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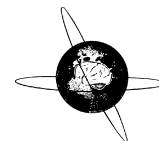
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## Using facial electromyography to detect preserved emotional processing in disorders of consciousness: A proof-of-principle study



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### HIGHLIGHTS

- A vegetative state patient can show intact emotional responses as measured by facial muscle activity.
- Responses in a VS patient mirrored the pattern of muscle activity observed in healthy controls.
- This methodology may serve as a feasible bedside tool to probe emotion in VS patients.

### ABSTRACT

**Objective:** To examine whether emotional functioning can be observed in patients who are behaviourally non-responsive using peripheral markers of emotional functioning.

**Method:** We tested two patients, both diagnosed as being in a vegetative state (VS) following hypoxia secondary to cardiac arrest. Thirty-seven healthy participants with no history of neurological illness served as a control group. The activity of two facial muscles (*zygomaticus major*, *corrugator supercillii*) was measured using facial electromyography (EMG) to probe for patterned responses that differentiate between auditorily presented joke and non-joke stimuli in VS patients.

**Results:** One of the two VS patients we tested demonstrated greater *zygomatic* and reduced *corrugator* activity in response to jokes compared with non-jokes. Critically, these responses followed the pattern and temporal profile of muscle activity observed in our healthy control sample.

**Conclusions:** Despite their behaviourally non-responsive profile, some patients diagnosed as VS appear to retain some aspects of emotional experience.

**Significance:** Our findings represent, to our knowledge, the first demonstration that a patient diagnosed as VS can exhibit intact emotional responses to humor as assessed by facial EMG. Therefore, our approach may constitute a feasible bedside tool capable of providing novel insight into the mental and emotional lives of patients who are behaviourally non-responsive.

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## 1. Introduction

There is now a substantial body of evidence suggesting that patients with disorders of consciousness (DOC) can exhibit a remarkable degree of residual cognitive functioning despite their inability to respond overtly to command (Owen, 2013). For example, studies using modern neuroimaging methods (e.g., fMRI, EEG) have demonstrated that some DOC patients can wilfully modulate their brain activity in response to external commands (Monti et al., 2010; Owen et al., 2006; Cruse et al., 2011), and often exhibit a

strikingly similar pattern of neural activity to that of healthy controls across a wide range of cognitive tasks (Naci et al., 2014; Coleman et al., 2007). These advances have made the prospect of communication with these individuals possible through brain-computer interfaces (BCIs) that map reliable patterns of neural activity on to yes/no answers and other forms of wilful communicative responses in such patients (Monti et al., 2010; Cruse et al., 2011; see Naci et al., 2012, for review). Together, these findings have challenged the notion that such patients lack conscious awareness, suggesting instead that some of them are actively engaged with their environment despite their unresponsive outward appearance.

In the present study, we aim to build on these findings with a different methodological approach that uses recordings of facial

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muscle activity to examine emotional functioning in patients diagnosed as being in a vegetative state (VS). This approach is based on an extensive body of research demonstrating that different emotional states are associated with unique patterns of facial muscle activity (Ekman, 1992, 1993; Ekman et al., 1990; Cacioppo et al., 1986). Happiness, for instance, is characterized by the contraction of the *zygomaticus major* and *orbicularis oculi* muscles, which raise the corners of the mouth to create a smile, and raise the cheeks to form wrinkles around the eyes, respectively (Ekman et al., 1990). For the current purposes, these documented emotion-specific changes in facial muscle activity afford a unique and powerful opportunity to examine whether residual emotional processing in VS includes patterned changes in peripheral physiology. The advantages of this approach are threefold. First, the use of facial electromyography (EMG) at bedside to probe muscle activity in these patients provides a more feasible approach to assessing residual emotional processing compared with more invasive and costly neuroimaging methods. Second, the established specificity of facial muscle activity with respect to different emotional states (Cacioppo et al., 1986) may offer insight into the nature of the elicited emotional response in patients. Third, facial EMG provides a sensitive index of muscle activity, and can detect potentially subtle muscle responses in VS patients that cannot be discerned through visual observation (Cacioppo et al., 1986). These properties suggest that facial EMG may provide a promising window into emotional functioning in VS.

