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Motherhood and oxytocin receptor genetic variation are associated with selective changes in electrocortical responses to infant facial expressions

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Abstract

Recent studies suggest that parental caregiving is associated with adaptive changes in neurocognitive responses to emotional cues and oxytocin function, possibly reflecting the increased need of parents to monitor infants' emotional states. In the current study, we investigated whether the changes associated with motherhood and oxytocin receptor genetic variation rs53576 are specific to the processing of infant cues as opposed to a more general increase in responsiveness to emotional cues. We measured event-related brain potentials (ERPs) and behavioral recognition responses from mothers of young infants (n = 48) and nulliparous females (n = 46) to infant and adult faces displaying strong and mild intensity emotional expressions. Mothers and GG allele carriers of the *OXTR* gene showed an early-latency (~100 ms) differential frontal ERP response to strong intensity facial expressions, and mothers also showed modulation of the posterior EPN waveform by negative valence. The early frontal ERP modulation was associated with faster emotion recognition performance across participants. Most importantly, these effects were highly specific to infant facial expressions. The results point to a dissociable neurocognitive system that is involved in monitoring infants' emotional cues and may be important in supporting parental caregiving in humans.

KEYWORDS: Motherhood; Oxytocin receptor gene; Event-related potentials; Facial expressions

Young infants are equipped with a limited but highly evocative repertoire of facial emotional expressions (Camras & Shutter, 2010) that effectively elicit parental caregiving and proximity. Recent research suggests that adaptive changes in adults' natural tendency to attend to infant faces are important for mediating parental caregiving (Rilling, 2013). Studies measuring rapid neural responses with event-related potentials (ERP) or magnetoencephalography (MEG) have shown increased neural activity elicited by infant vs. adult faces in early (~130 ms) orbitofrontal (Kringelbach et al., 2008) as well as in later occipitotemporal ERP responses reflecting the activity of face-sensitive cortical structures (Proverbio, Riva, Zani, & Martin, 2011) and attention allocation (Grasso, Moser, Dozier, & Simons, 2009). Recording ERPs in parents and non-parents in response to infants' facial expression stimuli, Proverbio, Brignone, Matarazzo, Del Zotto, and Zani (2006) showed that parenting amplifies neural responses to infants' emotional signals. Specifically, posterior ERP responses (375-600 ms after stimulus onset) reflecting the degree of attention allocation were larger in parents than in non-parents and the differences were most pronounced to highly intense negative expressions signaling infant distress. Such automatic attention to salient affect signals may be functionally very important in the context of parenting young infants given the continuous need to promptly respond to distress signals in order to mitigate infants' negative arousal. Our aim in this study was to replicate these (Proverbio et al., 2006) findings with a larger group of mothers and to test whether the ERP effects are specific to infant stimuli, which would substantiate the functional significance of increased neural activity in response to infant emotion expressions.

Similarly to other mammals (Carter, 1998; Insel & Young, 2001), the neuropeptide oxytocin (OT) appears to mediate parenting behaviors and sensitivity to the affective signals of the offspring also in humans. Parent-infant interaction increases parents' salivary OT levels (Feldman, Gordon, Schneiderman, Weisman, & Zagoory-Sharon, 2010), and higher OT levels throughout pregnancy and early postpartum correlate with observed caregiving sensitivity (Feldman, Weller,

Zagoory-Sharon, & Levine, 2007). In this context, it is highly interesting that the efficiency of central oxytocinergic transmission may be modulated by common variations in the gene encoding the oxytocin receptor (*OXTR*). The most extensively studied of these polymorphisms is a guanine (G) to adenine (A) substitution in the third intron of the oxytocin receptor gene (rs53576). This single nucleotide polymorphism is suggested to associate with the number, organization, or functioning of OT receptors in the brain, with the GG genotype (vs. AA or AG) presumably relating to more efficient OT signaling (Yamasue et al., 2012). Riem and colleagues showed that within a group of nulliparous females, the GG genotype was associated with increased cardiac reactivity when listening to infant cry sounds, suggestive of higher sensitivity to infant distress signals (Riem, Pieper, Out, Bakermans-Kranenburg, & van IJzendoorn, 2011). This interpretation is supported by data showing higher observed caregiving sensitivity in mothers homozygous for the GG allele as compared to non-GG mothers (Bakermans-Kranenburg & van IJzendoorn, 2008). We investigated whether the modulatory effects of the putatively more efficient GG variant of the *OXTR* gene extend to ERP responses to infants' facial expressions and provide a potential parallel to the effects of motherhood on ERPs.

Our aim was thus to determine the effects of motherhood and oxytocin receptor genetic variation on ERP responses to infants' facial expressions. We were particularly interested in whether these effects are specific to infant expressions rather than a more generalized bias to any emotional expressions. Indeed, the late stages of pregnancy (Pearson, Lightman, & Evans, 2009; Roos et al., 2012) and increased OT levels following intranasal OT administration (Van IJzendoorn & Bakermans-Kranenburg, 2012) are associated with increased sensitivity to adults' facial expressions, suggesting a generalized bias. To address the infant specificity, we measured responses also to adults' fearful and happy expressions which have been consistently associated with distinct ERP effects (i.e., larger responses to fearful vs. happy faces; Batty & Taylor, 2003; Leppänen, Moulson, Vogel-Farley, & Nelson, 2007). EEG was recorded while mothers and non-mothers were

presented with a stream of infant and adult facial expression stimuli. To maintain a sufficient level of attention to the stimuli and to manipulate the participants' attentional focus (infant vs. adult faces), a counting task requiring the participants to count the number of infant and adult faces in separate blocks was used. Testing whether infant faces are subject to enhanced processing regardless of whether they are actively attended can help to estimate the robustness of the effects. A separate emotion recognition task with the same stimuli was presented to examine whether the putative modulatory effects of maternal status and *OXTR* genotype on ERPs are linked with the speed and accuracy of categorizing emotional expressions.

