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Physiological responses to dyadic interactions are influenced by neurotypical adults' levels of autistic and empathy traits

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Keywords: Social Processing; ASD

HIGHLIGHTS

- Atypical responses to social situations are a feature of ASD.
- In the general population individuals vary for their social abilities.
- Distinct heart rate responses underlie high vs low autistic and empathy traits.
- Distinct temperature response underlie high vs low empathy traits.
- Physiological responses can be biomarkers for autistic traits and social abilities.

Abstract

Autistic traits are distributed on a continuum that ranges from non-clinical to clinical condition. Atypical responses to social situations represent a core feature of the Autism Spectrum Disorders phenotype. Here, we hypothesize that atypical physiological responses to social stimuli may predict non-clinical autistic and empathy traits levels. We measured physiological responses (heart rate, facial temperature) of 40 adults (20F) while showing them 24 movies representing dyadic interactions. Autistic traits were assessed through Autism Quotient questionnaire (AQ), while empathy traits were measured using the Empathy Quotient questionnaire (EQ). Opposite correlations between AQ and EQ scores and physiological responses were found. Analysis of physiological responses revealed that individuals with better social abilities, low AQ and high EQ, show opposite activation patterns compared to people with high AQ and low EQ. Findings show that physiological responses could be biomarkers for people's autistic traits and social abilities.

1.Introduction

1.1 Social stimuli processing in ASD

Autism Spectrum Disorders (ASDs) are characterized by multi-trait symptoms. A key feature of the disorders which characterize the spectrum is the presence of impairments in social interactions [1,2]. ASDs infants show atypical behavioral displays, which impair the construction of a typical relation between caregivers. For example, ASDs infants cry differently from typical development infants and do not adjust their body posture as expected when caregivers pick them up, therefore caregivers cannot respond to ASDs infants needs as naturally as they do for typical infants [3–5]. Since early social interactions are basic for the overall development of individuals, this initial impairments will negatively influence ASDs infants' development throughout social, emotional and cognitive domains. Researchers demonstrated that ASDs impairments in social interactions are characterized by at least two components: anomalous processing of social vs non-social stimuli and lack of comprehension of others' intentions and feelings. Individuals with ASDs show atypical responses when processing social cues, such as during face or emotion recognition tasks [6–9]. Enticott and colleagues (2014) [10] showed that there is a poorer performance in participants with ASD compared to matching controls, in a face categorization task using emotional cues. Furthermore, within the ASD group, participants had greater difficulty completing the task when they were shown dynamic stimuli, such as faces which changed their emotional expressions, as opposed to still faces. Being able to read the changes in facial expressions forms the basis of effective social interactions. People with ASDs show impairment in performance in response to dynamic social displays [10,11], which suggests that they will have increased difficulties in producing socially appropriate behavioral responses. Moreover ASDs' social impairments involved deficits in empathetic abilities, defined as the ability to understand and share others' feelings [12–14]. A recent study by Gu and colleagues (2015) [15] showed that ASDs individuals have lower levels of empathy traits measured with the Empathy Quotient questionnaire and that their ability in discriminate situations in which the subject of a picture was going to feel pain vs neutral situation was lower compared to typical developing individuals. Together with decrease empathetic abilities, ASDs individuals show a higher systemizing, defined as a drift towards the analysis and the exploration of rule-governed systems, where rules are fixed and easily inferred. This systemizing drift is highlighted by the high and often selective attention that ASDs individuals show towards objects [16].

1.2. Autistic traits: a continuum in the population

This differential processing between social and physical stimuli found in ASD individuals has also been found in people with neurotypical development with varying levels of autistic traits. Atypical characteristics commonly found in ASD patients can also be found in neurotypical individuals who present high

levels of autistic traits [17–19]. The presence of autistic traits in the neurotypical population supports the idea that autistic traits are distributed as a continuum in the population and individuals with higher levels of traits easily develop atypically within the Autism Spectrum Disorders [20,21]. Indeed, in parents and siblings of individuals diagnosed with ASDs usually high levels of autistic traits are found, such as differential social stimuli processing, higher systemizing and lower empathy. The continuum of traits allow us to investigate and better understand autistic traits, their characteristics and the possible interventions within a neurotypical population, without the possible confounding effect of developmental impairments. Moreover, a good assessment of typical adults' autistic traits levels could detect in the population which individuals are located within the higher extreme of the continuum and, thus, it could help in identify infants at risk for the development of ASDs.

