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FERNANDO MAX LIMA

COMPARATIVE EVALUATION OF METHODS FOR THE DETECTION OF ELECTRODERMAL RESPONSES TO MULTILEVEL INTENSITY THERMAL NOXIOUS STIMULI

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PhD thesis presented to the Faculty of Electrical Engineering of the Federal University of Uberlandia, as a partial requirement for obtaining the degree of Doctor in Sciences.

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"So everything that could've happened does happen, just in another universe … and the history of this particular universe, and this particular version of you in it, has led where you're now."

Stephen Hawking

ABSTRACT

Pain is a subjective sensation, only fairly related to tissue damage or any other pathological condition. While conscious and communicative subjects can provide accurate description about its location, characteristics and intensity by means of scales and questionnaires, realistic measures on non-communicative patients are a way more complicated, and they are usually dependent on the physicians and allied health professionals observations and annotations. To provide more precise assumptions on someone's painful experience, computer based algorithms, image and sensor based measurements has been extensively studied in the last decades. By far, skin conductance has been one of the most evaluated signals due to its direct correlation to the activity of the sympathetic nervous system, which is normally increased in cases of sustained pain or external noxious stimulation. Although measures of skin electric properties dates from the past century, new methodologies have been developed to infer the nerve activity of the sudo-motor nerves branches in order to provide more robust analyses of the skin conductance itself. In this sense, this work aimed to evaluate whether these newly developed techniques could provide further discriminative power to the study of painful sensations and noxious stimulation intensities or not. Therefore, a 3 x 4 factors study design was used to evaluate how the skin conductance analysis is affected by the usage of three different methods in four different noxious stimulation intensities. For this study, electrodermal activity data from the Biovid Heat Pain Databese were used. The database consists of data from 96 healthy participants aged between 18 and 65 years old (40.9 ± 14.9 years, mean ± standard deviation), equally distributed by gender. The participants went through heat noxious stimulation at the backside of the forearm 20 times at each of the four different intensities (participant's pain threshold temperature, participant's pain tolerance temperature, and two other equally distributed temperatures between the threshold and the tolerance temperatures) in a random order. From baseline temperature (32ºC), temperature increased for two seconds until reach the target temperature, kept there for four seconds, and then decreased back to the baseline temperature as fast as possible. Randomized pauses of 8 to 12 seconds were applied between two consecutive stimuli. Raw electrdermal signals were filtered with 1st order Butter-worth low-pass filter with a 5 Hz cutoff frequency, downsampled to 64 Hz, and analyzed using Ledalab software in Matlab. Continuous decomposition analysis (CDA), discrete decomposition analysis (DDA) and trough-to-peak (TTP) analysis were compared. Within the 8 second-long analysis window following each single stimulation, CDA and DDA methods identified more electrodermal responses between 1 and 9 seconds after

noxious stimulation than TTP for all stimulation intensities. However, the occurrence rates of at least one ER.EDR after noxious stimulation were similar among the three methods and tended to increase with increasing intensities. Among the common features, amplitude sum had better discriminative power for differentiating noxious stimulation intensities regardless of the method. The results suggest that all investigated methods performed similarly in identifying electrodermal changes in response to high-intensity thermal noxious stimuli. In the experimental conditions of cutaneous heat stimulation of nociceptors, good discrimination among stimulation intensities was found using the amplitude sum feature. In conclusion, although CDA and DDA were more sensitive than TTP for identifying ER.EDRs, neither CDA nor DDA brought further discriminative power to the study of noxious stimulation intensities when compared to the traditional TTP method.

Keywords: galvanic skin response, pain, pain measurement, nociception, nociceptive pain, psychophysiology.

RESUMO

A dor é uma sensação subjetiva, apenas parcialmente relacionada com danos teciduais ou qualquer outra condição patológica. Embora pessoas conscientes e comunicativas possam fornecer uma descrição exata sobre sua localização, características e intensidade por meio de escalas e questionários, medidas fidedignas em pacientes não comunicativos são mais complicadas, e geralmente são dependentes das observações e anotações dos médicos e dos profissionais de saúde. Para fornecer informações mais precisas sobre a experiência dolorosa dessas pessoas, algoritmos baseados em imagens e medições de sensores diversos têm sido extensivamente estudados nas últimas décadas. De longe, a condutância da pele tem sido um dos sinais mais estudados devido à sua correlação direta com a atividade do sistema nervoso simpático, que normalmente é aumentada em casos de dor sustentada ou estimulação externa nociva. Embora os estudos das propriedades elétricas da pele datem do século passado, novas metodologias têm sido desenvolvidas para estimar a atividade dos nervos sudomotores a fim de fornecer uma análise mais robusta da condutância da pele. Nesse sentido, este trabalho objetivou avaliar se essas técnicas recém-desenvolvidas poderiam fornecer melhor poder discriminativo ao estudo de sensações dolorosas e intensidades de estimulação nocivas ou não. Consequentemente, uma análise de 3 x 4 fatores foi usada para avaliar como a condutância da pele é afetada utilizando três métodos distintos de análise em quatro intensidades diferentes de estimulação nociceptiva. Para este estudo foram utilizados dados de atividade eletrodérmica da pele extraídos da base de dados "Biovid Heat Pain Database". O banco de dados consiste dados de 96 participantes saudáveis com idades entre 18 e 65 anos (40,9 ± 14,9 anos, média ± desvio padrão), distribuídos igualmente por gênero. Os participantes foram estimulados com energia térmica na parte de trás do antebraço por 20 vezes em cada uma das quatro intensidades distintas (limiar de dor do participante, limiar de tolerância da dor, e duas outras intensidades igualmente distribuídas entre os dois limiares) numa ordem aleatória. Partindo da temperatura basal (32 ° C), a temperatura aumentava por cerca de dois segundos até atingir a temperatura alvo, a qual era mantida por quatro segundos, e depois diminuía para a temperatura basal o mais rápido possível. Pausas aleatórias de 8 a 12 segundos eram aplicadas entre dois estímulos consecutivos. Os sinais brutos da condutância da pele foram filtrados com filtros passa-baixa Butterworth de primeira ordem, com frequência de corte de 5 Hz, reamostrados para 64 Hz, e analisados usando o software gratuito Ledalab em ambiente MATLAB. Foram comparadas os métodos "*Continuous Decomposition Analysis*" (CDA), "*Discrete Decomposition Analysis*" (DDA) e "*Trough-to-peak Analysis*" (TTP). Dentro de

uma janela de análise de 8 segundos de duração após cada estimulação, os métodos CDA e DDA identificaram mais respostas eletrodérmicas entre 1 e 9 segundos após estimulação nociva do que o método TTP para todas as intensidades de estimulação avaliadas. No entanto, as taxas de ocorrência de pelo menos uma resposta eletrodérmica após estimulação nociva foram semelhantes entre os três métodos e tenderam a aumentar com intensidades crescentes. Entre as características comuns ao três métodos, a soma das amplitudes teve o melhor poder discriminativo para diferenciar intensidades nocivas de estimulação, independente do método utilizado. Os resultados sugerem que todos os métodos investigados apresentaram resultados semelhante na identificação de alterações eletrodérmicas em resposta a estímulos térmicos nocivos de alta intensidade. Em condições experimentais de estimulação térmica dos nociceptores da pele, observou-se boa discriminação entre as várias intensidades de estimulação utilizando-se a característica soma das amplitudes. Concluindo, embora CDA e DDA sejam mais sensíveis do que TTP para identificar o ER.EDRs, nem o método CDA nem o método DDA forneceram maior poder discriminativo para o estudo de quatro intensidades de estimulação nocivas quando comparados ao método tradicional, TTP.

Palavras-chave: resposta galvânica da pele, dor, mensuração da dor, nocicepção, dor nociceptiva, psicofisiologia.

SUMMARY

1 INTRODUCTION

Pain is a very individual sensation and difficult to interpret without any communication from the patient. There is only a weak correlation between the subjectively reported pain and the tissue lesions or other pathological alterations, in certain circumstances. In other cases, pain may even be completely independent of any other alteration found. Therefore, somatic pathology does not allow any conclusion on the patient's subjective pain intensities (Niese, Al-Hamadi, & Michaelis, 2007; Nilges and Traue, 2007).

Typically in hospitals and intensive care units, there is a priority to control the pain and discomfort of their patients. And the only way to ensure that patients receive the best possible relief is to count on what clinicians and researchers call the most reliable indicator of pain: the patient's self-report of the pain sensation, whenever it can be obtained. Thus, pain assessment is important for establishing the efficacy of planning and treatment programs implemented for each individual (Jaywant and Pai, 2003; Norman & Judkins, 2004). A careful pain management plan helps to circumvent potential risks for critically ill and psychologically disturbed patients (Malenfant et al., 1996; Martin-Herz, Thurber, & Patterson, 2000; Ullrich, Askay, & Patterson, 2009).

For the monitoring of pain and for faster communication between patients and their caregivers, it is common to use one-dimensional pain scales such as the Visual Analogue Scale (VAS) or the Numerical Rating Scale (NRS) (Silva and Ribeiro-Filho, 2006). However, the main problem related to the use of pain scales is the fact that there is no easy method to measure pain directly nowadays. The examiner should always count on the qualitative description of the patient about the location, quality and intensity of the sensation of pain. These methods work, however, only when the patient is sufficiently alert and cooperative, which is not always the case in the field of medicine (for example, in the post-surgical phases and comatous states induced or not). In general, although useful, all methods have limitations and still need to be improved. In addition, the development of tools for the objective and automatic measurement of pain would be able to provide continuous monitoring of the patient's condition and not only in moments of pain exacerbation as can occur when scales are used by clinicians for the monitoring of pain intensity (Lucey et al., 2011).

Although one-dimensional scales are probably the most commonly used tools to assess the intensity of pain in the daily clinic (Sari, Gulbandilar, & Cimbiz, 2012), they are still prone to bias by other factors such as mood and predetermined levels of pain, leading to imprecise or dubious outcomes, once it is impossible to blind the patient when using self-reported one-dimensional scales (Tang et al., 2008; Wagemakers et al., 2019).

Since 1990s the American Pain Society (APS) had advocated a campaign that supported the necessity of monitoring the pain sensation more often in daily routines as it was normally done with the four vital signs (body temperature, blood pressure levels, pulse and respiratory rate), in order to make it an important measure of well-being (American Pain Society Quality of Care Committee, 1995). Thus, they advocated the widespread usage of one-dimensional tools to reduce the under assessment and inadequate treatment of pain in the United States of America, and they were supported by many medical societies, regulatory organizations and pharmaceutical companies over the following years (Mandell, 2016). However, it is said that this campaign relying on the one-dimensional pain scores had also unintentionally fostered the opioid epidemics of the past decades in America (Becker & Fiellin, 2017). In addition, they recognized in 2016 that although they strongly recommend the use of those validated scoring systems, they acknowledge that the evidences on their usage are week and other points should be addressed as well, as it is done in other multi-dimensional tools (Levy, Sturgess, & Mills, 2018).

In this sense, the search for new tools that could be able to provide quantitative information on someone's pain sensation, regardless of the conscious person's ability to perceive and to describe his or her feelings and regardless of the guesses and the observational skills of clinicians and allied health professionals, would be desirable to study the pain phenomenon, to establish patterns, normal and abnormal values and to correlate them with potential risks and better treatment outcomes. Many authors support the idea that heart rate variability (HRV), functional magnetic resonance imaging (fMRI), electroencephalography (EEG) and electromyography (EMG) are generally considered to be more accurate and objective measures for pain quantification than conventional one-dimensional pain measurement tools (Wagemakers et al., 2019).

A recent review from Ledowski (2019) on the commercially available objective nociception monitors, which are commonly used in surgical centers to monitor nociception and anesthesia

levels, shows that out of seven devices, one is based solely on skin conductance measures (Skin conductance, MedStorm innovations, AS, Oslo, Norway), and another one (NOL index, Medasense, Ramat Gan, Israel) encompass four different sensors, including galvanic skin responses, and the others usually use at least one measure of sympathetic activity. Although the author states the necessity of automated and objective anesthesia states monitoring in intra-operative settings, he claims that despite reflecting intraoperative stimuli slightly better than traditional blood pressure and heart rate (HR) parameters, none of them shown clinically relevant benefits to be used in clinical routines yet.

Recordings of electrodermal responses (EDRs) are mainly based on the measurement of skin conductance. Skin conductance changes with the activity of the sweat glands. This response of the skin is referred to as electrodermal activity (EDA), and it is considered to reflect the activity of the sympathetic branch of the autonomic nervous system. EDR is a way to measure sympathetic responses to mental or physical stimulation, particularly in response to noxious stimulation (Bach, 2014; Leknes, Brooks, Wiech, & Tracey, 2008). EDR can be described as a slowly varying monophasic signal that may change to a fast increase in its amplitude with slow recovering tails. These abrupt deflections can be related to either internal arousal processes or external stimuli (event-related electrodermal responses - ER.EDRs). Due to the slow recovery process following an EDR, studies using varying interstimulus intervals (ISIs) showed that responses following stimulation with ISIs as low as 2 s cannot be properly handled by the traditional trough-to-peak analysis (TTP) because such stimulation elicits responses that are superimposed on one another and appears as a single EDR in the raw signal (Benedek & Kaernbach, 2010a, 2010b). To overcome this problem, decomposition methods by means of deconvolution can be applied to sudomotor nerve activity (SNA) data (Bach, Daunizeau, Friston, & Dolan, 2010; Bach & Friston, 2013; Benedek & Kaernbach, 2010a, 2010b).

SNA and EDR may serve as important sources of information, especially in scenarios involving non-communicative patients in which pain is often inadequately identified and treated (Lichtner et al. 2014; Lucas et al. 2016; Walther-Larsen et al. 2017). Experimental noxious stimulation of healthy participants is used to better understand the nociceptive mechanisms involved in pain perception. This stimulation can be done by means of different agents, including intense pressure, subcutaneous injection of chemical substances, electrical stimulation, and extreme temperatures in both cold and hot domains. One of the most used

modalities is the heat stimulation, in which temperatures above 40 to 45 °C are expected to activate thermal nociceptors and lead to the perception of pain depending on the frequency of action potentials, temporal summation, and central modulation of afferent signals (Dubin & Patapoutian, 2010).

Although some studies have mentioned the low specificity and low predictive values of EDR analysis in assessing pain phenomena or the response to noxious stimulation (Ahmed et al., 2012; Ledowski, Ang, Schmarbeck, & Rhodes, 2009; Strehle & Gray, 2013; Valkenburg, Niehof, van Dijk, Verhaar, & Tibboel, 2012), many studies have analyzed EDRs by themselves or in combination with other physiological responses to monitor the pain sensation and try to provide more objective measurements (Ahmed et al., 2012; de Jesus, Campos Júnior, Storm, Da Rocha, & Tristão, 2015; Günther et al., 2016; Kächele, Thiam, et al., 2015; Kächele, Werner, et al., 2015; Loggia, Juneau, Bushnell, & Bushnell, 2011; Sabourdin et al., 2013; Storm, 2013; Strehle & Gray, 2013; Treister, Kliger, Zuckerman, Aryeh, & Eisenberg, 2012; Walter et al., 2014; Werner et al., 2014).

1.1 Motivation and justification for conducting this doctoral research

Until the present moment, there is no commercial equipment capable of inferring the intensity of pain that someone is feeling or felt at any given time. So far, we have seen only devices designed to be used in surgical centers in order to control the levels of anesthesia. Several studies have been evaluating the correlation of a large amount of potentially useful markers, solely or combined, with the self-reported perceived pain intensity or with the observed estimation of someone's pain intensity. The objective measurement of pain will allow the accurate diagnosis and to remotely monitor at least the approximate intensity of the pain sensation someone is experiencing, as well as it is already possible to be done with the other vital signs currently with the advent of smart watches and many other portable devices. Obtaining and storing accurate pain data will enable the appropriate treatment of collaborative and non-collaborative patients in the future. To the best of our knowledge, skin conductance is widely used an accepted as one promising candidate to fulfill the requirements for a commercial portable pain monitoring device, especially because of its inexpensive hardware requirements, as well as its simple analytic procedures. Although controversy information about its efficacy and specificity still exist, devices to monitor anesthesia status are already

commercially available, and therefore, we strongly believe that this is a worthwhile methodology to be further analyzed for pain measurement purposes.

1.2 Problem to be addressed

Although skin conductance might be an easy-to-use tool even in clinical settings, recent papers addressed the benefits of decomposition methods and new data processing techniques on the numeric features extracted from the EDA. So far, the authors are not aware of how this new techniques have been implemented on the study of pain sensation. Therefore we compared two decomposition methods with traditional though to peak analysis in order to see if they provide further discriminative power among several noxious stimulation intensities.

1.3 Objectives

1.3.1 General objective

Our aim is to compare the efficacy of two new methodologies in the field of EDA analysis against the traditional methodology, in detecting and discriminating four different noxious heat thermal stimulation intensities.

1.3.2 Specific objectives

- To study the performance of new features on detecting a noxious stimulation;
- To study the performance of new features on discriminating four noxious stimulation intensities, and;
- To compare the performance of commonly extracted features using traditional method against two new decomposition methods.