The ERP analyses focused on ERP components reflecting visual and attentional processing of facial expressions. First, the N1 component is an early-peaking (100-150 ms) response in frontal electrode sites and among the earliest responses differentiating facial expressions, particularly fearful and neutral/happy adult expressions (e.g., Holmes, Vuilleumier, & Eimer, 2003; Luo, Feng, He, Wang, & Luo, 2010). Emotional modulation of the N1 has been suggested to arise from coarse significance detection by the orbitofrontal cortex (Eimer & Holmes, 2007; Luo et al., 2010). From occipitotemporal electrode sites we analyzed the N170 component which is related to the encoding of structural information from faces (Bentin, Allison, Puce, Perez, & McCarthy, 1996). The N170 is modulated by adult facial expressions (Batty & Taylor, 2003; Leppänen et al., 2007; Williams, Palmer, Liddell, Song, & Gordon, 2006; but see Eimer & Holmes, 2002, for an absence of facial expression effects) and this modulation has been shown to correlate with the ability to detect emotional signals from faces (Zhang, Wang, Luo, & Luo, 2012). The degree of attention allocation to emotional expressions was investigated by measuring amplitude modulation in the early posterior negativity (EPN) and late positive potential (LPP) waveforms from posterior recording sites 240-450 ms after stimulus onset. Both the EPN and LPP are associated with attention capture by motivationally significant stimuli such as fearful and angry

facial expressions and other high-arousing emotional images (Leppänen, Kauppinen, Peltola, & Hietanen, 2007; Schupp et al., 2004; Schupp et al., 2007).

Methods

Participants

The participants were mothers (n = 48; mean age = 30.3 years) of 7-month-old infants and nulliparous females (non-mothers; n = 46; mean age = 23.9 years) who had no children of their own. All participants were of Finnish origin and Caucasian ethnicity. The final sample size varied slightly between different analyses as some participants had missing DNA (n = 8 of the original participants) or ERP data (due to EEG artefacts) in some of the analyses. Mothers were recruited through local child welfare clinics and a Population Register Center database. Non-mothers were recruited from university courses. On average, mothers were significantly older than non-mothers, $t(83) = 6.79$, $p < .001$, and, therefore, the potential effects of this difference were statistically controlled by including participant age as a covariate in the analyses where age was significantly associated with the statistical difference between the groups.

Genotyping

For the DNA extraction, 9.0 ml EDTA whole blood was taken from the participants and stored at -20 °C. Genomic DNA was extracted from peripheral blood leukocytes by using QIAamp® DNA Blood Minikit and automated biorobot M48 extraction (Qiagen; Hilden, Germany). The variant rs53576 in the oxytocin receptor gene *OXTR* was genotyped by using Taqman SNP Genotyping Assays (C___3290335_10) and ABI Prism 7900HT Sequence Detection System (Applied Biosystems; Foster City, CA). No discrepancies were detected in the genotyping results of duplicate samples. As in previous studies (Bakermans-Kranenburg & van IJzendoorn, 2008; Rodrigues, Saslow, Garcia, John, & Keltner, 2009), we combined the AG and AA genotypes into a single non-GG genotype. The allele distribution within mothers was GG: n = 13; non-GG: n = 30, and in non-mothers GG: n = 20; non-GG: n = 23. The allele frequencies (GG: n = 33; non-

GG: $n = 53$) deviated from Hardy-Weinberg equilibrium, $\chi 2(1) = 5.0$, $p = .03$, possibly owing to the relatively small sample size in the analysis.

Stimuli

The stimuli were black-and-white photographs of adults' and infants' facial expressions grouped by valence (positive and negative) and intensity (strong and mild). The adult faces displayed open mouth happy (strong positive), neutral (mild positive[1]), closed mouth fearful (mild negative), and open mouth fearful (strong negative) expressions, taken from the NimStim Face Stimulus Set (Tottenham et al., 2009). Infant stimuli were also obtained from an existing stimulus set (see Proverbio, Matarazzo, Brignone, Del Zotto, & Zani, 2007, for illustrations and information on the standardization of this stimulus set). The infant faces displayed pleasure (strong positive), neutral (mild positive), discomfort (mild negative), and distress (strong negative) expressions. Thus, while the infant and adult faces were similar in terms of emotional valence and intensity, we did not aim to match the faces in terms of the specific emotional signals transmitted by the negative expressions (i.e., fear vs. distress). Indeed, it is argued that infants do not express discrete emotional signals (e.g., fear) similarly as adults during the first year (Camras & Shutter, 2010). It is noteworthy, however, that the adult fear expression may be likened to a distress cue in the sense that it initiates helping and prosocial behaviors (Marsh, Kozak, & Ambady, 2007), and both of these cues are associated with distinct ERP effects (Batty & Taylor, 2003; Proverbio et al., 2006). Eighteen models (9 females, 9 males) were chosen from the adult stimulus set, with each model showing all 4 expressions, each shown 4 times during the ERP recording. The identity variation was larger in the infant stimulus set because none of the infants appeared in all expression categories. From the original stimulus database of 208 images we sampled 18 representative exemplars to each expression category and excluded those stimuli in which the model's head was averted or the model's age or expression intensity appeared to deviate from other images within the

category. With a viewing distance of 60 cm, the faces subtended 11° and 7° horizontally and vertically, respectively.

Laboratory Procedure

EEG was recorded while adult and infant faces were presented in a random order. To manipulate attentional focus, two separate counting tasks were administered with the instruction to silently count the number of either adult or infant faces within each stimulus block, irrespective of the emotional expressions. A total of 144 adult and 144 infant faces were presented in each counting task, adding to a total of 576 trials presented during the ERP recording. Both counting tasks were divided to 4 stimulus blocks consisting of 72 faces. The number of attended faces (adult or infant) ranged from 32 to 40 within each block. The participant was asked to report the number of attended faces after each block of 72 faces before the next block and to start counting from zero in the beginning of each block. After 4 blocks, a short break was allowed and the participant was instructed to count faces of the other age condition during the next 4 blocks. The stimuli were presented on a black background for 1000 ms each with a 700-ms blank screen inter-stimulus interval. Presentation order of the counting tasks (adult and infant) was counterbalanced across participants within each group (i.e., mothers and non-mothers) separately. Completing the ERP tasks lasted ca. 20 min.