1.3. Biomarkers and dynamic stimuli: reliable and ecological measurements

Diagnosis of ASDs as well as autistic traits assessments in the neurotypical population are carried out through behavioral observations and measurements. To diagnose a ASDs clinicians need to observe patients' behaviors delaying the diagnosis, because ASDs signs become visible at about 18-months of age, or even later if the disorder is not severe. An earlier diagnosis would be of great importance in order to start as soon as possible with the interventions and, thus, to enhance infants probability of gaining a typical development. Also, to assess the presence of autistic traits in the neurotypical population, behavioral measures are available, such as the Autism Quotient (AQ) questionnaire [22]. However, self-reported questionnaires are easily biased by misunderstandings or by the desire to give socially appropriate answers [23], and these may be a challenge for the interpretation of results [24]. Therefore, it would be of great importance to add to behavioral assessments univocal objective and reliable biomarkers in order to obtain earlier diagnosis in infants and to better assess autistic traits in adults.

Moreover, usually, research that involves social stimuli is conducted using static pictures or simple dynamic social stimuli, such as still faces with expression changes. However, social stimuli in daily life are embedded in dynamic and complex contexts. Since autistic traits and symptoms are expressed within real social interactions It would be useful to assess the presence of biomarkers in response to more ecological but still controlled scenarios, such as movies representing interactions between people [25].

1.4. Physiological findings

From a physiological perspective, researchers found differential activations in both the central and the autonomic nervous system in ASDs individuals in response to social stimuli. Concerning central nervous system responses, ASDs behavioral responses to social stimuli have been found to rely on differential patterns of brain electric activity [15, 26]. Indeed, there is an atypical ERP pattern of the cortical activation in typically developing

individuals and ASD affected individuals in response to facial expression of emotion [27]. Moreover, the atypical hemodynamic and electrical responses to social stimuli measured with fMRI and ERP in the ASD population compared to the neurotypical population points to atypical cerebral activations and suggests an impaired social understanding [26]. Also, the activations of the Autonomic Nervous System (ANS) have been found to highlight the different behavioral responses in ASDs for social stimuli and objects. The ANS is formed by two branches, the Parasympathetic Nervous System (PNS) and the Sympathetic Nervous System (SNS). The two ANS branches handle different situations and are constantly on balance. The PNS maintains the homeostasis of the body and it is active during resting states, during the PNS activation the heart rate slows down and all the bodily functions, such as the digestion, are activated. Index of the activation of the PNS is, for example, a decrease in the heart rate. Whereas the SNS is responsible for the activation of the body during threatening or dangerous situations, the so-called “fight or flight response”. During the SNS activation the heart rate speeds up, the blood is displaced towards limb muscles in order to prepare the body to react to the situation increasing the peripheral temperature and the sweat glands in the hands enhance their activity heightening the quantity of sweat on the hands. Indexes of the activation of the SNS are an increase in the heart rate and in peripheral skin temperature, and an enhanced electrodermal activity. The electrodermal activity on the hands measures how fast the skin conducts electricity on its surface, the more sweat there is on the hands, the higher is skin conductance. Since the sweat on hands increases during stressful situations, a higher skin conductance is an index for SNS activation. The atypical processing of social and physical stimuli found in ASD affected individuals can also be seen by different Autonomic Nervous System (ANS) activations. Indeed, by measuring the electrodermal activity in ASD affected children, Hirstein and colleagues (2001) [28] found a poor sympathetic activation in response to mothers' faces compared to inanimate objects, such as cups. It would also be important to find physiological biomarkers of autistic traits in the neurotypical population. In a recent study, Singleton and colleagues (2014) [29] investigated the sympathetic nervous system activation in response to social and non-social static stimuli in neurotypical adults with varying levels of autistic traits, measured with the self-reported Autism Quotient (AQ) questionnaire [22]. Authors measured skin conductance variations in response to social stimuli (picture of faces and stylized faces) and non-social stimuli (picture of objects). They found that the higher the level of autistic traits, the higher the sympathetic response in reaction to a picture of an object, and the lower their sympathetic response to stylized faces. Conversely, individuals with low levels of autistic traits showed a lower sympathetic response to pictures of objects and a higher sympathetic response to pictures of stylized faces. Findings show a higher arousal in response to non-social condition in people with high autistic traits while people with fewer autistic traits displayed a greater arousal in response to stylized social images. This highlights the presence of physiological biomarkers that reflect the different processing for social and physical stimuli also in a neurotypical population with distinct autistic traits levels, however it would be useful to see whether it is possible to find physiological signs also for different empathetic abilities in a neurotypical population. Then, if findings are

