The clinical diagnostic utility of electrophysiological techniques in assessment of patients with disorders of consciousness following acquired brain injury –A systematic review

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Key words (MeSH):

Consciousness Disorder, Persistent vegetative state, Minimally conscious state, Electrophysiology, Electroencephalography, Sensitivity Specificity, Diagnostic errors, Attention, Cognition

Objective: To investigate the diagnostic utility of electrophysiological recordings during active cognitive tasks in detecting residual cognitive capacities in patients with disorders of consciousness (DoC) after severe acquired brain injury.

Design: Systematic review of empirical research in Medline, Embase, PsycINFO, and Cochrane from January 2002 to March 2016.

Main Measures: Data extracted included sample size, type of electrophysiological technique and task design, rate of cognitive responders, false negatives and positives, and excluded subjects from study analysis. The Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2) was used for quality appraisal of the retrieved literature.

Results: Twenty-four studies examining electrophysiological signs of command-following in patients with DoC were identified. Sensitivity rates in healthy controls demonstrated variable accuracy across the studies, ranging from 71% to 100%. In patients with DoC, specificity and sensitivity rates varied in the included studies, ranging from 0% to 100%. Pronounced heterogeneity was found between studies regarding methodological approaches, task design and procedures of analysis, rendering comparison between studies challenging.

Conclusion: We are still far from establishing precise recommendations for standardized electrophysiological diagnostic procedures in DoC, but electrophysiological methods may add supplemental diagnostic information of covert cognition in some patients with DoC.

INTRODUCTION

Background

Developments in neuroimaging and electrophysiological methods have allowed both structural and functional studies of the living brain, enabling online monitoring of mental processes, including the neural correlates of human behavior.[1] Hence, much of contemporary evidence and theories of brain processes are informed by neuroimaging techniques, offering insight into age-old questions about brain-behavior relationships, and an emerging understanding of underlying neural mechanisms.[2-4] Although previously regarded as scientifically intractable, consciousness can now be studied with modern neuroscientific techniques, such as positron emission tomography (PET)[5], functional (fMRI)[6], and structural (diffusion tensor imaging; DTI)[7] Magnetic Resonance Imaging (MRI), and electrophysiological techniques.[8]

In parallel with this methodological development, a great increase in scientific interest has taken place with respect to patients with disorders of consciousness (DoC) following severe acquired brain injury, i.e. patients in either a vegetative (VS), also referred to as the “unresponsive wakefulness syndrome” (UWS),[9] or a minimally conscious state (MCS). Whereas the VS is characterized by absence of any behavioral signs of awareness, but regained intermittent wakefulness, the MCS, by contrast, is characterized by the presence of inconsistent, but clearly discernible behavioral evidence of awareness of self or the environment (i.e. visual pursuit, localization to pain, or reproducible command-following).[10,11] Recently, the MCS entity has been suggested to be divided into MCS+ and MCS-, depending on the complexity of behavioral responses. While MCS+ is characterized by more complex cognitive capacities, i.e presence of command-following, MCS-, is on the other hand characterized by nonlinguistic and simple signs of conscious awareness. However, consensus on a clear definition of MCS + and MCS- is currently lacking.[12,13] Novel neuroimaging and electrophysiological techniques have offered new insight and enhanced theoretical understanding of these patients` level of consciousness, brain connectivity, metabolic and cognitive functioning.[14]

The current standard approach to clinical diagnosis of DoC is based upon behavioral assessment strategies, along with patient history and structural brain imaging.[15] Notably, rates of misdiagnosis in DoC have been estimated to be as high as ≈40%.[16-18] The lack of a 'gold standard' for detection of conscious awareness in DoC is a prominent confounding factor for accurate diagnostic assessment, and it is recommended to apply standardized neurobehavioral rating scales designed to detect subtle, but clinically significant signs of consciousness.[19,20] In a comprehensive evidence-based review of the psychometric properties of existing assessment scales, the Coma Recovery Scale-Revised (CRS-R) was recommended with minor reservation, while the Sensory Modality Assessment Technique (SMART), Western Neuro Sensory Stimulation Profile (WNSSP), Sensory Stimulation Assessment Measure (SSAM), Wessex Head Injury Matrix (WHIM), and Disorders of Consciousness Scale (DOCS) were recommended with moderate reservations.[21]

Clinical diagnostic utility of electrophysiological methods in patients with DoC

Advances in neuroscientific methodology has led to optimism regarding potential clinical utility in diagnostic and prognostic considerations in patients with DoC, [22-24] in part due to several studies indicating that cognitive processing can be detected with imaging techniques in the absence of behavioral signs of consciousness.[5,25-29] These studies applied tasks that require subjects to exert mental responses to command,[30,31] in contrast to merely passive paradigms eliciting only "automatic" responses. Hence, in order to infer consciousness, it is necessary to include tasks involving active cognitive processing in combination with functional neuroimaging- and electrophysiological methods.[32] However, functional imaging-methods, such as fMRI and PET require high levels of technical skills, are expensive, and most often not readily accessible in rehabilitation facilities. On the other hand, electrophysiological techniques are more readily available by having the benefit of being of low-cost, noninvasive, and can be conducted repeatedly at bedside. Herein, Event-related potentials (ERPs) represent time-locked electroencephalographic (EEG) activity elicited by external events, thus providing a neurophysiological correlate of cognitive processing at the millisecond level, from early and largely sensory components to later and cognitively mediated waveforms, such as the P3.[33-35] Task-related systematic changes in oscillatory variation can also be an index of cognitive effort, and can be detected through the analysis of frequency bands, i.e. event-related desynchronisation (ERD).[36,37] Such electrophysiological features or activation patterns can also be applied in machine learning systems that allow quantification of differences in neural responses at an individual level.[38,39] Surface electromyogram (EMG) is, on the other hand, recordings of electrical activity in muscles, and is a commonly used tool to study physiological principles of muscles related to movement generation.[40,41]

