EEG-based effective connectivity distinguishes between unresponsive states with and without report of conscious experience and correlates with brain complexity

BE Juel[a], TR Bremnes[a,b], O Gosseries[e], M Rosanova[d], M Boly[e], S Laureys[e], PG Larsson[c], M Massimini[d] , JF Storm[a]*

[a]*Department of Molecular Medicine, University of Oslo, Norway;*
[b]*Department of Clinical Medicine, University of Tromsø, Norway;*
[c]*Department of Neurosurgery, Oslo University Hospital, Norway;*
[d]*Department of Biomedical and Clinical Sciences, University of Milan, Italy;*
[e]*Coma Science Group, Cyclotron Research Centre and Neurology Department, University and University Hospital of Liège, 4000 Liège, Belgium.*

***Corresponding Author**: Johan Frederik Storm (e-mail: j.f.storm@medisin.uio.no), Department of Molecular Medicine, University of Oslo, Domus Medica, Sognsvannsveien 9, 0372 Oslo, Norway

ABSTRACT

Objective methods for distinguishing conscious from unconscious states in humans are of key importance for clinical evaluation of general anesthesia and patients with disorders or consciousness, as well as for consciousness research. The Directed Transfer Function (DTF) – a measure of effective connectivity - was recently shown to accurately classify patients undergoing anesthesia as conscious or unconscious based on 1-second segments of raw EEG. Here, we test the generalizability of the DTF-based classification algorithm as an objective measure of conscious experience during anesthesia and correlate it with a well-tested index of consciousness: the Perturbational Complexity Index (PCI). We reanalyzed EEG data from an experimental study in which 18 healthy volunteers were randomly assigned to one of three types of general anesthesia: propofol, xenon, and ketamine. EEG was recorded before and during anesthesia, and DTF was calculated from every 1-second segment of the EEG data to quantify the effective connectivity between channel pairs. This was used to classify the state of each participant as either conscious or unconscious, and the classifications were compared with the participant's delayed report of experience. Finally, classifications were correlated with the PCI values obtained in the original study. The DTF analysis yielded two distinct patterns of DTF-based connectivity that corresponded well to the participants' reports about subjective experience, and was sufficient to accurately classify the state of the participants as conscious or unconscious. The algorithm was more likely to classify participants as conscious in the awake state than during propofol and xenon anesthesia ($p<0.05$), but not during ketamine anesthesia ($p>0.05$). Furthermore, the DTF-based confidence of being classified as conscious was highly correlated with PCI ($r^2=0.48$, $p<0.05$). The DTF-based measure distinguished between conscious and unconscious states during different forms of general anesthesia and wakefulness, and it correlates strongly with PCI. These results provide further support for the notion that effective connectivity measured between EEG electrodes can be used to distinguish between conscious and unconscious states in humans.

Key words *Consciousness • Anesthesia • EEG • Directed Transfer Function*

INTRODUCTION

Objective methods for distinguishing conscious from unconscious states in humans are of key importance for consciousness research as well as for clinical evaluation of general anesthesia and patients with disorders or consciousness (1–4). For example, estimates indicate that 26,000 patients undergoing general anesthesia experience unintended and undetected awakenings during surgery, per year, in the US alone (5). To avoid this, measures that can aid clinicians in determining the state and level of consciousness in the most difficult situations are needed. In the last few decades, the search for such measures has intensified and researchers have uncovered a variety of candidate measures that are, to various degrees, capable of objectively distinguishing between reportedly conscious and unconscious states (3,6–13).

In particular, the Perturbational Complexity Index (PCI), a measure inspired by the Integrated information theory (IIT) of consciousness (14,15), has shown highly promising performance (3). The PCI has repeatedly been shown to accurately distinguish conscious from unconscious states in healthy volunteers under controlled conditions such as sleep and anesthesia, and was recently shown to be remarkably sensitive in classification when applied to patients with disorders of consciousness (16). In addition to the PCI's strengths in classification, the method is particularly interesting because it probes the brain's causal dynamics directly without relying on the integrity of sensory or motor pathways, subjective reports, or participation of the test subject (17). Even though the PCI seems very reliable as an index of conscious state in humans, it is unfortunately suboptimal as a tool in certain clinical situations as it requires considerable expertise, trained personnel, and expensive equipment. Furthermore, the data acquisition and analysis are relatively time consuming, requiring several minutes of recording and hours of analysis for a single data point, thus preventing real-time monitoring with high time resolution.

Recently, we proposed a different objective measure that can be calculated automatically from 1-second segments of raw, spontaneous EEG data, which may overcome some of these limitations (8). Our measure is based on the Directed Transfer Function (DTF; (18)), a measure of effective connectivity in the Granger causality family, which can be applied to EEG data and is robust to noise and artefacts (19–21). Therefore, it is potentially well suited to form a basis for an automatic algorithm for classifying the conscious state of individuals in near real time directly from raw clinical EEG data. However, the DTF has only been tested in this context for a dataset comprising 8 patients undergoing anesthesia so far (8). Previously, other studies have shown that EEG-based effective connectivity, as quantified by the DTF, is different between sleep and wakefulness (22–24), and between healthy individuals and patients suffering from disorders of consciousness (25). Together, these findings indicate that a DTF-based measure might be useful as a general marker of consciousness.

In a recent study, Sarasso et al. (26) compared PCI in three different types of general anesthesia, propofol, xenon, and ketamine, at doses that rendered the participants behaviorally unresponsive. They found high PCI values for participants undergoing ketamine anesthesia (indicating consciousness), whereas the PCI values during xenon and propofol anesthesia were low (indicating unconsciousness). Importantly, this reflected the reports of the participants in the study; all participants undergoing ketamine anesthesia reported vivid dreams during the anesthesia, while the participants undergoing xenon and propofol anesthesia did not. These results provide evidence for PCI being able to detect conscious states, even in behaviorally unresponsive subjects. Also in this study we define consciousness as having any subjective experience. Thus, if an individual reports having some subjective experience, including dreaming, she/he is considered conscious.

