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Title: Three-year-olds' rapid facial EMG responses to emotional facial expressions and body postures

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Abstract: Rapid Facial Reactions (RFRs) to observed emotional expressions are proposed to be involved in a wide array of socioemotional skills, from empathy to social communication. Two of the most persuasive theoretical accounts propose RFRs to rely either on motor resonance mechanisms or on more complex mechanisms involving affective processes. Previous studies demonstrated that presentation of facial and bodily expressions can generate rapid changes in adult and school age children's muscle activity. However, up to date, there is little to no evidence to suggest the existence of emotional RFRs from infancy to preschool age. To investigate whether RFRs are driven by motor mimicry or could also be a result of emotional appraisal processes, we recorded facial electromyographic (EMG) activation from the zygomaticus major and frontalis medialis muscles to presentation of static facial and bodily expressions of emotions (i.e, happiness, anger, fear and neutral) in 3-years old children. Results showed no specific EMG activation in response to bodily emotion expressions. However, observing others' happy faces lead to the increased activation of the zygomaticus major and decreased activation of the frontalis medialis, while observing angry faces elicited the opposite pattern of activation. This study suggests that RFRs are the result of complex mechanisms in which both affective processes and motor resonance may play an important role.

1) Cover Letter

Dear Prof. Bjorklund,

Please find attached a revised version of the manuscript JECP-D-15-00298, “Three-year-olds' rapid facial EMG responses to emotional facial expressions and body postures”, which was submitted to the Journal of Experimental Child Psychology.

We would like to thank you and the reviewers for the constructive comments and suggestions, which were very helpful in revising the manuscript. We have carefully analysed and responded to all the points that were raised. Below you will find a detailed response to each of these issues, together with indications of how the document has been revised to address them.

The revised manuscript has also been checked for formatting, writing style and manuscript content, as per journal’s suggestions.

We hope that in its current form, the manuscript is suitable for publication in the Journal of Experimental Child Psychology.

Looking forward to hearing back your decision,

Elena Geangu and colleagues.

***1a) Detailed Response to Reviewers**

We wish to thank to the editor and to both reviewers for their helpful comments in revising our manuscript. We addressed each of the raised issues as described below.

<u>**Reviewer #1:**</u>

1. Concerns to body posture stimuli, do the participants recognize the emotion of those stimuli? The authors referred the previous studies, but how do the authors certify that the participants understood the each emotion of postures in the present study.

We would like to thank the reviewer for raising this point. Indeed, as we mentioned in the original manuscript, studies from different laboratories using both behavioural (Zieber et al., 2014) and EEG measures (Missana et al. 2014) now converge to suggest that static emotional body postures are discriminated by about 6-8 months of age. Nonetheless, we agree with the reviewer that assessing participants' affect knowledge with regards to the emotional body postures would be a useful step forward in investigating the factors contributing to children's rapid facial responses to others' emotions. We have now revised the manuscript to explicitly present this information and emphasize this idea (see the Discussion, paragraph 7). Part of Paragraph 7 now reads:

"While 3-year-old children are able to correctly associate an emotional facial expression of a person with the events most likely causing the associated affective state, they fail to do so for emotional body postures. This may be due to the difference in emotional information that the body postures communicate (Ekman, 1965). The ability to interpret such information may develop at a different pace than faces, potentially explaining the lack of emotionally specific RFRs to emotionally body postures in 3-year-old children. In our current study we did not include any measure of affect knowledge to assess whether 3-year-old children discriminate, label, and understand the meaning of different means of emotional expressivity. Further studies in which other emotional expression modalities than those included in this study are used (i.e., emotional prosody) together with measures of affect knowledge could help us understand whether the lack of selective RFRs for emotional expressions other than faces reflect the presence of perception-action mechanisms, affective processes or a combination of both."

2. And I wondered are there any individual differences of the results of EMG.

This is an interesting point. The descriptive statistics presented in Table 1 and in the Figures (2 from the original manuscript and 3 from the revised version) suggest a certain degree of variability in the EMG responses. This could be due to individual differences. So far, none of the existent studies we found in the literature explored individual differences in the facial EMG of typically developing populations. However, it would be interesting for future studies to explore whether these are related to temperamental characteristics and/or differences in emotional discrimination abilities. We have revised the Discussion section to include these suggestions. The end of paragraph 3 now reads:

"Further investigations in which measures of emotional arousal (e.g., heart rate, pupil dilation, galvanic skin response) are recorded simultaneously with facial EMG from all three muscles, could help elucidate whether the 3-year-olds' RFRs to others' emotional facial expressions are associated with a change in the affective state. This association may also depend on the extent to which different children respond emotionally to socioemotional events and the efficiency with which they regulate their emotions, since the temperamental characteristics recorded during the first years of life largely explain the variability in empathy development (van der Mark, van IJzendoorn, & Bakermans-Kranenburg, 2002; Young, Fox, Zahn-Waxler, 1999)."

3. Are there any correlation between EMG for the body postures and those for facial expressions?

We thank the reviewer for this suggestion of analysing the EMG data in our study. The analysis of the muscles predicted to show selective activation did not reveal any significant relations between the RFRs to facial and bodily expressions. More specifically, the average amplitude of the zygomaticus major when observing happy faces was not significantly related to the average amplitude recorded when observing happy bodies (r = .016, p = .943). Also, the average amplitude of the frontalis medialis recorded in response to angry faces did not significantly relate to the same muscle activation for angry body postures (r = .223, p = .318). In the same manner, the activation of the frontalis medialis in response to fearful faces and in response to bodies were not related (r = .392, p = .071). Neither the amplitude of the zygomaticus major (r = -.005, p = .981) nor the amplitude of the frontalis medialis (r = -.045, r = .844) in response to neutral faces correlated to those in response to emotionally neutral body postures. These results are in line with those of the statistical analysis already included in the initial manuscript, which shows different patterns of results for EMG responses to emotional faces and bodies. In light of this, we considered it would be perhaps unnecessary to include the correlational analysis in the revised manuscript. However, if the Reviewers and the Editor consider it would bring important information to the results we present, we can include it.

4. The authors should put into the graph of EMG for body postures even there is no significant result.

Thank you for this suggestion. The revised manuscript now includes Figure 3 illustrating the EMG recorded in response to body postures.

<u>**Reviewer #2:**</u>

1. The analyses that are provided clearly indicate that observation of facial happiness and anger activated different facial muscles in children. However, what is not provided are any analyses comparing these values to neutral, or baseline performance. Although the zygomaticus effects would probably be significant in such a comparison, the frontalis may not. I don't think this would reduce the quality of the work - the predicted effects are still present in direct comparisons - but I do think it's important to present such findings and to discuss them. I could see this as possibly adding to the already rich discussion of potential cognitive processes that mediate emotion recognition that are still under development in children. Perhaps recognition of happiness is consistent across kids, or more primary, and recognition of anger is more variable across kids, or can elicit multiple emotions... perhaps, sometimes, even anger itself (as that couldn't be measured accurately in the present experiment). I'm out of my element here for relevant discussion, but I think the differences from baseline should be analyzed and discussed.