After the ERP recording, the electrode cap was removed and a brief (5 min) behavioral two-alternative forced choice facial expression recognition task with adult and infant stimuli was administered. Adult and infant faces were presented separately in two blocks whose presentation order was the same as the counting order during the ERP task. In one block, a total of 72 adult or infant faces (i.e., 18 stimuli for each expression) were presented on a black background for 200 ms each. The instruction was to respond as quickly as possible whether the facial expression was emotionally negative (i.e., strong and mild negative expressions) or positive/neutral (i.e., strong and mild positive expressions). Responses were made with the mouse buttons and the order of the

response buttons was counterbalanced across participants. Following response, the next stimulus was presented after a 1000 ms blank screen inter-trial interval.

Acquisition and Analysis of the ERPs

For the first 52 participants, continuous EEG was recorded using a 64-channel electrode cap (Quik-Cap; Neuroscan, El Paso, TX). Due to damage to the electrode cap, EEG from the remaining 42 participants was recorded using a 21-channel cap (Electro-Cap; Electro-Cap International, Eaton, OH). The type of electrode cap was uniformly distributed across mothers and non-mothers, $\chi^2(1) = 2.30$, $p > .13$, and the *OXTR* genotypes, $\chi^2(2) = 0.19$, $p > .91$. In all measurements, the left mastoid served as an on-line reference. Vertical (VEOG) and horizontal (HEOG) electrooculogram was monitored bipolarly from sites above and below the left eye and beside the outer canthus of each eye. Electrode impedances were generally below 10 kΩ in the beginning of the recording. The EEG was band-pass filtered (0.05-200 Hz), amplified with a gain of 5000, and stored at a sample rate of 1000 Hz. An independent component analysis (ICA) was used to detect and remove the independent components produced by eye blinks. ICA was performed using the logistic infomax ICA algorithm included in the EEGLAB toolbox (Makeig, Bell, Jung, & Sejnowski, 1996). Removal of bad channels was performed by using visual inspection and by automated rejection of high-amplitude or improbable channels (probability threshold > 5 SD) using the EEGLAB toolbox. Next, epochs with EEG amplitude ≥ 150 μV were rejected resulting in an average inclusion of 33.8 epochs per experimental condition (93.9 % of all epochs, *SD* = 8.2 %). The number of included epochs was similar between mothers (*M* = 33.9, *SD* = 3.0) and non-mothers (*M* = 33.6, *SD* = 3.0), $F(1, 185) = .50$, $p = .48$. Continuous EEG signal was filtered off-line using a 30-Hz lowpass filter and segmented to 1000-ms epochs with a 200-ms prestimulus baseline. For the analyses of the transient components in the 80-210-ms latency range (N1 and N170), a 1-Hz highpass filter was further applied. The epochs were baseline-corrected against the mean voltage during the 200-ms baseline and average waveforms for each individual participant within each

experimental condition were calculated. Average waveforms were then re-referenced to an average of all recording channels excluding HEOG and VEOG. Finally, to accommodate the two different electrode caps to the same statistical analyses, the EEG data were reduced to averages from distinct electrode sites in the anterior and posterior scalp regions which covered equivalent cortical areas across the two electrode montages. The channel sets are overlaid on scalp topographies of ERPs in Figure 1.

Time windows for the extraction of different ERP components were determined by their temporal occurrence in the grand average waveforms. From the frontal channel set, the amplitude and latency of the N1 component was measured by determining the minimum peak amplitude within a time window of 80-150 ms post-stimulus. The N170 component was the minimum peak amplitude within 120-210 ms post-stimulus in the posterior channel set. The latency and the amplitude from the N1 and the N170 components were extracted using an automated peak-picking algorithm. Early posterior negativity (EPN) waveform is the subsequent negative shift following the positive deflection peaking sharply after the N170 component and it was calculated as the mean activity between 240-300 ms in the posterior channel set. More negative mean amplitudes of the EPN waveform correspond with increased attention (cf. Leppänen et al., 2007; Schupp et al., 2004; Schupp et al., 2007). Finally, the LPP waveform was determined as the mean activity between 300-450 ms in the posterior channel set, with more positive amplitudes corresponding with increased attention (Bradley, 2009; Olofsson, Nordin, Sequeira, & Polich, 2008).

Analysis of the Behavioral Data

From the facial expression recognition data, responses with reaction times (RTs) exceeding 5000 ms were first discarded as outliers (4.9 % of trials). Accuracy was then calculated as the percentage of correct responses out of the total number of responses separately in each stimulus condition. For RT analyses, a further filtering step was applied to remove correct responses

with RTs exceeding the time window of individual mean RT ±2 standard deviations. The mean RT in each stimulus condition was then calculated for each participant.

Statistical Analyses

The statistical analyses were run separately for adult and infant faces. In all results reported below, participant age did not influence the observed statistical effects. First, we compared the ERP responses and behavioral emotion recognition performance between mothers and non-mothers. For the ERP components, the amplitude (all components) and latency (N1 and N170) scores were analyzed with a repeated measures ANOVA with Attention (attended, unattended), Valence (positive, negative), Intensity (mild, strong), and Hemisphere (left, right) as within-subjects factors and Motherhood (mothers, non-mothers) as a between-subjects factor. The behavioral accuracy and RT scores were analyzed with Valence and Intensity as within-subjects factors and Motherhood as a between-subjects factor. In the *OXTR* genotype analyses, the same set of analyses was run with Genotype (GG, non-GG) replacing Motherhood as the between-subjects factor. The analyses of the associations between ERPs and behavioral data (accuracy and RT) were confined only to the infant facial expression recognition data due to the lack of group effects on ERPs to adult faces (see below). RT was initially entered as a continuous covariate in the ANOVA. Subsequently, a median split was used to divide the participants into low vs. high accuracy and slow vs. fast RT groups to allow for delineating the direction and size of the effects within groups.