Objectives of the systematic review

Although modern functional imaging and EEG-based techniques have given rise to hopes of improved diagnostic accuracy in DoC,[19,42] the body of existing systematic reviews and overview articles have various shortcomings in providing a sufficient estimate of the clinical usefulness of neurophysiological measures. A major limitation of existing reviews is the lack of reports regarding rates of responders, meaning subjects showing signs of active mental effort during electrophysiological assessment, both in healthy subjects and patients with DoC, and also an insufficient account of false negatives,[43-49] i.e. the rate of persons who do not display clear signs of cognitive effort in electrophysiological assessments, despite definite voluntary behavioral responses. Some reviews lack a representative body of included studies, either due to overly strict study inclusion criteria regarding sensitivity/specificity,[50] while others have not required use of active paradigms, rendering degree of consciousness uninterpretable.[44,51] Yet other papers only provide a topical overview without explicit systematic literature search strategies.[38,46,48,49,52-54] In addition, no existing review provides an overview over the rate of excluded subjects across studies due to methodological artifacts, which is quite common in electrophysiological methods in general, and might be expected to be even higher in groups known to have ample muscle artifact, and lack cooperative abilities in the engaged test-situation.

In summary, it is still not well described to what extent the combination of experimental paradigms with active conditions during electrophysiological recordings can complement standardized neurobehavioral assessment, or which type of experimental procedure or neurophysiological measure may be best suited. Both are paramount in order to establish the diagnostic value of the methods in clinical practice, where correct assessment of the level of consciousness in patients with DoC is crucial, but challenging. In a clinical context, it is necessary to establish to what extent we can gain additional diagnostic information from electrophysiological assessments at an individual patient level. The aim of this review was to examine the diagnostic utility of electrophysiological methods where active cognitive tasks have been applied to detect covert cognition in patients with DoC due to mixed etiologies. In order to evaluate the potential for clinical translation, two main issues were explored: Firstly, the experimental robustness of various published experimental paradigms was explored in healthy volunteers, who are by definition perfectly conscious. Secondly, the rate of patients with DoC who show electrophysiological responses indicating command following (responders) was assessed, as well as the rate of false negatives and positives.

METHODS

Inclusion criteria

Methods of the analysis and inclusion criteria were specified in advance and documented in a protocol, adhering to established recommendations for conducting systematic reviews,[55-57] including the PRISMA guidelines.[58-60] The full review protocol can be accessed in the Supplemental Digital Content 1, as well as PRISMA checklist in Content 2. Studies were included in the systematic review if they involved electrophysiological methods used in combination with experimental paradigms encompassing active conditions. Furthermore, only English empirical studies with more than five subjects were included. Studies were included if they investigated patients who met the diagnostic criteria for VS and MCS after acquired brain injury, where level of consciousness was established with a standardized behavioral assessment tool with acceptable psychometric properties, i.e. either the CRS-R, WHIM, SSAM, WNSSP, DOCS or SMART scales.[21] A further inclusion criterion required publication after the consensus-based criteria for diagnosing MCS, published in 2002.[10] Literature reviews and systematic reviews were excluded.

Search method for identification of studies

We undertook a systematic review of the literature and selected relevant studies published between January 2002 and March 2016 in the following databases: Medline, Embase, PsycINFO, Database of Abstracts of reviews of effects (Cochrane Library), and Cochrane Central Register of Controlled Trials (Cochrane Library). Primary search terms used defining DoC were: *Consciousness disorder, disorder of consciousness, vegetative state, persistent* *vegetative state, unresponsive wakefulness syndrome, or minimally conscious state.* Primary terms were paired with secondary terms defining aspects of electrophysiological measurement*: electrodiagnosis, electrophysiology, neurophysiology, electroencephalography, encephalogram, EEG, myography, or electromyography.* These were furthermore paired with third terms related to measure outcome: *Event Related Potentials, ERP, evoked potentials, P300, active task/condition/paradigm, residual function, covert attention/awareness/cognition or command-following.* We last searched the electronic databases on March 7th, 2016. See Supplemental Digital Content 3 for a full description of Medline search strategy. As studies were identified, researchers also checked for additional relevant articles being cited.

Study selection and analysis

Selection of studies

Titles and abstracts were reviewed first, and when indicating relevance, full text articles were assessed using the inclusion and exclusion criteria to exclude those papers that were not relevant to this review. The initial selection was conducted by one author (SLH), and double-checked by an independent second author (ML). Any disagreements were resolved by consensus, and if no agreement could be reached, it was planned that a third author would decide (author SA). One study author was contacted for additional information regarding clarification of the included study sample. Data was extracted by author SLH, and verified by author ML.

Quality appraisal of retrieved literature

Quality appraisal of the retrieved literature was conducted using the Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2). The initial assessment was conducted by author SLH, and verified by a second author (ML). The QUADAS-2 checklist assesses the risk of bias and concerns regarding applicability over four domains: patient selection, index test, reference standard, and flow and timing,[61] see Supplemental Digital Content 4 for QUADAS-2 questions. Patient selection was regarded to be at high risk of bias if the study did not primarily include patients in a medically stable phase, or in cases of insufficient differential diagnosis, i.e. from coma or Locked-In-Syndrome, was not based on a consecutive or random sample, or did not clearly avoid inappropriate exclusion, i.e. outpatients or concurrent referrals. Unblinded interpretation of the electrophysiological assessment, and lack of detailed descriptions of procedures for processing of EEG-data and experimental procedures was considered to represent a high risk of bias concerning the electrophysiological index test. The reference standard was considered to be at high risk if the behaviorally based diagnostic conclusion did not adhere to established consensus-based diagnostic criteria for VS and MCS,[10,11] and if the interpretation of the behavioral assessment was not blinded to the results of the electrophysiological assessment. Concerns regarding applicability were related to the representativeness of the studies in relation to the review questions, such as sample representatives, clearness and relevance of processing and interpretation of electrophysiological data in assessing consciousness, and adherence to diagnostic criteria for DoC.

