

# Neurophysiology of intra- and inter-species emotional interactions. Personality trait effect, P300 and N300 ERPs measures

Michela Balconi<sup>1,2</sup> - Davide Crivelli<sup>1,2</sup> - Maria Elide Vanutelli<sup>1,2,3</sup>

<sup>1</sup> Department of Psychology, Catholic University of the Sacred Heart, Milan, Italy

<sup>2</sup> Research Unit in Affective and Social Neuroscience, Catholic University of the Sacred Heart, Milan, Italy

<sup>3</sup> Department of Philosophy, University of Milan, Milan, Italy

DOI: <http://dx.doi.org/10.7358/neur-2018-023-bal2>      [michela.balconi@unicatt.it](mailto:michela.balconi@unicatt.it)

---

## ABSTRACT

*Emotional empathy plays a crucial role in social intra-species and inter-species interactions. However the role of interspecies interactions and of some personality components was underestimated. The present research explored electrophysiological correlates of affective processing in relation to emotionally valenced human-human (HH) and human-animal (HA) interactions. Further, we explored the link between such cortical responses and personality empathic profile as measured by the Balanced Emotional Empathy Scale (BEES) and the Interpersonal Reactivity Index (IRI). Both HH and HA interactions was associated to a significant increase of N300 and P300 deflections in response to positive and negative compared with neutral interactions. However, whereas N300 was mainly influenced by stimuli valence and was frontally distributed, P300 seemed to be mainly modulated by the stimuli attentional relevance and showed even a posterior distribution. Finally, a significant association was found between emotional empathy trait (BEES) and N300 amplitude. Results are discussed in light of the significance of empathic traits in mediating species-specific and species-aspecific relationships.*

*Keywords: Emotional empathy; intra/inter-species; BEES; IRI; N300/P300*

---

## 1. INTRODUCTION

Emotional responsiveness plays a fundamental role in interpersonal behaviour and comprehending the processes underlying such skill is crucial to understand why and how we engage in social interactions (Westbury & Neumann, 2008). The ability to adopt other people's perspective, monitoring and self-regulatory mechanisms that keep track of the origins of our own and others' emotions, and the ability to affectively respond to others' emotional state – which often entails the capacity to share this state (Balconi, Bortolotti, & Gonzaga, 2011; Preston & de Waal, 2002) – are basic components of the behavioural response to interpersonal situations. In this light, empathy and personality trait components seem related to the ability to understand others' emotions and feelings, with “resonance” mechanisms that mediate a direct form of understanding between the observer and the observed person (Balconi & Bortolotti, 2014; Balconi & Canavesio, 2014). An interesting approach rooted in ethology and evolutionary neuroscience was proposed by Preston and de Waal (Preston & de Waal, 2002), that is the Perception-Action Model (PAM). According to the PAM, the observation of another's emotional states automatically and unconsciously triggers corresponding neural representations in the observer. Importantly, the more similar and close two individuals are, the easier the tuning (de Waal, 2008). Additionally, the PAM outlines some other factors that could mediate emotional sharing and empathic mechanisms. In studies involving humans, several factors that can increase the perception of closeness and similarity, and consequently emotional understanding, have been investigated, with cultural similarity, sentience or social circumstance being the most influential (Westbury & Neumann, 2008). Nonetheless, while those affective mechanisms and their underlying cortical networks have been explored in the last years in human contexts (Balconi, Bortolotti, & Crivelli, 2012; Balconi & Canavesio, 2013b; Decety & Grèzes, 2006; Gallese, 2003), limited research focused on non-human social relationships (Balconi & Vanutelli, 2016). In the present study the role of different contexts in influencing affective tuning has been compared with respect to intra-species and inter-species relationships. Indeed, it is still not clear to which extent humans are able to present emotional and emphatic responses with respect to conspecifics involved in human-animal interaction scenes representing positive or negative exchanges. In fact, according to the biological significance of this kind of relationship, there is a need to improve our knowledge about the nature and the cortical correlates of human emotional behaviour in response to inter-species affective interactions where humans and animals are represented together.

Indeed, only a few studies focused on humans' emotional response to different species (Westbury & Neumann, 2008) or to humans' behaviour

observing specific situations where animals are implicated (such as pain perception) (Franklin et al., 2013), without a specific attention on the emotional significance and valence of the interactions between different species. Therefore, the significance that inter-species relationships may have in processing emotions is unexplored. Thus, with the present research, we aimed at examining the brain contribution to emotional behaviour in response to interactive situations including humans (HH) or humans and animals (HA) taking into account the specific role of cortical structures in processing different emotionally valenced species-specific and species-aspecific interactions. Such additional focus on interactions between different species may shed light on the hypothesis concerning the homogeneity of the emotional behaviour and of the emotional brain response even to inter-species interactional contexts. According to previous results, we think that the ability to respond to emotionally connoted interpersonal situations may be modulated by prefrontal functioning, since this cortical area could act as a regulator and “mediator” of emotional mechanisms (Balconi et al., 2011; Balconi & Bortolotti, 2012a, 2012b, 2012c; Balconi & Caldiroli, 2011; Balconi, Falbo, & Conte, 2012; Rameson & Lieberman, 2009). In other words, this cortical activity may have a direct influence in modulating subjects’ responsiveness to the interpersonal situations, independently from HH and HA conditions.

