

UNIVERSIDADE FEDERAL DE UBERLÂNDIA
INSTITUTO DE CIÊNCIAS AGRÁRIAS
PROGRAMA DE PÓS-GRADUAÇÃO EM AGRONOMIA

HUGO GABRIEL PERES

ASSOCIATION OF SILICON SUPPLY AND ARBUSCULAR MYCORRHIZAL FUNGI
INOCULATION IN SWEET CORN SUBJECTED TO DIFFERENT WATER REPOSITION
LEVELS

UBERLÂNDIA

2023

HUGO GABRIEL PERES

ASSOCIATION OF SILICON SUPPLY AND ARBUSCULAR MYCORRHIZAL FUNGI
INOCULATION IN SWEET CORN SUBJECTED TO DIFFERENT WATER REPOSITION
LEVELS

Dissertação apresentada ao Programa de
Pós-graduação em Agronomia da
Universidade Federal de Uberlândia como
requisito parcial para a obtenção do título
de ‘Mestre em Agronomia’.

Orientador: Prof. Dr. Douglas José
Marques.

UBERLÂNDIA

2023



UNIVERSIDADE FEDERAL DE UBERLÂNDIA

Secretaria da Coordenação do Programa de Pós-Graduação em Agronomia

Rodovia BR 050, Km 78, Bloco 1CCG, Sala 206 - Bairro Glória, Uberlândia-MG, CEP 38400-902

Telefone: (34) 2512-6715/6716 - www.ppga.iciag.ufu.br - posagro@ufu.br



ATA DE DEFESA - PÓS-GRADUAÇÃO

Programa de Pós-Graduação em:	Agronomia				
Defesa de:	Dissertação de Mestrado Acadêmico, 019/2022, PPGAGRO				
Data:	Oito de dezembro de dois mil e vinte e dois	Hora de início:	13:30	Hora de encerramento:	16:20
Matrícula do Discente:	12012AGR021				
Nome do Discente:	Hugo Gabriel Peres				
Título do Trabalho:	Association of arbuscular mycorrhizal fungi inoculation and Silicon supply in sweet corn subjected to different water reposition levels				
Área de concentração:	Produção Vegetal				
Linha de pesquisa:	Produção Vegetal em Áreas de Cerrado				

Reuniu-se por videoconferência, a Banca Examinadora, designada pelo Colegiado do Programa de Pós-graduação em Agronomia, assim composta: Professores Doutores: Lucas Carvalho Basílio de Azevedo - UFU; Eusímio Felisbino Fraga Júnior - UFU; Adriano Valentim Diotto - UFLA; Douglas José Marques - UFU orientador(a) do(a) candidato(a).

Iniciando os trabalhos o(a) presidente da mesa, Dr(a). Douglas José Marques, apresentou a Comissão Examinadora e o candidato(a), agradeceu a presença do público, e concedeu ao Discente a palavra para a exposição do seu trabalho. A duração da apresentação do Discente e o tempo de arguição e resposta foram conforme as normas do Programa.

A seguir o senhor(a) presidente concedeu a palavra, pela ordem sucessivamente, aos(às) examinadores(as), que passaram a arguir o(a) candidato(a). Ultimada a arguição, que se desenvolveu dentro dos termos regimentais, a Banca, em sessão secreta, atribuiu o resultado final, considerando o(a) candidato(a):

Aprovado.

Esta defesa faz parte dos requisitos necessários à obtenção do título de Mestre.

O competente diploma será expedido após cumprimento dos demais requisitos, conforme as normas do Programa, a legislação pertinente e a regulamentação interna da UFU.

Nada mais havendo a tratar foram encerrados os trabalhos. Foi lavrada a presente ata que após lida e achada conforme foi assinada pela Banca Examinadora.



Documento assinado eletronicamente por **Eusímio Felisbino Fraga Junior, Professor(a) do Magistério Superior**, em 08/12/2022, às 16:30, conforme horário oficial de Brasília, com fundamento no art. 6º, § 1º, do [Decreto nº 8.539, de 8 de outubro de 2015](#).



Documento assinado eletronicamente por **Lucas Carvalho Basílio de Azevedo, Professor(a) do Magistério Superior**, em 08/12/2022, às 16:35, conforme horário oficial de Brasília, com fundamento no art. 6º, § 1º, do [Decreto nº 8.539, de 8 de outubro de 2015](#).



Documento assinado eletronicamente por **Adriano Valentim Diotto, Usuário Externo**, em 08/12/2022, às 16:57, conforme horário oficial de Brasília, com fundamento no art. 6º, § 1º, do [Decreto nº 8.539, de 8 de outubro de 2015](#).



Documento assinado eletronicamente por **Douglas José Marques, Professor(a) do Magistério Superior**, em 08/12/2022, às 19:44, conforme horário oficial de Brasília, com fundamento no art. 6º, § 1º, do [Decreto nº 8.539, de 8 de outubro de 2015](#).



A autenticidade deste documento pode ser conferida no site https://www.sei.ufu.br/sei/controlador_externo.php?acao=documento_conferir&id_orgao_acesso_externo=0, informando o código verificador **4118052** e o código CRC **8479A498**.

Dados Internacionais de Catalogação na Publicação (CIP)
Sistema de Bibliotecas da UFU, MG, Brasil.

P437a Peres, Hugo Gabriel, 1997-
2023 Association of silicon supply and arbuscular mycorrhizal fungi inoculation in sweet corn subjected to different water reposition levels [recurso eletrônico] / Hugo Gabriel Peres. - 2023.

Orientador: Douglas José Marques.
Dissertação (Mestrado) - Universidade Federal de Uberlândia, Programa de Pós-Graduação em Agronomia.
Modo de acesso: Internet.
Disponível em: <http://doi.org/10.14393/ufu.di.2023.8021>
Inclui bibliografia.
Inclui ilustrações.

1. Agronomia. I. Marques, Douglas José, 1980-, (Orient.). II. Universidade Federal de Uberlândia. Programa de Pós-Graduação em Agronomia. III. Título.

CDU: 631

André Carlos Francisco
Bibliotecário - CRB-6/3408

AGRADECIMENTOS

O presente trabalho foi realizado com o apoio do Conselho Nacional de Desenvolvimento Científico e Tecnológico – Brasil. Desta forma, agradeço ao CNPq pela bolsa de estudos concedida nos primeiros meses do curso e também pelas demais formas de apoio ao projeto.

Aos meus pais, irmão, demais familiares e amigos.

Ao meu orientador, Prof. Douglas José Marques.

Aos professores Ana Paula Nogueira, Denise Garcia, Fábio Janoni, Flávio Lemes, Gilberto Mendes, João Paulo Ribeiro, Marcus Vinicius Sampaio e Mayara Stanger por transferirem seus conhecimentos nas aulas da pós-graduação.

Àqueles que, de alguma forma, contribuíram para o desenvolvimento do trabalho, em especial aos professores Eusímio Fraga Jr., Sidney Stürmer, Edmar de Melo, Denis Coelho, Lucas Basílio e Ana Carolina Siquieroli, aos alunos de graduação Bruno Garcia, Gustavo Lemos, Mateus Tondini, Marcello Isquizado, Maria Clara e Pedro Bataglia, ao pós-graduando João Paulo Borges e aos técnicos de laboratório Daiane e Daniel.

Deixo aqui o meu muito obrigado!

RESUMO

PERES, Hugo Gabriel. **Associação de silício e inoculação de fungos micorrízicos arbusculares em milho-doce submetido a diferentes níveis de reposição hídrica.** Orientador: Douglas José Marques. Dissertação (Mestrado em Agronomia) – Universidade Federal de Uberlândia, Uberlândia, 2023.

O estresse de seca afeta processos-chave do metabolismo vegetal, sendo um dos principais problemas para a segurança alimentar mundial. Fungos micorrízicos arbusculares (FMA's) e Silício (Si) são duas estratégias amplamente reconhecidas por mitigar o estresse de seca em plantas. Entretanto, não há informação acerca da sua associação em plantas de milho-doce. Levantou-se a hipótese de que a associação de FMAs e Si poderia ser uma forma de mitigar o estresse de seca em milho-doce. Um experimento foi conduzido em casa-de-vegetação sob esquema fatorial triplo (5x2x2), incluindo 5 níveis de reposição hídrica (40, 60, 80, 100 e 120% de reposição da capacidade de campo), dois níveis de inoculação de FMAs (presença/+FMA e ausência/-FMA) e 2 níveis de fornecimento de Si (presença/+Si e ausência/-Si). Foram avaliados parâmetros de crescimento, produção, fotossíntese (SPAD e Fv/Fm), teor de Si nas folhas e colonização micorrízica. A restrição hídrica levou a redução do desempenho do milho-doce. Em condições de estresse de seca severo (40% de reposição), a inoculação de FMAs aumentou o crescimento (parte aérea e raiz), e o Si aumentou o índice SPAD. De forma geral, os FMAs e o Si, isolados ou em associação, aumentaram os índices fotossintéticos SPAD e Fv/Fm. Em plantas em que não foi fornecido Si, a inoculação dos FMAs aumentou o teor de Si na planta, mas isto não foi observado em plantas em que o Si foi fornecido via solo. Em condições de seca severa, a inoculação de FMAs e o fornecimento de Si podem ser formas de mitigar os efeitos deletérios da seca. No entanto, frente a complexidade das interações planta-Si-FMAs-ambiente, novas pesquisas devem ser conduzidas visando estudar melhor estes efeitos.

Palavras-chave: *Zea mays* L.; seca; simbiose; silicato de cálcio.

ABSTRACT

PERES, Hugo Gabriel. **Association of silicon supply and arbuscular mycorrhizal fungi inoculation in sweet corn subjected to different water reposition levels.** Supervisor: Douglas José Marques. Dissertation (Master's in Agronomy) – Universidade Federal de Uberlândia, Uberlândia, 2023.

Drought affects plant key process, being a main constraint for food security worldwide. Arbuscular mycorrhizal fungi (AMF) and Silicon (Si) are well-known strategies to mitigate drought stress in plants. However, lacks information regarding it is association in sweet corn plants. We hypothesized that Si and AMF association could be a potential way to mitigate drought stress in sweet corn. A greenhouse experiment was conducted under a triple factorial (5x2x2) scheme, including five water reposition levels (40, 60, 80, 100 and 120% of reposition), two AMF inoculation levels (presence/+AMF, and absence/-AMF) and two Silicon (Si) supply levels (presence/+Si and absence/-Si). We evaluated plant growth, yield, photosynthesis parameters (SPAD, and Fv/Fm), Si content in leaves and mycorrhizal colonization. Water restriction led to reduction in sweet corn performance. In severe drought stress conditions (40% of reposition), AMF inoculation improved growth (shoot and root) and Si supply improved SPAD. Overall, AMF and Si, isolate or in association, increased SPAD and efficiency of PSII. In non-Si-supplied plants, AMF increased Si content in leaves, but no AMF-mediated uptake of Si was found in Si-supplied plants. In severe drought conditions, AMF and Si may be potential ways to mitigate the defective effects of drought. However, in front of the complexity of AMF-plant-Si-environment interactions, further research needs to be conducted in order to investigate better these effects.