2 THEORETICAL BACKGROUND

2.1 Pain and nociception

According to the definition of the International Association for the Study of Pain, pain is a multidimensional and unpleasant experience, involving not only a sensory component, but also an emotional component, which are related to a concrete or a potential tissue damage. As part of the process for the recognition of pain, nociception represents the processing by the nervous system of harmful or potentially harmful stimuli detected by specific sensory afferent neurons present both in the skin and in the internal organs, the nociceptors (IASP, 2017).

Nociceptors are primary afferents with poorly specialized terminations and elevated activation thresholds (free nerve endings - FNE) capable of recognizing mechanical, chemical or thermal stimuli, whose cell bodies are in the dorsal root ganglion or in the trigeminal ganglion and whose central extensions rise through the posterior horn of the spinal cord. Therefore, their function is to recognize and to conduct information of potentially harmful stimuli to the biological tissues such as skin, joints, muscles and viscera (Lopes, 2003).

There are basically three types of sensitive fibers, of which only two conduct information of harmful stimuli in normal situations. Aδ fibers are poorly myelinated and they usually make direct connections with second-order neurons, which transmit information to the upper centers leading information of immediate acute pain, while C-type fibers are amiellin and usually communicate indirectly with second-order neurons through interneurons presented in the medulla, conducting more diffuse information about the harmful stimulus. Finally, the Aβ fibers are highly myelinated and transmit information of mechanical stimuli of more specialized nerve endings with low activation threshold such as tact and vibration. However, the latter may be involved in the perception of pain during inflammatory processes or after the restoration of traumatic injuries (Babos, Grady, Wisnoff, & McGhee, 2013; Milligan & Watkins, 2009).

The information from the nociceptors is processed at spinal and supraspinal levels, providing detailed information about nature, intensity, location and duration of the harmful stimulus. The stimuli from the nociceptors are driven to the brain by three major ascending tracts in the

white matter of the spinal cord: the neospinothalamic tract; the paleospinothalamic tract, and; the spinoreticular tract.

The neospinothalamic tract transmits information directly to the posterior lateral ventro nucleus of the thalamus, providing discriminative sensory information of pain perception, while the paleospinothalamic tract has communications with the periaqueductal gray matter, with the hypothalamus and with the reticular system in the mesencephalon before reaching the medial portion of the thalamus, mediating autonomic and emotional components of pain. The spinoreticular tract goes from the spine towards the reticular formation of the medulla to the pons and mediates aversive components of pain. Third-order neurons protrude from the lateral and medial posterior ventral nuclei of the thalamus to the higher centers such as the somatosensory cortex and the limbic system, respectively (Fong & Schug, 2014; Lopes, 2003).

Currently, researchers consider the existence of three endogenous mechanisms involved in the modulation of pain perception: segmental inhibition; the endogenous opioid system, and; the downward modulation. The gate control theory says that the upward information of Aβ sensory fibers competes with the signs of the nociceptors at the spinal level, inhibiting the propagation of harmful stimuli to the upper centers (Babos et al., 2013).

More specifically on the descending pathways, they modulate the conduction and processing of information from nociceptors in the posterior horn of the medulla by releasing excitatory neurotransmitters such as glutamate, P substance and peptide related to the calcitonin gene (CGRP), or by the release of inhibitory neurotransmitters such as GABA, endogenous opioid agonists such as enkephalins and endorphins, norepinephrine and serotonin. They are stimulated mainly by areas of the mesencephalon, such as the periaqueductal gray matter, the rostroventromedial medulla and the tegument of the pons, but also by higher structures such as the amydala and the limbic cortex in case of inhibitory modulation. The endogenous opioid system consists of chemical mediators such as enkephalins, endorphins and dimorphins, which act directly on specific receptors of the neurons of the periaqueductal gray matter and the ventral medulla, as well as in several points of the spinal cord (Fong & Schug, 2014; Lopes, 2003).

Figure 1 illustrates in a schematic way a large part of the anatomical structures involved in the

processing of harmful stimuli and in the perception of pain and its interrelations for a better understanding of what was presented.

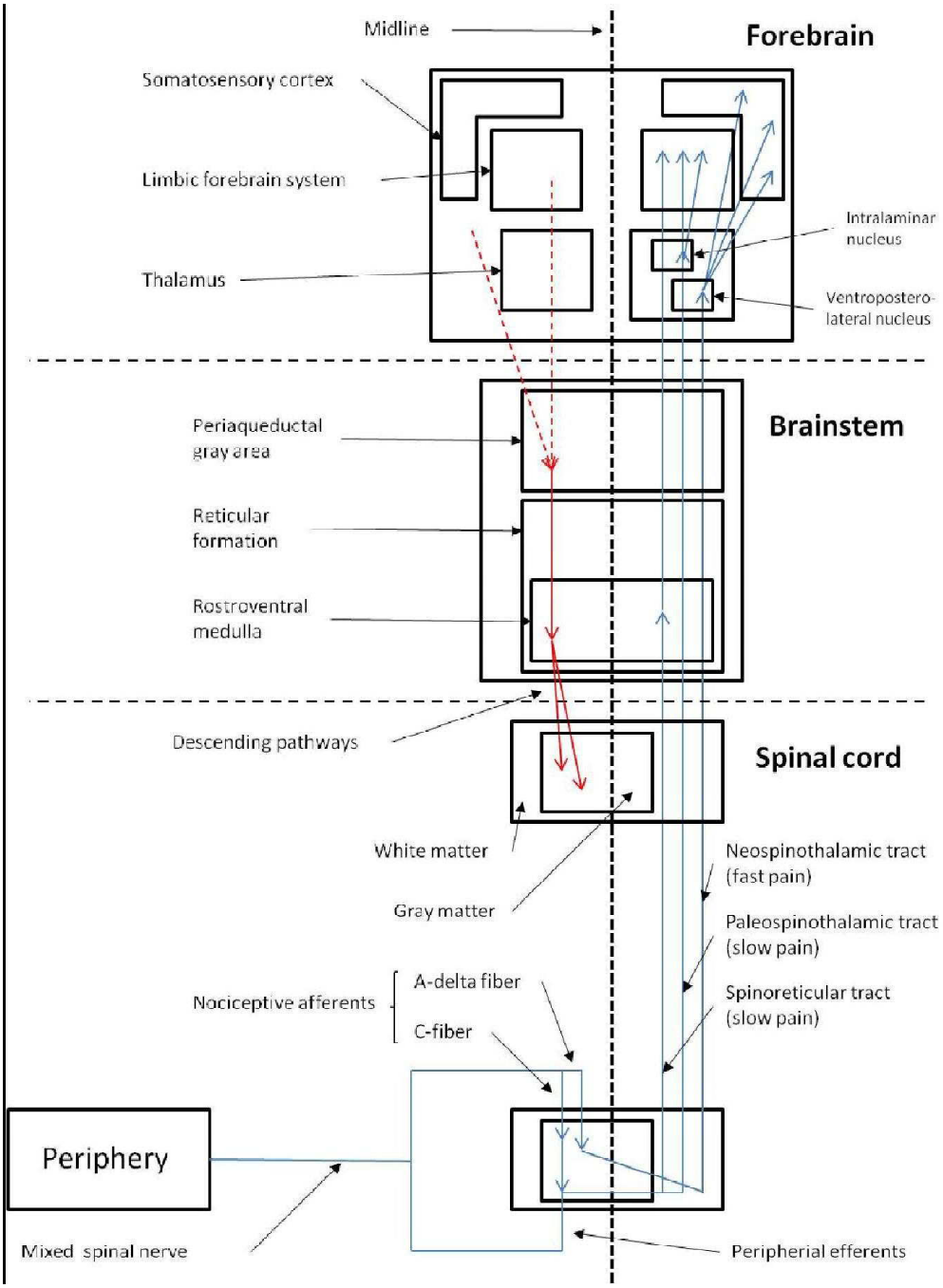

Figure 1. Schematic illustration of several structures involved in pain perception and modulation. The afferent pathways in blue represent several neurons and their central nervous system connection sites. In red, the descending modulatory pathways are represented.

In summary, this topic aimed to describe the main physiological mechanisms and anatomical structures involved in the perception of pain in humans. Thus, we observed that pain perception involves processes of stimulus recognition, information conduction, modulation (sensory, affective and autonomic) and awareness. Although the identification of the harmful stimulus and the presence of protective reflexes are present even in the absence of detailed conscious information about the triggering stimulus, the subjective sensation of pain experienced and described by the patient is influenced by other time-dependent aspects, which tend to worsen with the persistence of the triggering agent, and may persist in the future, even after the initial source of pain is extinguished. This information is supported by the involvement of several central nervous system (CNS) areas related to emotions and to the neurovegetative system, and by studies involving chronic pain issues. Since the experience of pain is unpleasant and aversive, human beings seek to avoid it or minimize it in most cases (Committee on Recognition and Alleviation of Pain in Laboratory Animals, 2009)

2.2 Assessment tools for the study of pain sensation

Although the experience of pain is complex and subjective, it is important all health professionals to be able to accurately monitor and record the pain experience of their patients, since this information will be necessary to guide physical treatments, to monitor the patient's evolution and to evaluate the efficacy of the treatments that were implemented. And although there is still no reliable and valid form for the objective measurement of pain, these records can be made by several available instruments, which provide at least an estimate of the pain experience of a patient, either by self-report or by clinical observation (Younger, McCue, & Mackey, 2009).

2.2.1 One-dimensional scales

One-dimensional scales are the most used tools for the clinical evaluation of pain (especially acute pain) due to the simplicity of use and the speed of obtaining information, and can be used frequently in reassessments without the need of much effort or time. However, they are used to monitor only one component of pain (usually pain intensity). Among them are the NRE, the VAS, the verbal rating scale (VRS) and the facial pain scale (FS). In the first two,

values between 0 and 10 or 0 and 100 represent the intensity of pain ranging from the absence of pain to the worst possible pain at their extremes.

They work simply by asking the patient to speak aloud a number or to mark somewhere in between the extremities of the tool markers, which are usually composed by a 10 cm straight segment without any visible numerical marking for the patient, being the distance between the starting point and the marking made by the patient measured by the examiner afterwards. In the last two cases, verbal descriptors such as “no pain”, “mild pain”, “moderate pain” and “severe pain” and images representing such descriptors by means of face designs are used to graduate the patient's pain experience when the patient presents any difficulty in translating his or her sensation into a number (Da Silva, Ribeiro-Filho, & Matsushima, 2010).

2.2.2 Multidimensional scales

Multidimensional scales encompass the evaluation of multiple dimensions of the pain experience, such as intensity, functional limitations, social limitations, affective impairments, emotional state and quality of life. They are usually presented in the form of questionnaires, inventories and/or formularies. Because they provide a broader approach, they are often used to map relationships between disabilities and other aspects rather than the intensity of pain, such as social participation, psychological impairments, and many others. Among the multidimensional scales, one can list the initial pain assessment inventory (IPAI), the brief pain inventory (BPI), the pain perception profile (PPP), the remembered pain assessment scale (MPAC), the descriptor differentiating scale (DDS), the research of pain treatment results (TOPS), the multidimensional pain inventory (MPI) and the McGill pain questionnaire (MPQ-long version-and and SFMPQ-short version) (Da Silva et al., 2010; Younger et al., 2009).

The MPQ was developed by Ronald Melzack in 1975, and is considered by many authors to be the most widely used tool for pain assessment. In its short version, developed in the following decade (Melzack, 1987) are presented 15 descriptors that represent the sensory (11 first descriptors) and affective (4 last descriptors) dimensions of pain. Each descriptor is classified by the patient on a scale of 4 points (0 = none; 1 = mild; 2 = moderate, and; 3 = severe) according to the level that best represents his/her pain at the time of evaluation. From them, three measures can be obtained: PRI-S - sum of the sensory descriptors (ranges from 0

to 33); PRI-A – Sum of the affective descriptors (ranges from 0 to 12), and; PRI-T- sum of PRI-S and PRI-A (ranges from 0 to 45). There is also a VAS in which the patient should indicate the intensity of pain he or her is currently experiencing. Finally, the patient makes an overall assessment of his painful experience (evaluative dimension of pain), choosing one of the following alternatives on a scale of 6 points: 0 = no pain; 1 = mild; 2 = discomforting; 3 = distressing; 4 = horrible, and; 5 = excruciating. In total, the application of the short version of MPQ takes about 10 minutes (Menezes Costa et al., 2011; Da Silva et al., 2010).

2.2.3 Behavioral scales

There are also behavioral scales, usually employed with unconscious or non-communicative patients, for which it is impossible to obtain a self-report on the sensation of pain. They can be both uni- or multidimensional, in which health professionals should evaluate facial expressions, body movements and vocalizations, among other things. However, although they are necessary in specific cases, they are fairly accepted for the publication of clinical trials. Among them, the behavioral scale of pain assessment (BPRS), the behavioral scale of pain (BPS), the behavioral pain assessment tool (PBAT), the critical care pain observation tool (CPOT), the non-verbal pain scale (NVPS), and the pain assessment and intervention rating algorithm (Pain Algorithm) (Li, Puntillo, & Miaskowski, 2008; Younger et al., 2009).

2.2.4 Bio and physiological indicators of pain

In the case of biological or physiological indicators of pain, changes in HR, respiration, sweating, muscular tension and blood pressure were already reported in scientific papers with regards to increased painful sensation. However, all these indicators tend to return to values close to normality as the body tries to regain homeostasis, even though the pain persists (Da Silva et al., 2010).

Cowen and Collaborators (Cowen, Stasiowska, Laycock, & Bantel, 2015) published an excellent review on the biological indicators of pain, which were classified into five different strategies for the objective measurement of pain: alterations of the autonomic nervous system; biopotentials; neuroimaging; biological markers, and; compound algorithms.

2.2.4.1 Autonomic function

Among the measures of the autonomic function most commonly used for the study of pain are measures of the variability of HR, pressure alterations and peripheral pulsation, EDA and pupilal reflexes. The main justifications for the use of these measures are evidence of an overlap between the nociceptive pathways and the autonomic pathways and the increase of circulating hormones related to stress in response to pain (Chang, Ma, Tsay, & Jong, 2012; Cowen et al., 2015; Gruenewald et al., 2013; Koenig, Jarczok, Ellis, Hillecke, & Thayer, 2013; Loggia et al., 2011).

2.2.4.2 Biopotentials

Biopotentials are electrical potentials that lead and transfer information between living cells, and that can be measured by means of sensors both on the body surface and by implanted sensors, very close to the cells. They have been used to monitor withdrawal reflex thresholds, evoked potentials after specific sensory stimuli, levels of cerebral activity in specific regions and the bispectral index (derived from cortical encephalography parameters) (Coleman et al., 2013; Cowen et al., 2015; Fabrizi et al., 2011; France, Rhudy, & McGlone, 2009; Zhang, Hu, Hung, Mouraux, & Iannetti, 2012).

2.2.4.3 Neuroimaging

Neuroimaging methods evaluate the neuronal function correlated with the morphology of the nervous system, allowing to map the activated areas during the experience of pain in terms of intensity, location, quality and intensity of the stimulus. As shown in the item (2.1), several regions of the cerebral cortex, the midbrain and the brainstem are activated repeatedly during harmful stimuli. Among the neuroimaging methods used to map these regions of the nervous system, we can cite the Positron Emission Tomography (PET), the fMRI and the Near Infrared Spectrometry (NIRS). These techniques measure changes in blood flow and in the consumption of glucose and oxygen in the central nervous system through different techniques. PET detects radioactive isotopes injected into the bloodstream by gamma ray emission. FMRI identifies changes in the magnetic properties of oxyhemoglobin and of deoxyhemoglobin or of water protons present in arterial blood marked by radiofrequency

pulses. Finally, NIRS detects changes in the absorption level of two infrared spectra by Oxyhemoglobin and Deoxyhemoglobin (Cowen et al., 2015)

2.2.4.4 Biological markers

Biomarkers or biological markers are practically anything that can be objectively measured and evaluated as an indicator of the occurrence of a physiological process. Thus, they can be represented by many things from genetic mappings to simple clinical scales. But their main contribution in the study of pain is related to the possibility of studying molecules and metabolic products at the cellular level. The main substances that have been analyzed as potential markers of autonomic function related to pain have been blood levels of catecholamines, cortisol, adenocorticotropic hormone and free fatty acids in addition to alpha-amylase and insulin sensitivity. Another point of interest is the study of pro- and anti-inflammatory cytokines (Angst et al., 2008; Cowen et al., 2015; Ferrara et al., 2013; Krikava, Kalla, Yamamotová, & Rokyta, 2004; Ledowski, Reimer, Chavez, Kapoor, & Wenk, 2012; Üçeyler, Häuser, & Sommer, 2011).

2.2.4.5 Compound algorithms

The last approach analyzed for pain quantification refers to the use of algorithms, encompassing different parameters, which has been suggested to better represent the complex nature of the sensation of pain. Thus, combinations of multimodal signals, including HRV, EDA, photoplethysmography, facial expressions among others have been performed to generate a single index capable of representing the sensation of pain or the nociception processes of post-surgical patients, anesthetized and during experimental pain induction (Ben-Israel, Kliger, Zuckerman, Katz, & Edry, 2013; Cowen et al., 2015; Sarén-Koivuniemi, Yli-Hankala, & Van Gils, 2011; Treister et al., 2012; Walter et al., 2013).