Secondly a direct comparison between emotion-related positive and negative patterns was considered. To explore these points, to investigate neural correlates underlying reactions to empathic-behaviours, and to study the relation between cortical responses and interpersonal context, we applied event-related potentials analysis (ERPs). Two ERP deflections were specifically analyzed, i.e. the P300 and N300 components. These deflections were deemed as specific markers of the emotional value, the relevance and the salience of a situation, as well as of the emotional involvement (arousal) of the subject (Balconi & Caldiroli, 2011). Indeed, as for the first positive deflection, Cuthbert and colleagues (Cuthbert, Schupp, Bradley, Birbaumer, & Lang, 2000) suggested that P300 can be evoked during picture viewing and augmented for emotional laden stimuli, mirroring an increase in resource allocation to motivationally relevant cues. Affective stimuli capture attention and receive increased resources, which facilitate their processing compared to neutral material, and such facilitation seems to relate to the modulation in the P300 (Rossignol, Philippot, Douilliez, Crommelinck, & Campanella, 2005). Enhanced late positive going waveforms have indeed been observed in response to the presentation of pleasant and unpleasant pictures from the International Affective Picture System (IAPS) (Cuthbert et al., 2000; Schupp et al., 2000; Schutter, De Haan, & Van Honk, 2004). It was also shown that the P300 is often modulated by the arousing content and cognitive engagement of stimuli

(Ruz, Madrid, & Tudela, 2013). Moreover several previous studies reported that P300 amplitude depends on the subjective relevance of information conveyed by the stimulus, including its emotional connotation (Amrhein, Mühlberger, Pauli, & Wiedemann, 2004; Conroy & Polich, 2007; Delplanque, Silvert, Hot, & Sequeira, 2005; Palomba, Angrilli, & Mini, 1997). Thus, for example Delplanque and colleagues (2005) reported an increase in P300 amplitude for pleasant and unpleasant novel stimuli relative to neutral novel ones, which suggests that attentional processes indexed by P300 are influenced by the affective valence and arousal features of a stimulus. Therefore we may suppose that this positive deflection subserves a specific emotion-related neural mechanism, which mirrors the salience and the attentional demand for the processed stimuli more than their specific hedonic valence.

As for the second deflection, the N300 component has been linked to the affective evaluation of stimuli (Carretié & Iglesias, 1995; Rossignol, Philippot, Douilliez, Crommelinck, & Campanella, 2005), and it is supposed to mirror the depth of emotional processing or the affective significance of the stimulus. According to Halgren and Marinkovich (Halgren & Marinkovich, 1994) the initial non-discriminatory step of the emotional reaction – the so called orienting complex (reflected in N200; Näätänen & Gaillard, 1983) – is succeeded by the ‘emotional event integration’ step, which is influenced by the emotional connotation of the stimuli. This second step is indeed associated, among other mechanisms, to the integration of the perceived emotional stimulus and the models filed in long-term memory. Moreover, in relation to stimulus valence, several studies indicate that negative events elicit more rapid and more prominent responses than neutral or positive events. This ‘negativity bias’ is manifested through different response systems, including those related to cognitive, emotional, and social behaviour (Balconi & Mazza, 2009; Cacioppo & Gardner, 1999; Carretié, Mercado, Tapia, & Hinojosa, 2001). Therefore, we expected significantly increased P300/N300 amplitudes in response to interpersonal negative and menacing interactions. Greater P300/N300 deflections were also expected for HA negative situations, since a potential increase in attentional relevance is likely induced by depicted threats.

Thirdly, the direct relation between cortical responses to emotional contexts and personality profiles and, specifically, ERPs modulations in relation to the empathic attitude shown by the participants. Empathy-related traits were measured by the BEES scale (Balance Emotional Empathy Scale; Mehrabian & Epstein, 1972). In the context of personality measurement, it describes individual differences in the tendency to show emotional empathy towards others. In addition, BEES has been found to reflect interpersonal attitudes. The Interpersonal Reactivity Index (IRI; Davis, 1980) was also administered to test the multidimensional aspects of empathic behaviour and the cognitive

component of empathy. IRI taps on four separate aspects of empathy and its relations to social functioning measures, self-esteem, emotionality, and sensitivity to others. Each of the four subscales displays a distinctive and predictable pattern of relations with those measures, as well as with previous unidimensional empathy measures. We supposed that those effects would have been accentuated in high empathic trait participants with respect to low empathic trait ones in case of both HH and HA interactions.

## 2. MATERIALS AND METHODS

### 2.1 Subjects

16 subjects, 8 females and 8 males ( $M_{age} = 26.17$ ;  $SD = 2.20$ ; range = 23-33) took part in the experiment. All subjects were right-handed, with normal or corrected-to-normal visual acuity. Exclusion criteria were neurological or psychiatric pathologies. They gave informed written consent for participating in the study and the research was approved by the ethical committee of the institution (Department of Psychology, Catholic University of Milan) where the work was carried out. The experiment was conducted in accordance with the Declaration of Helsinki and all the procedure was carried out with adequate understanding of the subjects, who read and signed the Research Consent Form before participating in this research. No payment was provided for their performance. One female participant was not included in the final set of analyses ( $N = 15$ ) because of EEG artifacts. No other participants or data were excluded from analyses and we report evidences concerning all manipulations in the study.