Currently, there is limited evidence pertaining to this issue, although a handful of neuroimaging studies have hinted that some forms of emotional processing may be at least partially preserved (Bekinschtein et al., 2004; Yu et al., 2013; Zhu et al., 2009; Di et al., 2007; Staffen et al., 2006). One such study reported activity in emotion-related brain regions, including the amygdala and insula, in a minimally conscious (MCS) patient that was unique to hearing a story read by the patient's mother as compared to a stranger (Bekinschtein et al., 2004). Similarly, a recent fMRI study in a group of VS patients found activity in 30% of them in the affective component of the pain matrix (i.e., network of regions associated with pain perception) in response to hearing another person crying out in pain (Yu et al., 2013). While these studies are certainly suggestive of residual emotional functioning in some VS patients, the interpretation of these neural responses remains somewhat unclear. Moreover, it is yet to be determined whether responses to emotional stimuli in this patient population are also associated with changes in peripheral physiology. Indeed, emotional experience in healthy adults is characterized not only by activation of affective centers in the brain, but also by patterned changes in autonomic and skeletal muscle activity (see Kreibig, 2010; Stephens et al., 2010; Levenson et al., 1990).

To examine whether any residual emotional functioning in VS can be observed with measures of peripheral physiological signals, we used facial EMG to measure activity from the *zygomaticus major* (smiling) and *corrugator supercilii* (frowning) muscles while patients and healthy controls listened to jokes and carefully matched non-joke stimuli. This approach was previously validated in a group of healthy control participants (Fiacconi and Owen, 2015), in which it was demonstrated that joke relative to non-joke stimuli elicited a robust increase in *zygomatic* activity and a reduction in *corrugator* activity. Note that one advantage of this approach is that, in addition to providing insight into emotional functioning, joke stimuli also necessarily draw on high-level language comprehension processes that underlie humor appreciation. We chose to use these stimuli based, in part, on previous neuroimaging work that has shown that some VS patients are in fact capable of complex language processing (Coleman et al., 2007, 2009). Therefore, an observed increase in *zygomatic* muscle activity in response to jokes would imply the preservation of complex

speech perception and language comprehension processes as well as intact emotional responses to humour. Here, we examined whether these abilities are preserved in some VS patients, as reflected in similar patterns of facial muscle activity to that of healthy controls.

## 2. Method

### 2.1. Healthy control participants

Thirty-seven healthy undergraduate students from Western University participated in this study in exchange for monetary compensation or course credit. There were 21 females ( $M = 22.8$  years,  $SD = 5.48$  years) with ages ranging from 17 to 41 years. No participants declared any history of neurological or psychiatric illness. One healthy control participant was excluded from all EMG analyses due to the presence of many large artifacts in the EMG signal from both the *zygomatic* and *corrugator* muscles. Seven participants were excluded from our analysis of onset latencies of the *zygomatic* response to jokes, as these participants' responses did not exceed the required slope threshold (see Section 2.5) during the time-window used for estimating onset latency. Data from this sample of healthy control participants was collected as part of a previous study (Fiacconi and Owen, 2015) investigating the relationship between muscle activity and cardiovascular changes associated with humor. All currently reported analyses and results concerning changes in muscle activity in the current study differ from those presented in our prior study.

### 2.2. Patients

We tested two patients (ages 35 and 49), both of whom received a diagnosis of VS based on behavioural assessment with the JFK coma recovery scale – revised (CRS-R) on the day of testing. The relevant demographic and clinical characteristics of each patient as well as their CRS-R scores are provided in Table 1. Patients were selected from a convenience sample to be closely matched in etiology (anoxia secondary to cardiac arrest) and behavioral profile. All experimental procedures were conducted after receiving informed consent from the legal guardian of the patient. All experimental procedures were approved by the Medical Research Ethics Board at Western University.

### 2.3. Materials

Stimuli consisted of 88 different sentences, half of which were jokes, and the other half non-jokes (Bekinschtein et al., 2011; Fiacconi and Owen, 2015). Both of these stimulus categories were made up of sentences that followed a common syntactic structure including an initial setup line, followed by a punchline. We operationally-defined the punchline as the critical phrase or word that allowed the global meaning of the sentence to be understood (e.g., Do you know what happens when frogs park illegally? They get towed; see Fiacconi and Owen (2015) for additional examples). Both stimulus types were closely matched in number of words, syllables, and syntactic structure. The duration of each sentence was on average 5.02 s, and ranged from 3.4 to 7.8 s. All sentences were recorded by a native English speaker using a lively prosody, rhythm, and intonation. For healthy control participants, the sound intensity level was set such that all participants heard the stimuli at a comfortable and clearly perceptible intensity. For patients, the sound level was adjusted to be slightly louder than that used in casual experimenter-patient interactions.