Results

ERPs to Infant Faces: Mothers vs. Non-mothers

A Motherhood × Intensity × Hemisphere interaction was observed in the frontal N1 latency data, $F(1, 90) = 5.60$, $p < .05$, due to mothers' N1 latency being shorter to strong (113.0 ms) vs. mild (115.9 ms) intensity infant faces in the right hemisphere, $F(1, 45) = 8.73$, $p < .01$, partial $\eta^2 = .16$, while no intensity modulation of N1 latency was observed in non-mothers (Figure 2a). No significant age-independent interactions involving Motherhood were observed in the N1 amplitude

data. At the level of the N170 component, a Valence × Attention interaction in the amplitude data was found, $F(1, 80) = 9.38$, $p < .01$. Replicating the main effect from Proverbio et al. (2006), the N170 amplitude elicited by infant faces during focused attention was larger to negative (-3.0 μV) vs. positive (-2.8 μV) expressions regardless of maternal status, $F(1, 92) = 11.33$, $p < .001$, partial $\eta^2 = .11$. In the 240-300-ms time window corresponding to the EPN waveform, we observed a Motherhood × Valence × Attention interaction, $F(1, 90) = 4.70$, $p < .05$, due to the EPN being enhanced (i.e., more negative) by negative (5.7 μV) as opposed to positive (6.0 μV) valence, $F(1, 46) = 8.32$, $p < .01$, partial $\eta^2 = .15$, in mothers when infant faces were attended (Figure 3). There were no effects of Valence or Intensity on EPN in mothers when infant faces were unattended or in non-mothers during both counting tasks, *p*s > .27. Finally, no interactions involving motherhood were observed in the LPP amplitude data.

Figures 1, 2, and 3 here

ERPs to Infant Faces: *OXTR* Genotype

A significant Genotype × Attention × Intensity interaction was observed in frontal N1 latency, $F(1, 82) = 4.93$, $p < .05$ (Figure 2b). This was due to a shorter latency to strong (110.5 ms) vs. mild (113.3 ms) intensity infant faces in the GG carriers when infant faces were attended, $F(1, 32) = 5.38$, $p < .05$, partial $\eta^2 = .14$, whereas no intensity modulation of N1 responses was observed in non-GG carriers, $F(1, 51) = 1.90$, $p > .18$. No other effects of *OXTR* genotype on ERPs were found.

ERPs to Adult Faces

None of the differences between mothers and non-mothers or *OXTR* genotypes elicited by infant faces (i.e., N1 and EPN) were paralleled by similar interactions in the adult stimulus data[2], highlighting the infant-specificity of the effects of motherhood and *OXTR*.

Across participants, main effects of Valence were observed in frontal N1 amplitude, $F(1, 90) = 8.82$, $p < .01$, partial $\eta^2 = .09$ (larger amplitude to negative [-2.2 μV] than to positive [-2.1 μV] faces), N170 latency, $F(1, 90) = 7.03$, $p < .01$, partial $\eta^2 = .07$ (longer latency to negative [162.6 ms] than to positive [161.5 ms] faces), and EPN amplitude, $F(1, 90) = 13.51$, $p < .001$, partial $\eta^2 = .13$, (increased negativity to negative [6.4 μV] as opposed to positive [6.7 μV] faces). Likewise, Intensity showed main effects in N170 latency, $F(1, 90) = 4.74$, $p < .05$, partial $\eta^2 = .05$ (longer latency to strong [162.6 ms] than to mild [161.5 ms] intensity faces), N170 amplitude, $F(1, 90) = 6.34$, $p < .05$, partial $\eta^2 = .07$ (larger amplitude to strong [-3.0 μV] than to mild [-2.8 μV] intensity faces), EPN amplitude, $F(1, 90) = 32.72$, $p < .001$, partial $\eta^2 = .27$ (increased negativity to strong [6.4 μV] as opposed to mild [6.8 μV] intensity faces), and LPP amplitude, $F(1, 90) = 34.96$, $p < .001$, partial $\eta^2 = .28$ (larger amplitude to mild [5.3 μV] than to strong [4.8 μV] intensity faces).

Emotion Recognition Performance

Main effects of valence and intensity emerged both in the accuracy and the RT data, *p*s < .01, indicating that positive valence and stronger intensity were related to more accurate and faster responding to both adult and infant stimuli. No main or interaction effects involving Motherhood or *OXTR* genotype were observed in the accuracy and the RT data of both adult and infant stimuli.

ERPs and Emotion Recognition Performance

Using the RT to infant faces as a covariate, an RT × Intensity interaction emerged in the frontal N1 latency data, $F(1, 89) = 5.05$, $p < .05$, partial $\eta^2 = .05$. This was due to a shorter N1 latency to strong (111.7 ms) vs. mild (113.6 ms) intensity infant faces in the fast-RT group (i.e., whose RTs to recognize the infant stimuli were at or below the median), $F(1, 45) = 5.44$, $p < .05$, partial $\eta^2 = .11$, whereas no modulation of N1 latency by intensity was observed in the slow-RT group, $F(1, 44) = 0.03$, $p > .86$. ERPs and RTs were also associated in terms of a linear correlation between the N1 latency effect (i.e., mean frontal N1 latency in the strong intensity condition minus

latency in the mild intensity condition) and mean behavioral RT to infant stimuli, $r = .26$, $p < .05$, $N = 92$. No corresponding correlation was observed in case of adult stimuli, $r = .07$, $p > .52$, $N = 93$. The interaction was further qualified by a three-way interaction between RT, Intensity, and Motherhood, $F(1, 87) = 7.18$, $p < .01$. This effect emerged from strong (113.8 ms) vs. mild (116.6 ms) intensity infant faces shortening the N1 latencies in slow-RT mothers, $F(1, 20) = 6.76$, $p < .05$, partial $\eta^2 = .25$, but not in fast-RT mothers, $F(1, 24) = 1.00$, $p > .33$, who tended to show overall shorter N1 latencies than slow-RT mothers, (112 vs. 116 ms). Regarding emotion recognition accuracy, an interaction between Accuracy, Valence, and Motherhood was observed in posterior LPP amplitudes (Figure 4) to infant faces, $F(1, 84) = 4.67$, $p < .05$. The more accurate mothers showed a marginally larger LPP amplitude to negative (5.7 μV) than to positive (5.5 μV) infant expressions, $F(1, 27) = 4.13$, $p = .052$, partial $\eta^2 = .13$, whereas mothers with accuracy scores below the median showed a larger LPP amplitude to positive (4.6 μV) as opposed to negative (4.2 μV) infant expressions, $F(1, 15) = 5.72$, $p < .05$, partial $\eta^2 = .28$. No effects of accuracy on LPP amplitudes were observed in non-mothers, ps > .27.

