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Measurement of the N170 during facial neuromuscular electrical stimulation (fNMES)

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ABSTRACT

Background: Studies on facial feedback effects typically employ props or posed facial expressions, which often lack temporal precision and muscle specificity.
New method: Facial Neuromuscular Electrical Stimulation (fNMES) allows for a controlled influence of contractions of facial muscles, and may be used to advance our understanding of facial feedback effects, especially when combined with Electroencephalography (EEG). However, electrical stimulation introduces significant interference that can mask underlying brain dynamics. Whether established signal processing methods can allow for a reduction of said interference whilst retaining effects of interest, remains unexplored.
Results: We addressed these questions focusing on the classic N170 visual evoked potential, a face-sensitive brain component: 20 participants viewed images of houses, and of sad, happy, and neutral faces. On half of the trials, fNMES was delivered to bilateral lower-face muscles during the presentation of visual stimuli. A larger N170 amplitude was found for faces relative to houses. Interestingly, this was the case both without and during fNMES, regardless of whether the fNMES artefact was removed or not. Moreover, sad facial expressions elicited a larger N170 amplitude relative to neutral facial expressions, both with and without fNMES.
Comparison with existing methods: fNMES offers a more precise way of manipulating proprioceptive feedback from facial muscles, which affords greater diversity in experimental design for studies on facial feedback effects.
Conclusions: We show that the combining of fNMES and EEG can be achieved and may serve as a powerful means of exploring the impact of controlled proprioceptive inputs on various types of cognitive processing.

1. Introduction

Electroencephalography (EEG) is a powerful – yet relatively inexpensive and thus practical – method to non-invasively measure brain activity with high-temporal resolution. For nearly a century, EEG, and its derived method of event-related potentials (ERPs) have been successfully used to investigate numerous aspects pertaining to cognitive neuroscience (Luck, 2014), including about the role of facial feedback in facial emotion recognition (Birch-Hurst et al., 2022; Schiano Lomoriello et al., 2021).

Facial feedback refers to afferent proprioceptive input from facial muscles to the central nervous system, and is believed by theories of embodied cognition (Niedenthal, 2007) to contribute to our felt emotions (Coles et al., 2019; J. I. Davis et al., 2009; Finzi and Rosenthal, 2016; Zamanian et al., 2017), the perception/recognition of emotional faces (Korb et al., 2016; Sel et al., 2015; Wood, 2016), and the interpretation of emotional stimuli in general (Strack et al., 1988; but see Wagenmakers et al., 2016). Indeed, experimental (pen in the mouth) or neurological conditions (face paralysis) affecting facial muscle movements can reduce accuracy and/or speed of facial emotion recognition (Korb et al., 2016; Oberman et al., 2007). They also affect EEG and ERP measures in response to faces (Birch-Hurst et al., 2022; J. D. Davis et al., 2017; Sessa et al., 2022). For example, the N170 visual-evoked potential (VEP), which reflects early face processing stages (Eimer, 2000), and is sometimes found to be larger for emotional than neutral facial expressions (Batty and Taylor, 2003; Hinojosa et al., 2015), increases in

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amplitude when participants smile to neutral faces, compared to when they observe the same faces while keeping a neutral expression (Sel et al., 2015; but see Schiano Lomoriello et al., 2021 who found facial feedback effects in high alexithymic participants on early visual responses but not on N170). This suggests that facial feedback can affect the N170 and potentially other aspects of the ERP/EEG in response to faces. However, the naturally or medically occurring changes in facial feedback (paralysis, Botox), and the experimental means to temporarily induce or change facial feedback (posing or blocking of facial expressions), often lack temporal and/or muscle specificity.

Facial neuromuscular electrical stimulation (fNMES) is an alternative way to directly manipulate afferent proprioceptive input from the face to the central nervous system (Pilurzi et al., 2020; Saito et al., 2013; Sasaki et al., 2017). It involves the direct activation of facial muscles via computer-controlled electrical impulses, and offers promising therapeutic effects for conditions such as pain (De Giorgi et al., 2017; Mummolo et al., 2020), weakening muscle strength (Choi, 2016; Safi et al., 2017), and unilateral facial paralysis (de Sire et al., 2022; Ilves et al., 2019; Makela et al., 2020). Moreover, fNMES can be delivered for short durations (e.g. 500 ms) at specific instances (e.g. before, during, or after the onset of a visually presented face). As such, fNMES bears enormous potential to study the effects of proprioceptive facial inputs in healthy participants, but remains largely unknown to both researchers and volunteers (Efthimiou, Hanel et al., 2022; Efthimiou, Perusquía--Hernández et al., 2022).

Preliminary evidence from participants' behaviour and self-report suggests that changes in felt emotion can indeed be induced with fNMES (Goto et al., 2018; Kapadia et al., 2019; Yen-Chin et al., 2017; Zariffa et al., 2014). However, the neural correlates of these effects have not been investigated. As mentioned above, physiological measures of brain activity, such as EEG, can reveal how the nervous system processes and integrates such proprioceptive inputs (Ding et al., 2021; Goldenkoff et al., 2021; Sel et al., 2015; Tayeb et al., 2021). Given the putative role of facial feedback for the processing and recognition of emotion in faces, a straight-forward question is whether fNMES modulates the N170.

N170 amplitude is consistently shown to be enlarged during the perception of face stimuli relative to non-face stimuli (Bentin et al., 1996; Eimer, 2011; Gao et al., 2019; Kanwisher et al., 1997; Rossion, 2014). Though N170 amplitude is sensitive to low-level visual features such as spatial frequency distributions (Eimer, 2011; Thierry et al., 2007), when such parameters are tightly controlled, it is still shown to be particularly sensitive to face stimuli (Rossion and Jacques, 2008). The exact mechanism that is indexed by N170 is debated, however it is generally accepted that it reflects early stages of face processing (such as structural encoding, Eimer, 2000).

The study of electrophysiological correlates to faces and their modulation by fNMES could be complicated given that it involves the delivery of a high amplitude, high frequency current to the body. Alternative electromagnetic stimulation techniques such as transcranial magnetic stimulation (TMS) and transcranial direct-current stimulation (tDCS) present a number of challenges when analysing EEG data. Stimulation currents can manifest as large bursts of interference in the EEG signal, and can distort accurate measurement of underlying brain dynamics (Gebodh et al., 2019). Many signal processing methods have been designed and implemented to reduce EEG artefacts induced by stimulation techniques, e.g. adaptive filtering (Mancini et al., 2015) and linear decomposition (Bai et al., 2016; Hernandez-Pavon et al., 2022), and so such methods might prove useful for reducing any potential artefacts introduced by fNMES. It should be noted however, that a liberal cleaning of EEG data can have minimal, and even detrimental effects, on the measurement of brain components (Delorme, 2022).

The purpose of the present study was to investigate the feasibility of combining fNMES and EEG. Specifically, we aimed to examine whether condition differences observed in trials without fNMES could also be observed during the application of fNMES. Given that the exact manifestation of the fNMES artefact in the EEG signal is unknown, we were also interested in whether the active removal of the fNMES artefact would impact any observed condition differences. We presented images of houses, as well as faces that displayed neutral, happy, and sad expressions, for half a second. An orthogonal task served to ensure participants' attention. fNMES was delivered to the lower facial muscles (between the lip corners and chin) for the duration of the presented visual stimulus in 50% of the trials. We aimed to establish whether differences in N170 amplitude could be observed both across stimulus types (houses and faces), and within stimulus types (different emotional expressions) both with and without fNMES, and whether the removal of the fNMES artefact would impact said differences. The reason for also examining within-stimulus type contrasts (different expressions) was to explore whether relatively small differences (compared to across-stimulus type contrasts, expected to elicit larger differences) would be observable during fNMES. Although it could be expected that stimulation of the DAO muscles could specifically modulate a particular expression type (e.g. sad), the current study was not motivated or designed to explicitly explore facial feedback effects, which have sometimes been found on the N170 (Sel et al., 2015; but see Schiano Lomoriello et al., 2021). The size of facial feedback effects has also been described to be generally small (Coles et al., 2019), and thus larger sample sizes are likely to be necessary to investigate them with fNMES.

Specifically, we hypothesized that, in the absence of fNMES, larger N170 amplitudes would be observed during the viewing of neutral facial expressions relative to images of houses, and that valenced facial expressions (happy and sad) would result in a larger N170 amplitude relative to neutral expressions. We were unsure as to whether these relative differences would be observed during the application of fNMES, however we expected that if they were, an active removal of the fNMES artefact would be necessary to observe them.

2. Methods

2.1. Participants

The participants were 20 adults (12 women, mean age = 30.8, SD = 6.6, range 20–44) with normal or corrected to normal vision, no current use of prescribed medication or history of illicit drug use, no history of neurological or psychiatric illness, no or shaved facial hair, and no known heart conditions. Participants were compensated with a voucher for their time. The participants were informed that the goal of the study was to investigate the effects of fNMES artefacts on EEG, gave written informed consent before taking part, and were debriefed on conclusion of the experiment. The study was approved by the local ethics committee (ETH2122–1966).