### 2.2 Stimuli

Subjects were presented with affective pictures depicting human-human (HH) and human-animal (HA) interactions. The stimuli set was constituted by 48 coloured realistic images representing positive, negative and neutral interaction between humans (24 pictures) and humans and animals (24 pictures). Pictures were equally divided with respect to emotional valence (positive, negative and neutral scenes). Positive pictures represented positive and cooperative interactions between HH or HA. Negative pictures represented negative and uncooperative interactions between HH and HA, Neutral pictures represented interactions without a specific valence between HH and HA (*Figure 1*). All pictures had the same size (14 cm x 10 cm) and they were similar for perceptual features such as luminance, as measured with a photometer, and complexity (i.e. number of details in the scene as assessed by six

judges in the validation phase, see ahead). No significant effects emerged with respect to species or valence. Scenes were also completely balanced with respect to actors' gender and animals' species (dogs and cats).

Stimuli were selected according to pre-experimental validation scores. Each scene was evaluated by six judges with respect to valence and arousal dimensions, using the Self-Assessment Manikin Scale with a five-point Likert Scale (Bradley & Lang, 1994, 2007). Also, they were asked to rate pictures for complexity. Ratings were averaged across all presented pictures for each condition. For what concerns SAM ratings the statistical analysis (ANOVA) showed that they differed in term of valence (more positive for positive pictures than the other pictures for HH and HA; more negative for negative pictures than the other pictures for HH and HA; with intermediate values for neutral pictures than the other pictures for HH and HA; all significant contrast comparisons:  $P \leq .05$ ) and arousal (more arousing for positive and negative pictures than neutral pictures for HH and HA;  $P \leq .05$ ). For what concerns complexity, no significant differences emerged with respect to the different conditions ( $P \geq .05$ ).



Figure 1. Some examples of emotional interaction scenes. Top: HH pictures; from left to right: positive, negative, neutral stimuli. Bottom: HA pictures; from left to right: positive, negative, neutral stimuli

### 2.3 Procedure

Subjects were seated in a dimly lit room, facing a computer monitor (distance: 70 cm). Stimuli were presented using E-Prime 2.0 software (Psychology Software Tools Inc., Sharpsburg, PA, USA) running on a personal computer with a 15-inch screen. Participants were asked to observe each stimulus during EEG measure recording, and to attend to the images for the entire time of exposure. Moreover, they were asked to try to enter into the other persons' situation (*“try to empathize with the people represented entering in his/her*

*feelings*”). In order to facilitate empathic resonance with the reproduced scene, actors were similar in age to the experimental participants. A specific questionnaire was used in order to assess the subjects’ self-rating on key aspects of subjective evaluation to the empathic task. The questionnaire was used in a post-experiment section. The aspects examined included degree of experienced empathy (“*How much did you enter into the actor’s feelings and situations?*”), personal emotional involvement (“*How much did you feel emotionally involved in the situation?*”), semantic attribution of the situation (positive, negative and neutral) (“*How do you classify the interpersonal situation?*”), emotional significance (high or low) (“*Did you perceive an emotional significance (response by the participants) of the situation?*”). All subjects experienced a high sense of empathy, they were emotionally engaged in the task and they were able to attribute a coherent emotional value to the pictures.

Pictures were randomly presented at the centre of the computer monitor for 6 seconds, with an inter-stimulus interval of 8 seconds. Each picture was presented for three times (24 stimuli for each category), for a total of 72 presentations for both the HH and the HA condition.

120 seconds resting baseline was recorded at the beginning of the experiment. After the experimental phase, subjects were asked to rate pictures hedonic valence and arousal dimensions on the SAM scale (five-points version). As shown by statistical analysis (repeated measures ANOVA with independent factor valence, 3, x condition, 2 – HH vs. HA) pictures differed in terms of valence (all paired comparisons  $P \leq .05$ ; for positive HH:  $M = 4.31$ ,  $SD = .03$ ; HA:  $M = 4.23$ ,  $SD = .04$ ; negative HH:  $M = 2.07$ ,  $SD = .02$ ; HA:  $M = 1.97$ ,  $SD = .03$ ; neutral HH:  $M = 3.08$ ,  $SD = .04$ ; HA:  $M = 3.32$ ,  $SD = .03$ ) and arousal (for the positive HH:  $M = 3.24$ ,  $SD = .02$ ; HA:  $M = 4.04$ ,  $SD = .03$ ; negative HH:  $M = 3.22$ ,  $SD = .28$ ; HA:  $M = 3.57$ ,  $SD = .29$ ; neutral HH:  $M = 2.28$ ,  $SD = .04$ ; HA:  $M = 2.38$ ,  $SD = .02$ ; with significant differences only for neutral vs. positive and neutral vs. negative scenes for both HH and HA,  $P \leq .05$ ). Finally subjects were asked to complete the BEES and IRI questionnaire (see *Figure. 2*).