Keywords: *Zea mays* L.; drought; symbiosis; calcium silicate.

SUMÁRIO

RESUMO.....	4
ABSTRACT	5
1 INTRODUCTION	5
2 MATERIAL AND METHODS	6
2.1 Overview of the plant growth experiment.....	6
2.2 Soil traits and Silicon supply	7
2.3 Arbuscular mycorrhizal fungi inoculum.....	8
2.4 Irrigation management	9
2.5 Plant growth and sweet corn yield	11
2.6 Photosynthesis-related measurements.....	11
2.7 Silicon analysis in plant tissue	11
2.8 Mycorrhizal colonization	12
2.9 Statistical analysis.....	12
3 RESULTS.....	12
3.1 Agronomic assessments.....	12
3.2 Silicon content in plant.....	15
3.3 Mycorrhizal colonization	16
3.4 Photosynthesis.....	17
3.5 Principal components analysis (PCA).....	17
4 DISCUSSION.....	18
5 CONCLUSIONS.....	23
REFERENCES	24

1 INTRODUCTION

Global demand for food supply is increasing, considering that Earth's population recently reached 8 billion people. However, drought is becoming a large problem for crop production and economy as a result of global climate changes and represents a major constraint on global food security (BRÁS *et al.*, 2021; FAHAD *et al.*, 2017; LIEBER *et al.*, 2022; NAUMANN *et al.*, 2021). Drought severely inhibits plant growth by affecting key metabolic processes, such as nutrient acquisition and photosynthesis (SELEIMAN *et al.*, 2021). Therefore, agricultural practices to counterbalance the drought effects, such as the addition of nutrients and microbial inoculants that help plants cope with drought should be considered in crop production to reduce plant stress and crop losses.

Silicon (Si) is a nutrient commonly used for improving plant resistance to drought. This quasi-essential metalloid is responsible for altering the cell wall composition, turning it more resistant. The absorption of Si from soil and distribution in plant organs results in amorphous silica deposition under the leaf cuticle, which comprises an important trait for improving leaf-atmosphere water relations (LUYCKX *et al.*, 2017; MALIK *et al.*, 2021). Moreover, Si was also attributed to play roles in photosynthesis and plant antioxidant responses (MALIK *et al.*, 2021; WANG *et al.*, 2021).

Plants have differences in the capacity to accumulate Si, ranging from less than 0.1% from up to 10% of dry weight (EPSTEIN, 1999) and the use of Si has been extensively studied in grasses (*Poaceae*) because of their high capacity of Si accumulation (PUTRA *et al.*, 2020). Si beneficial roles are closely related with the plant capacity to accumulate Si (YAN *et al.*, 2018). Intense weathering processes in tropical acid soils results in soils with low levels of available Si to plants (BUOL, 2009; CAMARGO *et al.*, 2013; DE TOMBEUR, 2021). To bypass this, calcium silicate is applied during crop production as a source of Si and to correct soil acidity (BIANCHINI; MARQUES, 2019; CAMARGO *et al.*, 2013; MARQUES *et al.*, 2022).

Microorganisms also help plants to cope with drought and represent sustainable tools for modulating crop tolerance under stressed conditions (KOUR *et al.*, 2022). Arbuscular mycorrhizal fungi (AMF) are well known to establish the arbuscular mycorrhizal symbiosis with ca. 72% of plant species (BRUNDRETT; TEDERSOO, 2018). AMF colonize plant root cortex and extend their hyphae into the soil, forming the extra-radicular mycelium, which explore the bulk soil for water and nutrients. AMF-plant symbiosis plays several ecological

roles that benefits both the plant host and the soil environment, by improving plant water and nutrient uptake, promoting plant tolerance to drought and other abiotic and biotic stresses, and contributing to soil aggregation (GIANINAZZI *et al.*, 2010). Since there is variation in response of soil and plant variables for each AMF strain, it is interesting to apply a consortium of species to guarantee the beneficial effects of the inoculum (SHARMA *et al.*, 2009).

The addition of microbial inoculants associated with beneficial nutrients, such as Si and AMF, is emerging as a sustainable alternative for mitigate drought stress in crops (VEGA *et al.*, 2021). The use of Si and mycorrhizal symbiosis as a strategy to mitigate drought stress have been studied for strawberry (MORADTALAB *et al.*, 2019), rice (DAS *et al.*, 2021), licorice (HAGHIGHI *et al.*, 2022; HAGHIGHI; SAHARKHIZ, 2022), and Indian mustard (SRIVASTAVA *et al.* 2022). It is important to highlight that the most pronounced beneficial effects of Si occur in accumulator plants (YAN *et al.*, 2018) and all the plants already used to explore the AMF-Si association as a tool to manage drought stress, with exception of rice, are not good Si accumulators. Thus, studies with the AMF-Si association should consider Si accumulating plants as the Poaceae.

Sweet corn (*Poaceae*) is a special type of maize that is used only for human consumption. The crop is high input and high value. Although drought is the main abiotic stress affecting sweet corn (REVILLA *et al.*, 2021), there is no report on the effects of the association of AMF inoculation and Si supply in sweet corn under drought stress. Thus, we hypothesized that the application of Si and a consortium of six AMF species could be a potential way to mitigate drought stress in sweet corn. To check the hypothesis, we assessed plant growth traits, yield, and photosynthesis parameters. Additionally, we assessed arbuscular mycorrhizal colonization in roots and Si content in leaves.

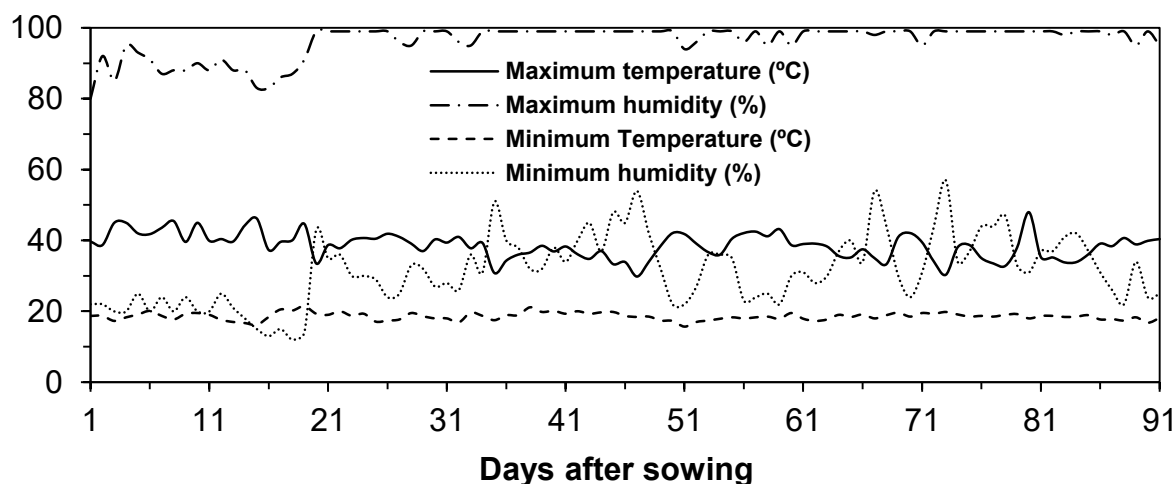
2 MATERIAL AND METHODS

2.1 Overview of the plant growth experiment

A greenhouse experiment was conducted at the Federal University of Uberlândia (UFU) in Monte Carmelo, Brazil. From November 24th, 2021, to March 24th, 2022, plants of sweet corn cv. FT 2015 (single-cross commercial hybrid) were grown in 25-liter pots containing 21.5 kg of soil. On average, the environment inside the greenhouse had a maximum temperature of 38.6 °C (29.8-48.0 °C) and minimum temperature of 18.5 °C (21.4-15.7 °C). Maximum

humidity values were always higher than 80% (80-99%) and minimum humidity ranging from 12-54% (Figure 1).

Figure 1 - Environmental conditions inside the greenhouse during the experiment.



Source: The author.

The experiment was set up in a randomized block design with four replications. Treatments comprised a complete factorial scheme (2x2x5): 2 levels of Si (with and without Si supply as soil acidity corrective) x 2 levels of AMF inoculation (with: +AMF and without: -AMF inoculation) x 5 water reposition levels (40%: severe drought; 60%: moderate drought; 80%: light drought; 100%: field capacity reposition; 120%: light water excess).

2.2 Soil traits and Silicon supply

The soil used to fill the pots correspond to a 0-20cm layer of a Ferralsol (World Reference Base - FAO), collected in a non-cultivated area, adjacent to native Cerrado vegetation at Monte Carmelo, Minas Gerais, Brazil (18°32'33.76"S; 47°29'10.65"O; 799 m.a.s.l.). Soil chemical and textural characterization (Table 1) shows a low natural fertility and clayey soil with low silt content, typical of Cerrado Biome (BUOL, 2009).

The dose of soil correctives was calculated as recommended by van Raij and Cantarella (1997) and adapted by Bianchini and Marques (2019) for corn plants. Si was supplied (+Si) using calcium silicate (CaO: 12%; SiO₂: 78%; ENV: 86%) as source (0.787 g dm⁻³). This dose represents a potential supply of 287 mg dm⁻³ of Si (or 5.5 g pot⁻¹). In control pots (-Si), an equivalent dose (0.714 g dm⁻³) of dolomitic limestone (CaO: 38.5%; MgO: 10.9%; ENV: 95%)

was applied, considering the effective neutralizing values (ENV) of each corrective. After the application of Silicate and limestone, the pots received water until reach field capacity and were incubated for 45 days to promote acidity correction reactions before fertilization.

Table 1 - Chemical and textural traits of soil before correction and fertilization.