We thank the Reviewer for suggesting this further analysis. We have now compared the zygomaticus major and the frontalis medialis mean activation during the 800-1300ms post-stimulus with the mean activation during the pre-stimulus baseline (when a fixation cross was displayed) as a follow-up analysis for the significant interactions between Muscle and Emotion for the Face Stimuli. For this purpose we used paired t-tests at .05 level of significance (two-tailed). As the Reviewer anticipated, these revealed interesting results, which converge with the findings we already included in the original manuscript. We revised the Results section to include these new results:

"Face stimuli
When compared to the baseline, observing happy facial expressions elicited an increased activation of the zygomaticus major (t(21) = 2.392, p = .026), while the angry faces led to a decrease in the activation of the same muscle (t(21) = -2.501, p = .021). In contrast, observing happy faces led to a

decrease in the activity of the frontalis muscle from the baseline levels (t(21) = -2.688, p = .014), while the same muscle tended to show an increased activation in response to angry faces when compared to the baseline, although it was marginally significant (t(21) = 1.947, p = .066). No other significant differences emerged."

We also revised the Discussion section to reflect these findings, see paragraphs 2 and 4.

Other minor things:

1. At the end of the first paragraph, the term "rapid facial responses" is first used, and is not defined. Use it earlier in the paragraph and define it then.

We have now revised the Introduction to clarify what we mean by 'rapid facial responses' (see paragraph 1)
"The covert responses can themselves vary from being extended to long periods activity to being very rapid and subtle, also called rapid facial responses (RFR)."

2. I found paragraphs in the paper very long and blocky, and hard to track the main points. Break these up into smaller digestible bits.

We have revised the manuscript trying to break down the paragraphs into smaller ones. We hope this now facilitates the reading and comprehension of the text.

3. For Western publication, the y axis on the graph should use a period to denote the decimal, instead of the comma.

We have revised both graphs to use a period for denoting the decimal.

4. Typos:
We have corrected the typos throughout the manuscript as recommended.

*2) Title Page (WITH Author Details)

Three-year-olds' rapid facial EMG responses to emotional facial expressions and body postures

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Three-year-olds' rapid facial EMG responses to emotional facial expressions and body postures

Word count: text = 6246 words; references = 2685

Seeing the emotional expressions of the people we interact with elicits most often similar expressions in us as observers. One of the most common examples is when we smile in response to seeing other people smile. Our responses can vary from being overt, observable with the naked eye, to being covert and only detectable by using specific electrophysiological measurements (i.e., electromyography – EMG) of the muscles involved in generating these expressions. The covert responses can themselves vary from being extended to long periods of activity to being very rapid and subtle, also called rapid facial responses (RFRs). Forms of emotional expression congruency can be recorded in humans from the first months of infancy (e.g., Haviland & Lelwica, 1987), throughout childhood (e.g., Beall et al., 2008; Deschamps et al., 2013; de Wied et al., 2006; Oberman, Winkielman, & Ramachandran, 2009) and adulthood (e.g., Bavelas et al., 1986; Hess & Blairy, 2001; Magnee et al., 2007b), and have been documented for facial, vocal, and postural modes of emotional expressivity (Hatfield & Cacioppo, 1994). Importantly, these expressivity matching responses have been attributed essential socio-emotional functions, with relevance for emotional contagion (Hatfield & Cacioppo, 1994), empathy (Decety & Jackson, 2004; de Vignemont & Singer, 2006), social communication (Hess & Burgeois, 2010), as well as social coordination through affiliation (Lakin & Chartrand, 2003), to name just a few. Despite a large body of research investigating the mechanisms underlying the variety of these abilities and their functions in adults, we still have limited knowledge about their development (Beall et al., 2008; Jones, 2007). The current paper aims to address this limitation by investigating the development of rapid facial responses (RFRs) to others' emotions in 3-year-old children.

Two main theoretical assumptions have been put forward with regards to the mechanisms underlying the RFRs. On one hand side, several researchers regard the RFRs as being simple motor responses, triggered by observing others' facial expression, without any direct affective underpinnings, usually coined as mimicry (Bavelas et al., 1986; Chartrand & Bargh, 1999; Hoffman, 1984; Meltzoff & Moore, 1977). Mimicking others' emotional displays is presumed to rely on perception-action matching mechanisms, whereby perceiving the pattern of motor behaviour specific for expressing different emotions activates in the observer the same motor response (Lipps, 1907; Hatfield & Cacioppo, 1994; Meltzoff, 2007; de Waal, 2009). At the neural level, the mirror neuron system is hypothesised to be involved in eliciting these motor resonance responses (Carr et al., 2003). Analogous to the neurons first described in the ventral premotor cortex and the inferior parietal lobule of the macaque brain (Ferrari et al., 2003; Gallese et al., 1996; Umiltà et al., 2001), the human mirror neuron system (including the pars opercularis of the inferior frontal gyrus, the ventral premotor cortex, and the anterior inferior parietal lobule) has been found responsive both when adults perform and observe simple goal-directed motor acts (e.g.; Buccino et al., 2001, 2004; Iacoboni et al., 1999; Iacoboni & Dapretto, 2006; Rizzolatti & Craighero; 2004), including emotional facial expressions (Pfeifer et al., 2008; Lee et al., 2006, 2008). According to this theoretical account, once elicited, the RFRs can lead to a change in the affective state of the observer through associations with previously experienced emotions, generating emotional contagion (Cappella, 1993; Hoffman, 1984; Laird et al., 1994; Lipps, 1907).

In support of this view, it has been shown that adults' vocal (Hatfield et al., 1995), facial (Davis et al., 2010; Manstead, 1988; Matsumoto, 1987), and postural (Duclos et al., 1989; Stepper & Strack, 1993) posing of emotional displays influences their experienced emotional state as well as their evaluation of the emotional stimuli (Strack et al., 1988).

However, the change in the affective state is not mandatory in all social situations. Emotional mimicry has also been proposed to serve communicative functions and to be guided by cultural norms (Lakin et al., 2003; Hess & Burgeois, 2010). Smiling in response to others' smiles can signal acknowledgement of affiliative intentions as well as the desire to affiliate, and may not necessarily lead to a change in the observers' affective state (Hess & Blairy, 2001; Hess & Burgeois, 2010; Hess et al., 2000; Knutson, 1996).

In contrast to the automatic mimicry view of RFRs to others' emotions, more recent theoretical perspectives suggest that these responses may be the result of more complex mechanisms involving a combination of motor, affective, and cognitive processes (Beall et al., 2008; Burgeois & Hess, 2008; Hess et al., 1998; Jones, 2007; Moody & McIntosh, 2006, 2011; Moody et al., 2007). The emotions of other people are usually highly salient for us, conveying important information for our social success and survival. Processing such emotional information can elicit a change in our affective states as observers, which is further expressed through face, body posture, and prosody. According to this view, the change in affective state and the corresponding RFRs will not necessarily be congruent with the observed facial expression, but rather congruent with the emotional interpretation and the affective state of the observer. Moreover, any emotional expression modality and any emotional information can elicit such responses.

