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Somatosensory Attention Identifies Both Overt and Covert Awareness in Disorders of Consciousness

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Objective: Some patients diagnosed with disorders of consciousness retain sensory and cognitive abilities beyond those apparent from their overt behavior. Characterizing these covert abilities is crucial for diagnosis, prognosis, and medical ethics. This multimodal study investigates the relationship between electroencephalographic evidence for perceptual/cognitive preservation and both overt and covert markers of awareness.
Methods: Fourteen patients with severe brain injuries were evaluated with an electroencephalographic vibrotactile attention task designed to identify a hierarchy of residual somatosensory and cognitive abilities: (1) somatosensory steady-state evoked responses, (2) bottom-up attention orienting (P3a event-related potential), and (3) top-down attention (P3b event-related potential). Each patient was also assessed with a clinical behavioral scale and 2 functional magnetic resonance imaging assessments of covert command following.
Results: Six patients produced only sensory responses, with no evidence of cognitive event-related potentials. A further 8 patients demonstrated reliable bottom-up attention-orienting responses (P3a). No patient showed evidence of top-down attention (P3b). Only those patients who followed commands, whether overtly with behavior or covertly with functional neuroimaging, also demonstrated event-related potential evidence of attentional orienting.
Interpretation: Somatosensory attention-orienting event-related potentials differentiated patients who could follow commands from those who could not. Crucially, this differentiation was irrespective of whether command following was evident through overt external behavior, or through covert functional neuroimaging methods. Bedside electroencephalographic methods may corroborate more expensive and challenging methods such as functional neuroimaging, and thereby assist in the accurate diagnosis of awareness.

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Disorders of consciousness (DoC) are states that a person may enter when they emerge from coma following a severe brain injury. Patients in a vegetative state (VS) do not demonstrate purposeful behavior and are considered to lack awareness.[1–3] In contrast, patients in a minimally conscious state (MCS) are considered to have fluctuating awareness and demonstrate variable, but reproducible, purposeful behavior.[4] Furthermore, the MCS can be subdivided into MCS$^+$ or MCS$^-$ on the basis of the patient's ability to follow commands.[5] Patients who demonstrate accurate communication and/or functional object use are considered emergent from an MCS (EMCS).[4] However, the accurate identification of a patient's diagnostic group comprises a considerable clinical challenge.[1–3,6–8]

To facilitate more accurate diagnosis of DoC, researchers have developed brain imaging paradigms to assess volition and command following in the absence of outward responsiveness.[9–14] Patients who produce behavior consistent with a VS, but who exhibit evidence of covert awareness with functional neuroimaging—such as imagining movements to command[6,9,10,13,15–17]—have

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been considered to exhibit a nonbehavioral MCS.[18] However, in both behavioral and neuroimaging-based assessments, a patient may produce a false negative due to fatigue or insufficient cognitive resources to successfully complete the demanding diagnostic task.[8,19]

Researchers have developed assessments of brain function to place a patient along a hierarchy of increasingly complex attentional information processing.[20–24] However, there are inconsistencies in the prognostic value of the event-related potentials used in these hierarchical approaches; some investigators have reported positive prognostic value in these attentional markers,[25] whereas others have not.[26] These inconsistencies may have occurred because multimodal assessments were not used to identify patients in a nonbehavioral MCS. Therefore, 15% of the patient sample considered to be in a VS may have possessed a nonbehavioral MCS and consequently may have misrepresented the diagnostic category.[27] Similarly, most studies of patients with DoC employ auditory stimulation because many patients lack oculomotor control; however, this tendency limits the characterization of a patient's sensory abilities to the auditory domain.

We report a hierarchical cognitive assessment in a sample of 14 patients with severe brain injuries using vibrotactile stimulation. The assessment employed an oddball paradigm to elicit steady-state evoked responses of sensory processing and event-related potential (ERP) markers of bottom-up and top-down attention (the P3a and P3b, respectively).[28] As with previous hierarchical designs, this approach discretizes a patient's sensory and cognitive abilities. A novel aspect of our method is the assessment of a patient's ability to sense and attend to touch. Importantly, patients were also evaluated using 2 previously established neuroimaging-based assessments of covert command following—mental imagery[6,9,10,13,15–17] and selective auditory attention[29,30]—and a clinical behavioral assessment.[31] By identifying patients with covert command-following abilities, these additional assessments ensured a more accurate representation of each patient's level of awareness. Furthermore, we were in a position to test the divergence and convergence of these methods. It was expected that ERP markers of higher-order attention would be evident in patients who were aware, expressed either overtly in their behavior, or covertly by willful modulations of brain activity detected with neuroimaging.

Subjects and Methods

Participants

Fourteen patients (mean age = 41 [range = 19–58] years) contributed sufficient data for inclusion in this investigation. Seven patients were diagnosed as VS,[3] 4 patients were diagnosed as MCS, 2 patients were diagnosed as EMCS[4], and 1 patient was diagnosed with locked-in syndrome (LIS).[32] Six patients had sustained traumatic brain injuries from motor vehicle accidents. The remaining 8 patients had sustained nontraumatic brain injuries from different etiologies including cardiac arrest (3 cases) and near-drowning (1 case; see Supplementary Table 1). Each patient's surrogate decision maker provided informed, written consent for the patient's participation in the study. Ethical approval was obtained from the University of Western Ontario's Health Sciences Research Ethics Board (London, Ontario, Canada).

As a scientific control, a sample of 15 healthy volunteers also participated in the somatosensory selective attention task. These participants ranged in age from 17 to 23 years (mean age = 18 years). All healthy volunteers provided informed written consent and received course credit for their participation. The Psychology Research Ethics Board of the University of Western Ontario (London, Ontario, Canada) provided ethical approval for the control study. Control studies of the other neuroimaging paradigms have been reported elsewhere.[15,30,33]

Procedure

For each patient, participation in this study comprised assessments with: (1) electroencephalography (EEG) during their completion of a somatosensory selective attention paradigm, (2) functional magnetic resonance imaging (fMRI) during their completion of a mental imagery paradigm,[6,9,10,13,15–17] (3) fMRI during their completion of an auditory selective attention paradigm,[29,30] and (4) the Coma Recovery Scale–Revised (CRS-R[31]; see Supplementary Tables 1 and 2). fMRI data from Patient EMCS2 could not be analyzed due to excessive motion artifacts. However, this patient was included in this investigation because his ability to follow simple commands and communicate was evident from his overt behavior. Similarly, the data for Patient VS7 from 1 fMRI session (selective auditory attention) were discarded due to excessive movement. This patient was included in the current investigation because useable data were obtained from this patient for the other 3 paradigms.