2.2.5 Conclusions

Despite the numerous pain assessment tools currently available, there is still no validated and accepted tool for the objective measurement of pain in the most diverse contexts, in the same

way that there is no consensus on the tools, results and crucial domains needed for future research in the objective measurement of pain and pain management in clinical settings (Cowen et al., 2015; Mahar et al., 2012).

2.3 Author's preliminary investigations on the study of pain and pain intensity measurement

In this section we would like to highlight other pieces of work that were developed by the doctoral candidate along with the main research project. Hence, we provide a short abstract of every paper that was written and presented in scientific conferences or published in scientific journals as a consequence of our studies in the fields of biomedical engineering, biomedical signal processing, pain assessment and measurement. These include studies in both clinical and experimental settings, as well as reviews and epidemiological studies with regards to pathological conditions and treatments related to pain assessment and management.

2.3.1 Perspectives on the Objective Pain Assessment and Measurement Through Multimodal Signals in Burn Patients (2014)

Major thermal injuries are a significant cause of pain and disability. All burn victims experience pain that can be modulated by psychological factors. In many cases, the treatment provided to patients is the continuous intravenous infusions of opioids. An overdose of such drugs may produce side effects. Subjective description of pain is the most common assessment to control drug's dosage. In this context, it's proposed to develop and assess a method, based on the analysis of multimodal signals, for the objective quantification of pain in patients after severe burn. The objective measurement of pain is still a problem that has not been adequately solved. The estimate of objective pain patterns may be used in many circumstances, such as for the controlling of automated infusion pumps, the decision of the type of analgesic administered to patients, and assessment of results of psychophysical therapies and other treatments.

Keywords: Biomedical Engineering; Pain; Pain measurement; Emotions; Burns; Biomedical Signals; Vital Signs; Facial Expression; Pose and paralinguistic signals.

2.3.2 On the use of biomedical parameters for the objective pain assessment in severe burn patients: a case study (2014)

Pain is a major problem in severe burn patients and it causes great physical and psychological suffering together with the burn injury. Biological parameters such as vital signs have been suggested as pain indicators to monitor pain, but just a few studies have attempted to study their variability as a possible source of information. Therefore, we aimed to evaluate different biomedical parameters for pain recognition in severe burn patients based on a case study. Vital signs, oximetry and blood pressure were collected together with Verbal Numeric Scale for four distinct conditions from a burn inpatient. Variables were arranged in ascending order regarding Pain Sensation values. Significant differences were found for the four different conditions. Heart Rate was found to be significantly different between baseline and increased pain values in three of them. Pulse and Respiration Rate variances showed consistent patterns in all four situations. All in all, we conclude that variance values of respiration rate and Pulse might be probably the main parameters among the analyzed biological parameters that might be used to identify and monitor increasing pain sensation in burn patients. Despite finding significant differences in other parameters, they may fluctuate during different conditions and should be further analyzed in a larger sample to bring more insights to the field.

Keywords: Burns, Oximetry, Pain, Pain Measurement, Vital Signs.

2.3.3 Assessment of the linear correlation between vital signs and perceptual pain in the context of burn individuals (2014)

Burns are one of the worst injuries to humans, due to intense pain related to initial injury itself, wound care and other physical treatments which can least from days to months. It can lead even to death or provoke physical impairments and psychological suffering. A number of studies suggest the existence of correlations between biological signals and the presence of pain and its severity. Therefore, we aim to assess the linear correlation between vital signs and perceptual pain in the context of burn individuals. Vital signs, oximetry and blood pressure were collected together with the Verbal Numeric Scale (VNS) pain measure for four different conditions (wound care [P1], physiotherapy [P2], and at rest 2 [R1] and 5 [R2] hours after

opiate medication administration) from one inpatient at a the Burn Unit of the Clinical Hospital of the Federal University of Uberlandia, Uberlandia, Brazil. Linear regression and correlation coefficients were evaluated to compare linear trend models of each variable among all four conditions. Moderate correlations were found for Pulse and HR in conditions R1 (negative correlation) and R2 (positive correlation), for MAP and DBP in condition R1 (positive correlation) and for O2 in condition R2 (negative correlation). Larger correlation and slope were found for Pulse and HR at rest in the absence of medication effects. We conclude that painful procedures and opiate medication may influence the linear trend of biological parameters in the context of burn patients, which should be further analyzed.

Keywords: Burn, Oximetry, Pain, Pain Measurement, Vital Signs.

2.3.4 Correlation between pain and biomedical signals in severely burnt individuals: preliminary results (2014)

The objective measurement of subjective, multi-dimensionally experienced pain is a problem for which there has not been found an adequate solution yet. Although verbal methods (e.g., pain scales and questionnaires) are commonly used to measure clinical pain, they tend to lack objectivity, reliability, or validity when applied to e.g. mentally impaired individuals. Biomedical signals and behavioral parameters may represent a solution. Such coding systems already exist, but they are either very costly or time-consuming, or have been not been sufficiently evaluated. In this context, we measured multimodal biomedical signals during the treatment of a patient with severe pain in the Burn Unit of the Clinical Hospital of the Federal University of Uberlandia (Brazil). We found convincing correlation between pain and vital signs related to the cardiac activity (e.g., blood pressure). Our goal is to carry out a large clinical study with severely burnt victims with the aim of developing a multimodal automatic burn recognition technology to medicate patients as early as possible.

Keywords: multimodal automatic pain recognition, burn victims, analgesics.

2.3.5 Calibrating the open source E-health sensor platform developed by Cooking-Hacks, Spain (2015)

The aim of this study is to describe the calibration process and results of the E-health Sensor Platform v2.0. Pulse oximeter (pulse rate and oximetry) and temperature sensor readings were tested against standard certified medical devices by means of statistical correlations and linear models evaluation, using the company's provided libraries over Arduino board. Strong correlations were found for the three evaluated parameters. However, issues regarding pulse oximeter readings within specific ranges were also found. The sensor platform provides reliable data, nonetheless, calibration process is always needed in order to identify malfunctions in advance.

Keywords: Body Temperature; Calibration; Heart Rate; Oximetry; Pulse; Skin Temperature.

2.3.6 Physical Therapy Pain Management in Burn Patients: a Systematic Review (2015)

This systematic review aimed to summarize physical therapy knowledge contributing to pain management of severe burn patients. Therefore, a systematic search for the combined terms "Pain Management", "Burns" and "Physical Therapy" or "Physiotherapy" was carried out in October, 2013 within PUBMED, IEEExplore and PEDro databases. From a total of 10 papers, three were included. They were all about Virtual Reality interventions. As a result was concluded that there is a lack of studies exploring pain relief possibilities of physical therapy techniques in burn patients. Virtual Reality has been proof to be a reliable tool to help burn patients especially during painful activities. Other techniques such massages, biofeedback, Transcutaneous Electrical Nerve Stimulation, muscle relaxation and exercises have been used, but poorly reported.

Keywords: Physical Therapy; Pain Management; Burns.

2.3.7 Burns in Triângulo Mineiro (Brazil): epidemiological study of a burn unit (2016)

Objective: To characterize the Burn Unit and its inpatients between January 2006 and December 2013. Methods: A descriptive, observational, retrospective epidemiological study with quantitative approach of the Burn Unit and its inpatients was performed with data from the hospital statistical service. Results: In general, the profile of the Burn Unit inpatients seen in the past eight years are adults in working age and children under 10 years old, victims of accidents with flammable and heated liquids, presenting second degree burns in about 10 to 19% of body surface area, especially on the trunk. The highest number of new admissions (81) occurred in 2010, with the highest occupancy rate in 2007 (67.2%) and lowest in 2008 (33.14%), the same year in which the highest mortality rate (7.2%) was observed. In contrast, the lowest mortality rate was observed in 2006 (1.6%). About 40% of the total admissions lasted less than ten days. From the 517 surgical procedures carried out inside the unit, 20% were dermal-epidermal grafts. Conclusion: There were little changes on the epidemiological profile of the delivered treatments in this unit when compared to the previous decade. However, physical and methodological adjustments should focus on child and on adults' occupational perspective.

Keywords: Burns. Epidemiology, Descriptive. Burn Units.

2.3.8 Pain-related heart rate variability responses in the context of experimental pain stimulation (2016)

Objective pain assessment is still an ongoing issue in health sciences, and the analysis of Heart Rate Variability (HRV) is considered a promising tool to provide quantitative information regarding pain sensation. However, commonly used HRV analysis comprises features determined in longer time-periods (at least five minute length recordings), which might be impractical for the development of some online monitoring systems. Thus, we aimed to analyze 10 time-domain features using ultra-short HRV analysis in the context of thermal-induced painful stimulation. Electrocardiographic data from 85 participants from the Biovid Pain Database were analyzed. Ten time-domain features were extracted from pre- and post-stimulation periods using 10-second duration windows. Then, after normal distribution assessment with Lilliefors normality test, we compared intra-group variability by means of paired t-test. Three features (SDNN, RANGE and HR) significantly increased after stimulation, while two features (pNN20 and MEAN) decreased. Further, the higher the

stimulation intensity, the larger the number of sensitive features. Although we suppose that the analysis of lower intensities stimulation was impaired by the short inter-stimuli interval duration, SDNN, RANGE, pNN20, MEAN and HR seem to be feasible time-domain features to assess experimental pain by means of ultra-short HRV analysis.

Keywords: Biomedical Signal Processing, Heart Rate Variability, Pain Measurement.

2.3.9 Facial and shoulder electromypgraphic response to heat stimulation at distinct intensity levels (2016)

Reliable objective pain assessment is necessary to provide adequate pain relief treatments, especially in case of non-communicative patients. In this sense we address the use of electromyography (EMG) in the context of experimental pain induction. Thus, we aimed to analyze whether thermal painful stimulation can elicit stable electromyographic responses among several random stimulations for the Corrugator, Zygomaticus and Upper Trapezius muscles. Bipolar differential surface EMG were acquired with a sampling rate of 512 Hz, gain of 24, and ADC resolution of 24 bits. Raw data were processed in Matlab with 4th order Butterworth bandpass (5-256 Hz) filter and 20th order polynomial filters. Then, the Hilbert transform was used to estimate the instantaneous amplitude, which was processed by a thresholding algorithm, so that only responses with prominences larger than 5% of the entire signal's range were counted. Among Corrugator, Zygomaticus and Trapezius muscles, Zygomaticus was the muscle in which the responses were more often identified (over 47%), regardless of the stimulation intensity. Thermal painful stimulation at the painful threshold level elicited electromyographic responses in only 20.7% (Corrugator), 31.3% (Zygomaticus) and 32.7% (Trapezius) of the total number of stimuli, while thermal painful stimulation at the tolerance level elicited electromyographic responses in 56.2%, 69.8%, and 54.2% of the total number of stimuli, respectively. We observed that the higher the stimulation intensity, the larger the recognition rates of electromyographic responses for all the three muscles. We concluded that, although using randomized time and intensity stimulation design, we didn't get stable EMG responses with respect to thermal pain stimulation.

Keywords: Biomedical Signal Processing, Electromyography, Pain Measurement.

2.3.10 Increased flexibility, pain reduction and unaltered levels of IL-10 and CD11b + lymphocytes in patients with systemic lupus erythematosus were associated with kinesiotherapy (2018)

The effect of physical activity on the immune system is still poorly understood in cases of systemic lupus erythematosus (SLE). Therefore, our aim was to investigate differences in the serum levels of cytokines (IL-2, IL-5, IL-6, IL-8, IL-10 and TNF-α) and the numbers of CD11b+ CXCR2+ neutrophils and lymphocytes in women with SLE undergoing drug treatment, without (n=9) or with (n=5) 4 months of kinesiotherapy. Parameters related to functional capacity were also analyzed. In the case of the patients who were not submitted to kinesiotherapy, there were reductions in the levels of IL-5, IL-6 and IL-10, and an increase in the number of CD11b+ leukocytes, in addition to an increase in abdominal circumference after the monitoring time. Patients submitted to kinesiotherapy did not present changes in serum cytokines or in the numbers of CD11b+ and CXCR2+ neutrophils and lymphocytes, but there were increases of flexibility and strength, as well as a reduction in pain sensation after the monitoring time. In conclusion, kinesiotherapy was able to increase flexibility and reduce pain in SLE patients without influencing immune parameters.

Keywords: Systemic lupus erythematosus; physical exercise; kinesiotherapy; cytokines; CXCR2; CD11b; phagocytosis; functional capacity

2.3.11 Comparative evaluation of methods for the detection of electrodermal responses to multilevel intensity thermal noxious stimuli (2019)

Purpose: In this study, three different methods to identify event-related electrodermal responses (ER.EDRs) were applied, comparing noxious heat stimulation at four different levels. Although the level of heat leading to subjective pain is individual, temperatures above 40 to 45 °C are normally experienced as painful due to the stimulation of heat-sensitive nociceptors. Methods: Skin conductance (SC) data from 96 healthy participants aged between 18 and 65 years old were analyzed by means of continuous decomposition analysis (CDA), discrete decomposition analysis (DDA), and trough-to-peak analysis (TTP). Several EDR features estimated from these methods were statistically compared. Results: Within the analysis window following each single stimulation, CDA and DDA methods identified more

electrodermal responses between 1 and 9 s after noxious stimulation than TTP for all stimulation intensities. However, the occurrence rates of at least one ER.EDR after noxious stimulation were similar among the three methods and tended to increase with increasing intensities. Among the common features, amplitude sum had better discriminative power for differentiating noxious stimulation intensities regardless of the method. Conclusion: The results suggest that all investigated methods performed similarly in identifying electrodermal changes in response to high-intensity thermal noxious stimuli. In the experimental conditions of cutaneous heat stimulation of nociceptors, good discrimination among stimulation intensities was found using the amplitude sum feature. In conclusion, although CDA and DDA were more sensitive than TTP for identifying ER.EDRs, neither CDA nor DDA brought further discriminative power to the study of noxious stimulation intensities when compared with the traditional TTP method.

Keywords Galvanic skin response. Nociception. Nociceptive pain. Psychophysiology

2.4 Electrodermal activity (EDA)

The term EDA was first introduced by Johnson and Lubnin (1966) as all electrical phenomena occurring in the skin and its appendages. EDA recordings can be either endosomatic, when only potential differences originating in the skin itself are monitored, or exosomatic, when direct (DC) or alternated currents (AC) are applied to the skin in order to measure other changes in electrical skin properties over the time. Main EDA recordings are: skin potential (SP); skin resistance (SR); skin conductance (SC); skin impedance (SZ), and; skin admittance (SY). Nowadays SP can be recorded by either endosomatic or AC methodologies, while SR and SC can be assessed by either DC or AC methodologies. SZ and SY can only be assessed by means of AC (Boucsein, 2012).

These recordings can be divided into tonic (level) and phasic (response). While phasic data are usually related with the existence of a distinct relationship between a known stimulus and an observable EDR in the electrodermal recording, tonic measures are normally obtained in response-free recording windows or as the count of non-specific electrodermal responses (NS.EDRs) that occurs within a recording window in the absence of any traceable

endogenous or exogenous stimulation (Boucsein, 2012; Boucsein et al., 2012; Handler, Nelson, Krapohl, & Honts, 2010).

Changes in those electrodermal recordings have been addressed to the activity of the sweat glands, and the relationship between their activity and the current flow in the skin dates from 1870s with the studies of Hermann and Luchsinger (1878) in Switzerland. By that time, there were also other theories which associated changes in electrdermal recordings with skin blood flow and involuntary muscle contractions. However these theories were overcome by the beginning of the 19th century, when correlations between EDRs and either plethysmographic changes or muscle tremors could not be proved (Boucsein, 2012).

First observations of the relationship between the electrodermal phenomena and psychological factors date from the 1880s, while monitoring hysterical patients and the use o anesthesia (Vigouroux, 1879), sensory and emotional stimulation (Féré, 1888), and while monitoring mental arithmetic, expectation, voluntary muscle contractions, imagination and sensory stimulation (Tarchanoff, 1889). Since then, it is one of the most used biosignals in psychophysiology due to the use of inexpensive equipment and the ease of obtaining distinct EDRs in response to external stimulation even in poorly controlled scenarios. Despite the widespread use of EDA measurements, their basic mechanisms related to anatomy, physiology, physics and psychology are not yet fully understood (Boucsein, 2012).

2.4.1 Anatomy and physiology of skin

The skin is the body's largest organ, and it corresponds to approximately 15% of the total adult body weight. It composes the integumentary system together with its derivate structures. From the inner to the outer level, the skin is composed by tree layers: the subcutaneous tissue or *panniculus*; the *dermis*, and: the *epidermis*. It is continuous with the mucous membranes lining the body's surface, and it performs many vital functions, including physical, chemical and biological protection, thermoregulation, prevention of excess water loss and sense function (Handler et al., 2010; Kolarsick, Kolarsick, & Goodwin, 2011).