One particular strong argument in favour of this latter perspective comes from studies investigating RFRs to expressions of anger. Expressions of anger are perceived by children and adults as signalling threat, and elicit increased allocation of attention and fast activation of the limbic system, similar to perceiving expressions of fear (Kret et al., 2011; Monk et al., 2008; Nelson & Nugent, 1990; Pichon et al., 2009). Feeling fear in response to others' anger has a potentially adaptive value, since it can facilitate flight in front of danger (LeDoux, 2000; Moody et al., 2007). It has been shown that adults in a high state of fear respond very fast to observing pictures of angry faces, with an increased activation of the facial muscles involved in expressing fear (Moody et al., 2007). This suggests that the RFRs are more congruent with the felt emotion rather than with the observed expression. Adult RFRs specific to fear are also elicited by images depicting environmental threat, like snakes (Dimberg, 1997), and by seeing bodily expressions of fear (Magnée et al., 2007; Tamietto & de Gelder, 2008). This indicates that, at least in certain situations, these responses are less likely to be the result of motor mimicry since the corresponding motor model is not present (Moody & McIntosh, 2006; Tamietto et al., 2009).

The RFRs relying on emotion-specific programs can also be to a certain degree automated. When adults are presented with masked emotional faces and body postures which they are not able to consciously see, they nevertheless show RFRs consistent with the emotional valence of the stimuli (Tamietto & de Gelder, 2008). Even the adults who are unable to consciously perceive visual information, due to unilateral destruction of the visual cortex, show RFRs congruent with the emotional valence of the facial and bodily expressions of emotions (Tamietto et al., 2009). In contrast, the RFRs which mimic the observed emotional facial expressions tend to be associated with increased allocation of attention, as indexed by changes in the electrical cortical activity (Achaibou et al., 2008), similar to other instances of non-emotional motor resonance (Chong et al., 2009). The modulation of the RFRs by early cognitive processes may explain the dissociation in the chain of processes elicited by perceiving others' emotions, activating either perception-action matching mechanisms or affect related processes. Functional magnetic resonance imaging (fMRI) studies show that both the emotion related circuitries and cortical networks typically associated with perception-action matching mechanisms are activated during imitation and passive viewing of facial expressions of emotions. However, due to the poor spatial resolution of the method they cannot disambiguate which mechanism has primacy (Carr et al., 2003; Lee et al., 2006, 2008; Pfeifer et al., 2008).

Although it is widely agreed that at least beginning with the age of 20 months children systematically reproduce in a spontaneous manner various non-emotional motor gestures observed in adults (Flynn & Whiten, 2008; Hopper et al., 2010; Jones, 2007), a less clear picture emerged so far with regards to their facial responses to others' expressions of emotions. Some clues are provided by the research investigating children's abilities to empathize (see Eisenberg, 2000, for a recent review). In most of these studies, changes in children's facial, vocal, and postural expressivity as a result of observing other's emotions are typically measured in order to establish the presence of empathic responses. The evidence converges in showing that children respond to others' affect, most often negative affect, with congruent emotional states (Decety & Svetlova, 2012; Eisenberg, 2000).

Only few studies specifically investigated children's RFRs to others' emotional displays by using EMG recordings of the facial muscles. One of the most important findings resulting from these studies is that children between the ages of 6- and 12-years show changes in their facial muscles activity in response to observing a variety of adult and child emotional facial expressions (i.e., happiness, anger, sadness, fear, and disgust) presented in either a static or dynamic way (Beall et al., 2008; Deschamps et al., 2014; de Wied et al., 2006; Oberman, Winkielman, & Ramachandran, 2009). Most of these studies assume that children's RFRs are the result of motor matching mechanisms (Deschamps et al., 2014; de Wied et al., 2006; Oberman, Winkielman, & Ramachandran, 2009), due to the selective activation of those facial muscles involved in the observed facial expression. Children's passive viewing of emotional facial expressions also leads to a small increase in the hemodynamic response of the cortical areas typically associated with the mirror neurons system (Pfeifer et al., 2008). One study, however, suggests that children's RFRs may also involve affective processes. Beall et al. (2008) presented 7- to 12-year-old children with static adult facial displays of happiness, anger, and fear, while the activity of the muscles specifically involved in expressing each of these emotions (i.e., zygomaticus major, corrugator supercilii, and medial frontalis respectively) was recorded using EMG. Similar to the other studies, an increased activity in the zygomaticus major, the smiling muscle, was recorded when children looked at happy faces. Unlike in the other developmental studies, but similar to some adult investigations (Moody et al., 2007; Magnée, de Gelder, Van Engeland, & Kemner, 2007), seeing angry faces elicited a selective increased activation of the medial frontalis muscle typically involved in raising the eye-brows while expressing fear (Darwin, 2002; Ekman, 1979). Therefore, children seem to display a facial expression that matches their affective state, in this case fear, in response to anger as a potential threat (Monk et al., 2008; Nelson & Nugent, 1990).

Several possible explanations could account for these discrepant results. Most of the studies in which children react with RFRs matching the perceived expression use active tasks in which the participants are asked to specifically pay attention to the emotional expression, to identify it and to verbally label it (Oberman et al., 2009; de Wied et al., 2006). This increased attention to the emotional expressions may have influenced subsequent processing, activating those mechanisms involved in mimicry, as suggested by the adult findings (Achaibou et al., 2008). Indeed, when adults and children specifically focus their attention on mimicking a facial expression, the activation of the cortical areas associate with the mirror neuron system is higher than during passive viewing (Pfeifer et al., 2008). One solution which could help further reduce the ambiguity regarding the mechanisms involved in children's RFRs is to present them with emotional stimuli containing cues about the motor acts required for mimicking the associated expression (i.e., faces) and emotional stimuli in which such information is absent (e.g., emotional body postures, emotional prosody). If affect processes are primarily responsible for observing the RFRs, then one would expect that they are similarly present for both types of stimuli (Magnée et al., 2007; de Gelder et al., 2004; Tamietto & de Gelder, 2008).

The current study aims to advance our understanding of RFRs development in two respects. First, we investigate whether such responses are present in childhood earlier than previously shown. Although evidence suggests that at least from the age of 2 years children can spontaneously reproduce the non-emotional motor gestures observed in others (e.g., Jones, 2007), most research on emotional RFRs has focussed on children over 6-years of age. Our study aims to reduce this gap by testing 3-years-old children's RFRs using EMG measurements of facial muscles activity. Second, the current study investigates whether the 3-year-old children's pattern of RFRs is consistent solely with motor mimicry interpretation or could also be regarded as a result of emotional appraisal processes. To help delineate between the two processes, we present children with static images of both faces and body postures displaying happy, anger, fear, and neutral emotional expressions. By the age of 3 years, children recognize and label body expressions of emotions with the same accuracy as for facial expressions (Nelson & Russell, 2011), suggesting good abilities to process the emotional information expressed this way. Recording the selective activation of the facial muscle representative for a certain emotional expression (i.e., zygomaticus major for happiness, corrugator supercilii for anger; frontalis medialis for fear) in response to both faces and body postures would be more consistent with an emotional processing interpretation (Magnee et al., 2007b; Tamietto & de Gelder, 2008). This idea will be further supported by finding that observing displays of anger elicit the selective activation of the frontalis medialis, the facial muscle specific for expressing fear (Beall et al., 2008).

Methods

Participants

A total of 22 healthy 3-year-old children (10 females; mean age = 40.42 months, age range = 36.50 – 47.57 months) were included in the final analysis. Nineteen additional children were tested, but then discharged from the final sample because they refused to watch the stimuli (n = 7), moved too much during trial presentation (n = 8) and did not complete the minimum number of trials required for data analysis (n = 4). The protocol was carried out in accordance with the ethical standards of the Declaration of Helsinki (BMJ 1991; 302:1194) and approved by the Ethical Committee of the University. Parents gave written informed consent for their children to participate in the study.