In this study, we reanalyzed data reported on in Sarasso et al. (26), by applying the DTF-based classification algorithm to the spontaneous EEG data. This was done in order to test the DTF-based algorithm as an objective measure for separating states of consciousness during anesthesia, and to correlate it with a well-tested index of consciousness: the PCI. We hypothesized that our DTF-based measure would separate conscious from unconscious states in accordance with the participant´s own report of (un)conscious state, and the PCI measure.

METHODS

PARTICIPANTS

The data analyzed were obtained in a previous study conducted at the Centre Hospitalier Universitaire (CHU) in Liege, Belgium (26). Eighteen healthy volunteers (10 females, 8 males, age 18-28 years) were recruited and participated in the study. Before the experiment, all volunteers gave their written informed consent for participation, and physical examinations were performed to exclude potential participants with medical conditions that were incompatible with the anesthesia or the experimental procedure. If no exclusion criteria were met, participants were randomly assigned to one of the three experimental protocols (propofol, xenon, or ketamine anesthesia; n=6 for each). The experimental protocol was approved by the local ethical committee of the University of Liège (Liège, Belgium).

PROCEDURE

As described in the original publication (26), 60 channel EEG was recorded from all participants before, during, and after the administration of the anesthetic, using a TMS-compatible 60-channel EEG amplifier (Nexstim Plc., Finland). The impedance at all electrodes was kept below 5 kΩ. EEG signals were referenced to an additional electrode on the forehead, filtered upon acquisition (0.1-350 Hz), and sampled at 1450 Hz. Two additional sensors were used to record the electrooculogram (EOG) activity. In both the awake and unresponsive conditions, EEG was recorded with and without concurrent single pulse transcranial magnetic stimulation (TMS). During the responsive wakefulness condition, a 10-minute spontaneous EEG recording was performed before TMS-EEG was acquired, while during the drug-induced unresponsiveness condition spontaneous EEG was continuously acquired starting ~3 min before and ending ~3 min after TMS-EEG recordings.

All anesthesia protocols were conducted by a certified senior anesthesiologist. Throughout the procedure, the participant's electrocardiogram (ECG), non-invasive blood pressure, SaO_2, exhaled CO_2, and axillary skin temperature were continuously monitored, and did not show significantly abnormal values during any of the experiments. Only one type of anesthetic was administered to a given participant, but before the beginning of all experimental procedures, participants were given metoclopramide (2 mg) to minimize possible complications caused by the anesthetic drug, such as nausea and vomiting.

The procedures for the three anesthetics aimed at reaching a common behavioral state, i.e. unresponsiveness, systematically assessed by means of repeated Ramsey Scale administrations (27). Thus, a Ramsay Scale score of 6 (deep unresponsiveness, corresponding to no response to external stimuli) was obtained for all the subjects in the three protocols (26). Following drug administration, repeated assessments of responsiveness were performed at 30 second intervals until three consecutive assessments obtained a Ramsay scale score of 6.

To assess the presence/absence of conscious experience during anesthesia-induced behavioral unresponsiveness, retrospective reports were collected for all participants after awakening. For this purpose, after participants recovered responsiveness (three consecutive Ramsey Scale scores of 2), they were asked to report their previous conscious experience during the period of anesthesia ("what was going on through your mind before awakening?"). Participants were asked to confirm their retrospective reports one hour after recovering responsiveness. Experience was defined as "any kind of mental activity," which included thoughts, dreams, images, emotions, etc. Responses were recorded and lumped into two categories: 1) no conscious experience/no recall; 2) conscious experiences, when the participant could describe the content of the experiences.

Data Processing and Analysis

Here, we focused our attention on the spontaneous EEG data recorded before and after the anesthetic administration. Each data-file was visually inspected using Besa Research software (BESA GmbH, 82166 Graefelfing, Germany). Specifically, individual EEG-channels with activity patterns deviating from the normal (high amplitude or variance noise over extended periods, or flat channels) were marked for removal.

The EEG files were loaded into Matlab R2015a using the EEGlab analysis toolbox (28). They were downsampled to 512 Hz, and recordings from bad channels were removed and replaced by standardized Gaussian noise. After this, the EEG file was divided into 1-second epochs from which the DTF was calculated using the 'DTF' function in the open source toolbox eConnectome (29). Care was taken to stay as close to the original analysis pipeline as possible (8). The only deviations from the original, beyond hardware differences, were that the current data used a slightly different set of 25 EEG channels (Fp1, F3, F7, F9, T7, T9, Fp2, F4, F8, F10, T8, T10, Fz, C3, Cz, C4, P3, Pz, P4, P7, P8, P9, P10, O1, O2; down-sampled from the 60 channels in the original recordings), and that we now focused on the theta frequency range (3-7 Hz) rather than the alpha range (8-12 Hz) to reduce confounds caused by eyes being open/closed.

The DTF was calculated for all channel pairs for every 1-second segment of EEG, yielding a channel-by-channel-by-frequency matrix of values quantifying the directed information flow between all channels for each frequency of interest. By taking the median across the frequencies we obtained a 25-by-25 matrix of the DTF values, quantifying the directed information flow for all channel pairs in the theta range. Also, since the DTF values are normalized to reflect the influence of one channel on another relative to the total influence all channels have on that channel, it makes sense to consider the logarithm of the DTF (LDTF) when investigating the relative differences in strength of information flow. By compressing this matrix further, taking the median across all influences from a given channel, we obtained a vector for each channel indicating the typical influence of that channel on the other channels. Later, we refer to the results of this operation as the outgoing connection strengths from the channels, or the source LDTF (sLDTF).