2.2. Apparatus and stimulation parameters

On each trial containing fNMES (see Fig. 1), fNMES was delivered for 500 ms at 70 Hz, as a train of 35 biphasic square pulses, with a pulse width of 100 μs, and a delay of 14 ms between pulses, using two constant-current electrical stimulators (Digitimer DS5, Welwyn Garden City, UK) and disposable Ag/AgCL 16 × 19 mm self-adhesive surface electrodes (Ambu BlueSensor BRS). We targeted the left and right Depressor Anguli Oris (DAO) muscles as the DAO is used during expressions of sadness, and could be a suitable target muscle for future studies on facial feedback effects. Electrode placement was based on existing EMG guidelines (Fridlund and Cacioppo, 1986) and experimenter visual verification. It was also verified after testing, based on video recordings. The amplitude (in mA) of the stimulation was determined for each individual by a configuration procedure defined below. These stimulation parameters were chosen based on our own pilot data that aimed to minimise discomfort whilst engaging facial muscles, and on previous studies using fNMES (e.g. Kapadia et al., 2019; for a review of the literature see Efthimiou, Perusquía-Hernández et al., 2022). The 70 Hz frequency is also convenient for other reasons, as it facilitates

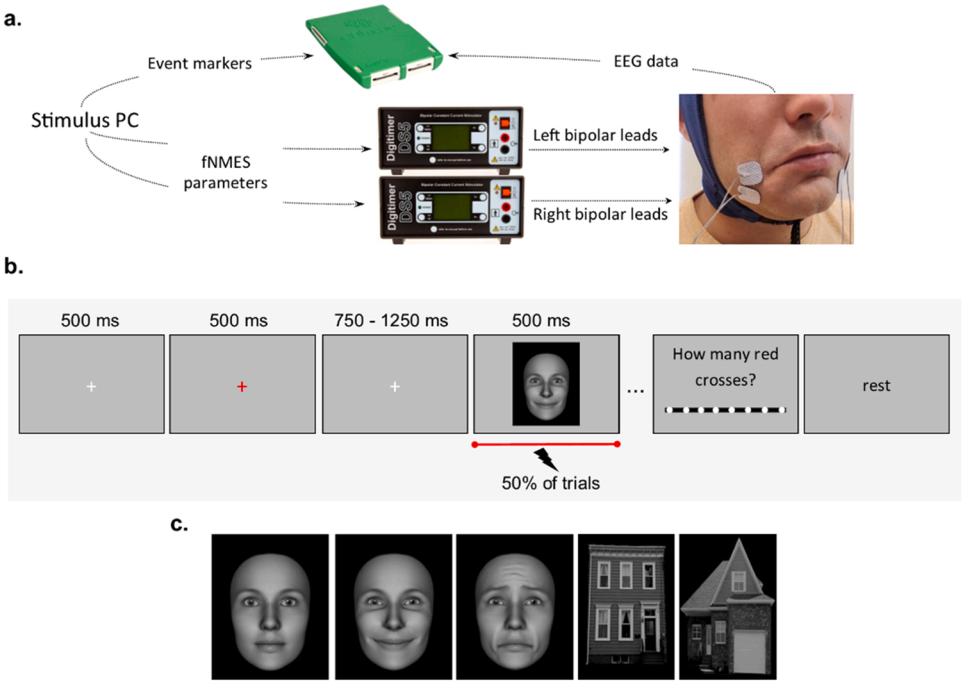

Fig. 1. Technical setup and procedure. (a) Schematic representation of the equipment used during the task. fNMES was delivered by two stimulators (one per muscle), which were controlled by the stimulus PC. (b) Timing of the paradigm. After each block, participants reported how many red crosses they had detected. fNMES was delivered for the entire duration of the stimulus on 50% of the trials. (c) Example stimuli used during the task: from left to right a neutral, happy, sad face, as well as two examples of houses.

estimation and removal of the stimulation artefacts, which will mostly lie outside the frequency range that is typically analysed for ERPs (1–30 Hz), and will also differ from line noise (50/60 Hz).

EEG data were acquired using an eego sports 64 channel amplifier (ANT Neuro, The Netherlands) in combination with ANT Neuro Waveguard 64 channel EEG caps in the licenced eego sports recording software environment.

2.3. Stimuli

The visual stimulus set was highly controlled, and thus ideal for ERP research. It consisted of 10 avatars (5 female) generated using the commercial software FaceGen (www.facegen.com) (see Fig. 1). The avatars were generated to have symmetrical faces with variance in the skin tone and face shape. All avatars were placed on a black background, with hair and ears removed. The generated faces were then manipulated using a second software FACSGen (Krumhuber et al., 2013; Roesch et al., 2011), which operates based on the facial action coding system (Ekman et al., 2002). The faces expressed three different emotional expressions (neutral, happy, sad), of which the happy and sad stimuli presented fully fledged expressions (i.e. non-ambiguous). Said expressions were produced by manipulating the relative intensity of Action units (AU). Specifically, happy expressions engaged AU6 (cheek raiser) at 100% intensity, AU7 (lids tightener) at 25% intensity, and AU12 (lip corner puller) at 100% intensity. For sad expressions, AU1 (inner brow raiser) and AU4 (brow lowerer) were engaged at 100% intensity, AU7 (lids tightener) at 80% intensity, AU11 (nasolabial deepener) at 60% intensity, and AU15 (lip corner depressor) at 100% intensity.

In total there were 30 unique images of faces (see Fig. 1c). The house stimuli were selected from the DalHouses database (Filliter et al., 2016). Houses were selected from the available set based on the lowest degree of similarity to faces (ratings provided by Filliter et al., 2016) so as to minimise the processing of any face-like features in the house stimuli. The 10 lowest rated 'face-like' houses were selected for the study. All final images were then turned to greyscale, equalised in luminance (mean and SD) using the MATLAB toolbox SHINE (Willenbockel et al., 2010) and resized to 800 × 1200 pixels.

2.4. Experimental procedure and task design

Participants were sat on a comfortable and stable chair with their eyes positioned approximately 60 cm from the centre of a 24.5 in. screen (Alienware aw2521h) with a resolution of 1920 × 1080 pixels and a 360 Hz refresh rate. Prior to beginning of the task, participants were introduced to the fNMES procedure so as to gain initial experience with the electrical stimulation of facial muscles, and to allow the experimenter to configure the appropriate fNMES amplitudes to use during the task. An alcohol wipe (70% isopropyl) was used to clean the skin, before the disposable surface electrodes were placed above the approximate location of the DAO muscle on each side of the face (guided by EMG guidelines, for a step-by-step guide see Efthimiou, Perusquía-Hernández et al., 2022). The experimenter systematically applied fNMES to each side of the face independently, starting with a low current. Current amplitude was gradually increased and the electrodes repositioned (if necessary) until a visible movement of the DAO was established, whilst maintaining acceptable levels of comfort for the participant. Current amplitudes of the established motor threshold were later used to apply fNMES during the task (left DAO: M = 20.4 mA, SD = 4.1 mA, right DAO: M = 20.7 mA, SD = 3.6 mA). Following the fNMES preparation, an EEG cap was placed over the head of the participant, and conductive Signa gel was applied until an impedance below 20 kΩ at each electrode was achieved. Participants were then informed that the task would begin shortly and that their face was being recorded throughout the session via a webcam. Video recordings were used in order to closely monitor the activation of the DAO muscles during the task, to ensure that the electrodes did not fall from the skin, and to verify appropriate electrode positioning when viewing after the session.

The task was programmed and run using Psychopy3 (v3.2.4) for Windows (Peirce et al., 2019). It required participants to covertly count the number of red fixation crosses presented throughout a given block of trials. Each trial began with a white fixation cross (horizontal = 1.19° and vertical = 1.19° of visual angle) presented at the centre of the display for 500 ms, which was followed by a second fixation cross presented for 500 ms that was either white or red. A final white fixation cross was then presented for 750 – 1200 ms before a face or house stimulus (horizontal = 14.25° and vertical = 20.77° of visual angle) appeared for 500 ms. In each trial, the colour of the fixation cross could

thus remain white, or briefly turn red before becoming white again. At the end of each block, participants were asked how many red crosses they had detected, and provided a response with a mouse click by selecting a number from a list on the screen. There were on average 12% of trials within each block that presented a red fixation cross. The purpose of this task was to maintain concentration throughout the block and to ensure that participants were attending to the centre of the screen during the presentation of the images. Accuracy values were not recorded, however the experimenter continuously monitored performance so as to ensure participants were actively engaged throughout each block.

Participants completed four blocks of trials in total, consisting of two types of blocks presented in a counterbalanced order (each type presented twice). In the houses-and-faces block, 40 images of each neutral facial expressions and houses were presented in a pseudo random order, ensuring that no more than three trials of the same type were shown consecutively. In the emotional-faces block, 40 images of each neutral, happy, and sad facial expressions were presented in a pseudo random order (10 identities displaying 3 expressions presented each 4 times), again ensuring that no more than three trials of the same type were presented consecutively. In all blocks, each unique stimulus was presented four times. As such, 80 trials were presented in each of the houses-and-faces blocks, and 120 trials were presented in each of the emotional-faces blocks. In 50% of all trials, fNMES was delivered for the duration of the presented image (500 ms). Pseudo randomisation of trial lists also ensured that no more than four trials containing fNMES were presented consecutively.

2.5. EEG data acquisition and signal processing

EEG data were acquired with 64 Ag/AgCl electrodes in the international 10–20 configuration at 512 Hz and digitized with 24-bit resolution. Data were referenced online to electrode CPz with the ground electrode at AFz, leaving 62 electrodes for further analyses. EEG data were imported and processed using functions from the EEGLAB (v2022.1) environment (Delorme and Makeig, 2004) for MATLAB (The Mathworks, Inc.). Continuous data were high-pass filtered (linear finite impulse response filter, filter order = 1690) at 1 Hz, which has been demonstrated to be optimal for independent component analysis (ICA) (Klug and Gramann, 2021), and low-pass filtered at 80 Hz, so as to retain the fNMES artefact in the data for the ICA decomposition (described below). Line noise (50 Hz and 100 Hz harmonic) was removed from the data using a combination of both Zapline (de Cheveigné, 2020) and CleanLine (Mullen, 2012) functions. Using a combination of spatio-spectral and time domain methods has been demonstrated to improve line noise removal (Miyakoshi et al., 2021). Channels that were considered noisy (e.g. contained extreme values or residual high frequencies) were interpolated using spherical interpolation (average of 4 per participant). Finally, data were epoched from − 500–1000 ms surrounding each stimulus onset. Epochs were then visually inspected for artefacts and were removed if they were considered to contain low-frequency drifts and/or unusual high frequency activity. Following epoch rejection, the average number of trials per condition was 39.61 (SD =0.54).