extended to an ASD population, the physiological responses to social and nonsocial stimuli could be part of the mechanism that underlies the increased systemizing and decreased empathizing found in ASD individuals.

1.5. Aims of the study

In our study we aimed to investigate how individuals' autistic and empathy traits influence physiological responses to dynamic social interactions. We showed participants movies representing interactions between two actors and within the exchanges we changed the topic of the interaction. Each interaction focused either on a social norm which people are expected to follow or on a physical norm which objects are expected to follow. A social norm, for example, states that if a person is trying to step on movable platform to change a light bulb, and risks falling, we expect the second person to aid him/her by holding the chair and not walk away (see Fig. 1(A-C)). A physical norm, instead, states that if one person lays a box on a table out of its centre of gravity, the box must fall down and not stand still (see Fig. 1(D-F)).

Each norm, social or physical, was represented in two versions: (i) in the appropriate scenario, where the social behavior or the physical object follow the social or physical norm as expected, while (ii) in the inappropriate scenario, where the social behavior or the physical object do not follow the social or physical norm as expected. We changed the focus within the interaction because we expected the focus, social vs physical event, to differentially enhance participants' physiological responses according to their levels of autistic and empathy traits, even if the social or physical focus was linked to a dyadic interaction. Specifically, we expected people with low autistic traits and high empathy traits to show higher sympathetic responses to inappropriate dyadic interactions focused on a social norm, whereas we expected people with high autistic traits and low empathy traits to show higher sympathetic responses in reaction to inappropriate interactions focused on a physical norm. Unlike Singleton and colleagues [29], we included multiple measurements of indexes of the ANS components (PNS and ANS) in order to gain an overall picture of the autonomic mechanisms underlying individuals' responses to social interactions. Moreover we added a measure of individuals' empathy and used as stimuli dynamic scenarios representing realistic interactions between people. An abnormal activation of the ANS in response to socially or physically focused interactions in individuals with higher levels of autistic traits would be an important physiological biomarker for the presence of autistic traits which allows for a more objective and reliable measure in the assessment of autistic traits.

2. Material and methods

2.1. Participants

Forty neurotypical adults (20 males, 20 females) who were undergraduates at the University of Trento (northern part of Italy) were recruited as participants. Participants' mean age was 21.8 years old (SD = 3.26), all were Caucasians.

Participants were given university credits. Informed consent was obtained from all individual participants included in the study. The study was conducted in accordance with the Helsinki Declaration as revised in 2000.

2.2. Stimuli

We used dynamic video clips representing realistic situations as the stimuli in our experiment. We created 48 video clips depicting realistic dyadic interactions between two male student actors (with similar bodily features) who were recruited from the Department of Psychology and Cognitive Science of Trento University. The video clips were filmed in a closed environment with artificial central light and a white background. In each video clip at the beginning there is only one actor in the scene, while towards the end of the video, a second actor enters the scene and the two actors start to interact. The onset of the interaction, defined as the moment the two actors look at each other, occurred