Figure 4 here

Discussion

Testing the associations of motherhood and oxytocin receptor genetic variation with neural and behavioral responses to emotional expressions of infants and adults, it was found that mothers (vs. non-mothers) and individuals carrying the rs53576 GG variant of the *OXTR* gene (vs. A-carriers) showed enhanced ERP differentiation of infants' strong vs. mild intensity facial expressions (i.e., pleasure and distress vs. comfort and discomfort). Both motherhood and *OXTR* modulated the latency of the early N1 response measured over frontal electrode sites, with faster-peaking responses to strong vs. mild intensity infant faces in mothers and participants with the GG

genotype. In mothers, the posterior EPN waveform was further enhanced by infant faces displaying negative valence. Furthermore, across participants, the peak latency of the frontal N1 response to strong vs. mild intensity infant faces correlated with the speed of correctly classifying infant faces as positive or negative in a separate task. The pattern of findings was highly selective to infant facial expressions and none of these ERP effects related to motherhood or *OXTR* polymorphism were paralleled by similar effects in response to adult facial expressions. At group level, ERPs to adult expressions showed effects that were generally consistent with previous research showing enhanced ERP responses to adult faces depicting negative valence or strong vs. mild intensity (e.g., Batty & Taylor, 2003; Leppänen et al., 2007; Schupp et al., 2004).

The ERP data are in line with the view that mothering young infants is associated with enhanced sensitivity to encode variations in infant affective signals (cf. Rilling, 2013). Only mothers showed differential early ERPs in frontal areas (N1) in response to infant facial expressions and this early modulation was not dependent on whether attention was focused on infant or adult faces. The spatiotemporal pattern of the N1 response is reminiscent of the infant-specific orbitofrontal response observed by Kringelbach et al. (2008), which was suggested to have a role in a rapid "tagging" of infant faces as motivationally significant and subsequent top-down amplification of later visual and attentional processing in the occipitotemporal areas (Bar et al., 2006; Barrett & Bar, 2009; Kringelbach et al., 2008). In our data, mothers also showed a differential posterior EPN response starting ~240 ms after stimulus onset in occipitotemporal areas to negatively valenced infant faces when the task required focusing attention on infant faces. The EPN is consistently associated with attention to emotionally salient (Leppänen et al., 2007; Schupp et al., 2007) as well as task-relevant (Hillyard & Anllo-Vento, 1998) stimuli. In contrast to Proverbio et al. (2006), we did not observe a main effect of motherhood on later positive components (LPP). Whether the absence of LPP modulation reflects task-related differences between the studies (counting vs. emotion recognition task) or some other factor is unclear. Overall, the results

nevertheless suggest that motherhood is associated with enhanced processing of variations in infants' emotional expressions during early (N1) and later (EPN) stages of visual processing.

An interesting question relates to the functional significance of the rapid ERP modulations, i.e., whether they are associated with variation in mothers' sensitive responsiveness to infants' affect signals. Our data are consistent with this possibility in showing the peak latency of the frontal N1 response correlated with the speed of behavioral classification of infant faces as positive or negative. Further research is required to examine the replicability of this result and to determine the relationship between this neural marker and more naturalistic measures of parental sensitivity.

The effects of rs53576 variation in the *OXTR* gene paralleled the effects of motherhood in showing that GG homozygotes and A-allele carriers exhibited differences in frontal N1 latency in response to strong vs. mild intensity infant expressions. Thus, the sensitivity to variation in infants' expression intensity that may be accomplished through experience in mothers may likewise emerge through naturally occurring genetic variation in the efficiency of oxytocinergic transmission also in nulliparous females not having extensive experience of interacting with infants. The effects of *OXTR* variation were more modest, however, in that the N1 latency effect observed in GG homozygotes was contingent on focused attention whereas in mothers the modulation occurred irrespective of attentional focus, and genotype had no effects on other ERP components. The data nevertheless raise the possibility that similarly as motherhood, the rs53576 GG variant of the *OXTR* polymorphism putatively related to more efficient oxytocinergic signaling yields rapid infant-specific effects on frontal activity that may be linked to coarse detection of emotional significance by the OFC (Kringelbach et al., 2008). The absence of similar effects in response to adult faces fits with the pivotal role of OT in promoting parental as well as alloparental affiliative behaviors toward infants and children (Bakermans-Kranenburg & van IJzendoorn, 2008; Marsh et al., 2012; Ross et al., 2009).

The *OXTR* data can be seen to contradict previous studies showing an enhancement of behavioral (Lischke et al., 2012; Van IJzendoorn & Bakermans-Kranenburg, 2012) and ERP (Huffmeijer et al., 2013) responses to adults' emotional expressions following intranasal OT administration as no direct effects of *OXTR* on responses to adult stimuli were observed. It may well be, however, that the effects of intranasal OT administration on social information processing are considerably larger than the direct effects of natural genetic variation of OT. Indeed, recent meta-analytic evidence indicates that whereas intranasal OT administration produces significant effects of on emotion recognition (Van IJzendoorn & Bakermans-Kranenburg, 2012), the direct effects of *OXTR* variation on different domains of social behavior tend to be inconsistent and often small in magnitude (Bakermans-Kranenburg & van IJzendoorn, in press). We believe that the neural responses to children's emotional signals represent a promising candidate to study the effects of *OXTR* variation on social behavior. Across mammals, the most prominent function of OT is to mediate parenting behaviors (Carter, 1998) and from this perspective, ERPs to infant communicative and emotional signals can be considered to reflect the elementary steps in the information processing stream upon which more complex parent-child interactive processes are likely build.

Although the effects of *OXTR* on ERPs to infant faces are in line with the established role of OT in early parenting behaviors, it should be pointed out that one of the major limitations of the present study is the focus on a single genetic variant. The direct effects of individual genetic polymorphisms on social information processing are likely to be small and dependent on interactions with multiple genetic variations across the genome. In the future, independent replications should analyze the cumulative effects of different genetic variants affecting central OT transmission as well as the effects of genetic variants affecting other (e.g., serotonergic) neurotransmitter systems implicated in social information processing to test whether the processing of emotional information from infant signals is most strongly associated with OT system genes.

