------------ 87
dqu-mir-252a/1-111      ACGGCU---------- 111
                        20.......130....
```

Supplementary figure 4: Alignment of mir-252a in Ampullariidae species and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculata*, mcr - *M. cornuarietis*, lny - *L. nyassanus*, sko - *Saccoglossus kowalevskii*, bbe - *Branchiostoma belcherimle*, dqu - *Dinoponera quadríceps*, pmi - *Patiria miniata*, tca - *Tribolium castaneum*

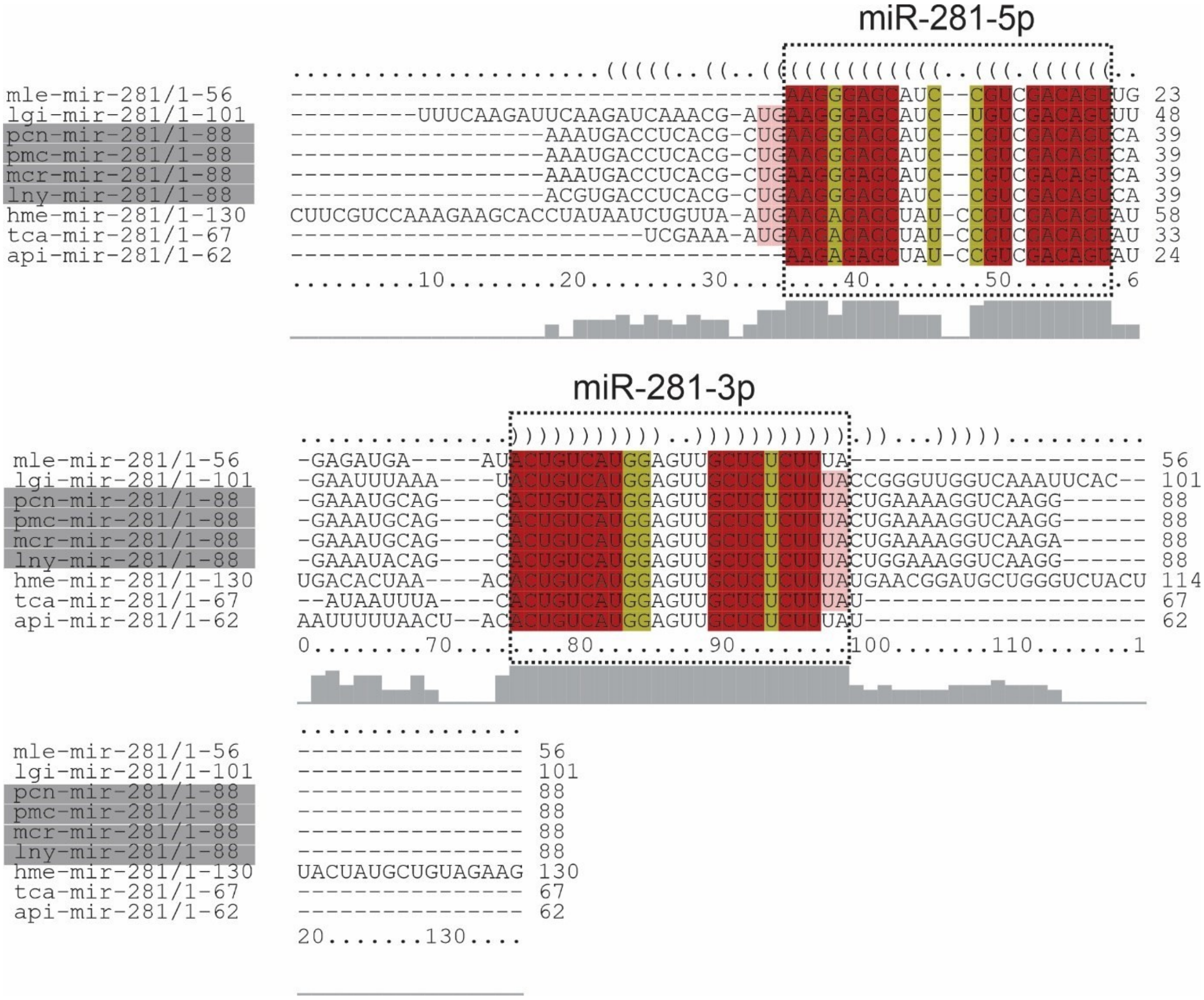


Supplementary figure 5: Secondary structure of mir-252a in Ampullariidae species and their orthologs; pcn - *P. canaliculata*, pmc - *P. maculata*, mcr - *M. cornuarietis*, lny - *L. nyassanus*, sko - *Saccoglossus kowalevskii,* bbe - *Branchiostoma belcherimle*, dqu - *Dinoponera quadríceps*, pmi - *Patiria miniata*, tca - *Tribolium castaneum.*



Supplementary figure 6: Phylogenetic distribution of mir-252a in Ampullariidae species and their orthologs.

mir-281



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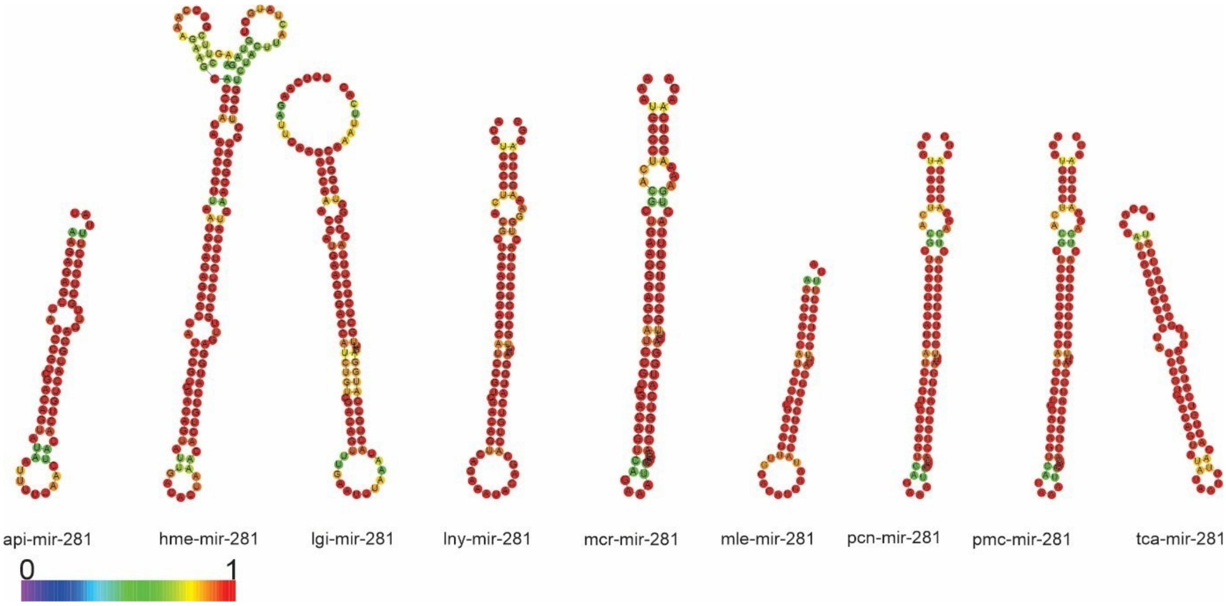
                     ......................(((((..((..(((((((((((((..(((.((((((..
mle-mir-281/1-56     -----------------------------------AAGGGAGCAUC--CGUCGACAGUUG 23
lgi-mir-281/1-101    ---------UUUCAAGAUUCAAGAUCAAACG-AUGAAGGGAGCAUC--UGUCGACAGUUU 48
pcn-mir-281/1-88     ------------------AAAUGACCUCACG-CUGAAGGGAGCAUC--CGUCGACAGUCA 39
pmc-mir-281/1-88     ------------------AAAUGACCUCACG-CUGAAGGGAGCAUC--CGUCGACAGUCA 39
mcr-mir-281/1-88     ------------------AAAUGACCUCACG-CUGAAGGGAGCAUC--CGUCGACAGUCA 39
lny-mir-281/1-88     ------------------ACGUGACCUCACG-CUGAAGGGAGCAUC--CGUCGACAGUCA 39
hme-mir-281/1-130    CUUCGUCCAAAGAAGCACCUAUAAUCUGUUA-AUGAAGAGAGCUAU-CCGUCGACAGUAU 58
tca-mir-281/1-67     -------------------------UCGAAA-AUGAAGAGAGCUAU-CCGUCGACAGUAU 33
api-mir-281/1-62     -----------------------------------AAGAGAGCUAU-CCGUCGACAGUAU 24
                     .........10........20........30........40........50........6

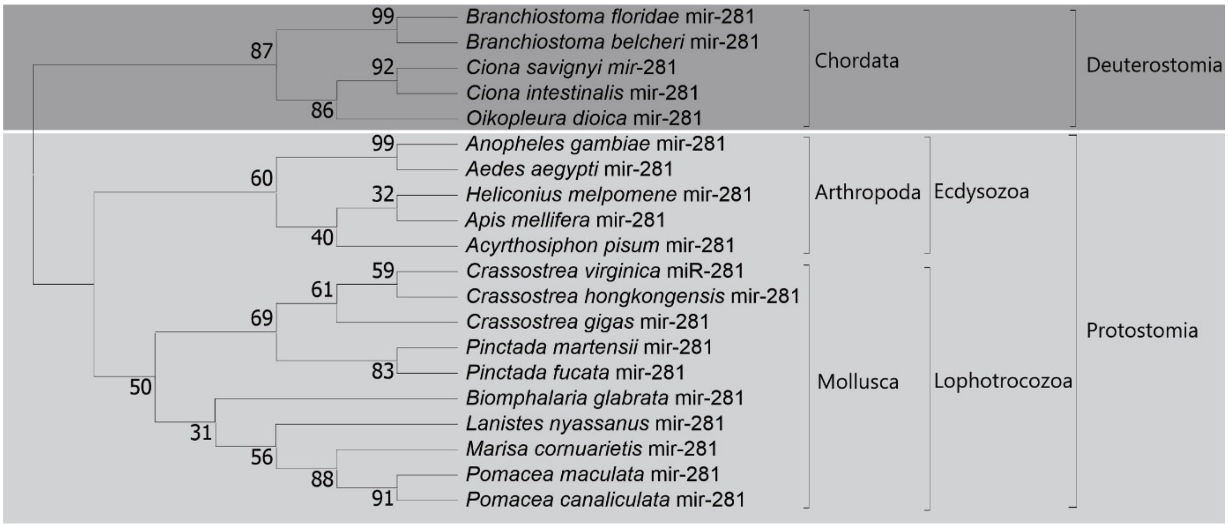
                     ...............)))))))))))..))))))))))).))...))))))..........
mle-mir-281/1-56     -GAGAUGA-----AUACUGUCAUGGAGUUGCUCUCUUUA--------------------- 56
lgi-mir-281/1-101    -GAAUUUAAA----UACUGUCAUGGAGUUGCUCUCUUUACCGGGUUGGUCAAAUUCAC-- 101
pcn-mir-281/1-88     -GAAAUGCAG----CACUGUCAUGGAGUUGCUCUCUUUACUGAAAAGGUCAAGG------ 88
pmc-mir-281/1-88     -GAAAUGCAG----CACUGUCAUGGAGUUGCUCUCUUUACUGAAAAGGUCAAGG------ 88
mcr-mir-281/1-88     -GAAAUGCAG----CACUGUCAUGGAGUUGCUCUCUUUACUGAAAAGGUCAAGA------ 88
lny-mir-281/1-88     -GAAAUACAG----CACUGUCAUGGAGUUGCUCUCUUUACUGGAAAGGUCAAGG------ 88
hme-mir-281/1-130    UGACACUAA----ACACUGUCAUGGAGUUGCUCUCUUUAUGAACGGAUGCUGGGUCUACU 114
tca-mir-281/1-67     --AUAAUUUA----CACUGUCAUGGAGUUGCUCUCUUUAU-------------------- 67
api-mir-281/1-62     AAUUUUUAACU--ACACUGUCAUGGAGUUGCUCUCUUUAU-------------------- 62
                     0........70........80........90........100.......110.......1

                     ................
mle-mir-281/1-56     ---------------- 56
lgi-mir-281/1-101    ---------------- 101
pcn-mir-281/1-88     ---------------- 88
pmc-mir-281/1-88     ---------------- 88
mcr-mir-281/1-88     ---------------- 88
lny-mir-281/1-88     ---------------- 88
hme-mir-281/1-130    UACUAUGCUGUAGAAG 130
tca-mir-281/1-67     ---------------- 67
api-mir-281/1-62     ---------------- 62
                     20.......130....
```

Supplementary figure 7: Alignment of mir-281 in Ampullariidae species and their orthologs; api - *Acyrthosiphon pisum*, hme - *Heliconius melpomene*, lny - *L. nyassanus*, lgi - *L. gigantea,* mcr - *M. cornuarietis*, mle - *M. leonina,* pcn - *P. canaliculata*, pmc - *P. maculata*, tca - *Tribolium castaneum*.