To quantify the overall pattern of outgoing connection strengths and compare differences within and between groups, we calculated the root mean squared difference (RMSD) between channel sLDTF's across the scalp. This results in a number indicating the degree of heterogeneity in outgoing connection strength across the scalp. We refer to this measure as the sLDTF heterogeneity (sLDTF-het). The RMSD can also be used to quantify the degree of dissimilarity in the outgoing connection strength across the scalp between groups. This gives a measure (abbreviated ΔsLDTF) of the dissimilarity between sLDTF topographies in the different groups.

Classification Algorithm

In a previous study, we implemented a data-driven algorithm to classify the states of patients undergoing anesthesia as awake or anesthetized based on LDTF computed from 25 channel clinical EEG (8). This resulted in a database of LDTF values labeled as awake or anesthetized by a trained clinical anesthesiologist. The database was used to generate descriptive statistics of the distributions of LDTF values in both the awake and anesthetized condition. Here, we used the database from that study as the basis for classifying the states of the subjects in the present study under the different protocols tested, as conscious or unconscious. In other words, we used the distributions of LDTF values from a different set of subjects, undergoing a different experimental protocol, as the basis for classifying the current participants as conscious or unconscious.

For each 25x25 matrix of LDTF-values computed from a 1-second segment of EEG in the current data set, the likelihood of it being drawn from either the conscious, L_C, or unconscious, L_{UC}, distribution was calculated. From this, a rating indicating the confidence of being classified as conscious, *C(t),* was calculated for the current time point, *t*:

$$C(t) = \frac{L_C(t)}{L_C(t) + L_{UC}(t)}$$

For each participant in a given state, the algorithm's average confidence in classification was calculated to give an indication of the overall confidence for classifying the participant as conscious:

$$\bar{C} = \frac{1}{n}\sum_t C(t)$$

This measure of average confidence in classifying the participant as conscious was used as our final metric for this study. A $\bar{C}$ close to 1 indicates that the algorithm had a high confidence in classifying the participant as conscious for all time points. While a low $\bar{C}$ (close to 0) indicates that the algorithm had a low confidence in classifying the participant as conscious for all time points, or similarly that it had a high confidence in classifying the participant as unconscious.

To investigate whether the algorithm was more likely to classify participants as conscious in the awake than the unresponsive state, we ran a paired t-tests comparing $\bar{C}$ between states. Next, we compared the algorithm's classification with the participant's own report of subjective experience, and performed a ROC analysis to find the optimal threshold confidence for classification as conscious for this dataset. To find this optimum, we set as our criteria that there were to be no false negatives (participants with subjective report classified as unconscious), while minimizing the false positive rate. Finally, we correlated the overall confidence, $\bar{C}$, with the PCI value computed from the same state and subject, as reported by Sarasso et al. (26).

RESULTS

The data from one participant in each condition had to be excluded due to missing data, leaving us with n=5 participants from each condition for the analysis (total: n=15). For each participant, an EEG segment was analyzed from the awake condition (median length [min:sec]: 5:07; range [min:sec]: 1:32-21:32), and the anesthetized condition (median length [min:sec]: 5:24; range [min:sec]: 2:07-17:04). The processing steps outlined above yielded a mapping of the outgoing connection strength for every second of EEG. Figure 1 shows the median outgoing connectivity strengths, sLDTF, across subjects for all four conditions.

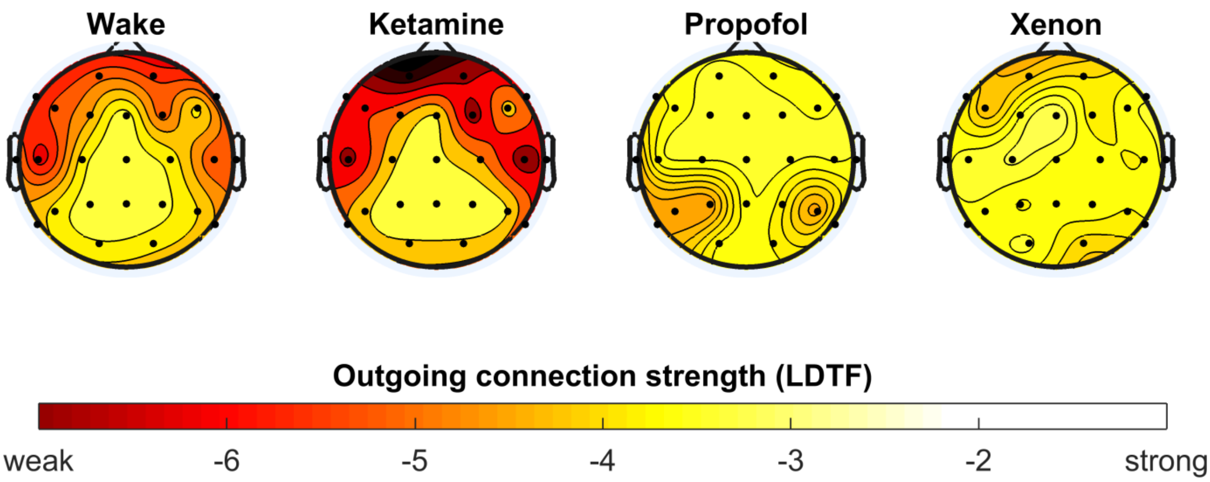

Figure 1: Patterns of outgoing connection strengths between awake state and different forms of general anesthesia; ketamine, propofol, and xenon. *For each state, a topographical visualization of the sLDTF-based median outgoing connection strength, across all subjects within the given state, is shown. Strength of outgoing connectivity is indicated by the color bar (from weak (-6) to strong (-1) connection strength). Head direction is indicated by the black overlay (nose pointing up).*