**Table 1**  
Relevant demographic and clinical characteristics for Patients 1 and 2.

Patient	Age/sex	Diagnosis/etiology	Time of Testing (days post-ictus)	CRS – R						Total score
				Auditory function	Visual function	Motor function	Oromotor/Verbal	Communication	Arousal	
1	36/M	VS/hypoxia secondary to cardiac arrest	6175	1 – auditory startle	1 – visual startle	2 – flexion withdrawal	1 – oral reflexive movement	0 – none	1 – eye opening w/stim.	6
2	51/F	VS/anoxia secondary to cardiac arrest	325	1 – auditory startle	0 – none	1 – abnormal posturing	1 – oral reflexive movement	0 – none	1 – eye opening w/stim.	4

Facial EMG was recorded from the *zygomaticus major* and *corrugator supercilii* muscles, with a bipolar electrode setup consisting of two 8 mm Ag/AgCl electrodes placed over each participants' left cheek (*zygomatic*), and left brow (*corrugator*), with each pair separated by 1 cm. A single reference electrode was placed on the forehead at the midline. Each electrode was fitted with an adhesive collar and filled with conductive gel (5% NaCl) to increase the signal-to-noise ratio. Electrodes were placed on the skin in muscle-specific locations specified by published guidelines (Fridlund and Cacioppo, 1986). All signals were recorded using a BIOPAC MP150 system equipped with EMG100C-MRI amplifiers.

The experiment was conducted on a Dell laptop computer running E-prime 2.0 software, and stimuli were presented through a pair of noise-cancelling headphones.

#### 2.4. Experimental procedures

For healthy controls, the experimental procedures are outlined in detail in a prior publication (Fiacconi and Owen, 2015). Here, we provide a summary of the most important procedural details. Participants were first set up with the aforementioned psychophysiological measures, and were then asked to listen carefully to each sentence, and to rate the perceived degree of humor for each sentence on a scale from 1 to 7, with 1 indicating not funny, and 7 corresponding to extremely funny. To allow participants to acclimatize to the experimental settings, they first heard four practice sentences that were similar in structure to the remaining experimental sentences. Following the rating response on each trial, there was a 12–18 s inter-stimulus interval (ISI) to facilitate the acquisition of the psychophysiological data. The order of presentation for both sentence categories was randomly intermixed. The experiment took roughly 50 min, after which time participants were debriefed.

For patients, the experimental setup was similar with a few exceptions. First, patients were not asked to provide a rating as to how funny they perceived the sentence. Second, to shorten the overall length of the experiment for patients, we used a subset of the joke and non-joke stimuli. Specifically, we chose the funniest 25 jokes, and the least funny 25 non-jokes as rated by healthy controls, with the goal of maximizing the likelihood of detecting preserved emotional responses in patients. All other experimental parameters were identical to those used for healthy controls.

#### 2.5. Data reduction and analysis

EMG data from both muscle sites were recorded with a sampling rate of 2000 Hz, amplified, and band-pass filtered from 30 to 500 Hz. Ambient line-noise (60 Hz) was subsequently removed using a notch-filter with cutoffs at 59.5 and 60.5 Hz. The filtered data were then full-wave rectified by computing the absolute value of the filtered EMG signal, and low-pass filtered at 2 Hz to obtain the envelope of the signal. The resulting data were then baseline-corrected by subtracting the mean EMG activity from the 1-s interval preceding the punchline onset from that occurring post-punchline. We excluded trials with extensive artifacts during

either the baseline period or the subsequent response period on the basis of visual inspection, blind to condition. For the healthy controls 16.6% and 19.6%, of trials were excluded for the *zygomatic* and *corrugator* muscles, respectively. For patients, 11.0% and 16.0% of trials were excluded for the *zygomatic* and *corrugator* muscles, respectively. Changes in muscle activity in response to each sentence were assessed on a trial-by-trial basis by time-locking our analyses to the onset of the critical punchline. Specifically, we subtracted the mean baseline activity (1 s prior to the punchline) from all muscle activity occurring post-punchline, and then averaged this baseline-corrected activity over the first 6 s following the punchline onset to obtain an index of activity change to jokes and non-jokes (see Bradley et al., 2001; Fiacconi and Owen, 2015, for a similar approach).