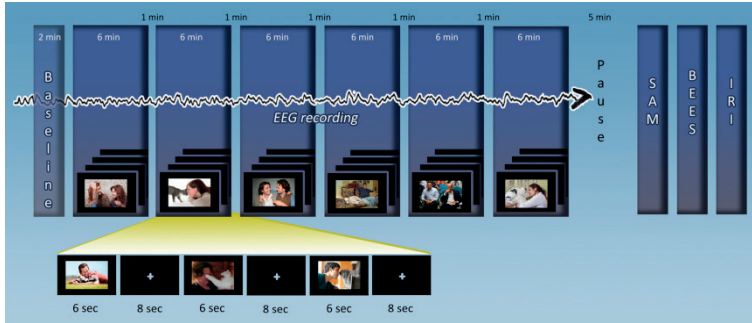


Figure 2. Experimental design and EEG recording steps

#### 2.4 BEES Measure

Trait empathy was assessed by the BEES questionnaire (Mehrabian & Epstein, 1972). It consists of 30 items, structured as Likert scales ranging from -4 (*very strong disagreement*) to +4 (*very strong agreement*). Higher scores represent higher levels of emotional empathy. The sample mean score was equal to 40.62 ( $SD = 5.98$ , range = -4 – 81). Inter-item Cronbach's alpha was calculated for BEES data (total 0.88).

#### 2.5 IRI measure

The Interpersonal Reactivity Index (IRI) is a questionnaire used to measure both cognitive and emotional components of empathy. It consists of 28 items, each including a 5-point Likert scale ranging from "It does not describe me well" to "It describes me very well". The questionnaire has 4 subscales constituted by seven different items each: *Perspective Taking*, *Fantasy*, *Empathic Concern*, *Personal Distress* (Davis, 1983). The sample mean score was equal to 61.22 ( $SD = 7.39$ , range = 58-93). We used the Italian validated version of the questionnaire (Albiero, Ingoglia, & Lo Coco, 2006).

#### 2.6 EEG recording and data reduction

A 16-channel portable EEG system (*V-Amp*, Brain Products GmbH, Gilching, Germany) was used for data acquisition. An EEG cap with Ag/AgCl electrodes was used to record EEG from active scalp sites referred to earlobes (10-5 system of electrode placement). EEG activity was recorded from the following positions: AFF3, AFF4, Fz, AFp1, AFp2, C3, C4, Cz, P3, P4, Pz, T7, T8, O1, O2. The cap was fixed with a chin strap to prevent movement during the task. Data were sampled at 1000 Hz, with a bandpass input filter set from 0.01 Hz



to 100 Hz and with a 50 Hz notch filter. The impedance of recording electrodes was monitored for each subject prior to data collection and it was always kept below 5 k $\Omega$ . Additionally, EOG electrodes were sited on the outer canthi to detect blinks and eye movements for subsequent ocular artifacts correction. After offline filtering through a 0.1-30 Hz bandpass filter, data were corrected by an ICA-based algorithm (Jung et al., 2000) and a computerized artifact rejection criterion excluded trials when the peak-to-peak amplitude exceeded 40  $\mu$ V. The signal was then visually checked to reject all residual ocular, muscular, or movement artifacts and to increase specificity. After performing EOG correction and visual inspection, only artifact-free trials were considered (rejected epochs, 3%). An averaged waveform (off-line) was obtained for each condition (not less than 20 epochs per condition entered the computation). The peak amplitude (from the baseline) was quantified relative to the 150 ms pre-stimulus window by identifying the most negative and positive peaks within the temporal window from 230 to 400 ms post-stimulus. The onset was coincident with the appearance of the stimulus on the screen. Morphological analysis of the EEG profile confirmed that the peak deflections were included within those time ranges. Peak amplitude and latency data were extracted respectively for P300 and N300 components, and distinct analyses were applied to each of the average profiles. Subsequently, localization (three sites: frontal, AFF3/AFF4 and AFp1/AFp2; temporo-central, C3/C4 and T7/T8 and parietal, P3/P4) and lateralization (two sides: left and right electrode positions) factors were explored by focused statistical analyses. Preliminary tests did not highlight significant latency differences across conditions. We then did not include this measure within the final set of analyses. The mean latency of the two components was approximately 360 ms and 300 ms.

### 3. RESULTS

A set of analyses was performed on the data. A first set of ANOVAs was applied to the peak amplitude of N300 and P300 ERP. A second set of correlation analyses was applied to BEES and IRI measures and N300/P300 peak amplitude.

#### *3.1 ANOVA*

ERPs data were entered into four-ways repeated measure ANOVAs. Condition (2, HH and HA), lateralization (2, left and right electrode sites), valence (3, positive, negative, neutral), and localization (3, frontal, temporo-central, and parietal sites) were included as factors of the statistical models applied to the peak

amplitude variable, distinctly for N300 and P300 components. Type I errors associated with inhomogeneity of variance were controlled by decreasing the degrees of freedom using the Greenhouse-Geisser epsilon. Bonferroni corrections for multiple comparisons were applied. Post-hoc comparisons were successively applied to the data (contrast analyses). The normal distribution of the data was assessed by using skewness and kurtosis test in a preliminary statistical phase.

