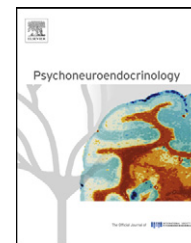




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Intranasal oxytocin enhances emotion recognition from dynamic facial expressions and leaves eye-gaze unaffected

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Summary Previous studies have shown that oxytocin improves the encoding and recognition of facial expressions, which has been proposed to be mediated by an increased exploration of the eye region during face processing. In the present study, we used eye tracking to assess visual attention to the eye region while participants performed a dynamic facial emotion recognition task. In a double-blind, placebo-controlled between-subjects design participants received 24 IU intranasal oxytocin ($n = 23$) or a placebo ($n = 24$). Although oxytocin administration had no effect on participants' visual scanning of emotional faces, it generally enhanced recognition performance, as the oxytocin group recognized emotional expressions at lower intensity levels. These findings suggest that oxytocin-induced improvement of facial emotion recognition is independent of modulations in overt visual attention.

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1. Introduction

The neuropeptide oxytocin (OT) is well known for its fundamental role in the regulation of social behavior and social

cognition in humans (Heinrichs et al., 2009). Considering that the ability to decode another's facial expression is a prerequisite for social interaction, it is not surprising that OT improves the recognition of emotional expressions in static (Domes et al., 2007; Di Simplicio et al., 2009; Guastella et al., 2010; Marsh et al., 2010; Schulze et al., in press) and dynamic (Fischer-Shofty et al., 2010) images of faces; although it remains inconclusive whether emotion recognition is specifically improved for positive (Di Simplicio et al., 2009; Marsh et al., 2010) or negative expressions (Fischer-Shofty et al., 2010). Other studies have shown that enhanced exploration of the eye region is likely to improve emotion

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recognition because this region conveys the most relevant cues for emotion discrimination (Schyns et al., 2002, 2007, 2009; Smith et al., 2005). Based on this, it seems possible that OT might improve emotion recognition by directing attention to the eyes. Although the proposed mediating effect of visual attention for the positive effect of OT on emotion recognition has yet not been tested explicitly, it is supported by the finding that OT increases gaze to the eye region of neutral (Guastella et al., 2008a; Andari et al., 2010) and emotional expressions (Gamer et al., 2010) and that OT enhances the ability to infer an opponent's mental state from subtle cues around the eye region (Domes et al., 2007; Guastella et al., 2010). Although the neural mechanisms for the influence of OT on attention to the eyes remain to be investigated, recent data suggests the involvement of the amygdala in attention to facial features in general (Adolphs and Spezio, 2006; Gamer and Buchel, 2009) and a role for the superior colliculi in the modulation of attention to facial features by OT (Gamer et al., 2010). Encouraged by these prior results, we assessed overt visual attention using eye tracking during a dynamic emotion recognition task, which allowed to directly investigate whether visual attention mediates the effects of OT on facial emotion recognition. We hypothesized that OT would generally promote emotion recognition from dynamic facial expressions and would enhance visual attention to the eye region of these dynamic facial expressions. In addition, we expected a positive association between attention to eye region and emotion recognition performance.

2. Materials and methods

2.1. Participants

In a double-blind placebo-controlled between-subjects design, forty-seven healthy male adults (age: $M = 26.09$, $SD = 3.41$) were randomly assigned to receive a nasal spray either containing 24 international units (IU) of OT ($N = 23$; Syntocinon Spray, Novartis, Basel, Switzerland) or placebo ($N = 24$; containing all ingredients except for the neuropeptide). Randomization was done by the compounding pharmacist. There were no differences between the groups in age, general intelligence, self-reported empathy and psychopathology [see supplementary Table S1, all $p > .23$]. Exclusion criteria for all participants were physical or mental illness, use of medication, substance abuse and smoking. All participants provided written, informed consent and were paid for participation. The study was carried out in accordance with the Declaration of Helsinki and was approved by the ethical committee of the University of Rostock.