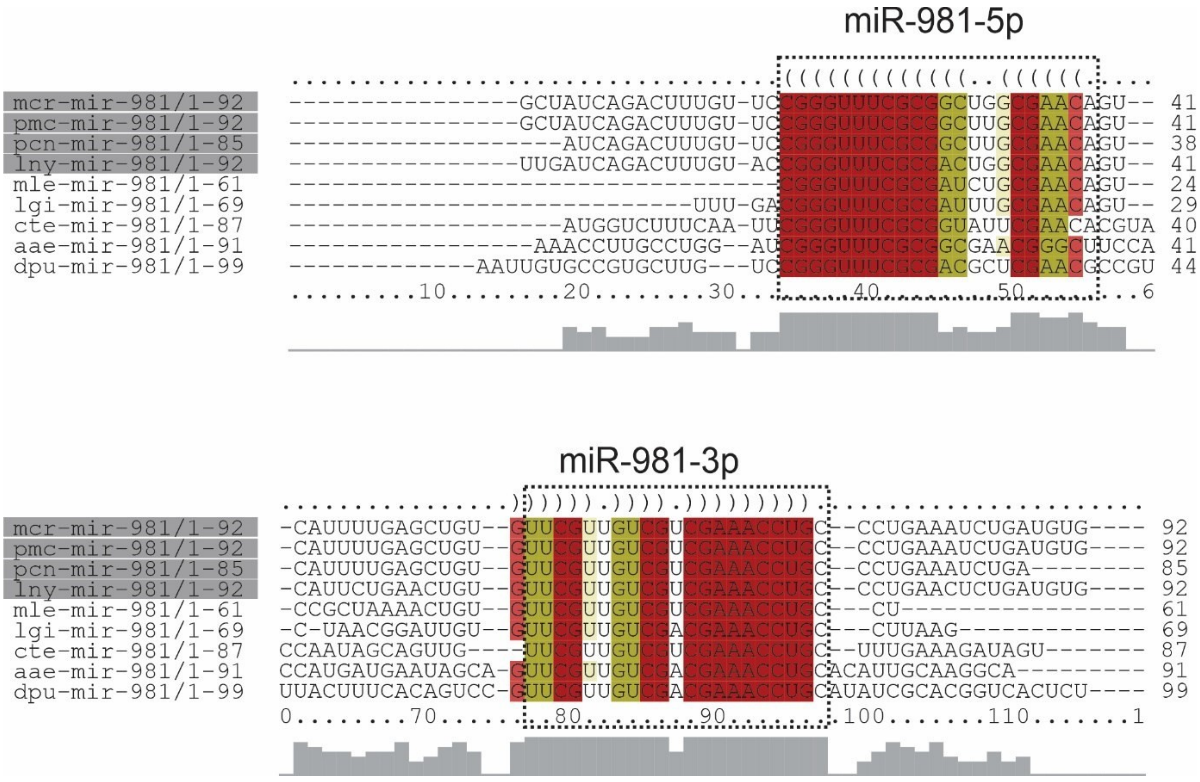


Supplementary figure 8: Secondary structure of mir-281 in Ampullariidae species and their orthologs; api - *Acyrthosiphon pisum*, hme - *Heliconius melpomene*, lny - *L. nyassanus*, lgi - *L. gigantea,* mcr - *M. cornuarietis*, mle - *M. leonina,* pcn - *P. canaliculata*, pmc - *P. maculata*, tca - *Tribolium castaneum.*



Supplementary figure 9: Phylogenetic distribution of mir-281 in Ampullariidae species and their orthologs.

mir-981



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                    ..................................(((((((((((((..((((((.....
mcr-mir-981/1-92    ----------------GCUAUCAGACUUUGU-UCCGGGUUUCGCGGCUGGCGAACAGU--  41
pmc-mir-981/1-92    ----------------GCUAUCAGACUUUGU-UCCGGGUUUCGCGGCUUGCGAACAGU--  41
pcn-mir-981/1-85    -------------------AUCAGACUUUGU-UCCGGGUUUCGCGGCUUGCGAACAGU--  38
lny-mir-981/1-92    ----------------UUGAUCAGACUUUGU-ACCGGGUUUCGCGACUGGCGAACAGU--  41
mle-mir-981/1-61    ----------------------------------CGGGUUUCGCGAUCUGCGAACAGU--  24
lgi-mir-981/1-69    ----------------------------UUU-GACGGGUUUCGCGAUUUGCGAACAGU--  29
cte-mir-981/1-87    -------------------AUGGUCUUUCAA-UUCGGGUUUCGCGGUAUUCGAACACGUA  40
aae-mir-981/1-91    -----------------AAACCUUGCCUGG--AUCGGGUUUCGCGGCGAACGGGCUUCCA  41
dpu-mir-981/1-99    -------------AAUUGUGCCGUGCUUG---UCCGGGUUUCGCGACGCUCGAACGCCGU  44
                    ........10........20........30........40........50.........6
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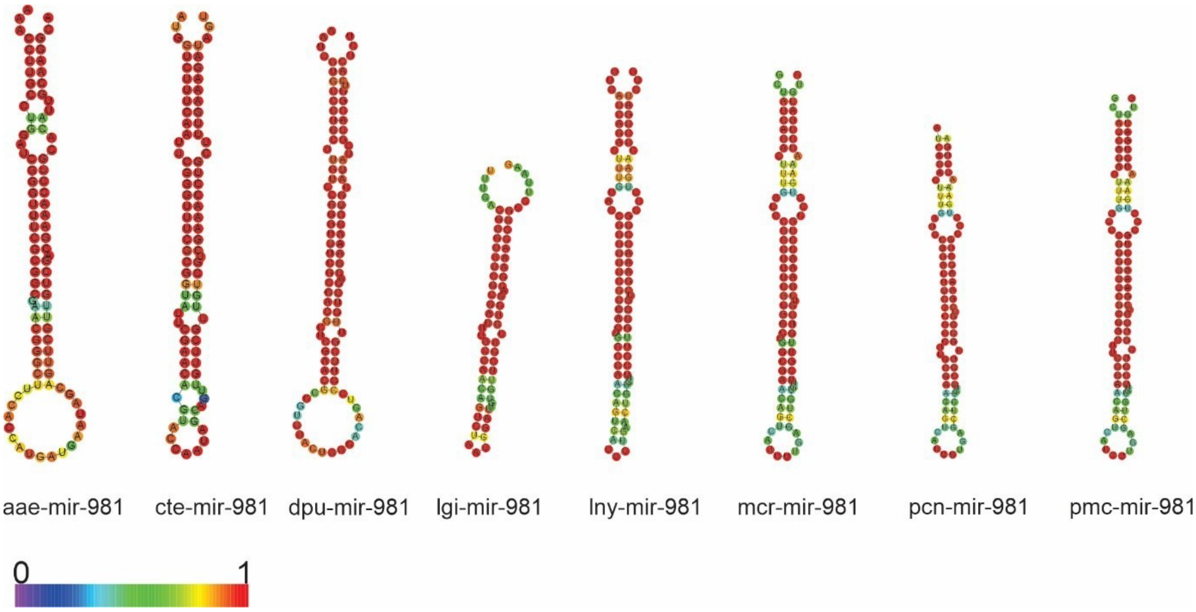
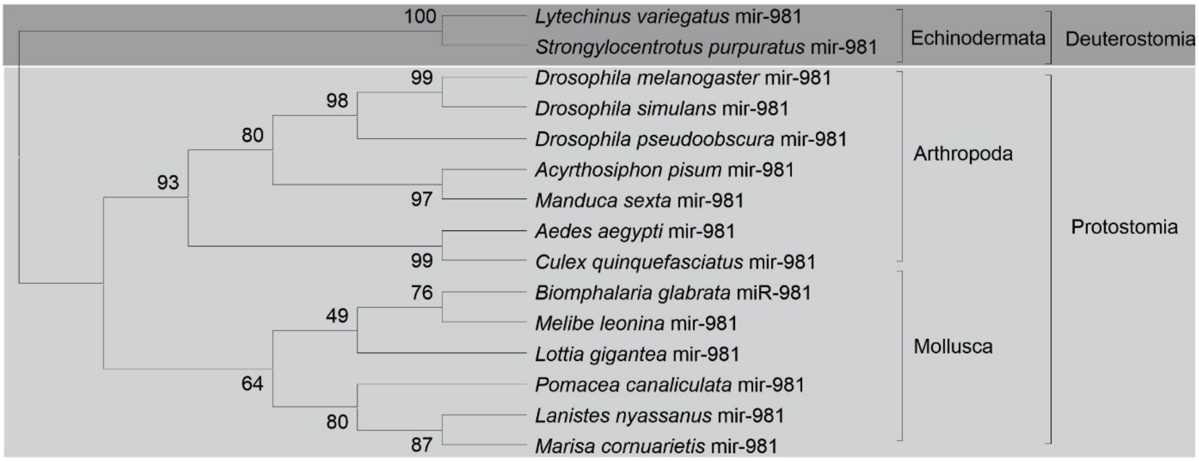
                    ................)))))).)))).))))))))).......................
mcr-mir-981/1-92    -CAUUUUGAGCUGU--GUUCGUUGUCGUCGAAACCUGC--CCUGAAAUCUGAUGUG----  92
pmc-mir-981/1-92    -CAUUUUGAGCUGU--GUUCGUUGUCGUCGAAACCUGC--CCUGAAAUCUGAUGUG----  92
pcn-mir-981/1-85    -CAUUUUGAGCUGU--GUUCGUUGUCGUCGAAACCUGC--CCUGAAAUCUGA--------  85
lny-mir-981/1-92    -CAUUCUGAACUGU--GUUCGUUGUCGUCGAAACCUGC--CCUGAACUCUGAUGUG----  92
mle-mir-981/1-61    -CCGCUAAAACUGU--GUUCGUUGUCGUCGAAACCUGC---CU-----------------  61
lgi-mir-981/1-69    -C-UAACGGAUUGU--GUUCGUUGUCGACGAAACCUGC---CUUAAG-------------  69
cte-mir-981/1-87    CCAAUAGCAGUUG----UUCGUUGUCGUCGAAACCUGC--UUUGAAAGAUAGU-------  87
aae-mir-981/1-91    CCAUGAUGAAUAGCA-GUUCGUUGUCGACGAAACCUGCACAUUGCAAGGCA---------  91
dpu-mir-981/1-99    UUACUUUCACAGUCC-GUUCGUUGUCGACGAAACCUGCAUAUCGCACGGUCACUCU----  99
                    0.......70........80........90.......100.......110.........1
```

Supplementary figure 10: Alignment of mir-981 in Ampullariidae species and their orthologs; aae - Aedes aegypti, dpu - Daphnia pulex, mle – M. leonina, lny - L. nyassanus, lgi - L. gigantea, mcr - M. cornuarietis, pcn - P. canaliculata, pmc - P. maculata, cte - C. teleta.



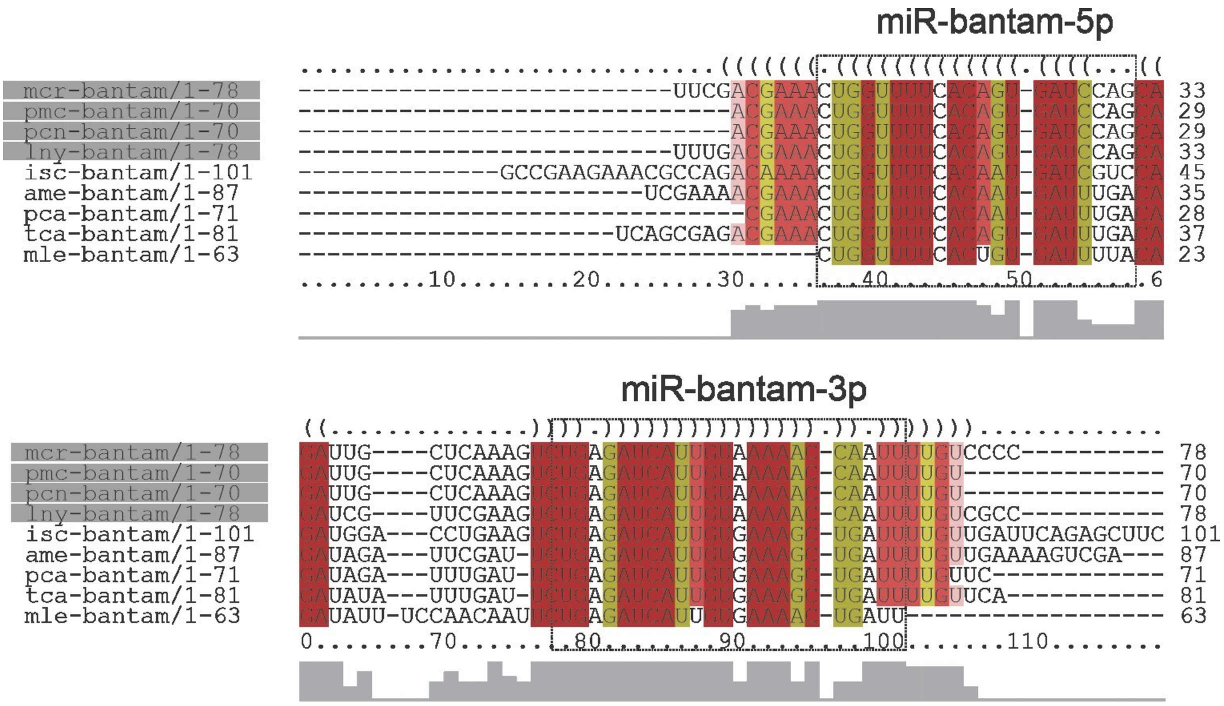
Supplementary figure 11: Secondary structure of mir-981 in Ampullariidae species and their orthologs; aae - *Aedes aegypti,* dpu - *Daphnia pulex,* cte - *C. teleta,* lny - *L. nyassanus,* lgi - *L. gigantea,* mcr - *M. cornuarietis,* pcn - *P. canaliculata,* pmc - *P. maculata.*



Supplementary figure 12: Phylogenetic distribution of mir-981 in Ampullariidae species and their orthologs.

Protostome specific miRNAs

mir-bantam



miR-bantam-5p

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                     ..............................((((((.(((((((((((((.((((...((
mcr-bantam/1-78      --------------------------UUCGACGAAACUGGUUUUCACAGU-GAUCCAGCA 33
pmc-bantam/1-70      ------------------------------ACGAAACUGGUUUUCACAGU-GAUCCAGCA 29
pcn-bantam/1-70      ------------------------------ACGAAACUGGUUUUCACAGU-GAUCCAGCA 29
lny-bantam/1-78      --------------------------UUUGACGAAACUGGUUUUCACAGU-GAUCCAGCA 33
isc-bantam/1-101     --------------GCCGAAGAAACGCCAGACAAAACUGGUUUUCACAAU-GAUCGUCCA 45
ame-bantam/1-87      ------------------------UCGAAAACGAAACUGGUUUUCACAAU-GAUUUGACA 35
pca-bantam/1-71      -------------------------------CGAAACUGGUUUUCACAAU-GAUUUGACA 28
tca-bantam/1-81      ----------------------UCAGCGAGACGAAACUGGUUUUCACAGU-GAUUUGACA 37
mle-bantam/1-63      ------------------------------------CUGGUUUUCACUGU-GAUUUUACA 23
                     ........10........20........30........40........50........6
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miR-bantam-3p

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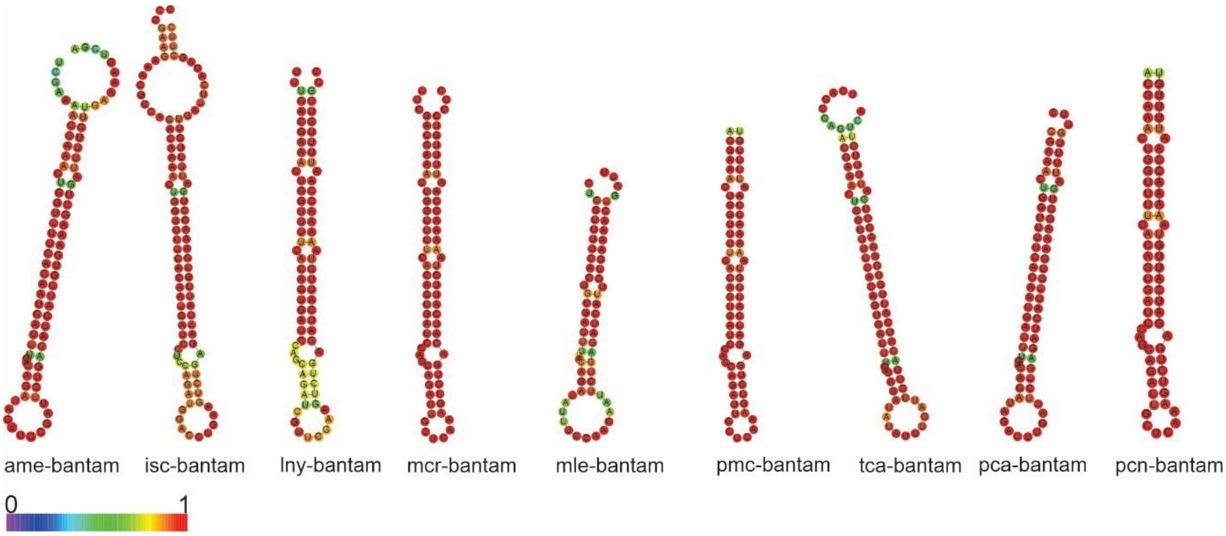
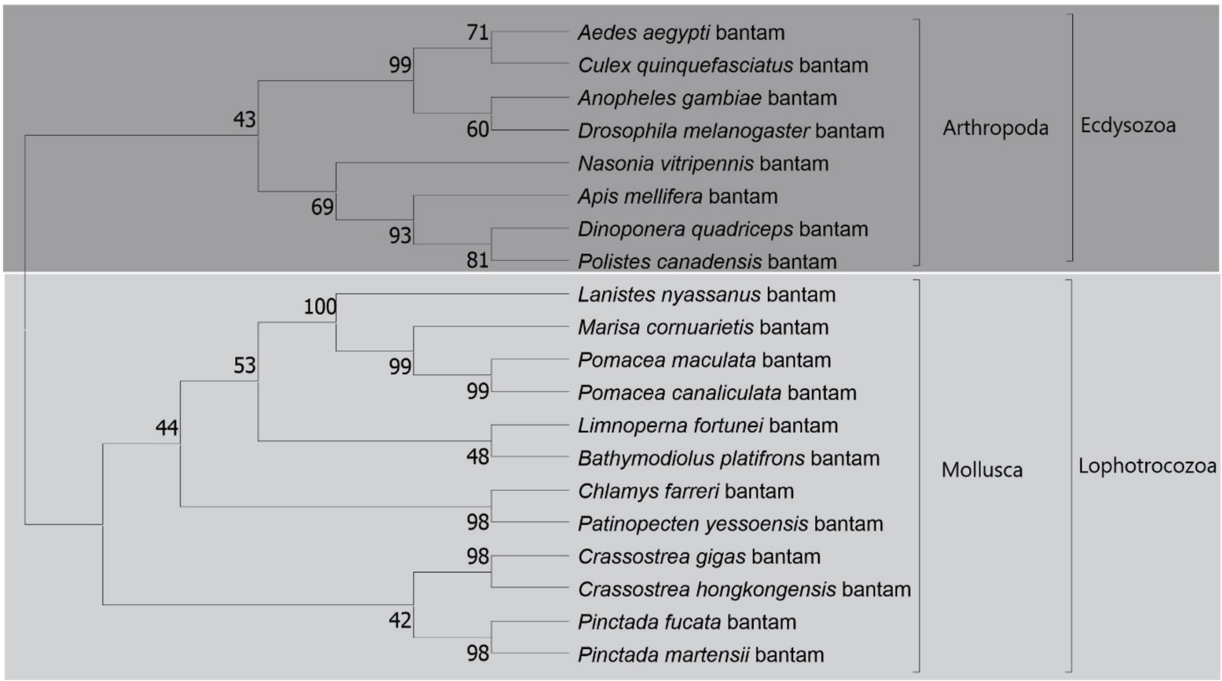
                     ((..............)))).)))))))))))).)).)))))))))..............
mcr-bantam/1-78      GAUUG----CUCAAAGUCUGAGAUCAUUGUAAAAAC-CAAUUUUGUCCCC---------- 78
pmc-bantam/1-70      GAUUG----CUCAAAGUCUGAGAUCAUUGUAAAAAC-CAAUUUUGU-------------- 70
pcn-bantam/1-70      GAUUG----CUCAAAGUCUGAGAUCAUUGUAAAAAC-CAAUUUUGU-------------- 70
lny-bantam/1-78      GAUCG----UUCGAAGUCUGAGAUCAUUGUAAAAAC-CAAUUUUGUCGCC---------- 78
isc-bantam/1-101     GAUGGA---CCUGAAGUCUGAGAUCAUUGUGAAAGC-UGAUUUUGUUGAUUCAGAGCUUC 101
ame-bantam/1-87      GAUAGA---UUCGAU-UCUGAGAUCAUUGUGAAAGC-UGAUUUUGUUGAAAAGUCGA--- 87
pca-bantam/1-71      GAUAGA---UUUGAU-UCUGAGAUCAUUGUGAAAGC-UGAUUUUGUUC----------- 71
tca-bantam/1-81      GAUAUA---UUUGAU-UCUGAGAUCAUUGUGAAAGC-UGAUUUUGUUCA---------- 81
mle-bantam/1-63      GAUAUU-UCCAACAAUUCUGAGAUCAUUGUGAAAAC-UGAUU----------------- 63
                     0........70........80........90........100.......110........
```

