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Electrodermal activity spectral and nonlinear analysis – potential biomarkers for sympathetic dysregulation in autism

Zuzana Visnovcova[1,2], Nikola Ferencova[1,2], Marian Grendar[1], Igor Ondrejka[3], Lucia Bona Olexova[1,2], Iveta Bujnakova[2,4] and Ingrid Tonhajzerova[1,2]

[1] *Biomedical Center Martin, Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava, Martin, Slovakia*
[2] *Department of Physiology, Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava, Martin, Slovakia*
[3] *Psychiatric Clinic, Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava, University Hospital Martin, Martin, Slovakia*
[4] *Society to Help People with Autism (SPOSA – Turiec), Martin, Slovakia*

Abstract. Autism spectrum disorder (ASD) is a neurodevelopmental disease characterized by emotional and social deficits, which can be associated with sympathetic dysregulation. Thus, we aimed to analyze the electrodermal activity (EDA) using time, and novel spectral and nonlinear indices in ASD. The cohort consisted of 45 ASD boys and 45 age-matched controls. EDA was continuously recorded at rest. The EDA indices were evaluated by time-, spectral-, and nonlinear-domain analysis. Our results revealed increased non-specific skin conductance responses, spectral parameters in high and very-high frequency bands, approximate and symbolic information entropy indicating sympathetic overactivity in ASD *vs*. controls ($p < 0.05$, for all). Surprisingly, the nonlinear index from detrended fluctuation analysis α1 was lower in ASD *vs.* controls ($p = 0.024$) providing thus distinct information about qualitative features of complex sympathetic regulation. Concluding, the complex time, spectral, and nonlinear EDA indices revealed discrete abnormalities in sympathetic cholinergic regulation as one of the potential pathomechanisms contributing to cardiovascular complications in ASD.

Key words: Electrodermal activity — Autistic spectrum disorder — Nonlinear analysis — Spectral-domain analysis — Time-domain analysis

Introduction

Electrodermal phenomena represent spontaneous changes in the electrical features of the skin, particularly in the sweat glands activity (Boucsein 2012; Gersak and Drnovsek 2020). More specifically, the external stimulation of sweat glands by weak electrical current spreading through two tactile electrodes into the skin surface causes the imbalance between positive and negative ions in the sweat resulting in measurable changes of electrodermal activity (EDA) (Boucsein 2012). In physics analogy, the sweat glands symbolise variable resistors, i.e. the amount of glands activated during perspiration represents the number of resistors involved in EDA (Hugdahl 1998). The potential differences – voltage (U) arises between two electrically charged particles. By connecting these two parts to the conductor, an electric current (I) flows through the conductor until the potentials are equalized. The quotient of voltage and current is termed resistance (R). The relations between these parameters are quantified by Ohm´s law, where resistance corresponds with electrical voltage between two electrodes localized on skin surface divided by constant electrical current passing through the skin: R = U/I (Dawson et al. 2007). The conductance, as an

Correspondence to: Ingrid Tonhajzerova, Department of Physiology, Biomedical Center Martin, Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava, Mala Hora 11161/4C, 036 01 Martin, Slovakia
E-mail: ingrid.tonhajzerova@uniba.sk; ingridtonhajzerova@gmail.com

inverse of resistance, is the result of current flow when using constant voltage (Hugdahl 1998; Dawson et al. 2007).

From the physiological perspective, the skin conductance directly depends on the amount of activated sweat glands regulated by the sympathetic nervous system (Dawson et al. 2007; Boucsein 2012). More specifically, EDA represents momentary changes in the electrical conductance of the skin reflecting the sudomotor activity of the eccrine sweat glands regulated by the sympathetic cholinergic activity (Andreassi 2000). Further, EDA is considered as an important index assessing the level of cognitive and emotional arousal (Critchley et al. 2000; Boucsein 2012) as well as sympathetic arousal to stress, i.e. everyday stress responses in natural environment (Vavrinsky et al. 2021). Thus, EDA measurement as a non-invasive and sensitive method is increasingly used in psychophysiological research (Posada-Quintero et al. 2016; Aldosky 2019; Posada-Quintero and Chon 2020).