Statistical analysis

Individual responder rates in both healthy controls and patient groups were described with actual numbers of subjects and percentage per study. Patients who displayed unequivocal behavioral signs of command-following were classified as MCS+, while patients with no reproducible behavioral response to command were classified as MCS-, in accordance with the definition provided by Bruno et al.[13] Sensitivity and specificity were computed using data from the published articles and calculated with 95% confidence intervals (CI) per study, with the behavioral assessment as the reference standard and VS and MCS- as the disorder of interest. Sensitivity was understood as the ability of the electrophysiological assessment to detect command-following in patients behaviorally classified as MCS+. Specificity was understood as the ability of electrophysiological techniques to confirm the behaviorally based VS or MCS- diagnosis, by the lack of electrophysiological signs of command-following. However, accurate calculation of sensitivity and specificity in patients with DoC is difficult, due to the lack of a true gold standard measure of level of consciousness.

RESULTS

Characteristics of the included studies

As illustrated in Figure 1, a total of 832 articles were initially identified from the search process, and nine were identified through other sources. Twenty-four studies were finally included for review. The characteristics of these studies are summarized in table 1.

Included study samples. Of the 24 studies, seven did not include a healthy control group for the active paradigm,[62-68] whereof four referred to previously published healthy control data.[62,63,65,67] The studies varied considerably with regard to sample sizes, from only six included patients[62,63] to a total of 167 electrophysiological recordings acquired from 113 patients in the largest study.[69] Overall, many studies were characterized by small sample sizes.

Behavioral assessment tool. All studies applied the CRS-R as the behavioral assessment scale of choice, except for one, where WHIM was applied.[64] Hence, the included studies represented uniform and sound procedures for behavioral diagnosis of consciousness.

Electrophysiological techniques. The included studies displayed a wide variation with regard to applied index tests. The majority of the studies applied EEG-based technology, while two included studies used experimental tasks with EMG.[64,70] Ten studies applied systems using EEG in combination with machine-learning, where algorithms were used to identify "patterns" of brain activity using a classifier method (for a review of classifier methods, see[71]). A subgroup of studies applied complex multivariate classifier methods, integrating data from a variety of electrophysiological features based on recordings during active tasks, e.g. ERPs, frequency power, complexity and connectivity measures.[69,72]

Design/task. The systematic review revealed considerable heterogeneity with regard to types of active experimental paradigms applied. The majority of tasks fell into two main categories; either imagery tasks, or tasks requiring counting an auditory target stimuli, while only three studies involved visual stimuli.[73-75] Five imagery tasks included instructions to imagine motor movements, e.g. squeezing hand, moving toes, or moving arm towards an object.[62,63,65,73,76] Fourteen studies included the active instruction to count either a target name or word,[27,66,77-80] occurrence of deviant tones,[81,82] or a target global deviant.[67-69,72,83,84] The latter has been repeatedly studied in a "local-global" paradigm consisting of series of tone sequences containing a two-level structure of occasional irregularities in short-term ("local") violations within a five-sound sequence, and long-term ("global") violations of the expectancies of such sequences.[83] Seven studies included subjectively relevant stimuli, e.g. photo of the subject,[74] a customized familiar motor imagery task,[62] or the subject`s own name (SON), where SON was applied in five studies.[26,27,66,77,78] All experimental tasks included verbally delivered instructions.

Excluded subjects. Not all studies provided information of whether subjects were excluded from analysis or not. Notably, some studies reported high rates of excluded subjects in the patient group. For example, Gibson and colleagues reported exclusion of five of 11 patients from the EEG-analysis.[62] Chennu and colleagues reported exclusion of nine out of 30 recruited patients,[80] and in the study of Faugeras and colleagues,[67] a total of 35 out of 100 patient EEG-recordings were excluded. Data exclusion was mainly due to low quality of EEG-recordings, and excessive noise artifacts in patients with DoC, demonstrating one of the intrinsic limitations of this approach. Also, exclusion of EEG-data from healthy controls due to artifacts was explicitly reported in two studies.[78,84]

Diagnostic performance. Table 2 illustrates calculated rates of sensitivity and specificity per study in healthy subjects and patients with DoC, except from five studies, due to results only confined to the group level,[73,77] lack of reports on individual patient behavioral responses,[81,82] or because comparison between EEG-responses and behaviorally based diagnosis was not possible.[75] Sensitivity and specificity rates in patients with DoC were calculated with the behavioral assessment as the reference standard, although a true gold standard to confirm consciousness level is nonexistent. In healthy controls, the studies displayed a relatively wide variability with regard to sensitivity rates, ranging from 71% to 100%. A high false negative rate up to 29% showed that the electrophysiological test failed to detect active mental effort in a considerable number of healthy subjects, while other studies identified all control subjects as responders.[27,74,75,83,84] There was also a wide variety in sensitivity rates in the patient group, ranging from 0% to 100%. Here, a sensitivity rate of 0% showed that none of the included patients with discernible behavioral evidence of command-following (MCS+) were classified as responders in the active task.[63,79,80] Of notice, a sensitivity rate of 100% was in several studies the result of samples consisting of one single MCS+ -responder. Specificity rates in the patient groups also ranged from 0% to 100%, the latter again due to one single patient.[63] Notably, eight studies[27,64,69,74,76,78,83,85] demonstrated specificity rates of 80% or below, illustrating that more than 20% of patients who could not demonstrate response to command behaviorally, did so in the electrophysiological assessment.

Insert figure 1 here (PRISMA).