Prior to finalizing our pre-processing strategy, we investigated whether simply applying a 40 Hz low-pass filter to the data would be sufficient in removing the fNMES artefact (without removal of any fNMES artefact components). Although the 70 Hz noise was removed with said filter, it introduced a DC shift during the stimulation period, a shift which was inconsistent across participants (some presented a positive shift, whilst others a negative shift). We provide an additional figure in the supplementary materials to demonstrate this (Fig. 1 s).

Epoched EEG data were subjected to ICA decomposition (Infomax ICA; Bell and Sejnowski, 1995) using the EEGLAB's runica function. The reason for performing ICA on epoched data (and not continuous data) was to restrict ICA to time periods of interest in order to attempt to maximise the sensitivity of the algorithm in identifying the fNMES artefact. ICA linearly decomposes the observed EEG signal into temporally independent sources that are assigned a spatial and temporal structure. ICA allows for both the analysis of data in source space (as opposed to sensor space) and for the identification and removal of estimated contributing sources that might be considered to be originating outside of the brain. As such, ICA is a powerful method for the reduction of stimulation related artefacts in the EEG signal (Bai et al., 2016), and for artefacts typically present in EEG data. Following decomposition, the time course, spectra, and topography of each component were explored (see Fig. 3c).

The data were analysed at two different stages of the processing pipeline (see Fig. 2). That is, following only the removal of blink components ('fNMES artefact-included'), and also following the removal of both blink components and identified fNMES components ('fNMES artefact-excluded'). Both fNMES and no-fNMES trials were included in both analyses.

For both analyses, a component was labelled for removal if it presented low frequency non-time-locked fluctuations with strong power towards the front of the head (indicative of blinks, see Onton et al., 2006), or time-locked fluctuations with strong power at frontal sites only present in trials with fNMES (indicative of a stimulation-induced blink). For the fNMES artefact-excluded analysis only, components presenting a 70 Hz peak in the component spectra with strong power at bilateral frontal sites only present in trials with fNMES (indicative of the stimulation artefact) were also removed (see supplementary materials). An average of 1.35 (range = [1 4]) fNMES artefact components per participant were removed (average of 7.80% percent variance accounted for, per component). Following this inspection, suspect components were removed, and the data were back-projected to the scalp, thus removing their contribution to the observed EEG. Finally, data were referenced to an average of all channels and trials were baseline corrected using the pre-stimulus period. In the case of the fNMES artefact-excluded analysis, a 40 Hz low pass filter was applied. No such low pass filter was applied in the artefact-included analysis. An additional 40 Hz low pass filter was applied because even following the removal of the fNMES artefacts, residual 70 Hz noise could still be observed for some participants (albeit much smaller in magnitude than prior to the removal of components, see supplementary materials).

2.6. ERP analyses

In order to establish which electrode sites to extract N170 amplitudes at, we first concatenated all datasets (following removal of blink artefacts) and plotted scalp topography between 100 and 200 ms post stimulus onset. This revealed a bilateral occipital cluster of electrodes (P7, P8, PO7, PO8) that presented a negative peak at around 150 ms post stimulus onset. We then averaged together the identified set of channels which confirmed the presence of the N170 component peaking at 154 ms. In order to quantify N170 amplitude, we calculated the mean amplitude between 130 and 210 ms post stimulus onset for each condition. This time range was selected based on the grand average of all participants and conditions, following recommendations in the field (Luck and Gaspelin, 2017). Visual inspection of each participant's average ERP waveform (average of all conditions) confirmed that this defined time range appropriately captured the N170 peak in each case.

N170 amplitudes were extracted at two different points in the processing pipeline. That is, before (fNMES artefact-included) and after (fNMES artefact-excluded) the removal of the fNMES artefact (via linear decomposition and a 40 Hz low pass filter, see Section 2.5. EEG data acquisition and signal processing). Statistical analyses were conducted with SPSS 28, Bonferroni correction was applied to post-hoc tests, and Hedges' correction was applied to estimated effect sizes (Cohen's d). Non-significant t-tests were further investigated with Bayesian t-tests in JASP (v.017.1) using default parameters. The EEG pre-processing and analysis scripts, as well as raw data and stimulus materials are available

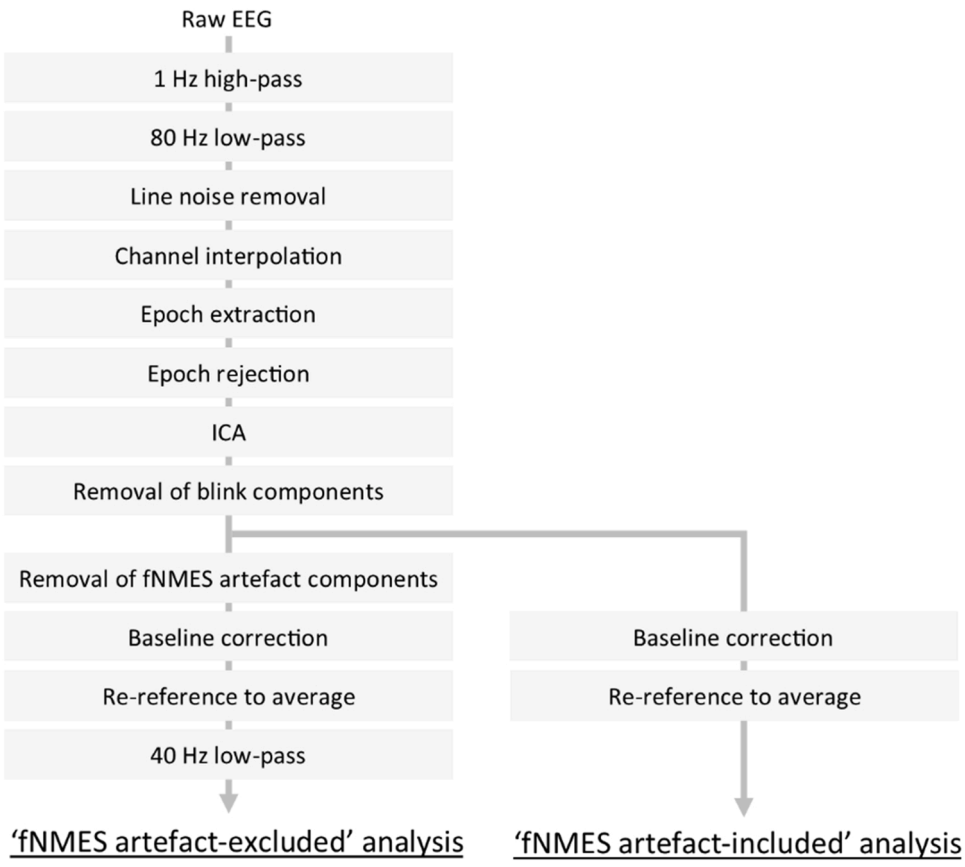

Fig. 2. Signal processing pipeline. Data were analysed at two different stages; following only the removal of blink components (fNMES artefact-included), and also following the additional removal of the fNMES artefact components and a 40 Hz low-pass filter (fNMES artefact-excluded). All trials (both fNMES and no-fNMES trials) were included in both analysis strategies.

online (https://osf.io/t4wa5/?view_only=5e066a7ab20b45ffac68d1d2288bdbea).

3. Results

3.1. N170 amplitudes for houses and faces

N170 amplitudes for houses and neutral faces were first analysed using a 2 (fNMES: off, on) x 2 (stimulus type: houses, faces) repeated measures ANOVA. The analysis was conducted on fNMES artefact-included and fNMES artefact-excluded amplitudes separately. For the fNMES artefact-included dataset (Fig. 4a), there was a significant main effect of stimulus type (F (1, 19) = 60.96, $p < .001$, $\eta_p^2 = .762$), whereby face stimuli resulted in a larger N170 amplitude (M = −1.11, SD = 2.47) relative to house stimuli (M = 1.24, SD = 2.39). Post-hoc paired sample t-tests revealed that this was the case both in the absence of fNMES (t (19) = 6.97, $p < .001$, d = 1.49, 95% CI [0.85, 2.12]; faces: M = −0.54, SD = 1.98, houses: M = 1.77, SD = 2.22) and when fNMES was applied (t (19) = 6.63, $p < .001$, d = 1.42, 95% CI [0.80, 2.03]; faces: M = −1.67, SD = 3.48, houses: M = 0.71, SD = 3.54). There was no significant main effect of fNMES (p = .113), nor a significant interaction between fNMES and stimulus type (p = .849). Similarly, for the fNMES artefact-excluded dataset (Fig. 4b), there was a significant main effect of stimulus type (F (1, 19) = 58.4, $p < .001$, $\eta_p^2 = .755$), whereby face stimuli resulted in a larger N170 amplitude [M = −0.92, SD = 1.83] relative to house stimuli [M = 1.26, SD = 1.82]. Post-hoc paired sample t-tests revealed that this was the case both in the absence of fNMES (t (19) = 6.76, $p < .001$, d = 1.45, 95% CI [0.82, 2.06]; faces: M = −0.43, SD = 1.81, houses: M = 1.70, SD = 2.05) and when fNMES was applied (t (19) = 6.69, $p < .001$, d = 1.43, 95% CI [0.81, 2.04]; faces: M = −1.42, SD = 2.4, houses: M = 0.81, SD = 2.53). There was no significant main effect of fNMES (p = .084), nor a significant interaction between fNMES and stimulus type (p = .753).

Finally, we explored the effect of removing the fNMES artefact on the observed differences between N170 amplitudes to face and house stimuli (we subtracted the value for faces from that for houses). Removing the fNMES artefact did not modulate the difference between house and face stimuli either without fNMES (p = .161, d =0.31, 95% CI [−0.12,0.74]), or with fNMES (p = .087, d =0.38, 95% CI [−0.05,0.82]).