To assess the reliability of differential EMG responses to jokes and non-jokes for both individual healthy control participants and patients, we used a bootstrapping approach (see Oruc et al., 2011, for a similar approach applied to ERP data). After calculating the mean activity over 6 s following punchline onset for each trial in each condition (i.e., jokes, non-jokes), we randomly selected (with replacement) a new sample of trial values for each condition and computed the mean EMG response across these newly sampled trial values. This procedure was then repeated 10,000 times. For each of these iterations, difference scores for the *zygomatic* muscle were created by subtracting the mean activity elicited by non-joke stimuli from the mean activity elicited by joke stimuli for a given iteration. For the *corrugator* muscle, these difference scores were calculated by subtracting the response to jokes from that to non-jokes. Based on this distribution of bootstrapped mean difference scores, we then constructed 95% confidence intervals to assess the reliability of the difference scores. If the *zygomatic* EMG response to jokes is greater than that to non-jokes, we expected that our confidence interval around the mean difference score between these conditions should not include zero. Similarly, if non-jokes elicit greater *corrugator* EMG activity than jokes, then our obtained confidence interval should also exclude zero.

We operationally defined the initial inflection point in the *zygomatic* response to the joke stimuli as the point in time at which the instantaneous slope of the baseline-corrected EMG signal first exceeded 2  $\mu\text{V/s}$  within a 4-s window following punchline onset. This criterion was chosen based on visual inspection of the healthy control data, and was calculated for both patients and controls based on their overall trial-averaged waveforms. Seven healthy controls were excluded from analysis of the mean initial inflection point because the instantaneous slope of their trial-averaged waveforms did not exceed 2  $\mu\text{V/s}$  during the critical time-window of interest. These seven participants tended to exhibit a relatively small, if any, smiling response to the joke stimuli.

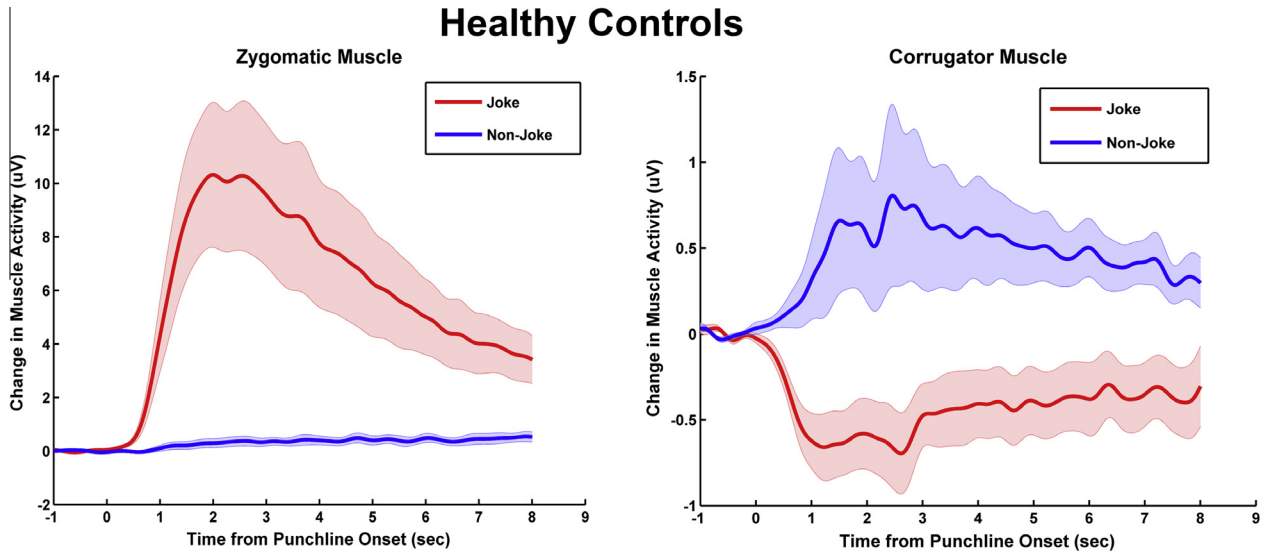
### 3. Results

To facilitate direct comparison with the patient data, we analyzed only the subset of joke and non-joke stimuli that were presented to the patients in our sample of healthy controls.