Chemical ¹	Unit	Value
pH in H ₂ O	-log [H ⁺]	5.20
pH in CaCl ₂	-log [H ⁺]	4.70
Phosphorus (Melich-1)	mg dm ⁻³	5.40
Exchangeable Potassium (K ⁺)	cmol _c dm ⁻³	0.10
Exchangeable Calcium (Ca ²⁺)	cmol _c dm ⁻³	0.18
Exchangeable Magnesium (Mg ²⁺)	cmol _c dm ⁻³	0.04
Sulphate (S-SO ₄ ²⁻)	mg dm ⁻³	1.00
Exchangeable Aluminium (Al ³⁺)	cmol _c dm ⁻³	0.26
Potential acidity (H ⁺ +Al ³⁺)	cmol _c dm ⁻³	2.20
Sum of bases (Ca ²⁺ + Mg ²⁺ + K ⁺)	cmol _c dm ⁻³	0.31
Cation exchange capacity (T)	cmol _c dm ⁻³	2.52
Base saturation (V)	%	12.00
Aluminum saturation (m)	%	45.00
Organic matter (O.M.)	%	1.40
Silicon (Si) ¹	mg dm ⁻³	8.30
Copper (Cu)	mg dm ⁻³	1.00
Iron (Fe)	mg dm ⁻³	16.00
Boron (B)	mg dm ⁻³	0.13
Manganese (Mn)	mg dm ⁻³	7.70
Zinc (Zn)	mg dm ⁻³	0.70
Physical	Unit	Value
Clay ²	%	64.50
Silt ²	%	2.50
Sand ²	%	33.00
Global density ³	g cm ⁻³	1.12

¹Determined with CaCl₂. 2H₂O 0,01 mol L⁻¹; ²Determined with the pipette method ³Determined in an undisturbed soil sample.

Source: The author.

Fertilization followed the recommendations of Novais *et al.* (1991). Two days before sowing, the base fertilization was mixed in the soil. Three top-dressing fertilizations of potassium (K) and nitrogen (N) were performed at 10, 20, and 30 days after sowing.

2.3 Arbuscular mycorrhizal fungi inoculum

AMF isolates were obtained at the International Culture Collection of Glomeromycota (www.furb.br/cicg) at the Universidade Regional de Blumenau (FURB). Inoculum for each

isolate was obtained by mixing stock cultures (containing spores, hyphae and mycorrhizal root pieces) with a sterilized substrate of a soil: sand mix (1:1, v/v, pH 6.0), placed in 0.8 Kg plastic pots and seeded with *Sorghum bicolor* L. as the host. Pots containing only the sterilized soil:sand mix were established to obtain appropriate non-mycorrhizal inoculum used in the plant growth experiment. Before sowing, 12g of AMF inoculum (Table 2) or control were applied in the depth 5 cm below the seeds.

Table 2 - AMF species with their respective isolate code used to inoculate sweet corn grown in different water reposition levels and Si supply.

Strain	Concentration in inoculum (spores g ⁻¹)
<i>Gigaspora albida</i> PRN200A	183
<i>Dentiscutata heterogama</i> MGR610A	227
<i>Rhizophagus clarus</i> SCT720A	22
<i>Rhizophagus intraradices</i> SCT736J	152
<i>Acaulospora mellea</i> SCT063B	14
<i>Acaulospora longula</i> PNB101A	144
Total	742 spores g ⁻¹

Source: The author.

2.4 Irrigation management

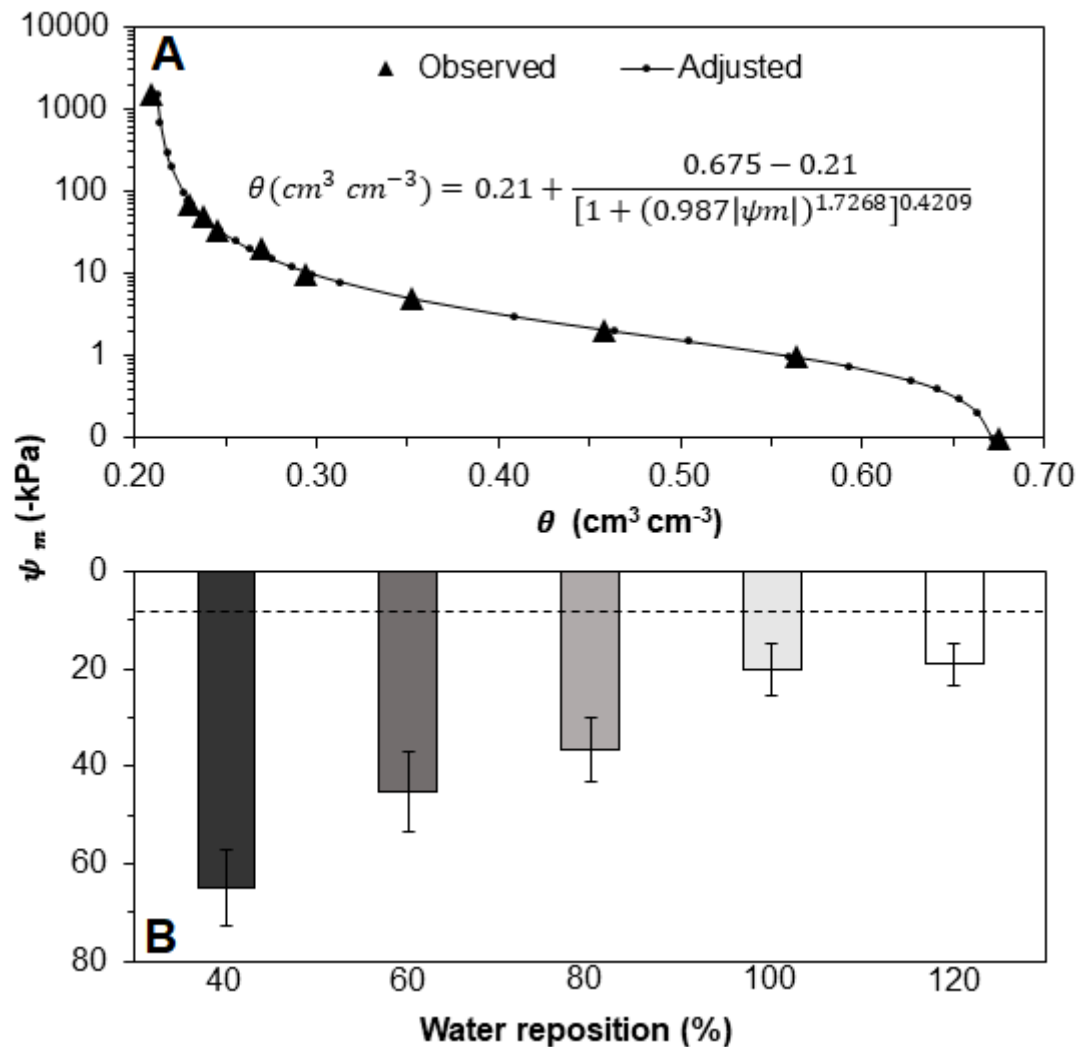
Irrigation was performed self-compensating drippers (Netafim PCJ-CNL. working pressure range: 70-400 kPa), micro tubes, and stakes. A digital controller with a 1-minute resolution (RainBird ESP-RZXe) was used to open/close the solenoid valve and turn on/off the pump system. Before the experiment starts, end-of-line pressure and statistical uniformity coefficient of system were determined as 220 kPa and 96%, respectively.

Soil water retention curve was obtained from adjustment of physical-hydric data of an undisturbed sample to van Genuchten (1980) equation (Figure 2A). The field capacity of tropical soils usually ranges from -6 to -10 kPa (Andrade and Stone 2011). Field capacity of this soil were estimated at 0.3124 cm³ cm⁻³ (-8kPa) by a pot-based approach: soil was saturated at end of day and soil matric potential measured after water percolation with minimum evapotranspiration (morning of next day).

Application of water reposition treatments started 30 days after sowing, 12 tensiometers were installed at 15cm depth (soil depth in pots was 30cm) for monitoring soil matric potential

in all reposition levels: 40% (2), 60% (2), 80% (2), 100% (4), and 120% (2). Matric potential readings were performed daily, and the critical value adopted for irrigation was -20kPa in the reference level (100% of reposition). The amount of water applied in 100% reposition corresponds to field capacity reposition, and the same irrigation time was adopted for all reposition levels. The reposition levels were achieved by variation of flow rate in pots. as follows: 40% (2 L h⁻¹ pot⁻¹), 60% (3 L h⁻¹ pot⁻¹), 80% (4 L h⁻¹ pot⁻¹), 100% (5 L h⁻¹ pot⁻¹), and 120% (6 L h⁻¹ pot⁻¹). Means of matric potential before irrigation were -65, -45, -37, -20, and -19 kPa for 40, 60, 80, 100, and 120% reposition levels, respectively (Figure 2B).

Figure 2 - Soil water retention curve adjusted by van Genuchten (1980) equation (A) and means of nine weeks monitoring of soil matric potential (ψ_m) before irrigations in the five water reposition levels (B). The dotted line represents the matric potential at the field capacity (-8 kPa).



Source: The author.

2.5 Plant growth and sweet corn yield

After the first blooming signal, days to male flowering (DMF) were recorded daily. Plant height (PH) were measured after tasseling with a tape measure, corresponding from the distance between the soil level and the top of tassel. Fully expanded ears were harvested and husks removed. The ears without husks were weighted to obtain yield of fresh-market sweet corn (YLD). After harvest, we hold irrigation until the soil drying, and the roots were washed and placed to dry at room temperature for 1 week. Shoots were dried in an oven at 60°C until reached constant mass. Dry roots and shoots were placed separately in a 0.1 g resolution balance to obtain root (RDM) and shoot (SDM) dry mass.

2.6 Photosynthesis-related measurements

At 60 days after sowing, Soil Plant Analysis Development (SPAD) index was measured with a SPAD-502 (Konica Minolta, Japan) chlorophyll meter in three leaves from the middle third of each plant. Three days after SPAD measurements, chlorophyll fluorescence analysis was performed on the leaves of each treatment ($n=3$ for each treatment) using a modulated fluorescence imaging apparatus. Handy Fluorcam PSI (Photo Systems Instrument, Czech Republic) to obtain the F_v/F_m (maximum PSII quantum yield in the dark-adapted state, where $F_v = F_m - F_0$). The maintenance of the integrity of the electron chains and the capacity of the plants for photosynthesizing were demonstrated after a dark-adapted (30 minutes). Both SPAD and fluorescence analysis were performed from 8:00 to 10:00 A.M. to avoid environmental disruptions.

2.7 Silicon analysis in plant tissue

For determination of Si content in plant, leaves were harvested at flowering stage, washed to remove soil particles, dried, central nervure removed. grinded and sieved (0.5 mm). Digestion of organic fraction was performed with H₂O₂ and NaOH. Silicon content in leaves [Si] was determined by absorbance readings at 410nm after colorimetric reaction with molibdate (ELLIOT; SNYDER, 1991; KORNDÖRFER *et al.*, 2004). Results were expressed in mg of Si per dry mass (mg g⁻¹).