All patients completed the 2 fMRI paradigms within a 2-day period. Ten patients completed the fMRI assessments within 2 days of their EEG assessments (see Supplementary Table 2). The other 4 patients completed the EEG assessments after the fMRI assessment with the following delay: 1.5 months (EMCS1); 7.5 months (MCS3); 1 year (VS3); and 3.5 years (VS7). Only Patient MCS3 demonstrated a clinical status change between assessments with EEG and fMRI (MCS^{-} to MCS^{+}). Given the etiology, age, and time postictus of those patients with 1 year or more between assessments (see Supplementary Table 2), it is unlikely (although not impossible) that either of these patients underwent a change in their conscious states between assessments.[1–3] Patients VS3 and VS7 demonstrated overt behavior consistent with a VS at all assessments.

Somatosensory Selective Attention Paradigm

Participants completed a short somatosensory selective attention task as their EEGs were recorded. One stimulator was affixed

TABLE 1. Number of Trials Available for the Analyses of the Electroencephalographic Data from the Somatosensory Selective Attention Paradigm following Artifact Rejection

Subjects	Stimulus Type[a] M (MIN–MAX)			
	Upper Back	Target Wrist	Nontarget Wrist	Trials Rejected, %
Patients, n = 14	2,614 (1,591–3,246)	313 (188–384)	311 (180–388)	35 (20–59)
Controls, n = 15	2,890 (2,718–5,026)	345 (327–363)	345 (321–359)	25 (20–32)

[a]A 2 × 3 chi-square goodness of fit test indicated that the minimum number of trials in each of the 3 stimulus types did not significantly differ between the controls and patients, $\chi^2(2) = 0.21$, $p = 0.9$.
M = mean; MAX = maximum; MIN = minimum.

to each wrist and the upper back (3 total). Each stimulator administered nonpainful vibrotactile stimuli via a motor housed in a rubberized casing.[34] A similar paradigm has also been evaluated for patients with LIS.[35] The experiment comprised 14 blocks. Participants were presented with a series of vibrations alternating among their wrists (10% per wrist) and upper back (80%). A vibration occurred every 200 milliseconds and lasted for 50 milliseconds. The number of vibrations presented to each wrist in a block was selected on a random uniform interval from 28 to 32. There was always a minimum of 3 (maximum = 21) upper back stimuli between wrist vibrations; on average, 49% (standard deviation = 13%) of the wrist stimuli followed exactly 3 upper back stimuli. Participants were instructed to count the vibrations presented only to the target wrist. The experimenter touched the patient's target wrist after the instruction. The right wrist was always the target wrist for the first block and subsequently alternated between the left and right wrists. The healthy volunteers reported their count at the end of each block; these participants reported the correct number of vibrations for 12 of 14 blocks on average (all reports were within ±3 of the true number of targets). One block of trials lasted for approximately 1 minute.

Mental Imagery Paradigm

During an fMRI scan, patients were asked to engage in 2 mental imagery paradigms.[6,9,10,13,15–17] In the motor imagery task, patients were instructed to imagine swinging their right arm to hit a tennis ball. In the spatial navigation task, patients were instructed to imagine walking from room to room in their house and visualize all objects they would encounter. Instructions were delivered with noise cancellation headphones (Silent Scan [Avotec, Stuart, FL] for patients scanned in the Trio system, as well as Patient VS6 [first visit], and Sensimetrics S14 [Sensimetrics Corporation, Malden, MA] for the patients scanned in the Prisma system, including Patient VS6 [second visit]). Patients VS1, VS2, VS4, VS5, VS6 (second visit), MCS4, and EMCS1 completed 2 sessions of each task, whereas patients VS3, VS6 (first visit), VS7, MCS1, MCS2, MCS3, and LIS1 completed only 1 session due to scanner availability or patient fatigue. Each task alternated five 30-second blocks of mental imagery and five 30-second blocks of rest for a total of 5 minutes.

Auditory Selective Attention Paradigm

The fMRI selective auditory attention paradigm has been previously described in healthy individuals[30] and patients with DoC,[29] and is designed to identify an ability to follow commands to selectively attend to stimuli—that is, top-down attention. On each trial, participants were instructed either to count a target word ("yes" or "no") presented among pseudorandom distractors (spoken digits 1–9), or to relax. Each trial had an on/off design: sound (~22.5 seconds) followed by silence (10 seconds). The scan lasted 5 minutes, including instructions.

Replication Data

Patients VS4, MCS3, and EMCS1 participated in second assessments with the somatosensory selective attention task and the CRS-R. These assessments occurred from 2 to 3.5 months following their initial participation. Patient VS6 completed a second assessment with all paradigms (CRS-R, fMRI, and EEG) 22 months after her initial assessment. All 4 patients maintained their clinical status at follow-up (see Supplementary Table 2).

EEG Data Acquisition and Preprocessing

EEG data were recorded at sites FC1, Fz, FC2, C3, Cz, C4, CP1, CP2, Pz, Oz, PO7, and PO8 using an electrode cap with the g.Gamma active electrode system (g.tec Medical Engineering, Schiedlberg, Austria). This montage was selected following a previous study conducted in patients with LIS[35] and previous work concerning optimal P300 classification.[36] Data were sampled at 256Hz and filtered between 0.5 and 30Hz using a digital Butterworth filter. Stimuli were presented with the g.VIBROstim box (g.tec Medical Engineering) using a custom MATLAB script for Simulink (MathWorks, Natick, MA). The recordings were referenced to the right earlobe with a forehead (Fpz) ground. Impedances were kept below 5kΩ. Data processing was conducted with EEGLAB.[37] The data were segmented into 1-second epochs with a 200-millisecond prestimulus period, and linear detrending and baseline correction were applied to each epoch. For artifact correction, all trials containing data with voltages exceeding ±100 μV were rejected. In a second step, the kurtosis of the signal across all channels was calculated for each stimulus type separately, and all trials exceeding 2.5

standard deviations of the mean were rejected. Final trial numbers are reported in Table 1.