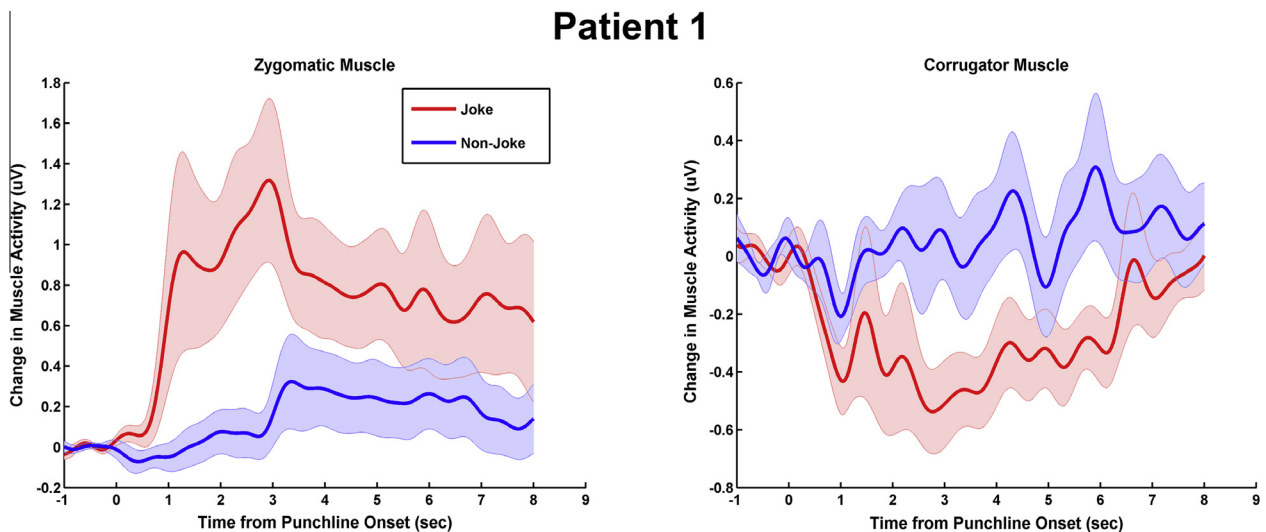
The mean EMG activity time-locked to the onset of the punchline for each muscle site, for patients 1 and 2, along with that of healthy controls, is depicted in Figs. 1–3. The results for each muscle site are reported separately.

For the *zygomatic* muscle, jokes elicited greater mean EMG activity during the first 6-s post-punchline than non-jokes in healthy control participants  $t(35) = 3.38, p < .001$  (one-tailed), indicating that jokes elicit a greater smiling response. At the individual participant level, this effect was statistically significant in 31 of our 36 healthy controls (~86%) as determined by bootstrapped 95% confidence intervals calculated for each participant. To estimate the “moment of insight” in humor comprehension, we measured the latency of the initial inflection point of *zygomatic* activity following the punchline of joke (see Section 2.5). For healthy control participants, this “moment of insight” was found to occur roughly 650 ms ( $M = 663$  ms, 95% CI [470, 856]) following punchline onset. With respect to Patient 1, *zygomatic* muscle activity for jokes was also greater than that for non-jokes (95% CI for difference score

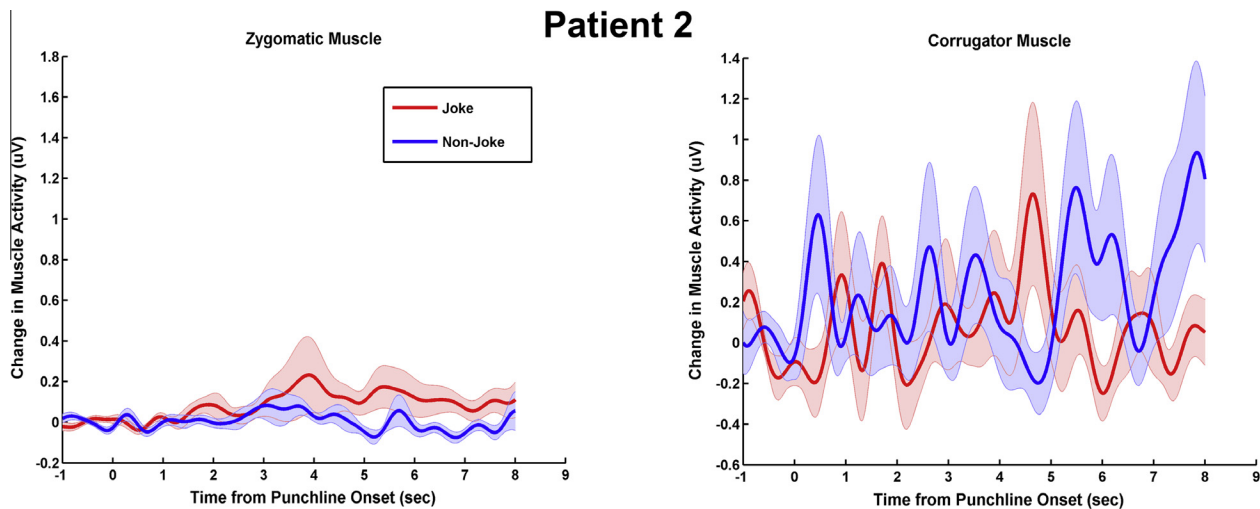
[.14, 1.21]), and followed a similar, albeit slightly delayed temporal profile (estimated smile onset latency = 887 ms) as compared to healthy participants. By contrast, for Patient 2, we did not observe any reliable differences in *zygomatic* muscle activity for jokes as compared to non-jokes (95% CI for difference score [−.01, .20]). To quantify the similarity of the *zygomatic* EMG response profile to jokes between Patient 1 and the healthy controls, we computed a Pearson’s correlation coefficient between the mean time course observed in Patient 1 and that obtained in each of our healthy control participants. This procedure was also repeated with mean time course obtained from Patient 2. We therefore obtained a series of 36 correlation coefficients for each patient representing the similarity between their *zygomatic* response profile to jokes and that for each healthy control participant. Given that Pearson correlation coefficients are non-normally distributed, we applied a Fisher’s  $z$ -transformation to all coefficients prior to statistical analyses. For ease of interpretation, however, we report descriptively the Pearson correlation coefficient derived from the mean  $z$ -transformed