The subcutaneous tissue is basically composed by fat cells, or lipocytes, and fibrous septa made up of collagen and large blood vessels. It functions as a storehouse of energy and its thickness varies depending on the location and anatomy of the body. It also plays an

important function as an endocrine organ by producing leptin, which helps to regulate body weight, and by converting androstenedione into estrone (Kolarsick et al., 2011).

The *dermis* is basically made up of collagen, which are fibrillar structural proteins that composes the connective tissues, which accommodates nerves and blood vessels networks, epidermal appendages, fibroblasts, macrophages and mast cells in the *dermis*. It is responsible for most of the skin pliability, elasticity and tensile strength. It also contains receptors of sensory stimuli (touch, pressure, temperature, pain and itch sensation), protects the body from mechanical injury, binds water and aids in thermal regulation (Kolarsick et al., 2011).

Between the *dermis* and the *epidermis*, there is a porous basement membrane zone, called the dermal-epidermal junction. The dermal-epidermal junction offers support for the *epidermis*, helps to establish cell polarity and direction of growth, directs the cytoskeleton structure in epidermal basal cells, provides developmental signals, and functions as a semi-permeable barrier between two layers (Kolarsick et al., 2011).

The *epidermis* is the most external layer, and it is normally divided into four stratified layers in accordance with the differentiation of its main cells, the keratinocytes. Many functional derivative structures such as nails, sweat glands and pilosebaceous apparatuses are originated in the *epidermis*. The basal layer is attached to the dermal-epidermal membrane. It is the primary location of mitotic activity that provides cells to the outer layers. It also contains the melanin pigments produced by the melanocytes, which distinguish the human skin color. Their cells form a single cell layer, and they are strongly adhered to one another, to the basal membrane and to the squamous cells in the second layer. The second layer is a 5 to 10 cells thick layer with a variety of shapes, structures and subcellullar properties. Its intercellular space is abundant in desmossomes that promote mechanical coupling and provide resistance to physical stresses. The third layer, the granular layer, is the last layer where we find living cells. They are flattened and rich in keratohyaline granules. The layer thickness can vary from 1-3 cells to up to 30 cells. The keratohyaline granules are important for the formation of the interfibrillary matrix and the inner lining of the horny cells, which maintain the keratin filaments together. Enzimatic action of the keratohyaline garnules also changes the softness or hardness (nails and hair) of the keratin filaments. The fourth layer is the cornified layer. The large, flat and polyhedral-shaped corneocytes have no longer nuclei, and they are technically considered to be dead. They are rich in protein and the continuous surrounding extracellular

matrix is rich in lipids. Thus, they provide good mechanical protection and water loss prevention (Kolarsick et al., 2011).

Finally, the skin *adnexa* group all epidermally derived appendages, including sweat glands, hair follicles, sebaceous glands and nails.

2.4.2 Sweat glands and perspiration

Perspiration takes places in many regions of the skin. It helps to keep the skin moist, contributing to the body's flexibility and to thermoregulatory processes. Sweating cools the outer skin surface by removing the latent heat from the outer layers when the liquid water evaporates. However, some regions like the palms on the hands and the soles on the feet are less likely to sweat in response to high ambient temperature. On the other hand, they are more responsive to CNS activation than the other areas (Handler et al., 2010).

Sweating has also a variety of other critical homeostatic functions unrelated to thermoregulation. Although not clear yet, sweat glands may play an important excretory function, similar to that of the renal system, clearing excess micronutrients, metabolic waste, and toxins from the body. In extenuous conditions sweating may also leads to micro-nutrient imbalances. Finally, preliminary immuno-histo-chemistry studies have being suggesting that sweat glands may produce and excrete antimicrobial peptides such as dermcidin, cathelicidin, and lactoferrin, pointing out to a potential role of sweating in host defense against skin infection (Baker, 2019).

So far, there are three different sweat gland types described in literature. The first one, the eccrine sweat glands, are spread all over the body, but especially concentrated on the palms, soles and forehead, and they are estimated to be around 2 and 5 million units throughout the body. They consist of a coiled secretory portion located either deep in the *dermis* or in the subcutaneous tissue, and a ductal discharge tube straight though the *dermis* and with a spiral course through the *epidermis* until their pore directly on the skin surface. Their ducts are formed by two to three layers of cells in the *dermis* and the epidermal portion, which is called acrosyringum, is only a coiled ducted surrounded by epidermal cells. This thin cell layer enables to hydrate the entire epidermis, improving its ability to conduct electrical currents (Handler et al., 2010). The secretory coil is composed of glycogen-rich clear secretory cells,

dark mucoidal cells, and myoepithelial cells with contractile properties. The initial formation of sweat in response to a sympathetic stimulation occurs in the glycogen-rich epithelial cells, while the darker mucoidal cells reabsorb sodium from sweat in the ducts, resulting in the extremely hypotonic solution that is emitted onto skin (Kolarsick et al., 2011). The highest gland densities are on the palms and soles (~250–550 glands/cm^2) and respond to both emotional and thermal stimuli. The density of eccrine glands on non-glabrous skin, such as face, trunk, and limbs is approximately 2 to 5 times lower than that of glabrous skin, and they are primarily responsible for thermoregulation. Eccrine sweat is mostly water and NaCl, but also contains a mixture of many other chemicals originating from the interstitial fluid and the eccrine gland itself (Baker, 2019).

The second type, the apocrine sweat gland, is rather involved in scent release than in thermal regulation, thus, being located mainly in the axillae and perineum regions. Their secretory coil is located exclusively in the subcutaneous tissue and it is composed of secretory cells solely with proteinaceous, lipid-rich and viscous content, which are activated during puberty. The intraepithelial ducts discharges directly into the pilosebaceous follicles (Baker, 2019; Kolarsick et al., 2011).

Another type of sweat gland is the apoeccrine sweat gland. They develop from eccrine sweat glands during the puberty and they are present in the axillae of aduts, representing up to 45% of the total amount of axilary glands at the age of 18 (Baker, 2019). They are similar to eccrine sweat glands, opening directly to the skin surface, but with a secretory rate many times higher than those eccrine ones. Thus, they have been associated with axillary hyperhidrosis, an abnormal condition with increased rates of perspiration (Kolarsick et al., 2011).

2.4.2.1 The sweating action of the eccrine sweat glands

Eccrine sweat glands primarily respond to thermal stimuli, including increased core temperature, skin temperature and increased skin blood flow. Central thermoreceptors in the abdominal region and in the muscles as well as skin thermoreceptors send information to be processed by the preoptic area of the hypothalamus that trigger the sudomotor response, which is mediated mainly by sympathetic cholinergic stimulation. Sweating is stimulated

through the release of acetylcholine from nonmyelinated class C sympathetic postganglionic fibers, which binds to muscarinic receptors on the sweat gland. Eccrine glands may also be stimulated by adrenergic stimulation. In addition, eccrine sweat glands also respond to exercise-related stimuli mediated by feed-forward mechanisms related to central command, muscle metabo- and mechanoreceptors, osmoreceptors, and baroreceptors (Baker, 2019).

Sweating rate over the whole body surface area is related to both density of active sweat glands and the secretion rate per gland. First response is a rapid increase in sweat gland recruitment, followed by a gradual increase in sweat secretion per gland. Two important aspects are involved in thermoregulatory sweating, namely the body core temperature threshold shifts, which are supposed to occur in hypothalamus and the sensitivity of the sweat glands' response to hyperthermia, which is supposed to takes place locally in peripheries (Baker, 2019).

2.4.2.2 Physiological functions of palm and sole sweating and its relations with Electrodermal activity

Main function of sweating of palm and sole is to prevent slippage while performing delicate tasks and grasping. This idea is supported by the observation of other mammals, such as monkeys, in which a high density of sweat glands is found in the finger pulps similar to what happens in human beings, and also on the underside of the distal portion of the tail that is also used for gripping. It also improves tactile sense and enhances grip as an aspect of the fight or flight response in humans (Asahina, Poudel, & Hirano, 2015; Baker, 2019).

Considered a preliminary escape response to face and to escape from danger situations, sweating of the palm and soles can be elicited by emotions, such as anxiety and fear. But rather than being directly related to emotion solely, it can also be elicited by several factors such as local tactile stimulation, deep inspiration, voluntary physical exercises and mental activities (Asahina et al., 2015).

Secondly, it is also a useful index to evaluate sympathetic and limbic system functions related to cognition, attention, and emotional processing. When an emotionally arousing stimulus is experienced, eccrine sweat glands produce sweat, which is an efficient conductor of current and, as a result, the electrical properties of the skin change. The more emotionally arousing

the stimulus is, the more sweat is secreted and the more the electrical properties of the skin change (Boucsein, 2012; Caruelle, Gustafsson, Shams, & Lervik-Olsen, 2019).

Therefore, sweating on the palm and the sole has become a clue for clarifying both complex human psychological and autonomic function. In addition, with help of EDA measurements it helped to overcome three intrinsic limitations related to self-reports of emotions: the difficulty of obtaining a continuous measurement; the subject's inability or unwillingness of reporting their emotions accurately, and; the impossibility of capturing unconscious emotions (Asahina et al., 2015; Caruelle et al., 2019).

2.4.2.3 Central and peripherial mechanisms involved in the electrodermal activity

Pathways controlling sweating and EDA have not been fully delineated in detail yet. Afferent pathways include sensory nerve fibers, central nuclei, the amygdale and the limbic system in accordance with the eliciting stimulus. Generally, the hypothalamus is regarded as the control center for all vegetative functions, including vasomotor activity and sweat secretion, and electrical stimulation in its paraventricular and posterior nuclei, always produce sympathetic reactions and sweat secretion (Asahina et al., 2015; Boucsein, 2012).

The CNS elicitation of EDA points to the existence of two different origins above reticular level: a limbic–hypothalamic source for thermoregulatory and emotionally influenced ipsilateral EDA, and; a premotor-basal ganglia source for contralateral EDA regarding the preparation of specific motor actions. The premotor ganglia source includes the basal ganglia, the thalamus, and the Brodmann area 6 of the frontal lobe taking part in eliciting sweat gland activity. The pathways stemming from these structures cross in the medulla oblongata to the contralateral side, while the fibers originating in the limbic-hypothalamic source descend via the tegmentum and the ventrolateral reticular formation to the ipsilateral sympathetic anterolateral pathway. In addition, general arousal efferent responses may also start directly at the brainstem reticular formation. This system may be also responsible for inhibitory influences on EDA, which may be either ipsi- or contralateral. Figure 2 depicts main central areas involved in this process (Asahina et al., 2015; Boucsein, 2012).

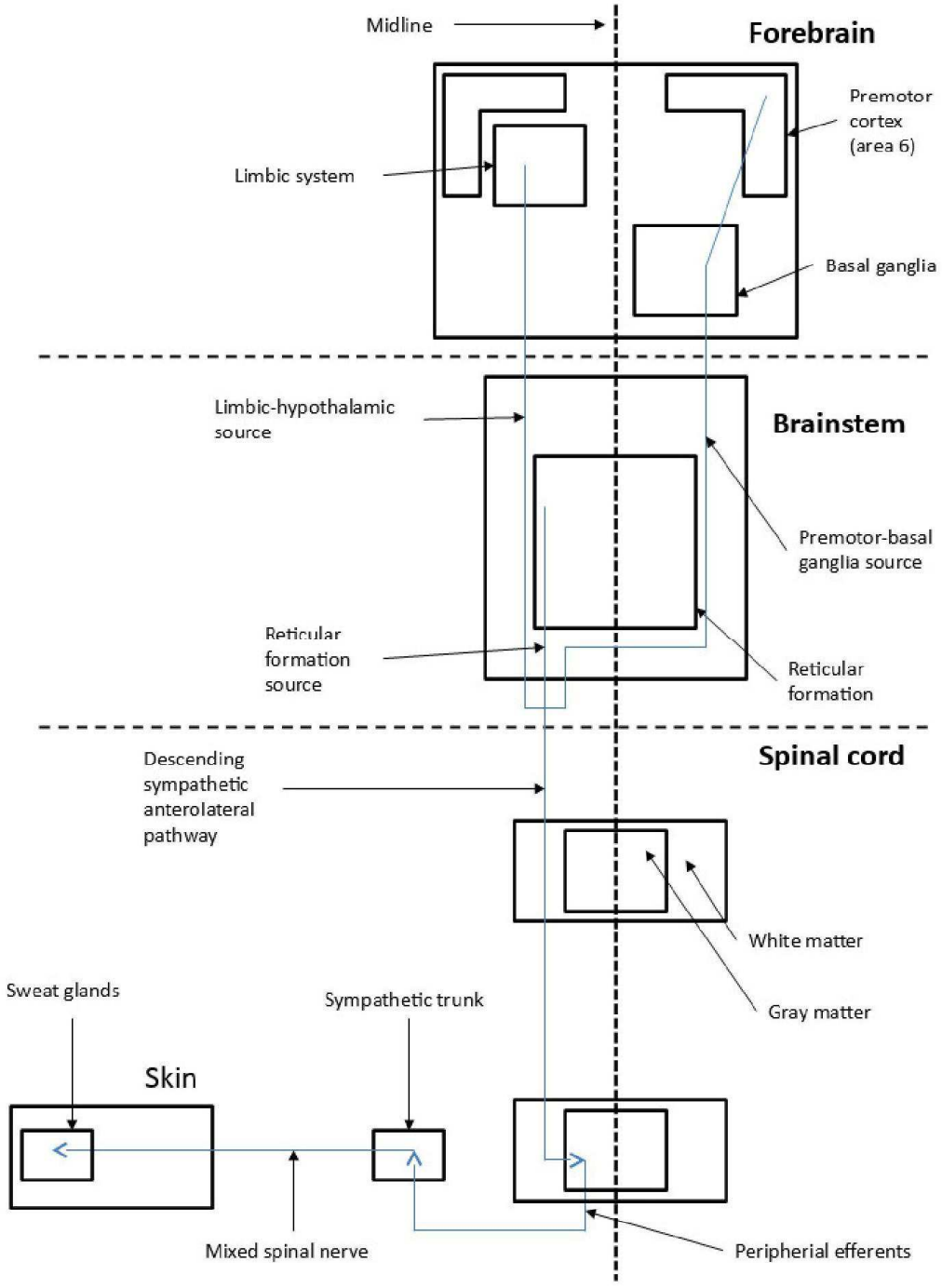

Figure 2. Three main pathways for the EDA elicitation by the central nervous system, including an ipsilateral influence from the limbic system via hypothalamic thermoregulatory areas, a contralateral pathway via premotor cortical and basal ganglia areas and a modulatory pathway originating in the reticular formation.

In a recent review, Asahina et al. (2015) reported that patients with bilateral amygdale lesions showed no sweat response on the palm during either deep respiration, exercise or local tactile stimulation, suggesting that the limbic system may be indispensable for the generation of sweating on the palm and sole in response to both emotional and non-emotional stimuli. In addition, the frontal cortex has strong connections with the amygdala and the entire limbic system, which may underlie the effects of emotional state on cognition. In fact, electrical stimulation of the prefrontal area increases sweat gland activity on the palm, although the reaction is far weaker than when the amygdale or cingulate cortex is stimulated. The latter also participates in the control of emotion-related physiological behavior, cognition, and attentional processes, and possibly also in habituation if any. In the medulla oblongata the raphe nuclei of the rostral medulla has been linked to thermoregulatory function as well as pain responses, while rostral ventral medial medulla plays a more important role in emotional sweating (Asahina et al., 2015).

Signals originating in those areas descend down the sympathetic nervous system pathway through the medial part of the lateral column until the intermediolateral nucleus of the spinal cord leaving it through the ventral root together with motoric efferents, projecting to postganglionic sympathetic neurons in the mixed spinal nerve to innervate the secretory part of the eccrine glands in a wide spread net of sympathetic fibers. Though postganglionic sympathetic transmission is normally adrenergic, these postganglionic sudomotor transmission by nonmyelinated class C fibers is cholinergic, using acetylcholine as a synaptic transmitter (Asahina et al., 2015; Boucsein, 2012).

2.4.3 The EDA signal

Main differences with regards to the EDA signal trace, is due to the usage of endosomatic or exosomatic measurements. Following a given stimulus, an EDR is expected to happen between 1 to 5 s after the stimulus onset. While exosomatic measures (either with DC or AC) will always present monophasic EDRs superposed over a slow varying monophasic signal (the EDL), the endosomatic measurements will present either mono-, bi- or triphasic EDRs. Following stimulation, the first negative deflection in SPRs is usually called the a-wave, and it might be followed by b- and c-waves. Although they are usually negative, positive

monophasic SPRs are also possible due to the interaction of underlying processes under the recording sites (Boucsein, 2012).

On the other hand, an exosomatic EDR will appear either as a steep decrease in the skin resistance or a steep increase in the skin conductance until it reaches its minima or its maximum after a short period and then tends to return to its previous values in a flatter recovery manner than its onset (Boucsein, 2012).