Secondly, as OT levels are associated with lactation (White-Traut et al., 2009; but see Feldman et al., 2012), and breastfeeding could thereby influence social behaviors for which OT is important, another limitation of the study is the lack of information on whether or not the mothers were still breastfeeding at the time of the study. Therefore, we cannot rule out the possibilities of breastfeeding-related variation in ERP and behavioral responses within the mothers or potential interactions between breastfeeding and *OXTR* genotype in modulating the *OXTR* effects. Thirdly, due to the small subgroups, no reliable analyses of the potential interactions between motherhood and *OXTR* genotype could be conducted on the present data. An important goal for future research is to investigate the relative contribution of *OXTR* genetic variation and motherhood in the neural response to infant cues.

In conclusion, the present study showed that both motherhood and the GG variant of the *OXTR* gene are associated with enhanced neural responses specifically to emotionally evocative infant – but not adult – facial expressions. The early frontal ERP modulation was also sensitive to variation in behavioral recognition speed of infant faces, providing a neural marker most consistently related to the efficiency of processing emotional signals from infant faces. In future, a larger sample of mothers is needed to determine whether the behavioral effects of oxytocinergic variation in mothers (Bakermans-Kranenburg & van IJzendoorn, 2008; Feldman et al., 2007) are linked to differential neural responses to infants' emotional signals. Also important will be to extend similar correlational analyses to more naturalistic measures of maternal sensitivity and include mothers from a sufficient range of the sensitivity spectrum.

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Author notes

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Footnotes

[1]Neutral instead of mild (i.e., closed mouth) happy expressions were selected for this category to match the adult stimuli with the expressionless (or neutral) facial expressions in the infant mild positive emotion category.

[2]The age of the stimulus face was not included as a factor in the original analyses given various differences in the infant and adult stimuli. When the three interactions observed in the infant stimulus data (i.e., Motherhood × Intensity × Hemisphere, Motherhood × Valence × Attention, and Genotype × Attention × Intensity) were tested by including Face Age (adult vs. infant) as an additional factor, marginal interactions were found between Face Age and the above three effects (p = 0.055, p = 0.10, and p = 0.052, respectively), suggesting specificity of the observed effects for infant facial expressions. Furthermore, none of these effects were significant in the ERPs elicited by adult stimuli (p = .77, p = .74, and p = .58, respectively).

Figure Captions

Figure 1. Topographical plots of the N1, N170, EPN, and LPP responses to infant faces (averaged across participants). The channels used in extracting the responses are overlaid with rectangles on each plot. The N1 was derived from anterior channels (top row) and the N170, EPN, and LPP components from posterior channels.

Figure 2. Modulation of the N1 latency by expression intensity of infant stimuli in A) mothers vs. non-mothers (data from the right hemisphere) and B) different *OXTR* rs53576 allele carriers (data averaged across hemispheres). Error bars indicate the standard error of mean.

Figure 3. Event-related potentials elicited by infant stimuli in the attended condition (i.e., where participants counted the infant faces) in mothers and in non-mothers derived from the posterior electrodes. LH and RH denote the left and right hemisphere, respectively. IMN: infant mild negative; ISN: infant strong negative; IMP: infant mild positive; ISP: infant strong positive.

Figure 4. Event-related potentials elicited by infant stimuli in accurate (left) and less accurate mothers (right) derived from the posterior electrodes. Data is aggregated across task conditions as well as the left and the right hemispheric electrode locations. ERPs elicited by negative and positive facial expressions are indicated with blue and red lines, respectively.

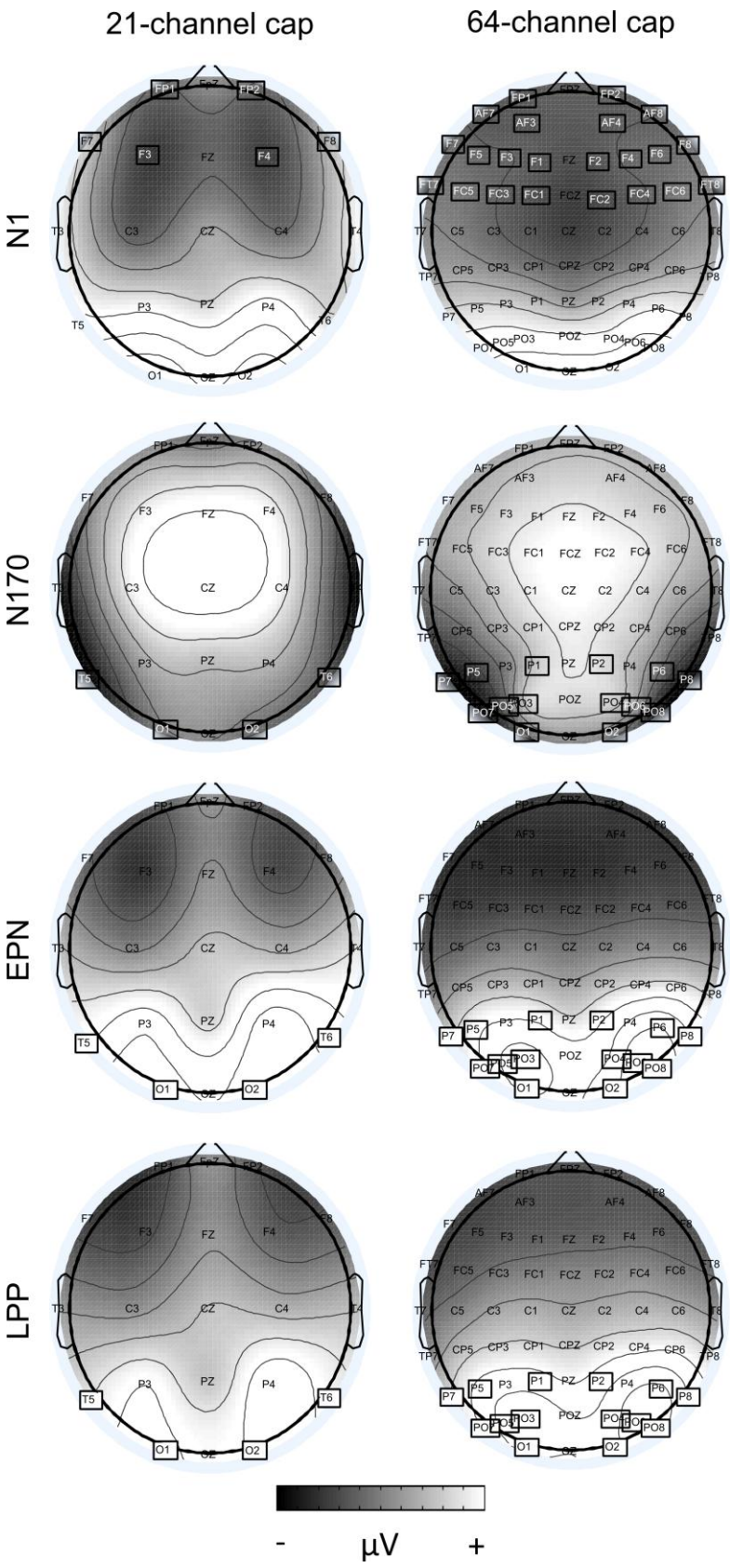

Figure 1.