fMRI Data Acquisition and Preprocessing

The MRI data were acquired with a 3T Siemens scanner (Siemens, Erlangen, Germany) with a Siemens 32-channel head-coil at the Centre for Functional and Metabolic Mapping at Robarts Research Institute, University of Western Ontario, London, Ontario, Canada. The patients were recruited over 30 months, in which time the 3T scanner was upgraded. Three patients (VS3, VS7, and MCS3) were scanned in a Magnetom Trio system. All other patients were scanned in a Magnetom Prisma system. Functional echo-planar images of 36 slices covering the whole brain were acquired (repetition time = 2,000 milliseconds, echo time = 30 milliseconds, matrix size = 420 × 420, slice thickness = 3mm, in-plane resolution = 3 × 3mm, flip angle = 78°; for patients VS6 and LIS1 only, matrix size = 384 × 384 and flip angle = 75°). High-resolution T1-weighted 3-dimensional images were acquired in the same session (Trio system: repetition time = 2,300 milliseconds, echo time = 2.98 milliseconds, inversion time = 900 milliseconds, matrix size = 256 × 240, voxel size 1 × 1 × 1mm, flip angle = 9°; Prisma system: repetition time = 2,300 milliseconds, echo time = 2.32 milliseconds, inversion time = 900 milliseconds, matrix size = 256 × 256, flip angle = 8°; for patients VS6 and LIS1 only, matrix size = 240 × 256 and flip angle = 9°). Data from the mental imagery paradigm were preprocessed using SPM8 (http://www.fil.ion.ucl.ac.uk/spm), as described elsewhere.[13] For the selective attention paradigm, preprocessing was performed with AA software.[38]

Statistical Analyses

EEG RESPONSES. The EEG data were assessed for the presence of a steady-state evoked potential to the repetitive vibrotactile stimulation. As 1 vibration occurred every 200 milliseconds, an evoked response was considered present when the averaged peak of the frequency spectrum of the data at the stimulation rate (5Hz) and its first harmonic (10Hz) was significantly higher than the background noise.[39] A frequency spectrum was calculated with a discrete Fourier transform over the entire 1-second epoch from the average of all trials using data only from site Pz.[40,41] An F ratio (alpha = 0.05; $F_{2,20} \geq 3.49$) was computed to compare the power at 5 and 10Hz with the average power in the 10 adjacent ∼1Hz frequency bins (2–4Hz, 6–9Hz, and 11–13Hz).[39]

Two analyses of the EEG data were conducted to identify the attention-based ERPs. For the bottom-up attention effect (P3a), responses to wrist (deviant) and upper back (standard) stimuli were compared. A random subset of the standard stimuli (equal in number to the deviant stimuli) was selected, because there were many more standard than deviant stimuli. For the top-down attention effect (P3b), responses to the target and nontarget wrist stimuli were compared. Trial numbers were matched between the target and nontarget trials. Data from 50 to 750 milliseconds poststimulus were analyzed using the cluster-mass procedure[42] of the MATLAB toolbox FieldTrip.[43] This technique has been described in detail previously.[42,44] In the first step, data were compared at each time point using a t test. In the second step, t values of adjacent spatiotemporal points with $p < 0.05$ were clustered together by summating their t values. The largest cluster was retained. This entire procedure was repeated 1,000 times with recombination and randomized resampling of the ERP data. This Monte Carlo method generated a nonparametric estimate of the p-value representing the statistical significance of the originally identified cluster.

BLOOD OXYGEN LEVEL–DEPENDENT MENTAL IMAGERY RESPONSES. Single-subject fixed-effect analyses were performed for each patient. The analysis was based on the general linear model using the canonical hemodynamic response function[45] implemented with SPM8 (http://www.fil.ion.ucl.ac.uk/spm). The analysis pipeline was previously reported.[13] Linear contrasts were used to obtain subject-specific estimates, and results were thresholded at a voxel level, familywise error (FWE), whole-brain $p < 0.05$. When no significant activations were found at this level, the statistical threshold was reduced to an uncorrected $p < 0.001$ because of the strong anatomical a priori hypotheses.[6,9,10,13,15–17] This less conservative threshold excluded the possibility of failing to detect more subtle changes in the signal.[45,46]

Blood Oxygen Level–Dependent Auditory Selective Attention Responses

The general linear model (SPM8) was used to explore effects of interest. Two event types were defined corresponding to the on/off periods (count/relax; ∼22.5 seconds, or vice-versa). The silent period (10 seconds) served as an implicit baseline for all trials. Events for these regressors were modeled by convolving boxcar functions with the canonical hemodynamic response function. Also included in the general linear model were the following nuisance variables: the movement parameters in the 3 directions of motion and 3 degrees of rotation, and the mean of each scan. Linear contrasts were used to obtain subject-specific estimates for the effect of interest. Clusters that survived the $p < 0.05$ threshold after the FWE correction were reported as significant.

Results

All patient outcomes are summarized in Figure 1 and Supplementary Table 3.

EEG Responses

A steady-state evoked potential was detected in the EEG data of all patients (n = 14) and all healthy volunteers (n = 15; Fig 2).

Bottom-up attention effects (deviant vs standard stimuli) were detected in 8 patients and all of the healthy volunteers (n = 15; Fig 3). All patients who demonstrated a differential response to the deviant versus standard stimuli also demonstrated evidence of command

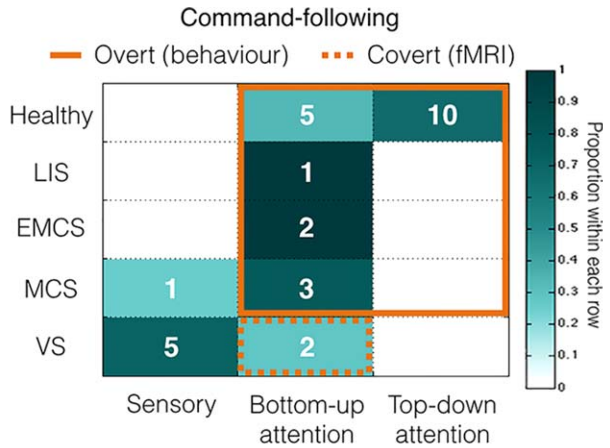

FIGURE 1: Summary of the relationship between command following and outcomes on the selective somatosensory attention task. The summary depicts the number of patients and healthy volunteers who generated each of the 3 possible outcomes on the somatosensory selective attention task. EMCS = emergent from a minimally conscious state; fMRI = functional magnetic resonance imaging; LIS = locked-in syndrome; MCS = minimally conscious state; VS = vegetative state. [Color figure can be viewed in the online issue, which is available at www.annalsofneurology.org.]