Supplementary figure 13: Alignment of mi-bantam in Ampullariidae species and their orthologs; ame - *Apis melífera,* isc - *Ixodes scapularis,* lny - *L. nyassanus,* mcr - *M. cornuarietis,* pca - *Polistes canadenses,* pcn - *P. canaliculata,* pmc - *P. maculata*, tca - *Tribolium castaneum*, mle - *M. leonina.*

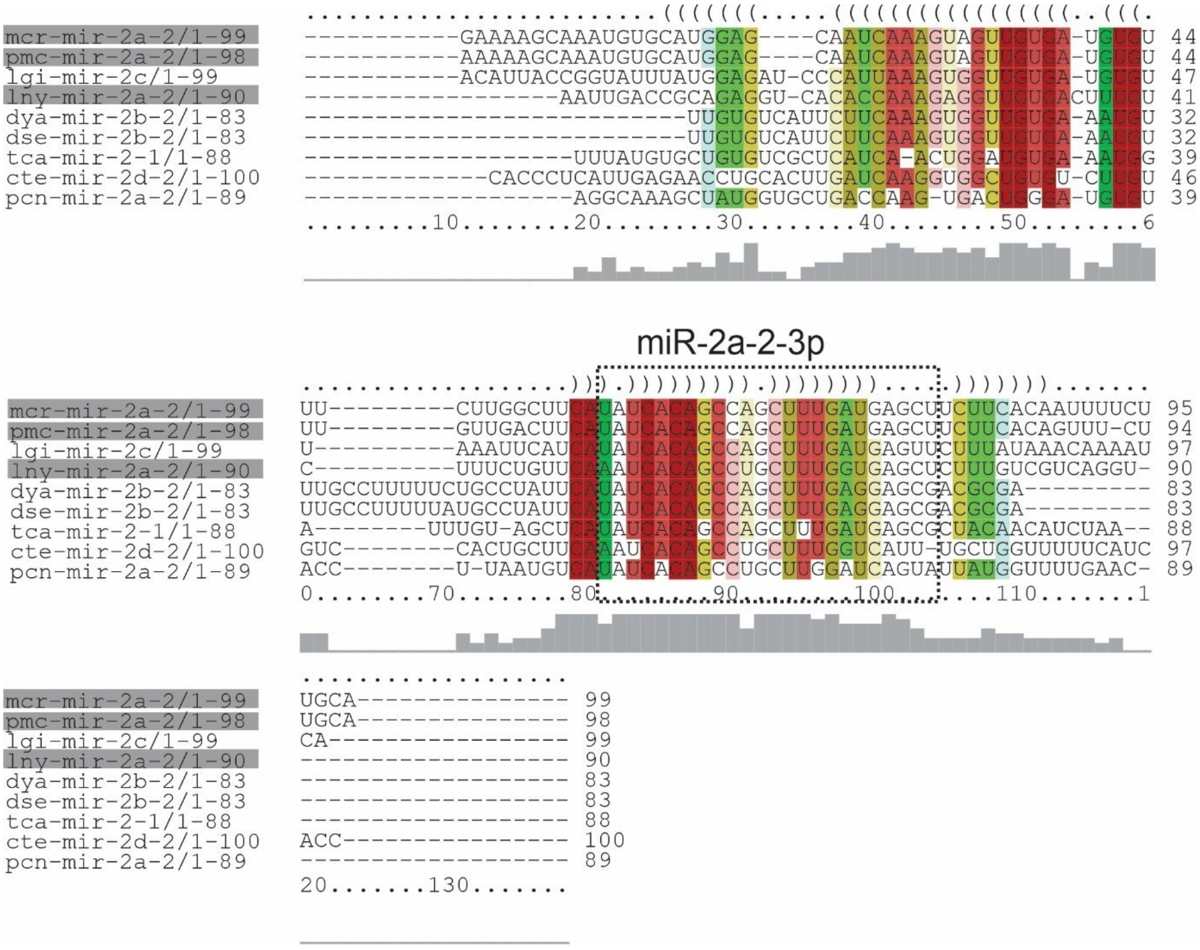


Supplementary figure 14: Secondary structure of mir-bantam in Ampullariidae species and their orthologs; ame – *Apis melífera,* isc - *Ixodes scapularis,* lny - *L. nyassanus,* mcr - *M. cornuarietis,* pca - *Polistes canadenses,* pcn - *P. canaliculata,* pmc - *P. maculata*, tca - *Tribolium castaneum*, mle – *M. leonina.*



Supplementary figure 15: Phylogenetic distribution of mir-bantam in Ampullariidae species and their orthologs.