**Fig. 1.** Facial EMG data for healthy control participants. Group-averaged facial EMG responses obtained in healthy control participants for each muscle site and condition. Shaded region depicts standard error of the mean.



**Fig. 2.** Facial EMG data for Patient 1. Trial-averaged facial EMG responses plotted separately for each muscle site and condition. Note the different scaling on y-axis relative to Fig. 1. Shaded region depicts standard error of the mean.



**Fig. 3.** Facial EMG data for Patient 2. Trial-averaged facial EMG responses plotted separately for each muscle site and condition. Note the different scaling on y-axis relative to Fig. 1. Shaded region depicts standard error of the mean.

coefficient for each patient. As expected, the mean correlation coefficient describing the response similarity between Patient 1 and healthy controls (.77) was significantly greater than that describing the response similarity between Patient 2 and healthy controls (.46)<sup>1</sup>,  $t(70) = 5.42$ ,  $p < .001$ . These results confirm that the observed *zygomatic* response to jokes in Patient 1 was more similar to healthy controls than the observed response in Patient 2.

For *corrugator* EMG activity, jokes elicited less mean EMG activity during the first 6-s post-punchline than non-jokes in healthy control participants,  $t(35) = 2.51$ ,  $p < .017$ . Of our sample of 36 healthy controls, 18 (50%) demonstrated this effect reliably based on bootstrapped 95% confidence intervals calculated for each individual. These findings validate the EMG data collected from the *zygomaticus* muscle, suggesting that the observed activity in this muscle did indeed reflect smiling and not simply a global increase in muscle activity. Again, similar to healthy controls, Patient 1 demonstrated less *corrugator* muscle activity in response to jokes relative to non-jokes (95% CI for difference score [.08, .71]). Such a pattern was not observed for Patient 2, who failed to elicit differential *corrugator* activity between these two types of stimuli (95% CI for difference score [−.30, .51]). Following the same procedure used to assess response similarity for the *zygomatic* muscle, we also quantified the similarity of the *corrugator* EMG response profile to jokes between each patient and healthy controls. Again, we found that the mean correlation coefficient describing the response similarity between Patient 1 and healthy controls (.32) was significantly greater than that describing the response similarity between Patient 2 and healthy controls (−.04),  $t(70) = 3.60$ ,  $p < .001$ , suggesting that the observed *corrugator* response to jokes

in Patient 1 was more similar to healthy controls than the observed response in Patient 2.