always at 22 s after the start of the movie. In order to investigate participants' reactions to the presence of empathy-activating or systemizing activating events within the social interaction, in each video clip the interaction focused either on a social behavior, which is supposed to follow a social norm and is played out within the social exchange, or focused on an object, which is supposed to follow a physical law. A social norm, for example, states that if a person is trying to stand on a movable platform to change a light bulb, risking to fall, we are expected to aid him/her holding the chair and not to walk away. A physical norm, instead, states that if one person lays a box on a table out of its centre of gravity, the box must fall down and not stand still. Each norm, social or physical, was represented in two versions: (i) in the appropriate scenario, where the social behavior or the physical object follow the social or physical norm as expected, while (ii) in the inappropriate scenario, where the social behavior or the physical object do not follow the social or physical norms expected. In the social scenario described above, for example, the appropriate event is represented by the second person helping the first person holding the movable platform, whereas in the inappropriate event the second person, after the aid request, simply walked away (See [Fig. 1\(A-C\)](#)). For the physical scenario described above, in the appropriate scenario the actor lays the box completely on the table, whereas in the inappropriate scenario, the actor lays the box on the table out of its centre of gravity and the box does not fall down (See [Fig. 1\(D-F\)](#)). To sum up, there were four video-clip conditions; (i) appropriate social norm (the second person holds the wheelchair); (ii) inappropriate social norm (the second person walks away without helping); (iii) appropriate physical norm (the box rests completely on the table); (iv) inappropriate physical norm (the box does not fall even if it lays on the table out of its gravity centre). The manipulation regarding the appropriateness of the norms (social or physical) was included to control for possible confounding physiological activation triggered by the surprise that might result from something new happening in each clip. Moreover, each scene was shown two times, one with verbal communication between the actors, the other with non-verbal communication between the actors. We used both verbal and non-verbal interactions for three reasons: (i) Verbal language could have affected the physiological responses, hence the non-verbal conditions controlled for

the effect of the verbal communication. (ii) We wanted to find out if non-verbal communication clips are more difficult to understand than verbal communication clips. (iii) The study will be extended to a Japanese sample, hence the condition with nonverbal communication will allow us to continue the study in cross-cultural contexts using the same stimuli used in Italy. Each video clip was 27 s long, with two seconds allotted for the title of the scene and 25 s

for the video. The target event (social or physical, appropriate or inappropriate) occurred at the twenty-second second. The video clips continued for five additional seconds after the target event occurred because in the pilot study we determined that 5 s was the most appropriate post-event timing to both obtain the physiological measurements and to maintain participants' attention. In a subsequent pilot study 20 participants were asked to rate forty-eight video clips on 7-points Likert items for how comprehensible they were. Participants were asked three questions for each video-clip: (i) what the clip represented, (ii) whether something unexpected happened during the clip, coded as 1 (something unexpected happened) or 0 (nothing unexpected happened) (inappropriate: $M = 0.78$, $SD = 0.09$; appropriate: $M = 0.11$, $SD = 0.08$), (iii) how comprehensible the scene was on a 7-points Likert item. The twenty-four video clips rated as most comprehensible were selected as stimuli for the present research. The mean of the comprehensibility rating for the chosen appropriate videos was 5.8 ($SD = 0.4$), while the mean value for the comprehensibility scores of the appropriate discarded videos was 3 ($SD = 1$).

Fig. 1. Examples of the possible social interactions represented. (A–C) Example of social event: Holding the movable platform. (A) One person tries to stand on a movable platform to change a light-bulb and risks falling. A second person arrives and the first person asks him for help. (B) Appropriate continuation of a social event: the second person helps the first one by holding the movable chair. (C) Inappropriate continuation of a social event: the second person pauses to stare at the first one, and leaves the scene without helping. (D–F) Example of physical event: Box delivering. (D) One actor is working at the computer, the second actor enters the scene delivering a box to the first one by placing it on the table. (E) Appropriate continuation of a physical event: the second actor places the box completely on the table. (F) Inappropriate continuation of a physical event: the second actor places the box on the table out of its centre of gravity and the box does not fall down.

2.3. Procedure

The experiment was conducted in two parts: the first part involved completion of an online survey for autistic and empathy traits assessment, and the second part was carried out in a laboratory in the Psychology Department, at the University of Trento. In the laboratory, the participants were seated approximately one and a half meters away from the wall where the video clips were projected (the screen dimensions on the wall were: 1.50 × 1.00 m). Each participant viewed a sequence of the twenty-four video clips. The video clips presentation order was randomized three times, creating three presentation sequences. Each participant was assigned one presentation sequence among the three sequences available, so that the presentation order was counterbalanced across participants. Overall, all participants viewed all the video clips. After each video clip, a screen with the Likert items appeared and participants were asked to give the two behavioral judgments. Participants verbally gave their judgments, which were recorded by a visible video camera. Participants' heart rate and peripheral temperature were measured throughout

the entire presentation of the twenty-four video clips. Overall, the experimental session lasted 40min.