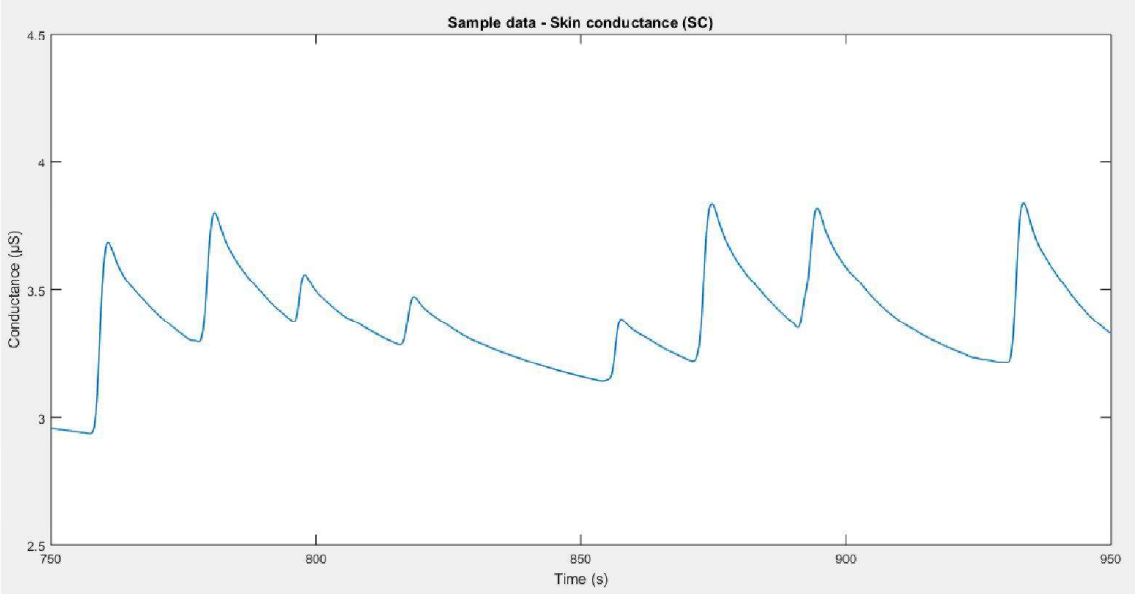

Figure 3. Example of an exosomatic recording of the skin conductance data.

The biggest advantages of using exosomatic measures instead of endosomatic measures are: the easiness to analyze the data since they are always unidirectional; they are less affected by electrode artifacts; an inactive reference electrode is not required at an abraded site; SCL is less sensitive to hydration status than SPL, and; the majority of studies already points out to many correlations between psychological processes and exosomatic measures (Boucsein, 2012).

When comparing constant current techniques and constant voltages techniques, the former have the following advantages: less amplification than constant voltage techniques is needed, and; the danger of electrode polarization is reduced due to limited current flow density through the electrodes. By contrast, constant voltage methods have the following advantages: there is no danger of sweat glands damage due to current concentration over a few secretory

ducts; the currents on both electrodes are independent and there is no need of matching current densities to the electrodes' areas even if electrodes of different sizes are used, and; little influence of the EDL upon the EDR amp is noticed (Boucsein, 2012).

On the other hand, endosomatic method is regarded as being more physiological due to the absence of external currents, preventing electrode polarization in long-term recordings. In addition, no special EDA coupler is needed when using a general-purpose bio-signal amplification system with sufficiently sensitive high-ohmic amplifiers. And, in a practical application, counts of NS.EDRs may be more sensitive in SP measurements than in SC measurements (Boucsein, 2012).

2.4.4 Biophysics of Electrodermal Measurements

Recommended units to be used in EDA analysis and reports are: milivolts (mV) for SP; microsiemens (μS) for SC and SY, and; kiloohms (kΩ) for SR and SZ (Boucsein, 2012).

In any electrical system, there is a potential difference *U* measured in Volts (V) between any two bodies with electrical changes (*Q*) of different sizes, usually measured in Coulombs (C). If they are connected by a conductor, an electrical current (*I*), measured in Amperes (A), will flow until the charges are equally distributed between the two bodies and the *U* disappear. Theoretically, U and I are proportional, and their quotient is constant, defined as the electrical resistance (*R*), given in Ohms (Ω), so that one V equals one Ω times one A. This relationship is known as the Ohm's law.

$$R = \frac{U}{I} \tag{1}$$

The reciprocal of *R* is the conductance (*G*) given in Siemens (S), which states that given a constant voltage, conductance is dependent on the intensity of electrical current flowing on the system, as given below:

$$G = \frac{1}{R} = \frac{1}{\left(\frac{U}{I}\right)} = \frac{I}{U} \tag{2}$$

Since most biological process involves resistances in the order of kiloohms (kΩ), conductance is commonly measured in microssiemens (μS). If resistance and conductance are changing

over time, we have a relationship so that increases in resistance lead to decreases in conductance, as follows:

$$\Delta G = \frac{1}{R2} - \frac{1}{R1} = \frac{-\Delta R}{R1 * R2} \quad (3)$$

Similarly, when using alternating current we can use electrical Impedance (*Z*) given in Ω, to measure the opposition that a circuit presents to a current when a voltage is applied. Impedance extends the concept of resistance to AC circuits, and possesses both magnitude and phase, unlike resistance, which has only magnitude. When a circuit is driven with direct current (DC), there is no distinction between impedance and resistance, so that the latter can be thought of as impedance with zero phase angle. Similarly, Admittance (*Y),* given either in S or in ʊ, is a measure of how easily a circuit or device allows a current to flow when a voltage is applied, and can be compared to the conductance of a system driven with direct current. It is defined as the reciprocal of impedance:

$$Y(f) = \frac{1}{Z(f)}. \quad (4)$$

An ohmic resistor to which direct current is applied will implement electrical resistance to a circuit. This means that it uses up electrical energy to transform it into heat. In electronic circuits they may be used to reduce current flow, to adjust signal levels, to divide voltages among other uses. They can provide either fixed or variable resistance depending on their characteristics and the necessity to model a specific system. In contrast, capacitors are used to store energy in a system. When voltage is applied to it, it become charged and builds up an electrical field. Once it is fully charged, no more charging current will flow. If the voltage source is removed, the full voltage remains until they are short circuited through a load. During the discharge process, the current will flow in the opposite direction to the charging current, until the voltage between its components reaches zero. The capacitance (*C*) of a capacitor indicates its ability to store an electrical charge (Boucsein, 2012).

2.4.4.1 Electrophysical properties of skin and sweat glands

When an external current is applied to a biological tissue like the skin, the skin acts like electrical networks built of resistors and capacitors. This means that the skin can be modeled as a RC circuit in which capacitors (C) are charged and discharged through one or more resistors (R). Some structures of the skin and its appendages are likely to act similar to resistive elements while other structures will rather act as capacitive elements (Boucsein, 2012).

2.4.4.1.1 Active properties of skin and sweat glands

Cellular membranes present selective permeability to certain ions and, thus, form an obstacle for the free movement of ions involved in the current flow. This storage of ions at the boundaries is followed by the buildup of a potential difference across the cell membranes. These membranes having polarization capacities, and hence capacitor-like or potential-like properties, are presumed to be located at the sweat gland membranes, at the dermal-epidermal boundary membrane and in the epidermis. All those properties together are supposed to form the active sources for electrodermal phenomena that is measured in EDA endosomatic measures, or skin potentials (SP) (Boucsein, 2012).

2.4.4.1.2 Resistive properties of skin and sweat glands

The abundant supplies of blood and interstitial fluids in the dermis and hypodermis layers possess good electrical conductivity. Having different conductivities dependent on their ionic concentration, they act as variable resistors. In addition, the lower corneal zone is relatively impermeable to water and solutions, and thus, it is thought to be mainly responsible for the skin's resistance (Fowles, 1974). Additionally, the epidermal Malpighian layer and the stratum intermedium are relatively conductive structures. Therefore, the whole stratum corneum is regarded as being a variable resistor, depending on its degree of hydration (Boucsein, 2012).

Under normal conditions, the corneum is always partially hydrated, depending on the environment's temperature and relative humidity and on the person's hydration status. With an increase in sweating, corneal hydration also increases, which causes tonic skin resistance to decrease. On the other hand, if the corneum becomes dry as a result of aging, or by

spontaneous reabsorption of water into the underlying dermis, tonic skin resistance increases. In addition, it is more likely that the conductivity of the stratum corneum depends on its electrolyte content than on its humidity (Boucsein, 2012).

Additionally, sweat gland ducts act as electrical shunts through the stratum corneum. It is supposed that skin conductance increases with the height of the ductal sweat column. Models of skin that focus on its resistive properties regard each sweat gland as single resistors with fixed values that can be switched on or off. Slow declines in SCL in the absence of EDRs may reflect a gradual dissipation of sweat in the ducts due to reabsorption mechanisms. These models are adequate to explain a fast rise and fall of ductal sweat, leaving the gradual changes of conductance owing to the corneal moistening (Boucsein, 2012).

In this sense, skin and sweat gland resistive properties can be depicted as several serial- and parallel-connected resistors.

2.4.4.1.3 Capacitive properties of the skin and sweat glands

The capacitive properties of skin and sweat glands haven't been as investigated as the resistive ones. Studying capacitive properties requires exosomatic measurements with AC, which is far less common than DC measurement (Boucsein, 2012).

When an external current is applied to the skin, the cell membranes exhibit their polarization capacities, storing electric potentials like technical capacitors. Whole cell assemblages such as epidermal layers may act selectively to some degree on the influx of ions of different sizes because of the cell structures extending into the intercellular spaces. Therefore, the whole epidermis will react to an external current like a network built from RC links connected in parallel and in series (Boucsein, 2012).

Other active membranes such as nerve, muscle, and glandular cells membranes are also present in the skin. These membranes have a resting charge which becomes reversed when stimulated. In addition, when an external current is applied they also show capacitive properties. Active membranes that act as capacitors with respect to EDA are located mainly in the secretory part of the sweat gland. Further, other capacitive properties may stem from

membrane polarization and depolarization in the blood capillaries, the pilo-erecting muscles, and the myoepithelia surrounding the sweat glands (Boucsein, 2012).

In summary, although investigating the AC properties of the skin and its appendages is much more complex than investigating the DC properties, AC investigations are indispensable for modeling the capacitive properties of the electrodermal system. By doing so with a wide spectrum of AC frequencies, it should be possible to quantitatively study the electrical properties of single components of the electrodermal system, and to describe the tonic and phasic components of EDA with regards to their origins and interactions. But, up to now, a simple resistive model is enough to explain most of the observed phenomena.

2.4.5 Electrodermal activity acquisition

Acquisition of EDA is possible with simple, portable, non-invasive and wearable sensors that measures skin electrical properties to track correlates of autonomic nervous system activity. The recording equipment can be either exosomatic or endosomatic, depending on whether an external voltage is applied to the skin or not, respectively. Exosomatic recording is done by applying a direct current (DC) or an alternating current (AC). Furthermore, some equipment allows recording phasic EDA separately from tonic EDA, with the help of an AC-coupled amplifier (Boucsein, 2012; Caruelle et al., 2019).

The exosomatic recording technique consists of passing current across the skin, between two electrodes that are part of the EDA recording equipment. Frequently used locations for positioning the electrodes include the fingers, the palms, and the wrists. Fingers, palms and soles should be preferred to other locations, because of their higher density of eccrine sweat glands (Boucsein, 2012). Shoulders were also shown to be good alternatives, whereas upper arms, backs, and armpits should be avoided. When electrodes are placed on the fingers, palms, or wrists, it is usually done in the non-dominant hand in order to limit movement artifacts during the recording. In addition, researchers can explicitly instruct participants to avoid movements of the equipped hand whenever it is possible (Caruelle et al., 2019).

Recording EDA during a resting period to serve as a baseline is not usually necessary since phasic EDA values do not depend on tonic EDA values, however rest intervals might be

useful whenever someone intends to compare SCL among several distinct conditions or stimuli. In addition, is always good to keep in mind that pathologies and demographics (age, gender, and ethnicity) can affect EDA. However, using demographic characteristics as recruitment criteria might considerably limit the external validity of the research findings (Boucsein, 2012; Caruelle et al., 2019).

2.4.6 Electrodermal signal decomposition techniques

It is already well accepted that EDA consists of a slow varying component which might be disrupted by fast inflections with slower recovering time than its outset. In order for a better understanding of the electrodermal phenomena and to overcome technical issues related to the analysis of EDL and EDRs, such as amplitude measures of overlaping EDRs and related SCLs, some techniques have been proposed.

Barry et al. (1993) first attempt to fix amplitude measurements was to correct the initial baseline values by subtracting each EDR from the extensions of previous EDRs. Then, Lim et al. (1997) developed first a four-parameter sigmoid-exponential SCR model that describes the entire response for a single pure EDR (gain, response onset time, rise time, and decay time constant), and then extended it to a eight-parameter skin conductance model to account for preceding EDRs tails and overlapped EDRs. By doing so, improvements in EDR amplitudes of up to 15% and in latency periods of up to 140 ms were achieved.

After that, Alexander et al. (2005) introduced the decomposition of EDA signal by means of deconvolution of the EDA signal into a sudomotor nerve activity signal with distinct bursts, with a biexponential IRF that accounts for the basic shape of an EDR. After identifying and isolating single peaks in the sudomotor drive, they are individually convoluted with the IRF to reconstruct individual EDRs, from which amplitude features could be computed more accurately.

Later, Bach et al. (2009) proposed the convolution of discrete time events with a general canonical IRF constant within each individual and extracted by means of principal component analysis (PCA) over all available EDRs for a given time-series. Then, they were also able to deconvolve the EDA signal into distinct events by using this canonical IRF. Main advantages

pointed out by the authors are the possibility to account artifacts and signal quality in their models.

Based on the ideas of Lim et al. (1997) and Alexander et al. (2005), a new idea to overcome problems regarding invariant IRF was proposed by Benedeck & Kaernbach (2010b). Because inter-individual and intra-individual EDR's shapes present significant variability, and because higher time-constants would lead to negative driver signals, which could not be interpreted in terms of sudomotor nerve activity, they proposed a nonnegative deconvolution method to deal also with varying EDR shapes, claiming the no negativity of the driver and maximal compactness of its impulses. Using optimization approaches, the nonnegative deconvolution, results in a tonic component, a nonnegative phasic driver with a zero baseline and predominantly distinct peaks, and a remainder which captures all deviations from the standard EDRs shape and signal artifacts. When the phasic signal is convolved once again with the IRF, the reminder and the tonic component, the full EDA signal is recovered.

The same year, Benedeck & Kaernbach (2010a) also proposed a continuous measure of the EDA. In order to obtain a continuous measure of the EDA by means of response window integration, they abandoned the concept of single and discrete responses. Conventional deconvolution as proposed by Alexander et al. (2005) is performed with the same parameters used by them, then tonic and phasic activities are estimated, and a value of goodness is computed based on the reminder signal. Following, an optimization is performed changing the two time parameters of the IRF in order to obtain the lowest value of goodness in a series of trials. Differently than what happens in nonnegative deconvolution, the phasic driver can take on negative values, which are related to sub-optimal IRFs or artifacts in the original EDA signal, and instead of refusing it, it is meant to minimize it.

Differences in the decomposition effects using both conventional and non-negative deconvolution on skin conductance levels are shown in figure 4.

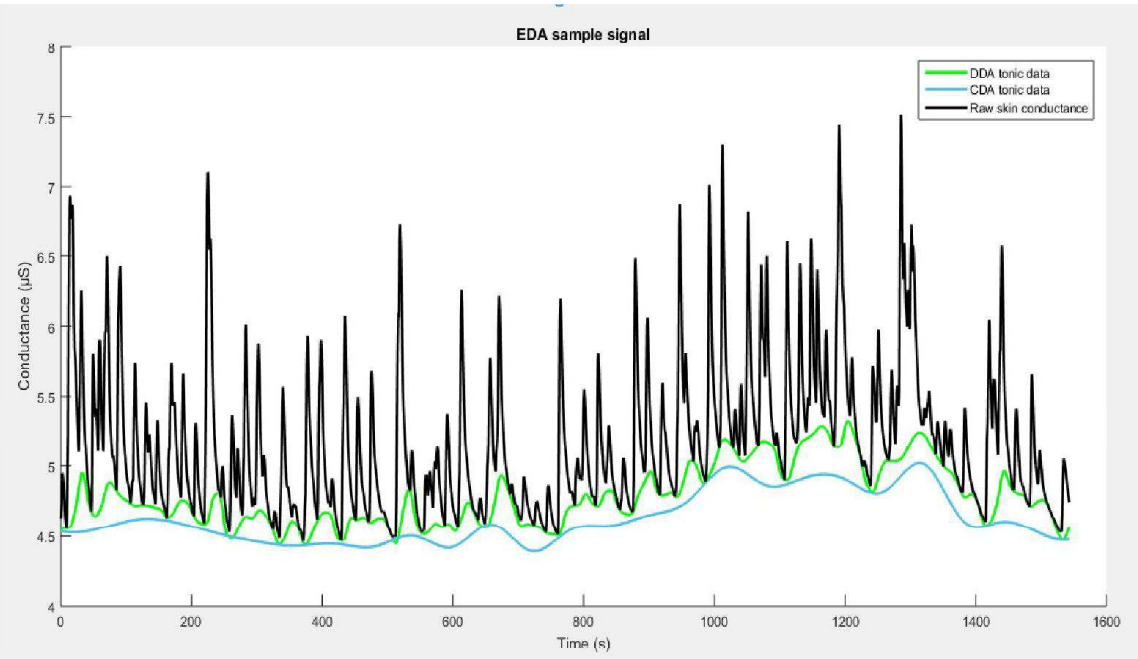

Figure 4. Skin conductance responses (black) and estimation of skin conductance levels by means of conventional deconvolution using continuous decomposition analysis, CDA (blue), and non-negative deconvolution using discrete decomposition analysis, DDA (green).