### 3.2 N300

As shown by ANOVA, the peak amplitude was modulated by valence ( $F(2,14) = 11.50, P = 0.001, \eta^2 = .44$ ), and valence x localization ( $F(4,27) = 9.03, P = 0.001, \eta^2 = .40$ ). No other main or interaction effect was statistically significant. Post-hoc comparison revealed an increased peak amplitude for positive ( $F(1,14) = 8.13, P = 0.001, \eta^2 = .38$ ) and negative ( $F(1,14) = 7.89, P = 0.001, \eta^2 = .34$ ) in comparison with neutral conditions. A significant difference was also found for negative (higher N300) compared to positive ( $F(1,14) = 6.58, P = 0.001, \eta^2 = .32$ ) conditions. Moreover, the frontal area involving AFF3, AFF4, AFp1 and AFp2 showed a higher peak amplitude for positive and negative conditions compared to other cortical sites: respectively compared to the temporo-central (for positive  $F(1,14) = 8.54, P = 0.001, \eta^2 = .40$ ; for negative  $F(1,14) = 9.32, P = 0.001, \eta^2 = .42$ ); and to parietal (for positive  $F(1,14) = 8.23, P = 0.001, \eta^2 = .40$ ; for negative  $F(1,14) = 8.04, P = 0.001, \eta^2 = .39$ ) sites.

### 3.3 P300

ANOVA showed significant effects for valence x localization ( $F(4,27) = 11.52, P = 0.001, \eta^2 = .45$ ) and condition x valence x localization ( $F(4,53) = 10.03, P = 0.001, \eta^2 = .43$ ). No other main or interaction effect was statistically significant. Indeed, firstly positive and negative conditions revealed an increased P300 amplitude than neutral conditions more localized on frontal (for positive  $F(1,14) = 8.16, P = 0.001, \eta^2 = .39$ ; for negative  $F(1,14) = 7.78, P = 0.001, \eta^2 = .37$ ) and parietal (for positive  $F(1,14) = 7.96, P = 0.001, \eta^2 = .37$ ; for negative  $F(1,14) = 9.13, P = 0.001, \eta^2 = .41$ ) sites. A significant difference was also found between HA and HH condition, with an increased P300 peak amplitude in HA for negative than positive and neutral conditions within the frontal areas over AFF3, AFF4, AFp1 and AFp2 (comparison negative vs. positive  $F(1,14) = 6.73, P = 0.001, \eta^2 = .34$ ; comparison negative vs. neutral  $F(1,14) = 6.98, P = 0.001, \eta^2 = .36$ ) and parietal areas over P3 and P4 (comparison negative vs. positive  $F(1,14) = 7.03, P = 0.001, \eta^2 = .37$ ; comparison negative vs. neutral  $F(1,14) = 8.15, P = 0.001, \eta^2 = .39$ ).

### 3.4 Correlation analyses

Pearson correlation values were applied to ERPs amplitude (N300 and P300) and BEES for HH and HA in response to positive, negative and neutral conditions. A second set of correlations was applied to ERPs and IRI distinctly for HH and HA and for positive, negative and neutral conditions.

For the first set (BEES measure), as shown by statistical results, BEES scores were significantly and positively correlated with N300 amplitude in response to negative conditions respectively for HH ( $r = 0.525$ ,  $P < .001$ ) and HA ( $r = 0.607$ ,  $P < .001$ ). Contrarily, no significant effect was found for P300. As for the second set of analyses (IRI sub-scales measures), no statistically significant correlations were found for both N300 and P300.

## 4. DISCUSSION

Empathic sensitivity to different positive vs. negative emotional situations in species-specific (HH) and species-aspecific (HA) relationships was investigated in the present research. This sensitivity was tested by using psychophysiological measures (N300 and P300 ERPs) in high-empathic and low-empathic subjects, according to BEES and IRI. Results confirmed the significance of emotional empathic behaviours in response to interpersonal situations in both HH and HA with respect to valence, while the later P300 component more highlighted some specificities with respect to HH and HA conditions. The direct link between these different levels of analysis (i.e. ERP measures and empathic personality traits) was discussed.

The present findings firstly supported the hypothesis that there is a relation between positive (cooperative) vs. negative (not cooperative) situations and brain response in both HH and HA interaction types. That is, different conditions evoked distinct ERP response patterns, with increased N300 activity over anterior frontal and frontopolar electrodes to positive and mainly negative scenes as compared to neutral ones in both HH and HA interactions. The P300 component, instead, was greater over both anterior frontal and frontopolar, but also parietal sites (P3, P4) and showed a more specific modulation in response to HA scenes exclusively for negative more than positive and neutral conditions.

For what concerns N300 deflection, the valence effect was significant mainly with respect to negative stimuli. Moreover, there was no distinction between HH and HA in these reactions. This component has been linked to the affective charge of visual stimuli (Carretié & Iglesias, 1995; Rossignol et al., 2005), and it is supposed to mirror the depth of emotional processing or the

affective significance of the stimulus. Moreover, in relation to stimulus valence, several studies indicate that negative events elicit more rapid and more prominent responses than neutral or positive events. This phenomenon has been called ‘negativity bias’ (Balconi & Mazza, 2009; Cacioppo & Gardner, 1999; Carretié et al., 2001). Moreover, the N300 in the anterior brain region (prefrontal cortex) has been positively associated with negative valenced stimuli. In accordance with its frontal localization, the N300 component might be involved in the detection and evaluation of relevant and threatening patterns (Posner & Raichle, 1997). These results also suggest that the prefrontal cortex is involved in broad regulatory control mechanisms, and again in “bringing” emotional representations online, in maintaining, and in regulating them (Balconi & Canavesio, 2013b; Ochsner et al., 2004). Also, the absence of relevant differences between HH and HA suggests that this emotional modulation was not species-specific.