mir- 2a-2



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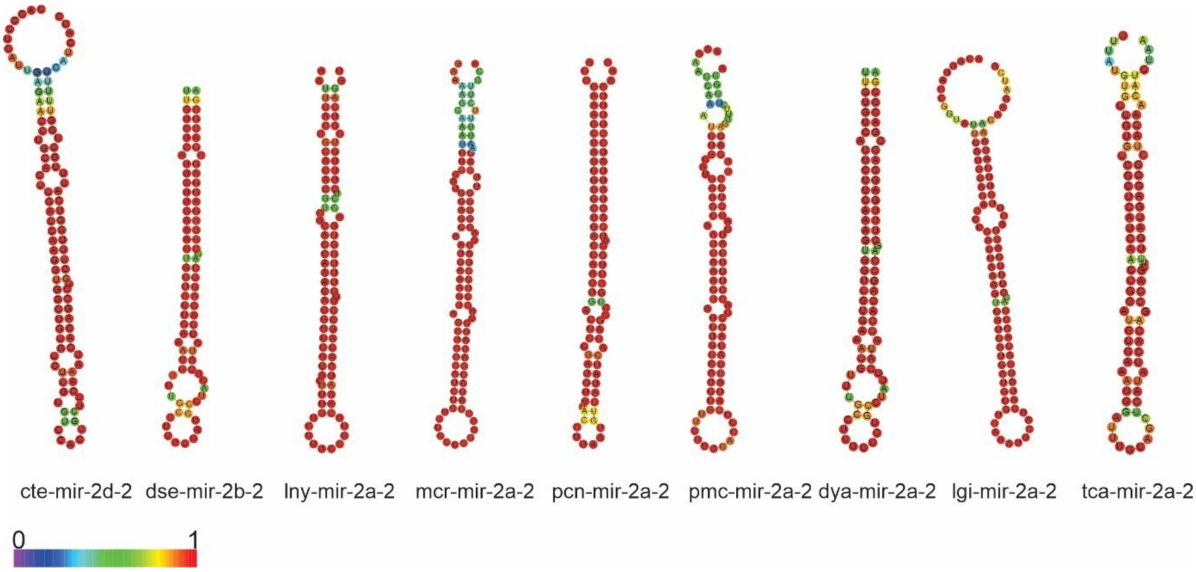
                   .........................(((((((.....(((((((((((((((((..(((.
mcr-mir-2a-2/1-99  -----------GAAAAGCAAAUGUGCAUGGAG----CAAUCAAAGUAGUUGUGA-UGUGU 44
pmc-mir-2a-2/1-98  -----------AAAAAGCAAAUGUGCAUGGAG----CAAUCAAAGUAGUUGUGA-UGUGU 44
lgi-mir-2c/1-99    -----------ACAUUACCGGUAUUUAUGGAGAU-CCCAUUAAAGUGGUUGUGA-UGUGU 47
lny-mir-2a-2/1-90  ------------------AAUUGACCGCAGAGGU-CACACCAAAGAGGUUGUGACUUUGU 41
dya-mir-2b-2/1-83  ---------------------------UUGUGUCAUUCUUCAAAGUGGUUGUGA-AAUGU 32
dse-mir-2b-2/1-83  ---------------------------UUGUGUCAUUCUUCAAAGUGGUUGUGA-AAUGU 32
tca-mir-2-1/1-88   -------------------UUUAUGUGCUGUGUCGCUCAUCA-ACUGGAUGUGA-AAUGG 39
cte-mir-2d-2/1-100 -------------CACCCUCAUUGAGAACCUGCACUUGAUCAAGGUGGCUGUGU-CUUGU 46
pcn-mir-2a-2/1-89  -------------------AGGCAAAGCUAUGGUGCUGACCAAG-UGACUGGGA-UGUGU 39
                   ........10........20........30........40........50.........6

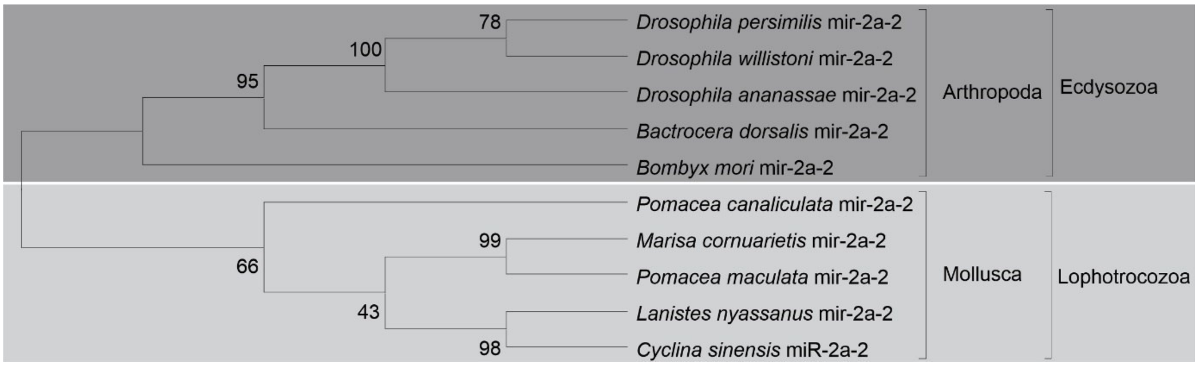
                   ...................))).)))))))))).)))))))).....)))))))).......
mcr-mir-2a-2/1-99  UU---------CUUGGCUUCAUAUCACAGCCAGCUUUGAUGAGCUUCUUCACAAUUUUCU 95
pmc-mir-2a-2/1-98  UU---------GUUGACUUCAUAUCACAGCCAGCUUUGAUGAGCUUCUUCACAGUUU-CU 94
lgi-mir-2c/1-99    U----------AAAUUCAUCAUAUCACAGCCAGCUUUGAUGAGUUCUUUAUAAACAAAAU 97
lny-mir-2a-2/1-90  C----------UUUCUGUUCAAAUCACAGCCUGCUUUGGUGAGCUCUUUGUCGUCAGGU- 90
dya-mir-2b-2/1-83  UUGCCUUUUUCUGCCUAUUCAUAUCACAGCCAGCUUUGAGGAGCGACGCGA--------- 83
dse-mir-2b-2/1-83  UUGCCUUUUUAUGCCUAUUCAUAUCACAGCCAGCUUUGAGGAGCGACGCGA--------- 83
tca-mir-2-1/1-88   A--------UUUGU-AGCUCAUAUCACAGCCAGCUUUGAUGAGCGCUACAACAUCUAA-- 88
cte-mir-2d-2/1-100 GUC--------CACUGCUUCAAAUCACAGCCUGCUUUGGUCAUU-UGCUGGUUUUUCAUC 97
pcn-mir-2a-2/1-89  ACC--------U-UAAUGUCAUAUCACAGCCUGCUUGGAUCAGUAUUAUGGUUUUGAAC- 89
                   0.......70........80........90.......100.......110.........1