Qualitatively, the topographic sLDTF maps for the wake and ketamine groups seemed to resemble each other. Both showed a heterogeneous pattern with regions of relatively stronger sources in the medial posterior (parietal and occipital) areas and weaker sources in the front and in the periphery (Figure 1, left). In contrast, the corresponding maps for the propofol and xenon groups both showed a more homogenous pattern, where most regions had similarly strong outgoing connection strengths (Figure 1, right). The values of sLDTF appeared to be less variable across the scalp in propofol and xenon anesthesia, than in wakefulness and ketamine anesthesia. The measure of heterogeneity (sLDTF-het) reflected this qualitative difference; wake: 1.01, ketamine: 1.26, propofol: 0.63, xenon: 0.49.

The dissimilarities between the topographies, as assessed by ΔsLDTF, were lower between propofol and xenon anesthesia (0.469) and between wake and ketamine (0.799), than they were between any other combination of conditions (1.038 - 1.789) (Table 1). In other words, the topographical maps of sLDTF in wakefulness and ketamine anesthesia were relatively similar. As were the outgoing connectivity maps in propofol and xenon anesthesia. Taken together, the measures of heterogeneity indicate that the topographical maps of sLDTF values could be used to distinguish between participants with and without reports of experience (awake and ketamine anesthesia vs. propofol and xenon anesthesia).

ΔsLDTF	Wake	Ketamine	Propofol	Xenon
Wake	0			
Ketamine	0.799	0		
Propofol	1.178	1.789	0	
Xenon	1.038	1.615	0.469	0

Table 1: *Values indicating the root mean square difference of outgoing connectivity strength (ΔsLDTF) between DTF topography maps in different conditions. Condition pairs with similar DTF topography maps (ΔsLDTF < 1) are marked by green color, while condition pairs with different DTF topography maps (ΔsLDTF > 1) are marked by red color.*

This grouping of states – with wakefulness and ketamine anesthesia in one group, and propofol and xenon anesthesia in another – seemed to still hold at the individual level. The pattern of weaker outgoing connectivity from frontal peripheral channels in wakefulness and ketamine anesthesia was clear in the topographical visualizations (Figure 2, right). Furthermore, the patterns seemed to be stable over time, as can be seen from the time course plots (Figure 2, left).