In contrast P300 was presumably related to attentive behaviour we adopt to regulate our response to salient, emotional cues. This salient and potentially dangerous situation may have determined a higher-level attention demand: the perception of an “uncontrolled” situation in the case of an animal aggression may have plausibly triggered the interactions as highly salient and aversive. The cortical localization of this ERP (anterior and parietal) may confirm this attentional hypothesis, consistently with previous research that have demonstrated that P300 amplitude may mirror increases in resource allocation to motivationally relevant incentives (for a recent review see van Dinteren, Arns, Jongma, & Kessels, 2014). Thus, for example Delplanque and colleagues (2005) reported an increase in P300 amplitude for pleasant and unpleasant novel stimuli relative to neutral novel ones, which suggests that attentional processes indexed by P300 are influenced by the affective valence and arousal features of a stimulus. Therefore we may suppose that this positive deflection subserves a specific emotion-related neural mechanism, which mirrors the salience and the attentional demand for the processed stimuli more than their specific hedonic valence. More generally, an emotion-vigilance network of attention consisting in frontal and parietal cortices is argued to maintain a state of alertness when salient stimuli are encountered in high relevant (empathic) situations. In fact, aggressive and negative interactions are important socially aversive and potentially threatening conditions (Balconi & Bortolotti, 2012a, 2012b; Ohman, Flykt, & Esteves, 2001), hence the involvement of the vigilance network in response to such situations can be plausibly assumed, in particular since alertness and action preparation are essential for appropriate responses.

Within this network, the N300 amplitude appeared to be significantly modulated over the anterior brain areas and to be valence-sensitive (Carretié, Iglesias, & García, 1997). In contrast, the P300 could be more directly modulated by the level of alertness independently from valence, on the basis of a

mechanism for attention resource allocation. The cooperative – but even more the uncooperative and conflictual human-human and human-animal interactions – are highly relevant. Both of them are attentionally critical (as revealed by higher P300 for both HH and HA) but the negative situations seemed to be able to elicit an increased emotional response when subjects observed aggressive HA interactions. This may be due to the potential lack of control on the situation which the subject has to face: in case of HA scenes, the unpredictable outcomes of aggressive inter-species interactions may be evaluated as more dangerous than those of intra-species ones.

In addition, the role of the empathic profile was found to be relevant, as confirmed mainly by BEES measure. In fact the emotional response mirrored by the N300 deflection was greater in high- with respect to low-empathy individuals. Thus, this pattern of responses confirms the direct link between trait empathy and brain responses. However this trait-modulation was not generic but context- and valence-related. Indeed this result was obtained only in response to negative situations in the case of both HH and HA interaction scenes, and highly empathic and lowly empathic participants did not differ in terms of “emotional resonance” in positive or neutral situations. That is, the aggressive situations elicited a consistent and intense response in highly empathic subjects when a human is threatened independently from the eliciting context (HH and HA).

In contrast, no significant association was found between trait empathy and P300 ERP deflection. The reason why the P300 modulation was not directly related to empathic trait is presumably linked to the functional (cognitive and attentional) significance of this ERP deflection, without a specific relation with the emotional valence per se, but to the relevance of the situations. Moreover, the absence of significant correlations between ERPs and IRI measures may be explained by considering the limited sensitivity of this measure in respect to the emotional content of the empathic situations as compared to the BEES. IRI indeed more specifically tests “cognitive” empathic components and includes heterogeneous elements, such as cognitive perspective-taking or personal distress. Future research should better explore this issue, for example by directly comparing and investigating the relative role of different components of the empathy construct (e.g. emotional or cognitive ones).

To summarize, our results highlight the potential of using psychophysiological and personality indices to measure emotional empathy in contexts with different biological and evolutionary meanings, involving species-specific and species-aspecific relationships (Balconi & Canavesio, 2013a, 2013b; Carlo, Hausmann, Christiansen, & Randall, 2003; Chiu Loke, Evans, & Lee, 2011). Moreover, brain activity was analysed to investigate markers of affective processes supporting emotional “resonance mechanisms” that are activated when we face an emotionally-connoted situation (Balconi & Bortolotti, 2012a). These

mechanisms may be plausibly considered as relatively independent from the actors involved in an interaction (i.e. would they be only humans or humans and animals), as they may be considered phylogenetically and evolutionary acquired to respond to emotional situations where at least a consimilar is implicated. Nonetheless, P300 component proved to be sensitive to the species condition, since it signalled the relevance of HA interactions when a negative context was considered. According to the role of this kind of stimuli in eliciting uncontrolled, primitive and orienting responses, significant higher responses emerged in the case of dog/cat attacks toward humans.

Finally, psychophysiological indices differed depending on individual empathy traits. High-empathy participants, as compared to their low-empathy counterparts, exhibited greater brain responsiveness to emotional situations for both species-specific and species-aspecific interactions.

However, an ampler sample should be used to make the main effects we found more robust. As for ERPs significance, the roles of the N300 component and of a possible anticipated N250 deflection should be distinguished. Indeed species-specific and species-aspecific relationships require an emotional component and recent observations underlined that N250 amplitude might mirror the increased effort in attributing emotions to inanimate objects. Due to the latency feature in the present paper we may not totally exclude the possibility that the observed negative deflection could be a later N250 phenomenon. However the cortical localization and the latency values (later than 300 msec) could suggest a later than early deflection. Finally, the familiarity with animal interactions may be taken in consideration to better elucidate the effect that previous relationships with other species has on the emotional response to inter-species contexts.