2.8 Mycorrhizal colonization

At harvest time, roots were collected in two points (a 2x2cm slice) per pot, distanced 5 cm from stem, at 5 cm depth using a blade of stainless steel. Root samples were washed with tap water to remove soil particles and kept on falcon tubes containing ethanol (70%) until clear and staining processes. For clarification, roots were kept in KOH (10%) at room temperature for 16 h, followed by an additional step at 60°C for 10 min with a renewed solution. Then, fungi structures within roots were stained with blue ink (Parker™) (VIERHEILIG *et al.*, 1998). Stained roots were preserved in acid lactoglycerol and the mycorrhizal colonization (%) were determined by the grid-line intersection method (GIOVANETTI; MOSSE, 1980).

2.9 Statistical analysis

In order to check the assumptions of analysis of variance (ANOVA), data was submitted to Levene, Kolmogorov-Smirnov/Lilliefors, and Tukey tests for homogeneity of variances, normality of residuals, and additivity of treatments and blocks, respectively ($\alpha=0.01$). Then, a three-way ANOVA ($\alpha=0.05$) was applied to check simple factor and interactive effects.

Significant differences in means of AMF and Si factors were also checked with Tukey HSD test and when suitable, regression analysis was used to study effects of water reposition ($\alpha=0.05$). When triple interaction detected, means were clustered with Scott-Knott test ($\alpha=0.05$). In case of violation of any assumption, data (x) were transformed to log (x) to run ANOVA, but original data (means and standard deviations) are shown in figures. In addition, principal components analysis (PCA) was performed with standardized data.

3 RESULTS

3.1 Agronomic assessments

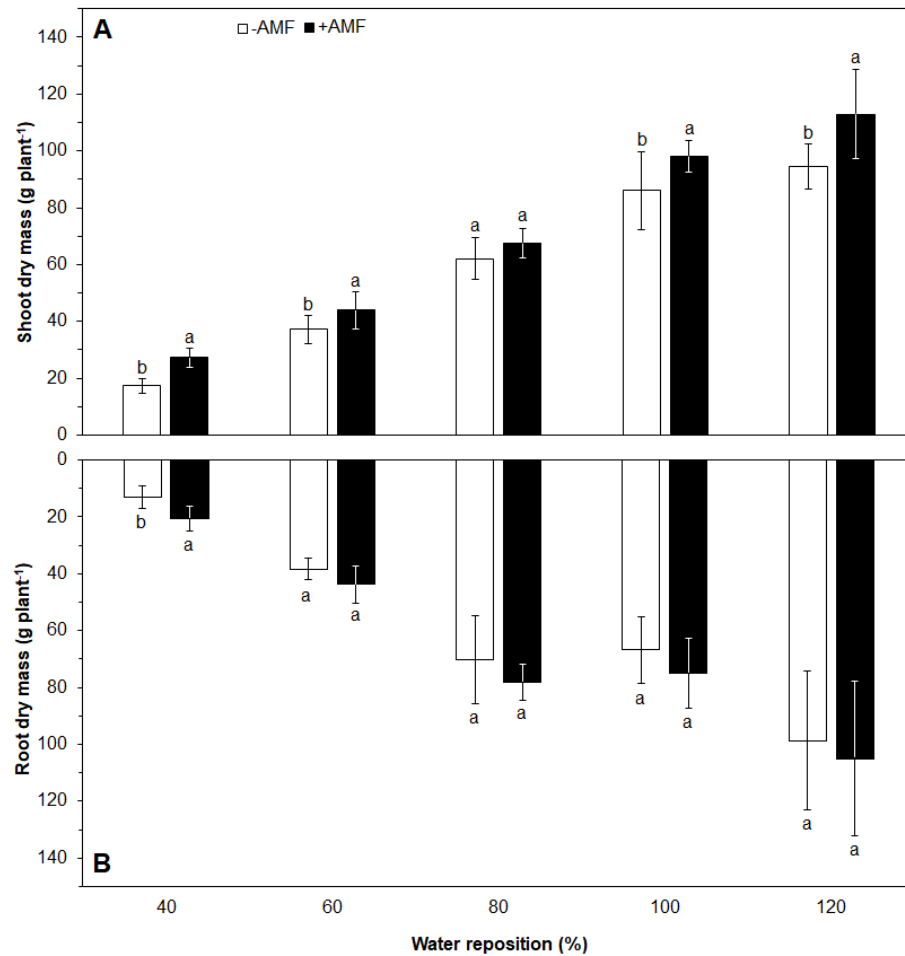
Drought stress reduced most of the agronomic parameters evaluated, with exception to days to male flowering. Overall, plants inoculated with AMF were significantly higher ($p=0.049$) than non-inoculated plants (data not shown). For shoot dry mass, a significant interaction was found between AMF and water reposition. Except for the 80% reposition level

($p=0.125$), AMF inoculation promoted higher shoot dry mass, reaching the highest promotion in 40% reposition level (56% in comparison to the non-inoculated plants). For 60, 100, and 120% water reposition levels, the relative increase promoted by inoculation were 18, 14, and 19%, respectively. Taking the 40% (severe drought) and the 100% (control) reposition levels as references, drought stress reduced 79.8% of shoot biomass in non-inoculated plants versus 72.3% in inoculated plants (Figure 3A).

AMF inoculation also promoted significant increases in root growth, but only in severe drought (40% of reposition). In this situation, the root system of inoculated plants accumulated 56% more biomass than non-inoculated plants (Figures 3B and 4). Considering 40 and 100% reposition levels, a pattern very similar to shoots was observed in roots: drought stress reduced 80.4% of root biomass in non-inoculated plants versus 72.5% in inoculated plants.

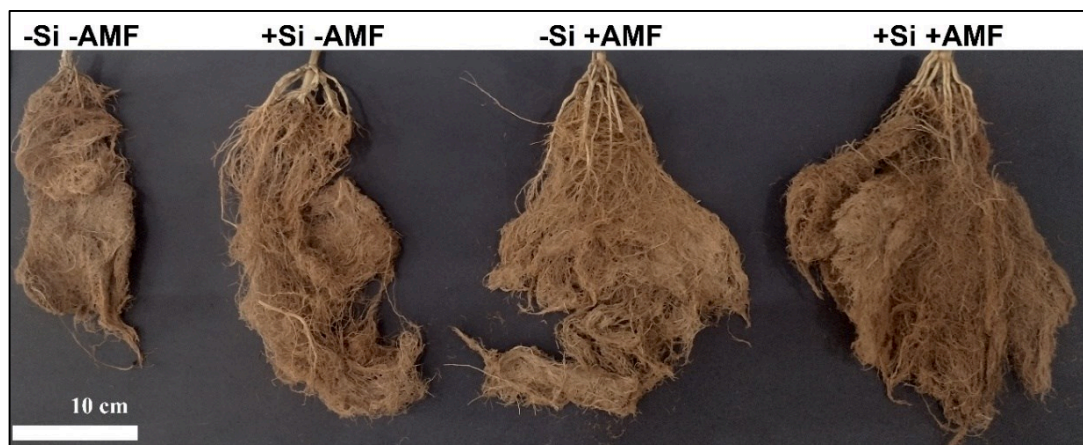
Water restriction led to a 7-days delay in male flowering, comparing the severe restriction in relation to well-watered plants. A quadratic polynomial regression fitted well to data, indicating that 100 and 120% reposition levels were similar for this trait (Figure 5A). In either presence (+Si) or absence (-Si) of Si supply, AMF-inoculated plants presented higher yields on average in severe drought stress (40% of water reposition). However, no statistical differences were detected ($p_{-Si}=0.39$ and $p_{+Si}=0.08$). In optimal water reposition conditions (100%), AMF inoculation in plants with no Si supply stands out in relation to other treatments, being the only plants in this water condition capable of filling the second ear completely (data not shown). Plants in light water excess (120%) also were able to fill the second ear completely, being similar to -Si+AMF plants with 100% reposition (Figure 5B).

Figure 3 - Effect of water reposition levels and AMF inoculation in shoot (A) and root (B) dry weight of sweet corn plants. Means in a same water reposition level followed by different lowercase letters differ by Tukey HSD test ($p < 0.05$).



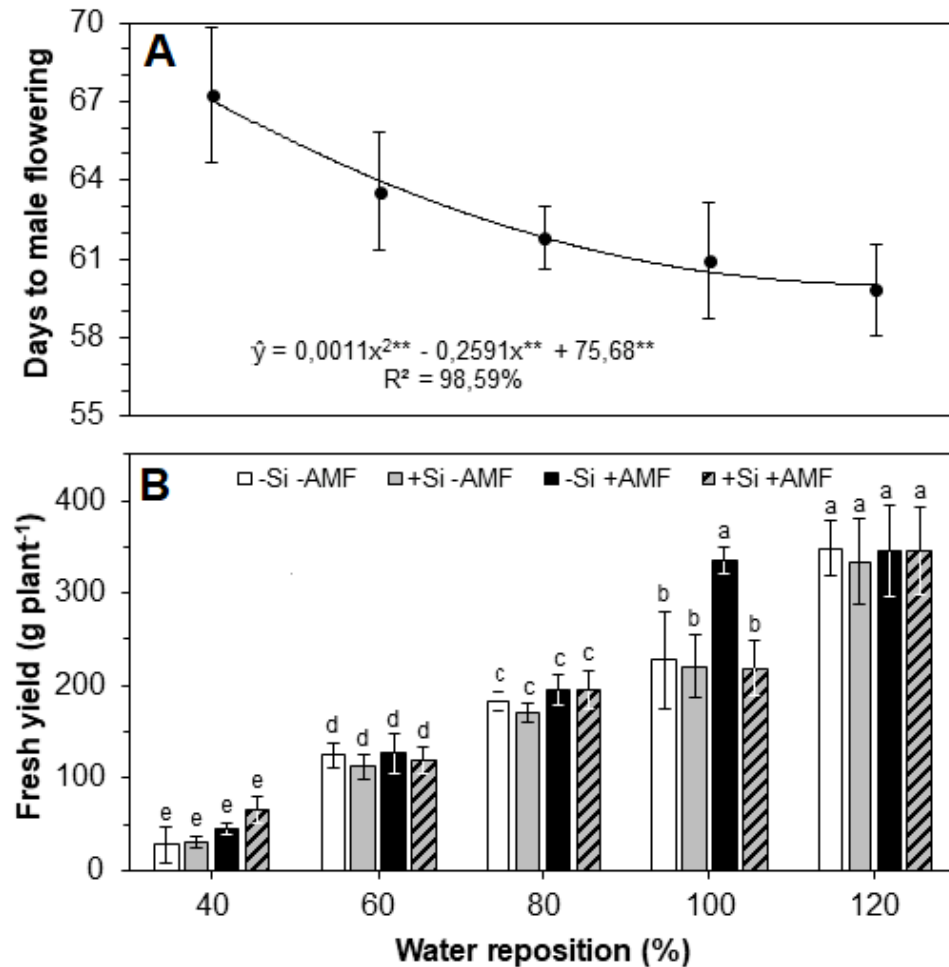
Source: The author.