2.2. Experimental procedure

Following a standardized protocol (Domes et al., 2010), the participants self-administered the nasal spray (three puffs per nostril, each containing 4 IU of OT or placebo) 45 min before the start of the emotion recognition task (Born et al., 2002). Substance-induced changes in mood, calmness and wakefulness were tracked by administering a multidimensional mood questionnaire, the MDBF (Steyer et al., 1997), directly before substance application and directly before the

emotion recognition task. After the experimental session, self-report revealed that participants were not able to discriminate OT and placebo. In addition, participants did not report any side-effects of drug administration in the present study. After arrival at the laboratory, a venous catheter was inserted into the participants' forearm (30 min before OT was administered). In order to control for differences in OT resorption, blood samples were drawn 5 min before and 45 min after substance application into 5 ml EDTA vacuatainer tubes. Immediately after collection, the samples were cooled in ice-chilled water at 4 °C, 15 min later centrifuged at 4000 rpm (for 5 min at 4 °C) and finally stored in a freezer at -20 °C. After completion of the study, the samples were shipped on dry ice at -20 °C to the Department of Behavioral and Molecular Neuroendocrinology at the University of Regensburg, Germany, where they were extracted and analyzed using a radioimmunoassay (Landgraf, 1981). The assay detection limit was 0.1 pg/sample, cross-reactivity with other related neuropeptides was <0.7%, intra-assay precision was 7–10% and inter-assay variability was eliminated by measuring all samples within the same assay.

2.3. Emotion recognition task

Forty-five minutes after substance application, the participants performed the emotion recognition task. The task involved the use of computer-manipulated images of faces, whose neutral expression was gradually and continuously changing into an emotional one (see Domes et al., 2008). The images were selected on basis of a validation study from the FACES database (Ebner et al., 2010) and consisted of six young male and six young female individuals, each depicting a neutral, sad, angry, fearful and happy expression.

Adobe Photoshop CS4 (Adobe Systems Inc., San Jose, CA, USA) and Matlab 7.7 (MathWorks Inc., Natick, MA, USA) were used to convert the images into gray-scales and equalize them in size. Finally, the cumulative brightness was normalized across all images using an in-house script written in MATLAB. Thereafter, each image was enclosed within an elliptic mask (411 × 570 pixel) that only revealed the face itself. Winmorph 3.01 (<http://www.debugmode.com/winformorph/>) was used to morph each emotional expression with a neutral expression in 5% increments, resulting in expressions varying in emotional intensity from 0% (neutral) to 100% (emotion). For each individual face four sets (sad, angry, fearful, and happy expression) were obtained, each containing 21 images. Each image of a set of expressions was presented for 800 ms, ranging from 0% to 100% of intensity. Each image was presented on a 20-in. computer screen (screen size: 30.6 cm × 40.8 cm; resolution: 1024 × 768 pixel) against a gray background at a viewing distance of 55 cm (vertical/horizontal visual angle of approx. 17°/23.5°).

Participants were instructed to press a stop button as soon as they recognized the expression of the face. After stopping the presentation, the face remained visible on the screen and participants had to identify the particular expression by making a forced-choice between four possible emotion labels (happy, sad, angry and fearful). No time limit was given for the choice of the emotion label. Number of correct answers and intensity levels at which the presentation was stopped in correct trials only were used as the dependent variables.

2.4. Eye tracking

During the emotion recognition task, participants' eye movements were recorded with a remote infra-red eye-tracker with a chin-rest (PC-60 Head Fixed; Arrington Research Inc.; Scottsdale, AZ, USA). Raw data were collected at 60 Hz sampling rate with a spatial resolution of approx. 0.15° and $0.25\text{--}1.0^\circ$ for gaze position accuracy. Eyetracking data were analyzed with ILAB (Gitelman, 2002). After smoothing of the raw data with a Gaussian filter, blink and artifact detection was performed. Three participants of the OT group and one of the placebo group had to be excluded from eye tracking analyses because of more than 20% lost data. There were no differences between the remaining groups with regard to valid data (all $p > .18$). Fixations were coded when gaze remained for at least 100 ms within an area with a diameter of 1° . Image-specific templates were used to specify the whole face (elliptic template with 411×570 pixels), the eye region (rectangular template of 354×123 pixels) and the mouth (rectangular template of 223×87 pixels). For the whole trial, we calculated average percent fixation duration to the whole face relative to the screen, and average percent fixation duration to the eye region and the mouth region relative to the whole face. In order to investigate the initial allocation of attention, we calculated relative fixation duration for the areas of interest (face, eyes, mouth) also for the first 1600 ms of each trial.