Grecco et al. (2016) also reported a novel algorithm for the analysis of EDA. In their model, EDA is also considered as a sum of three terms: the phasic component, which is the convolution of an Infinite Impulse Response (IIR) function and the sparse non-negative phasic driver; the tonic component, and; a Gaussian noise term. The latter component is conceived as a model prediction and data acquisition error estimation. The model is grounded on Bayesian statistics and convex optimization approach, and they claimed that the use of an IIR exhibited better accuracy and greater computational efficiency when compared to an IRF.

So far, these are the main studies with regards to performing signal decomposition techniques prior to EDA feature extraction.

2.4.7 Electrodermal activity processing

Preprocessing of EDA recordings can be performed in order to facilitate analyzing the EDA data. This step includes downsampling the data to speed up the analysis process and smoothing the data or applying a low-pass filter in order to reduce high-frequency noise produced by the device, and to reduce small movement artifacts. Artifact removal should also

be performed manually or with the help of available software with a customized algorithm to do it, after carrying a cautious visual inspection. Whenever an artifact is identified, it should be removed and replaced by a reconstructed data using interpolation techniques. If it is not possible to delete an artifact, that time frame containing it should not be considered for further analysis (Caruelle et al., 2019).

Researchers must also determine which time window is of interest. Normally if someone is looking for elicited responses, analysis windows should include the period between 1 and 4 seconds after stimulation is applied, since elicited responses usually have an onset latency within this period. In addition, a minimum amplitude criterion must be defined in order to take only significant signal changes into account. Generally signal fluctuations ranging from 0.01 to 0.05 μS are set to distinguish significant response from other small artifacts. Thresholds as low as 0.01 can be used in controlled trials which take places in experimental settings, with low chances of movement and electrical artifacts, however, higher thresholds might be preferred if the EDA data are going to be recorded less controlled environment, such as an ambulatory setting or a field study (Boucsein et al., 2012; Caruelle et al., 2019).

Habituation of the SCR must also be considered, since the responses to a repetitive stimulation might become smaller and smaller as the individual is exposed to consecutive stimuli, with possible negative influences on the quantification of EDA measures. Habituation is particularly likely to arise in studies with repeated-measure designs. Therefore, researchers who employ a repeated-measure design must keep in mind that this process should be addressed properly in their works (Caruelle et al., 2019).

2.4.7.1 Selecting EDA metrics

Once we acquired the signal and stored it, several metrics of features can be computed, and then subject to statistical analysis. However, they raise concerns about some statistical tests. A first concern relates to inter-individual differences in EDL and in EDR amplitude. Absolute EDR amplitude itself offers little insight because a given EDR amplitude can correspond to a large response for one person and to a small response for another person. Therefore, it is recommended to transform the EDA metrics to make them more comparable across

individuals. Standardization and range-correction transformation are pointed in the psychophysiology literature as possible solutions (Boucsein et al., 2012; Caruelle et al., 2019).

A second concern for the statistical analysis of EDA data relates to the distribution of SCR amplitudes, which is often positively skewed (Benedek & Kaernbach, 2010a, 2010b; Boucsein, 2012). Typically in an EDA recording, most EDRs have low amplitude and few EDRs have high amplitude, which results in a positively-skewed distribution of the EDR amplitudes. Therefore, some transformations might be performed to obtain a non-skewed distribution if that is a requirement for the statistical analysis.

When an emotionally arousing stimulus is experienced, eccrine sweat glands produce sweat, which is an efficient conductor of current and, as a result, the electrical properties of the skin change. The more emotionally arousing the stimulus is, the more sweat is secreted and the more the electrical properties of the skin change. Measuring EDA consists of measuring either electrical conductance, resistance, impedance, or admittance (depending on the recording method) of the skin. The increase in skin conductance starts 1 to 4 s after stimulus exposure, and it persists for 1 to 3 s (Boucsein, 2012; Caruelle et al., 2019).

EDA measurement helped to overcome three limitations inherent to self-report of emotions: the difficulty of obtaining a continuous measurement; subjects' inability or unwillingness to accurately report their emotions, and; the impossibility of capturing unconscious emotions. (Caruelle et al., 2019).

For EDA analysis, the raw signal is usually seen as a three-component signal, as previous mentioned: EDL; EDRs, and; artifacts. The first component refers to slowly fluctuating tonic levels which are typically used to evaluate general trends in activity and levels of activation. It typically varies between 2-20 μS, within and between individuals due to environmental and personal factors. On the other hand EDRs refer to quick, high-frequency responses superimposed on the tonic level. They can be characterized by rise time, amplitude, latency, and half recovery time. In healthy adults, rise time is usually between 1 and 3 seconds, amplitude often varies, and half recovery time is typically between 2 and 10 seconds. Artifacts are usually similar in shape and phase to EDRs, making their identification challenging. Main causes include physical movements, environmental factors, and electrical noise (Kelsey et al., 2017).

2.4.7.2 Electrodermal features

2.4.7.2.1 Time domain features

Time domain features include ordinary statistical parameters of the windowed signal such as signal mean value, standard deviation, kurtosis, or skewness. In addition, other event-related or attributes of the short-term responses following stimulation might be computed. These include the presence or absence of a SCR, with regards to a predefined threshold in order to restrict the analysis only to non-negligible responses, EDR amplitudes, EDR peaks count, mean EDR rise time, or the sum of EDR areas. Traditional thresholds for EDR amplitude has been set between 0.01 and 0.05μS (Shukla, Barreda-Angeles, Oliver, Nandi, & Puig, 2019).

2.4.7.2.2 Frequency domain features

Although only a few researches had focused on the predictive power of frequency domain EDA features, the frequency domain analysis has shown superior capability for the gradient component's detection of individual SCR over traditional amplitude analysis (Shimomura et al., 2008). Fast Fourier Transform (FFT), Short-time Fourier Transform (STFT) and power spectral density (PSD) estimation using Welch's method were the most commonly used algorithms to obtain the frequency domain representations of the signal. Lately, previous research has also considered statistical aspects such as variance, range, signal magnitude area, skewness, kurtosis, harmonics summation and spectrum power of five frequency bands ranging from 0.045 to 0.37 Hz (Alberdi, Aztiria, & Basarab, 2016; Ghaderyan & Abbasi, 2016; Posada-Quintero et al., 2018; Shukla et al., 2019).

2.4.7.2.3 Time frequency domain features

Due to the non-stationary behavior of EDA signals, wavelets have been suggested as suitable techniques to model EDA activity. In these sense, Discrete Wavelet Transform (DWT) and Stationary Wavelet Transform (SWT) were successfully implemented for emotional state classification and for denoising EDA signals. Mel-Frequency Cepstrum features was also suggested once to be useful in dealing with superimposed EDRs due to its abilities to magnify small amplitude variations and due to the assumption that the EDA signal might be a

convolution of a neural driver signal and a skin response driven by those underlying neuronal bursts. It is already a well-established technique to analyze speech signals with fairly no applications in EDA analysis yet (Ghaderyan & Abbasi, 2016; Shukla et al., 2019).

3 METHODS

3.1 Database and participants

This work was developed using previously recorded data from the Biovid Pain Database project (ethics committee approval: 196/10-UBB/bal, Ulm University, Ulm, BW, Germany) (Walter et al., 2013).

This database consists of a multimodal dataset obtained by monitoring several biopotentials during the application of noxious thermal stimulation on the forearm skin at four different intensities. Although the database encompass several data including biopotentials (EDA, electrocardiogram, electromyogram and electroencephalogram), video recordings of facial expression and head movements and stimulation temperatures during noxious heat stimulation and emotion elicitation (with pictures and video clips) solely and combined, only electrodermal signals in combination with stimulation temperature data in the absence of any emotion elicitation were investigated. Data from 96 healthy participants equally distributed by gender and aged between 18 to 65 years old (40.9 ± 14.9 years, mean ± standard deviation) were available for analysis.

3.2 Noxious Stimulation

Thermal stimuli were applied to the backside of the right forearm while the participant sat in a chair with both arms resting on a table. The experiment was performed using a thermode (PAHTWAY, Medoc, Israel). For each participant, the subjective heat pain threshold (P1) and tolerance threshold (P4) stimulus intensities were obtained by continuously increasing the thermode's surface temperature. P1 is defined as the lowest stimulus intensity able to cause pain, while P4 is defined as the maximum temperature the participant can stand. The following instructions were originally read aloud to each participant prior to the commencement of the calibration procedure at the beginning of the experiment to establish P1 and P4 levels: (i) Instruction for heat pain threshold - "*Please immediately press the stop button when a burning, stinging, drilling, or drawing feeling occurs in addition to the feeling of heat"*, and: (ii) Instruction for heat tolerance threshold - *"Please immediately press the stop button when you can no longer tolerate the heat regarding the burning, stinging, drilling, or*

drawing". A maximum temperature of 50.5 °C was not exceeded in order not to harm skin integrity. Thereafter, two equally distributed intermediate temperatures (P2 and P3) were calculated in order to obtain four distinct stimulation intensities. Finally, each temperature (P1, P2, P3, and P4) was randomly applied 20 times and sustained for 4 s, with randomized pauses of 8 to 12 s between consecutive stimuli (baseline temperature of 32 °C) (Walter et al., 2013).

3.3 Data acquisition

EDR was recorded using a Nexus-32 amplifier (Humakarigar Pvt. Ltd., India), with a sampling frequency of 512 Hz, and event-related data (i.e., thermal stimulus) were simultaneously recorded using Biotrace+ software (Mind Media, Netherlands).

3.4 Data Analysis

ER.EDRs were analyzed in MATLAB R2015a (MathWorks Inc., USA) using the open-source software Ledalab v3.4.8 (available at http://www.ledalab.de), with 8-s analysis window (between 1 and 9 s; see Eq. 3). Data were filtered with 1st order Butter-worth low-pass filter with a 5 Hz cutoff frequency, down-sampled to 64 Hz, and analyzed using the three methods described below. A block diagram depicting the data analysis sequence is provided in figure 5.

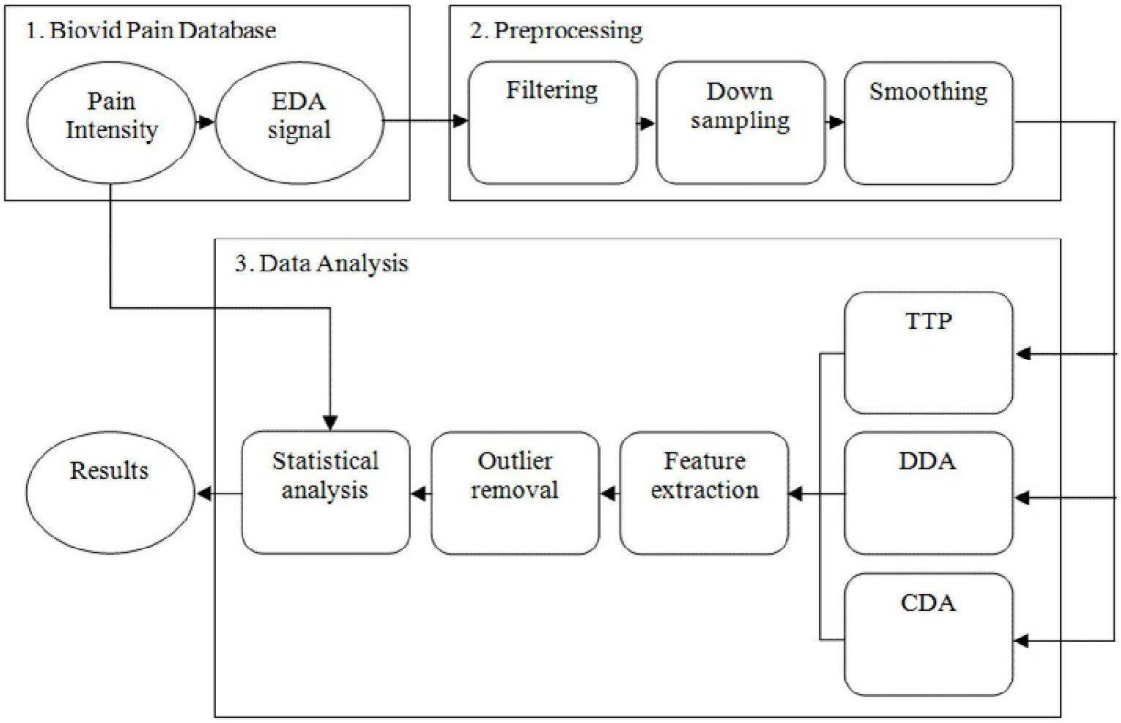

Figure 5. Block diagram depicting the sequence of steps employed for data analysis: CDA, continuous decomposition analysis; DDA, discrete decomposition analysis; EDA, electrodermal activity; TTP, trough-to-peak analysis. Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

3.4.1 Trough-to-peak analysis

TTP analysis is the traditional and simplest technique for identifying EDRs. It consists of running peak detection algorithms over the EDA signal to detect significant deflections on it. Thresholds of 0.01 μS or 0.05 μS are normally used to detect significant EDRs. Commonly estimated features for the characterization of EDRs include amplitude, rise time, half recovery time, and latency (Boucsein, 2012; Boucsein et al., 2012; Cowley & Torniainen, 2016).

3.4.2 Discrete decomposition analysis

Decomposition techniques developed by Benedek and Kaernbach (2010b) are based on the works of Alexander et al. (2005) and Lim et al. (1997), in which the electrodermal signal is composed of a slowly varying tonic activity and a phasic component, which is expressed as

the convolution of the underlying SNA and an impulse-response function (IRF). The main advantage of discrete decomposition analysis (DDA) is the possibility to detect deviations in the EDR shape with regard to a fixed response function (Benedek & Kaernbach, 2010b).

Therefore, the authors assume that the electrodermal signal consists of a nonnegative driver characterized by compact non-overlapping positive deflections over a zero-valued baseline that represents the SNA, an IRF, while the remainder that accounts for variation in EDR shapes is supposed to be related to the pore opening processes of the sweat glands (Benedek and Kaernbach 2010b). The IRF -$b(t)$- consists of a biexponential function with two time constants,$\tau_1$andτ_2, which account for a steep onset and a slow recovery:

$$b(t) = g \cdot \left(e^{\frac{-t}{\tau_1}} - e^{\frac{-t}{\tau_2}}\right). \quad (5)$$

A full description of the method can be found elsewhere (Benedek & Kaernbach, 2010b).

3.4.3 Continuous decomposition analysis

Continuous decomposition analysis (CDA) was developed by Benedeck and Kaernbach and is fully described in their work (Benedek & Kaernbach, 2010a). The EDA signal is estimated as follows:

$$EDA = SCL + EDRs = Driver * IRF = \left(Driver_{tonic} + Driver_{phasic}\right) * IRF, \quad (6)$$

in whichSCL(skin conductance level) is the tonic component of the EDA signal, and the IRF is given in (5).

3.4.4 Description of the set of features

The following steps summarize how features are calculated:

- Set the beginning of the analysis window 1 second after the beginning of stimulation;
- Set the end of the analysis window 9 seconds after the beginning of stimulation;
- Find all local minima followed by a local maxima within the analysis window, if any;

- Exclude pairs of local minima and local maxima that have differences less than 0.01 µS;
- Proceed with analysis for any pair of local minima and local maxima with differences greater than 0.01 µS.

Let$S$be the windowed signal as a function of the finite time set$T$, which are both ordered sets, described as follows:

$$T = \left\{t_n = n \cdot (1/64) \middle| n \in N \quad \wedge 1 \leq t_n \leq 9\right\} \tag{7}$$

$$S = \{s_n = s(t_n) | t_n \in T\}. \tag{8}$$

Given π and υ, $V$and$P$are ordered sets containing valleys and peaks of S, and TV and $TP$are ordered sets containing the timestamps in which valleys and peaks occur, so that:

$$\pi: N \to K \subset N; \upsilon: N \to J \subset N \tag{9}$$

$$V = \left\{v_j = s(t_n) \middle| s(t_n) \leq s(t_{n-1}) \wedge s(t_n) < s(t_{n+1}) \wedge n = \upsilon(j)\right\} \tag{10}$$

$$P = \{p_k = s(t_n) | s(t_n) > s(t_{n-1}) \wedge s(t_n) \geq s(t_{n+1}) \wedge n = \pi(k)\} \tag{11}$$

$$TV = \left\{t_n \in T \middle| n = \upsilon(j) \wedge v_j \in V\right\} \tag{12}$$

$$TP = \{t_n \in T | n = \pi(k) \wedge p_k \in P\}. \tag{13}$$

Whenever$V$and$P$are non-empty sets, significant responses can be retrieved by computing the relative amplitudes between each identified valley and the following peak whose differences are larger than a thresholdθ:

$$R = \{r_n = p_k - v_x | \exists p_k \in P, \exists v_x \in V: (p_k - v_x > \theta)\} \tag{14}$$

$$TR = \{t_n \in T | \exists p_k \in P, \exists v_x \in V: (p_k - v_x > \theta \wedge t_n = tv_x)\} \tag{15}$$

given:

$$x = max(\{j \in J | j < k\}). \quad (16)$$

After detecting significant peaks in the windowed signal, a number of features described in Table 1 are computed. Only EDRs with latencies between 1 and 9 seconds and relative amplitude larger than 0.01 μS were considered ER.EDRs. The occurrence rate (*OCR*) refers to the percentage of cases in which at least one ER.EDR is detected in a specific condition, so that if there were 20 stimuli at P1 level and significant ER.EDRs were found in only five of them, then OCR = 25%. This feature indicates the ability of the analysis to detect ER.EDR due to noxious stimulation.