Figure 4 - Root system of sweet corn plants subjected to severe drought stress (40% of water reposition). From left to right: control (-Si-AMF), Si-supplied (+Si-AMF), AMF inoculated (-Si+AMF), and association of Si supply and AMF inoculation (+Si+AMF)



Source: The author.

Figure 5 - Days to male flowering affected by water reposition levels (A) and fresh market yield of sweet corn subjected to different water reposition levels, Si supply and AMF inoculation (B). ** indicates significance of regression term ($p < 0.01$). Means followed by different letter belong to different cluster according with Scott-Knott test ($p < 0.05$)

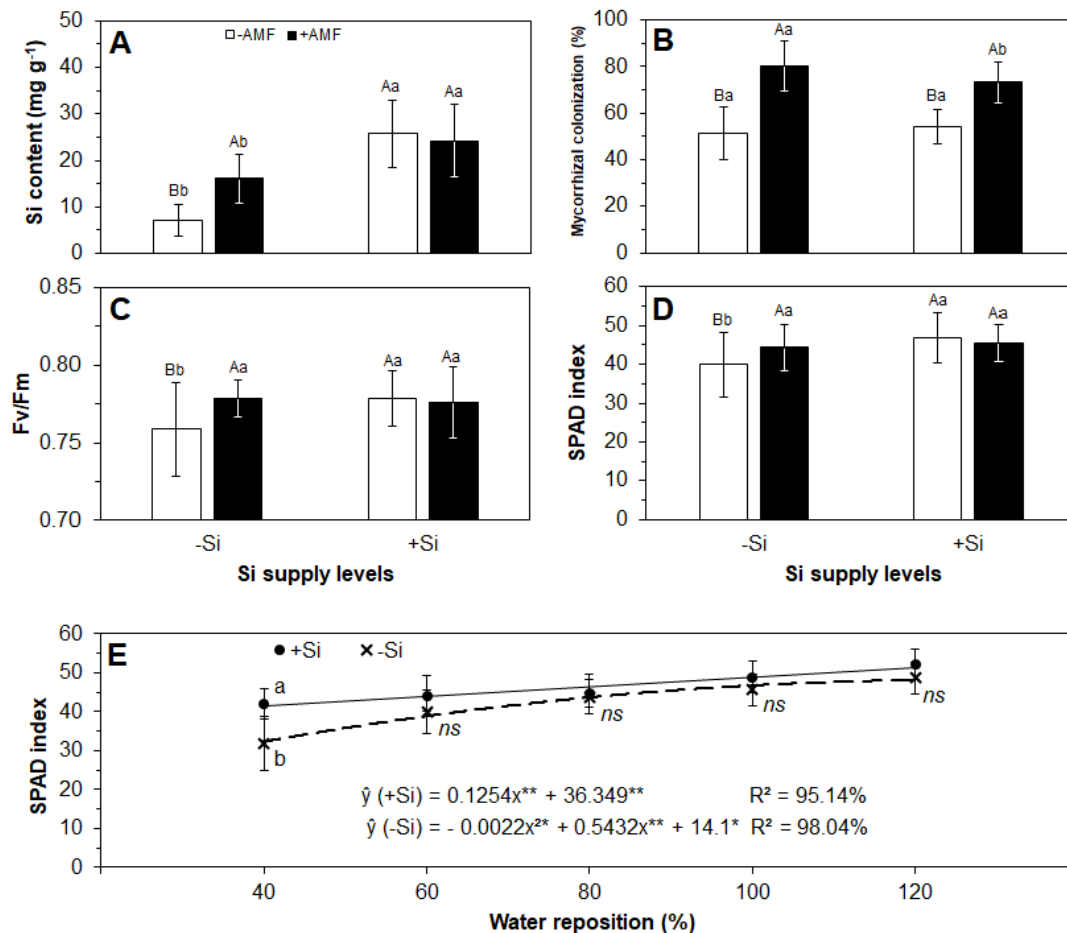


Source: The author.

3.2 Silicon content in plant

As expected, sweet corn accumulated more Si per dry mass when it was supplied to soil via acidity corrective ($p < 0.001$). In non-inoculated plants, was observed a concentration about 0.7% of dry weight in non-Si-supplied conditions *versus* 2.5% in Si-supplied plants. AMF inoculation improved the Si concentration in leaves, but only when no Si was supplied ($p < 0.001$), being the concentration 124% higher if compared with non-inoculated plants (Figure 6A).

Figure 6 - Interaction between Si supply and AMF inoculation in: A) Si content of leaves (mg g^{-1}); B) Mycorrhizal colonization (% of root segments); C) Maximum photochemical efficiency of photosystem II (Fv/Fm); D) Soil plant analysis development (SPAD) index; E) interaction between water reposition and Si supply for SPAD. Uppercase letters compare pairwise columns (AMF effect) and lowercase letters compare columns with same color or different dot styles (Si effect) by Tukey HSD test ($p < 0.05$). *ns*: non-significant. * and ** indicates significance of regression term by *t*-test at $p < 0.05$ and $p < 0.01$, respectively.



Source: The author.

3.3 Mycorrhizal colonization

An interactive effect was observed for AMF inoculation and Si supply ($p=0.0225$). Considerable colonization was observed even in non-inoculated plants. However, very high rates of colonization ($p < 0.001$) were observed in inoculated plants, which presented 36 and 57% more colonization in relation to non-inoculated plants with or without Si supply, respectively (Figure 6B). Si supply had no effect ($p=0.334$) in colonization of non-inoculated plants but reduced ($p=0.0226$) colonization in inoculated plants by 7% (Figure 6B). Curiously, water reposition levels did not affect mycorrhizal colonization ($p=0.188$).

3.4 Photosynthesis

SPAD measurements showed significant interactions ($p < 0.01$) between Si and water reposition and between Si and FMA. Overall, AMF and Si increased about of four SPAD units. in association or isolated (Figure 6C). Analyzing Si and water interaction, Si supplied plants had an increase of almost 10 SPAD units in 40% reposition ($p < 0.0001$), but no significant differences were detected in other water reposition levels. Regression analysis showed linear and quadratic patterns of +Si and -Si plants, respectively (Figure 6E). Chlorophyll fluorescence analysis revealed a pattern similar to SPAD for Si*AMF interaction, with an increase in quantum yield of PSII photochemistry (F_v/F_m), regardless of isolated or in association (Figure 6D).

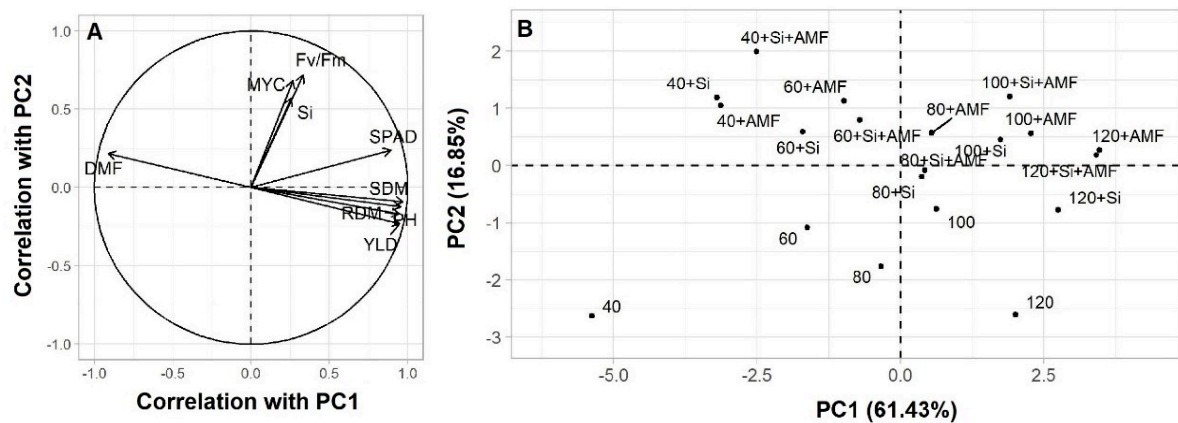
3.5 Principal components analysis (PCA)

The PCA showed the relationships among plant variables and the association with the treatments of drought levels. The two first principal components (PC1 and PC2) explained 77.63% of total variance. Positive and strong correlations were detected between the PC1 and the most of response variables, with exception of days to male flowering, which presented a strong negative correlation. Weakest correlations with the first PC were observed for Mycorrhizal colonization, Si content, and F_v/F_m , but these variables showed the strongest correlations with PC2 (Figure 8A).

In treatments scatterplot (Figure 8B), treatments with AMF inoculation were positively associated with root colonization, Si content, SPAD and F_v/F_m values. Treatments with Si supply and no inoculation (+Si/-AMF) presented intermediated values of these variables, while there is a reduction gradient in these variables from the highest to the lowest level of water reposition when neither Si and AMF (-Si/-AMF) was applied. Severe drought stress without any treatment (40% -Si-AMF) was the most divergent from others, indicating the potential of severe drought mitigation by AMF and Si, alone or in an association. Considering the plant growth (yield, shoot and root dry mass), the first principal component clearly shows that the main driver is the water reposition level, independently of Si and AMF application. The 120%

water reposition level was associated with the highest values and 40% was associated with the lowest plant growth values.

Figure 7 - Principal components analysis (PCA): correlation of variables with the PCs (A) and scatterplot of treatments (B). DMF: Days to male flowering; SDM: Shoot dry mass; RDM: Root dry mass; YLD: Yield; PH: Plant height; SPAD: Soil plant analysis development index; Si: Si content in leaves; MYC: Arbuscular mycorrhizal colonization of roots; Fv/Fm: maximum photochemical efficiency of PSII. Dots with 40, 60, 80, 100, and 120 indicate treatments with the percentage of water reposition levels. AMF: treatments with arbuscular mycorrhizal fungi inoculation; Si: silicon supply treatments.



Source: The author.

4 DISCUSSION

This is the first report on the association between Si and AMF aiming to mitigate drought stress on a C4 plant. In addition, this is the first study using an inoculum containing more than one AMF strain in association with Silicon in drought stress conditions. Water restriction affected sweet corn growth, cycle duration, fresh yield and photosynthesis. Our results indicate beneficial effects of AMF on growth (root and shoot) and Si on SPAD index in plants subjected to severe drought stress. In well-watered conditions, AMF increased sweet corn yield and shoot growth. Overall, both AMF and Si, alone or in association, resulted in improved photosynthesis (SPAD and Fv/Fm).