Insert table 1 here (study characteristics).

Insert table 2 here (CI calculations sensitivity/specificity).

Risk of bias

The QUADAS-2 assessment demonstrated that none of the 24 included studies had a low risk of bias or concerns regarding applicability across all domains (see table 3). Regarding patient selection, bias concern was found due to inclusion of patients in a very early sub-acute phase after severe acquired brain injury,[77,83] lack of information regarding time since injury,[75,81] only two studies clearly stated they were based on consecutive sample[66,67], and overall lack of clarifications about inappropriate exclusion avoided, i.e. outpatients or concurrent referrals. Applicability concerns regarding patient selection was due to potential sample representativity issues. Risk of bias was found with regard to the index and reference tests, as all studies, except one,[78] lacked clear statements of whether or not interpretation of the electrophysiological assessment was blinded to the behavioral assessment, or vice versa. Concern regarding applicability of the index test was thus found in all studies but one,[78] reflecting that there is no tradition of blinding in this field. Furthermore, the domain of flow and timing was overall of bias concern, as nine studies were scored as unclear or with high bias risk with regard to the time interval between the behavioral and electrophysiological assessment. Accordingly, this implicated a concern for the relation between behavioral and electrophysiological assessments.

Insert table 3 here.

DISCUSSION

Over the past decade, there has been increasing scientific effort aiming at assessing covert awareness in patients with DoC applying active paradigms during electrophysiological recordings. However, the diagnostic accuracy of electrophysiological methods is still not established. Furthermore, there is no consensus regarding which experimental designs and modes of analysis would be most applicable for clinical use at a single patient level. The aim of this systematic review was to identify existing studies and to explore the clinical utility of electrophysiological methods.

Task robustness of active paradigms in healthy control subjects

In order to evaluate the diagnostic potential of electrophysiological methods to detect remnant cognitive resources in DoC, a main aim was to establish the robustness of active experimental paradigms in healthy conscious subjects. This could not be done in the seven studies lacking a healthy control group. [62-68] However, the remaining studies had sensitivity rates in healthy controls varying from 71% to 100%. Of the three studies showing sensitivity rates below 80%,[26,76,78] two included an active condition with the instruction to listen for a change in pitch to the subject`s own name (SON).[26,78] The necessity of including personally relevant stimuli has previously been strongly emphasized, as the probability of electrophysiological responses in patients with DoC increases with salient self-referential stimuli,[86] and the person's own name (SON) has proven promising in this regard.[27,87-90] However, these results demonstrate that the cognitive content of the active condition is also of importance, as the instruction to count SON has proven to be more robust, with replicated high sensitivity rates in healthy subjects.[27,78] While SON is a complex meaningful salient stimulus, other studies have applied simple harmonic tones with the instruction to count a global auditory deviant, denoted as the "local-global" paradigm,[83] where high sensitivity rates in healthy subjects have been repeatedly demonstrated.[68,69,72,83,84] This review illustrates that far from all electrophysiological studies have shown 100% accuracy in healthy controls. In addition, even if a method is robust in healthy subjects, it remains a question whether the sensitivity will generalize to severe brain injury populations.

Diagnostic accuracy of electrophysiological measures in DoC

A second aim of this systematic review was to establish the rates of responders in patients with DoC, as well as the number of patients with behavioral command following that fail to show definite electrophysiological signs of active cognitive effort (false negatives). Sensitivity rates in patients with DoC varied markedly across the included studies, ranging from 0% to 100%, indicating on average that maybe as many as one third of patients that presented with unequivocal behavioral responses to command were not classified as responders based on their electrophysiological activity across studies. It is however challenging to disentangle whether lack of responsivity is due to patients' characteristics or the methodological limitations of the electrophysiological technique. Patients with DoC may suffer from severe underlying perceptual and cognitive impairments, such as deficits in language, working memory, attention, memory and executive functioning, potentially preventing them from responding in active tasks despite being conscious. Bias due to impaired hearing can be controlled for with auditory evoked potentials and by ensuring presence of the auditory N1 and/or mismatch negativity (MMN) components. Furthermore, the tasks in electrophysiological studies may demand higher cognitive abilities than what is required for displaying behavioral command-following, rendering CRS-R and electrophysiological results potentially incomparable. In addition, patients with DoC typically fluctuate both in their level of cognitive functioning and fatigue.[91] Also, active tasks containing verbal instructions to elicit willfully modulated mental processes are limited by the fact that they require language comprehension, constituting a comparable challenge to that inherent in all behavioral scales.[92] Consequently, negative EEG-findings in this patient group cannot be interpreted as evidence that the patient lacks awareness any more than a negative behavioral finding does so.[29,62,93]

Specificity rates also varied markedly, ranging from 0% to 100%, implying that some patients show signs of command-following in electrophysiological recordings, despite not doing so behaviorally (false positives). This could be related to small sample sizes, or might actually be due to the fact that behavioral measures, in some cases fail to detect the true level of functioning in the patient. Of note, the two largest studies containing 158 and 167 valid patient recordings, demonstrated false positive rates of 17% and 33%, respectively.[69,72] This highlights that, despite high rates of false negatives, covert signs of command-following have also been demonstrated. Notably, the number of patients showing electrophysiological signs of mental effort despite lack of behavioral command-following, is in line with those obtained in fMRI studies using active tasks.[38,94] In summary, the two large studies applying multivariate EEG-classifier systems most likely represent the method with best balance between rates of sensitivity and specificity.

Methodological issues

The review demonstrates heterogeneity with regard to the electrophysiological techniques applied. Even though EEG-based techniques were the most frequently applied method, with only two EMG-studies, there was variety in the mode of analysis, such as ERP and ERD, along with diversity in EEG features included in classifier methods, hence complicating comparison of results.