                   ...................
mcr-mir-2a-2/1-99  UGCA--------------- 99
pmc-mir-2a-2/1-98  UGCA--------------- 98
lgi-mir-2c/1-99    CA----------------- 99
lny-mir-2a-2/1-90  ------------------- 90
dya-mir-2b-2/1-83  ------------------- 83
dse-mir-2b-2/1-83  ------------------- 83
tca-mir-2-1/1-88   ------------------- 88
cte-mir-2d-2/1-100 ACC---------------- 100
pcn-mir-2a-2/1-89  ------------------- 89
                   20.......130.......
```

Supplementary figure 16: Alignment of mir-2a-2 in Ampullariidae species and their orthologs; cte - *C. teleta*, dse - *Drosophila sechellia*, lny - *L. nyassanus,* mcr - *M. cornuarietis,* pcn - *P. canaliculata,* pmc - *P. maculata*, dya - *Drosophila yakuba*, lgi- *L. gigantea,* tca - *Tribolium castaneum.*

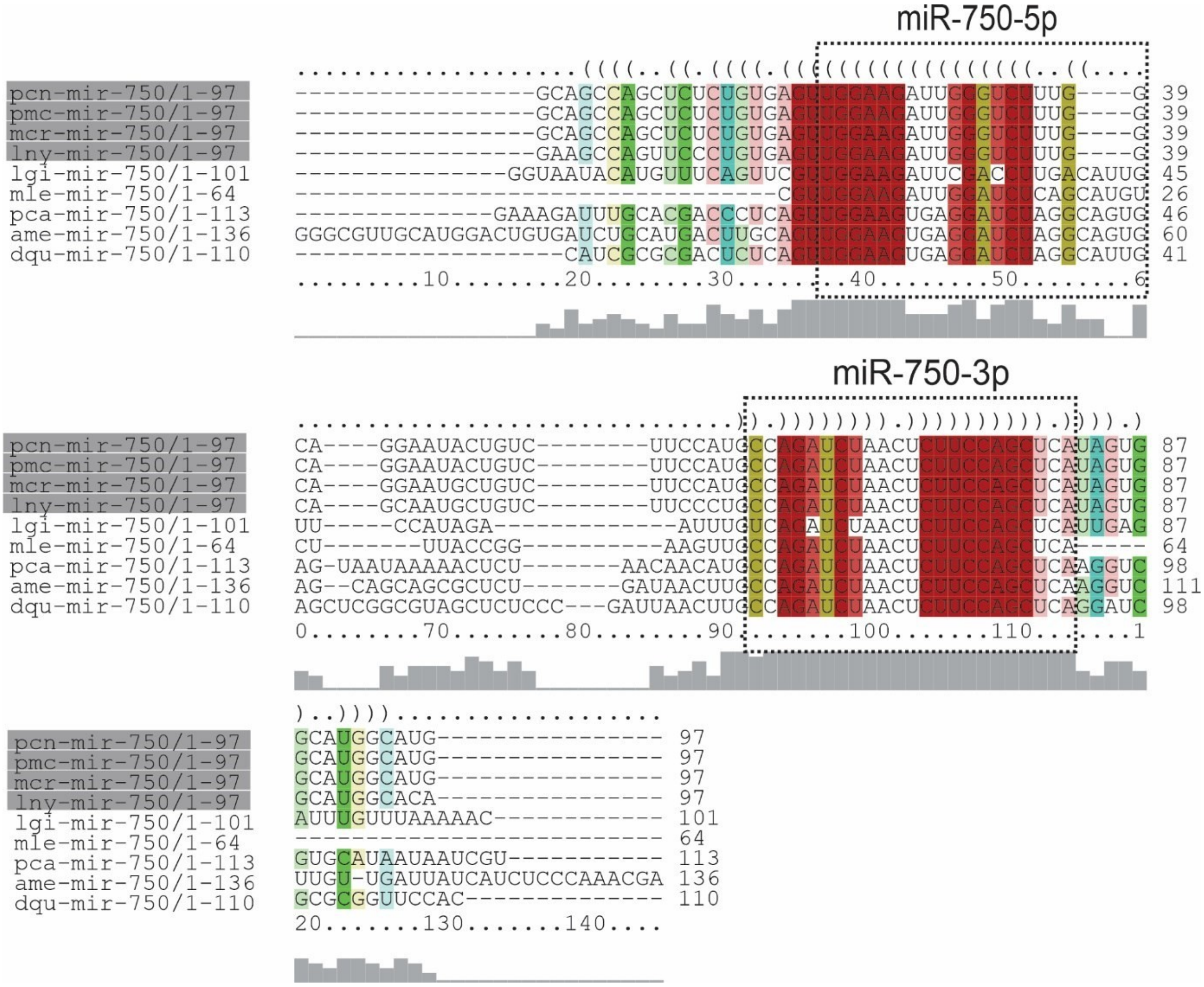


Supplementary figure 17: Secondary structure of mir-2a-2 in Ampullariidae species and their orthologs; cte - *C. teleta*, dse - *Drosophila sechellia*, lny - *L. nyassanus,* mcr - *M. cornuarietis,* pcn - *P. canaliculata,* pmc - *P. maculata*, dya - *Drosophila yakuba*, lgi- *L. gigantea,* tca - *Tribolium castaneum*



Supplementary figure 18: Phylogenetic distribution of mir-2a-2 in Ampullariidae species and their orthologs

mir-750



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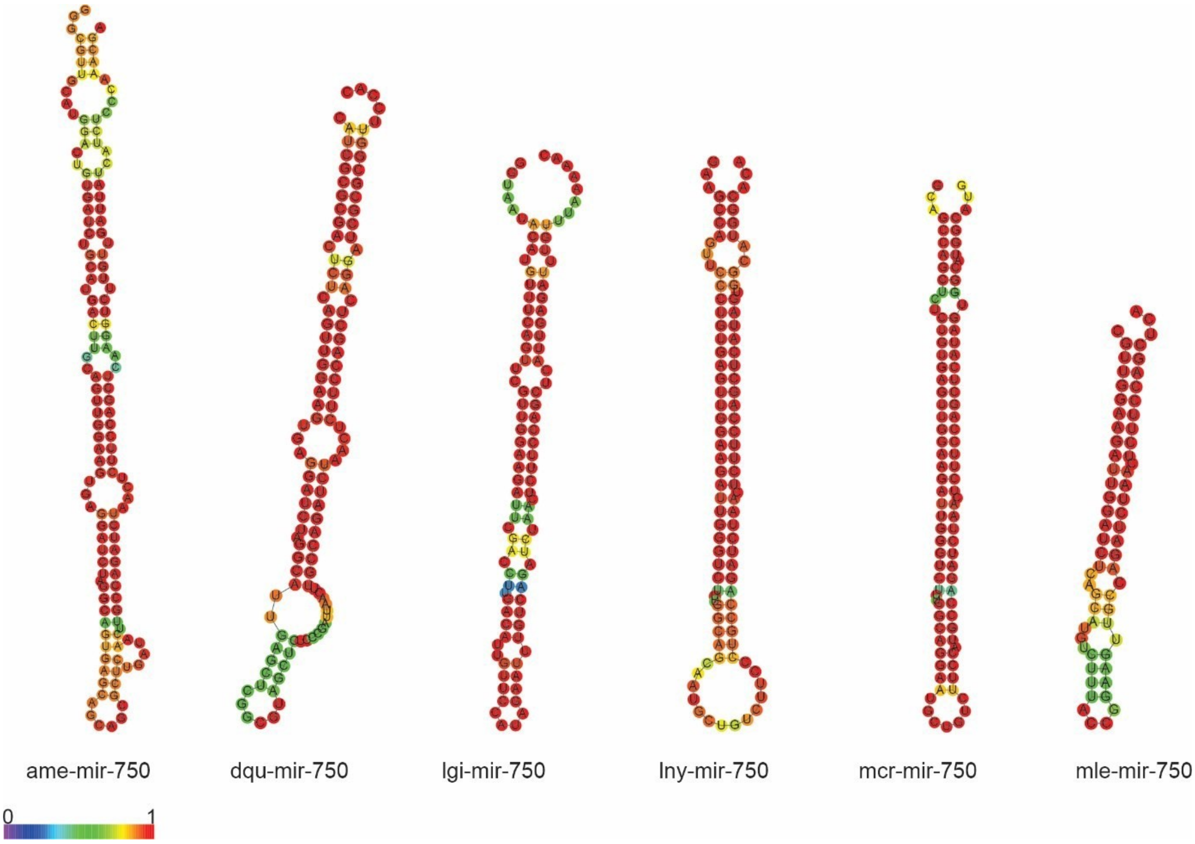
                     ....................((((..((.((((.((((((((((((((((((..((....
pcn-mir-750/1-97     -----------------GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUG----G 39
pmc-mir-750/1-97     -----------------GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUG----G 39
mcr-mir-750/1-97     -----------------GCAGCCAGCUCUCUGUGAGUUGGAAGAUUGGGUCUUUG----G 39
lny-mir-750/1-97     -----------------GAAGCCAGUUCCCUGUGAGUUGGAAGAUUGGGUCUUUG----G 39
lgi-mir-750/1-101    ---------------GGUAAUACAUGUUUCAGUUCGUUGGAAGAUUCGACCUUGACAUUG 45
mle-mir-750/1-64     ----------------------------------CGUUGGAAGAUUGGAUCUCAGCAUGU 26
pca-mir-750/1-113    --------------GAAAGAUUUGCACGACCCUCAGUUGGAAGUGAGGAUCUAGGCAGUG 46
ame-mir-750/1-136    GGGCGUUGCAUGGACUGUGAUCUGCAUGACUUGCAGUUGGAAGUGAGGAUCUAGGCAGUG 60
dqu-mir-750/1-110    -------------------CAUCGCGCGACUCUCAGUUGGAAGUGAGGAUCUAGGCAUUG 41
                     ........10........20........30........40........50.........6

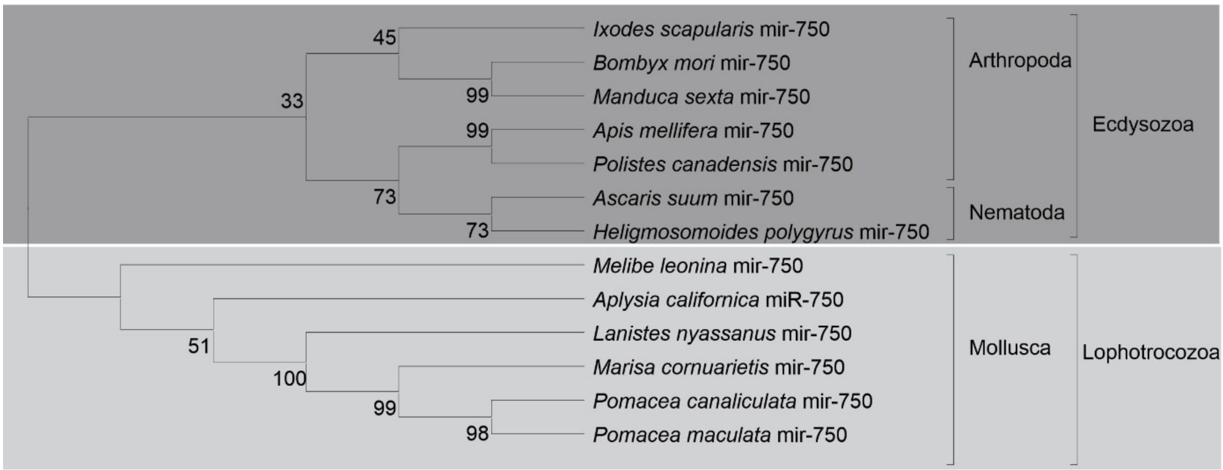
                     ...............................)).)))))))).)))))))))).)))).)
pcn-mir-750/1-97     CA----GGAAUACUGUC--------UUCCAUGCCAGAUCUAACUCUUCCAGCUCAUAGUG 87
pmc-mir-750/1-97     CA----GGAAUACUGUC--------UUCCAUGCCAGAUCUAACUCUUCCAGCUCAUAGUG 87
mcr-mir-750/1-97     CA----GGAAUGCUGUC--------UUCCAUGCCAGAUCUAACUCUUCCAGCUCAUAGUG 87
lny-mir-750/1-97     CA----GCAAUGCUGUC--------UUCCCUGCCAGAUCUAACUCUUCCAGCUCAUAGUG 87
lgi-mir-750/1-101    UU-----CCAUAGA-------------AUUUGUCAGAUCUAACUCUUCCAGCUCAUUGAG 87
mle-mir-750/1-64     CU-------UUACCGG----------AAGUUGCCAGAUCUAACUCUUCCAGCUCA----- 64
pca-mir-750/1-113    AG-UAAUAAAAACUCU-------AACAACAUGCCAGAUCUAACUCUUCCAGCUCAAGGUC 98
ame-mir-750/1-136    AG--CAGCAGCGCUCU-------GAUAACUUGCCAGAUCUAACUCUUCCAGCUCAAGGUC 111
dqu-mir-750/1-110    AGCUCGGCGUAGCUCUCCC---GAUUAACUUGCCAGAUCUAACUCUUCCAGCUCAGGAUC 98
                     0.......70........80........90.......100.......110.........1

                     )..)))))...................
pcn-mir-750/1-97     GCAUGGCAUG---------------- 97
pmc-mir-750/1-97     GCAUGGCAUG---------------- 97
mcr-mir-750/1-97     GCAUGGCAUG---------------- 97
lny-mir-750/1-97     GCAUGGCACA---------------- 97
lgi-mir-750/1-101    AUUUGUUUAAAAAC------------ 101
mle-mir-750/1-64     -------------------------- 64
pca-mir-750/1-113    GUGCAUAAUAAUCGU----------- 113
ame-mir-750/1-136    UUGU-UGAUUAUCAUCUCCCAAACGA 136
dqu-mir-750/1-110    GCGCGGUUCCAC-------------- 110
                     20.....130.......140....
```

Supplementary figure 19: Alignment of mi-750 in Ampullariidae species and their orthologs; pcn - *P. canaliculata*, lny - *L. nyassanus*, mcr - *M. cornuarietis*, pmc - *P. maculata*, ame – *Apis melífera*, dqu - *Dinoponera quadríceps*, pca - *Polistes canadenses*, lgi - *L. gigantea*, mle - *M. leonina*.



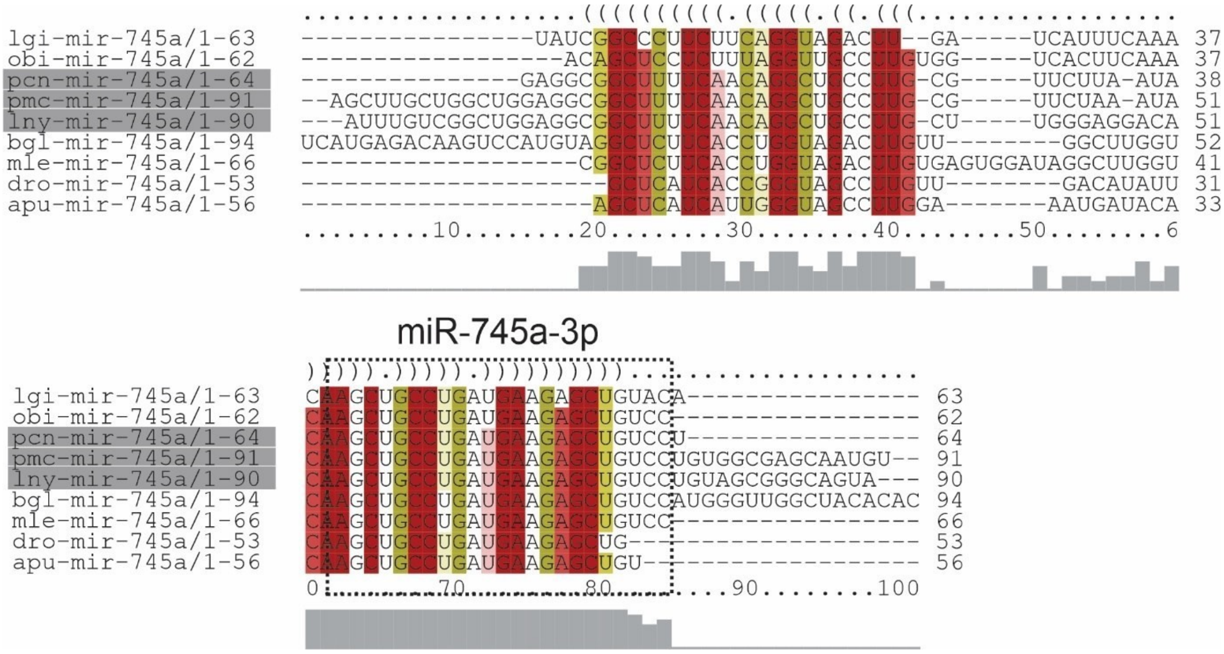
Supplementary figure 20: Secondary structure of mir-750 in Ampullariidae species and their orthologs; lny - *L. nyassanus,* mcr - *M. cornuarietis*, ame - *Apis melífera*, dqu - *Dinoponera quadriceps*, lgi - *L. gigantea*, mle - *M. leonina.*



Supplementary figure 21: Phylogenetic distribution of mir-750 in Ampullariidae species and their orthologs.

Lophotrochozoa specific miRNAs

mir-745[a]