Higher plant growth caused by AMF inoculation is a well-established knowledge in diverse conditions, including under drought (CHENG *et al.*, 2021). Those effects may come from the higher nutrient and water uptake from the soil explored by the external hyphae (MA *et al.*, 2022; FALL *et al.*, 2022). Improved root growth increases tolerance to drought and

mineral stresses, due to an increase in soil volume explored by the plant. Promotion of root growth is particularly interesting if we consider medium to long-term response in agriculture systems with non-tillage practices. In addition to increasing soil organic fraction after decomposition and promote nutrient cycling, root growth results in soil decompaction, called as “bio-tillage” (JABRO *et al.*, 2021; ZHANG *et al.*, 2021).

SPAD index and photosystem II indicator (Fv/Fm ratio) suggest that the application of Si and AMF reduces plant stress and improves N and photosynthesis efficiency. SPAD is a quick and non-destructive way to access nitrogen (N) content in corn leaves (RHEZALI; AISSAOUI, 2021). In field conditions, Si supply was previous reported to increase SPAD index of maize leaves both ideal and late (drought prone environment) sowing dates (ASADPOUR *et al.*, 2020). Several AMF are also reported to increase N content in corn plants (MA *et al.*, 2022).

Chlorophyll fluorescence analysis is one of the most reliable ways to evaluate the photosynthesis phenomena. The Fv/Fm ratio is a powerful and sensitive indicator of photoinhibition and other kinds of injuries caused in the PSII reaction centers (BRESTIC; ZIVCAK, 2013; KALAJI *et al.*, 2016). A healthy leaf, regardless of plant species, show values close to 0.8 for the Fv/Fm, and the reduction indicates that a proportion of PSII reaction centers are damaged (BJORKMAN; DEMMING, 1987). The drought stress effects on photosynthesis typically start with stomatal effects culminating in metabolic changes in a severe or long-lasting drought (JEDMOWSKI *et al.*, 2013; KALAJI *et al.*, 2016). At the end, the drought stress can be associated with enhanced photoprotective function, increasing the resistance of the PSII to water deficit (CHAVES *et al.*, 2009; LAURIANO *et al.*, 2006).

Even with the impressive photosynthesis efficiency of C4 metabolism in high light and temperatures resulting from morphoanatomical adaptation, which avoids photorespiration, C4 species presents higher susceptibility to photoinhibition when subjected to drought (GUIDI *et al.*, 2019). Here, we showed two sustainable alternatives (Si and AMF) to overcome the drought stress, since the treatments improved those important photosynthetic parameters.

The Si may have a crucial role in drought tolerance through amorphous silica deposition under the cuticle, which is an essential characteristic for enhancing leaf-atmosphere water interactions (LUYCKX *et al.*, 2017; MALIK *et al.*, 2021). Si was also thought to be involved in photosynthesis and the reactions of plants' antioxidants (MALIK *et al.*, 2021; WANG *et al.*, 2021). Si content in leaves of Si-supplied plants (2.5%) was similar to the findings of Boer *et al.* (2018) for grain corn hybrids P30F53YH and DKB310PRO2, which presented 3.1 and 2.7% of Si in leaves. respectively. Both hybrids presented higher Si contents than FT 2015 in non-supplied conditions (about 2%). Si content in leaves of non-Si supplied plants (0.72%) was

similar to those reported by Andrade *et al.* (2014) for two grain corn hybrids: Coodetec 384 (0.73%), and Pioneer BG7065H (0.53%). However, no data regarding Si content in soil are shown for both studies.

Overall, AMF resulted in improved Si content in leaves, but only when no Si was supplied. This is not the first report of enhanced acquisition of Si by AMF-corn. Kothari *et al.* (1990) owns the first report on increased Si uptake by corn plants grown in alkaline soil in response to AMF inoculation (*Glomus mosseae*). Using three AMF species (*G. etunicatum*, *G. diaphanum* and *G. intraradices*) and four soil types (2 acid and 2 alkaline), Clark; Zeto (1996; 2000) showed that AMF-maize symbiosis can increase corn Si content when grown in acid soils, but the intensity of Si acquisition is clearly affected by AMF species and soil type. Frew *et al.* (2017) evaluated the Si content of sugarcane in low and high Si soils using a commercial inoculum (*G. etunicatum*, *G. coronatum*, *G. intraradices*, and *G. mosseae*) and native AMF from respective soil. Both AMFs (commercial and native) increased Si content in roots, but only in low Si soil; shoot Si content was unaffected by AMF inoculation in both soils. In alkaline soil, *G. versiforme* was found to be more efficient to help tomato plants accumulate Si than *Funneliformis mosseae* and *A. delicata*. This study also revealed that different AMFs affect Si dynamics in soil, probably by differences in secretion (JU *et al.*, 2021). The differential composition of compounds in the rhizosphere of citrus was previously reported as a drought alleviation mechanism by AMF (CHENG *et al.*, 2022). Improved Si uptake in low-Si soil by AMF-plants could result in enhanced bioavailability of Si for plants at the medium-long term since phytoliths are a main bioavailable Si source (DE TOMBEUR *et al.*, 2021).

Johnson *et al.* (2022) investigated the reciprocal effects of AMF colonization and Si accumulation in *Brachypodium distachyon* (*Poaceae*). To check this, two contrasting genotypes regarding Si accumulation (wild-type/high Si-accumulator; Bdlsi1-1/low Si-accumulator) were submitted to AMF inoculation and five doses of Si supply. It was found that AMF did not have any effect on Si accumulation in leaves, but Si accumulation reduced AMF colonization in wild-type plants. Thus, they proposed that AMF contribution to Si uptake by plants that inherently have a high capacity to accumulate Si due to transporters, like corn (MITANI *et al.*, 2009) and other *Poaceae*, is weaker and more context dependent.

In agreement with Johnson *et al.* (2022), we also verified that Si accumulation reduced AMF colonization, but on a lower scale than the previous report. Suppression in root colonization by AMF Si-supplied plants may occur due to decrease in the C:Si stoichiometry (FRAZÃO *et al.*, 2020), since AMF acquires C from plants. Another possible mechanism is the mechanical barrier resulting from Si accumulation in cell walls, which probably decreased the

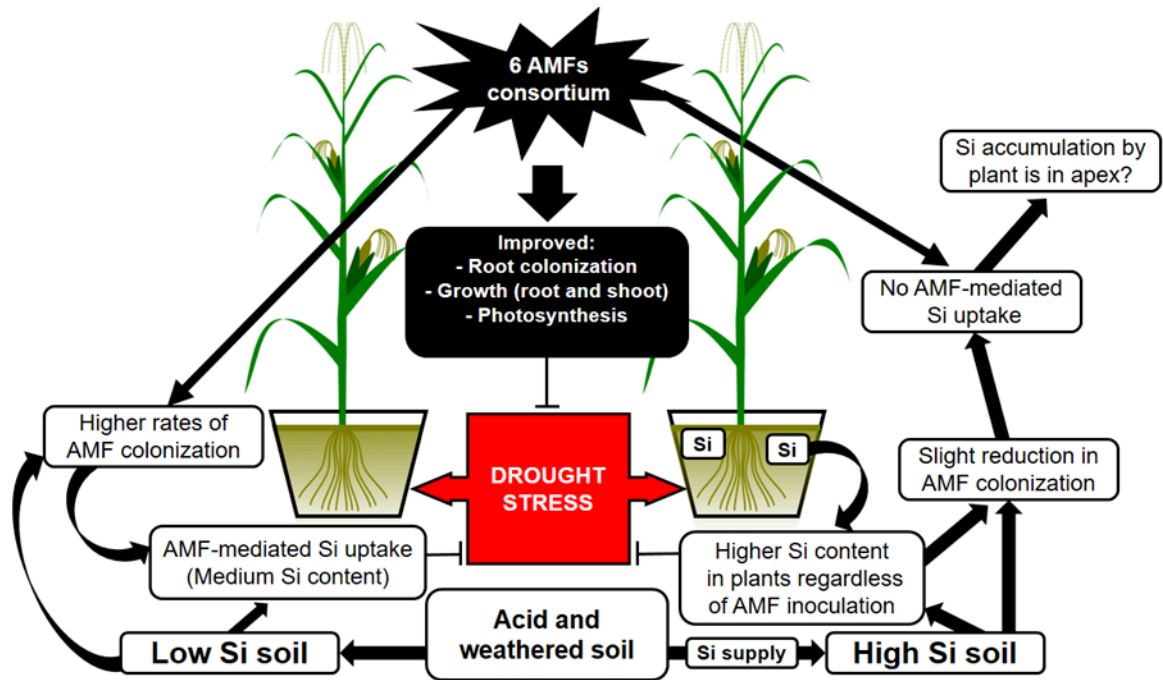
fungi penetration. Sweet corn presents some alleles that lacks some C metabolism enzymes present in grain corn, thus resulting in sugar accumulation in grains instead of starch (REVILLA *et al.*, 2021). Because of C metabolism differences, a different behavior of Si-AMF-plant interaction may occur in grain corn, requiring further studies to establish better these effects.

Regarding Si accumulation by inoculated plants, we observed a different pattern than Johnson *et al.* (2022), since AMF inoculated plants presented more Si content than non-inoculated plants, indicating that probably exists the context dependency suggested by these authors for AMF-mediated Si accumulation in *Poaceae*. A main factor affecting AMF-mediated Si uptake that may explains these differences is soil pH (CLARK; ZETO, 1996; 2000), since the soil used by Johnson *et al.* (2022) presents pH 7.4 and the soil used here is acid (pH = 5.2) and, even with soil correctives, is not expected to reach an alkaline pH.

Considering the set of findings and the literature studies, a model of interaction among plant-AMF-Si is suggested (Figure 8). The plant growing in low Si soil allows higher mycorrhizal colonization when selected AMF isolates are inoculated (Figure 6B). The higher mycorrhizal colonization provided by the inoculum results in greater Si uptake by plants (Figure 6A). The higher Si content in comparison to the uninoculated, in turn, leads to lower stress. which is indicated by the photosynthetic parameters (Figures 6C and 3). On the other hand, the application of Si in the soil also results in higher drought tolerance by plants. Therefore, inoculation of plants with selected AMF and with an appropriate Si content supplied through application results in higher drought tolerance and, consequently, improved plant growth under water stress (Figure 3).

However, the findings encompass extremely intricate phenomena of plant-Si-AMF-environment interactions affected by several factors. Thus, to further understand these consequences, additional research must be done. Future studies should consider variations in Si and AMF inoculum dosages, plant genotype, AMF strains, and Si sources. In addition, the molecular regulation between plant-Si-AMF can be discovered using molecular techniques like metagenomic, transcriptome, and metabolome analysis.

Figure 8 - Model suggested for sweet corn-AMF-Si interactions in drought stress conditions: extremely intricate phenomena



Source: The author.