Furthermore, the electrophysiological methods are characterized by variations in, e.g. choice of EEG-equipment, protocols for EEG-recordings, and methods for data analysis. Notably, there are studies where data have been re-analyzed, showing diverging results regarding rates of responders both in healthy controls and VS/MCS patients.[76,95,96] Additionally, studies performed in different scientific laboratories conducting similar experimental paradigms have generated conflicting results. Using a variant of the local-global experiment, a different research group found responses to global deviants in 10/24 comatose patients following cardiac arrest, but only in six out of 21 healthy controls,[97] thus challenging previous results where the global effect has been interpreted as only being present in conscious subjects.[69,72,83,84] These conflicting results have led to a debate about divergences in methodological approaches.[98,99]

Further methodological challenges are illustrated in the QUADAS-2 assessment, demonstrating a bias concern with regard to whether the interpretation of the electrophysiological assessment was masked to the behavioral assessment and vice versa. In clinical trials, blinding of assessors is a common requirement, while this is not tradition within electrophysiological research, likely because the electrophysiological recording is not expected to be biased by rater expectations. However, there is a fair amount of subjective evaluations in processing and interpretation of EEG-data, rendering reason for bias concern. Also, the QUADAS-2 assessment illustrated that flow of timing between the electrophysiological assessment and behavioral diagnostic measure was a concern in as many as nine studies, highlighting that the lack of standardized and uniformly accepted methodological approaches is a real concern and a prerequisite for successful clinical translation.

Unfortunately, not all studies reported on the rate of excluded subjects, while others reported relatively high exclusion numbers due to artifacts, even in healthy subjects. In clinical practice, this means that there is a relatively high risk that a time-consuming assessment will not provide interpretable data.

In summary, there are several general and method-specific advantages and disadvantages with electrophysiological techniques applied in the included studies. High levels of artifacts remain an issue of concern in all methods described. In particular, relying on motor responses in EMG-tasks is problematic due to frequent severe motor deficits such as paresis, spasticity and contractures. When it comes to EEG frequency analysis (e.g. ERD), this method alone has not per date provided strong evidence of clinical applicability, but has been included as one of several components in multivariate feature analysis. Regarding ERP, the P3 is the component of choice in this particular diagnostic context, but as noted, the chance of providing evidence of consciousness is highly dependent on the experimental paradigm applied. Additionally, applying multivariate EEG-classifier systems might be less influenced by subjective rater bias.

Conclusions and implications for future studies

Determining where patients lie on the spectrum of conscious awareness, and assessment of residual cognitive resources, is essential in accurate diagnosis of patients with DoC. Electrophysiological methods have the potential to make important contributions. However, we are still far from establishing precise recommendations for standardized electrophysiological diagnostic measures in DoC. A necessary step in future research is to initiate multi-center studies, as a means to establish comparable data sets with large sample sizes across laboratories, and to further establish sensitivity and specificity. Herein, ensuring systematic validation of electrophysiological paradigms in healthy controls is essential. Both false positive and false negative rates may have important implications for clinical decision-making, e.g. pain management, intensity of rehabilitation, and sometimes end-of-life decisions. In summary, one needs to cautiously balance the risk of false positive versus false negative diagnostic errors in individual assessments, as it is evident that a patient with discernible signs of behavioral command-following can appear as a false negative electrophysiologically. Thus, standardized behavioral measures still constitute the standard approach to diagnostic assessment. However, in cases where severe motor deficit may mask a patient’s true level of consciousness, or where other factors contribute to diagnostic uncertainty, electrophysiological methods can complement behavioral measures with valuable additional clinical information.

Limitations

The main limitation of this systematic review is the difficulty of study comparison. Subsequently, the review focused on a qualitative synthesis of identified studies, as meta-calculation of pooled sensitivities and specificities across methods and experimental conditions was considered ineffectual. Also, as there is no established veridical benchmark of level of consciousness, precaution should be taken in interpreting results as precise estimates of sensitivity and specificity in patients with DoC.

REFERENCES

1. Tononi G, Koch C. The neural correlates of consciousness: an update. *Ann. N. Y. Acad. Sci.* 2008;1124:239-261.

2. de Graaf TA, Hsieh PJ, Sack AT. The 'correlates' in neural correlates of consciousness. *Neurosci. Biobehav. Rev.* 2012;36(1):191-197.

3. Fins JJ, Illes J, Bernat JL, Hirsch J, Laureys S, Murphy E. Neuroimaging and disorders of consciousness: envisioning an ethical research agenda. *The American journal of bioethics: AJOB.* 2008;8(9):3-12.

4. Block N, Carmel D, Fleming SM, et al. Consciousness science: real progress and lingering misconceptions. *Trends in cognitive sciences.* 2014;18(11):556-557.

5. Stender J, Gosseries O, Bruno MA, et al. Diagnostic precision of PET imaging and functional MRI in disorders of consciousness: a clinical validation study. *Lancet (London, England).* 2014;384(9942):514-522.

6. Vanhaudenhuyse A, Demertzi A, Schabus M, et al. Two distinct neuronal networks mediate the awareness of environment and of self. *J. Cogn. Neurosci.* 2011;23(3):570-578.

7. Jang SH, Kwon HG. The direct pathway from the brainstem reticular formation to the cerebral cortex in the ascending reticular activating system: A diffusion tensor imaging study. *Neurosci. Lett.* 2015;606:200-203.

8. Boly M, Moran R, Murphy M, et al. Connectivity changes underlying spectral EEG changes during propofol-induced loss of consciousness. *J. Neurosci.* 2012;32(20):7082-7090.

9. Laureys S, Celesia GG, Cohadon F, et al. Unresponsive wakefulness syndrome: a new name for the vegetative state or apallic syndrome. *BMC medicine.* 2010;8:68.

10. Giacino JT, Ashwal S, Childs N, et al. The minimally conscious state: definition and diagnostic criteria. *Neurology.* 2002;58(3):349-353.