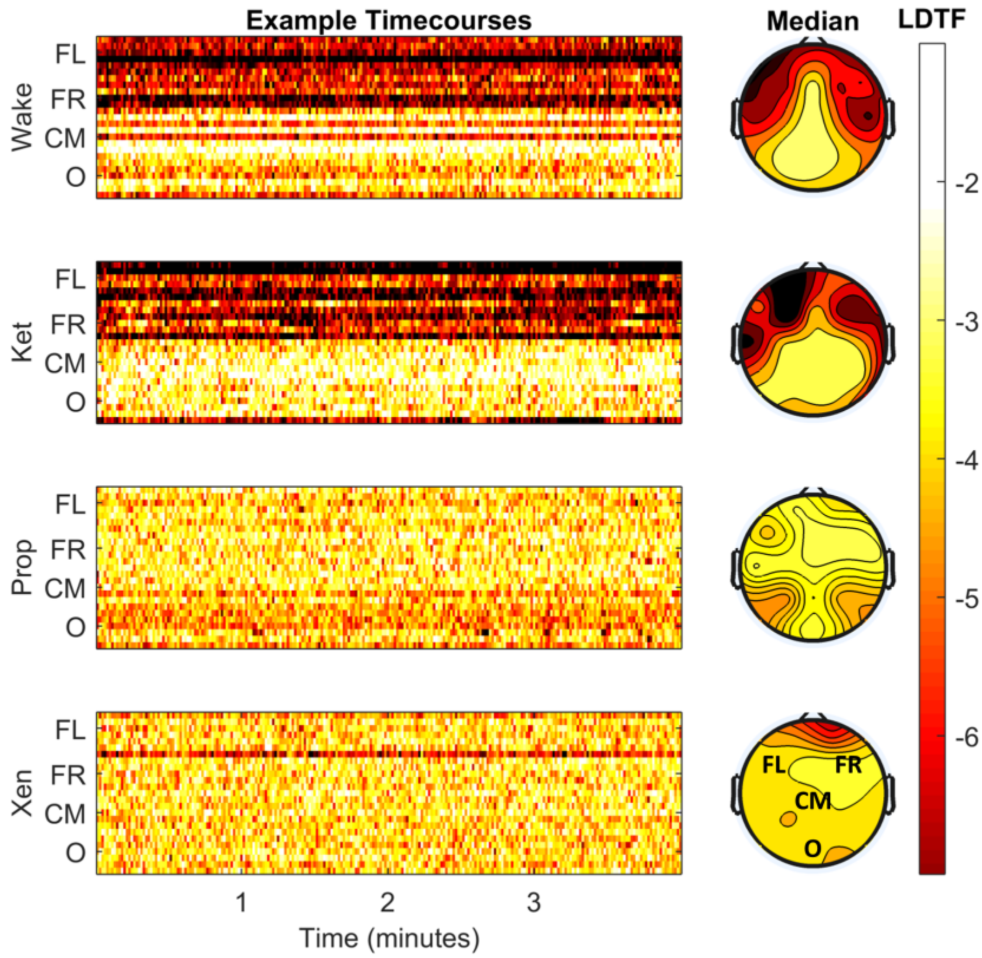

Figure 2: Time-courses for example data from one individual in each condition, with corresponding topographies of their median outgoing connection strength. *Left: timecourses of the outgoing connection strength of the channels for each individual are shown. In the panels on the left, timecourses of the outgoing connection strengths are shown, based on 4 minutes of spontaneous EEG recordings. Every EEG channel has a corresponding row in the timecourse, and the rows are ordered based on the position of the channel on the scalp (front left (FL): Fp1,F3,F7,F9,T7,T9; front right (FR): Fp2,F4,F8,F10,T8,T10; central medial (CM): Fz,C3,Cz,C4,P3,Pz,P4; Occipital (O): P7,P8,P9,O1,O2,P10). Right: the corresponding topography of the median outgoing connection strength over the whole segment for each example participant is shown. Strength of outgoing connectivity is indicated by the color bar (from weak (-6) to strong (-1) connection strength). Head direction is indicated by the black overlay (nose pointing up).*

The results from the DTF-based classification based on the LDTF values are summarized in Figure 3. Specifically, the average confidence of being classified as conscious was significantly smaller for subjects undergoing propofol and xenon anesthesia than for the awake state ($p<0.05$ for both). In addition, there was no significant difference in this classification confidence between awake subjects and subjects undergoing ketamine anesthesia. These trends could be seen both when considering the paired data from all participants (Figure 3, left) and when considering the pooled data for each condition (Figure 3, right). Furthermore, when comparing the variation in the confidence of being classified as conscious with the variation in PCI values reported by Sarasso et al. (26), we found a strong correlation ($r^2 = 0.48$; $p<0.05$, Figure 4).

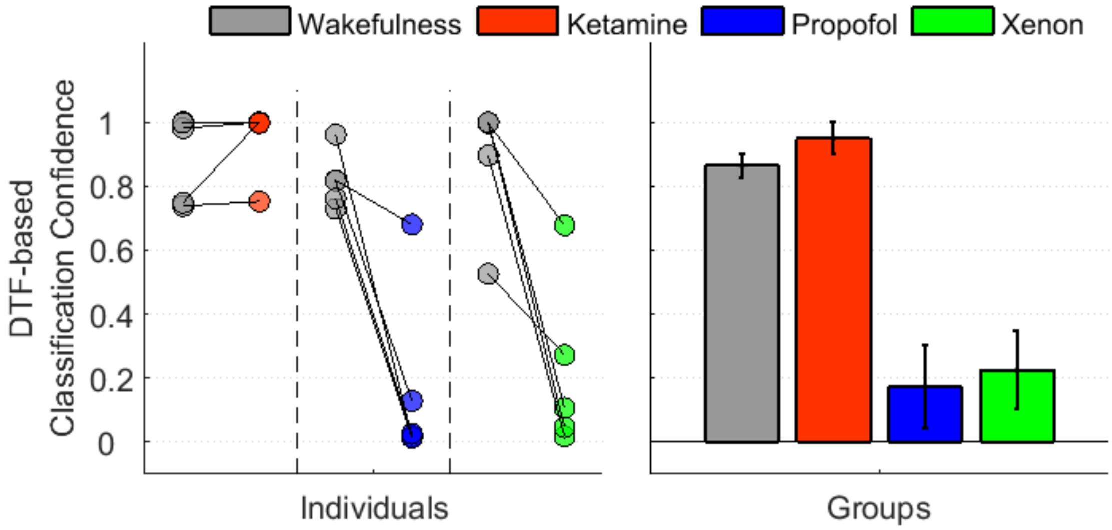

Figure 3: The DTF-based classification confidence for individual participants, and grouped. *The variation in the confidence of being classified as conscious for individual participants (left), and grouped (right).*

Based on a ROC-analysis, we found a range of cutoff values for classifying conscious states vs. unconscious states with 90% accuracy or higher. There was a wide choice of intermediate cutoff values (between 0.25 and 0.70) yielding high accuracy, sensitivity and specificity. Specifically, cutoff values between 0.25 and 0.45 yielded the maximal accuracy obtained with this dataset (93%) while maintaining a perfect sensitivity (no conscious individuals classified as unconscious). Without a larger study population we cannot currently draw any stronger conclusion on the optimal cutoff value.

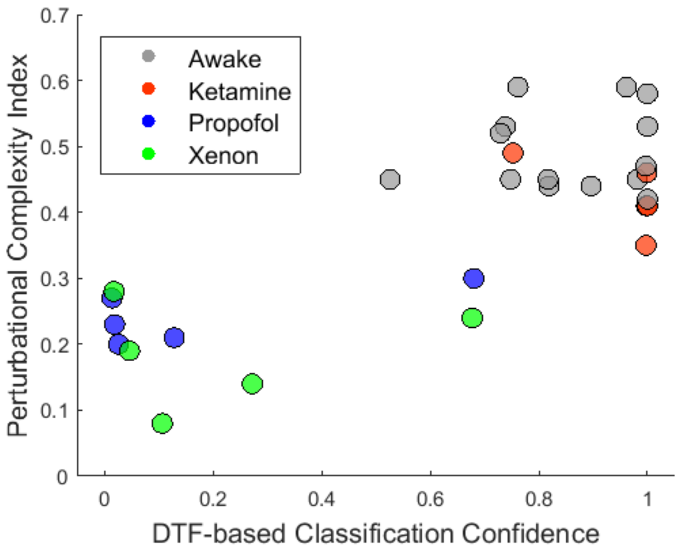

Figure 4: Comparison of the Perturbational Complexity Index values and the DTF-based classification confidence. *A scatter plot shows the relationship between the DTF-based confidence of being classified as conscious and the PCI values obtained for the same participants in Sarasso et al.* (26).