2.4. Measures

2.4.1. Autistic traits – the AQ

In order to assess participants' levels of autistic traits, they were asked to complete the 50-items Autism Quotient questionnaire [22] as part of an online survey that also collected demographic information such as age, gender, and educational level. For our study, we chose to rely on the Autism-spectrum Quotient because it covers the individual's behavior across five domains associated with the reaction towards social compared to physical stimuli: communication abilities, social skills, attention switching, imagination, and attention to details. The internal consistency of the Italian version of the AQ calculated with Cronbach's Alpha Coefficients was moderate to high (AQ total=0.76, communication = 0.64, social skills = 0.68, imagination = 0.52, local details = 0.58, attention switching= 0.54) [30]. Moreover the AQ questionnaire has consistent results both across time and cultures [31], and is able to stratify the neurotypical population accordingly to individuals' levels of autistic traits [22,29,32]. Answering each question was mandatory, thus there were no missing data. The results were calculated according to Baron-Cohen et al.'s scoring scheme [22], resulting in an "AQ score" for each participant. All participants were included in the study, regardless of their final AQ score.

2.4.2. Empathy traits – the EQ

Empathetic abilities are associated with the comprehension of others' behaviors and feelings and are required when participating or assisting in social interactions. Participants answered the 40-items Italian short version of the Empathy Quotient questionnaire as translated by Preti and colleagues [33,34]. The original version of the EQ has 60- items, however, it consists of 40 clinical items and 20 filler items, and in the short version, only the clinical items are included [32,34]. In the original research by Baron-Cohen and colleagues, EQ was proven to be a discriminating factor between neurotypical and ASDs adults since the latter group who completed the questionnaire scored significantly lower than control participants ($t(178) = -13.07$, $p < 0.001$) [32]. The results were calculated according to Baron-Cohen et al.'s scoring scheme, which is the same as the Italian version, obtaining an "EQ score" for each participant [34,35]. All participants were included in the study, regardless of their final EQ score.

2.4.3. Heart rate (HR)

As an index of both parasympathetic and sympathetic nervous system activation we assessed participants' heart rate using a pulse oximeter (CONTEC CMS60D) placed on participants' left index finger [36, 37]. The activation of the parasympathetic nervous system would lead to a decrease in the heart rate, whereas the activation of the sympathetic nervous system

could lead to either an increase or a decrease in the heart rate values. Depending on the stimuli valence, sudden possible threatening external stimuli makes the heart rate decrease [38], while in general attention-getting positive stimuli makes the heart rate increase [36]. The sampling rate of the Oximeter (CONTEC CMS60D) was 64 Hz and it recorded at a 1 HZ sampling rate (one value per second, where each measure represents the average of the previous 64 samples). In participant with lower autistic traits and higher empathy traits we expected to find a higher sympathetic activation (increase in heart rate) in response to socially-focused appropriate interactions. Whereas in participants with higher autistic traits and lower empathy traits we expected to find a higher sympathetic activation (increase in heart rate) in response to physically-focused appropriate interactions.

2.4.4. Temperature (T)

As an index of sympathetic activation we assessed participants' peripheral surface body temperature on the cheeks (both left and right) by placing two thermistors on participants' skin (Applent at4524 multi-channel temperature meter) [39]. The sampling rate of the thermistor (Applent at4524) was 64 Hz and it recorded at a 1 HZ sampling rate (one value per second, where each measure represents the average of the previous 64 samples). Literature findings about facial temperature are not univocal and it is shown that the activation of the sympathetic nervous system may be highlighted by either a decrease or an increase of skin temperature on the face. A decrease in nose temperature has been found in response to stress-eliciting stimuli, such as feelings of guilt or threat-related stimuli, in infants, adults and also non-human primates [40,41,42]. Pavlidis and colleagues (2000) [43] found a decrease on cheek temperature in response to sudden unexpected loud sounds. On the other hand, Hahn and colleagues (2016) [44] found an increase in nose temperature in response to social contact. They recorded participants' facial temperature while the experimenter was touching them on high-intimate (face, chest) or low-intimate (arm, palm) locations and they found a higher increase in facial temperature in response to high-intimate touch, especially in the case of opposite- sex experimenter. These differential directions of temperature changes consequent of sympathetic nervous system activations likely depend on the type of stimuli the organism is facing. Since we showed realistic dyadic social interactions between two adults and considering the attentional drift towards physical objects found in individuals' with high autistic traits, we expected participants with lower autistic traits and higher empathy traits to show greater increase in cheek temperature in response to socially-focused appropriate interactions compared with other conditions, while participants with higher autistic traits and lower empathy traits to show an increase in cheek temperature in response to physically-focused appropriate interactions compared with other conditions.