Autism spectrum disorder (ASD) represents a serious neurodevelopmental disorder characterized by deficiencies in social communication and behaviours. The exact definition of ASD is not clear yet, but ASD is thought to be a manifestation of complex genetic defects additionally influenced by external factors (Betancur 2011; Ostatnikova et al. 2016). In this context, the autonomic nervous system (ANS) plays a crucial role in the control of the physiological, emotional/cognitive and behavioural state during social interaction. Recently, scientists' interest in detecting relationships between autonomic dysfunction and ASD has been growing, but the exact pathomechanisms leading to autonomic abnormalities in ASD are still unclear (Bal et al. 2010; Ming et al. 2011; Plus 2019; Bharath et al. 2020). Moreover, the existing findings on the ASD-linked sympathetic regulation reflected by EDA are controversial, such as increased, i.e. associated with more social impairments (O'Haire et al. 2015; Neuhaus et al. 2016; Fenning et al. 2017), unaltered (Kong et al. 2021), decreased (linked with augmented externalizing behaviour complications in ASD children) (Bujnakova et al. 2016; Baker et al. 2018), or slower habituation of sympathetic skin response to auditory stimuli indicating a persistent predominant state of sympathetic activity in autism (Bharath et al. 2020). Furthermore, Baker et al. (2015) found positive associations of EDA and interpersonal variability (i.e. relationship between parent and ASD child) during naturalistic free play. More specifically, the ASD children with better relationship with their parent are characterized by faster EDA declining (Baker et al. 2015).

In addition to the standardly used EDA parameters (e.g. skin conductance level, non-specific skin conductance responses), the EDA changes can be also evaluated by spectral-domain and nonlinear indices (deterministic chaos, recurrence plot, detrended fluctuation analysis) (Lanata et al. 2012). In this context, our previous study evaluated the alterations between time and nonlinear EDA indices (approximate and symbolic information entropies) during and after cognitive stressors in healthy group. We revealed that nonlinear indices of EDA represent more sensitive tool for extraction of EDA characteristics as features of sympathetic control (Visnovcova et al. 2016). However, quantification of EDA by using spectral and nonlinear parameters in mental disorders is rare. Thus, we aimed to evaluate resting EDA by different parameters of the spectral and nonlinear analysis in children suffering from ASD. We hypothesized that complex EDA analysis could provide important and independent information related to sympathetic cholinergic regulatory network in children suffering from ASD. Further, we aimed to use the Random Forest (RF) machine learning algorithm to detect relevant EDA predictors for differentiating autism spectrum disorder. To the best of our knowledge, this is the first study to use EDA complex and detailed analysis associated with machine-learning algorithm in children suffering from ASD.

Materials and Methods

Ethics statement

The study was approved by the Ethics Committee of Jessenius Faculty of Medicine in Martin, Comenius University in Bratislava in accordance with the Helsinki declaration (2000) of the World Medical Association. All patients and their parents/legal guardians were carefully informed about study protocol. The written consent was provided by the legal guardian.

Participants

A total of 55 boys with ASD were examined, of which 10 ASD boys were excluded due to large number of movement artifacts. The final studied cohort consisted of 45 boys with ASD diagnosis (average age: 11.64 ± 0.44 years) and 45 aged and gender-matched apparently healthy control (HC) (average age: 11.72 ± 0.45 years). The ASD participants were recruited from the regional Autism Centre and Psychiatric Clinic University Hospital Martin. Diagnosis of ASD without comorbidities (e.g. attention deficit/hyperactivity disorder) was made by a specialist in child and adolescent psychiatry and corroborated with Diagnostic and statistical manual of mental disorder (DSM-5, APA 2013). In addition, the intellectual functioning measured by Wechsler Intelligence Scale for Children (WISC III) was performed by a licensed clinical psychologist. Inclusion criteria for ASD group: diagnosis of ASD by child psychiatrist, IQ above 70, without mediation treatment, ability to withstand the study protocol. Exclusion criteria for ASD and control groups were following: presence of a genetic or neurological disorders, acute or chronic res-