Stimuli and Procedure

Participants were presented with color photographs of human female *faces* and *bodies* displaying happy (HA), angry (AN), fear (FE) and neutral (NE) expressions on a 24” LCD monitor at a distance of approximately 80 cm. Face stimuli were selected from the Radboud Faces Database (RaFD; Langner et al., 2010), while body stimuli were extracted from the Bodily Expressive Action Stimulus Test database (BEAST; de Gelder & Van den Stock, 2011). Both face and body stimuli were screened and selected by 3 adult raters for their emotional valence. In order to ensure that the processing of the emotional information expressed through body postures is not influenced by the facial expression, all faces on the body stimuli were masked with an opaque patch (Figure 1). Each stimulus was presented at the center of the screen on a grey background for 500 ms and was preceded by an inter-stimulus interval of 2000 ms consisting of a grey screen with a central fixation cross, similar to previous studies using this paradigm (Oberman et al., 2009). In a completely within-subjects design, face and body stimuli were presented in alternating blocks. Each block consisted of 20 randomly presented stimuli (5 for each emotional expression), with the only constraint that the stimuli displaying the same emotion could not occur more than twice consecutively. The order of presentation was counterbalanced across participants, so that half of them started the experiment with the body and the other half with the face condition.

Figure 1. Examples of the face (a) and body (b) emotion expressions used as stimuli in the study.

Upon completing informed consent procedures, participants' faces were cleaned and scrubbed with NuPrep Gel to ensure good quality signal recording from the EMG electrodes. Children sat on a chair in a dimly lit, audiometric and electrically shielded cabin. An experimenter was present throughout the entire procedure so that participants' movements were minimized and their interest and attention were maintained. Children were instructed to relax, to not move or talk, and to watch the pictures on the screen. No other instruction was given to the participants. In order for the children to familiarize with the procedure and to ensure that they understood the instructions, each session started with 8 practice trials in which an equal number of faces and bodies were displayed. Total duration of the task was approximately 15 minutes and at the end of the session, participants received a small reward.

sEMG Recordings and Data reduction

EMG was used to record the levels of muscle activation for the zygomaticus major (raises the cheek), the medial frontalis (raises the brow), and the corrugator supercilii (knits brow). These muscles were chosen based on previous studies showing that their activation is a reliable marker for facial expressions of happiness (zygomaticus major), anger (corrugator supercilii), and fear (frontalis medialis) (Cacioppo et al., 1986; Ekman & Friesen, 1976; Frois-Wittman, 1930). A D360 Digitimer electromyograph was used to continuously record the EMG signal from the selected muscles using bipolar montages, following previously established guidelines (Tassinary & Cacioppo, 2000). Ambu Neuroline 700 surface adhesive 4 mm Ag-AgCl electrodes for pediatric use were placed on the child's face at locations corresponding to each muscle. The electrodes were positioned longitudinal to the muscle, with an inter-electrode distance of 10 mm between their centers. Electrodes were positioned on the left side of the face to obtain maximal reactions (Fridlund & Cacioppo, 1986). The reference electrode was positioned just below the hairline, ~3 cm above the nasion. Impedance was kept between 5 and 10 kΩ using a conductive EMG gel (Viasys Electrolyte Gel). The EMG signal was amplified online by a factor of 1000 and recorded at a sampling rate of 1 kHz with a 10-1000 Hz bandpass filter. The EMG signal was filtered offline (150 Hz; high-pass: 30 Hz), and further rectified for analysis using Spike2 software (Cambridge Electronic Design Ltd., Cambridge, UK). Because of difficulties and excessive noise recorded from the corrugator supercilii muscle, data acquired from this electrode site were excluded from further analysis. One consequence of the lack of data from this muscle is that it will make it difficult to draw conclusions regarding the presence of RFRs specific to anger. Nevertheless, considering our prediction of fear RFRs to the emotional stimuli expressing anger, intact recordings of the frontalis medialis will allow us meaningful interpretations of the results in this respect (Beall et al., 2008).

Children's looking time toward the stimuli was coded offline and trials in which they looked at the stimuli for less than 70% of its duration or were moving, were discarded. In order to avoid any spurious effect produced by participants' movements while watching the stimuli, trials were also discarded whenever signal noise and motion artifacts contaminated the EMG recordings. Only children with at least 4 trials per emotion/condition were included in the statistical analyses. Across participants, the mean number of trials contributing to the statistical analyses was 13.02 (HA: 13.09; AN: 12.77; FE: 13.59; NE: 12.64) per emotion in the face condition, and 12.98 (HA: 13.41; AN: 12.82; FE: 13.23; NE: 12.45) per emotion in the body condition. A similar number of trials contributed to the final analysis for each condition, $F(3,63) = 2.016$; $p > .12$

Average amplitude values were calculated for each 100 ms interval from 500 ms pre-stimulus onset to 1500 ms post-stimulus. In order to reduce the impact of extreme values and standardize the observed activation, we transformed raw data in Z scores within participants and muscle sites. Next, each 100 ms interval post-stimulus onset was baseline corrected by subtracting the average amplitude of the 500 ms pre-stimulus interval from the average amplitude of each 100 ms post-stimulus onset interval. Finally, trials of the same emotion and condition were averaged to obtain one value for each 100 ms interval of every trial type. Previous studies with children using a similar paradigm have shown that the facial muscles usually begin to show differentiated activation in response to facial expressions of emotions after 500 ms from stimulus onset, reaching the peak around 1000 ms in the case of longer stimulus presentations (Beall et al., 2008; Oberman et al., 2009), which is also consistent with adult studies (Dimberg, 1982; Dimberg & Petterson, 2000; Moody et al., 2007). Visual inspection of the data in the current study suggested a similar pattern, with the recorded muscles showing differentiated activation between 800 - 1300 ms post stimulus onset. The mean amplitude values for this time window were further analyzed using a 2 (Condition: bodies and faces) x 4 (Emotion: HA, AN, FE, NE) x 2 (Muscle: zygomaticus major and medial frontalis) repeated measures ANOVA. All statistical tests were conducted at .05 level of significance (two-tailed), and paired sample *t*-tests were corrected for multiple comparisons using the Holm-Bonferroni stepwise procedure. Furthermore, in order to confirm that the EMG activity of a specific muscle changed in response to a certain emotional stimulus, each significant Emotion x Muscle interaction was followed-up by a comparison of the non-baseline corrected EMG data of each condition during the 800-1300 ms post stimulus onset with that recorded during the pre-stimulus 500 ms baseline when a fixation cross was displayed. For this purpose, we used paired t-tests at .05 level of significance (two-tailed).

Results

Table 1 shows the mean activation (with SDs) for the zygomaticus and frontalis muscles across conditions. The results of the 2 (Condition: face stimuli, body stimuli) x 4 (Emotional expression: happy, anger, fear, neutral) x 2 (Muscle: Zygomaticus Major, Frontalis Medialis) repeated measures ANOVA show a significant interaction between condition, emotion, and muscle, $F(3,60) = 6.008$, $p = .001$, $\eta^2 = .231$. No other significant main effects or interactions were found ($p > .291$). In order to unpack this interaction 4 (Emotion: happy, anger, fear, neutral) x 2 (Muscle: Zygomaticus Major, Frontalis Medialis) repeated measures ANOVAs were performed separately for each condition.