3.2. N170 amplitudes for emotional facial expressions

N170 amplitudes for faces depicting emotional expressions were analysed using a 2 (fNMES: off, on) x 3 (expression: neutral, happy, sad) repeated-measures ANOVA. The analysis was conducted on fNMES artefact-included and fNMES artefact-excluded datasets separately. For the fNMES artefact-included dataset (Fig. 5a), there was no main effect or interaction involving fNMES (Fs < 1.84, ps > 0.05). There was however, a significant main effect of expression (F (2, 38) = 12.23, $p < .001$, $\eta_p^2 = .392$), whereby sad faces [M = −1.33, SD = 2.43] and happy faces [M = −0.96, SD = 1.96] elicited a larger N170 than neutral faces [M = −0.53, SD = 2.04] (both ps < 0.001). To investigate whether the differences between sad and happy, and neutral expressions were present both with and without fNMES, we performed exploratory paired sampled t-tests between each pair of emotions in both fNMES conditions separately. In the fNMES-off condition, N170 amplitude for sad expressions [M = −1.05, SD = 2.40] was larger than for neutral expressions [M = −0.14, SD = 1.63] (t (19) = 3.28, p = .024, d = 0.70, 95% CI [0.22, 1.17]); Fig. 5a). This difference was also observed when fNMES

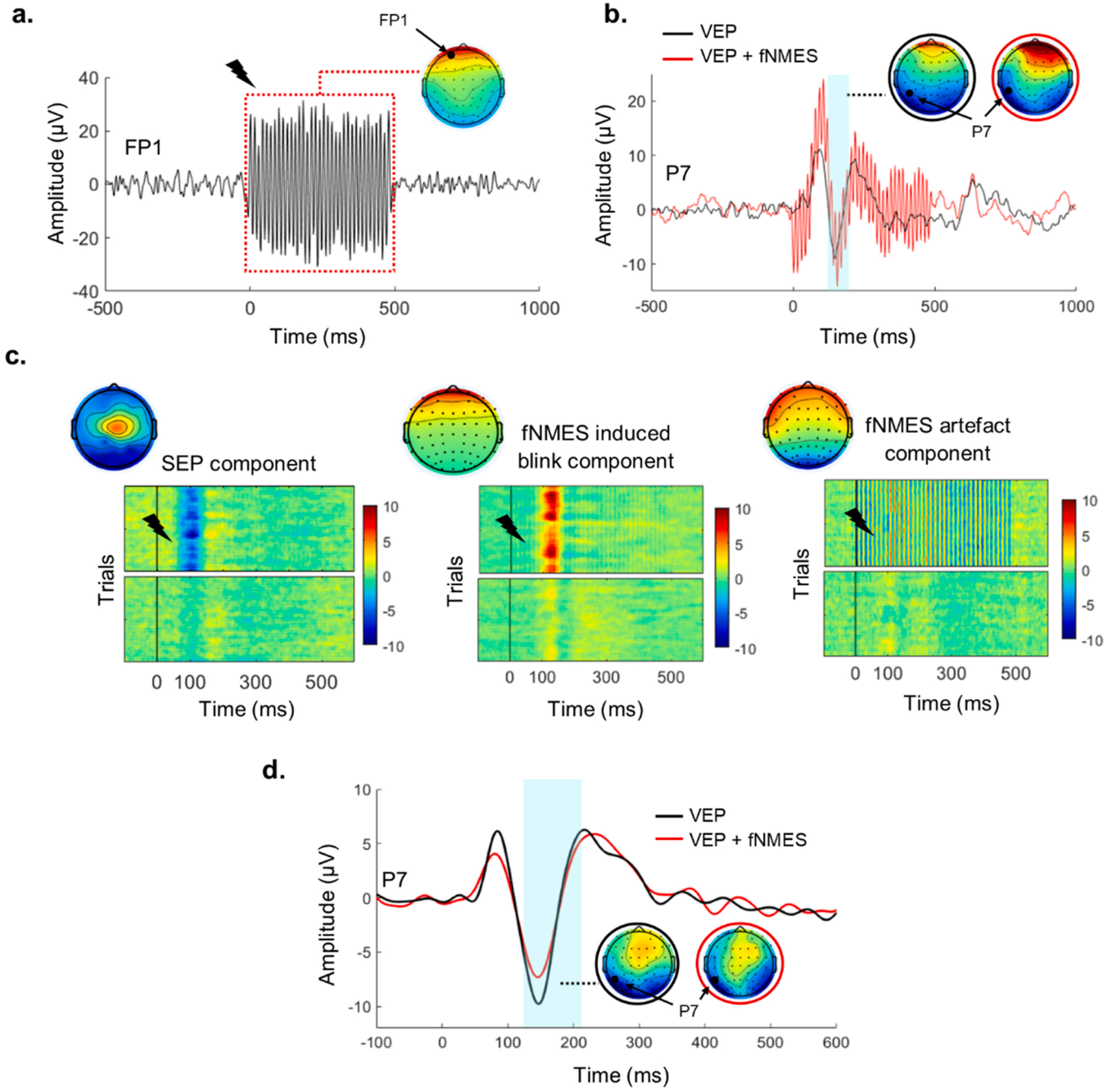

Fig. 3. Signal cleaning procedure and example independent components. (a) An example of the fNMES artefact at electrode FP1 (single trial of one subject). Scalp map shows average topography during the stimulation period (0 – 500 ms). (b) A visual evoked potential (VEP) without (black) and with (red) fNMES at electrode P7 showing average response to neutral face stimuli for one participant. Scalp maps show average topography between 130 and 210 ms post stimulus onset, reflecting an N170. (c) Exemplars of three independent components with corresponding scalp topographies and ERP images representing somatosensory evoked potentials (SEP) (left), fNMES-induced startle response (middle), and the fNMES artefact (right). The top rectangle of each ERP image shows trials with fNMES (as indicated by the lightning icon), the bottom rectangle shows trials without fNMES. (d) Back-projected averaged VEP for one participant to neutral faces without (black) and with (red) fNMES at P7 following removal of identified components and application of a 40 Hz low pass filter. Scalp maps show average topography between 130 and 210 ms post stimulus onset.

was applied (t (19) = 3.58, p = .012, d = 0.76, 95% CI [0.27, 1.24]) [sad: M = −1.62, SD = 3.12, neutral: M = −0.91, SD = 3.04]. N170 to happy expressions did not differ from neutral expressions either in the absence of fNMES (p = .156, d =0.44, 95% CI [−0.04,0.88]), or during fNMES (p = .087, d =0.50, 95% CI [0.05,0.95]). No differences were observed between happy and sad expressions either in the absence of fNMES (p = .210, d =0.48, 95% CI [0.03,0.92]) or during fNMES (p = .310, d =0.14, 95% CI [−0.28,0.56]).

For the fNMES artefact-excluded data (Fig. 5b), similar results were found. There was a significant main effect of expression (F (2, 38) = 7.80, p < .001, η_p^2 = .291), whereby sad faces [M = −1.04, SD = 1.81] and happy faces [M = −0.80, SD = 1.47] elicited a larger N170 than neutral faces [M = −0.45, SD = 1.48] (p < .001 and p = .005, respectively). Sad and happy expressions did not differ (p = .215). To investigate whether these differences were present both with and without fNMES, we performed exploratory paired sampled t-tests between sad