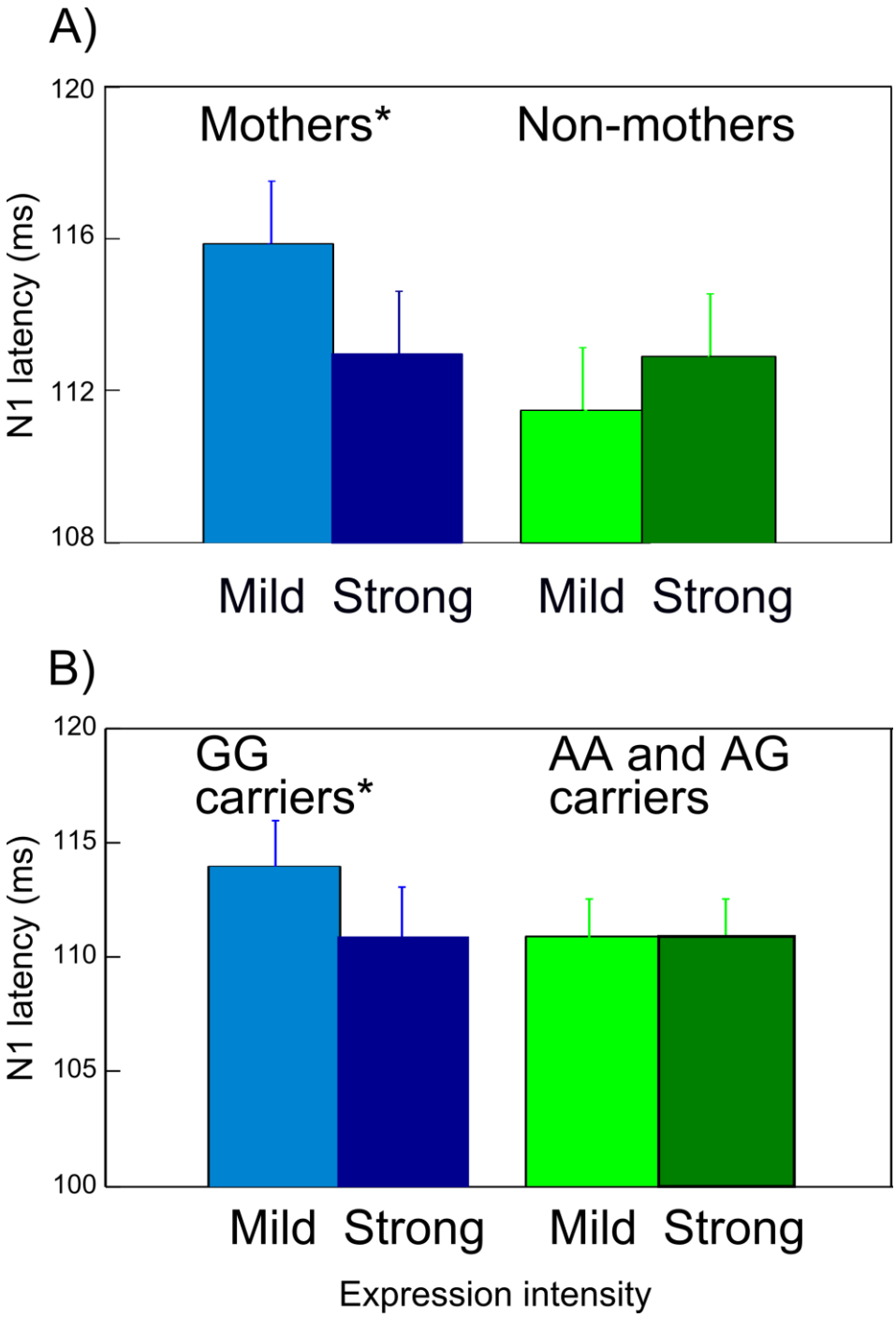
A)
120
116
112
108
N1 latency (ms)
Mothers*
Non-mothers
Mild
Strong
Mild
Strong
B)
120
115
110
105
100
N1 latency (ms)
GG carriers*
AA and AG carriers
Mild
Strong
Mild
Strong
Expression intensity

Figure 2.