2.5. Statistical analyses

All statistical analyses were performed using SPSS 15 (SPSS Inc., Chicago, IL, USA). Effects of OT and emotional valence of the presented faces on recognition performance were tested with two-way repeated measures ANOVAs. Eye tracking data were subject to three-way repeated-measures ANOVAs. Greenhouse–Geisser correction was applied whenever the sphericity assumption was violated. Post hoc analyses of significant main effects and interactions included t -tests (two-tailed) and two-way ANOVAs respectively. Multiple moderated regression analyses were performed for each emotional category to test whether OT treatment influenced the association between gaze to the eye region and emotion recognition. The level of significance for all analyses was set at $p < .05$. In case of significant effects, the effect size measure f was reported according to Cohen (1988).

3. Results

3.1. Peripheral OT-levels and mental state

To test whether the participants in the OT and placebo group differed in OT-levels over the time course of substance application, a two-way repeated measures ANOVA (Group \times Time) was run [see supplementary Table S2; main effect Time: $F(1,45) = 2.46$, $p = .12$; main effect Group: $F(1,45) = 3.79$, $p = .06$; interaction Time \times Group: $F(1,45) = 26.94$, $p < .001$, $f = 0.76$]. OT application resulted in a significant increase in peripheral OT (pre: 24.6 ± 19.0 ; post: 39.7 ± 20.1), whereas placebo had no such effect (pre: 27.2 ± 16.6 ; post: 22.8 ± 16.8).

Participants' mood and calmness was unaffected by time and OT [see supplementary Table S2 mood—main effect Time: $F(1,45) = 2.49$, $p = .12$; main effect Group: $F(1,45) = 1.78$, $p = .19$; interaction Group \times Time: $F(1,45) = 3.02$, $p = .09$; calmness—main effect Time: $F(1,45) = 1.31$, $p = .26$; main effect Group: $F(1,45) = 0.32$, $p = .86$; interaction Group \times Time: $F(1,45) = 0.03$, $p = .86$]. The trends towards a Group \times Time interaction for mood was due to slightly lower post-application mood in the OT group (PL: 4.5 ± 0.4 ; OT: 4.2 ± 0.6 ; $t(45) = 1.92$; $p = .07$). Although participants' wakefulness declined throughout the experiment, OT administration had no effect [main effect Time: $F(1,45) = 38.94$, $p < .001$, $f = 0.92$; main effect Group: $F(1,45) = 1.55$, $p = .22$; interaction Group \times Time: $F(1,45) = 0.09$, $p = .77$].

3.2. Emotion recognition

A two-way repeated measures ANOVA (Group \times Emotion) demonstrated that OT application decreased intensity levels at which the expressions were recognized regardless of the particular type of emotion [main effect Group: $M \pm SD$; Placebo: 44.2 ± 8.2 ; OT: 39.1 ± 7.6 ; $F(1,45) = 4.86$, $p = .03$, $f = 0.32$; interaction Group \times Emotion: $F(1,135) = 1.32$, $p = .27$]. In general, participants' recognized happy expressions at a lower intensity level, moderate for angry and fearful expressions and low for sad expressions [main effect emotion: $F(3,135) = 77.30$, $p > .001$, $f = 1.31$]. Exploratory follow-up analyses suggested that OT may have specifically lowered the recognition threshold for angry [$t(45) = 2.67$, $p < .01$, two-sided] and fearful [$t(45) = 2.01$, $p = .05$, two-sided] expressions – Fig. 1 and supplementary Table S3.

Another two-way repeated measures ANOVA (Group \times Emotion) indicated that OT application had no effect on participants' recognition accuracy [main effect Group: $F(1,45) = 0.29$, $p = .59$; interaction Group \times Emotion: $F(2,25,101.23) = 1.54$, $\epsilon = 0.75$, $p = .22$], which was generally high for happy expressions, moderate for angry and fearful expressions and low for sad expressions [main effect Emotion: $F(2,25,101.23) = 38.46$, $\epsilon = 0.75$, $p < .001$, $f = 0.92$]. However, exploratory single comparison suggested that OT specifically enhanced participants' recognition accuracy for fearful expressions [$t(45) = 2.36$, $p = .02$] but not for the other expressions (all $p > .50$) – Fig. 1 and supplementary Table S3.