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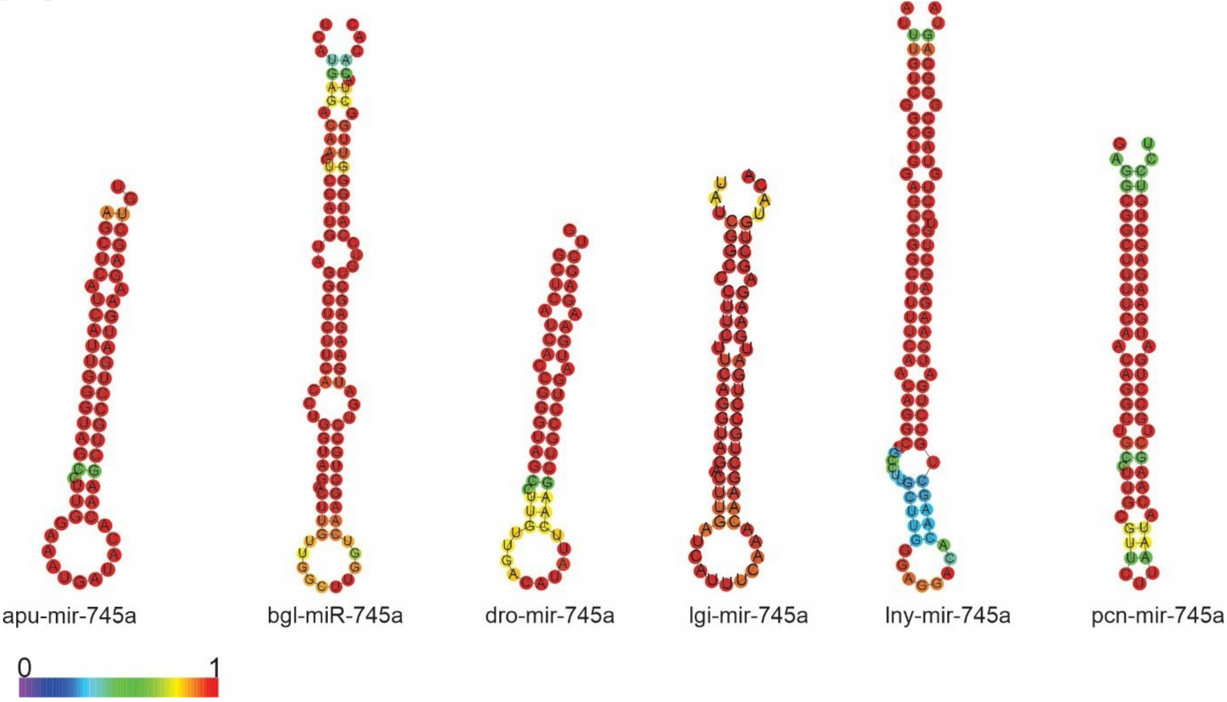
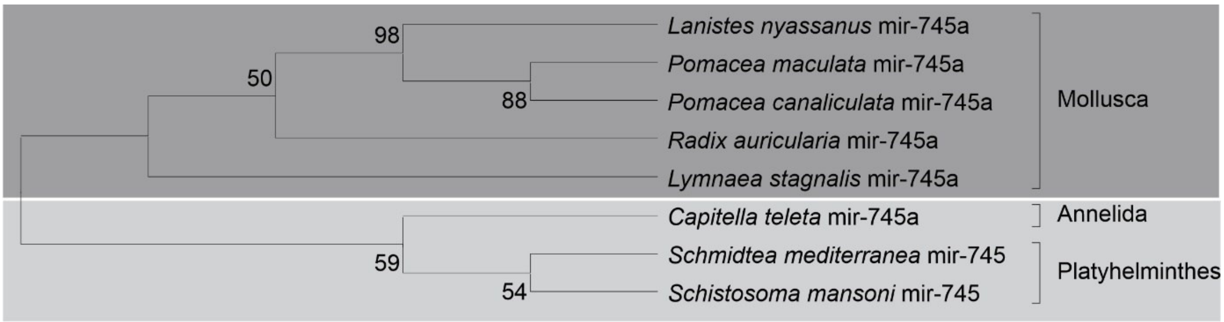
                     ....................(((((((((.(((((.((.(((..................
lgi-mir-745a/1-63    ----------------UAUCGGCCCUUCUUCAGGUAGACUU--GA-----UCAUUUCAAA 37
obi-mir-745a/1-62    ------------------ACAGCUCCUCUUUAGGUUGCCUUGUGG-----UCACUUCAAA 37
pcn-mir-745a/1-64    ---------------GAGGCGGCUUUUCAACAGGCUGCCUUG-CG-----UUCUUA-AUA 38
pmc-mir-745a/1-91    --AGCUUGCUGGCUGGAGGCGGCUUUUCAACAGGCUGCCUUG-CG-----UUCUAA-AUA 51
lny-mir-745a/1-90    ---AUUUGUCGGCUGGAGGCGGCUUUUCAACAGGCUGCCUUG-CU-----UGGGAGGACA 51
bgl-mir-745a/1-94    UCAUGAGACAAGUCCAUGUAGGCUCUUCACCUGGUAGACUUGUU--------GGCUUGGU 52
mle-mir-745a/1-66    -------------------CGGCUCUUCACCUGGUAGACUUGUGAGUGGAUAGGCUUGGU 41
dro-mir-745a/1-53    ---------------------GCUCAUCACCGGGUAGCCUUGUU--------GACAUAUU 31
apu-mir-745a/1-56    --------------------AGCUCAUCAUUGGGUAGCCUUGGA-------AAUGAUACA 33
                     .........10........20........30........40........50........6

                      miR-745a-3p
                     ))))).))))).))))))))))....................
lgi-mir-745a/1-63    CAAGCUGCCUGAUGAAGAGCUGUACA---------------- 63
obi-mir-745a/1-62    CAAGCUGCCUGAUGAAGAGCUGUCC----------------- 62
pcn-mir-745a/1-64    CAAGCUGCCUGAUGAAGAGCUGUCCU---------------- 64
pmc-mir-745a/1-91    CAAGCUGCCUGAUGAAGAGCUGUCCUGUGGCGAGCAAUGU-- 91
lny-mir-745a/1-90    CAAGCUGCCUGAUGAAGAGCUGUCCUGUAGCGGGCAGUA--- 90
bgl-mir-745a/1-94    CAAGCUGCCUGAUGAAGAGCUGUCCAUGGGUUGGCUACACAC 94
mle-mir-745a/1-66    CAAGCUGCCUGAUGAAGAGCUGUCC----------------- 66
dro-mir-745a/1-53    CAAGCUGCCUGAUGAAGAGCUG-------------------- 53
apu-mir-745a/1-56    CAAGCUGCCUGAUGAAGAGCUGU------------------- 56
                     0........70........80........90........100
```

Supplementary figure 22: Alignment of mir-745a in Ampullariidae species and their orthologs; pcn - *P. canaliculata,* lny - *L. nyassanus*, pmc - *P. maculata*, lgi - *L. gigantea*, mle - *M. leonina*, obi – *Octopus bimaculoides*, bgl – *B. glabrata*, dro - *Dreissena rostriformis*, apu - *Argopecten purpuratus.*

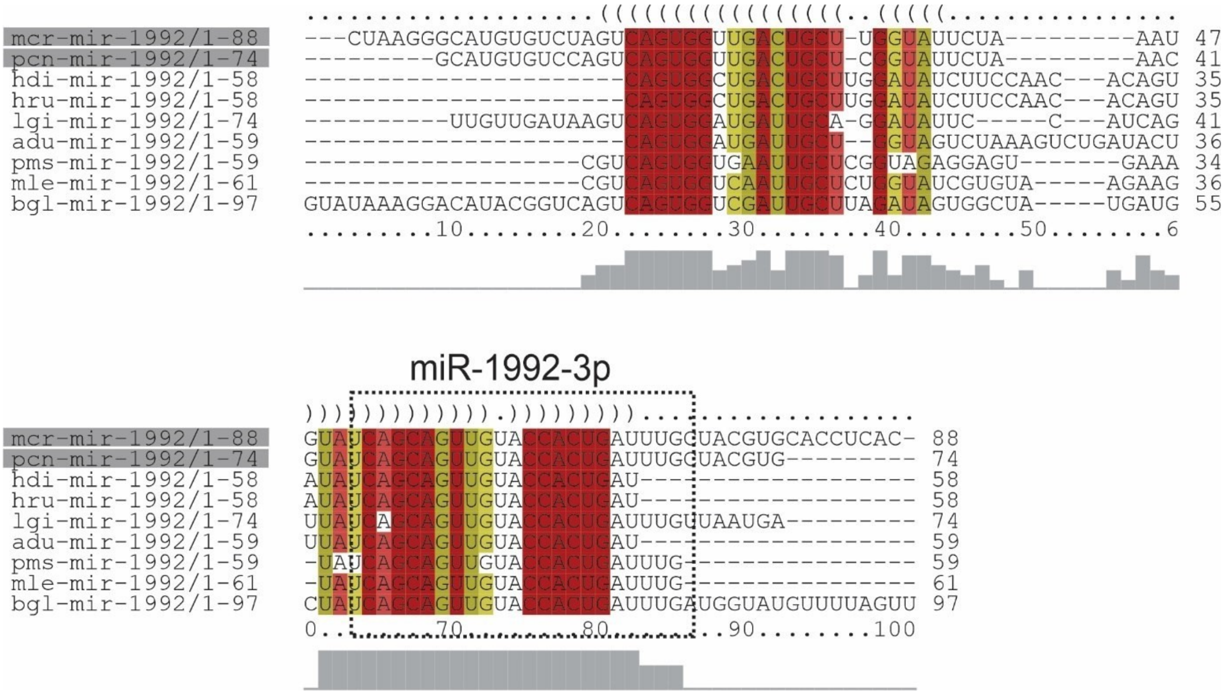


Supplementary figure 23: Secondary structure of mir-745a in Ampullariidae species and their orthologs; pcn - *P. canaliculata,* lny - *L. nyassanus*, lgi - *L. gigantea*, bgl – *B. glabrata*, dro - *Dreissena rostriformis*, apu - *Argopecten purpuratus.*

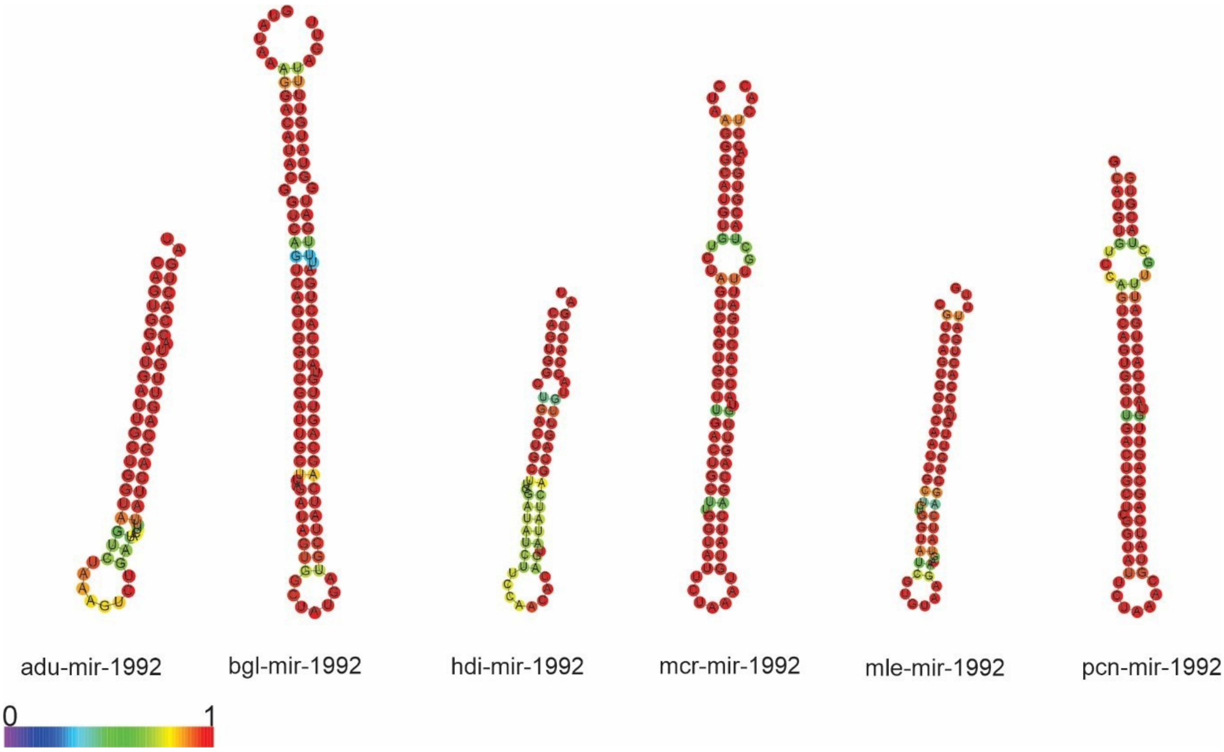


Supplementary figure 24: Phylogenetic distribution of mir-745a in Ampullariidae species and their orthologs.

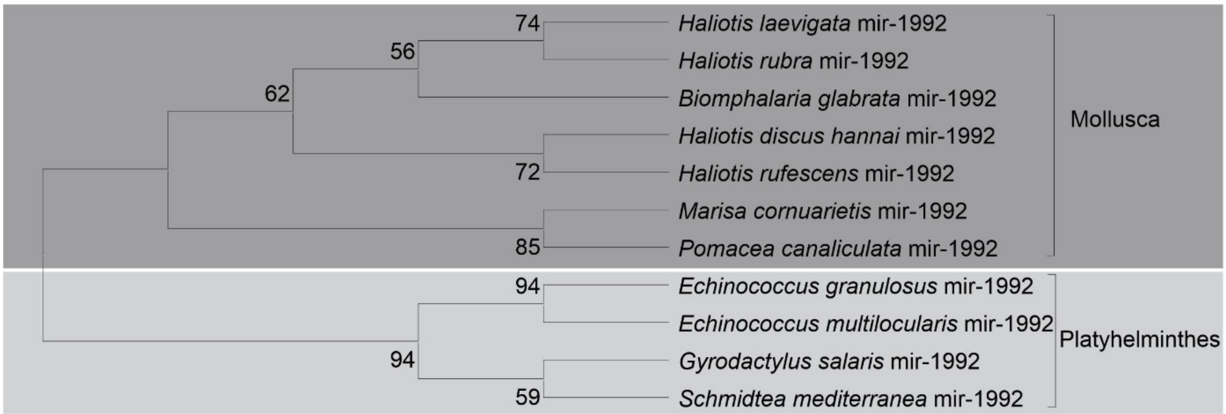
mir-1992



Supplementary figure 25: Alignment of mi-1992 in Ampullariidae species and their orthologs; mcr - *M. cornuarietis*, pcn- *P. canaliculata,* hdi - *Haliotis discus hannai*, hru – *Haliotis rufescens*, lgi - *L. gigantea*, adu - *Architeuthis dux*, pms - *Pinctada martensii*, mle - *M. leonina*, bgl – *B. glabrata.*

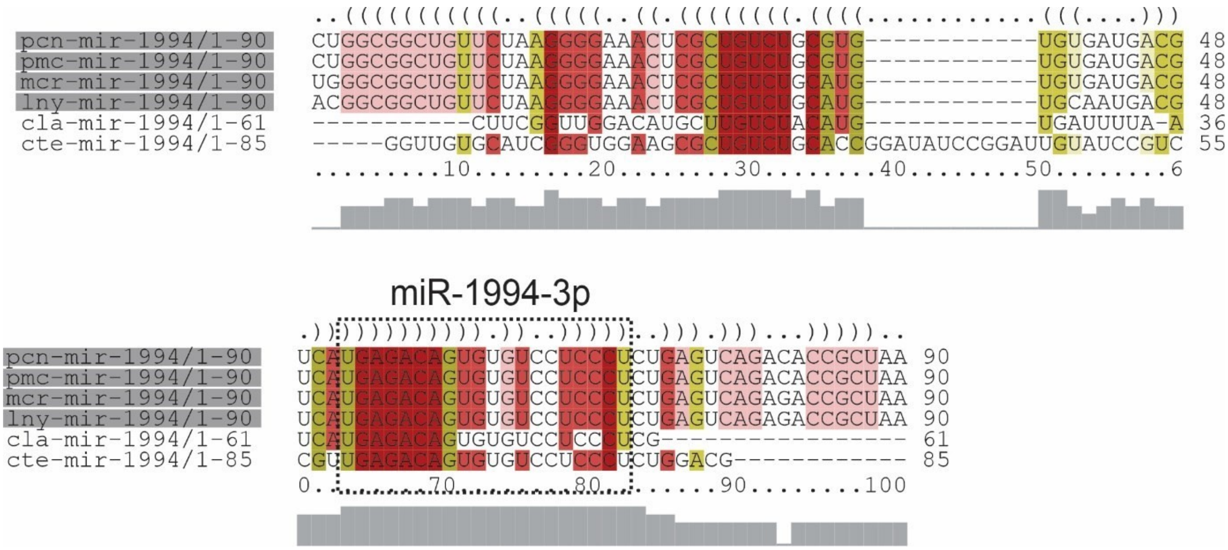


Supplementary figure 26: Secondary structure of mir-1992 in Ampullariidae species and their orthologs; mcr -*M. cornuarietis*, pcn- *P. canaliculata,* hdi - *Haliotis discus hannai*, lgi - *L. gigantea*, adu - *Architeuthis dux*, mle - *M. leonina*, bgl - *B. glabrata.*



Supplementary figure 27: Phylogenetic distribution of mir-1992 in Ampullariidae species and their orthologs.

mir-1994



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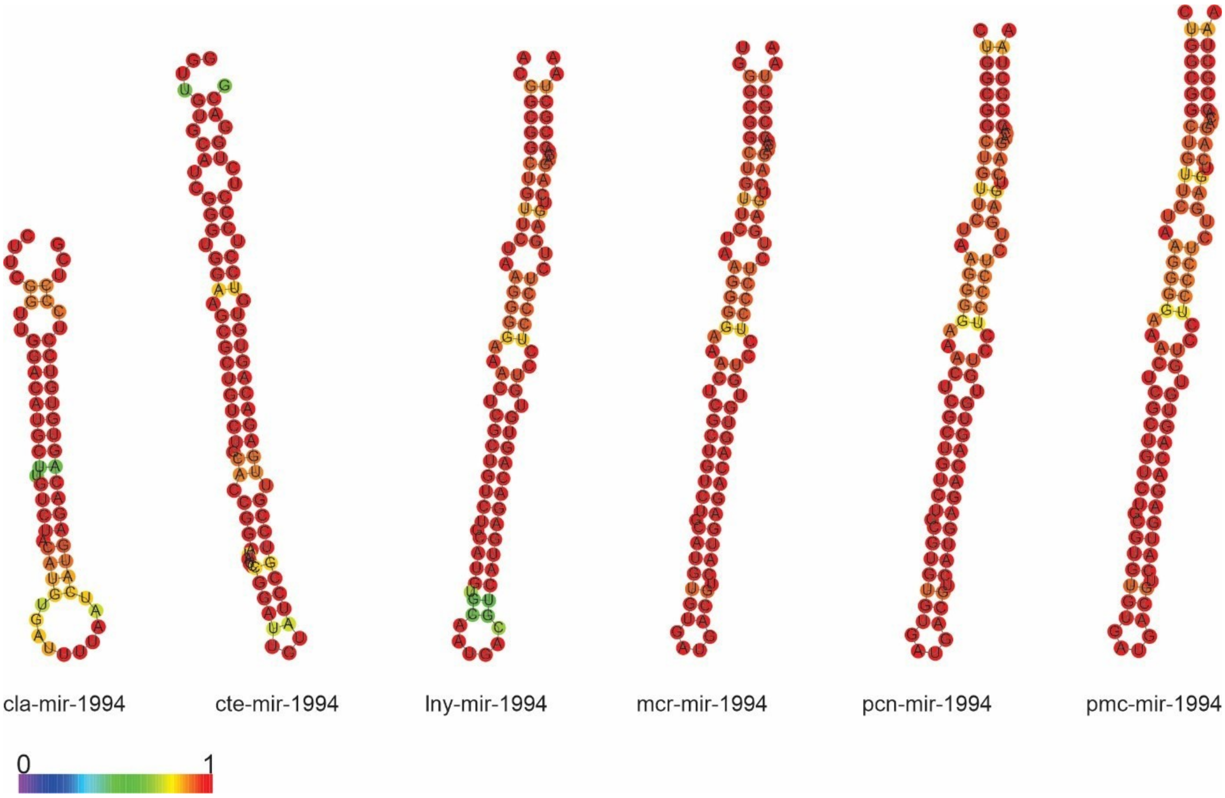
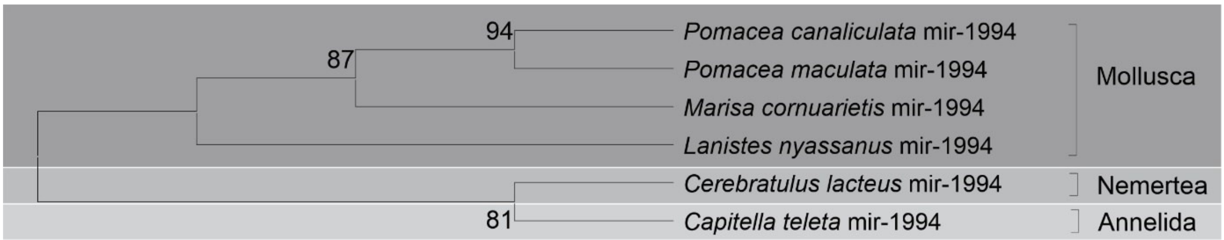
                      ..(((((((((((..(((((..((.((((((((.((((............(((....)))
pcn-mir-1994/1-90     CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUG------------UGUGAUGACG 48
pmc-mir-1994/1-90     CUGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCGUG------------UGUGAUGACG 48
mcr-mir-1994/1-90     UGGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUG------------UGUGAUGACG 48
lny-mir-1994/1-90     ACGGCGGCUGUUCUAAGGGGAAACUCGCUGUCUGCAUG------------UGCAAUGACG 48
cla-mir-1994/1-61     -----------CUUCGGUUGGACAUGCUUGUCUACAUG------------UGAUUUUA-A 36
cte-mir-1994/1-85     -----GGUUGUGCAUCGGGUGGAAGCGCUGUCUGCACCGGAUAUCCGGAUUGUAUCCGUC 55
                      ........10........20........30........40........50.........6

                      miR-1994-3p
                      .))))))))))))).))..)))))..))).)))...))))).. 
pcn-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGACACCGCUAA 90
pmc-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGACACCGCUAA 90
mcr-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGAGACCGCUAA 90
lny-mir-1994/1-90     UCAUGAGACAGUGUGUCCUCCCUCUGAGUCAGAGACCGCUAA 90
cla-mir-1994/1-61     UCAUGAGACAGUGUGUCCUCCCUCG----------------- 61
cte-mir-1994/1-85     CGUUGAGACAGUGUGUCCUCCCUCUGGACG------------ 85
                      0........70........80........90.........100
```