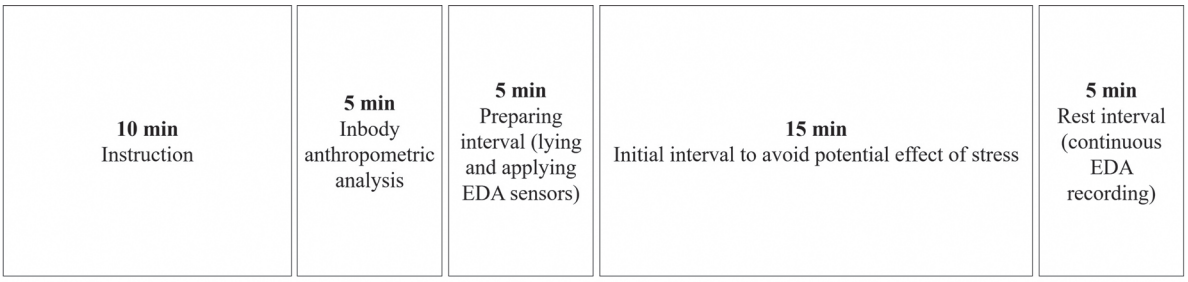

Figure 1. The time schedule of the examination protocol.

piratory, cardiovascular, endocrine diseases, other diseases potentially altering ANS activity (underweight, obesity, smoking, drug abuse). The control group did not suffer from any mental disorders. Additionally, body mass index (BMI) of both groups was calculated to exclude potential effect of under-/overweight on ANS (BMI: 20.00 ± 0.66 kg/m^2 in ASD; 19.15 ± 0.53 kg/m^2 in HC).

Protocol

The examination was performed in the Psychophysiological laboratory (Biomedical Center Martin, Jessenius Faculty of Medicine in Martin) under standard conditions with the minimization of internal/external stimuli, i.e. a quiet room, 22°C, air humidity around 50% in the morning between 8:00–11:30 a.m., after a light breakfast one hour before examination. The whole examination protocol lasted 40 min: initial 10 min necessary for all instructions, 5 min for the InBody anthropometric analysis (InBody 120, Biospace Co. Ltd. Soul, Korea), 5 min for lying comfortably on a special chair and placing the EDA electrodes. Afterwards, the initial phase lasting 15 min to avoid potential effect of stress was followed by 5 min of continuous EDA recording (Fig. 1).

Moreover, the ASD group´s examination protocol consisted of two periods. For the first time, the patients and their legal guardian came to get acquainted with the ambulance environment and the course of examination to minimization of external stress stimuli. For the second time, the ASD boys were examined according to protocol. For minimization of movement artifacts – we sufficiently instructed patients with ASD and controls to minimize any hand and fingers movements and to withstand the whole examination without any movement by positive motivation – small reward (e.g. chocolate) after examination. Continuous EDA was recorded with sampling frequency of 256 Hz (required by hardware) (FlexComp Infinity Biofeedback, Thought Technology, Canada). The EDA signal was monitored by two dry, Ag-AgCl, bipolar electrodes placed on the middle phalanges of two fingers on the left (non-dominant) hand (Blain et al. 2010; Poh et al. 2010; FlexComp Infinity Hardware Manual). The reusable EDA electrodes were carefully cleaned with an alcohol wipe before each examination. The baseline phase of the study lasted for 5 min. The typical time series of EDA in ASD and control group were shown in Figure 2.

Data analysis

Time- and spectral-domain parameters of EDA

Firstly, the tonic EDA was extracted by 10th order low-pass finite impulse response filter (cut off 0.0004 Hz) for time domain parameters. Next for spectral-domain indices, the raw EDA data were filtered with an 8th-order Chebyshev Type I low-pass filter and down-sampled to 2 Hz and to eliminate any trend the data were high-pass filtered with an 8th-order Butterworth filter (Posada-Quintero et al. 2016).

The skin conductance level (SCL) was calculated as a mean of tonic EDA. Typical values are ranged from 0 to 30 microsiemens (μS) according to electrodes size (Venables et al. 1980; Dawson et al. 2007; Boucsein 2012). Non-specific skin conductance responses (NS.SCRs) were evaluated as the rate of spontaneous skin conductance responses that occur without external stimuli (Boucsein 2012). The standard values are from 0 to 3 *per* min during baseline. The values over 3 indicate increased arousal states (Dawson et al. 2007). Index SCL expresses the level of mental arousal of the subject, while the NS.SCRs represent an amount of momentary arousal and define pulses in EDA signal (Figner and Murphy 2011; Braithwaite et al. 2013).