3.3. Visual attention

Initial allocation of attention to facial regions. A two-way repeated measures ANOVA (Group \times Emotion) showed that OT administration had no effect on the duration of initial fixations to the face [main effect group: $F(1,41) = 0.19$, $p = .67$; main effect emotion: $F(2,01,82.33) = 0.39$, $\epsilon = 0.67$, $p = .68$; Group \times Emotion: $F(2,01,82.33) = 1.27$, $\epsilon = 0.67$, $p = .29$]. A subsequent three-way repeated measures ANOVAs (Group \times Emotion \times Region) revealed that the duration [main effect group: $F(1,41) = 0.33$, $p = .57$; interaction Group \times Emotion: $F(3,123) = 0.84$, $p = .48$; interaction Group \times Emotion \times Region: $F(3,123) = 0.24$, $p = .87$] of initial fixations to the eye or mouth region were also unaffected by OT administration (Table 1).

Attention to facial regions over the whole trial. OT administration had no effect on the overall fixation duration

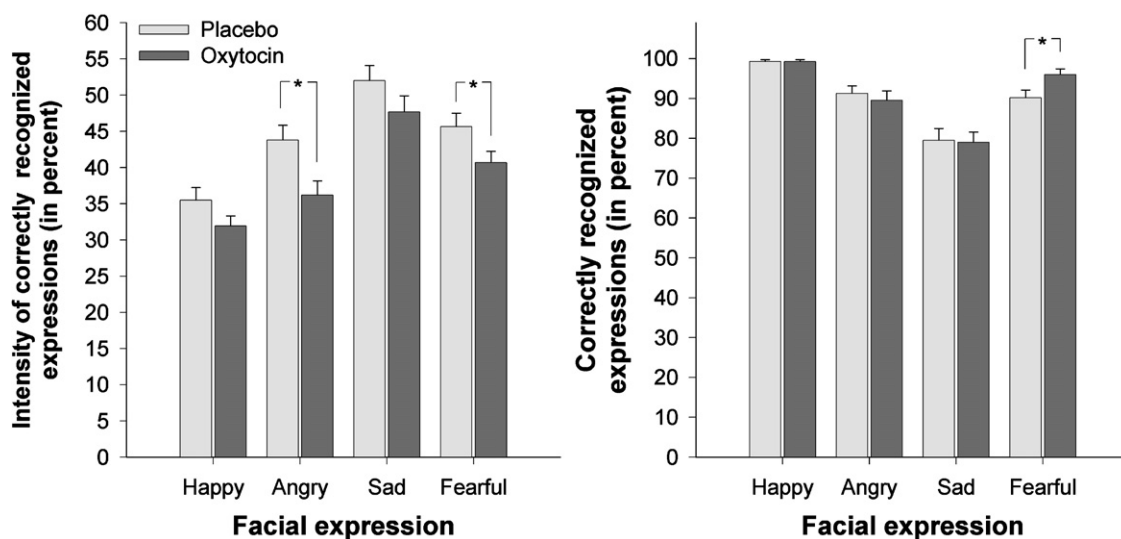


Figure 1 Intensity and number of correctly recognized facial expressions. Asterisks indicate significant post hoc single comparisons ($p < .05$, two-tailed). Bars present mean percent \pm standard error of mean.

to the whole face [main effect group: $F(1,41) = 0.10, p = .75$, main effect emotion: $F(1.96,80.19) = 0.24, \epsilon = 0.65, p = .78$; interaction Group \times Emotion: $F(1.96,80.19) = 0.78, \epsilon = 0.65, p = .46$]. Another three-way repeated measures ANOVA (Group \times Emotion \times Region) revealed that the overall duration of fixations to the eye or mouth region were also unaffected by OT administration [main effect group: $F(1,41) = 0.16, p = .70$; interaction Group \times Emotion: $F(3,123) = 0.76, p = .52$; interaction Group \times Emotion \times Region: $F(3,123) = 0.58, p = .63$] (Table 2).