Supplementary figure 28: Alignment of mi-1994 in Ampullariidae species and their orthologs; pcn - *P. canaliculata,* lny - *L. nyassanus,* mcr - *M. cornuarietis*, pmc - *P. maculata,* cte - *C. teleta,* cla - *Cerebratulus lacteus.*



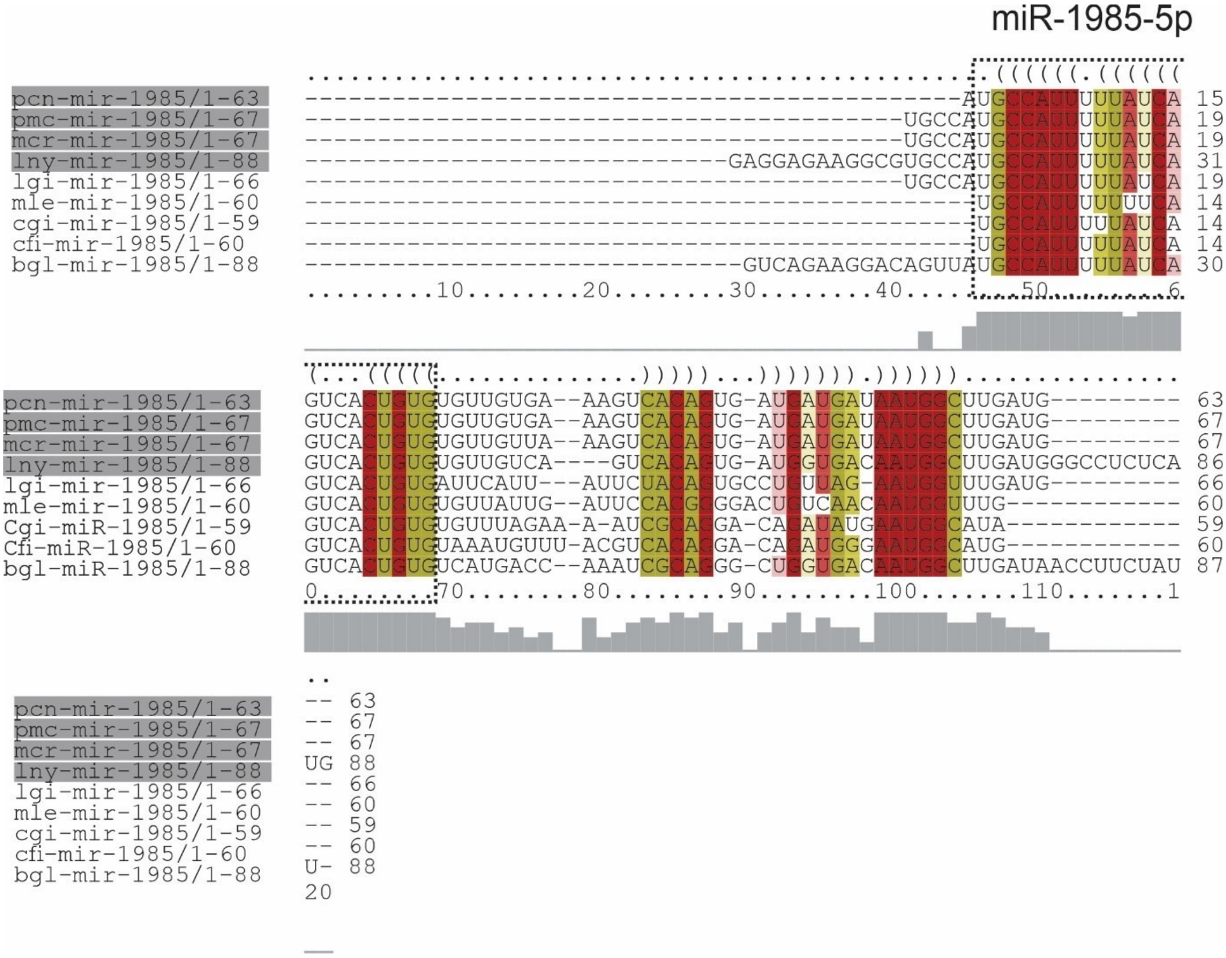
Supplementary figure 29: Secondary structure of mir-1994 in Ampullariidae species and their orthologs; pcn - *P. canaliculata,* lny - *L. nyassanus,* mcr - *M. cornuarietis*, pmc - *P. maculata,* cte - *C. teleta,* cla - *Cerebratulus lacteus.*



Supplementary figure 30: Phylogenetic distribution of mir-1994 in Ampullariidae species and their orthologs.