The power spectra of EDA signals were calculated using Welch's periodogram method with a 50% overlap. The mean power spectrum was evaluated by fast Fourier transform (with Blackman window length of 128 samples) and spectral powers (μS^2) in the fitting frequency bands (VLF: 0.000–0.045 Hz; EDA-Symp (sum of LF: 0.045–0.15 Hz and HF1: 0.15–0.25 Hz); HF: 0.25–0.40 Hz and VHF: 0.40–0.50 Hz) were achieved according to Posada-Quintero et al. (2016). Frequency-domain indices provide information about the

spectral distribution of sympathetic arousal in the skin (Posada-Quintero et al. 2016).

Entropy-based parameters of EDA

Approximate (ApEn) entropy defines the probability of similarity between vectors of length m (from time series of long N points) and vector of length m+1 within a given tolerance size r: ApEn (m, r, N) = $\Phi^{m}(r) - \Phi^{m+1}(r)$, where N is number of points, m is length of sequence (for N = 300 length of sequence m = 2), r is tolerance of similarity (r ϵ < 0.1 of standard deviation (SD); 0.25 SD>). Minimum values of ApEn (around zero) designate more predictable, regular system. In contrast, the elevated ApEn informs about a random and more intricate system (Pincus 1991; Pincus and Goldberger 1994).

Symbolic information entropy (SIE) is another entropy-based parameter that appears to be sufficient to detect the complexity of the systems and evaluate nonlinear characteristics of biological time series. Firstly, the time series is transformed by coarse-graining into symbolic sequences with a given alphabet into four numbers (0, 1, 2, 3) to identify the dynamic changes of data. Consequently, from these special number codes, the SIE could be generated (Yang and Liu 2014; Visnovcova et al. 2016). The numbers describe fluctuations of the time series, i.e., 0 and 1 specify slow and fast elevation of the data, respectively; 2 and 3 identify fast and slowly declining of the data, respectively. The patterns are arrayed into vectors of length L = 2 and they are matching into the group according to numbers of the vector. For instance, vector 20 means the change of SCL from a fast decrease to a slow increase in the waveform. Index SIE representing the uncertainty and messiness of the system can be defined as: SIE(m) = $-\Sigma^{M}_{i=1}(p_i * P_i)$, where p_i is probability of occurrence of individual vectors with length L in each groups; if $p_i > 0$, then $P_i = \log2(p_i)$, else if $p_i = 0$, then $P_i = 0$. Like ApEn, higher SIE values represent more unpredictable, irregular and chaotic signals, whereas standards around zero describe higher regularity and symmetry (Yang et al. 2014; Visnovcova et al. 2016).

Scaling-based parameters of EDA

Detrended fluctuation analysis (DFA) represents a scaling method for assessing the numerical self-similarity properties of the signals. DFA is used to endorse the presence of sustained long-term range relations in EDA signals. Generally, exponents α1 and α2 are estimated by linear regression as a trend line associating log F(n) to log (n) in the timescales. Indices α1 and α2 represent interactions over short (≤ 30 s) and long (> 30 s) timescales, respectively (Peng et al. 1995). The root-mean-square fluctuation F(n) of the signal was calculated according to Lanata et al. (2012) formula as:

$$F(n) = \sqrt{\frac{1}{N}\sum_{k=1}^{N}[y(k) - y_n(k)]^2}$$

Statistical analysis

Statistical analysis was performed by SYSTAT (SSI, Richmond, CA, USA). The Shapiro-Wilk normality statistical test was used for evaluation data distributions (Gaussian/