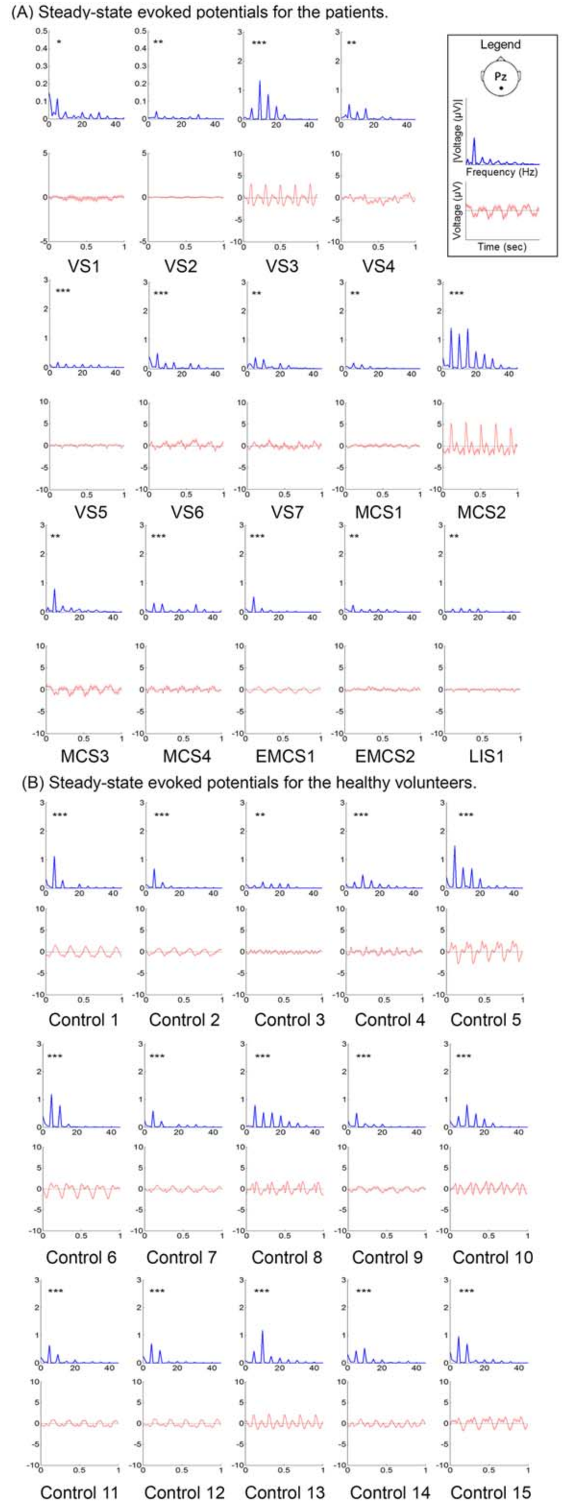

FIGURE 2: Steady-state evoked responses to the repetitive vibrotactile stimulation. (A) Single-sided amplitude spectra and (B) averaged electroencephalographic responses calculated over a period of 1 second. Analyses were conducted using the data recorded from site Pz only; each waveform (B) is depicted with ± 1 standard error of the mean. *$p < 0.05$, **$p < 0.01$; ***$p < 0.001$. EMCS = emergent from a minimally conscious state; LIS = locked-in syndrome; MCS = minimally conscious state; VS = vegetative state. [Color figure can be viewed in the online issue, which is available at www.annalsofneurology.org.]

following in either a behavioral or a neuroimaging-based assessment (see Fig 1 and Supplementary Table 3).

Top-down ERP attention effects (target vs nontarget wrist vibrations) were not detected in any of the patients. However, this ERP effect was evident for healthy volunteers at the group level (n = 15) and at the single-subject level, albeit with a hit rate of 67% (Fig 4). Hit rates of at least 80% (12 of 15) and 100% (15 of 15) have been reported for fMRI-detected mental imagery and selective attention, respectively.[30] Given the relatively lower sensitivity of the top-down attention ERP analysis (ie, 67%), additional post hoc comparisons were conducted. Although the number of trials available after artifact rejection did not differ across groups (see Table 1; $\chi^2[2] = 0.21$, $p = 0.9$), some patients had many fewer trials available than healthy individuals. The single-subject ERP analyses for the healthy volunteers were thus repeated in the post hoc analyses using only a pseudorandom subset of trials equal in number to the minimum number of trials available in the single-subject analyses of the patient data (180 trials, in the case of Patient MCS2).

Bottom-up attentional ERP effects were detected at the single-subject level for all healthy volunteers when as few as 180 trials were included for each stimulus type. However, top-down attentional ERP effects were detected from only 7 healthy volunteers. Subsequent analyses revealed that a minimum of 300 trials were required to detect the top-down attentional ERP effects from the same 10 healthy volunteers as in the a priori analyses. Four patients did not have enough trials available to