DISCUSSION

Qualitatively, the DTF topographies of the wake and ketamine groups showed regions of relatively stronger and weaker apparent sources of outgoing connections. In contrast, both the propofol and xenon groups showed a more heterogeneous pattern, with outgoing connections of similar strength from most regions (Figure 1). Thus, the heterogeneity of these patterns was high for the two conditions characterized by conscious experience (wake 1.01, ketamine 1.26), and low in the two unconscious states (propofol 0.63, xenon 0.49). The group level patterns of connectivity seemed to hold on the level of individual participants as well (Figure 2). Based on these differences, the DTF-based confidence of being classified as conscious was significantly lower when subjects underwent propofol or xenon anesthesia than when they were awake ($p<0.05$). In contrast, there were no statistically significant differences when comparing the DTF-based confidence of being classified as conscious during ketamine anesthesia to the awake state ($p>0.05$; Figure 3). In addition, we found a significant correlation between the DTF-based confidence of being classified as conscious and the PCI-values computed by Sarasso et al. (26). This indicates that states yielding high values of PCI also had a high average probability of being classified as conscious by the DTF-based classification, and vice versa.

At least four other studies have reported changes in EEG connectivity quantified by DTF in relation to changing states of consciousness (17,19,20,22). Höller et al. found that the DTF was among the strongest measures for the separating vegetative and minimally conscious patients (25). Earlier, Bertini et al. showed that interhemispheric EEG connectivity quantified by DTF changed between wakefulness and stage 2 sleep, particularly in posterior regions (22). Similarly, Gennaro et al. showed that the EEG connectivity from posterior electrodes to frontal electrodes was reduced just after sleep onset (23). Finally, Kaminski et al. reported a "*diminishing role of the posterior sources and an increasing effect of the anterior areas*" at the onset of sleep (17). These findings, together with our earlier results showing abrupt changes when patients are anesthetized, suggest that changes in DTF may reflect properties related to (un)consciousness (8). However, whether and how the DTF-based measure may reflect or be related to neural correlates of consciousness remains to be determined.

In contrast to our results, Lee et al. (26), using normalized symbolic transfer entropy, found that ketamine had effects on connectivity that resembled those of propofol and sevoflurane. Thus, they found that all three anesthetics selectively inhibited frontal-parietal feedback connectivity, indicating that diverse anesthetics disrupt frontal-parietal communication, despite molecular and neurophysiologic differences. This difference in results may be related to different conditions (surgery vs. experimental setting), subtle differences in anesthetic protocols, or just an artefact of the different measures and methods used for analysis.

The aim of this study was to test whether the DTF-based measure, applied to raw EEG-signals, could be used uses to objectively distinguish conscious from unconscious states in humans. Our results seem to indicate that our method was successful in this respect. However, neither PCI nor the DTF-based measure distinguishes between connected and disconnected forms of consciousness, i.e. consciousness without externally relevant content, such as vivid dreams during ketamine anesthesia (26,31). Thus, both measures show overlapping distributions of values for normal wakefulness and ketamine dreams. For practical purposes, such as assessing the level of consciousness in the clinic, it is clearly often important to be able to distinguish between connected and disconnected consciousness, since only the latter is acceptable for general, surgical anesthesia. Although, connected consciousness can often be directly assessed clinically, this is not always the case (1,27,28). Even though specially designed tests based on communication may reveal connected consciousness in some nonresponsive patients (1,32), such tests may fail because of sensory deficits. Thus, further research is needed to devise objective methods for detecting connected and disconnected consciousness in non-communicating patients.

Even though the DTF-based measure seems fairly robust, three data points fell outside the main pattern intended to separate conscious from unconscious states. Two participants who did not report having an experience, and who's PCI-value indicated unconsciousness, were classified as awake more often than not by the DTF-based classification. Also, one subject in the wakeful state, who reported subjective experience and had a PCI-value indicating consciousness, was classified as awake less than half the time. These "misclassifications" could have several explanations. Because PCI requires > 5 minutes with TMS-EEG data to give an overall value for the whole segment recorded (3), measurements from brief periods of transient conscious states might not significantly influence the value obtained from TMS-EEG data, e.g. unintended awakenings during anesthesia and dreams. For example, due to the DTF-based measure´s higher temporal resolution compared to PCI, it might conceivably capture brief periods that deviate from the clinically observed conscious state of the subject. One study found that up to 60% of all subjects undergoing anesthesia report some subjective experience (33). For this conceivable problem, the DTF-based method may be an important tool for detecting shorter segments of conscious state that may go undetected in clinical evaluation and PCI calculation, and thus be (mis)classified as unconscious. On the other hand, the DTF-based measure could also just be sensitive to irrelevant properties of the EEG-signal leading to misclassification, as has been shown to be the case for the most popular clinical monitor of anesthesia (34).

If we consider the subject's own report of experience as the ground truth about the subject's conscious state, our results indicate that the DTF-based classification algorithm classifies participants correctly as conscious with 100% sensitivity. The accuracy, however, peaks at 93.3%, as two unconscious participants were mistakenly classified as conscious. The lower specificity as compared to the PCI values may, for many purposes, be counterbalanced by the fact that the DTF-based classification is based on automatic processing steps, works directly on raw, spontaneous data, and can update its classification every second to probe the dynamics of an individual's state. A high DTF-based confidence of being classified as conscious indicates that the algorithm had a high probability of classifying the subject as awake throughout the time-course. In contrast, a low DTF-based based confidence of being classified as conscious indicates that the algorithm was unlikely to classify the subject as awake throughout the time-course. Unfortunately, an intermediate DTF-based value may reflect at least two different causes: either the algorithm was not always reliable throughout the time-course, or the confidence of the algorithm fluctuated (either rapidly or between stable states of confidence), or a combination of these.