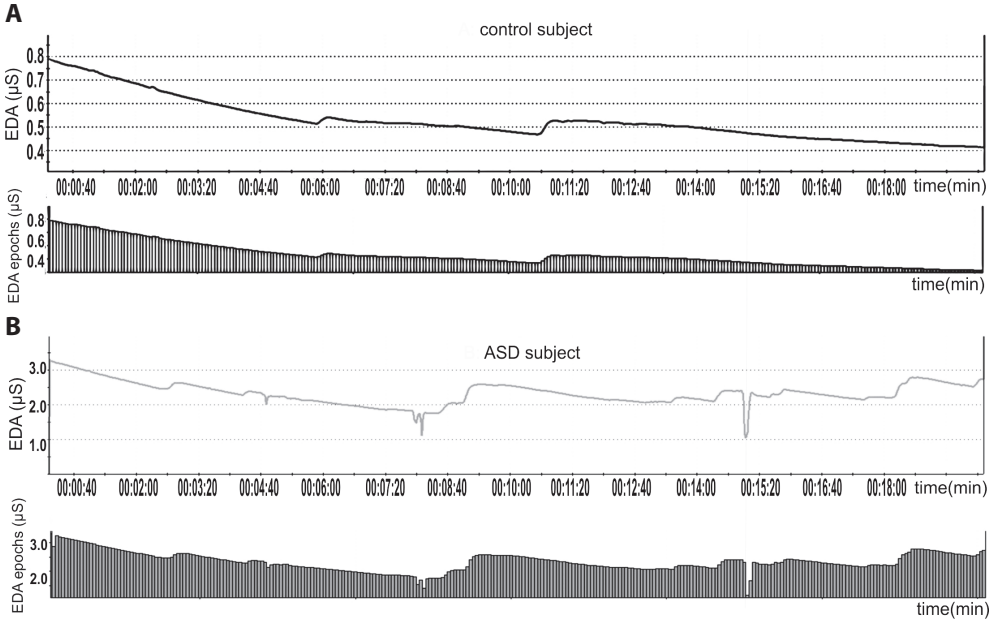

Figure 2. The typical time series and time epochs (averages every 4 s of record) of electrodermal activity (EDA). **A.** Control subject. **B.** Subject suffering from autism spectrum disorder (ASD).

non-Gaussian). Absolute values of spectral EDA indices (EDA-VLF, EDA-LF, EDA-HF1, EDA-Symp, EDA-HF2, EDA-VHF) differed greatly among individuals, therefore, they were logarithmically transformed for next statistical analysis. Consequently, the unpaired Student´s *t*-test was used for between-group comparison of EDA indices because data were normally distributed. Additionally, for multiple hypothesis testing was used the Benjamini-Hochberg (BH) correction of *p* value (p_{BH}). EDA parameters were expressed as mean ± SEM. The results are considered statistically significant if the following logical conjunction applies: $p < 0.05 \Lambda p_{BH} < 0.05 \Lambda p < p_{BH}$. The associations between the EDA parameters were analysed using Spearman´s rank-order correlation test, where a value of $p < 0.05$ (two-tailed) was considered statistically significant.

A predictive model of the case-control status was built using the RF machine learning algorithm. RF was trained on the data, and the nested cross-validation algorithm with the minimum graph depth criterion was used to select important features (EDA parameters). The receiver operating characteristic (ROC) curve generated by the RF with selected EDA indices was used to quantify their predictive ability. The ROC curve was constructed from the out-of-bag data. The data analyses were performed in R Core Team (The R Project for Statistical Computing, https://www.r-project.org/) v. 3.5.2, using libraries beeswarm (Eklund 2020), robustbase (Maechler et al. 2020), randomForestSRC (Ishwaran and Kagalur 2020), ggRandomForests (Ehrlinger 2020).

Results

The comparison between groups of time-domain EDA parameters

In general, boys with ASD had a slightly higher SCL than controls, however, these differences were not significant. The ASD patients had significantly higher number of NS.SCRs compared to controls ($p = 0.0070$; $p_{BH} = 0.0071$).

The comparison between groups of spectral-domain EDA parameters

The spectral indices lnEDA-HF2 and lnEDA-VHF were significantly increased in ASD compared to controls ($p = 0.0165$, $p_{BH} = 0.0213$; $p = 0.0138$, $p_{BH} = 0.0142$; respectively). No significant differences in remaining spectral EDA indices were found between groups.

The comparison between groups of nonlinear EDA parameters

Nonlinear entropy-based parameters ApEn and SIE revealed significant increasing in ASD compared to controls ($p = 0.0249$, $p_{BH} = 0.0425$; $p = 0.0224$, $p_{BH} = 0.0283$; respectively). Index α1 was significantly lower in ASD compared to controls ($p = 0.0236$, $p_{BH} = 0.0354$). No significant changes were found in remaining nonlinear parameters. All results are summarized in the Table 1.