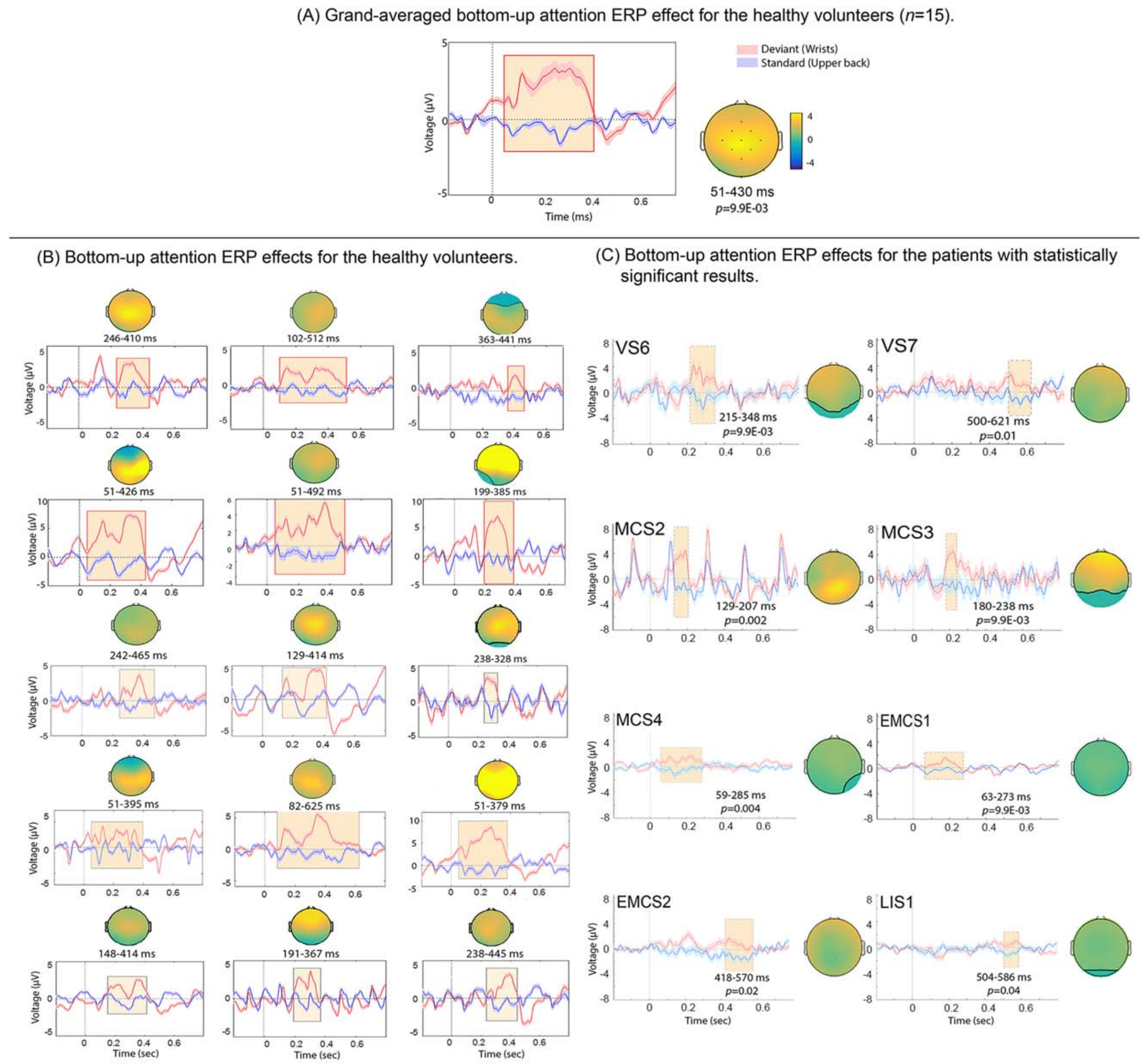

FIGURE 3: Bottom-up attention event-related potentials (ERPs) to the standard and deviant vibrotactile stimulation. Spatiotemporal clusters were calculated across all 12 electrodes and are depicted with ±1 standard error of the mean in matched shading. The electrodes included in the significant spatiotemporal cluster are enclosed with a black line on each topographic plot. The temporal boundaries and the probability value of each cluster are indicated with shading and inset text. (A) Grand-averaged ERP effect for the healthy volunteers. (B) Single-subject ERP effects for the healthy volunteers ($p = 9.9E\text{-}03$ in all cases). (C) Single-subject ERP effects for the patients with statistically significant results. EMCS = emergent from a minimally conscious state; LIS = locked-in syndrome; MCS = minimally conscious state; VS = vegetative state. [Color figure can be viewed in the online issue, which is available at www.annalsofneurology.org.]

meet this criterion. Overall, these analyses indicate that the top-down attentional ERP effect may not have been detected in some single-subject analyses due to low trial numbers. Nevertheless, the bottom-up attentional ERP effect was robust to data loss.

Blood Oxygen Level–Dependent Mental Imagery Responses

In her first visit, Patient VS6 produced reliable, appropriate activation during the motor imagery task in the supplementary motor area and cerebellum bilaterally at an uncorrected $p < 0.001$ (cluster level FWE-corrected $p < 0.05$). In her second visit, Patient VS6 produced reliable, isolated clusters of activation during the motor imagery and spatial navigation tasks in the left precentral gyrus at an uncorrected $p < 0.001$ (cluster level FWE-corrected $p < 0.05$). The patient was thus reclassified as in a nonbehavioral MCS.[18]

Patients VS7 showed high levels of motion requiring 37% and 37.5% of his data to be discarded (for