The following features are automatically calculated by the Ledalab software. The number of ER.EDRs or number of skin conductance responses (*NSCR*), *Latency,* and amplitude sum (*AmpSum*) were computed for all three methods (CDA, DDA, and TTP). The sum of identified ER.EDRs (*AreaSum*) was only computed for DDA, and average phasic driver (*SCR*), area of phasic driver (*ISCR*), and maximum value of detected ER.EDRs (*PhasicMax*) were only computed for CDA.

Table 1. Description of the features extracted from the electrodermal activity.

Feature	Unit	Description	Equation
$NSCR_{ttp}$ $NSCR_{dda}$ $NSCR_{cda}$	(a.u.)	Number of ER.EDRs identified within the analysis window	$NSCR = \lvert R \rvert$ (17) in which R is the set of significant responses identified in the windowed signal using either TTP, DDA, or CDA.
$Latency_{ttp}$ $Latency_{dda}$ $Latency_{cda}$	(s)	Latency of the first ER.EDR identified within the analysis window	$Latency = \{x \in TR \lvert x \leq y, \forall y \in TR\}$ (18) in which TR is the set of the onset of significant responses within the analysis window using either TTP, DDA, or CDA.
$AmpSum_{ttp}$ $AmpSum_{dda}$ $AmpSum_{cda}$	(μS)	Sum of the ER.EDR amplitudes within the analysis window	$AmpSum = \sum_{l=1}^{n} R_l, n = \lvert R \rvert$ (19) in which R is the set of the relative amplitudes of the responses within the analysis window using either TTP, DDA, or CDA.
$AreaSum_{dda}$	(μS*s)	Sum of the identified ER.EDR areas using DDA	$AreaSum_{dda} = \sum_{l=1}^{m} \lim_{\delta x \to 0} \sum_{x=t_1}^{t_n} s_x \delta x, m = \lvert R \rvert$ (20) in which S is the convolved signal of the phasic driver and the IRF, and t_1 and t_n are the bounds of the analysis window.
SCR_{cda}	(μS)	Average phasic driver estimated using CDA	$SCR_{cda} = \frac{1}{n} \sum_{i=1}^{n} s_i$ (21) in which s_i is the i^{th} sample of the phasic driver signal.
$ISCR_{cda}$	(μS*s)	Estimated area of the phasic driver using CDA	$ISCR_{cda} = \lim_{\delta x \to 0} \sum_{x=t_1}^{t_n} s_x \delta x$ (22) in which s_x are elements of the phasic driver signal (S), and t_1 and t_n are the bounds of the analysis window.
$PhasicMax_{cda}$	(μS)	Maximum value of the detected ER.EDRs using CDA	$PhasicMax_{cda} = \{x \in S \lvert x \geq y, \forall y \in S\}$ (23) in which S is the phasic activity signal.

CDA, continuous decomposition analysis; DDA, discrete decomposition analysis; ER.EDRs, event-related electrodermal responses; TTP, trough-to-peak analysis. Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

3.5 Statistical Analysis

Statistical analyses were performed using R Software version 3.5.2 (R Core Computing Team, 2017). Since Lilliefors normality test rejected the hypothesis of normal distribution of all variables, data from NSCR, Latency, Ampsum, and OCR features were transformed using the aligned rank transformation in order to enable the study of interactions in 4 (intensity: P1 vs. P2 vs. P3 vs. P4) by 3 (method: CDA vs. DDA vs. TTP) between-subject design. One-way and two-way analyses of variance were used on the aligned ranks to assess the main effects and interactions, respectively, as suggested in the literature (Wobbrock, Findlater, Gergle, & Higgins, 2011). The other features were not transformed, and the Friedman test was used to assess the main effects of stimulus intensity. Bonferroni correction for post-hoc test was used to discriminate significant differences between methods, intensities, and their interactions. The significance level was set at 0.05 for all comparisons.

4 RESULTS

Data from one participant were excluded due to a corrupted EDA signal. The final dataset consisted of 95 complete EDA signals. These main results were already published elsewhere (Lima et al., 2019).

The *NSCR* feature presented significant main effects of both intensity, $F(3.23) = 314.94$, $p < 0.0001$, and method, $F(2.23) = 276.12$, $p < 0.0001$. However, these main effects were qualified by a significant interaction between the two factors, $F(6.23) = 4.26$, $p = 0.0003$. Bonferroni-adjusted comparisons indicated that for the same intensity, *NSCR* values obtained with CDA and DDA were larger than those using TTP for all intensities at a significance level lower than 0.01%. *NSCR* values obtained with CDA and DDA only differed for intensities P1 ($p = 0.0051$) and P2 ($p = 0.0067$). For the same method, *NSCR* values obtained with DDA at P3 were larger than those at P1 ($p = 0.0261$) and at P2 ($p = 0.0446$). However, no significant differences for *NSCR* values obtained with CDA or TTP methods were observed. The probability density estimates of *NSCR* for each method of analysis and stimulus intensity are shown in figure 6.

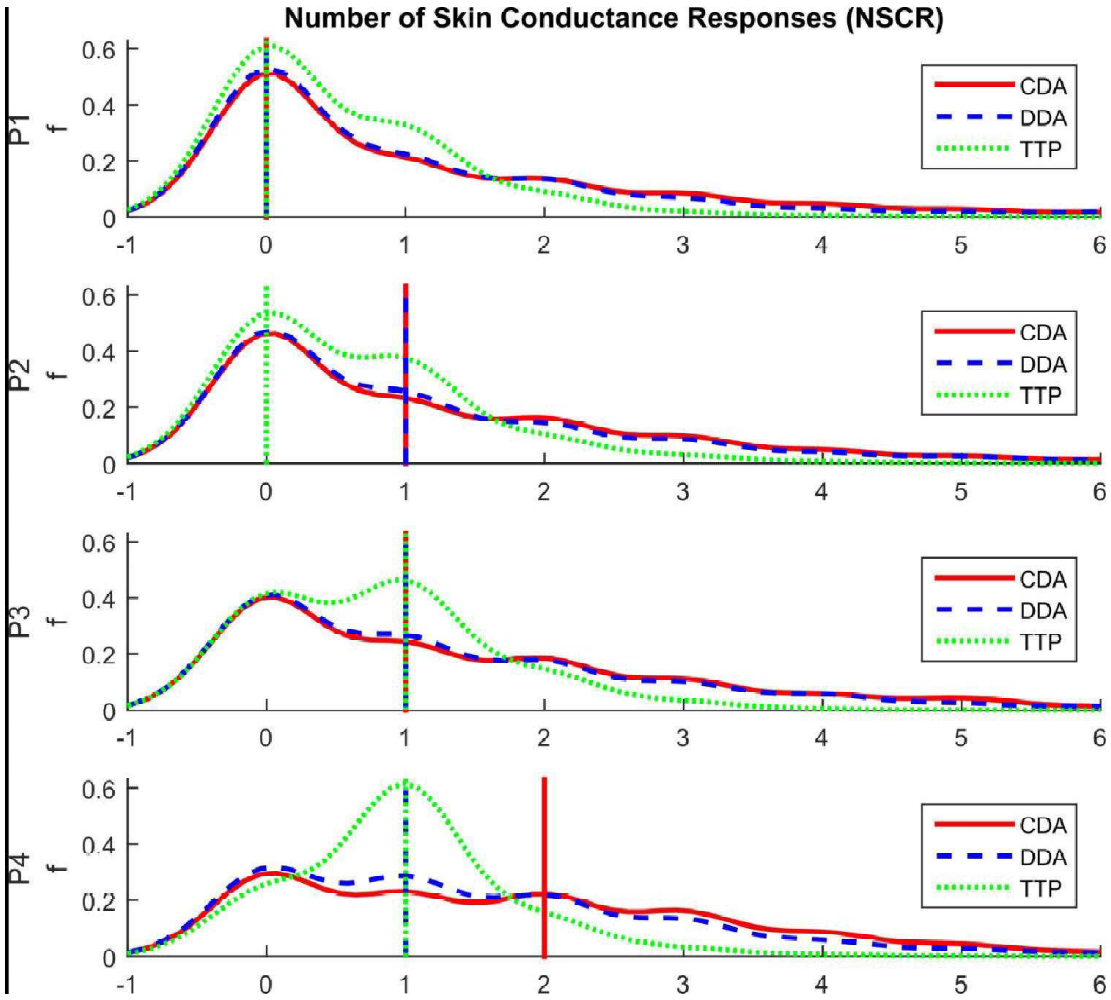

Figure 6. Probability density estimates of the number of skin conductance responses (NSCR) and their medians (vertical lines) after applying 80 random stimuli with four increasing intensities (P1, P2, P3, and P4, 20 stimuli each) detected by using continuous decomposition analysis (CDA), discrete decomposition analysis (DDA), and trough-to-peak analysis (TTP). Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

The *OCR* feature of significant responses presented significant main effects of the intensity factor, $F(3.11) = 41.31$, $p < 0.0001$, in which P3 and P4 values were larger than P1 ($p < 0.0001$ and $p < 0.0001$, respectively) and P2 ($p = 0.0110$ and $p < 0.0001$, respectively) values, but demonstrated no main effects of the method, $F(2.11) = 0.89$, $p = 0.4101$. Furthermore, no significant interactions were found between the two factors, $F(6.11) = 1.61$, $p = 0.1414$. The probability density estimates of *OCR* for each method of analysis and stimulus intensity are shown in figure 7.

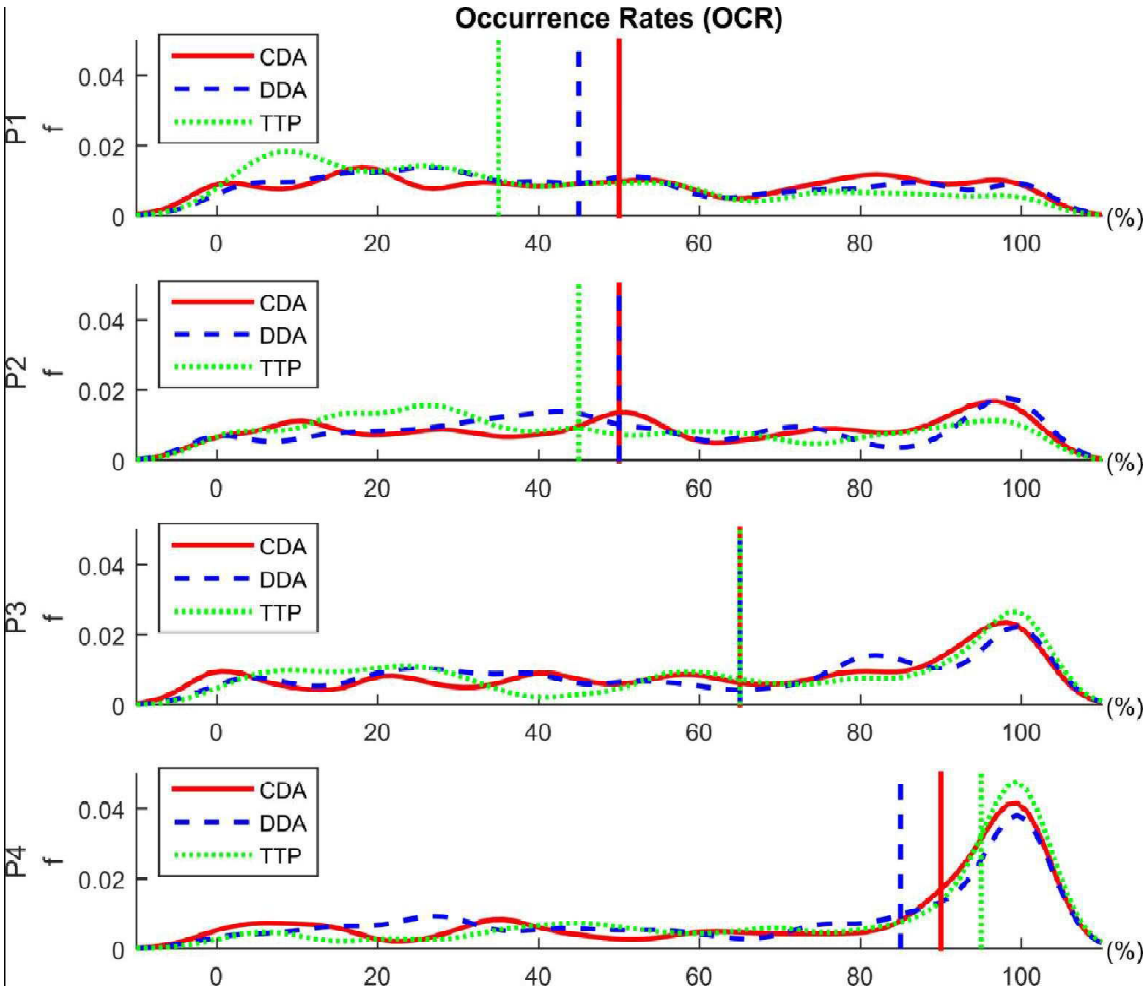

Figure 7. Probability density estimates of occurrence rates (OCR) of ER.EDRs and their medians (vertical lines) after applying 80 random stimuli with four increasing intensities (P1, P2, P3, and P4, 20 stimuli each) detected by using continuous decomposition analysis (CDA), discrete decomposition analysis (DDA), and trough-to-peak analysis (TTP). Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

The *AmpSum* feature presented significant main effects of both intensity, $F(3.23) = 516.52$, $p < 0.0001$, and method, $F(2.23) = 6.28$, $p = 0.0019$. However, these main effects were qualified by a significant interaction between the two factors, $F(6.23) = 8.07$, $p < 0.0001$. Bonferroni-adjusted comparisons indicated that for the same method, all intensities were significantly different for all three methods, with larger values for the higher intensities at a significance level equal to or less than 0.01%. For the same intensity, *AmpSum* values obtained with CDA and DDA were larger than those using TTP for intensities P1, P2, and P3 at a significance level equal to or lower than 0.01%, but their values were lower than those using TTP for P4

intensity ($p < 0.0001$). Furthermore, no significant differences between *AmpSum* values obtained with CDA and DDA for any of the four intensities were observed; however, in P1 ($p = 0.0216$), the values obtained with CDA were lower than those using DDA. The probability density estimates of *AmpSum* for each method of analysis and stimulus intensity are shown in figure 8.