11. Giacino J, Whyte J. The vegetative and minimally conscious states: current knowledge and remaining questions. *The Journal of head trauma rehabilitation.* 2005;20(1):30-50.

12. Bruno MA, Vanhaudenhuyse A, Thibaut A, Moonen G, Laureys S. From unresponsive wakefulness to minimally conscious PLUS and functional locked-in syndromes: recent advances in our understanding of disorders of consciousness. *J. Neurol.* 2011;258(7):1373-1384.

13. Bruno MA, Majerus S, Boly M, et al. Functional neuroanatomy underlying the clinical subcategorization of minimally conscious state patients. *Journal of neurology.* 2012;259(6):1087-1098.

14. Di Perri C, Heine L, Amico E, Soddu A, Laureys S, Demertzi A. Technology-based assessment in patients with disorders of consciousness. *Annali Dell'Istituto Superiore di Sanita.* 2014;50(3):209-220.

15. Peterson A, Cruse D, Naci L, Weijer C, Owen AM. Risk, diagnostic error, and the clinical science of consciousness. *NeuroImage Clinical.* 2015;7:588-597.

16. Andrews K, Murphy L, Munday R, Littlewood C. Misdiagnosis of the vegetative state: retrospective study in a rehabilitation unit. *BMJ (Clinical research ed.).* 1996;313(7048):13-16.

17. Childs NL, Mercer WN, Childs HW. Accuracy of diagnosis of persistent vegetative state. *Neurology.* 1993;43(8):1465-1467.

18. Schnakers C, Vanhaudenhuyse A, Giacino J, et al. Diagnostic accuracy of the vegetative and minimally conscious state: clinical consensus versus standardized neurobehavioral assessment. *BMC Neurol.* 2009;9:35.

19. Giacino JT, Fins JJ, Laureys S, Schiff ND. Disorders of consciousness after acquired brain injury: the state of the science. *Nat. Rev. Neurol.* 2014.

20. Giacino JT, Katz DI, Whyte J. Neurorehabilitation in disorders of consciousness. *Seminars in neurology.* 2013;33(2):142-156.

21. American Congress of Rehabilitation Medicine BI-ISIGDoCTF, Seel RT, Sherer M, et al. Assessment scales for disorders of consciousness: evidence-based recommendations for clinical practice and research. *Arch Phys Med Rehabil.* 2010;91(12):1795-1813.

22. Laureys S, Boly M. The changing spectrum of coma. *Nat. Clin. Pract. Neurol.* 2008;4(10):544-546.

23. Daltrozzo J, Wioland N, Mutschler V, Kotchoubey B. Predicting coma and other low responsive patients outcome using event-related brain potentials: a meta-analysis. *Clin. Neurophysiol.* 2007;118(3):606-614.

24. Bodart O, Laureys S, Gosseries O. Coma and disorders of consciousness: scientific advances and practical considerations for clinicians. *Semin. Neurol.* 2013;33(2):83-90.

25. Owen AM, Coleman MR, Boly M, Davis MH, Laureys S, Pickard JD. Detecting awareness in the vegetative state. *Science.* 2006;313(5792):1402.

26. Schnakers C, Giacino JT, Lovstad M, et al. Preserved covert cognition in noncommunicative patients with severe brain injury? *Neurorehabilitation and neural repair.* 2015;29(4):308-317.

27. Schnakers C, Perrin F, Schabus M, et al. Voluntary brain processing in disorders of consciousness. *Neurology.* 2008;71(20):1614-1620.

28. Monti MM, Vanhaudenhuyse A, Coleman MR, et al. Willful modulation of brain activity in disorders of consciousness. *N. Engl. J. Med.* 2010;362(7):579-589.

29. Bardin JC, Fins JJ, Katz DI, et al. Dissociations between behavioural and functional magnetic resonance imaging-based evaluations of cognitive function after brain injury. *Brain: a journal of neurology.* 2011;134(Pt 3):769-782.

30. Monti MM, Coleman MR, Owen AM. Neuroimaging and the vegetative state: resolving the behavioral assessment dilemma? *Ann. N. Y. Acad. Sci.* 2009;1157:81-89.

31. Owen AM, Schiff ND, Laureys S. A new era of coma and consciousness science. *Prog. Brain Res.* 2009;177:399-411.

32. Schnakers C. Clinical assessment of patients with disorders of consciousness. *Archives italiennes de biologie.* 2012;150(2-3):36-43.

33. Duncan CC, Barry RJ, Connolly JF, et al. Event-related potentials in clinical research: guidelines for eliciting, recording, and quantifying mismatch negativity, P300, and N400. *Clin. Neurophysiol.* 2009;120(11):1883-1908.

34. Reinvang I. Cognitive event-related potentials in neuropsychological assessment. *Neuropsychol. Rev.* 1999;9(4):231-248.

35. Soltani M, Knight RT. Neural origins of the P300. *Critical reviews in neurobiology.* 2000;14(3-4):199-224.

36. Pfurtscheller G, Aranibar A. Event-related cortical desynchronization detected by power measurements of scalp EEG. *Electroencephalogr. Clin. Neurophysiol.* 1977;42(6):817-826.

37. Sauseng P, Klimesch W. What does phase information of oscillatory brain activity tell us about cognitive processes? *Neurosci. Biobehav. Rev.* 2008;32(5):1001-1013.

38. Noirhomme Q, Brecheisen R, Lesenfants D, Antonopoulos G, Laureys S. "Look at my classifier's result": Disentangling unresponsive from (minimally) conscious patients. *Neuroimage.* 2015.

39. Naci L, Monti MM, Cruse D, et al. Brain-computer interfaces for communication with nonresponsive patients. *Annals of neurology.* 2012;72(3):312-323.