#### 4. General discussion

In the present study, we provide to our knowledge the first demonstration that a VS patient can exhibit intact emotional responses to humor as assessed by recordings of facial muscle activity. Paralleling the pattern of results observed in our sample of healthy control participants, we found that one of the two VS patients tested showed greater *zygomatic* and reduced *corrugator* muscle activity to auditorily presented jokes relative to non-jokes. These results suggest that in addition to increased positive affect, jokes may also be associated with less negative affect than non-jokes in both healthy controls and Patient 1 (see Larsen et al., 2003 for a discussion of the relative sensitivity of each muscle to positive and negative affect). Moreover, the temporal profile of the *zygomatic* response to jokes in Patient 1 was highly similar to that of our healthy controls, suggesting that the dynamics of the high-level language processing that support humor comprehension are also preserved. These findings stand in stark contrast with Patient 1's non-responsive behavioural profile as determined by bedside testing. Together, our results provide novel support for the growing consensus that some VS patients are capable of complex high-level cognitive processing despite their inability to respond to command, and point to facial EMG as a promising tool to detect differences in cognitive and emotional functioning between patients who are indistinguishable at the level of behavioural assessment.

Although some neuroimaging studies using fMRI in VS patients have reported activity in brain regions implicated in emotional processing (Bekinschtein et al., 2004; Yu et al., 2013; Staffen et al., 2006), there is little evidence to date on whether such patients can generate peripheral signatures of emotion, nor what these fMRI responses mean specifically. This question is of clinical and practical importance, as measures of peripheral changes are relatively inexpensive and less invasive compared with neuroimaging techniques (e.g., fMRI). Therefore, evidence that VS patients can in fact exhibit changes in autonomic and/or skeletal muscle activity in response to emotional stimuli would suggest that such measures may constitute a feasible bedside methodology for assessing emotional functioning in DOC, with the potential to detect covert awareness in some of these patients. Some previous

<sup>1</sup> Although the mean correlation coefficient (.46) for the *zygomatic* muscle that describes the response similarity between Patient 2 and healthy controls is relatively large (and statistically greater than zero), we feel that this result is somewhat misrepresentative of the response similarity to healthy controls in this patient. This correlation coefficient is likely inflated due to the tendency for the *zygomatic* muscle to preferentially increase in activity relative to baseline. As such, the range of variability with respect to the activity of this muscle is restricted, which likely results in artificially inflated correlation coefficients. For this reason, we believe the relative comparison between the mean correlation coefficients obtained for Patient 1 and Patient 2 is most informative. Moreover, given that we did not observe a differential EMG response to jokes as compared to non-jokes in Patient 2, it makes little sense to interpret the similarity in response profile to jokes between Patient 2 and healthy participants in and of itself. We include the mean correlation coefficient for Patient 2 for completeness, but caution against interpreting it as a meaningful metric of response profile similarity with healthy controls.

support for this possibility was reported in a recent study in coma patients (Daltrozzo et al., 2010), in which it was found that such patients demonstrated a larger skin conductance response to auditorily presented emotional stimuli, and that this differential response was positively related to behavioural indices of coma recovery. While this finding is certainly consistent with the present results, it does not directly address conscious emotional functioning in VS. Moreover, measures of overall autonomic arousal (i.e., skin conductance) provide little information on the valence of emotional response, and can be driven by cognitive as well as emotional factors (Dawson et al., 2000). By contrast, the use of facial EMG provides a direct index of the valence of emotional experience, and can therefore shed light on the nature of emotional responses in VS. In addition, by time-locking our EMG analyses to the critical word of each joke (i.e., the punchline), we were able to pinpoint the determinant of the observed responses, ensuring that any changes in muscle activity were in fact related to humor appreciation rather than prosody or intonation. These advantages highlight the potential breadth of insight into emotional functioning afforded by facial EMG.

As we alluded to earlier, changes in facial muscle activity related to humor not only allow for conclusions regarding the emotional functioning of VS patients, but also speak to the presence of residual speech perception and language comprehension processes. That such high-level processes were clearly preserved in Patient 1 is in line with previous neuroimaging studies documenting differential patterns of neural activity to semantically ambiguous versus unambiguous sentences in some VS patients (Coleman et al., 2007, 2009). Importantly, these differential patterns of neural activity mirrored that observed in healthy controls, implying that these patients were capable of comprehending the meaning of complex linguistic materials. These findings are particularly relevant to the current study, as the comedic value of many of our joke stimuli hinged on the presence of semantic ambiguity (see Fiacconi and Owen (2015) for specific examples). Therefore, our results build on previous neuroimaging evidence and suggest that Patient 1 can not only resolve semantic ambiguity, but can also appreciate the comedic value that arises from the unexpected juxtaposition of different meanings of the same word. While it is currently not possible to rule out that this could occur in unconscious states, given the cognitive complexity involved in this process, it seems highly unlikely. Thus, this simple and relatively cheap technique could add to the growing battery of tools available for detecting covert awareness in patients believed to be in a vegetative state (Owen et al., 2006; Cruse et al., 2011; Naci et al., 2014).