5 CONCLUSIONS

Si supply and specially AMF inoculation showed to be potential strategies to improve the resilience of sweet corn plants under stress conditions. In severely stressed plants, the strategies seem to be more effective than in light stress levels to promote plant growth and physiological tolerance to stress. However, further studies need to be conducted in order to investigate better these effects. Future research should consider variations in plant genotype, Si and AMF inoculum doses, as well as AMF strains and Si sources. Molecular approaches such as metagenomic, transcriptome, and metabolome can aid to reveal the molecular interaction among plant-Si-AMF.

A comparison of our findings with previous reports clearly shows the high complexity of plant-Si-AMF-environment interactions, which are strongly affected by several factors, such as soil traits, and plant, and fungi genotypes. Thus, to unravel the basis of these complex interactions and guarantee crop resilience under stressed conditions, mycologists, crop, and soil scientists, as well as breeders and geneticists, should join forces to result in significant knowledge that can be applied to agricultural systems.

REFERENCES

- ANDRADE, F. A.; ANDRADE JÚNIOR, O.; ANDRADE, C. G. T. J.; Miglioranza, E. Accumulation of silicon and arrangement and shapes of silica bodies in corn leaves. **Genetics and Molecular Research**, Ribeirão Preto, v. 13, p. 1690-1696, 2014. DOI: <http://dx.doi.org/10.4238/2014.January.24.6>.
- ANDRADE, R. S.; STONE, L. F. Estimativa da umidade na capacidade de campo em solos sob Cerrado. **Revista Brasileira de Engenharia Agrícola e Ambiental**, Campina Grande, v. 15, n. 2, p. 111-116. DOI: <https://doi.org/10.1590/S1415-43662011000200001>.
- ASADPOUR, S.; MADANI, H.; MOHAMMADI, G. N.; HERAVAN, I. M.; ABAD, H. H. S. Improving maize yield with advancing planting time and nano-silicon foliar spray alone or combined with zinc. **Silicon**, London, v. 14, p. 201–209, 2022. DOI: <https://doi.org/10.1007/s12633-020-00815-5>.
- BIANCHINI, H. C.; MARQUES, D. J. Tolerance to hydric stress on cultivars of silicon-fertilized corn crops: absorption and water-use efficiency. **Bioscience Journal**, Uberlândia, v. 35, n. 2, p. 527-539, 2019. DOI: <https://doi.org/10.14393/BJ-v35n2a20198-42347>.
- BJÖRKMAN, O.; DEMMIG, B. Photon yield of O₂ evolution and chlorophyll fluorescence characteristics at 77 K among vascular plants of diverse origins. **Planta**, Berlin, v. 170, n. 4, p. 489-504, 1987. DOI: <https://doi.org/10.1007/BF00402983>.
- BOER, C. A.; SAMPAIO, M. V.; PEREIRA, H. S. Silicon-mediated and constitutive resistance to *Rhopalosiphum maidis* (Hemiptera: Aphididae) in corn hybrids. **Bulletin of Entomological Research**, London, v. 109, p. 356-364, 2018. DOI: <https://doi.org/10.1017/S0007485318000585>.
- BRÁS, T.A.; SEIXAS, J.; CARVALHAIS, N.; JÄGERMEYR, J. Severity of drought and heatwave crop losses tripled over the last five decades in Europe. **Environmental Research Letters**, Bristol, v. 16, n. 6, 065012, 2021. DOI: <https://doi.org/10.1088/1748-9326/abf004>.
- BRESTIC, M.; ZIVCAK, M. PSII fluorescence techniques for measurement of drought and high temperature stress signal in plants: protocols and applications. In: ROUT, G. R.; Das, A. B. (ed.) **Molecular stress physiology of plants**. Dordrecht: Springer, 2013, p. 87-131. DOI: https://doi.org/10.1007/978-81-322-0807-5_4.
- BRUNDRETT, M. C.; TEDERSOO, L. Evolutionary history of mycorrhizal symbioses and global host plant diversity. **New Phytologist**, Oak Ridge, v. 220, p. 1108–1115, 2018. DOI: <https://doi.org/10.1111/nph.14976>.
- BUOL, S.W. Soils and agriculture in central-west and north Brazil. **Scientia Agricola**, Piracicaba, v. 66, p. 697-707, 2009. DOI: <https://doi.org/10.1590/S0103-90162009000500016>.

CAMARGO, M. S.; ROCHA, G.; KORNDÖRFER, G. H. Silicate fertilization of tropical soils: silicon availability and recovery index of sugarcane. **Revista Brasileira de Ciência do Solo**, Viçosa, v. 37, p. 1267–1275, 2013. DOI: <https://doi.org/10.1590/S0100-06832013000500016>.

CHAVES, M. M.; FLEXAS, J.; PINHEIRO, C. Photosynthesis under drought and salt stress: regulation mechanisms from whole plant to cell. **Annals of Botany**, Oxford, v. 103. n. 4. p. 551–560, 2009. DOI: <https://doi.org/10.1093/aob/mcn125>.

CHENG H, GIRI B, WU Q, ZOU Y, KUČA K. Arbuscular mycorrhizal fungi mitigate drought stress in citrus by modulating root microenvironment. **Archives of Agronomy and Soil Science**, London, v. 68, p. 1217–1228, 2022. DOI: <https://doi.org/10.1080/03650340.2021.1878497>.

CLARK, R. B.; ZETO, S. K. Mineral acquisition by mycorrhizal maize grown on acid and alkaline soil. **Soil Biology and Biochemistry**, Amsterdam, v. 28, p. 1495–1503, 1996. DOI: [https://doi.org/10.1016/S0038-0717\(96\)00163-0](https://doi.org/10.1016/S0038-0717(96)00163-0).

CLARK, R.B.; ZETO, S. K. Mineral acquisition by arbuscular mycorrhizal plants. **Journal of Plant Nutrition**, London, v. 23, p. 867–902, 2000. DOI: <https://doi.org/10.1080/01904160009382068>.

DAS, D.; BASAR, N. U.; ULLAH, H.; SALIN, K. R.; DATTA, A. Interactive effect of silicon and mycorrhizal inoculation on growth, yield and water productivity of rice under water-deficit stress. **Journal of Plant Nutrition**, London, v. 44, n. 18, p. 2756–2769, 2021. DOI: <https://doi.org/10.1080/01904167.2021.1927087>.

DE TOMBEUR, F.; ROUX, P.; CORNELIS, J. T. Silicon dynamics through the lens of soil-plant-animal interactions: perspectives for agricultural practices. **Plant and Soil**, Perth, v. 467, p. 1–28, 2021. DOI: <https://doi.org/10.1007/s11104-021-05076-8>.

ELLIOT, C.L.; SNYDER, G. H. Autoclave-induced digestion for the colorimetric determination of silicon in rice straw. **Journal of Agriculture and Food Chemistry**, Washington, v. 39, p. 1118–1119, 1991. DOI: <https://doi.org/10.1021/jf00006a024>.

EPSTEIN, E. Silicon. **Annual review of plant physiology and plant molecular biology**, Palo Alto, v. 50, p. 641–664, 1999. DOI: <https://doi.org/10.1146/annurev.arplant.50.1.641>.

FAHAD, S. *et al.* Crop production under drought and heat stress: plant responses and management options. **Frontiers in Plant Science**, Lausanne, v. 8, 1147, 2017. DOI: <https://doi.org/10.3389/fpls.2017.01147>.

FALL, A. F.; NAKABONGE, G.; SSEKANDI, J.; FOUNOUNE-MBOUP, H.; APORI, S.O.; NDIAYE, A.; BADJI, A.; NGOM, K. Roles of arbuscular mycorrhizal fungi on soil fertility:

contribution in the improvement of physical, chemical, and biological properties of the soil. **Frontiers in Fungal Biology**, Lausanne, v. 3, 723892, 2022. DOI: <https://doi.org/10.3389/ffunb.2022.723892>.

FRAZÃO, J. J.; DO PRADO, R. M.; SOUZA JÚNIOR, J. P. de.; ROSSATTO, D. R. Silicon changes C:N:P stoichiometry of sugarcane and its consequences for photosynthesis, biomass partitioning and plant growth. **Scientific Reports**, London, v. 10, 12492, 2020. DOI: <https://doi.org/10.1038/s41598-020-69310-6>.

FREW, A.; POWELL, J. R.; ALLSOPP, P. G.; SALLAM, N.; JOHNSON, S. N. Arbuscular mycorrhizal fungi promote silicon accumulation in plant roots. reducing the impacts of root herbivory. **Plant and Soil**, Perth, v. 419, p. 423–433, 2017. DOI: <https://doi.org/10.1007/s11104-017-3357-z>.

GIANINAZZI, S.; GOLLOTE, A.; BINET, M. N.; VAN TUINEN, D.; REDECKER, D.; Wipf, D. Agroecology: the key role of arbuscular mycorrhizas in ecosystem services. **Mycorrhiza**, Heidelberg, v. 20, n. 8, p. 519-530, 2010. DOI: <https://doi.org/10.1007/s00572-010-0333-3>.

GIOVANNETTI, M.; MOSSE, B. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection roots. **New Phytologist**, Oak Ridge, v. 84, p. 489-500, 1980. DOI: <https://doi.org/10.1111/j.1469-8137.1980.tb04556.x>.

GUIDI, L.; PICOLLO, E. L.; LANDI, M. Chlorophyll fluorescence. photoinhibition and abiotic stress: does it make any difference the fact to be a C3 or C4 species? **Frontiers in Plant Science**, Lausanne, v. 10, 174, 2019. DOI: <https://doi.org/10.3389/fpls.2019.00174>.

HAGHIGHI, T. M.; SAHARKHIZ, M. J. Mycorrhizal colonization and silicon nutrition mitigates drought stress in licorice (*Glycyrrhiza glabra* L.) with morphophysiological and biochemical perspectives. **Industrial Crops and Products**, Amsterdam, v. 178, 114650, 2022. DOI: <https://doi.org/10.1016/j.indcrop.2022.114650>.

HAGHIGHI, T. M.; SAHARKHIZ, M. J.; KAVOOSI, G.; JOWKAR, A. Monitoring amino acid profile and protein quality of Licorice (*Glycyrrhiza glabra* L.) under drought stress, silicon nutrition and mycorrhiza inoculation. **Scientia Horticulturae**, Amsterdam, v. 295, 110808, 2022. DOI: <https://doi.org/10.1016/j.scienta.2021.110808>.

JABRO, J. D.; ALLEN, B.L.; RAND, T.; DANGI, S. R.; CAMPBELL, J. W. Effect of previous crop roots on soil compaction in 2 yr rotations under a no-tillage system. **Land**, Basel, v. 10, n. 2, 202, 2021. DOI: <https://doi.org/10.3390/land10020202>.