Table 1. The parameters of electrodermal activity (EDA)

EDA parameter	df	Controls	ASD	*p*	p_{BH}
SCL (μS)	84.00	1.46 ± 0.17	1.61 ± 0.24	0.6000	0.0779
NS.SCRs	84.00	2.05 ± 0.47	4.60 ± 0.77	0.0070	0.0071
lnEDA-VLF(μS^2)	83.00	1.70 ± 0.18	1.95 ± 0.18	0.3310	0.0708
lnEDA-LF(μS^2)	83.00	1.88 ± 0.16	2.17 ± 0.18	0.2268	0.0638
lnEDA-HF1(μS^2)	83.00	0.81 ± 0.13	1.23 ± 0.18	0.0662	0.0495
lnEDA-Symp(μS^2)	83.00	1.94 ± 0.15	2.25 ± 0.18	0.2020	0.0567
lnEDA-HF2(μS^2)	83.00	0.22 ± 0.13	0.73 ± 0.16	0.0165	0.0213
lnEDA-VHF(μS^2)	83.00	−0.78 ± 0.16	−0.20 ± 0.17	0.0138	0.0142
ApEn	84.00	0.57 ± 0.03	0.68 ± 0.03	0.0249	0.0425
SIE	84.00	2.08 ± 0.08	2.37 ± 0.09	0.0224	0.0283
α1	84.00	1.23 ± 0.06	1.05 ± 0.05	0.0236	0.0354
α2	84.00	0.47 ± 0.06	0.50 ± 0.06	0.7311	0.0850

Values are expressed as mean ± SEM. SCL, skin conductance level; NS.SCRs, nonspecific skin conductance responses; EDA-VLF, electrodermal activity in very low frequency band; EDA-LF, electrodermal activity in low frequency band; EDA-HF1, electrodermal activity in high 1 frequency band; EDA-Symp, EDA in the sum of low and high 1 frequency bands; EDA-HF, EDA in high 2 frequency band; EDA-VHF, EDA in very high frequency band; ApEn, approximate entropy; SIE, symbolic information entropy; α1 and α2, indices of detrended fluctuation analysis short-term and long-term correlations, respectively; df, degree of freedom; ASD, autism spectrum disorder; BH, Benjamini Hochberg correction of *p* value. The *p* value expresses the comparison between groups. The results are considered statistically significant if the following logical conjunction applies: $p < 0.05 \Lambda p_{BH} < 0.05 \Lambda p < p_{BH}$.

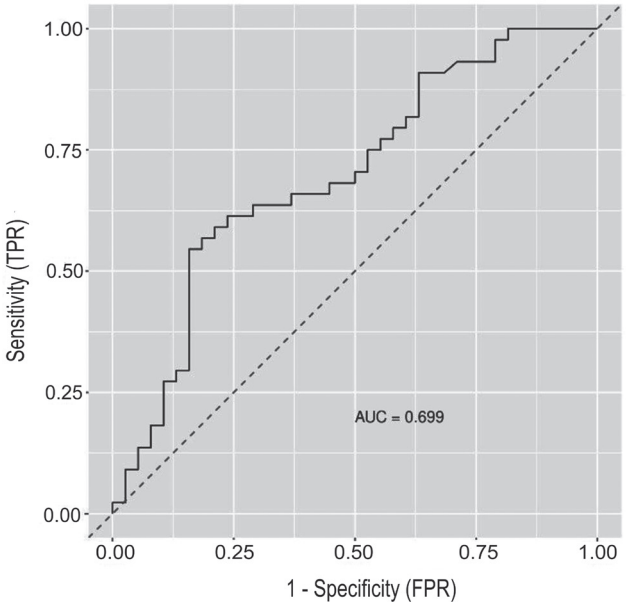

Figure 3. Receiver operating characteristic (ROC) curve analysis for the predictive potential of nonlinear (SIE, and α1) and spectral (EDA-HF1, EDA-HF2, EDA-VHF) indices and BMI (selected based on random forest analysis) in autism detection. The dashed line represents ROC curve of random classifier with AUC = 0.5; AUC, area under the curve; FPR, false positive rate (1 -Specificity); TPR, true positive rate (Sensitivity).