40. Mills KR. The basics of electromyography. *J. Neurol. Neurosurg. Psychiatry.* 2005;76 Suppl 2:ii32-35.

41. Enders H, Nigg BM. Measuring human locomotor control using EMG and EEG: Current knowledge, limitations and future considerations. *European journal of sport science.* 2015:1-11.

42. Owen AM. Using functional magnetic resonance imaging and electroencephalography to detect consciousness after severe brain injury. *Handb. Clin. Neurol.* 2015;127:277-293.

43. Bender A, Jox RJ, Grill E, Straube A, Lule D. Persistent vegetative state and minimally conscious state: a systematic review and meta-analysis of diagnostic procedures. *Deutsches Arzteblatt international.* 2015;112(14):235-242.

44. Kondziella D, Friberg CK, Frokjaer VG, Fabricius M, Moller K. Preserved consciousness in vegetative and minimal conscious states: systematic review and meta-analysis. *J. Neurol. Neurosurg. Psychiatry.* 2015.

45. Harrison AH, Connolly JF. Finding a way in: a review and practical evaluation of fMRI and EEG for detection and assessment in disorders of consciousness. *Neurosci. Biobehav. Rev.* 2013;37(8):1403-1419.

46. Peterson A, Cruse D, Naci L, Weijer C, Owen AM. Risk, diagnostic error, and the clinical science of consciousness. *NeuroImage. Clinical.* 2015;7:588-597.

47. Luaute J, Maucort-Boulch D, Tell L, et al. Long-term outcomes of chronic minimally conscious and vegetative states. *Neurology.* 2010;75(3):246-252.

48. Gosseries O, Zasler ND, Laureys S. Recent advances in disorders of consciousness: focus on the diagnosis. *Brain Inj.* 2014;28(9):1141-1150.

49. Laureys S, Schiff ND. Coma and consciousness: paradigms (re)framed by neuroimaging. *Neuroimage.* 2012;61(2):478-491.

50. Bender A, Jox RJ, Grill E, Straube A, Lule D. Persistent vegetative state and minimally conscious state: a systematic review and meta-analysis of diagnostic procedures. *Deutsches Arzteblatt International.* 2015;112(14):235-242.

51. De Salvo S, Bramanti P, Marino S. Clinical differentiation and outcome evaluation in vegetative and minimally conscious state patients: The neurophysiological approach. *Functional neurology.* 2012;28(3):155-162.

52. Luaute J, Morlet D, Mattout J. BCI in patients with disorders of consciousness: clinical perspectives. *Ann. Phys. Rehabil. Med.* 2015;58(1):29-34.

53. Gawryluk JR, D'Arcy RC, Connolly JF, Weaver DF. Improving the clinical assessment of consciousness with advances in electrophysiological and neuroimaging techniques. *BMC Neurol.* 2010;10:11.

54. Rosenbaum AM, Giacino JT. Clinical management of the minimally conscious state. *Handb. Clin. Neurol.* 2015;127:395-410.

55. Kable AK, Pich J, Maslin-Prothero SE. A structured approach to documenting a search strategy for publication: a 12 step guideline for authors. *Nurse Educ. Today.* 2012;32(8):878-886.

56. Moher D, Shamseer L, Clarke M, et al. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. *Systematic reviews.* 2015;4:1.

57. Deeks JJ WS, Davenport C. . *Chapter 4: Guide to the contents of a Cochrane Diagnostic Test Accuracy Protocol.* Available from: http://srdta.cochrane.org/. The Cochrane Collaboration; 2013.

58. Liberati A, Altman DG, Tetzlaff J, et al. The PRISMA statement for reporting systematic reviews and meta-analyses of studies that evaluate health care interventions: explanation and elaboration. *PLoS Med.* 2009;6(7):e1000100.

59. Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann. Intern. Med.* 2009;151(4):264-269, W264.

60. Shamseer L, Moher D, Clarke M, et al. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015: elaboration and explanation. *BMJ.* 2015;349:g7647.

61. Whiting PF, Rutjes AW, Westwood ME, et al. QUADAS-2: a revised tool for the quality assessment of diagnostic accuracy studies. *Ann. Intern. Med.* 2011;155(8):529-536.

62. Gibson RM, Fernandez-Espejo D, Gonzalez-Lara LE, et al. Multiple tasks and neuroimaging modalities increase the likelihood of detecting covert awareness in patients with disorders of consciousness. *Frontiers in Human Neuroscience.* 2014;8:950.

63. Horki P, Bauernfeind G, Klobassa DS, et al. Detection of mental imagery and attempted movements in patients with disorders of consciousness using EEG. 2014;8(DEC).

64. Bekinschtein TA, Coleman MR, Niklison IJ, Pickard JD, Manes FF. Can electromyography objectively detect voluntary movement in disorders of consciousness? *Journal of Neurology, Neurosurgery and Psychiatry.* 2008;79(7):826-828.

65. Cruse D, Chennu S, Chatelle C, et al. Relationship between etiology and covert cognition in the minimally conscious state. *Neurology.* 2012;78(11):816-822.

66. Risetti M, Formisano R, Toppi J, et al. On ERPs detection in disorders of consciousness rehabilitation. *Front Hum Neurosci.* 2013;7:775.

67. Faugeras F, Rohaut B, Weiss N, et al. Event related potentials elicited by violations of auditory regularities in patients with impaired consciousness. *Neuropsychologia.* 2012;50(3):403-418.

68. Rohaut B, Faugeras F, Chausson N, et al. Probing ERP correlates of verbal semantic processing in patients with impaired consciousness. *Neuropsychologia.* 2015;66:279-292.

69. Sitt JD, King J-R, El Karoui I, et al. Large scale screening of neural signatures of consciousness in patients in a vegetative or minimally conscious state. *Brain: A Journal of Neurology.* 2014;137(8):2258-2270.