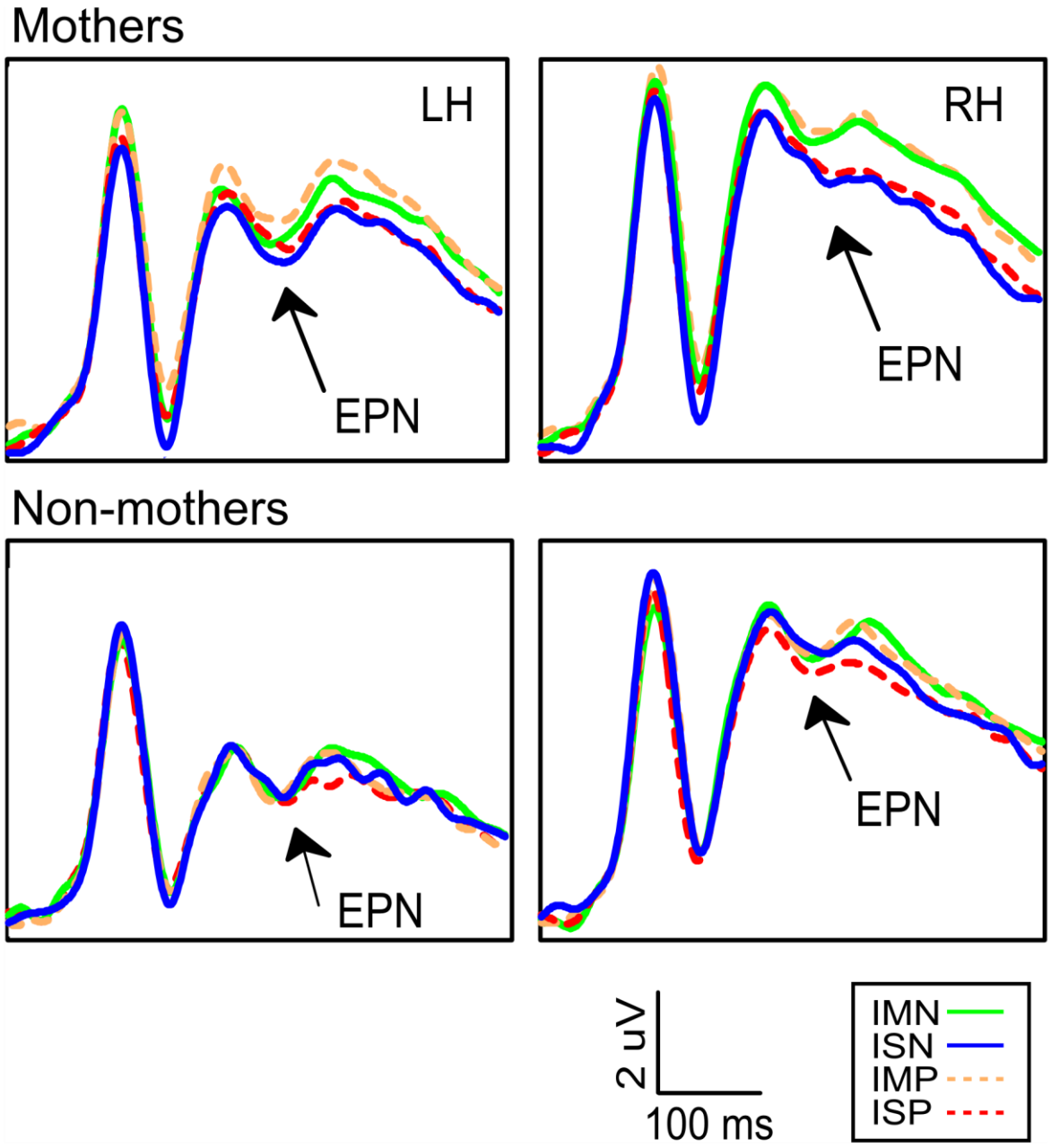
Mothers
LH
EPN
RH
EPN
Non-mothers
EPN
EPN
2 uV
100 ms
IMN
ISN
IMP
ISP

Figure 3.

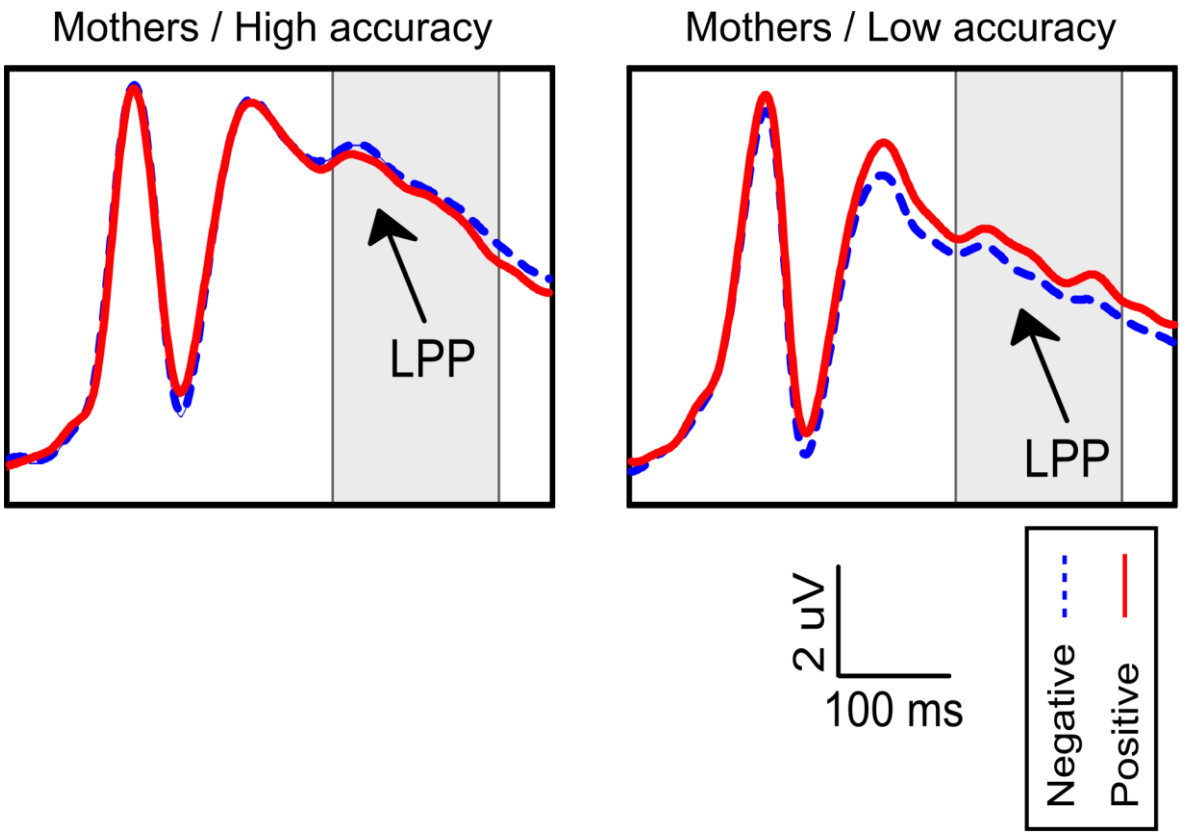
Mothers / High accuracy
Mothers / Low accuracy
LPP
LPP
2 uV
100 ms
Negative
Positive

Figure 4.