2.4.5. Behavioral judgment

During the experimental session participants were asked to rate on 5-point Likert items, how much each video clip was predictable and comprehensible. Each participant was asked, "How predictable was the scene?" and "How

comprehensible was the scene?”. We assessed participants' behavioral judgments in order to both investigate whether the expected ANS activation was reflected by behavioral differences and to maintain participants' active attention throughout the experimental session.

2.5. Preliminary analysis

Prior to data analysis, univariate and multivariate distributions of heart rate values, temperature values and behavioral judgment scores were examined for normality, homogeneity of variance, outliers, and influential cases [45]. To normalize the distribution of temperature values a cubic transformation was applied. After the cubic transformation all of these variables were normally distributed and absolute skewness values were $b1$ for all variables. The distance of each case to the centroid was evaluated to screen for multidimensional outliers [45]. There were no significant differences between female and male participants in physiological values nor behavioral judgements (HR: $t(1,38)=-1.9$, ns; T left cheek: $t(1,38)=-0.94$, ns; T right cheek: $t(1,38)=-0.98$, ns; Predictability judgments: $t(1,38) = -0.96$, ns; Intelligibility judgements: $t(1,38) = -0.10$, ns). For this reason, females and males were treated as a single group for analyses. No significant differences were found in the comprehensibility scores between verbal and non-verbal scenarios, thus we considered for the analysis only the non-verbal scenarios in order to avoid the effects of language on physiological responses. Autism Quotient scores and Empathy Quotient scores were, as expected, negatively correlated ($r = -0.29$, $p < 0.001$); however, these two variables shared only 8.25% of their common variance, and so we treated them independently.

2.6. Analysis

Significant threshold was set at $p < 0.05$. To measure participants' reactions to dyadic interactions, we considered the physiological responses in the 5 s before (BEFORE) and 5 s after (AFTER) the onset of interactions. This data analysis procedure was tested on some preliminary data (not used in the current report) where different timing for the time windows (3,5,7 s) were used the 3 s windows was proven to be too short and thus was excluded. The 5 and 7 s windows showed very similar patterns. However, because of the complexity of the perceptual stimulation occurring to the participants during the video presentation, we preferred among the two options to favour the shorter one, thus the 5 s window was selected. For each participant and each physiological response (heart rate, temperature on cheeks) we calculated the mean BEFORE and the mean AFTER in each condition (social vs physical, appropriate vs inappropriate). Then, for each subject in each condition, a single value for the difference AFTER-BEFORE was calculated. For each physiological measure, a general linear model (GLM) was performed with the difference AFTER-BEFORE, the scenario condition (social vs physical) and the appropriateness (appropriate vs inappropriate) as within-subjects factors and the AQ score as a continuous covariate. The covariate was centered throughout all participants [46]. The same set of analyses was repeated with the EQ score as a covariate. The same GLM was then performed with the behavioral judgments as dependent variables. Bonferroni correction was used

to analyze post hoc effects of significant factors, the regression coefficients were used to analyze the effect of the covariate on the dependent variable, and Cohen's d was used to evaluate the magnitude of significant effects.

3. Results

3.1. Heart rate

3.1.1. Autism Quotient

A main effect of Autism Quotient scores on heart rate values was found ($F(1.39) = 4.18$, $p < 0.05$, $d = 0.64$). A negative correlation was found between HR values of the AFTER-BEFORE difference and participants' AQ scores, $r = -0.24$. That is, the higher the level of autistic traits, the greater the decrease in HR values in response to the onset of any type of social interaction, regardless of the topic of the interaction itself, while the lower the level of autistic traits, the greater the increase in HR values in response to the onset of interactions [Fig. 2A].