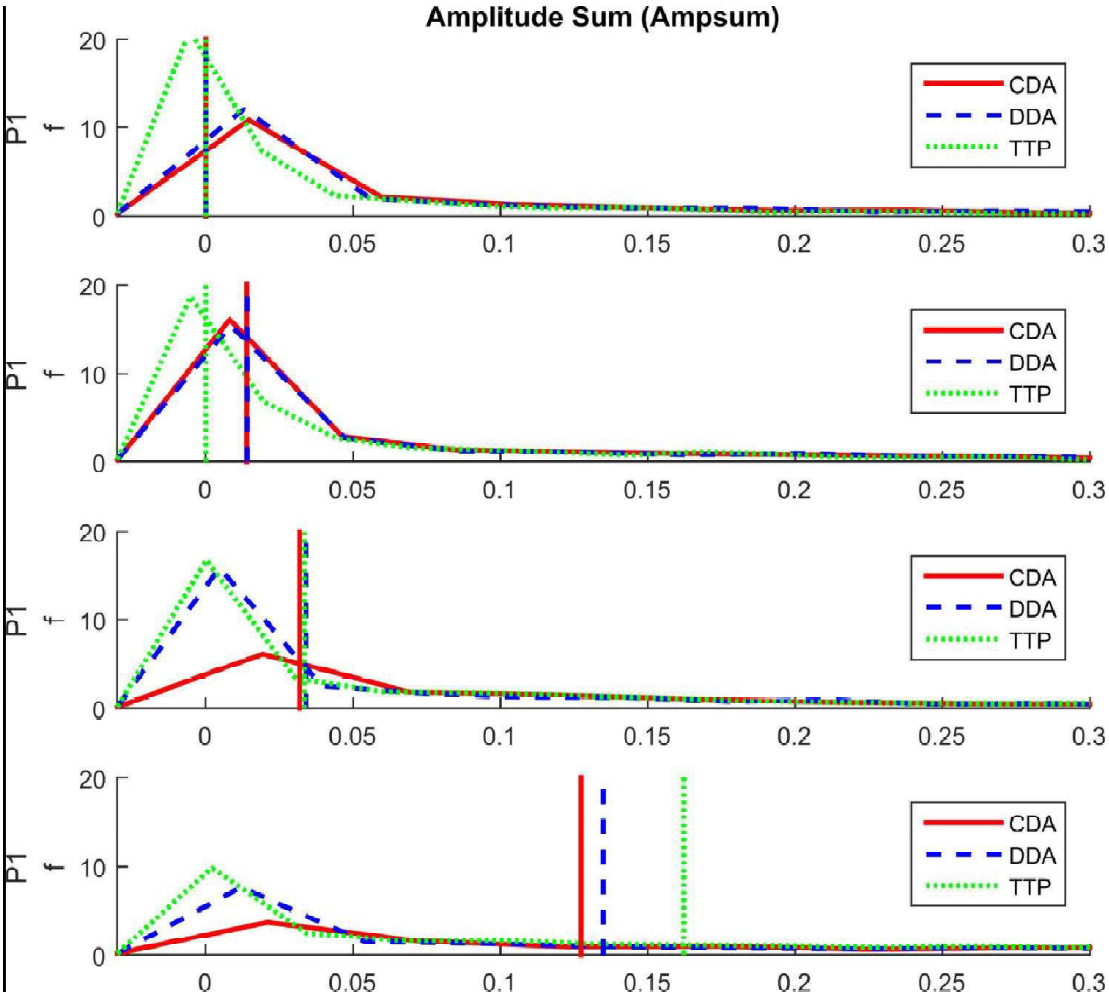

Figure 8. Probability density estimates of amplitude sum (AmpSum) of the ER.EDRs and their medians (vertical lines) after applying 80 random stimuli with four increasing intensities (P1, P2, P3, and P4, 20 stimuli each) detected by using continuous decomposition analysis (CDA), discrete decomposition analysis (DDA), and trough-to-peak analysis (TTP). Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

The *Latency* feature presented significant main effects of both intensity, $F(3.13) = 69.21$, $p < 0.0001$, and method, $F(2.13) = 90.91$, $p < 0.0001$. However, these main effects were qualified

by a significant interaction between the two factors, $F(6.13) = 5.31$, $p < 0.0001$. Bonferroni-adjusted comparisons indicated that at P4 intensity, *Latency* values obtained with CDA and DDA were lower than those using TTP ($p = 0.0409$ and $p = 0.0068$). For the same method, differences between intensities were only significant for Latency values obtained with TTP, in which P1 and P2 values were lower than P4 values ($p = 0.0078$ and $p = 0.0074$). The probability density estimates of *Latency* for each method of analysis and stimulus intensity are shown in figure 9.

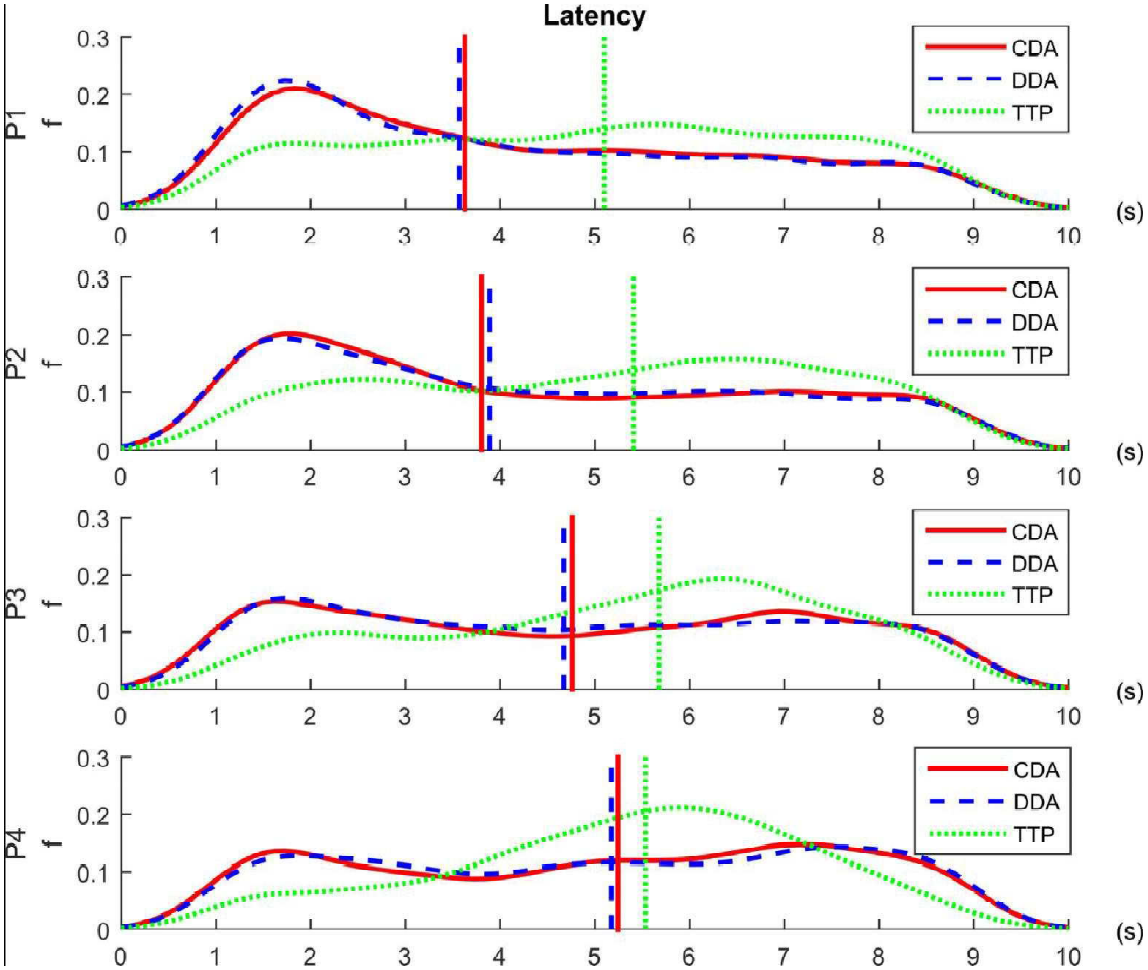

Figure 9. Probability density estimates of the latency of the ER.EDRs and their medians (vertical lines) after applying 80 random stimuli with four increasing intensities (P1, P2, P3, and P4, 20 stimuli each) detected by using continuous decomposition analysis (CDA), discrete decomposition analysis (DDA), and trough-to-peak analysis (TTP). Reprinted from Lima et al. (2019). Copyright (2019) by RBE-SBEB.

The other four variables that are specific to a single method, such as $AreaSum_{dda}$, $ISCR_{cda}$, SCR_{cda} and $PhasiMax_{cda}$, behaved similarly. For all of them, Friedman's test identified significant differences in the factor intensity at a significance level of less than 0.01%. Bonferroni-adjusted comparisons indicated that for the $AreaSum_{dda}$ feature, P4 values were larger than P1 ($p < 0.0001$), P2 ($p = 0.0001$), and P3 ($p = 0.0403$) values. For the $ISCR_{cda}$ feature, P4 values were also larger than P1 ($p < 0.0001$), P2 ($p < 0.0001$), and P3 ($p = 0.0085$) values. For the SCR_{cda} feature, P4 values were also larger than P1 ($p < 0.0001$), P2 ($p < 0.0001$), and P3 ($p = 0.0085$) values. Finally, for the $PhasiMax_{cda}$ feature, P4 values were larger than P1 ($p < 0.0001$), P2 ($p < 0.0001$), and P3 ($p = 0.0244$) values. The only difference here involved the P3 values, which were also larger than the P1 ($p = 0.0416$) values.

5 DISCUSSION

In this research, TTP and decomposition analyses were applied to signals resulting from skin conductance. The aim was to detect and discriminate noxious thermal stimuli at several intensities.

Although CDA and DDA presented significant improvements in the count of event-related EDRs (*NSCR*) when compared with TTP for all stimulus intensities, this fact did not present significant improvements in the *OCR* of ER.EDRs regardless of the intensity. This finding means that more EDRs were identified, but solely where at least one single response had already been identified using TTP analysis.

The second point was to assess whether CDA and DDA could provide better discrimination among noxious stimulation intensities than TTP. Our results showed an improvement in the number of identified responses, as well as in the amplitude features. However, these results did not support improvements in discrimination power among the four intensities when compared with TTP.

In general, some reviews had pointed out recently that although skin conductance has been used to assess pain in several different conditions such as intensive care unit patients, post-operative patients, patients undergoing anesthesia (Storm, Günther, Sackey, Bernhardsson, & Bjärtå, 2019) especially by means of skin conductance amplitude and by means of the number of skin conductance responses per second. In infants Hu et al. (2019) said that although skin conductance has been positively correlated with uni-dimensional pain assessment scales and crying time but not with multidimensional measurements, inconsistent findings on its validity still exist. Azevedo Santos & DeSantana (2018) also reported that, although behavioral scales are still considered the gold standard in assessing pain in mechanical ventilated patients in intensive care units, pupillometry, the bispectral index (BIS) and skin conductance have been used in this context to provide more objective measurements. However, due to the variety of factors influencing brain activity and sympatetic neural activity, more evidence is needed to their use in clinical practice.

Considering the *NSCR* feature, which the literature usually considers a feature relevant to pain evaluation, it tends to increase with increased pain or increased stimulation intensity. Clinical

studies have found from a moderate correlation with numeric rating scales in postoperative inpatients (Rago et al., 2015) to no correlation with behavioral scales in neonates during the heel prick procedure (de Jesus et al., 2015), while an experimental study using thermal stimulation found moderate discrimination power among different stimulation intensities in healthy participants (Treister et al., 2012). Similarly, a recent study showed that an increased *NSCR* is more closely related to the electric noxious stimulation than to positive or negative emotions elicited by pictures (Günther et al., 2016). Svalebjørg et al. (2018) also reported that the *NSCR* per second is sensitive to indicate moderate or severe procedural pain in perioperative conditions, but with low specificity, being unable to discriminate pain and other stressors before major surgeries. And Khanna et al. (2018) reported significant negative correlation between sedation status and the number of skin conductance fluctuations per second in postoperative patients and in patients with sepsis due to pulmonary diseases in intensive care units during procedural pain caused by patient positioning and tracheal suctioning. Roué et al. (2018) reported moderate correlations between *NSCR* per second and the Neonatal Facial Coding System after venopuncture. On the other hand, Storm et al. (2019) reported moderate correlations between *NSCR* and the strength of the electrical stimulation but no correlation to VAS scores, when the stimulation was modulated by means of pictorial emotional stimuli. Our findings corroborate most of these results, and, although values obtained with CDA and DDA methods were significantly larger than those obtained with TTP method for all stimulus intensities, if one intends to discriminate intensities of noxious stimuli by means of *NSCR* values, DDA method performed slightly better than CDA and TTP methods.

Most previous studies also corroborate the idea that amplitude values also tend to increase with increased thermal stimulation intensities but without good decimation power related to stimulus intensity or severity of pain (de Jesus et al., 2015; Loggia et al., 2011; Treister et al., 2012). In contrast, the study of Walter et al. (Walter et al., 2014) found a set of new EDR features related to amplitude that could provide discriminative power among four heat noxious stimulation intensities. Bari et al. (2018) found good discriminative power among three different electrical stimulation intensities in healthy subjects in a significant linear fashion, and that higher stimulus' intensities were also related with higher NRS scores. In the present work, only the *AmpSum* feature could accurately discriminate the four noxious stimulation intensities, regardless of the method used to compute it.

Other measures that are somehow related to the signal's amplitude ($AreaSum_{dda}$, $ISCR_{cda}$, SCR_{cda} and $PhasiMax_{cda}$) and were calculated only for the decomposition methods also increased with increased noxious stimulation intensities. Although they performed better than some other common features evaluated in this paper, they were only able to discriminate the higher stimulation intensities from the lower ones, but none of them could discriminate all four intensities of noxious stimulation.

Finally, CDA and DDA presented smaller *Latency* values than TTP, which may account for the identification and discrimination of hidden or superposed responses at the beginning of the stimulation phase. Although a subtle difference has been observed among the methods, only TTP could discriminate the stimulus intensities. In addition, no recent studies were found in which a relationship between latency and stimulus intensity has been investigated.

Using data from the same BioVid databese, Lopez-Martinez & Picard (2018) used DDA deconvoluted skin conductance and electrocardiogram data to design an automated algorithm to continuously estimate pain intensity with recurrent neural networks. They extracted six features from SC and seven features from HRV data, and they concluded that a SC-based algorithm outperformed even compound algorithms using SC and HRV data. Gruss et al. (2015) used electromyogram, electrocardiogram and skin conductance data to extract 159 different features and a support vector machine-based algorithm to discriminate the four different stimulation intensities. They reached from 79% to 90% of accuracy in discriminating non-painful stimulation from pain threshold and from non-painful stimulation to pain tolerance levels, respectively. However, no information on the ability of skin conductance solely to discriminate different stimulation intensities can be inferred. On the other hand, Susam et al. (2018) used skin conductance data solely and support vector machine-based algorithms to discriminate between pain and non-pain states in children. Their algorithm reached up to 77.66% of accuracy, with a sensitivity of 81.33%, and specificity of 74%, considering a threshold pain score level of 4 in 10, which is usually reported as moderate pain.

Considering these recent papers, it may be noted that amplitude values and *NSCR* can provide some reliable information on nociceptive perception. Using sustained thermal stimulation for a few seconds (approximately 6 s) and a comprehensive response window, our results partially support this statement. However, one drawback of using ER.EDRs to study how the human body reacts to the noxious heat stimulation is that it still relies on the lack of

specificity and on the low predictive power for a clinical application to quantify how strongly patients respond to different modalities of noxious stimuli. Even when using new methods to estimate the SNA instead of using EDR data itself, no advances were achieved in terms of discrimination of noxious stimulation intensity. All three methods presented similar discrimination power for the common features among them. Some authors (Loggia et al., 2011; Walter et al., 2014) suggest that searching for new features may provide better insight into the study of sympathetic responses to noxious stimulation, responses which may be related to the subjective experience of pain. In our study, the four features that were only computed for the CDA or DDA methods presented some tendency but poor discrimination power among the four stimulus intensities. In contrast, only the common feature *AmpSum* could discriminate all four intensities of noxious stimulation, regardless of the method. Therefore, future works should keep looking for new significant features while using more robust signal processing methods, such as CDA and DDA, but should not be restricted to them.

Regarding decomposition methods, to the best of our knowledge, at least two ready-to-use tools could be further analyzed to check our results on decomposition methods. Those include the PsPM, which uses a canonical skin conductance response function and a general linear convolution model (Bach et al., 2010), and the CvxEDA, which uses Bayesian statistics, a convex optimization approach, and an IRF based on an infinite impulse response (IIR) (Greco et al., 2016). Both authors claimed advantages on the use of their software against the Ledalab software. Greco et al. (2016) reported a better discrimination among four emotion arousal levels using pictures in favor of the CvxEDA when compared to CDA analysis. Similarly, Bach et al. (2014) used five contrast situations in a mix of positive, neural and negative arousing pictures to compare the performance of the previous SCRalyze version of their software, which is now part of the PsPM software, and the Ledalab software. He reported better results while using SCRalyze in four out of five situations, and comparative values on the fifth one.

Although the developers of these other methods have reported significant improvements in EDR analysis when compared with Ledalab software, we opted to use the Ledalab software, which combines the three different analytic methods, for our first trial. Therefore, it can be emphasized that different methodologies should be further evaluated, and generalization to other decomposition methods should be carefully done or altogether avoided. Furthermore,

with regard to the differences among the three methods, using the specified parameters, the CDA and TTP methods were faster than DDA.

6 CONCLUSION

After discussing the most updated literature and our own findings, it can be concluded that all the investigated methods, namely TTP, CDA and DDA, can be equally suitable to identify electrodermal skin responses to high-intensity noxious stimuli. Other methods and software such as the PsPM and the CvxEDA should be further analyzed before stating any conclusions.

CDA and DDA were more sensitive at identifying EDRs compared with the TTP method, indicating that they more efficiently identify slight activation of the sudomotor nerves that are not obvious or easily identified in the raw EDR signal. However, this information brought no further improvements to the identification of distinct stimulation intensities in our study. On the other hand, this might give them some advantages while using long-term monitoring recordings in clinical settings associated with counts of NS.EDRs.

Although EDR occurrence rates tended to increase with increased stimulus intensity regardless of the method of analysis, ranging from 40 to 78% of the time, no differences among them for the same intensities were observed.

The amplitude features showed to be the best choice to discriminate 4 stimulation intensities even when compared to the number of evoked responses, which is often used as a good predictor of the stimulation intensity or the pain intensity in clinical studies. They have also been cited by many authors as the best choices among EDA features lately.

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