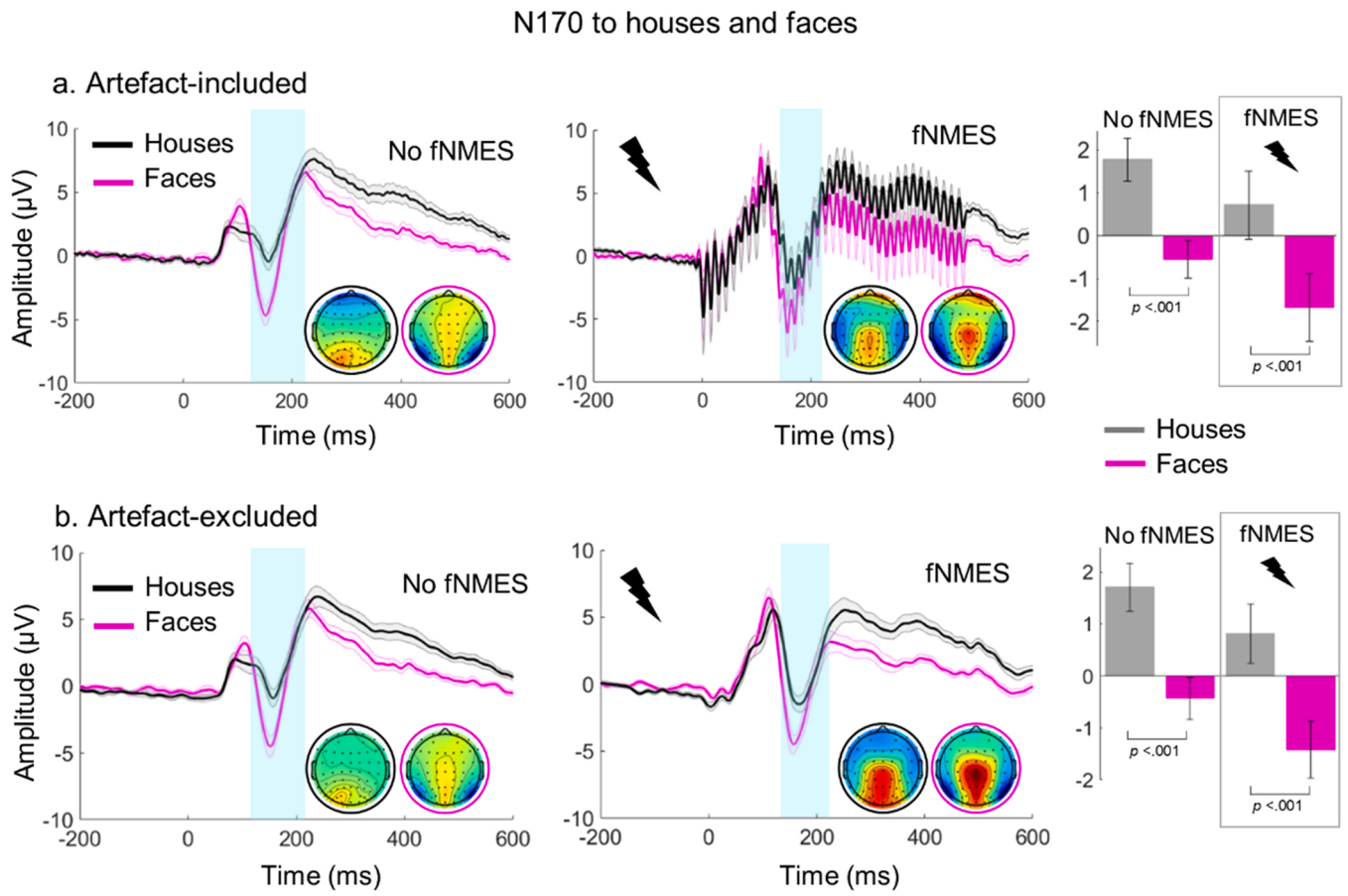

Fig. 4. N170 to houses and faces over bilateral occipital cluster (P7, P8, PO7, PO8). (a) N170 for house and face stimuli without fNMES (left) and during fNMES (right) for the artefact-included dataset (only blink components removed). N170 amplitude was greater for faces than houses in both conditions. (b) N170 for the artefact-excluded dataset (blink and fNMES components removed, 40 Hz low-pass filter) without fNMES (left) and during fNMES (right). Faces elicited larger N170 amplitudes than houses in both conditions. Scalp maps show average topography between 130 and 210 ms post stimulus onset. Shaded areas and error bars on bar plots (showing mean amplitudes) show standard error.

and neutral, and happy and neutral in both fNMES conditions separately. In the fNMES-off condition, N170 amplitude for sad expressions [M = −0.82, SD = 2.14] was larger than for neutral expressions [M = −0.07, SD = 1.30] (t (19) = 2.81, p = .044, d = 0.60, 95% CI [0.13, 1.06]). A similar finding was also observed when fNMES was applied (t (19) = 2.77, p = .048, d = 0.59, 95% CI [0.12, 1.05]) [sad: M = −1.27, SD = 2.13, neutral: M = −0.83, SD = 2.31]. Happy expressions did not differ from neutral expressions either without fNMES (p = .189, d =0.42, 95% CI [−0.02,0.86]), or with fNMES (p = .261, d =0.38, 95% CI [−0.05,0.82]). No differences were observed between happy and sad expressions either in the absence of fNMES (p = .171, d =0.43, 95% CI [−0.01,0.87]) or during fNMES (p = .971, d =0.008, 95% CI [−0.42,0.41]).

Finally, we explored the effect of removing the fNMES artefact on the observed differences between N170 amplitudes to sad and neutral expressions (Fig. 5c). When fNMES was applied, removing the artefact significantly reduced the difference between sad and neutral expressions (t (19) = 2.97, p = .008, d = 0.63, 95% CI [0.16, 1.09]) [artefact-included: M = 0.706, SD = 0.882, artefact-excluded: M = 0.440, SD = 7.10]. Surprisingly, this was also the case for trials without fNMES (t (19) = 2.73, p = .013, d = 0.58, 95% CI [0.12, 1.03]) [artefact-included: M = 0.905, SD = 1.23, artefact-excluded: M = 0.751, SD = 1.19].

In addition to the frequentist t-tests, we explored the non-significant pair-wise comparisons found between happy and neutral, and happy and sad expressions both in the absence and presence of fNMES (for both analysis strategies), with Bayesian paired-sample t-tests (artefact-included: [neutral no-fNMES – happy no-fNMES, BF_{10} = 1.34; happy no-fNMES – sad no-fNMES, BF_{10} = 1.82; neutral fNMES – happy fNMES, BF_{10} = 2.14; happy fNMES – sad fNMES, BF_{10} =0.28], artefact-excluded: [neutral no-fNMES – happy no-fNMES, BF_{10} = 1.16; happy no-fNMES – sad no-fNMES, BF_{10} = 1.26; neutral fNMES – happy fNMES, BF_{10} =0.90; happy fNMES – sad fNMES, BF_{10} =0.23]). All Bayes factor values were between.23 and 2.14, indicating at best anecdotal evidence in favour of H1.

4. Discussion

This study's primary goal was to investigate whether N170 amplitude differences between stimulus categories (faces and houses) and within stimulus categories (between emotional facial expressions) could be observable during the concurrent application of fNMES. Given that there are no previous studies that combine fNMES and EEG, we were unsure as to how the anticipated fNMES artefact would manifest in the EEG data, and whether the presence of an artefact would hinder any accurate measurement of N170 differences. As such, we also explored the impact of an active reduction of the fNMES artefact on any N170 amplitude differences observed during concurrent fNMES.

As expected, an artefact induced by fNMES was visible during the stimulation period (see Fig. 3a), which can also be seen to be superimposed over the visual response to stimuli (see Fig. 3b). Linear decomposition of the data (via ICA) revealed a number of components associated with fNMES (see Fig. 3c). These included components directly related to the high-frequency current (large peak in power at the stimulation frequency), and components that indicated a blink-induced

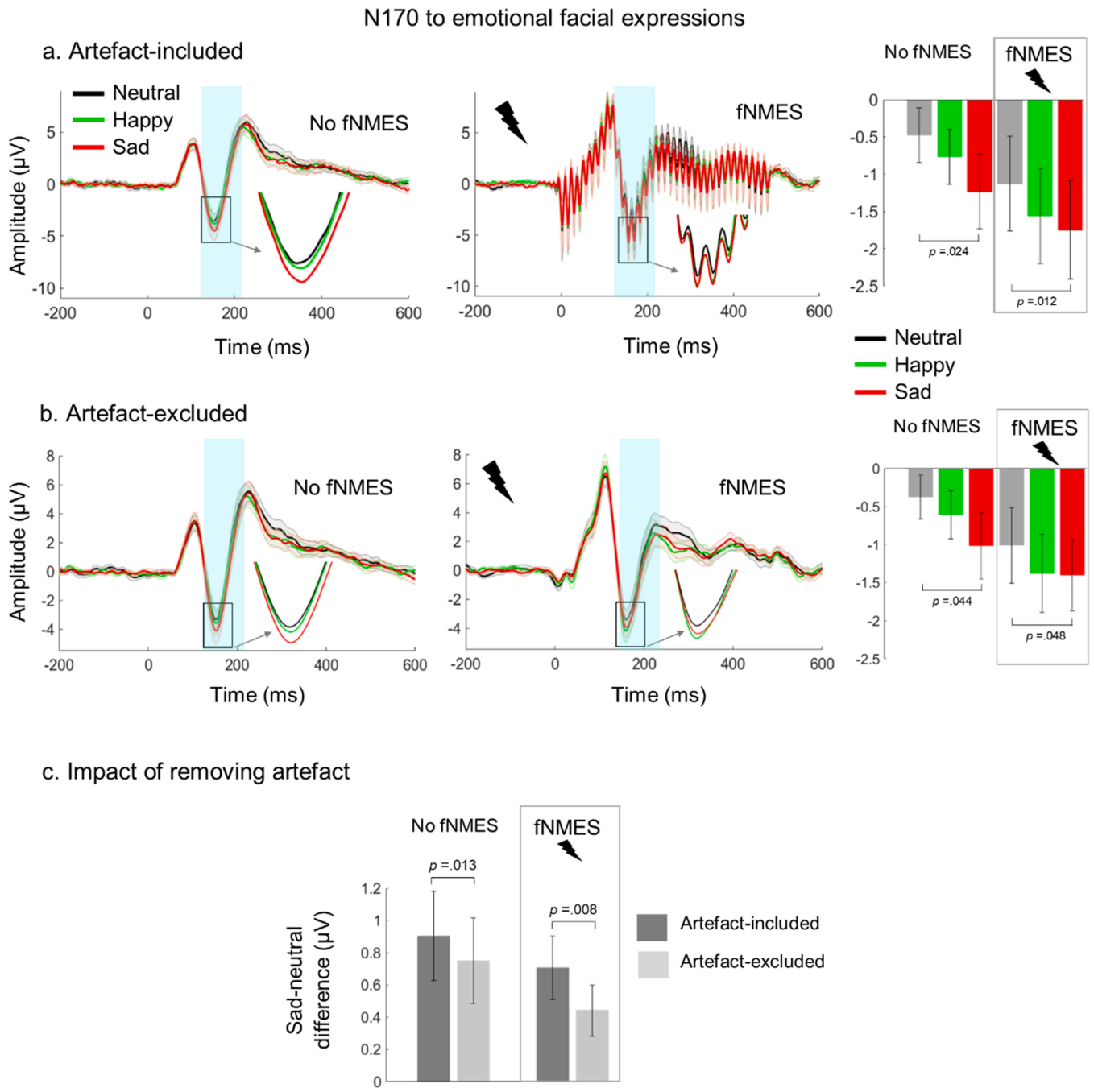

Fig. 5. N170 to emotional facial expressions over bilateral occipital cluster (P7, P8, PO7, PO8). (a) ERP without fNMES (left) and during fNMES (right) for the artefact-included dataset (only blink components removed). N170 amplitude was greater for sad faces than neutral faces in both conditions. (b) ERP for the artefact-excluded dataset (blink and fNMES components removed, 40 Hz low-pass filter applied) without fNMES (left) and during fNMES (right). Sad expressions elicited a larger N170 than neutral faces in both conditions. (c) Difference in N170 amplitude between sad and neutral expressions without fNMES (left) and with fNMES (right) with darker bars representing the artefact-included dataset. Removing the fNMES artefact significantly reduced the difference between sad and neutral expressions, even during trials without fNMES. Shaded areas and error bars on bar plots (showing mean amplitudes) show standard error.

startle response at the onset of stimulation. In addition, a somatosensory evoked potential (SEP) component was identified in the majority of participants (see supplementary materials for additional analyses of SEP). As such, ICA was able to accurately decompose the fNMES artefact (s) in the present study. Given that the artefact was observable at all in the averaged data, suggests that our stimulation onset times were extremely precise. If the stimulation delivery system was to introduce variability in onset times, then the artefact might not be visible in the averaged data, and ICA might not perform as well in isolating artefact-specific components.