## REFERENCES

- Albiero, P., Ingolia, S., & Lo Coco, A. (2006). Contributo all'adattamento italiano dell'Interpersonal Reactivity Index. *Testing Psicometria Metodologia*, 13(2), 107-125.
- Amrhein, C., Mühlberger, A., Pauli, P., & Wiedemann, G. (2004). Modulation of event-related brain potentials during affective picture processing: A complement to startle reflex and skin conductance response? *International Journal of Psychophysiology*, 54(3), 231-240.
- Balconi, M., & Bortolotti, A. (2012a). Detection of the facial expression of emotion and self-report measures in empathic situations are influenced by

- sensorimotor circuit inhibition by low-frequency rTMS. *Brain Stimulation*, 5(3), 330-336.
- Balconi, M., & Bortolotti, A. (2012b). Empathy in cooperative versus non-cooperative situations: The contribution of self-report measures and autonomic responses. *Applied Psychophysiology Biofeedback*, 37(3), 161-169.
- Balconi, M., & Bortolotti, A. (2012c). Resonance mechanism in empathic behavior. BEES, BIS/BAS and psychophysiological contribution. *Physiology and Behavior*, 105(2), 298-304.
- Balconi, M., & Bortolotti, A. (2014). Self-report, personality and autonomic system modulation in response to empathic conflictual versus non conflictual situation. *Cognition & Emotion*, 28(1), 153-62.
- Balconi, M., Bortolotti, A., & Crivelli, D. (2012). Self-report measures, facial feedback, and personality differences (BEES) in cooperative vs. noncooperative situations: Contribution of the mimic system to the sense of empathy. *International Journal of Psychology*, 48(4), 631-640.
- Balconi, M., Bortolotti, A., & Gonzaga, L. (2011). Emotional face recognition, EMG response, and medial prefrontal activity in empathic behaviour. *Neuroscience Research*, 71(3), 251-259.
- Balconi, M., & Canavesio, Y. (2013a). High-frequency rTMS improves facial mimicry and detection responses in an empathic emotional task. *Neuroscience*, 236, 12-20.
- Balconi, M., & Canavesio, Y. (2013b). Prosocial attitudes and empathic behavior in emotional positive versus negative situations: brain response (ERPs) and source localization (LORETA) analysis. *Cognitive Processing*, 14(1), 63-72.
- Balconi, M., & Canavesio, Y. (2014). High-frequency rTMS on DLPFC increases prosocial attitude in case of decision to support people. *Social Neuroscience*, 9(1), 82-93.
- Balconi, M., & Caldiroli, C. (2011). Semantic violation effect on object-related action comprehension. N400-like event-related potentials for unusual and incorrect use. *Neuroscience*, 197, 191-199.
- Balconi, M., Falbo, L., & Conte, V. A. (2012). BIS and BAS correlates with psychophysiological and cortical response systems during aversive and appetitive emotional stimuli processing. *Motivation and Emotion*, 36(2), 218-231.
- Balconi, M., & Mazza, G. (2009). Brain oscillations and BIS/BAS (behavioral inhibition/activation system) effects on processing masked emotional cues. ERS/ERD and coherence measures of alpha band. *International Journal of Psychophysiology*, 74(2), 158-165.

- Balconi, M., & Vanutelli, M.E (2016). Hemodynamic (fNIRS) and EEG (N200) correlates of emotional inter-species interactions modulated by visual and auditory stimulation. *Scientific Reports*, 6, 23086.
- Bradley, M. M., & Lang, P. J. (1994). Measuring emotion: the Self-Assessment Manikin and the Semantic Differential. *Journal of Behavior Therapy and Experimental Psychiatry*, 25(1), 49–59.
- Bradley, M. M., & Lang, P. J. (2007). The International Affective Picture System (IAPS) in the study of emotion and attention. In J. A. Coan & J. J. B. Allen (Eds.), *Handbook of emotion elicitation and assessment* (pp. 39–46). New York: Oxford Univ Press.
- Cacioppo, J. T., & Gardner, W. L. (1999). Emotion. *Annual Review of Psychology*, 50, 191–214.
- Carlo, G., Hausmann, A., Christiansen, S., & Randall, B. A. (2003). Sociocognitive and Behavioral Correlates of a Measure of Prosocial Tendencies for Adolescents. *Journal of Early Adolescence*, 23(1), 107–134.
- Carretié, L., & Iglesias, J. (1995). An ERP study on the specificity of facial expression processing. *International Journal of Psychophysiology*, 19(3), 183–192.
- Carretié, L., Iglesias, J., & García, T. (1997). A study on the emotional-processing of visual stimuli through event-related potentials. *Brain and Cognition*, 34(2), 207–217.
- Carretié, L., Mercado, F., Tapia, M., & Hinojosa, J. A. (2001). Emotion, attention, and the “negativity bias”, studied through event-related potentials. *International Journal of Psychophysiology*, 41(1), 75–85.
- Chiu Loke, I., Evans, A. D., & Lee, K. (2011). The neural correlates of reasoning about prosocial-helping decisions: An event-related brain potentials study. *Brain Research*, 1369, 140–148.
- Conroy, M. A., & Polich, J. (2007). Affective valence and P300 when stimulus arousal level is controlled. *Cognition & Emotion*, 21(4), 891–901.
- Cuthbert, B. N., Schupp, H. T., Bradley, M. M., Birbaumer, N., & Lang, P. J. (2000). Brain potentials in affective picture processing: Covariation with autonomic arousal and affective report. *Biological Psychology*, 52(2), 95–111.
- Davis, M. H. (1980). A multidimensional approach to individual differences in empathy. *JSAS Catalog of Selected Documents in Psychology*, 10, 85.
- Davis, M. H. (1983). Measuring individual differences in empathy: Evidence for a multidimensional approach. *Journal of Personality and Social Psychology*, 44(1), 113–126.