Table 1. Mean (M) and standard deviation (SD) values of the electromyography activation recorded from the zygomaticus and frontalis muscles in response to facial and bodily expressions of emotion in the 800-1300 ms time window.

		Zygomaticus (Z scores)	Frontalis (Z scores)
		M (SD)	*M (SD)*
Anger	Face	-.075 (.147)	.060 (.122)
	Body	.059 (.208)	-.073 (.219)
Happiness	Face	.090 (.160)	-.057 (.112)
	Body	.013 (.106)	.045 (.123)
Fear	Face	-.038 (.158)	-.024 (.141)
	Body	-.022 (.196)	-.006 (.172)
Neutral	Face	.024 (.163)	.038 (.144)
	Body	-.023 (.234)	.022 (.128)

Face stimuli

A significant interaction between emotional expression and muscle emerged, $F(3,60) = 5.310$, $p = .003$, $\eta^2 = .210$, suggesting a selective activation of the recorded muscles for specific emotional expressions. Post-hoc pairwise comparisons revealed that observing facial expressions of happiness elicits increased activation of the zygomaticus major ($M = .090$; $SD = .160$) compared to observing angry faces ($M = -.075$; $SD = .147$), $t(21) = 3.452$, $p = .026$. In contrast, observing facial expressions of anger led to an increased activation of the frontalis ($M = .060$, $SD = .122$) compared to observing happy faces ($M = -.056$, $SD = .112$), *t*(21) = 3.396, *p* = .036 (Figure 2). The use of standardized Z scores also allowed us to compare the level of activation between muscles. The analysis of the difference in activation for both zygomaticus and frontalis within emotion expression, further supports the results of selective activation by showing that observing facial expressions of happiness leads to activation of the muscle responsible for smiling (zygomaticus major, *M* = .090; *SD* = .160) and deactivation of the muscle which raises the eye-brows (frontalis medialis, *M* = -.056, *SD* = .112), *t*(21) = 3.696, *p* = .014, while observing angry faces leads to activation of the frontalis (*M* = .060, *SD* = .122) and deactivation of the zygomaticus (*M* = -.075; *SD* = .147), *t*(21) = 3.387, *p* = .036. When compared to the baseline, observing happy facial expressions elicited an increased activation of the zygomaticus major (*t*(21) = 2.392, *p* = .026), while the angry faces led to a decrease in the activation of the same muscle (*t*(21) = -2.501, *p* = .021). In contrast, observing happy faces led to a decrease in the activity of the frontalis muscle from the baseline levels (*t*(21) = -2.688, *p* = .014), while the same muscle tended to show an increased activation in response to angry faces when compared to the baseline, although it was marginally significant (*t*(21) = 1.947, *p* = .066). No other significant differences emerged.

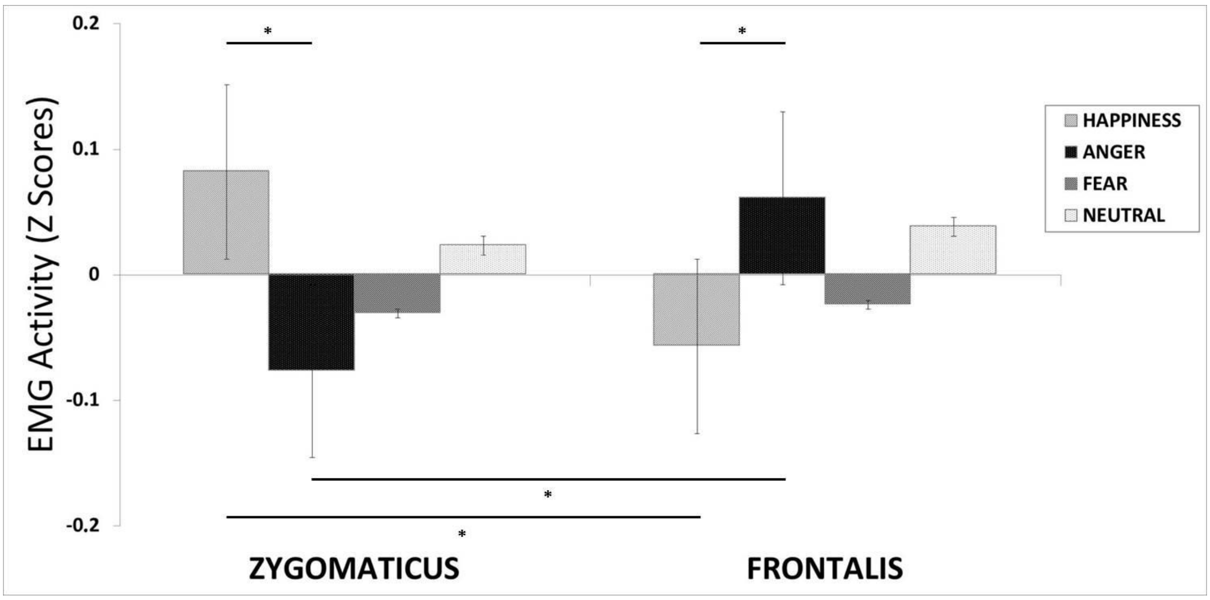

Figure 2. Electromyographic activation recorded from the zygomaticus (left) and frontalis (right) muscles in response to facial expressions of emotion in the 800-1300 ms time window. The error bars represent the standard errors.

Body stimuli

The analysis of the average muscle activation recorded in response to observing body postures did not show a significant interaction between the emotional expression and the type of muscle, Emotion x Muscle, $F(3,60) = 2.355$, $p = .100$, $\eta^2 = .105$ (Figure 3). Similar levels of activation of both zygomaticus major and frontalis medialis were recorded in response to all types of body postures, $p > .960$.