JEDMOWSKI, C.; ASHOUB, A.; BRUGGEMANN, W. Reactions of Egyptian landraces of *Hordeum vulgare* and *Sorghum bicolor* to drought stress, evaluated by the OJIP fluorescence

transient analysis. **Acta Physiologiae Plantarum**, Kraków, v. 35, n. 2, p. 345–354, 2013. DOI: <https://doi.org/10.1007/s11738-012-1077-9>.

JOHNSON, S. N.; POWEL, J.R.; FREW, A.; CIBILS–STEWART, X. Silicon accumulation suppresses arbuscular mycorrhizal fungal colonisation in the model grass *Brachypodium distachyon*. **Plant and Soil**, Perth, v. 477, p. 219–232, 2022. DOI: <https://doi.org/10.1007/s11104-022-05463-9>.

JU, S.; WANG, N.; CHEN, M.; WANG, J. Arbuscular mycorrhizal fungi regulate tomato silicon absorption. **Soil Science and Plant Nutrition**, Tokyo, v. 67, p. 408–414, 2021. DOI: <https://doi.org/10.1080/00380768.2021.1927833>.

KALAJI, H. M.; JAJOO, A.; OUKARROUM, A.; BRESTIC, M.; ZIVCAK, M.; SAMBORSKA, I. A.; CETNER, M. D.; ŁUKASIK, A.; GOLTSEV, V.; LADLE, R. J. Chlorophyll a fluorescence as a tool to monitor physiological status of plants under abiotic stress conditions. **Acta Physiologiae Plantarum**, Kraków, v. 38, 102, 2016. <https://doi.org/10.1007/s11738-016-2113-y>.

KOUR, D.; KHAN, S. S.; KAUR, T.; KOUR, H.; SINGH, G.; YADAV, A.; YADAV, A. N. Drought adaptive microbes as bioinoculants for the horticultural crops. **Heliyon**, Cambrigde, v. 8, e09493, 2022. DOI: <https://doi.org/10.1016/j.heliyon.2022.e09493>.

KOTHARI, S. K.; MARSCHNER, H.; ROMHELD, V. Direct and indirect effects of VA mycorrhizal fungi and rhizosphere microorganisms on acquisition of mineral nutrients by maize (*Zea mays* L.) in a calcareous soil. **New Phytologist**, Oak Ridge, v. 116, p. 637–645, 1990. DOI: <https://doi.org/10.1111/j.1469-8137.1990.tb00549.x>.

KORNDÖRFER, G. H.; PEREIRA, H. S.; NOLLA, A. **Análise de Silício: solo, planta e fertilizante**. Uberlândia: UFU, 2004. 50 p.

LAURIANO, J. A.; RAMALHO, J. C.; LIDON, F. C.; CÉU MATOS, M. Mechanisms of energy dissipation in peanut under water stress. **Photosynthetica**, Praha, v. 44, n. 3, p. 404–410, 2006. DOI: <https://doi.org/10.1007/s11099-006-0043-4>.

LIEBER, M.; CHIN-HONG, P.; KELLY, K.; DANDU, M.; WEISER, S. D. A systematic review and meta-analysis assessing the impact of droughts, flooding, and climate variability on malnutrition. **Global Public Health**, London, v. 17, n. 1, p. 68–82, 2022. DOI: <https://doi.org/10.1080/17441692.2020.1860247>.

LUYCKX, M.; HAUSMAN, J. F.; LUTTS, S.; GUERRIERO, G. Silicon and Plants: Current Knowledge and Technological Perspectives. **Frontiers in Plant Science**, Lausanne, v. 8, 411, 2017. DOI: <https://doi.org/10.3389/fpls.2017.00411>.

MA, J.; WANG, W.; YANG, J.; QIN, S.; YANG, Y.; SUN, C.; PEI, G.; ZEESHAN, M.; LIAO, H.; LIU, L.; HUANG, J. Mycorrhizal symbiosis promotes the nutrient content accumulation and affects the root exudates in maize. **BMC Plant Biology**, London, v. 22, 64, 2022. DOI: <https://doi.org/10.1186/s12870-021-03370-2>.

MALIK, M. A.; WANI, A. H.; MIR, S. H.; REHMAN, I. U.; TAHIR, I.; AHMAD, P.; RASHID, I. Elucidating the role of silicon in drought stress tolerance in plants. **Plant Physiology and Biochemistry**, Paris, v. 165, p. 187-195, 2021. <https://doi.org/10.1016/j.plaphy.2021.04.021>.

MARQUES, D. J.; BIANCHINI, H. C.; MACIEL, G. M.; MENDONÇA, T. F. N.; SILVA, M. F. Morphophysiological changes resulting from the application of silicon in corn plants under water stress. **Journal of Plant Growth Regulation**, Basel, v. 41, p. 569–584, 2022. DOI: <https://doi.org/10.1007/s00344-021-10322-5>.

MITANI, N.; YAMAJI, N.; MA, J. F. Identification of maize silicon influx transporters. **Plant & Cell Physiology**, Oxford, v. 50, p. 5-12, 2009. DOI: <https://doi.org/10.1093/pcp/pcn110>.

MORADTALAB, N.; HAJIBOLAND, R.; ALIASGHARZAD, N.; HARTMANN, T. E.; NEUMANN, G. Silicon and the association with an arbuscular-mycorrhizal fungus (*Rhizophagus clarus*) mitigate the adverse effects of drought stress on strawberry. **Agronomy**, Basel, v. 9, 41, 2019. DOI: <https://doi.org/10.3390/agronomy9010041>.

NAUMANN, G.; CAMMALLERI, C.; MENTASCHI, L.; FEYEN, L. Increased economic drought impacts in Europe with anthropogenic warming. **Nature Climate Change**, London, v. 11, n. 6, p. 485-491, 2021. DOI: <https://doi.org/10.1038/s41558-021-01044-3>.

NOVAIS, R. F.; NEVES, J. C. L.; BARROS, N. F. Ensaio em ambiente controlado. *In*: NOVAIS, R. F.; NEVES, J. C. L.; BARROS, N. F.; OLIVEIRA, A. J.; GARRIDO, W. E.; ARAÚJO, J. D.; LOURENÇO, A. S. **Métodos de pesquisa em fertilidade do solo**. 3. ed. Brasília: EMBRAPA-SEA, 1991, p. 189-253.

PUTRA, R.; POWELL, J. R.; HARTLEY, S. E.; JOHNSON, S. N. Is it time to include legumes in plant silicon research? **Functional Ecology**, London, v. 34, p. 1142–1157, 2020. DOI: <https://doi.org/10.1111/1365-2435.13565>.

REVILLA, P.; ANIBAS, C. M.; TRACY, W. F. Sweet corn research around the world 2015–2020. **Agronomy**, Basel, v. 11, 534, 2021. DOI: <https://doi.org/10.3390/agronomy11030534>.

RHEZALI, A.; AISSAOUI, A. E. Feasibility study of using absolute SPAD values for standardized evaluation of corn nitrogen status. **Nitrogen**, Basel, v. 2, n. 3, p. 298-307, 2021. DOI: <https://doi.org/10.3390/nitrogen2030020>.

SELEIMAN, M. F.; AL-SUHAIBANI, N.; ALI, N.; AKMAL, M.; ALOTAIBI, M.; REFAY, Y.; DINDAROGLU, T.; ABDUL-WAJID, H. H.; BATTAGLIA, M. L. Drought stress impacts on plants and different approaches to alleviate its adverse effects. **Plants**, Basel, v. 10, n. 2, 259, 2021. DOI: <https://doi.org/10.3390/plants10020259>.

SHARMA, D.; KAPOOR, R.; BHATNAGAR, A. K. DIFFERENTIAL growth response of *Curculigo orchoides* to native arbuscular mycorrhizal fungal (AMF) communities varying in number and fungal components. **European Journal of Soil Biology**, Paris, v. 45, n. 4, p. 328-333, 2009. DOI: <https://doi.org/10.1016/j.ejsobi.2009.04.005>.

SRIVASTAVA, A.; SHARMA, K. V.; KAUSHIK, S. P.; EL-SHEIKH, M. A.; QADIR, S.; MANSOOR, S. Effect of silicon application with mycorrhizal inoculation on *Brassica juncea* cultivated under water stress. **PLOS ONE**, San Francisco, v. 17, e0261569, 2022. DOI: <https://doi.org/10.1371/journal.pone.0261569>.

VAN GENUTCHEN, M. T. A closed-form equation for predicting the hydraulic conductivity of unsaturated Soils. **Soil Science Society of America Journal**, Madison, v. 44, p. 892-898, 1980. DOI: <https://doi.org/10.2136/sssaj1980.03615995004400050002x>.

VAN RAIJ, B.; CANTARELLA, H. Milho para grão e silagem. In: VAN RAIJ, B.; CANTARELLA, H.; QUAGGIO, J.A.; FURLANI, A. M. C. (ed.) **Recomendações de adubação e calagem para o estado de São Paulo**. 2. ed. Campinas: Instituto Agrônomo, p. 56-59, 1997.

VEGA, I.; PONTIGO, S.; NUNES-NESI, A.; DE LA LUZ, M. M.; MEIER, S.; CARTES, P. Interaction between silicon and arbuscular mycorrhizal symbiosis: An ecologically sustainable tool to improve crop fitness under a drought scenario? **Journal of Soil Science and Plant Nutrition**, Temuco, 2021. DOI: <https://doi.org/10.1007/s42729-021-00701-y>.

Vierheilig, H.; Coughlan, A.P.; Wyss, U.; Piché, Y. Ink and vinegar. a simple staining technique for arbuscular-mycorrhizal fungi. **Applied Environmental Microbiology**, Washington, v. 64, p. 5004-5007, 1998. DOI: <https://doi.org/10.1128/AEM.64.12.5004-5007.1998>.

WANG, M.; WANG, R.; MUR, L. A. J. *et al.* Functions of silicon in plant drought stress responses. **Horticulture Research**, Nanjing, v. 8, n. 254, 2021. DOI: <https://doi.org/10.1038/s41438-021-00681-1>.

YAN, G.; NIKOLIC, M.; YE, M.; XIAO, Z.; LIANG, Y. Silicon acquisition and accumulation in plant and its significance for agriculture. **Journal of Integrative Agriculture**, Beijing, v. 17, n. 10, p. 2138-2150, 2018. DOI: [https://doi.org/10.1016/S2095-3119\(18\)62037-4](https://doi.org/10.1016/S2095-3119(18)62037-4).

ZHANG, Z.; YAN, L.; WANG, Y.; RUAN, R.; XIONG, P.; PENG, X. Bio-tillage improves soil physical properties and maize growth in a compacted Vertisol by cover crops. **Soil Science Society of America Journal**, Madison, v. 86, n. 2, p. 324-337, 2021. DOI: <https://doi.org/10.1002/saj2.20368>.