Association between gaze to the eye region and emotion recognition. Separate multiple regression analyses for the different emotions displayed, we found a moderation of the association between fixation duration to the eye region and emotion recognition threshold for sad expressions

($T = -2.14; p < .05$). Fig. 2 illustrates this moderation: in the OT group there was a significant correlation between gaze to the eye region between the intensity at which sadness was recognized ($r = -.52$) whereas this association was absent in the placebo group ($r = .09$) – Fig. 2. No such moderation was found for the number of correctly identified expressions as the dependent variable.

4. Discussion

The first aim of the present study focused on the effects of intranasal OT on emotion recognition from dynamic facial expressions. In the present study, OT decreased the critical

Table 1 Visual attention during the initial 1600 ms of a trial.

	Placebo (N = 23)		Oxytocin (N = 20) ^a	
	M	SD	M	SD
Happy expressions				
Face/screen	.96	.12	.98	.04
Eye/face	.54	.16	.61	.15
Mouth/face	.17	.15	.15	.10
Angry expressions				
Face/screen	.97	.08	.99	.02
Eye/face	.67	.18	.69	.18
Mouth/face	.08	.08	.07	.11
Sad expressions				
Face/screen	.97	.07	.97	.06
Eye/face	.64	.24	.69	.19
Mouth/face	.08	.08	.07	.09
Fearful expressions				
Face/screen	.98	.03	.97	.07
Eye/face	.66	.15	.69	.17
Mouth/face	.11	.11	.06	.07

Table 2 Visual attention during the whole trial.

	Placebo (N = 23)		Oxytocin (N = 20) ^a	
	M	SD	M	SD
Happy expressions				
Face/screen	.96	.14	.97	.04
Eye/face	.44	.15	.50	.16
Mouth/face	.26	.12	.23	.14
Angry expressions				
Face/screen	.96	.08	.99	.03
Eye/face	.59	.20	.66	.16
Mouth/face	.13	.10	.09	.07
Sad expressions				
Face/screen	.96	.08	.97	.08
Eye/face	.60	.20	.63	.14
Mouth/face	.12	.08	.10	.08
Fearful expressions				
Face/screen	.98	.04	.96	.07
Eye/face	.62	.16	.63	.15
Mouth/face	.15	.09	.11	.09

^a Data from one participant of the placebo and three participants of the oxytocin group were excluded because of artifacts.

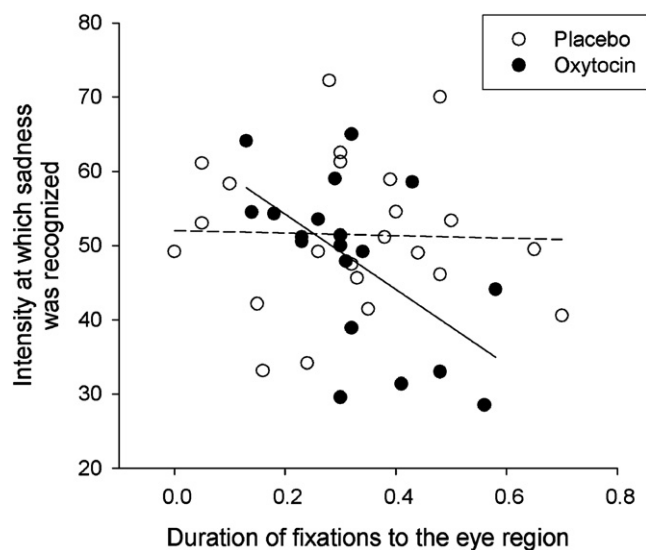


Figure 2 Linear association between fixation duration to the eye region and the intensity at which sadness was detected as a function of drug treatment. Lines represent linear regressions for the OT and the placebo group.