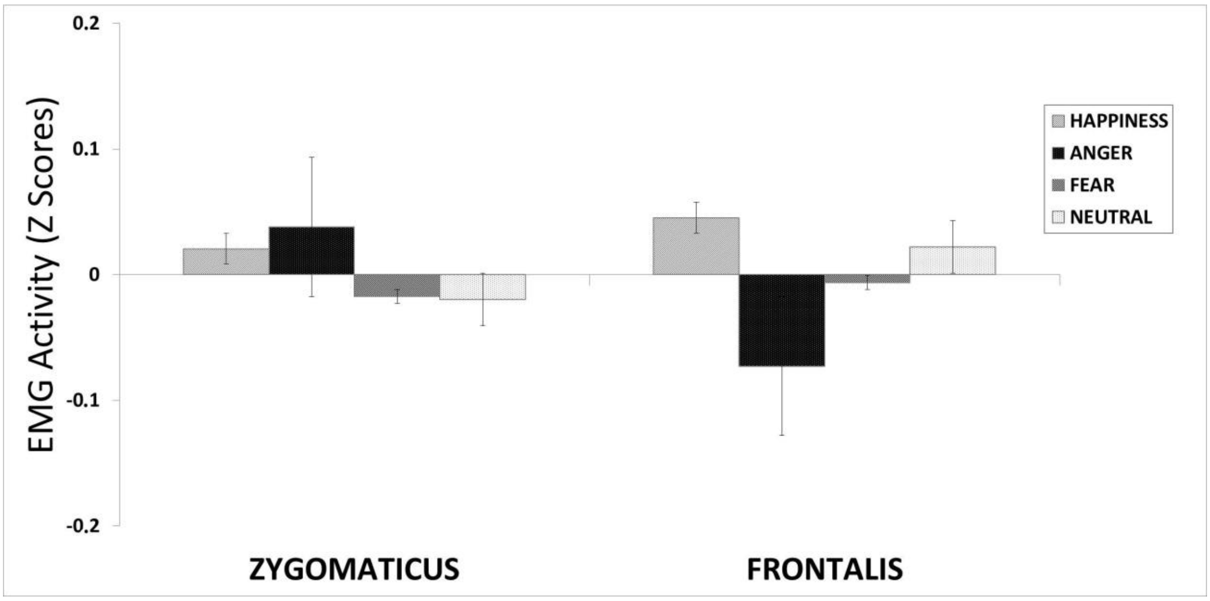

Figure 3. Electromyographic activation recorded from the zygomaticus (left) and frontalis (right) muscles in response to body expressions of emotion in the 800-1300 ms time window. The error bars represent the standard errors.

Discussions

The aim of our study was to investigate whether 3-year-olds show RFRs to others' expressions of emotions, and to explore the mechanisms underlying these responses. Towards this aim we presented children with static images of faces and bodies displaying happiness, fear, anger, and emotionally neutral expressions. RFRs were recorded using EMG from the zygomaticus major, the muscle involved in pulling the corners of the mouth in a smile, typically associated with expressing happiness, and from the frontalis medialis, the muscle which raises the eye brows, typically involved in expressing fear.

Convergent with previous studies with older children (Beall et al., 2008) and adults (Moody et al., 2007), we have shown for the first time that 3-years-old children manifest selective RFRs, as measured by EMG, to static facial expressions of happiness and anger. More specifically, observing others' happy faces lead to the increased activation of the zygomaticus major and decreased activation of the frontalis medialis. Observing angry faces triggered an opposite pattern of activation. These findings were supported by the analysis of the EMG responses both when conditions were directly compared one with the other and when each condition was compared to the baseline.

The RFRs to angry facial expressions suggest that affective processes may also be involved and thus do not rely solely on perception-action matching mechanisms (Beall et al., 2008; Burgeois & Hess, 2008; Hess et al., 1998; Jones, 2007; Moody & McIntosh, 2006, 2011; Moody et al., 2007). Based on the responses to happy facial expressions alone, such an interpretation would be hazardous, given that both types of processes would result in similar responses. Seeing someone smiling could be processed as a cue for pleasant social interaction leading to a happy response in the observer, usually expressed through smile.

Mimicking the observed smile in order to acknowledge others' affiliative intentions would also lead to this response (Hess & Blairy, 2001; Hess & Burgeois, 2010; Hess et al., 2000; Knutson, 1996). However, the fact that angry faces led to a change in facial muscle activation specific to fear is more in line with interpreting RFRs as involving the emotional interpretation of the stimuli (Beall et al., 2008; Moody et al., 2007). An angry face with the eye gaze directed at the perceiver is usually regarded as threatening and potentially elicits fear (Öhmann, 2005). The fact that we were not able to provide information about the response of the corrugator muscle to static angry faces, may be regarded as limiting our conclusions. However, the activation of the frontalis, with or without the associated activity of the corrugator, is specific for expressing fear, not anger (Eckman & Friesen, 1978; Boxtel, 2010). Further investigations in which measures of emotional arousal (e.g., heart rate, pupil dilation, galvanic skin response) are recorded simultaneously with facial EMG from all three muscles, could help elucidate whether the 3-year-olds' RFRs to others' emotional facial expressions are associated with a change in the affective state. This association may also depend on the extent to which different children respond emotionally to socioemotional events and the efficiency with which they regulate their emotions, since the temperamental characteristics recorded during the first years of life largely explain the variability in empathy development (van der Mark, van IJzendoorn, & Bakermans-Kranenburg, 2002; Young, Fox, Zahn-Waxler, 1999).

Neither the emotionally neutral nor the fearful faces elicited selective activation of the recorded facial muscles. The fact that in our study static fearful faces did not elicit selective RFRs in 3-year-old children is in line with Beall et al. (2008) findings for 7- to 12-year-old children and Moody et al. (2007) findings for adults. However, they are in contrast to those of Deschamp et al. (2014) and Oberman et al. (2009). Facial expressions of fear are typically regarded as cues for threat (Adams, Gordon, Baird, Ambady, & Kleck, 2003; Pessoa, Japee, & Ungerleider, 2005), which capture attention and elicit fear (Öhmann, 2005; Vuilleumier, 2002). One possible explanation for the lack of selective RFRs in our study could be that 3-year-old children's abilities to process fearful facial expressions are not sufficiently mature. In terms of processing the specific facial features, humans are able to discriminate fearful from other emotionally positive and negative facial expressions both visually and at the neural level as early as 5- to 7-months after birth (Schwartz, Izard, & Ansul, 1985; Hoehl & Striano, 2008). Notwithstanding infants' sophisticated abilities to process others' emotional expressions, the literatures converge to suggest that it takes many years before children reach the adults' level of accuracy and speed in recognizing facial expressions. In particular, children's sensitivity to fearful expressions continues to improve till 10 years of age (Herba & Phillips, 2004; Gao & Maurer, 2009, 2010). Moreover, it is possible that 3-year-old children experience less negative than positive emotional expressions, and in particular they may encounter fewer instance during everyday life of other people manifesting fearful than happy and even angry facial expressions (Gao & Maurer, 2010; Grossman, Striano, Federici, 2007). Our findings that the frontalis muscle tends to show less change from baseline in response to angry faces than the response of zygomaticus in response to happy faces could be regarded as indirectly supporting the idea that a differential amount of experience with certain emotional expression may have an impact on children's RFRs. The most experienced emotional expressions could trigger more easily RFRs than the less experienced ones.

Another different interpretation for the lack of RFRs for fearful facial expressions might suggest the involvement of affect mechanisms. Beyond infancy, more complex knowledge about emotions, including fear, emerges. For example, the ability to verbally label emotional expressions is manifest more systematically for happiness and anger around the age of 3-years, while for fear more so towards the age of 5-years (Widen & Russel, 2003). The knowledge about the events that could potentially cause fear, although present to a certain extent by the age of 2-years, continues to improve beyond the age of 3-years (Denham & Couchoud, 1990; Mondloch, Horner, & Mian, 2012). Thus, one possibility could be that insufficient affect knowledge about fear impairs 3-year-old RFRs to these emotional expressions. However, this explanation is less likely to account for the same findings in Beall et al. (2008), since by the age of 7- to 12-years affect knowledge is advanced. Future studies in which measures of affect knowledge are included could help testing this hypothesis.