Correlation analysis

Correlation analysis in the total group (control group and ASD group together) revealed significant positive correlations between time index SCL and spectral indices EDA-VLF, EDA-LF, EDA-Symp, EDA-VHF. SCL showed significant negative correlation with DFA indices α1 and α2. Index NS.SCRs significantly positively correlated with all spectral-domain indices (EDA-VLF, EDA-LF, EDA-HF1, EDA-Symp, EDA-HF2, EDA-VHF). Spectral indices (EDA-HF1, EDA-Symp, EDA-HF2 and EDA-VHF) significantly positive correlated with SIE. Spectral indices EDA-VLF, EDA-LF, EDA-Symp and EDA-VHF significantly negative correlated with DFA α2 index. Significantly negative correlations were found between nonlinear entropy-based parameter ApEn and nonlinear scaling DFA parameter α1. No significant correlations were found in the remaining parameters. All results from correlation analysis are summarized in the Table 2.

Random forest machine learning analysis

To assess the predictive value of the studied EDA indices, ROC curve was generated. Based on random forest analysis, nonlinear (SIE, and α1) and spectral (EDA-HF1, EDA-HF2, EDA-VHF) indices and BMI were selected as important predictors. According to the ROC curve (Fig. 3) and area under the curve (AUC, 0.699), the predictive ability of these EDA indices is moderate.

Discussion

The main results can be summarized as follows: (1) several parameters of time (NS.SCRs), frequency (EDA-HF2 and EDA-VHF) and entropy-based nonlinear (ApEn and SIE) analysis were significantly higher indicating sympathetic overactivity in boys suffering from autism compared to controls; (2) in contrast, the nonlinear scaling index of DFA – α1 showed different pattern compared to the time, frequency and entropy-based EDA characteristics – it was decreased in

Table 2. The correlation analysis of the EDA parameters in total group (control group, and ASD group together)

	SCL	NS.SCRs	lnEDA-VLF	lnEDA-LF	lnEDA-HF1	lnEDA-Symp	lnEDA-HF2	lnEDA-VHF	ApEn	SIE	α1
NS.SCRs	0.217	–									
lnEDA-VLF	0.557**	0.677**	–								
lnEDA-LF	0.452**	0.737**	0.927**	–							
lnEDA-HF1	0.144	0.698**	0.735**	0.880**	–						
lnEDA-Symp	0.436**	0.743**	0.919**	0.998**	0.898**	–					
lnEDA-HF2	−0.114	0.644**	0.534**	0.708**	0.911**	0.729**	–				
lnEDA-VHF	0.289*	0.503**	0.316*	0.480**	0.652**	0.496**	0.846**	–			
ApEn	0.109	0.154	0.201	0.217	0.237	0.220	0.230	0.151	–		
SIE	−0.131	0.146	0.147	0.247	0.364**	0.409**	0.409**	0.369**	0.152	–	
α1	−0.453**	−0.097	−0.096	−0.076	0.024	0.007	0.069	0.007	−0.424**	−0.035	–
α2	−0.908**	−0.174	−0.406**	−0.331*	−0.083	−0.310*	−0.145	−0.310*	−0.193	0.045	0.551**

Values are expressed as Spearman´s rank-order correlation coefficient. NS.SCRs, nonspecific skin conductance responses; ApEn, approximate entropy. For other abbreviations, see Table 1. * correlation is significant at the level 0.05; ** correlation is significant at the level 0.001.

ASD providing thus distinct information about complexity of sympathetic cholinergic control network in autism; (3) in addition, the correlation analysis revealed negative correlation between nonlinear scaling index DFA and time, spectral and entropy-based parameters.

It seems that ASD is characterized by the sympathetic overactivity indexed by the several EDA quantitative time (NS-SCRs) and frequency (lnEDA-HF2 and lnEDA-VHF) domain indices and qualitative entropy (ApEn and SIE)-based nonlinear indices. This assumption is in agreement with other studies which revealed tachycardia (Kushki et al. 2013; Bujnakova et al. 2016; Ming et al. 2016), shortened pre-ejection time (Neuhaus et al. 2016), and elevated diastolic blood pressure (Ming et al. 2016) indicating higher sympathetic activity in children suffering from autism. In contrast, Bricout et al. (2018) revealed attenuated sympathetic vasomotor regulation indexed by blood pressure variability during head-up tilt test in ASD children. In this respect, EDA indices could represent another part of the assessment of sympathetic activity, i.e. cholinergic innervation from sudomotor fibres associated with sympathetic control (Posada-Quintero and Chon 2020), and complete the complex sympathetic response in autism.