The DTF-based measure distinguishes between conscious and unconscious states during different forms of general anesthesia and wakefulness, and it correlates strongly with PCI. It may overcome some of the limitations of the PCI method related to its relatively low temporal resolution. PCI requires more expensive equipment (navigated TMS), and also extensive training of personnel for experimental and analysis tasks, as well as significant computational effort in data processing and analysis (3,26). Since the DTF-based confidence of being classified as conscious is defined for all 1-second segments in a given data set, the temporal resolution is relatively high. This can be important in many clinical situations, such as monitoring patients undergoing anesthesia during surgery, or bedside monitoring of patients suffering from disorders of consciousness. Furthermore, the DTF-based measure could presumably easily be made automatic, and used as the basis for real-time, low-cost monitoring of the state of consciousness.

There are several remaining challenges which must be tackled before general claims about the relation between the DTF-based measure and consciousness can be made. First and foremost, more studies including a variety of distinct states of consciousness need to be completed, and the number of participants included in the studies should be increased. Furthermore, the choice parameters used for the DTF analysis should be investigated in depth to uncover the relationship between the scalp-level EEG connectivity and the underlying brain connectivity. Our choices of parameters (such as choice of channels, model order, frequency band etc.), as well as the fact that pre-processing steps such as filtering and artefact rejection were omitted deliberately in this study, may have had an impact on the precision of inference about the brain connectivity (35,36). That said, whether the scalp level connectivity estimates are good estimators of the underlying, neural connectivity is disputed (37,38). However, as previously stated, the aims here were to test the DTF-based algorithm as an objective measure for separating states of consciousness during anesthesia, not to investigate whether the DTF reflects real changes in brain connectivity.

To this end, the DTF-based measure was able to distinguish between conscious and unconscious states in accordance with the participant´s own report of (un)conscious state. This was achieved using an automatic algorithm to short segments of spontaneous EEG. Furthermore, the DTF-based measure correlated with the PCI measure as it classified participants experiencing dreams during ketamine anesthesia as conscious, but classified participants undergoing xenon and propofol anesthesia as unconscious.

ACKNOWLEDGMENTS

This work was supported by the Human Brain Project (HBP: 720270-HBP SGA1-H2020) and the Norwegian Research Council (NRC: 262950/F20 and 214079/F20). We would also like to thank Cecilie P. Oksvold for her continued assistance in the lab.

REFERENCES

1. Owen AM, Coleman MR, Boly M, Davis MH, Laureys S, Pickard JD. Detecting Awareness in the Vegetative State. Science. 2006 Sep 8;313(5792):1402–1402.

2. Sanders RD, Tononi G, Laureys S, Sleigh J. Unresponsiveness ≠ Unconsciousness. Anesthesiology. 2012 Apr;116(4):946–59.

3. Casali AG, Gosseries O, Rosanova M, Boly M, Sarasso S, Casali KR, et al. A Theoretically Based Index of Consciousness Independent of Sensory Processing and Behavior. Sci Transl Med. 2013 Aug 14;5(198):198ra105–98ra105.

4. Koch C, Massimini M, Boly M, Tononi G. Neural correlates of consciousness: progress and problems. Nat Rev Neurosci. 2016 May;17(5):307–21.

5. Sebel PS, Bowdle TA, Ghoneim MM, Rampil IJ, Padilla RE, Gan TJ, et al. The Incidence of Awareness During Anesthesia: A Multicenter United States Study: Anesth Analg. 2004 Sep;99(3):833–9.

6. Barrett AB, Seth AK. Practical Measures of Integrated Information for Time-Series Data. PLoS Comput Biol [Internet]. 2011 Jan [cited 2014 Jul 1];7(1). Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3024259/

7. Boly M, Garrido MI, Gosseries O, Bruno M-A, Boveroux P, Schnakers C, et al. Preserved Feedforward But Impaired Top-Down Processes in the Vegetative State. Science. 2011 May 13;332(6031):858–62.

8. Juel BE, Romunstad LG, Kolstad L, Storm JF, Larsson PG. Classification of consciousness during propofol infusion in patients using the directed transfer function. Rev. 2017;

9. King J-R, Sitt JD, Faugeras F, Rohaut B, El Karoui I, Cohen L, et al. Information Sharing in the Brain Indexes Consciousness in Noncommunicative Patients. Curr Biol. 2013 Oct;23(19):1914–9.

10. Ku S-W, Lee U, Noh G-J, Jun I-G, Mashour GA. Preferential Inhibition of Frontal-to-Parietal Feedback Connectivity Is a Neurophysiologic Correlate of General Anesthesia in Surgical Patients. PLoS ONE [Internet]. 2011 Oct 5 [cited 2015 Aug 12];6(10). Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3187752/

11. Lee U, Mashour GA, Kim S, Noh G-J, Choi B-M. Propofol induction reduces the capacity for neural information integration: Implications for the mechanism of consciousness and general anesthesia. Conscious Cogn. 2009 Mar;18(1):56–64.

12. Maksimow A, Silfverhuth M, Långsjö J, Kaskinoro K, Georgiadis S, Jääskeläinen S, et al. Directional Connectivity between Frontal and Posterior Brain Regions Is Altered with Increasing Concentrations of Propofol. PLoS ONE. 2014 Nov 24;9(11):e113616.

13. Nicolaou N, Hourris S, Alexandrou P, Georgiou J. EEG-Based Automatic Classification of "Awake" versus "Anesthetized" State in General Anesthesia Using Granger Causality. PLoS ONE [Internet]. 2012 Mar 22 [cited 2015 Aug 12];7(3). Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3310868/

14. Tononi G. An information integration theory of consciousness. BMC Neurosci. 2004;5(1):42.

15. Tononi G, Boly M, Massimini M, Koch C. Integrated information theory: from consciousness to its physical substrate. Nat Rev Neurosci. 2016 May 26;17(7):nrn.2016.44.