The discrepant results in RFRs to fear may also be due to a difference in the saliency of the fearful expressions as cues for threat used in the current and previous studies. Oberman et al. (2009) asked children to verbally label and categorize the observed emotional expressions, while Deschamp et al. (2014) presented dynamic stimuli. These procedural aspects may have modulated children's processing of emotional expressions. In our study, similarly to Beall et al. (2008), we asked children to watch static facial expressions of fear with gaze directed towards the observer, without any further instructions. It is possible that in passive tasks using static stimuli that provide impoverished emotional information, the interpretation of fearful facial expressions as cues for threat is more dependent on certain features of the face or of the environment pointing to the source of threat, like the eye-gaze (Fox et al., 2007; Hoehl & Straino, 2008; Hoehl & Straino, 2010; Neath et al., 2013). Fearful faces with eye-gaze directed towards a specific aspect of the environment more clearly points to the specific source of threat and it is more meaningful than a fearful face with the eye-gaze oriented towards the observer. This typically influences participants' attentiveness and behaviour related to that object starting from infancy (Hoehl & Striano, 2010), and continuing throughout childhood and adulthood (Neath et al., 2013). It is thus possible that the static fearful stimuli used in our study and in Beall et al. (2008) were not sufficiently informative with respect to the potential threat. Future studies in which the orientation of the eye-gaze in fearful and angry faces is specifically manipulated, as well as the use of both static and dynamic stimuli, could greatly contribute to understanding the underlying mechanisms of RFRs to emotional faces in children.

As for the bodily expressions of emotions, we found that observing human bodies with happy, angry, fearful, and emotionally neutral postures resulted in non-selective RFRs. Taken in isolation from the pattern of EMG responses to facial expressions of emotions, these findings would suggest that 3-year-old children' RFRs could be the result of perception-action matching mechanisms (Bavelas et al., 1986; Chartrand & Bargh, 1999; Hoffman, 1984; Meltzoff & Moore, 1977). Nevertheless, since the RFRs to emotional facial expressions did not fully follow the pattern of muscle activation expected in case of mimicry (i.e., zygomaticus major for happiness, frontalis medialis for fear), this explanation is less likely to be the case. In adults, emotion specific facial muscle activity has been recorded in response to both faces and bodies expressing happiness and fear (Magnee et al., 2007a; Tamietto & deGelder, 2008). What could thus explain the difference in RFRs to static emotional body postures between adults and 3-year-old children? Although only few studies have investigated the development of processing emotional information expressed in body postures, they converge in showing that already by the age of 6-8 months after birth, infants discriminate visually and at the neural level between positive and negative emotional body postures (Zieber et al., 2014; Missana, Atkinson, & Grossmann, 2014). It is thus less likely that the lack of emotion specific RFRs in 3-year-old children is due to an inability to tell apart between different emotional body postures. Also, 3-years-old children correctly label emotional expressions both for bodies and faces (Nelson & Russell, 2011), suggesting that this ability might not necessarily account for the RFRs to body postures. One task in which 3-year-old children perform differently for facial expressions and body postures is the ability to relate emotional expressions observed in others with the events potentially causing them (Mondloch, Horner, & Mian, 2013). While 3-year-old children are able to correctly associate an emotional facial expression of a person with the events most likely causing the associated affective state, they fail to do so for emotional body postures. This may be due to the difference in emotional information that the body postures communicate (Ekman, 1965). The ability to interpret such information may develop at a different pace than faces, potentially explaining the lack of emotionally specific RFRs to emotionally body postures in 3-year-old children. In our current study we did not include any measure of affect knowledge to assess whether 3-year-old children discriminate, label, and understand the meaning of different means of emotional expressivity. Further studies in which other emotional expression modalities than those included in this study are used (i.e., emotional prosody) together with measures of affect knowledge could help us understand whether the lack of selective RFRs for emotional expressions other than faces reflect the presence of perception-action mechanisms, affective processes or a combination of both.

In sum, the findings of our study provide valuable insight into 3-year-old children's facial responses to others' emotions, particularly when displayed in static images, and show that EMG recordings can be a viable tool of investigation for this age group. The reported results speak in favour of RFRs as the result of complex mechanisms in which affective processes may play an important role. These findings add to a growing body of research on the development of complex social and emotional abilities like empathy (Decety & Svetlova, 2012; Decety, 2015; Geangu, 2015; Geangu et al., 2011) and social understanding (Meltzoff, 2007; Carpendale & Lewis, 2006). It will be particularly interesting to explore whether RFRs to others' emotions are related to children's abilities to share the emotional experiences of people around them or whether they contribute to how well children understand their own and others' emotions. In light of recent research showing that EMG is a valid tool to be used even with infants (Natale et al., 2014; Turati et al., 2013), the current findings open an important possibility for addressing long standing questions about infants' facial responses to others' emotional expressions (Field et al., 1983; Geangu et al., 2011; Kaitz et al., 1988; Haviland & Lelwicka, 1987; Ray & Heyes, 2011).

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*Highlights (for review)

- 3-years-old children show rapid facial responses (RFRs) to others' emotional faces
- Observing happy faces selectively activates the smiling muscle (zygomaticus major)
- Observing angry faces elicits RFRs specific for fear (frontalis medialis)
- Emotional bodies do not trigger emotion selective activation of the facial muscles
- Electromyography is a viable tool for investigating emotional RFRs in pre-schoolers

Figure captions

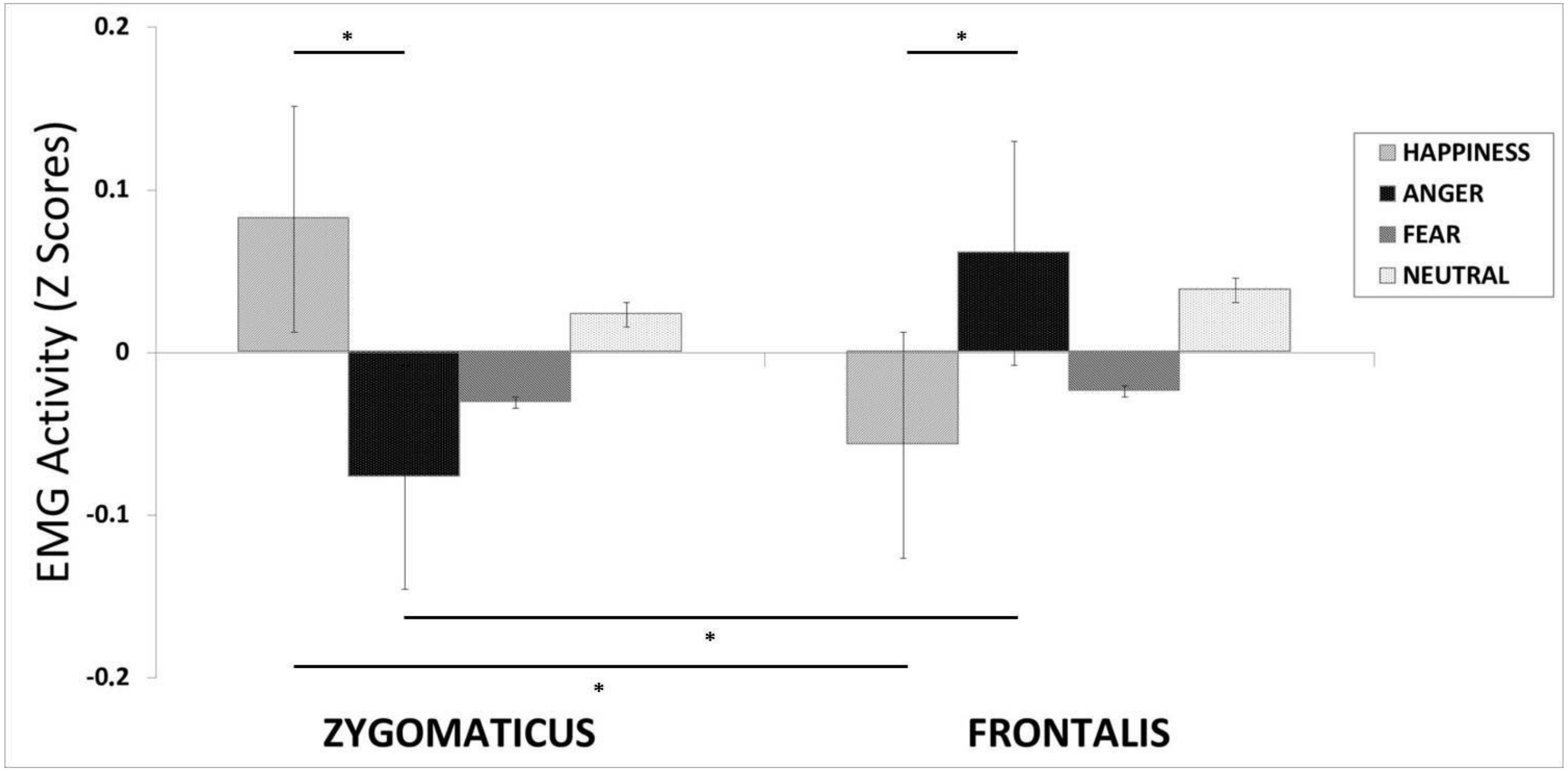
Figure 1. Examples of the face (a) and body (b) emotion expressions used as stimuli in the study.