Regarding comparisons between stimulus categories (houses and faces), in trials without fNMES, N170 amplitude was greater during the perception of faces relative to houses. This effect is commonplace in the face perception literature (Eimer, 2000; Key and Corbett, 2020), and confirms that our stimuli and paradigm were appropriate to elicit the classical N170. Interestingly, this same difference was observed during fNMES, whether or not the stimulation artefact was actively removed. This is surprising, given the clear presence of a high amplitude, high frequency noise component superimposed on the N170 (see Fig. 4a). It should be noted, however, that quantifying ERP amplitude using the

mean over a specified time window (here, 130–210 ms post stimulus onset) would necessarily smooth out the high frequency oscillations, thus reducing the impact of the artefact on the measurement of the ERP. There was no significant main effect of fNMES either prior to, or after artefact removal, however visual inspection indicated that N170 to both houses and faces became more negative during stimulation, thus the relative difference between houses and faces was maintained. As such, it is evident from our current findings that fNMES poses no significant challenges for observing robust effects such as a larger N170 to faces relative to non-face stimuli, whether the artefact is removed or not.

The reason for also including emotional faces in this experiment, is that differences in N170 amplitude between emotional and neutral faces are typically less pronounced (if they can be found at all), compared to those between faces and houses. That is, the physical properties of houses and faces differ more than do the physical properties of faces displaying different emotional expressions (for a related debate see Thierry et al., 2007)and Rossion (2014)), especially when low-level aspects of face stimuli are tightly controlled, as in the present stimulus set. We therefore aimed to see if these relatively smaller differences in N170 could be observed during fNMES. We first examined whether there existed any difference between expression types in the absence of stimulation and found that sad expressions resulted in larger N170 amplitudes than neutral expressions. Though the current study was not motivated for investigating the specific direction of any N170 differences between emotional facial expressions, a number of studies have shown that emotionally expressive faces elicit larger N170 amplitudes than do neutral faces (see Hinojosa et al., 2015). It should be noted that we analysed the power in low (<1 cycles/degree), medium (1–4 cycles/degree), and high (4–20 cycles/degree) spatial frequency bands for each expression, and found that sad expressions had significantly higher power in the medium spatial frequencies than did neutral expressions. Low-level visual features have indeed been shown to account for observed differences between conditions concerning affective stimuli (Delplanque et al., 2007), however a number of studies have demonstrated that this is not the case for the N170 (Bruchmann et al., 2020; Holmes et al., 2005). As such, we cannot rule out that an enhanced N170 to sad expressions was observed because of differences in power at medium spatial frequencies. Regardless of the cause for a larger N170 to sad expressions, we were primarily interested in whether the same difference would be observed during fNMES.

A pronounced N170 to sad facial expressions (relative to neutral) was also observed during fNMES when the stimulation artefact was present. Though we observed no main effect of fNMES or any interactions with expression type, numerically, N170 became more negative for all expressions during fNMES, thus the relative difference between sad and neutral expressions was maintained. Following the removal of the fNMES artefact, the aforementioned differences between sad and neutral expressions both without and with fNMES were again observed, however, to a smaller extent. Surprisingly, removing the artefact not only reduced the difference between sad and neutral expressions during fNMES trials, it also reduced the difference in trials that contained no stimulation (see Fig. 5c). As such, removing the fNMES artefact had an unexpected effect on non-fNMES trials. For the between stimulus types comparison (houses and faces), the removal of the fNMES artefact had no such effect. It should be noted that the removal of the fNMES artefact reduced the estimated effect size, Cohen's d, by about 0.1, thus a medium effect size for the sad and neutral contrast was observed in both analysis strategies. It is possible that the derived independent components contained a mix of source contributions (fNMES artefact and brain components), and thus the removal of an apparent fNMES artefact component could have suppressed the contribution of brain-specific activations to the observed ERP. If this were the case, then it could be argued that such an impact would be more evident/pronounced in data concerning much smaller differences between conditions (comparisons between expression types).

Although we observed significant differences between N170 amplitudes in response to neutral and sad facial expressions (both with and without stimulation) in the artefact-excluded analysis (and the impact of removing the artefact on said difference in both analysis strategies), a sensitivity analysis (two-tailed, $\alpha = 0.05$, power $=0.80$, $N = 20$) using G*Power (v.3.1.9.7) revealed that the effect sizes were below the bounds of what the current study was powered for (ds <0.66). As such, these findings should be taken with caution. The effect sizes for the significant across stimulus-type comparisons (houses and faces) were larger (all ds > 1.43). Future studies that aim to specifically examine the direction of differences within stimulus types (different emotional faces), should thus recruit a larger number of participants.

We did not find an effect of fNMES on N170 amplitudes in any condition, nor any interactions with expression or stimulus type. The purpose of the current study was to explore whether ERP components (such as the N170) could be observed at all during fNMES. As such, the study was not designed to explicitly explore facial feedback effects in emotional face perception. One of the advantages of using fNMES is that it allows for a high degree of specificity when modulating afferent proprioceptive signals to the central nervous system, signals of which could be integrated with other sensory inputs in order to aid in the interpretation of external events. In our case, the faces presented were of fully fledged emotional expressions, and therefore presented no ambiguity. The literature on multisensory integration demonstrates that the combining of multiple sensory signals can facilitate the resolving of sensory ambiguities (Green and Angelaki, 2010). As such, future studies that aim to use fNMES as a means of studying facial feedback effects should utilize ambiguous facial expressions. In addition, facial feedback effects are typically very small (Coles et al., 2019), and so future studies should aim to recruit a much larger sample than we included in the present study. It is also possible that evidence of any modulation of face processing due to proprioceptive signals induced by fNMES is masked by the stimulation artefact, and/or is removed during the artefact removal procedure. If this were the case, then this would severely limit the types of experimental designs that are afforded when combining fNMES and EEG, unless alternative artefact reduction methods are implemented.

Though we have successfully demonstrated that differences in N170 can be observed during concurrent fNMES, more attention should be given to the impact of artefact reduction procedures on the observation of underlying brain dynamics. Recently, Delorme (2022) demonstrated that many of the typical processing steps associated with reducing noise in EEG data (including ICA) have very little (and sometimes detrimental) effects on observing significant differences between conditions. The artefact reduction procedure that we have implemented here, significantly reduced the differences between N170 amplitudes to sad and neutral expressions, even during trials that contained no stimulation artefacts. Although these differences remained significant, the removal of the fNMES artefact under different experimental conditions and with different stimulation and recording apparatus might have undesired consequences.

Our findings need not encourage the choice to abandon the active removal of the fNMES artefact. It is standard practice to reduce the impact of stimulation artefacts on EEG data (such as those observed during TMS and tDCS) so that accurate measurements of underlying brain phenomena can be obtained. The EEG system used in the current study had active shielding that limits conductive interference from sources originating from outside of the head (e.g. line-noise), and thus the impact of fNMES might be minimal compared to systems that do not have such shielding. In addition, EEG analyses are not restricted to ERPs. The impact of fNMES on other measures such as time-frequency decompositions and spectral coherence measures might pose significant challenges, unless the artefact is indeed removed.

In sum, the combining of fNMES and EEG offers a relatively low-cost and practical method of investigating multisensory integration that ensures both temporal and spatial precision when inducing activation of facial muscles. We encourage researchers to adopt fNMES as a means of

investigating the neurophysiological processes underlying facial feedback effects.

CRediT authorship contribution statement

J. Baker: Conceptualization, Methodology, Software, Data curation, Writing – original draft preparation, Visualization, Formal analysis, Investigation. **T. Efthimiou:** Methodology, Writing – review & editing, Project administration, Software. **R. Scherer:** Conceptualization, Supervision, Writing – review & editing. **A. Gartus:** Methodology, Software, Resources, Writing – review & editing. **A. Elsenaar:** Methodology, Conceptualization, Funding acquisition, Writing – review & editing. **M. Mehu:** Methodology, Writing – review & editing, Funding acquisition. **S Korb:** Supervision, Conceptualization, Methodology, Writing – review & editing, Funding acquisition.

Declaration of Competing Interest

The authors declare no conflict of interest.

Data availability

data and code available on OSF (link in manuscript).

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jneumeth.2023.109877.

References

Bai, Y., Wan, X., Zeng, K., Ni, Y., Qiu, L., Li, X., 2016. Reduction hybrid artifacts of EMG–EOG in electroencephalography evoked by prefrontal transcranial magnetic stimulation. J. Neural Eng. 13 (6), 066016 https://doi.org/10.1088/1741-2560/13/6/066016.

Batty, M., Taylor, M.J., 2003. Early processing of the six basic facial emotional expressions. Cogn. Brain Res. 17 (3), 613–620. https://doi.org/10.1016/S0926-6410(03)00174-5.

Bell, A.J., Sejnowski, T.J., 1995. An information-maximization approach to blind separation and blind deconvolution. Neural Comput. 7 (6), 1129–1159. https://doi.org/10.1162/neco.1995.7.6.1129.