Furthermore, we used EDA nonlinear analysis revealing complex qualitative features of this physiological bio-signal. In contrast to nonlinear entropy-based parameters, the scaling DFA parameter α1 was decreased in boys suffering from autism. Moreover, the correlation analysis revealed negative correlations between DFA and other EDA indices (time, spectral and entropy-based parameters). This variation in assessment of complex and multipart characteristics of signal could be explained by mathematical point of view, where the fundamental principle of entropy is the finding the probabilities of distribution of similar vectors with certain length into time series (Richman and Moorman 2000), while α1 represents scaling method evaluating statistical self-similarity relationships over short-time scale (Lanata et al. 2012). The elevation of entropy-based parameters could be explained by ASD symptoms like stereotype and repetitive behaviours, which might result in increased distribution in similar EDA patterns needed for entropy analysis.

From neurophysiological point of view, the altered EDA could indicate the differences in sympathetic modulation to meet task demands associated with central/peripheral neurobiological differences in ASD (Panju et al. 2015; Haigh et al. 2020). More specifically, a widespread network of regions, including the amygdala, the prefrontal, anterior cingulate, and insular cortices, is included in the sympathetic regulation (Pina-Camacho et al. 2012). ASD has been associated with abnormalities in neuroanatomical functioning and connectivity in these regions affecting thus sympathetic cholinergic functioning also at the peripheral level (Panju et al. 2015). Therefore, we assume that the EDA analysis in time and frequency domains and entropy could represent a promising and sensitive tool for early ASD-linked alterations in sympathetic regulatory integrity.

Recently, the machine learning algorithms (e.g. RF) are more often used in clinical research to select relevant predictors potentially contributing to differentiation of mental disorders. In this context, the RF algorithm has identified three spectral EDA (EDA-HF1, EDA-HF2, EDA-VHF) and two nonlinear EDA (SIE, α1) indices as relevant predictors for ASD differentiation. The predictive power of EDA parameters resulting from the ROC curve revealed only moderate ability to discriminate patients suffering from autism. From this perspective, the predictive model of EDA indices based on RF algorithm is not specific enough to differentiate ASD patients, however, the potential inclusion of traditional ASD assessment such as questionnaires (e.g. ADOS II) (Raya et al. 2020) or other autonomic parameters (e.g. indices from the heart rate and blood pressure analysis) (Tonhajzerova et al. 2021) in the predictive model could improve its predictive power in ASD differentiation.

Conclusions

Our study revealed increased EDA time, spectral and nonlinear indices based on entropy indicating sympathetic overactivity in ASD boys. However, the nonlinear index α1 – as a short-time parameter of scaling method – showed different pattern of complexity of sympathetic cholinergic network in ASD. We suggest, that complex and detailed EDA analysis could reveal initial abnormalities of sympathetic regulation in ASD even before commonly used SCL assessment.

Limitations of the study

In present study, the cohort consisted of a relatively small homogenous sample of male adolescents with ASD; therefore, it needs to be validated in a larger cohort with respect to gender. The male group was used because the prevalence of ASD is four time higher in male compared to female population and to exclude sex differences (Loomes et al. 2017). Moreover, special questionnaires or any tests to obtain psychological profiles in ASD were not used in this study. We suggest that further research to study relations between ASD-linked psychological symptoms and sympathetically mediated indices based on EDA biosignal is needed. Further, EDA represents a noninvasive index of the sympathetic cholinergic system; therefore, it is not possible to apply these results in terms of general sympathetic dysregulation associated with autism spectrum disorder. Although sympathetic overactivity represents a potential pathomechanism contributing to cardiovascular complications, it is not possible to

determine cardiovascular risk based on altered sympathetic regulation indexed by EDA alone. Future research based on non-invasive continuous simultaneous and analysis of other physiological parameters providing information on sympathetic regulatory mechanisms (e.g. blood pressure variability, systolic time intervals) is necessary.

Conflicts of interest. The authors declare no conflict of interest.

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