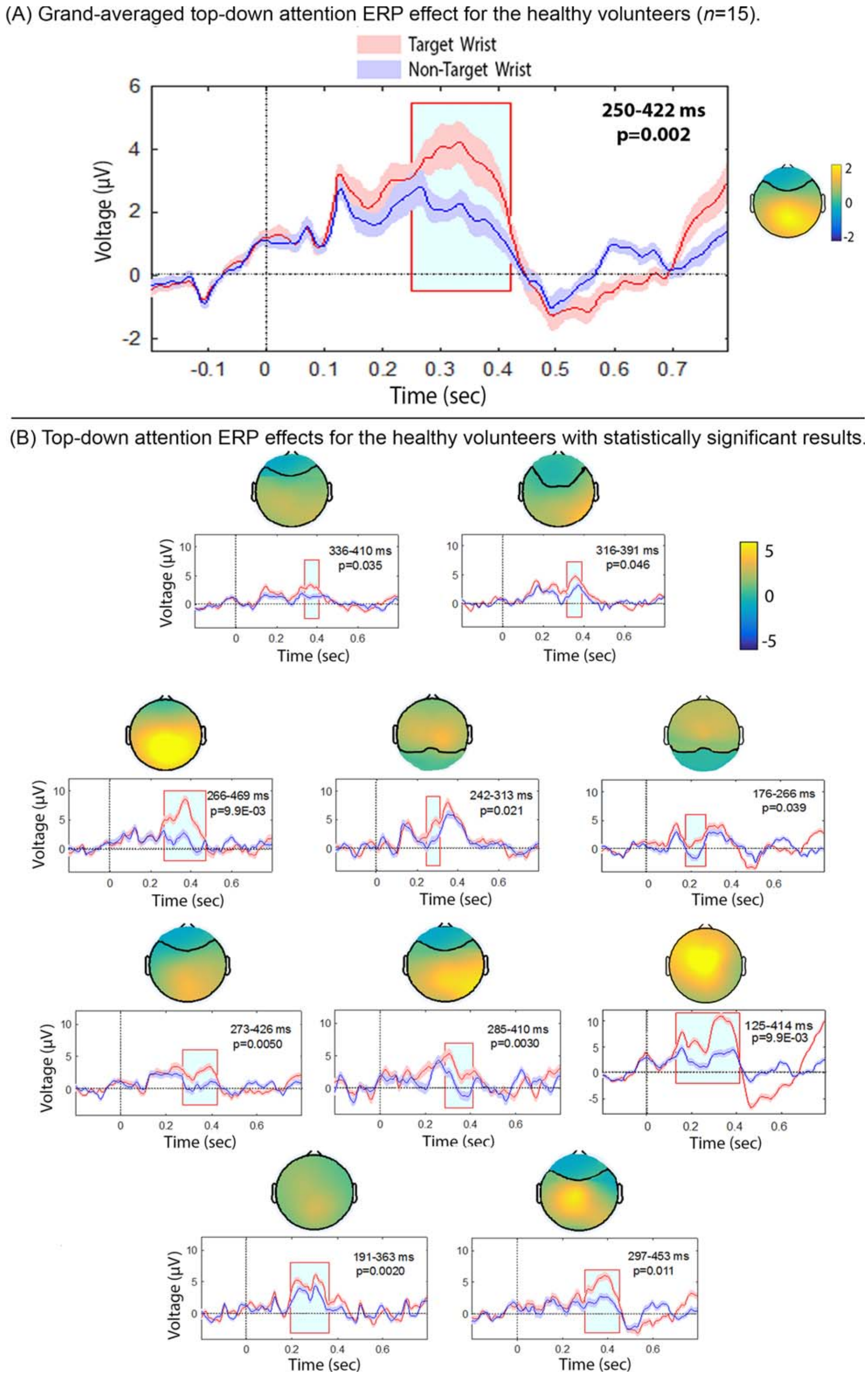

FIGURE 4: Top-down attention event-related potentials (ERPs) to the target and nontarget vibrotactile stimulation for the healthy volunteers. Spatiotemporal clusters were calculated across all 12 electrodes with each waveform depicted with ±1 standard error of the mean. The electrodes included in the significant spatiotemporal cluster are enclosed with a black outline on each topographic plot. The temporal boundaries and the probability value of each cluster are indicated with shading and inset text. (A) The grand-averaged result (n = 15). (B) For the single-subject results, only results from participants with statistically significant clusters are shown. [Color figure can be viewed in the online issue, which is available at www.annalsofneurology.org.]

motor imagery and spatial navigation, respectively). The analysis of the remaining data revealed appropriate activation during the spatial navigation task only (ie, the left occipitoparietal junction at uncorrected $p < 0.001$.). The patient was thus reclassified as in a nonbehavioral MCS.[18]

Patients MCS3, MCS4, EMCS1, and LIS1 showed reliable activation during the spatial navigation task only. This involved: bilateral occipitoparietal junction (uncorrected $p < 0.001$) for MCS3; right temporo-occipitoparietal junction (FWE-corrected $p < 0.05$), as well as right dorsal premotor cortex, right insular cortex, and right putamen (uncorrected $p < 0.001$) for MCS4; right occipitoparietal junction, a region in the boundaries between right lingual gyrus and parahippocampal cortex, and left precentral gyrus (comprising the supplementary and presupplementary motor areas), as well as some less typical areas such as the inferior frontal gyrus, the left superior temporal gyrus, and the left striatum (FWE-corrected $p < 0.05$) for EMCS1; and supplementary motor area, right precentral gyrus, occipitoparietal junction, posterior temporo-occipital region, and the cerebellum (uncorrected $p < 0.001$) for LIS1.

The remaining 7 patients (VS1–5, MCS1, and MCS2) showed no activation at the conservative FWE-corrected statistical threshold, or at uncorrected $p < 0.001$.

Blood Oxygen Level–Dependent Auditory Selective Attention Responses

Of the patients diagnosed as in a VS, only Patient VS6 showed significantly more activation following the instruction to count than to relax. This patient showed significant activation in the temporal and parietal cortex bilaterally (FWE-corrected at $p < 0.05$).

Patients MCS1–4 and LIS1 also showed significantly more activation following the instruction to count than to relax. Patient MCS1 showed significant activation in the frontotemporal and parietal cortex bilaterally. Patient MCS2 showed significant activation in the temporal cortex bilaterally (FWE-corrected at $p < 0.05$). Patient MCS3 showed significant activation in the parietal cortex bilaterally. Patient MCS4 showed significant activation in the frontotemporal and parietal cortex bilaterally (FWE-corrected at $p < 0.05$). Patient LIS1 produced significant brain activity in the frontotemporal cortex bilaterally (FWE-corrected at $p < 0.05$).

Of note, Patient EMCS1 did not show significant differences in activation in the command-following task, although she was able to follow commands with her overt behavior immediately prior to her assessment. Patients VS7 and EMCS2 were excluded from this analysis, because both patients moved excessively during their functional scans.

Correspondence between Command Following and EEG Responses

The main hypothesis in this investigation was that patients who were aware would exhibit concordant EEG markers of higher-order attention processing. Although top-down processing (P3b) was not detected in any patients, an interesting observation from the current data is the relationship between a specific marker of awareness—command following—and the bottom-up attention-orienting ERP effect, the P3a. A patient was considered to have evidence of such awareness if they demonstrated evidence of command following in any 1 of the 3 non-EEG assessments (selective auditory attention, mental imagery, or a behavioral assessment with the CRS-R). This approach is consistent with clinical behavioral guidelines in which a diagnosis of awareness (MCS) is given if a patient follows commands on 1 occasion across multiple assessments. A Fisher exact test revealed a significant positive association between evidence for command following and evidence for the P3a ($p = 0.007$; note $p = 0.0047$ if the 2 observations of Patient VS6 are not included to maintain the assumption of independence). This relationship is summarized in Figure 1.