3.1.2. Empathy Quotient

A main effect of Empathy Quotient scores on heart rate values was found ($F(1.39) = 6.25$, $p < 0.05$, $d = 0.79$). A positive correlation was found between the AFTER-BEFORE difference in heart rate values and participants' EQ scores, $r = 0.28$. That is, the higher the level of empathy traits, the greater the increase in heart rate in response to the onset of interactions, while the lower the level of empathy traits, the greater the decrease in heart rate values in response to the onset of the interactions [Fig. 2B].

3.2. Temperature

3.2.1. Autism Quotient

No effect of AQ scores on left or right cheek temperature was found (respectively: $F(1.39) = 0.47$, ns; $F(1.39) = 0.03$, ns).

3.2.2. Empathy Quotient

A significant main effect of EQ scores on left cheek temperature was found ($F(1.39) = 4.71$, $p < 0.05$, $d = 0.69$). In a post-hoc analysis, a positive correlation was found between the AFTER-BEFORE difference in left cheek temperature values and participants' EQ scores, $r = 0.24$. That is, the higher the level of empathy traits, the greater the increase in temperature in response to the onset of any type of interaction, regardless of the topic of the interaction, while the lower the level of empathy traits, the greater the decrease in temperature values in response to the onset of interactions [Fig. 3A]. Moreover a significant effect of the interaction between EQ scores and appropriateness of the norms (both social and physical) on right cheek temperature was found ($F(1.39) =$

5.90, $p < 0.05$, $d = 0.71$). Post-hoc analysis revealed a negative correlation between EQ scores and right cheek temperature in response to appropriate scenarios, while a positive correlation in response to inappropriate scenarios (respectively: $r = -0.31$; $r = 0.24$) [Fig. 3B].

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Fig. 2. Heart rate results. Correlations between HR responses (AFTER-BEFORE) and questionnaires scores. Black circles = HR responses BEFORE-AFTER. Black lines represent the linear models. In the figure the r -values represent Pearson's r . (A) Effect of AQ scores on HR responses to dyadic interactions. (B) Effect of EQ scores on HR responses to dyadic interactions. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

3.3. Behavioral judgments

Overall, participants scored as more predictable and more intelligible the appropriate scenarios compared to the inappropriate ones (respectively: $t(14) = -16.5$, $p < 0.001$; $t(14) = -4.9$, $p < 0.001$), with no difference between social-norm and physical-norm condition. However, no main effect of autistic and empathy trait levels was found on behavioral judgment scores (both on predictability and comprehensibility). Therefore, in this study behavioral judgements do not discriminate between distinct autistic and empathy traits levels.

Fig. 3. Temperature results. Correlations between cheeks temperature (AFTER-BEFORE) and EQ scores. Black circles = Temperature responses to all scenarios; black triangles = appropriate scenarios; gray triangles = inappropriate scenarios. In the figure the r -values represent Pearson's r . (A) Effect of EQ scores on left cheek temperature responses to dyadic interactions. Black line represents the linear model. (B) Effect of EQ scores on right cheek temperature responses to dyadic interactions. Black line represents the linear model for the appropriate scenarios. Gray line represents the linear model for the inappropriate scenarios. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

4. Discussion

Autism Spectrum Disorders include many traits that need to be better understood in order to elucidate the structure and mechanisms of ASDs [2]. Interestingly, autistic traits have been found to be distributed within the neurotypical population, which means there is a continuum of levels of autistic traits in the population [47]. The presence of autistic traits in the neurotypical population allows us to investigate the autistic traits without the possible confounding effect of difficulties caused by developmental impairments. Research has shown that neurotypical individuals with high levels of autistic traits, for example, display uncommon processing of social stimuli compared to physical stimuli, similar to the processing present in individuals with ASDs [10,18,19,21,27,29,48]. Physiological markers for the presence of autistic traits could allow us to enhance the reliability and the objectivity of people's autistic traits assessment. Singleton and colleagues (2015) [29] in a recent study found that higher levels of autistic traits are related to abnormal galvanic skin responses to social compared to physical static stimuli. Since ASDs lead to impairments in social interactions [2], it is also necessary to test how autistic traits affect individuals' responses to social dynamic interactions between people. In our work we aimed to investigate how levels of autistic and empathy traits influence individuals' sympathetic and parasympathetic responses to dynamic social interactions focused on social or physical events. Contrary to expectations we only found an effect of appropriateness of the interaction on right cheek temperature changes (appropriate vs inappropriate). Since no participant has a ASDs diagnosis, the lack of this expected influence