Bentin, S., Allison, T., Puce, A., Perez, E., McCarthy, G., 1996. Electrophysiological Studies of Face Perception in Humans. J. Cogn. Neurosci. 8 (6), 551–565.

Birch-Hurst, K., Rychlowska, M., Lewis, M.B., Vanderwert, R.E., 2022. Altering Facial Movements Abolishes Neural Mirroring of Facial Expressions. Cogn., Affect., Behav. Neurosci. 22 (2), 316–327. https://doi.org/10.3758/s13415-021-00956-z.

Bruchmann, M., Schindler, S., Straube, T., 2020. The spatial frequency spectrum of fearful faces modulates early and mid-latency ERPs but not the N170. Psychophysiology 57 (9), e13597. https://doi.org/10.1111/psyp.13597.

de Cheveigné, A., 2020. ZapLine: A simple and effective method to remove power line artifacts. NeuroImage 207, 116356. https://doi.org/10.1016/j.neuroimage.2019.116356.

Choi, J.-B., 2016. Effect of neuromuscular electrical stimulation on facial muscle strength and oral function in stroke patients with facial palsy. J. Phys. Ther. Sci. 28 (9), 2541–2543. https://doi.org/10.1589/jpts.28.2541.

Coles, N.A., Larsen, J.T., Lench, H.C., 2019. A meta-analysis of the facial feedback literature: Effects of facial feedback on emotional experience are small and variable. Psychol. Bull. 145 (6), 610–651. https://doi.org/10.1037/bul0000194.

Davis, J.D., Winkielman, P., Coulson, S., 2017. Sensorimotor simulation and emotion processing: Impairing facial action increases semantic retrieval demands. Cogn., Affect. Behav. Neurosci. 17 (3), 652–664. https://doi.org/10.3758/s13415-017-0503-2.

Davis, J.I., Senghas, A., Ochsner, K.N., 2009. How does facial feedback modulate emotional experience. J. Res. Personal. 43 (5), 822–829. https://doi.org/10.1016/j.jrp.2009.06.005.

De Giorgi, I., Castroflorio, T., Sartoris, B., Deregibus, A., 2017. The use of conventional transcutaneous electrical nerve stimulation in chronic facial myalgia patients. Clin. Oral. Investig. 21 (1), 275–280. https://doi.org/10.1007/s00784-016-1787-2.

Delorme, A., 2022. *EEG is better left alone* (p. 2022.12.03.518987). bioRxiv. https://doi.org/10.1101/2022.12.03.518987.

Delorme, A., Makeig, S., 2004. EEGLAB: An open source toolbox for analysis of single-trial EEG dynamics including independent component analysis. J. Neurosci. Methods 134 (1), 9–21. https://doi.org/10.1016/j.jneumeth.2003.10.009.

Delplanque, S., N'diaye, K., Scherer, K., Grandjean, D., 2007. Spatial frequencies or emotional effects?: A systematic measure of spatial frequencies for IAPS pictures by a discrete wavelet analysis. J. Neurosci. Methods 165 (1), 144–150. https://doi.org/10.1016/j.jneumeth.2007.05.030.

Ding, K., Dragomir, A., Bose, R., Osborn, L., Seet, M., Bezerianos, A., Thakor, N., 2021. Sensory Stimulation Enhances Functional Connectivity towards the Somatosensory Cortex in Upper Limb Amputation. 2021 10th Int. IEEE/EMBS Conf. Neural Eng. (NER) 226–229. https://doi.org/10.1109/NER49283.2021.9441295.

Efthimiou, T.N., Hanel, P.H.P., Korb, S., 2022. Volunteers' concerns about facial neuromuscular electrical stimulation. BMC Psychol. 10 (1), 117. https://doi.org/10.1186/s40359-022-00827-3.

Efthimiou, T.N., Perusquía-Hernández, M., Elsenaar, A., Mehu, M., Korb, S., 2022. facial Neuromuscular Electrical Stimulation (fNMES)—Review and recommendations. PsyArXiv. https://doi.org/10.31234/osf.io/fd8ce.

Eimer, M., 2000. The face-specific N170 component reflects late stages in the structural encoding of faces. NeuroReport 11 (10), 2319–2324.

Eimer, M., 2011. The Face-Sensitive N170 Component of the Event-Related Brain Potential. Oxf. Handb. Face Percept. https://doi.org/10.1093/oxfordhb/9780199559053.013.0017.

Ekman, P., Friesen, W.V., & Hager, J.C. (2002). *The Facial Action Coding System: A Technique for the Measurement of Facial Movement. San Francisco, CA: Consulting Psychologists Press.* San Francisco, CA: Consulting Psychologists Press.

Filliter, J.H., Glover, J.M., McMullen, P.A., Salmon, J.P., Johnson, S.A., 2016. The DalHouses: 100 new photographs of houses with ratings of typicality, familiarity, and degree of similarity to faces. Behav. Res. Methods 48 (1), 178–183. https://doi.org/10.3758/s13428-015-0561-8.

Finzi, E., Rosenthal, N.E., 2016. Emotional proprioception: Treatment of depression with afferent facial feedback. J. Psychiatr. Res. 80, 93–96. https://doi.org/10.1016/j.jpsychires.2016.06.009.

Fridlund, A.J., Cacioppo, J.T., 1986. Guidelines for human electromyographic research. Psychophysiology 23, 567–589. https://doi.org/10.1111/j.1469-8986.1986.tb00676.x.

Gao, C., Conte, S., Richards, J.E., Xie, W., Hanayik, T., 2019. The neural sources of N170: Understanding timing of activation in face-selective areas. Psychophysiology 56 (6), e13336. https://doi.org/10.1111/psyp.13336.

Gebodh, N., Esmaeilpour, Z., Adair, D., Chelette, K., Dmochowski, J., Woods, A.J., Kappenman, E.S., Parra, L.C., Bikson, M., 2019. Inherent physiological artifacts in EEG during tDCS. NeuroImage 185, 408–424. https://doi.org/10.1016/j.neuroimage.2018.10.025.

Goldenkoff, E.R., McGregor, H.R., Mergos, J., Gholizadeh, P., Bridenstine, J., Brown, M. J.N., Vesia, M., 2021. Reversal of Visual Feedback Modulates Somatosensory Plasticity. Neuroscience 452, 335–344. https://doi.org/10.1016/j.neuroscience.2020.10.033.

Goto, T., Tag, B., Kunze, K., Dingler, T., & Assoc Comp Machinery, 2018, Towards Enhancing Emotional Responses to Media using Auto-Calibrating Electric Muscle Stimulation (EMS) (WOS:000511407100028). AUGMENTED HUMAN 2018: PROCEEDINGS OF THE 9TH AUGMENTED HUMAN INTERNATIONAL CONFERENCE. https://doi.org/10.1145/3174910.3174939.

Green, A.M., Angelaki, D.E., 2010. Multisensory integration: Resolving sensory ambiguities to build novel representations. Curr. Opin. Neurobiol. 20 (3), 353–360. https://doi.org/10.1016/j.conb.2010.04.009.

Hernandez-Pavon, J.C., Kugiumtzis, D., Zrenner, C., Kimiskidis, V.K., Metsomaa, J., 2022. Removing artifacts from TMS-evoked EEG: A methods review and a unifying theoretical framework. J. Neurosci. Methods 376, 109591. https://doi.org/10.1016/j.jneumeth.2022.109591.

Hinojosa, J.A., Mercado, F., Carretié, L., 2015. N170 sensitivity to facial expression: A meta-analysis. Neurosci. Biobehav. Rev. 55, 498–509. https://doi.org/10.1016/j.neubiorev.2015.06.002.

Holmes, A., Winston, J.S., Eimer, M., 2005. The role of spatial frequency information for ERP components sensitive to faces and emotional facial expression. Cogn. Brain Res. 25 (2), 508–520. https://doi.org/10.1016/j.cogbrainres.2005.08.003.

Ilves, M., Lylykangas, J., Rantanen, V., Mäkelä, E., Vehkaoja, A., Verho, J., Lekkala, J., Rautiainen, M., Surakka, V., 2019. Facial muscle activations by functional electrical stimulation. Biomed. Signal Process. Control 48, 248–254. https://doi.org/10.1016/j.bspc.2018.10.015.

Kanwisher, N., McDermott, J., Chun, M.M., 1997. The fusiform face area: a module in human extrastriate cortex specialized for face perception. J. Neurosci. 17 (11), 4302–4311. https://doi.org/10.1523/JNEUROSCI.17-11-04302.1997.

Kapadia, N., Zivanovic, V., Moineau, B., Downar, J., Zariffa, J., Popovic, M.R., 2019. Functional electrical stimulation of the facial muscles to improve symptoms in individuals with major depressive disorder: Pilot feasibility study. Biomed. Eng. OnLine 18 (1), 109. https://doi.org/10.1186/s12938-019-0730-6.

Key, A.P., Corbett, B.A., 2020. The Unfulfilled Promise of the N170 as a Social Biomarker. Biol. Psychiatry.: Cogn. Neurosci. Neuroimaging 5 (3), 342–353. https://doi.org/10.1016/j.bpsc.2019.08.011.

Klug, M., Gramann, K., 2021. Identifying key factors for improving ICA-based decomposition of EEG data in mobile and stationary experiments. Eur. J. Neurosci. 54 (12), 8406–8420. https://doi.org/10.1111/ejn.14992.

Korb, S., Wood, A., Banks, C.A., Agoulnik, D., Hadlock, T.A., Niedenthal, P.M., 2016. Asymmetry of Facial Mimicry and Emotion Perception in Patients With Unilateral Facial Paralysis. JAMA Facial Plast. Surg. 18 (3), 222–227. https://doi.org/10.1001/jamafacial.2015.2347.