The preserved emotional response to humor observed in Patient 1 is also noteworthy when considered together with previous neuroimaging evidence obtained from this specific patient (Naci et al., 2014; Naci and Owen, 2013). In particular, this patient was able to follow commands as demonstrated by greater bilateral neural activity in temporal cortex to auditorily presented spoken words when instructed to count these words relative to when he was asked to relax during their presentation (Naci and Owen, 2013). Furthermore, dynamic changes in this patient's neural activity in auditory, visual, and executive control regions during viewing of a movie were found to correlate with that of healthy control participants in the very same brain areas (Naci et al., 2014). This latter finding is particularly intriguing, as this patient often attends the movie theatre with his father, who reports that the patient can in fact comprehend the narrative content of the film. Our results are especially informative in this context, as they suggest that this patient may in fact be able to appreciate the humorous content often depicted in movies. Further testing in this patient that combines facial EMG with movie stimuli may be able to confirm this possibility.

While Patient 1 did reliably distinguish between jokes and non-jokes on the basis of recorded *zygomatic* and *corrugator* muscle activity, the mean response amplitude for both muscles was smaller than that of our healthy controls. This is particularly obvious for the *zygomatic* response to jokes (see left panel of Figs. 1 and 2). The reason for this disparity in mean response activity is currently unclear, but it should be noted that 10 of our 36 healthy controls exhibited equal or lesser *zygomatic* activity to jokes than Patient 1. Therefore, while smaller on average, the *zygomatic* response to jokes observed in Patient 1 is still well within the range of response magnitudes elicited in our healthy control participants. With respect to *corrugator* activity, the mean response magnitude to jokes for Patient 1 was similar, albeit slightly less than the corresponding mean response in our healthy controls. For Patient 1, the differential response to jokes relative to non-jokes in this muscle appeared to be driven by a decrease in muscle activity to jokes, as non-jokes failed to elicit the same increase in muscle activity observed in the healthy controls.

Although we did not observe differential *zygomatic* or *corrugator* activity to jokes and non-jokes in Patient 2, we note that it is difficult to conclude with certainty that this patient was unable to comprehend the meaning of the presented sentences. In fact, 5 out of our 36 (~14%) healthy control participants also did not exhibit a reliable difference in *zygomatic* activity to jokes as compared to non-jokes. Such a conclusion is even more problematic with respect to the *corrugator* muscle, as a reliable difference in activity to jokes was detected in only 50% of our healthy controls. Therefore, it is possible that despite the observed lack of facial muscle activity differences between jokes and non-jokes in Patient 2, this patient may nonetheless comprehend our humorous sentences. On the other hand, these findings do suggest that *zygomatic* muscle activity provides a more sensitive marker of humor comprehension than *corrugator* activity, and that future studies using facial EMG to detect humor processing in behaviourally non-responsive patient populations may wish to prioritize the collection of data from the *zygomatic* muscle.

We should also note here that we are not claiming that the majority of VS patients are capable of the complex cognitive and emotional processing that we observed in Patient 1. Given that we tested only two patients, our sample size is not sufficient to draw conclusions regarding the proportion of such patients who exhibit intact emotional responses to humor. However, the point of our paper is to highlight that it is in fact possible to measure such responses in a VS patient that is entirely behaviourally non-responsive, and to suggest that the novel methodological approach employed here (i.e., facial EMG) could be of utility in examining emotional responses in other VS patients, as well as in other patient populations who are incapable of overt behavioural communication.

Although much of our understanding of the residual capacities of DOC patients has come from the use of modern neuroimaging tools (e.g., fMRI, EEG), the present study demonstrates the utility of facial EMG as a more cost-effective, and less invasive alternative methodology that can nevertheless reveal important insights into the mental lives of such patients. The observed differentiation between jokes and non-jokes at the level of facial muscle activity in Patient 1 also raises the question of whether such responses could be adapted for use as part of a BCI. Although facial EMG is not a direct measure of neural activity, it may nonetheless be possible to use such responses for the purpose of classifying stimuli in terms of their affective properties, and perhaps even for communication. Given the well-established links between specific emotional states and discrete patterns of facial muscle activity (Ekman, 1992, 1993; Ekman et al. 1990; Cacioppo et al., 1986), we propose that facial EMG holds the potential to provide a rich

window into emotional functioning in individuals who are incapable of overt behavioural communication.

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**Conflict of interest:** None.

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