of interaction topic on physiological responses could be due to the stimuli themselves: the presence of a dynamic dyadic interaction of any type, was probably sufficiently social to elicit a physiological response in neurotypical individuals, regardless of their autistic or empathy trait levels. However, we did find an influence of autistic and empathy traits on autonomic nervous system responses to dynamic interactions in general. For the Autism Quotient scores, a negative correlation was found between AQ scores and individuals' heart rate responses. For the Empathy Quotient, a positive correlation was found between EQ scores and individuals' heart rate responses. In people with high autistic traits and low empathy traits, the decrease in heart rate values found in response to interactions most likely reflects an orienting response [38]. The orienting response is part of individuals' defensive system: the onset of possible threatening stimuli induces a sudden cardiac deceleration which allows the individual to calm down for the time needed in order to understand what is going on and what would be the best plan of action. Also, a positive correlation between EQ values and individuals' left cheek temperature was found. Opposite patterns of activations were instead found on right cheek temperatures in response to appropriate versus inappropriate scenarios corresponding to participants' EQ scores. Specifically, in response to appropriate scenarios, a negative correlation between right cheek temperature and EQ scores was found, while in response to inappropriate scenarios a positive correlation between right cheek temperature and EQ scores was found. Therefore, the interaction shows that high empathy individuals tend to have higher right cheek temperature for inappropriate scenarios as opposed to appropriate scenarios, whereas the low empathy individuals do the opposite. Since a sympathetic activation could result either in an increase or decrease of temperature depending on the kind of stimuli that the organism is facing [40,41,42,43,44], this differential pattern of activations found in response to inappropriate and appropriate scenarios show that distinct levels of empathetic

abilities at the behavioral level may have its basis in the differential processing of social stimuli at the physiological level. Specifically, high versus low empathy people may interpret dyadic interactions in different ways according to the focus of the interaction. Findings show the presence of differential patterns of autonomic nervous system activations in response to social stimuli in people with distinct levels autistic and empathy traits.

4.1. Limits

This study has some limits, which point to future improvements. In our study only the AQ was used as questionnaire for autistic traits, however there are other reliable measures, such as the Social Responsiveness Scale (SRS), which has proven to correlate with behavioral measures collected to diagnose ASDs [49,50]. Also, the measure of empathy traits could be improved, for example by applying a recently published shorter version of the Italian Empathy Quotient questionnaire, which has better psychometric properties [51]. However, this shorter version was not available at the time we ran the present study. Facial temperature was measured on the cheeks, however, blood vessels on different areas of the face are differently regulated by the

sympathetic nervous system. Therefore, to obtain a better overview of the pattern of temperature responses on the face, further studies should evaluate temperature changes in other distinct and relevant facial areas, such as nose, pre-orbital, maxillary and forehead areas, are necessary [52]. No effect of autistic or empathy traits on predictability or comprehensibility judgements was found. This result shows how an influence of the levels of autistic and empathy traits on physiological reactions can be missed when investigating only behavioral responses. Thus, assessing the presence of autistic and empathy traits only with self-reported questionnaires does not present a full picture. In future studies we will include a clinical sample and more physiological responses will be added in order to better assess individuals' responses to social interactions: (i) electromyography, as indication about individuals' promptness to action, (ii) electrodermal activity, as index of individuals' stress levels, and (iii) electroencephalography, to assess individuals' cortical responses and attention to stimuli.

4.2. Conclusions

Our findings show that autonomic nervous system responses are influenced by the levels of autistic and empathy traits. Therefore, in future studies, the assessment of peripheral physiological responses could be added to the assessment of autistic traits and, more generally, people's social abilities, in order to have a more reliable and faster measure for the presence of autistic traits and social abilities. Since ASDs individuals are within the autistic traits continuum, the presence of distinct activation patterns within the continuum of autistic traits may underlie the differential processing of social stimuli which has been found in ASDs patients compared with the neurotypical population. An enhanced understanding of the physiological mechanisms underlying social difficulties in ASDs individuals may help us in finding objective and reliable biomarkers that are useful to obtain an earlier diagnosis of Autism Spectrum Disorders. An earlier diagnosis would then allow faster and more effective ASDs treatments.

Conflict of interest

The authors declare that they have no conflict of interest.

Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

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