16. Casarotto S, Comanducci A, Rosanova M, Sarasso S, Fecchio M, Napolitani M, et al. Stratification of unresponsive patients by an independently validated index of brain complexity. Ann Neurol. 2016 Nov;80(5):718–29.

17. Massimini M, Boly M, Casali A, Rosanova M, Tononi G. A perturbational approach for evaluating the brain's capacity for consciousness. Prog Brain Res. 2009;177:201–14.

18. Kaminski MJ, Blinowska KJ. A new method of the description of the information flow in the brain structures. Biol Cybern. 1991 Jul;65(3):203–10.

19. Astolfi L, Cincotti F, Mattia D, Marciani MG, Baccala LA, de Vico Fallani F, et al. Comparison of different cortical connectivity estimators for high-resolution EEG recordings. Hum Brain Mapp. 2007 Feb 1;28(2):143–57.

20. Kamiński M, Ding M, Truccolo WA, Bressler SL. Evaluating causal relations in neural systems: granger causality, directed transfer function and statistical assessment of significance. Biol Cybern. 2001 Aug;85(2):145–57.

21. Schumacher EM, Westvik AS, Larsson PG, Lindemann R, Westvik J, Stiris TA. Feasibility of long-term continuous EEG monitoring during the first days of life in preterm infants: an automated quantification of the EEG activity. Pediatr Res. 2011 May;69(5 Pt 1):413–7.

22. Bertini M, Ferrara M, De Gennaro L, Curcio G, Moroni F, Babiloni C, et al. Directional information flows between brain hemispheres across waking, non-REM and REM sleep states: An EEG study. Brain Res Bull. 2009 Mar 30;78(6):270–5.

23. Gennaro LD, Vecchio F, Ferrara M, Curcio G, Rossini PM, Babiloni C. Changes in fronto-posterior functional coupling at sleep onset in humans. J Sleep Res. 2004 Sep 1;13(3):209–17.

24. Kamiński M, Blinowska K, Szelenberger W. Investigation of coherence structure and EEG activity propagation during sleep. Acta Neurobiol Exp (Warsz). 1995;55(3):213–9.

25. Höller Y, Thomschewski A, Bergmann J, Kronbichler M, Crone JS, Schmid EV, et al. Connectivity biomarkers can differentiate patients with different levels of consciousness. Clin Neurophysiol. 2014 Aug;125(8):1545–55.

26. Sarasso S, Boly M, Napolitani M, Gosseries O, Charland-Verville V, Casarotto S, et al. Consciousness and Complexity during Unresponsiveness Induced by Propofol, Xenon, and Ketamine. Curr Biol. 2015 Jul 12;25(23):3099–105.

27. Ramsay MAE, Savege TM, Simpson BRJ, Goodwin R. Controlled Sedation with Alphaxalone-Alphadolone. Br Med J. 1974 Jun 22;2(5920):656–9.

28. Delorme A, Makeig S. EEGLAB: an open source toolbox for analysis of single-trial EEG dynamics including independent component analysis. J Neurosci Methods. 2004 Mar 15;134(1):9–21.

29. He B, Dai Y, Astolfi L, Babiloni F, Yuan H, Yang L. eConnectome: A MATLAB Toolbox for Mapping and Imaging of Brain Functional Connectivity. J Neurosci Methods. 2011 Feb 15;195(2):261–9.

30. Lee U, Ku S, Noh G, Baek S, Choi B, Mashour GA. Disruption of Frontal-Parietal Communication by Ketamine, Propofol, and Sevoflurane. Anesthesiology. 2013 Jun;118(6):1264–75.

31. Collier BB. Ketamine and the conscious mind. Anaesthesia. 1972 Apr 1;27(2):120–34.

32. Monti MM, Vanhaudenhuyse A, Coleman MR, Boly M, Pickard JD, Tshibanda L, et al. Willful Modulation of Brain Activity in Disorders of Consciousness. N Engl J Med. 2010;362(7):579–89.

33. Noreika V, Jylhänkangas L, Móró L, Valli K, Kaskinoro K, Aantaa R, et al. Consciousness lost and found: Subjective experiences in an unresponsive state. Brain Cogn. 2011 Dec;77(3):327–34.

34. Schuller PJ, Newell S, Strickland PA, Barry JJ. Response of bispectral index to neuromuscular block in awake volunteers. Br J Anaesth. 2015 Jul 1;115(suppl 1):i95–103.

35. Florin E, Gross J, Pfeifer J, Fink GR, Timmermann L. Reliability of multivariate causality measures for neural data. J Neurosci Methods. 2011 Jun 15;198(2):344–58.

36. Kaminski M, Blinowska KJ. Directed Transfer Function is not influenced by volume conduction—inexpedient pre-processing should be avoided. Front Comput Neurosci [Internet]. 2014 Jun 10 [cited 2015 Aug 20];8. Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4050361/

37. Brunner C, Billinger M, Seeber M, Mullen TR, Makeig S. Volume Conduction Influences Scalp-Based Connectivity Estimates. Front Comput Neurosci [Internet]. 2016 [cited 2017 Jan 3];10. Available from: http://journal.frontiersin.org/article/10.3389/fncom.2016.00121/abstract

38. Kaminski M, Blinowska KJ. The Influence of Volume Conduction on DTF Estimate and the Problem of Its Mitigation. Front Comput Neurosci [Internet]. 2017 May 12;11. Available from: http://www.ncbi.nlm.nih.gov/pmc/articles/PMC5427064/