Replication Data

The replication results are depicted in Figure 5. All patients exhibited consistent effects across assessments with the exception of Patient VS6, for whom a P3a was significant only during her initial assessment.

Discussion

We investigated a novel EEG method for the assessment of residual sensory and cognitive processing alongside 2 fMRI-based assessments of covert command following and 1 behavioral assessment of overt command following in a sample of 14 patients with severe brain injuries. The primary novel finding of this work is the relationship between an ERP marker of bottom-up attention orienting (the P3a) and command following such that all patients with a P3a response demonstrated positive evidence of command following. Similarly, most patients who did not generate a P3a response also did not demonstrate evidence of command following (see Fig 1 and Supplementary Table 3).

Some investigators have reported positive prognostic value in the presence of a P300 following traumatic brain injury.[25] There have also been reports of correlations between cognitive ERPs and behavioral markers of awareness,[14,24] as well as the prediction of recovery from DoC using cognitive ERPs.[26,47] Crucially, the current study included 2 neuroimaging-based assessments of covert command following. This step is important given that a recent meta-analysis estimates a 15% rate of covert awareness among patients diagnosed as in a VS.[27] Previous studies of the P300 in patients with DoC are likely to have included patients capable of covert command following, thus obscuring the relationship reported here.

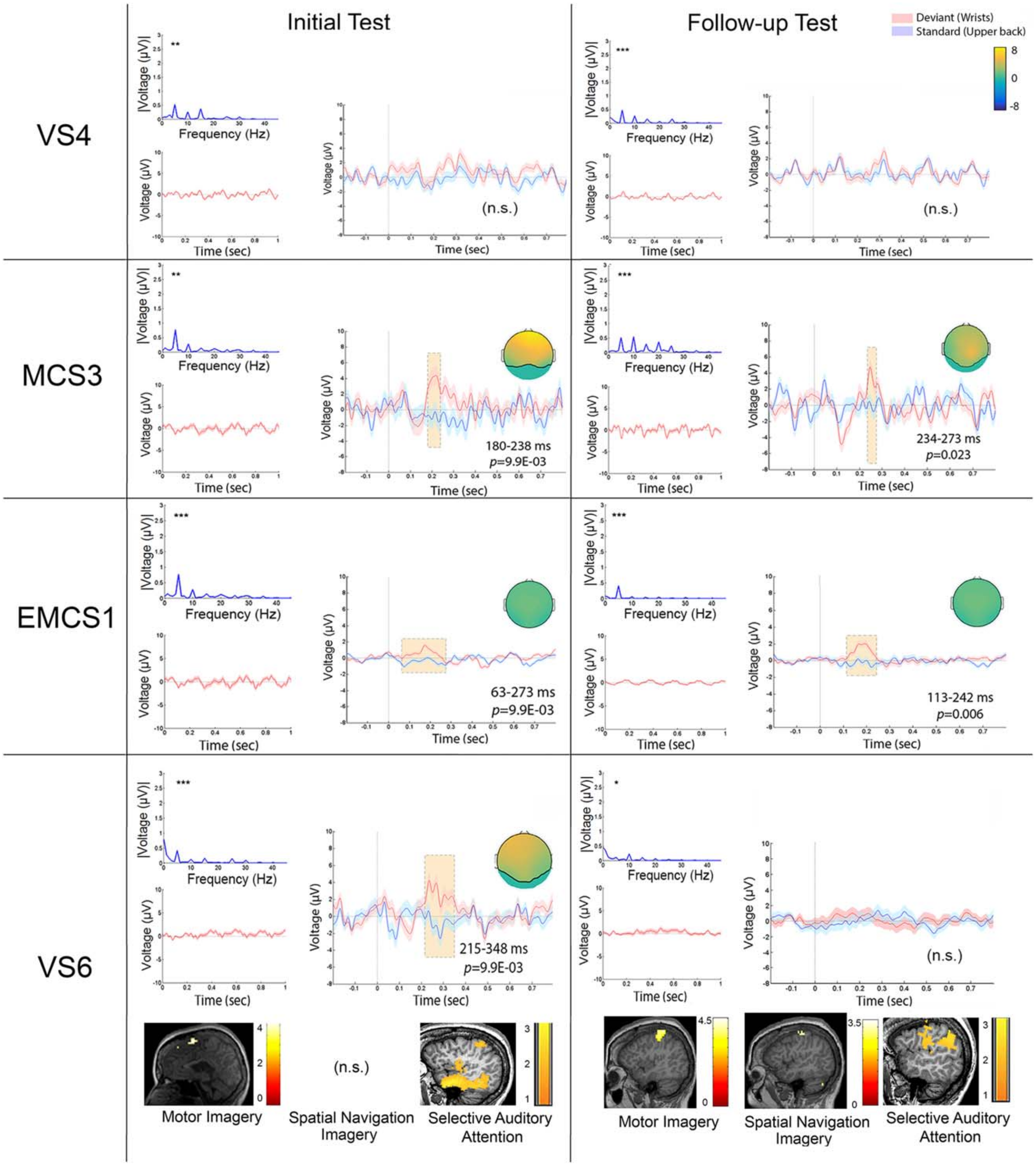

FIGURE 5: Replication data from the four patients with whom follow-up investigations were conducted. Data are depicted for the initial and follow up tests of Patients VS4, MCS3, EMCS1, and VS6, as labelled. For the steady-state evoked potentials, single-sided amplitude spectra (top left panels within each cell) and averaged EEG data (bottom left panels within each cell) were calculated over a period of 1-second. Analyses were conducted using the data recorded from site Pz only; each waveform (bottom left panels within each cell) is depicted with ±1 standard error of the mean. For the bottom-up attention ERP effects (right panels within each cell), spatiotemporal clusters were calculated across all twelve electrodes and are depicted with ±1 standard error of the mean. The electrodes included in the significant spatiotemporal cluster are enclosed with a black line on each topographic plot. The temporal boundaries and the probability value of each cluster are indicated with shading and inset text. For Patient VS6 only, two separate fMRI assessments were conducted at each testing session. For the fMRI mental imagery paradigm, significant task-related fMRI activation is depicted (Imagery>Rest), and results are thresholded at an uncorrected $p<0.001$. For the fMRI selective auditory attention task, only activation clusters within the attention network (Count>Relax) that survived the familywise error correction threshold of $p<0.05$ at the whole-brain level are displayed. The fMRI results are rendered on the patient's T1 anatomical MRI image, and scales depicting the *t*-value statistical maps are inset. *$p<0.05$; **$p<0.01$; ***$p<0.001$. EMCS = emergent from a minimally conscious state; MCS = minimally conscious state; VS = vegetative state; n.s. = not statistically significant. [Color figure can be viewed in the online issue, which is available at www.annalsofneurology.org.]