- de Waal, F. B. M. (2008). Putting the altruism back into altruism: the evolution of empathy. *Annual Review of Psychology*, *59*, 279–300.
- Decety, J., & Grèzes, J. (2006). The power of simulation: Imagining one's own and other's behavior. *Brain Research*, *1079*(1), 4–14.
- Delplanque, S., Silvert, L., Hot, P., & Sequeira, H. (2005). Event-related P3a and P3b in response to unpredictable emotional stimuli. *Biological Psychology*, *68*(2), 107–120.
- Franklin, R. G., Nelson, A. J., Baker, M., Beeney, J. E., Vescio, T. K., Lenz-Watson, A., & Adams, R. B. (2013). Neural responses to perceiving suffering in humans and animals. *Social Neuroscience*, *8*(3), 217–27.
- Gallese, V. (2003). The Roots of Empathy: The Shared Manifold Hypothesis and the Neural Basis of Intersubjectivity. *Psychopathology*, *36*(4), 171–180.
- Halgren, E., & Marinkovich, K. (1994). Neurophysiological networks integrating human emotions. In M. S. Gazzaniga (Ed.), *The Cognitive Neurosciences*. London: MIT Press.
- Jung, T.-P., Makeig, S., Humphries, C., Lee, T.-W., McKeown, M. J., Iragui, V., & Sejnowski, T. J. (2000). Removing electroencephalographic artifacts by blind source separation. *Psychophysiology*, *37*(2), 163–178. Retrieved from <https://www.cambridge.org/core/article/removing-electroencephalographic-artifacts-by-blind-source-separation/2548D35629CAE17E6956C2FFF1B6C8AB>
- Mehrabian, A., & Epstein, N. (1972). A measure of emotional empathy. *Journal of Personality*, *40*(4), 525–543.
- Näätänen, R., & Gaillard, A. W. K. (1983). The orienting reflex and the N2 deflection of the event-related potential (ERP). In A. W. K. Gaillard & W. Ritter (Eds.), *Tutorials in ERP Research: Endogenous Components*. Amsterdam: North-Holland.
- Ochsner, K. N., Ray, R. D., Cooper, J. C., Robertson, E. R., Chopra, S., Gabrieli, J. D. E., & Gross, J. J. (2004). For better or for worse: neural systems supporting the cognitive down- and up-regulation of negative emotion. *NeuroImage*, *23*(2), 483–499.
- Ohman, A., Flykt, A., & Esteves, F. (2001). Emotion drives attention: detecting the snake in the grass. *Journal of Experimental Psychology. General.*, *130*(3), 466–478.
- Palomba, D., Angrilli, A., & Mini, A. (1997). Visual evoked potentials, heart rate responses and memory to emotional pictorial stimuli. *International Journal of Psychophysiology: Official Journal of the International Organization of Psychophysiology*, *27*(1), 55–67.

- Posner, M. I., & Raichle, M. E. (1997). *Images of Mind*. New York: Scientific Medical Library.
- Preston, S. D., & de Waal, F. B. M. (2002). Empathy: its ultimate and proximate bases. *The Behavioral and Brain Sciences*, 25(1), 1–20.
- Rameson, L. T., & Lieberman, M. D. (2009). Empathy: a social cognitive neuroscience approach. *Social and Personality Psychology Compass*, 3(1), 94–110.
- Rossignol, M., Philippot, P., Douilliez, C., Crommelinck, M., & Campanella, S. (2005). The perception of fearful and happy facial expression is modulated by anxiety: An event-related potential study. *Neuroscience Letters*, 377(2), 115–120.
- Ruz, M., Madrid, E., & Tudela, P. (2013). Interactions between perceived emotions and executive attention in an interpersonal game. *Social Cognitive and Affective Neuroscience*, 8(7), 838–844.
- Schupp, H. T., Cuthbert, B. N., Bradley, M. M., Cacioppo, J. T., Ito, T., & Lang, P. J. (2000). Affective picture processing: the late positive potential is modulated by motivational relevance. *Psychophysiology*, 37(2), 257–261.
- Schutter, D. J. L. G., De Haan, E. H. F., & Van Honk, J. (2004). Functionally dissociated aspects in anterior and posterior electrocortical processing of facial threat. *International Journal of Psychophysiology*, 53(1), 29–36.
- van Dinteren, R., Arns, M., Jongasma, M. L. A., & Kessels, R. P. C. (2014). Combined frontal and parietal P300 amplitudes indicate compensated cognitive processing across the lifespan. *Frontiers in Aging Neuroscience*, 6(October), 1–9.
- Westbury, H. R., & Neumann, D. L. (2008). Empathy-related responses to moving film stimuli depicting human and non-human animal targets in negative circumstances. *Biological Psychology*, 78(1), 66–74.