Mollusc-specific miRNAs

mir-1985



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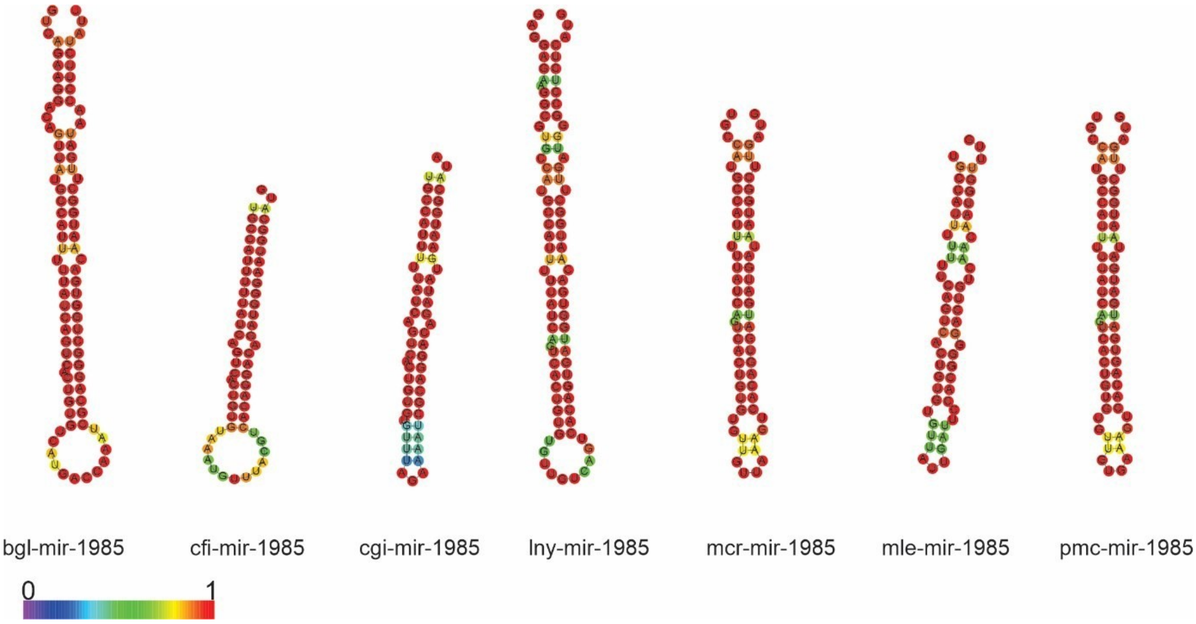
                                                                                  miR-1985-5p
                     .............................................((((((.((((((
pcn-mir-1985/1-63    ---------------------------------------------AUGCCAUUUUUAUCA 15
pmc-mir-1985/1-67    -----------------------------------------UGCCAUGCCAUUUUUAUCA 19
mcr-mir-1985/1-67    -----------------------------------------UGCCAUGCCAUUUUUAUCA 19
lny-mir-1985/1-88    -----------------------------GAGGAGAAGGCGUGCCAUGCCAUUUUUAUCA 31
lgi-mir-1985/1-66    -----------------------------------------UGCCAUGCCAUUUUUAUCA 19
mle-mir-1985/1-60    ----------------------------------------------UGCCAUUUUUUUCA 14
cgi-mir-1985/1-59    ----------------------------------------------UGCCAUUUUUAUCA 14
cfi-mir-1985/1-60    ----------------------------------------------UGCCAUUUUUAUCA 14
bgl-mir-1985/1-88    ------------------------------GUCAGAAGGACAGUUAUGCCAUUUUUAUCA 30
                     .........10........20........30........40........50........6

                     (...(((((...............)))))...))))))).))))))...............
pcn-mir-1985/1-63    GUCACUGUGUGUUGUGA--AAGUCACAGUG-AUGAUGAUAAUGGCUUGAUG--------- 63
pmc-mir-1985/1-67    GUCACUGUGUGUUGUGA--AAGUCACAGUG-AUGAUGAUAAUGGCUUGAUG--------- 67
mcr-mir-1985/1-67    GUCACUGUGUGUUGUUA--AAGUCACAGUG-AUGAUGAUAAUGGCUUGAUG--------- 67
lny-mir-1985/1-88    GUCACUGUGUGUUGUCA----GUCACAGUG-AUGGUGACAAUGGCUUGAUGGGCCUCUCA 86
lgi-mir-1985/1-66    GUCACUGUGAUUCAUU---AUUCUACAGUGCCUGUUAG-AAUGGUUUGAUG--------- 66
mle-mir-1985/1-60    GUCACUGUGUGUUAUUG--AUUCCACGGGGACUGUCAACAAUGGUUUG------------ 60
Cgi-miR-1985/1-59    GUCACUGUGUGUUUAGAA-A-AUCGCAGGA-CAGAUAUGAAUGGCAUA------------ 59
Cfi-miR-1985/1-60    GUCACUGUGUAAAUGUUU-ACGUCACAGGA-CAGAUGGGAAUGGCAUG------------ 60
bgl-miR-1985/1-88    GUCACUGUGUCAUGACC--AAAUCGCAGGG-CUGGUGACAAUGGCUUGAUAACCUUCUAU 87
                     0........70........80........90........100.......110.......1

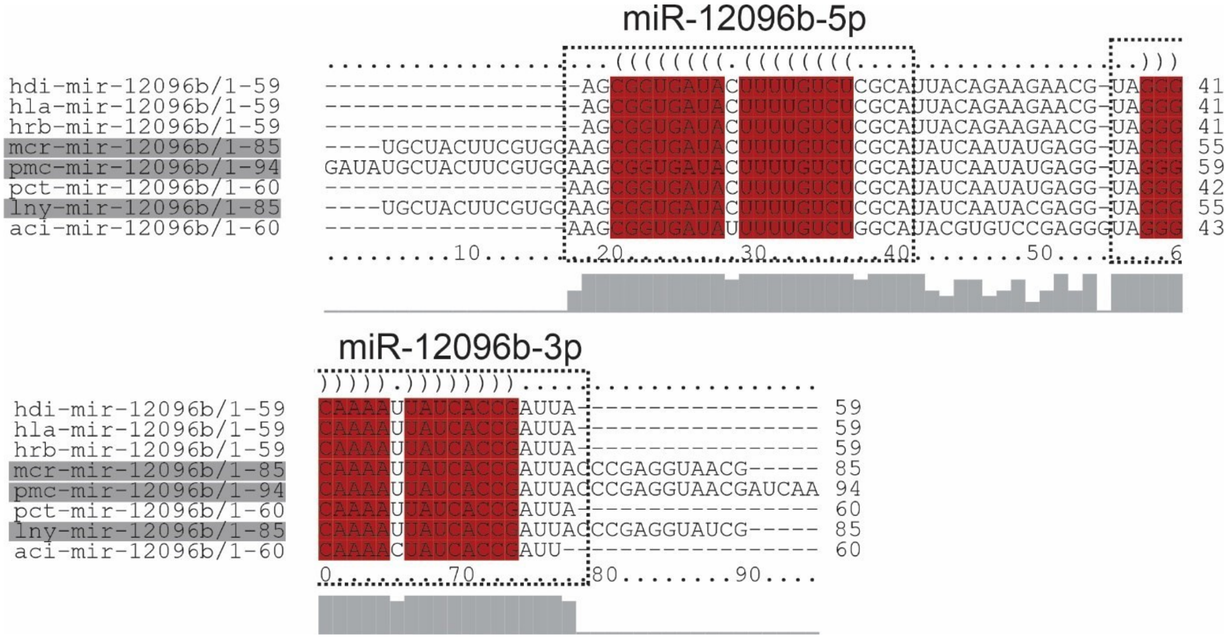
                     ..
pcn-mir-1985/1-63    -- 63
pmc-mir-1985/1-67    -- 67
mcr-mir-1985/1-67    -- 67
lny-mir-1985/1-88    UG 88
lgi-mir-1985/1-66    -- 66
mle-mir-1985/1-60    -- 60
cgi-mir-1985/1-59    -- 59
cfi-mir-1985/1-60    -- 60
bgl-mir-1985/1-88    U- 88
                     20
```

Supplementary figure 31: Alignment of mi- in Ampullariidae species and their orthologs; pcn- *P. canaliculata,* lny - *L. nyassanus,* mcr -*M. cornuarietis*, pmc - *P. maculata,* lgi - *L. gigantea,* mle - *M. leonina,* cgi - *C. gigas,* cfi - *Chlamys farreri,* bgl – *B. glabrata.*



Supplementary figure 32: Secondary structure of mir- in Ampullariidae species and their orthologs; lny - *L. nyassanus,* mcr -*M. cornuarietis*, pmc - *P. maculata,* mle - *M. leonina,* cgi - *C. gigas,* cfi - *Chlamys farreri,* bgl – *B. glabrata*

mir-12096b



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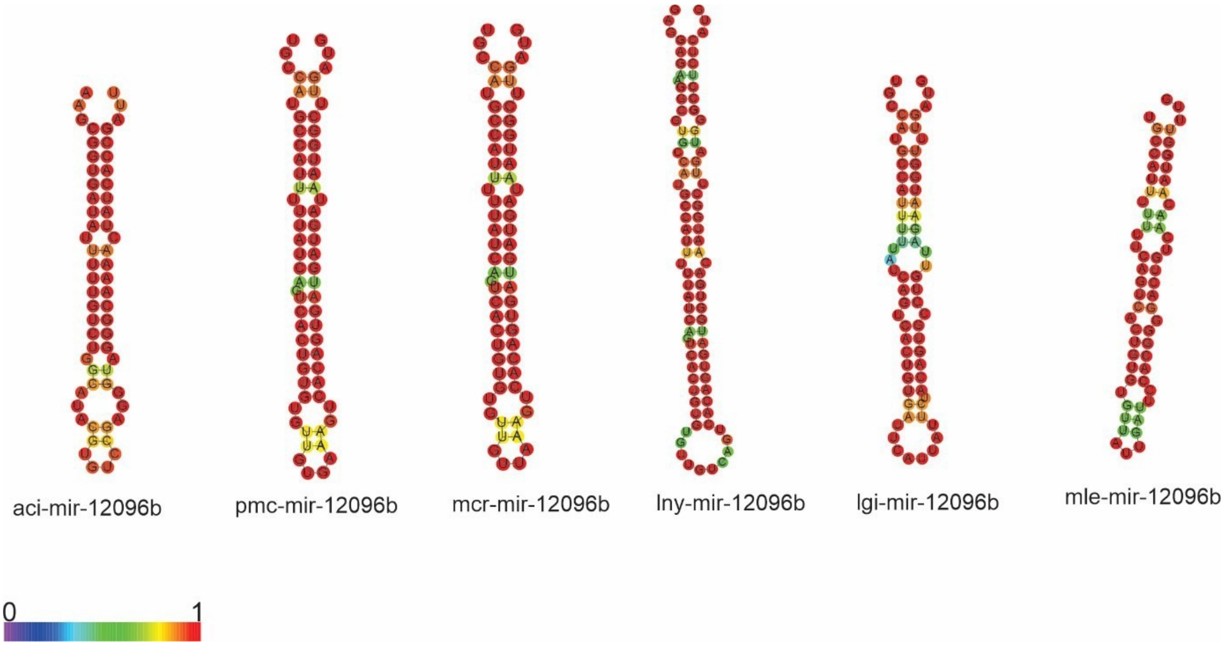
                      .................(((((((.((((((((.......................)))
hdi-mir-12096b/1-59   ------------------AGCGGUGAUACUUUUGUCUCGCAUUACAGAAGAACG-UAGGG 41
hla-mir-12096b/1-59   ------------------AGCGGUGAUACUUUUGUCUCGCAUUACAGAAGAACG-UAGGG 41
hrb-mir-12096b/1-59   ------------------AGCGGUGAUACUUUUGUCUCGCAUUACAGAAGAACG-UAGGG 41
mcr-mir-12096b/1-85   ----UGCUACUUCGUGGAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAGG-UAGGG 55
pmc-mir-12096b/1-94   GAUAUGCUACUUCGUGGAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAGG-UAGGG 59
pct-mir-12096b/1-60   -----------------AAGCGGUGAUACUUUUGUCUCGCAUAUCAAUAUGAGG-UAGGG 42
lny-mir-12096b/1-85   ----UGCUACUUCGUGGAAGCGGUGAUACUUUUGUCUCGCAUAUCAAUACGAGG-UAGGG 55
aci-mir-12096b/1-60   -----------------AAGCGGUGAUAUUUUUGUCUGGCAUACGUGUCCGAGGGUAGGG 43
                      .........10.......20........30........40........50........6

                      ))))).)))))))).......................
hdi-mir-12096b/1-59   CAAAAUUAUCACCGAUUA------------------- 59
hla-mir-12096b/1-59   CAAAAUUAUCACCGAUUA------------------- 59
hrb-mir-12096b/1-59   CAAAAUUAUCACCGAUUA------------------- 59
mcr-mir-12096b/1-85   CAAAAUUAUCACCGAUUACCCGAGGUAACG----- 85
pmc-mir-12096b/1-94   CAAAAUUAUCACCGAUUACCCGAGGUAACGAUCAA 94
pct-mir-12096b/1-60   CAAAAUUAUCACCGAUUA------------------- 60
lny-mir-12096b/1-85   CAAAAUUAUCACCGAUUACCCGAGGUAUCG----- 85
aci-mir-12096b/1-60   CAAAACUAUCACCGAUU-------------------- 60
                      0.........70........80........90....
```

Supplementary figure 33: Alignment of mir-12096b in Ampullariidae species and their orthologs; Aci- *Aplysia californica,* pmc - *P. maculata,* mcr - *M. cornuarietis,* lny - *L. nyassanus,* lgi - *L. gigantea,* hdi - *H. discushannai,* hla - *Haliotis laevigata,* hrb - *Haliotis rubra*



Supplementary figure 34: Secondary structure of mir-12096b in Ampullariidae species and their orthologs; Aci- *Aplysia californica,* pmc - *P. maculata,* mcr - *M. cornuarietis,* lny - *L. nyassanus,* lgi - *L. gigantea,* mle – *M. leonina.*