Although the feasibility of routine neuroimaging assessments in clinical practice is limited by important health, safety, and financial factors, the findings of this work suggest that these assessments are necessary to elucidate the relationship between a patient's conscious state and their residual sensory and cognitive abilities.

It is curious that an ERP marker of unconscious (or preconscious) processing (ie, the P3a) is closely linked to awareness in this work. The P3a can be elicited by unattended stimuli and during rapid eye movement sleep and deep sedation.[28,48] We speculate that the correspondence between the P3a and command following stems from the overlap of the neural networks that support attention, and those that are relatively more preserved in conscious patients.[49,50] Frontal lobe lesions have been associated with diminished P3a responses to auditory[51] and somatosensory[52] stimulation. Equally, this association suggests that a P3a response may be less informative for patients with specific frontal lobe injuries. Nevertheless, a P3a can be elicited without the explicit collaboration of the individual—that is, without following task instructions.[48] This feature is appealing, as it suggests that a passive assessment of attention orienting, which entails lower cognitive demands than active assessments of voluntary top-down attention, may be sufficient to identify patients with covert awareness.

The P3b marker of top-down attention in the current EEG task was not detected in any of the patients in this sample, as has been reported previously.[53] P3b responses in the current work were detected in only 67% (10 of 15) of the healthy volunteers. Post hoc analyses of the ERP data indicated that this low sensitivity may be exacerbated by the fewer usable trials in the patient data, as this comparison was sensitive to a reduced signal-to-noise ratio. Additionally, time-variant levels of arousal and fatigue characteristic of the DoC may have led to inconsistent engagement in the counting task needed to generate the top-down ERP effect.[8,19] In contrast to the fMRI-based selective attention task, the selective attention manipulation in the EEG task may have placed higher cognitive demands on participants due to the longer duration of the EEG task. Participants were required to sustain attention for 5 minutes in ~22.5-second blocks for both fMRI tasks, whereas the EEG task involved 15 minutes of attention in ~1-minute blocks. The EEG task was longer to ensure that a high EEG signal-to-noise ratio was achieved, and post hoc analyses confirmed that the top-down ERP effect was sensitive to trial numbers. Unfortunately, increased task duration requires participants to sustain attention for an even longer period, making it unlikely that this manipulation would increase the sensitivity of the task. Some investigators use machine learning to circumvent these issues and address possible spatiotemporal variations in the electrocortical responses of patients with brain injuries.[54] For simplicity of interpretation and consistency with clinical methods, we employed a more traditional approach to comparing scalp voltages. Although no false alarms were evident in the current sample, misses occurred with 2 patients; that is, patients demonstrated evidence of command following but no evidence of a P3a. As has been discussed elsewhere, signs of awareness in both behavioral and neuroimaging assessments may be missed due to fluctuating arousal.[13] Nevertheless, when a P3a is elicited, the current data suggest the sophisticated cognitive networks that underlie an ability to follow commands are also preserved.

The detection of awareness in DoC is a clinical standard of care. To provide sufficient evidence to influence clinical practice, it is essential to compare novel assessments to existing techniques. The current investigation allowed for a comparison of 2 previously reported neuroimaging-based assessments of covert command following, based on mental imagery[6,9,10,13,15–17] and selective auditory attention.[29,30] The results of these assessments converged for 9 of the 12 patients with useable data from both paradigms. Two patients demonstrated positive evidence of command following in only the selective auditory attention task, whereas one patient showed positive evidence of command following only in the mental imagery task. The behavioral profile of DoC—that is, time-variant fatigue and arousal—always affords the possibility that a patient did not demonstrate positive evidence of covert command following due to lack of voluntary engagement in the task. Likewise, false negatives occur in assessments of healthy volunteers.[11,55] Nevertheless, the less than perfect correspondence of the 2 covert fMRI command-following tasks may have occurred because the demands of one task were better suited to the patient. For example, some individuals find it difficult to engage in motor imagery,[56] and in some reports, brain–computer interfaces based on selective attention tasks are successfully operated by more users than those based on responses to motor imagery.[57,58] Accordingly, assessments of covert command following based on selective attention may be better suited to a general population. Overall, however, an optimal evaluation of a patient with one of the DoC should include multiple assessments to maximize the likelihood of detecting responses that are not evident from overt behavior.[13] In the absence of unambiguous ground truth, an investigation of the concordance between assessments may be the best way to improve diagnostic and prognostic accuracy.

In summary, the brain responses of 14 patients with severe brain injuries were assessed using an EEG-based somatosensory selective attention task, 2 fMRI-based assessments of covert command following, and 1 behavioral instrument. Although limited by a relatively small sample of patients, the data tentatively suggest that the detection of a somatosensory bottom-up P3a effect in a patient correlates with an ability to follow commands, as evaluated by multimodal assessments. This provides evidence that a bedside somatosensory oddball procedure can improve diagnostic accuracy in DoC and more accurately characterize the level of neurocognitive preservation. Overall, this work provides a valuable addition to neuroimaging batteries for the clinical assessment of patients with DoC and convergent, multimodal evidence for the utility of these techniques.

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Author Contributions

R.M.G., D.F.-E., L.N., and D.C. contributed to conception and design of the study, and to data collection and analysis. R.M.G., D.C., S.C., and A.M.O contributed to writing the manuscript.

Potential Conflicts of Interest

Nothing to report.

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