Figure 2. Mean electromyographic activation (with SE) recorded from the zygomaticus (left) and frontalis (right) muscles in response to facial expressions of emotion in the 800-1300 ms time window.

Figure 3. Mean electromyographic activation (with SE) recorded from the zygomaticus (left) and frontalis (right) muscles in response to body expressions of emotion in the 800-1300 ms time window.

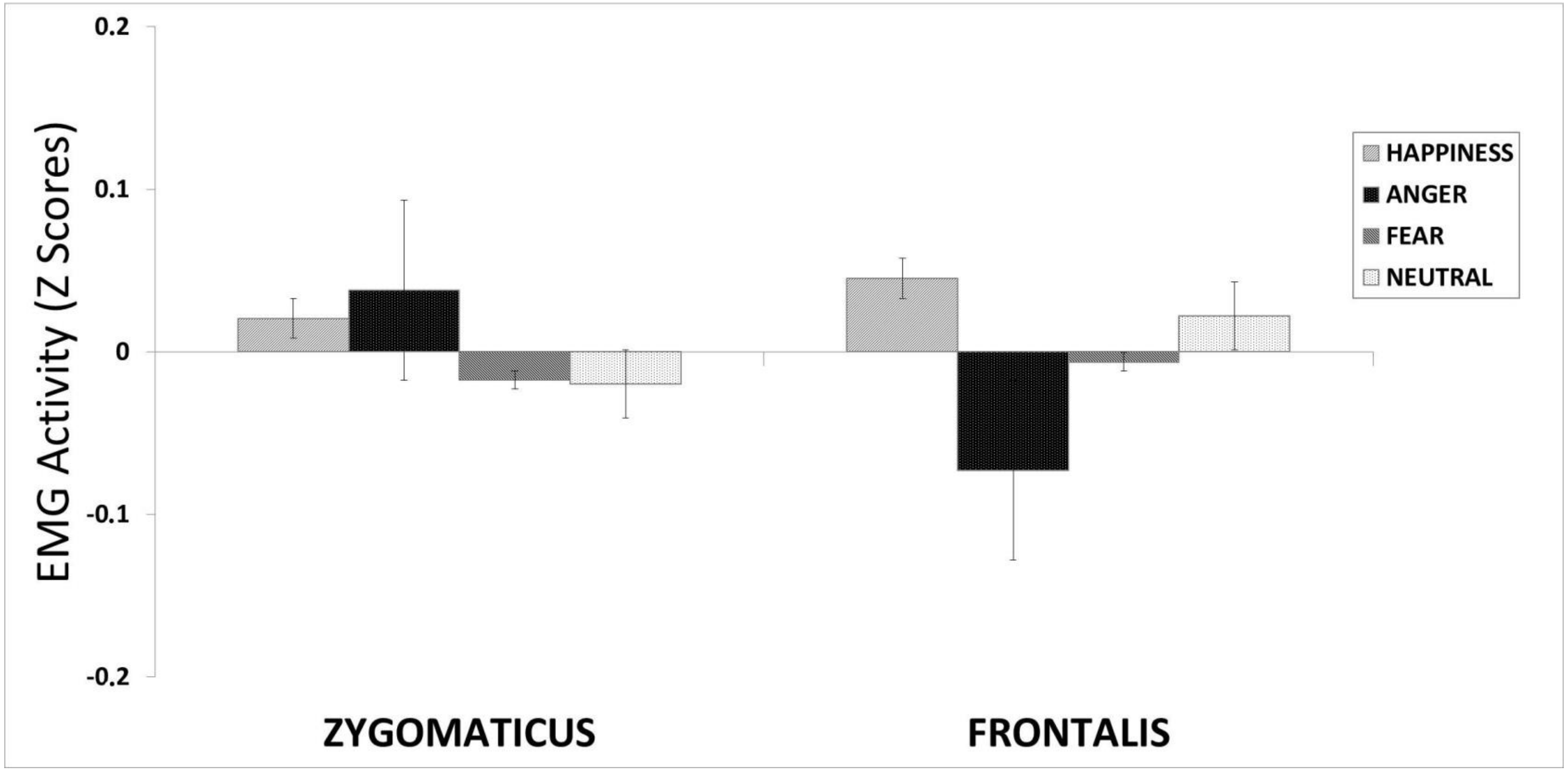
Figure1

Figure2



EMG Activity (Z Scores)
0.2
0.1
0
-0.1
-0.2
*
*
*
*
HAPPINESS
ANGER
FEAR
NEUTRAL
ZYGOMATICUS
FRONTALIS

Figure3


0.2
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-0.1
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EMG Activity (Z Scores)
HAPPINESS
ANGER
FEAR
NEUTRAL
ZYGOMATICUS
FRONTALIS

Table1

Table 1. Mean (M) and standard deviation (SD) values of the electromyography activation recorded from the zygomaticus and frontalis muscles in response to facial and bodily expressions of emotion in the 800-1300 ms time window.

		Zygomaticus (Z scores)	Frontalis (Z scores)
		M (SD)	*M (SD)*
Anger	Face	-.075 (.147)	.060 (.122)
	Body	.059 (.208)	-.073 (.219)
Happiness	Face	.090 (.160)	-.057 (.112)
	Body	.013 (.106)	.045 (.123)
Fear	Face	-.038 (.158)	-.024 (.141)
	Body	-.022 (.196)	-.006 (.172)
Neutral	Face	.024 (.163)	.038 (.144)
	Body	-.023 (.234)	.022 (.128)