intensity at which participants were able to recognize emotional expressions from dynamic facial stimuli. In other words, participants who had received 24 IU OT prior to the emotion recognition task, were able to correctly recognize emotional expressions at a lower intensity from facial stimuli regardless of the specific valence, although the effect seemed to be pronounced for angry and fearful expressions. This effect was not due to more liberal responding, i.e., participants in the OT group did not respond earlier during the presentation of the dynamic stimuli for the price of lower accuracy. Accuracy in general was quite high in the present study and participants with OT tended to make even fewer errors for the fearful expressions than the control group, which is in line with previous studies (Fischer-Shofty et al., 2010). In contrast to previous studies which suggested specific effects of OT on the recognition of positive (Di Simplicio et al., 2009; Marsh et al., 2010) or negative emotional expressions (Fischer-Shofty et al., 2010), the present study supports a more general view: OT seems to enhance emotion recognition from dynamic facial expressions irrespective of the valence, although the effect was marginally larger for angry and fearful faces. Differences in task sensitivity might in part explain the divergent results. In contrast to previous studies with static stimuli, the present study mainly relied on recognition threshold rather than error rates. Hence, differences in item difficulty might have been less relevant as compared to tasks using static images. For example, previous studies have shown that the same task might differ in sensitivity to detect subtle effects of OT in different samples (Domes et al., 2007; Guastella et al., 2010). Another possible explanation might be differences in the stimuli used. Fischer-Shofty et al. (2010) presented sections of the eye region, and found a specific effect of OT on the accuracy of detecting fearful expressions. In contrast, we used whole faces to investigate the role of visual attention to different section of the face during emotion recognition, and found an overall effect of OT on the recognition threshold (mean intensity) in addition to slightly enhanced accuracy for fearful expressions. The eye region seems to play a crucial role in the

recognition of fear, and thus might have been the most sensitive stimulus category in the previous study. Moreover, since OT has been found to improve memory for faces and facial expressions (Guastella et al., 2008b; Rimmele et al., 2009), it could be possible that in the present study, OT enhanced memory for prototypical facial expressions and thus enhanced emotion recognition by improving the matching of the presented facial expression to previously memorized expressions.

The second aim of the present study was to test whether OT promotes facial emotion recognition by enhancing overt visual attention to the eye region of facial expressions. In all, there was no effect of OT on fixation duration for the eye region or the presented faces. The lack of an effect on visual attention contradicts previous studies which suggested that OT promotes eye gaze during face processing (Guastella et al., 2008a; Andari et al., 2010; Gamer et al., 2010). Differences in image presentation could explain the disparity of findings. In contrast to previous studies, which presented static neutral faces (Guastella et al., 2008a), static emotional faces (Andari et al., 2010) or emotional expression with very short durations (Gamer et al., 2010), we used dynamic stimuli, which introduce the aspect of increasing emotional intensity over the stimulus presentation. In addition, the presentation time of the stimuli on average was much longer in the present study. In static faces, the eyes might be especially salient because they convey crucial information about an individual's emotional state (Emery, 2000), and attention might be captured by the eyes because they are assumed to be the most informative part of the face (Adolphs et al., 2002). OT might be especially involved in the initial allocation of attentional resources to social cues. This interpretation is in line with the above mentioned study of Gamer et al. (2010), who showed reflexive saccades towards the eyes even for facial expressions presented for 150 ms. In dynamic stimuli, the effect of OT on the initial orienting probably becomes less relevant with increasing duration of the stimulus. Thus, OT might facilitate eye gaze in static rather than dynamic faces, in particular when the stimulus is presented briefly. It should be noted that there was

a moderate association between duration of eye gaze and sensitivity for the recognition of sadness in the OT group.

In light of the limited sample size, the statistical power is too low to draw definite conclusions. Using more sensitive measures of emotion recognition and/or more fine-grained eye tracking methods might be a promising approach to investigate the association between visual attention and emotion recognition and the modulation by OT. Future studies could additionally focus on whether the changing features of dynamic facial stimuli modulate visual attention over time, and whether OT modulates attentional capture by these changing features. Another limitation is the fact that we excluded female participants. Future study could explicitly investigate the sexual dimorphism of OT effects in the central nervous system as suggested by previous studies (Carter, 2007; Domes et al., 2010).

In sum, this is the first study that explicitly investigated the possible role of visual attention for the effect of OT on facial emotion recognition. Consistent with previous studies, which showed positive effects of OT on facial emotion recognition, the present results suggest that OT promotes emotion recognition from dynamic facial expressions, which resemble a more naturalistic interpersonal situation than static facial expressions. With regard to the role of visual attention as a possible mediating factor, the present results suggest that the positive effect of OT on emotion recognition might be independent of modulations of visual attention to specific facial regions, at least during the processing of the dynamic facial expressions.

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Conflict of interest

The authors declare that they have no conflicts of interest.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at [doi:10.1016/j.psyneuen.2011.07.015](https://doi.org/10.1016/j.psyneuen.2011.07.015).

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