Krumhuber, E.G., Kappas, A., Manstead, A.S.R., 2013. Effects of Dynamic Aspects of Facial Expressions: A Review. Emot. Rev. 5 (1), 41–46. https://doi.org/10.1177/1754073912451349.

Luck, S.J. (2014). *An Introduction to the Event-Related Potential Technique, second edition*. MIT Press.

Luck, S.J., Gaspelin, N., 2017. How to get statistically significant effects in any ERP experiment (and why you shouldn't). Psychophysiology 54 (1), 146–157. https://doi.org/10.1111/psyp.12639.

Makela, E., Ilves, M., Venesvirta, H., Lylykangas, J., Rantanen, V., Vehkaoja, A., Verho, J., Lekkala, J., Surakka, V., Rautiainen, M., 2020. Effect of pulse waveforms on movement amplitudes and perceived discomfort in electric muscle stimulation in unresolved facial nerve palsy. Biomed. Phys. Eng. EXPRESS 6 (3). https://doi.org/10.1088/2057-1976/ab7eea.

Mancini, M., Pellicciari, M.C., Brignani, D., Mauri, P., De Marchis, C., Miniussi, C., Conforto, S., 2015. Automatic artifact suppression in simultaneous tDCS-EEG using adaptive filtering. 2015 37th Annu. Int. Conf. IEEE Eng. Med. Biol. Soc. (EMBC) 2729–2732. https://doi.org/10.1109/EMBC.2015.7318956.

Miyakoshi, M., Schmitt, L.M., Erickson, C.A., Sweeney, J.A., Pedapati, E.V., 2021. Can We Push the "Quasi-Perfect Artifact Rejection" Even Closer to Perfection? Front. Neuroinformatics 14, 597079. https://doi.org/10.3389/fninf.2020.597079.

Mummolo, S., Nota, A., Tecco, S., Caruso, S., Marchetti, E., Marzo, G., Cutilli, T., 2020. Ultra-low-frequency transcutaneous electric nerve stimulation (ULF-TENS) in subjects with craniofacial pain: A retrospective study. CRANIO® 38 (6), 396–401. https://doi.org/10.1080/08869634.2018.1526849.

Niedenthal, P.M., 2007. Embodying Emotion. Science 316 (5827), 1002–1005. https://doi.org/10.1126/science.1136930.

Oberman, L.M., Winkielman, P., Ramachandran, V.S., 2007. Face to face: Blocking facial mimicry can selectively impair recognition of emotional expressions. Soc. Neurosci. 2 (3–4), 167–178. https://doi.org/10.1080/17470910701391943.

Onton, J., Westerfield, M., Townsend, J., Makeig, S., 2006. Imaging human EEG dynamics using independent component analysis. Neurosci. Biobehav Rev. 30 (6), 808–822 https://doi.org/S0149-7634(06)00050-9 [pii] 10.1016/j.neubiorev.2006.06.007.

Peirce, J., Gray, J.R., Simpson, S., MacAskill, M., Höchenberger, R., Sogo, H., Kastman, E., Lindeløv, J.K., 2019. PsychoPy2: Experiments in behavior made easy. Behav. Res. Methods 51 (1), 195–203. https://doi.org/10.3758/s13428-018-01193-y.

Pilurzi, G., Ginatempo, F., Mercante, B., Cattaneo, L., Pavesi, G., Rothwell, J.C., Deriu, F., 2020. Role of cutaneous and proprioceptive inputs in sensorimotor integration and plasticity occurring in the facial primary motor cortex. J. Physiol. 598 (4), 839–851. https://doi.org/10.1113/JP278877.

Roesch, E.B., Tamarit, L., Reveret, L., Grandjean, D., Sander, D., Scherer, K.R., 2011. FACSGen: A Tool to Synthesize Emotional Facial Expressions Through Systematic Manipulation of Facial Action Units. J. Nonverbal Behav. 35 (1), 1–16. https://doi.org/10.1007/s10919-010-0095-9.

Rossion, B., 2014. Understanding face perception by means of human electrophysiology. Trends Cogn. Sci. 18 (6), 310–318. https://doi.org/10.1016/j.tics.2014.02.013.

Rossion, B., Jacques, C., 2008. Does physical interstimulus variance account for early electrophysiological face sensitive responses in the human brain? Ten lessons on the N170. NeuroImage 39 (4), 1959–1979. https://doi.org/10.1016/j.neuroimage.2007.10.011.

Safi, M.F., Wright-Harp, W., Lucker, J.R., Payne, J.C., 2017. Effect of surface neuromuscular electrical stimulation on labial and lingual muscles in healthy volunteers. Int. J. Rehabil. Res. Int. Z. Fur Rehabil. Rev. Int. De. Rech. De. Readapt. 40 (2), 119–126. https://doi.org/10.1097/MRR.0000000000000217.

Saito, K., Yamaguchi, T., Yoshida, N., Tanabe, S., Kondo, K., Sugawara, K., 2013. Combined effect of motor imagery and peripheral nerve electrical stimulation on the motor cortex. Exp. Brain Res. 227 (3), 333–342. https://doi.org/10.1007/s00221-013-3513-5.

Sasaki, R., Kotan, S., Nakagawa, M., Miyaguchi, S., Kojima, S., Saito, K., Inukai, Y., Onishi, H., 2017. Presence and Absence of Muscle Contraction Elicited by Peripheral Nerve Electrical Stimulation Differentially Modulate Primary Motor Cortex Excitability. *Front. Hum. Neurosci.* 11 https://doi.org/10.3389/fnhum.2017.00146.

Schiano Lomoriello, A., Maffei, A., Brigadoi, S., Sessa, P., 2021. Altering sensorimotor simulation impacts early stages of facial expression processing depending on individual differences in alexithymic traits. Brain Cogn. 148, 105678 https://doi.org/10.1016/j.bandc.2020.105678.

Sel, A., Calvo-Merino, B., Tuettenberg, S., Forster, B., 2015. When you smile, the world smiles at you: ERP evidence for self-expression effects on face processing. Soc. Cogn. Affect. Neurosci. 10 (10), 1316–1322. https://doi.org/10.1093/scan/nsv009.

Sessa, P., Schiano Lomoriello, A., Duma, G.M., Mento, G., De Stefani, E., Ferrari, P.F., 2022. Degenerate pathway for processing smile and other emotional expressions in congenital facial palsy: An hdEEG investigation. Philos. Trans. R. Soc. B: Biol. Sci. 377 (1863), 20210190. https://doi.org/10.1098/rstb.2021.0190.

de Sire, A., Marotta, N., Agostini, F., Drago Ferrante, V., Demeco, A., Ferrillo, M., Inzitari, M.T., Pellegrino, R., Russo, I., Ozyemisci Taskiran, O., Bernetti, A., Ammendolia, A., 2022. A Telerehabilitation Approach to Chronic Facial Paralysis in the COVID-19 Pandemic Scenario: What Role for Electromyography Assessment. J. Pers. Med. 12 (3) https://doi.org/10.3390/jpm12030497.

Strack, F., Martin, L.L., Stepper, S., 1988. Inhibiting and facilitating conditions of the human smile: A nonobtrusive test of the facial feedback hypothesis. J. Personal. Soc. Psychol. 54 (5), 768–777. https://doi.org/10.1037/0022-3514.54.5.768.

Tayeb, Z., Bose, R., Dragomir, A., Osborn, L.E., Thakor, N.V., Cheng, G., 2021. EEG Decoding of Pain Perception for a Real-Time Reflex System in Prostheses. In: Guger, C., Allison, B.Z., Gunduz, A. (Eds.), Brain-Computer Interface Research: A State-of-the-Art Summary, 10. Springer International Publishing, pp. 47–58 https://doi.org/10.1007/978-3-030-79287-9_5.

Thierry, G., Martin, C.D., Downing, P., Pegna, A.J., 2007. Controlling for interstimulus perceptual variance abolishes N170 face selectivity. In: Nature Neuroscience, 10. Article 4. https://doi.org/10.1038/nn1864.

Wagenmakers, E.-J., Beek, T., Dijkhoff, L., Gronau, Q.F., Acosta, A., Adams, R.B., Albohn, D.N., Allard, E.S., Benning, S.D., Blouin-Hudon, E.-M., Bulnes, L.C., Caldwell, T.L., Calin-Jageman, R.J., Capaldi, C.A., Carfagno, N.S., Chasten, K.T., Cleeremans, A., Connell, L., DeCicco, J.M., Zwaan, R.A., 2016. Registered Replication Report Strack, Martin, & Stepper (1988). *Perspectives on*. Psychol. Sci. 11 (6), 917–928. https://doi.org/10.1177/1745691616674458.

Willenbockel, V., Sadr, J., Fiset, D., Horne, G.O., Gosselin, F., Tanaka, J.W., 2010. Controlling low-level image properties: The SHINE toolbox. Behav. Res. Methods 42 (3), 671–684. https://doi.org/10.3758/BRM.42.3.671.

Wood, A., 2016. Altering sensorimotor feedback disrupts visual discrimination of facial expressions. Psychon. Bull. Rev. 7.

Yen-Chin, L., Feng, Y., Kunze, K., Shimizu, J., Nakao, T., 2017. Eyewear to Make Me Smile: Can Electric Muscle Stimulation increase Happiness? Proc. Elev. Int. Conf. Tangible, Embed., Embodied Interact. 579–582. https://doi.org/10.1145/3024969.3025097.

Zamanian, A., Jolfaei, A.G., Mehran, G., Azizian, Z., 2017. Efficacy of Botox versus placebo for treatment of patients with major depression. Iran. J. Public Health 46 (7), 982–984.

Zariffa, J., Hitzig, S.L., Popovic, M.R., 2014. Neuromodulation of Emotion Using Functional Electrical Stimulation Applied to Facial Muscles. Neuromodulation: Technol. Neural Interface 17 (1), 85–92. https://doi.org/10.1111/ner.12056.