70. Habbal D, Gosseries O, Noirhomme Q, et al. Volitional electromyographic responses in disorders of consciousness. *Brain injury.* 2014;28 (5-6):533.

71. Lotte F, Congedo M, Lecuyer A, Lamarche F, Arnaldi B. A review of classification algorithms for EEG-based brain-computer interfaces. *J Neural Eng.* 2007;4(2):R1-R13.

72. King J, Faugeras F, Gramfort A, et al. Single-trial decoding of auditory novelty responses facilitates the detection of residual consciousness. *NeuroImage.* 2013;83:726-738.

73. Lechinger J, Chwala-Schlegel N, Fellinger R, et al. Mirroring of a simple motor behavior in Disorders of Consciousness. *Clinical Neurophysiology.* 2013;124(1):27-34.

74. Pan J, Xie Q, He Y, et al. Detecting awareness in patients with disorders of consciousness using a hybrid brain-computer interface. *Journal of Neural Engineering.* 2014;11(5).

75. Li Y, Pan J, He Y, et al. Detecting number processing and mental calculation in patients with disorders of consciousness using a hybrid brain-computer interface system. *BMC Neurol.* 2015;15:259.

76. Cruse D, Chennu S, Chatelle C, et al. Bedside detection of awareness in the vegetative state: A cohort study. *The Lancet.* 2011;378(9809):2088-2094.

77. Fellinger R, Klimesch W, Schnakers C, et al. Cognitive processes in disorders of consciousness as revealed by EEG time-frequency analyses. *Clinical Neurophysiology.* 2011;122(11):2177-2184.

78. Hauger SL, Schnakers C, Andersson S, et al. Neurophysiological indicators of residual cognitive capacity in the minimally conscious state. *Behavioural Neurology.* 2015;2015:145913.

79. Lule D, Noirhomme Q, Kleih SC, et al. Probing command following in patients with disorders of consciousness using a brain-computer interface. *Clinical Neurophysiology.* 2013;124(1):101-106.

80. Chennu S, Finoia P, Kamau E, et al. Dissociable endogenous and exogenous attention in disorders of consciousness. *NeuroImage Clinical.* 2013;3:450-461.

81. Real RG, Veser S, Erlbeck H, et al. Information processing in patients in vegetative and minimally conscious states. *Clinical neurophysiology : official journal of the International Federation of Clinical Neurophysiology.* 2016;127(2):1395-1402.

82. Pokorny C, Klobassa DS, Pichler G, et al. The auditory P300-based single-switch brain-computer interface: paradigm transition from healthy subjects to minimally conscious patients. *Artificial intelligence in medicine.* 2013;59(2):81-90.

83. Bekinschtein TA, Dehaene S, Rohaut B, Tadel F, Cohen L, Naccache L. Neural signature of the conscious processing of auditory regularities. *PNAS Proceedings of* *the National Academy of Sciences of the United States of America.* 2009;106(5):1672-1677.

84. Faugeras F, Rohaut B, Weiss N, et al. Probing consciousness with event-related potentials in the vegetative state. *Neurology.* 2011;77(3):264-268.

85. Schnakers C, Giacino JT, Lovstad M, et al. Preserved covert cognition in noncommunicative patients with severe brain injury? *Neurorehabilitation and neural repair.* 2015;29(4):308-317.

86. Laureys S, Perrin F, Bredart S. Self-consciousness in non-communicative patients. *Consciousness and cognition.* 2007;16(3):722-741; discussion 742-725.

87. Fischer C, Dailler F, Morlet D. Novelty P3 elicited by the subject's own name in comatose patients. *Clinical neurophysiology : official journal of the International Federation of Clinical Neurophysiology.* 2008;119(10):2224-2230.

88. Fischer C, Luaute J, Morlet D. Event-related potentials (MMN and novelty P3) in permanent vegetative or minimally conscious states. *Clinical Neurophysiology.* 2010;121(7):1032-1042.

89. Perrin F, Schnakers C, Schabus M, et al. Brain response to one's own name in vegetative state, minimally conscious state, and locked-in syndrome. *Archives of neurology.* 2006;63(4):562-569.

90. Cavinato M, Volpato C, Silvoni S, Sacchetto M, Merico A, Piccione F. Event-related brain potential modulation in patients with severe brain damage. *Clinical Neurophysiology.* 2011;122(4):719-724.

91. Giacino JT, Schnakers C, Rodriguez-Moreno D, Kalmar K, Schiff N, Hirsch J. Behavioral assessment in patients with disorders of consciousness: gold standard or fool's gold? *Progress in brain research.* 2009;177:33-48.

92. Majerus S, Bruno MA, Schnakers C, Giacino JT, Laureys S. The problem of aphasia in the assessment of consciousness in brain-damaged patients. *Progress in brain research.* 2009;177:49-61.

93. Boly M, Coleman MR, Davis MH, et al. When thoughts become action: an fMRI paradigm to study volitional brain activity in non-communicative brain injured patients. *Neuroimage.* 2007;36(3):979-992.

94. Weijer C, Peterson A, Webster F, et al. Ethics of neuroimaging after serious brain injury. *BMC medical ethics.* 2014;15:41.

95. Goldfine AM, Bardin JC, Noirhomme Q, Fins JJ, Schiff ND, Victor JD. Reanalysis of "Bedside detection of awareness in the vegetative state: a cohort study". *Lancet (London, England).* 2013;381(9863):289-291.

96. Cruse D, Chennu S, Chatelle C, et al. Reanalysis of "Bedside detection of awareness in the vegetative state: A cohort study": Authors' reply. *The Lancet.* 2013;381(9863):291-292.

97. Tzovara A, Simonin A, Oddo M, Rossetti AO, De Lucia M. Neural detection of complex sound sequences in the absence of consciousness. *Brain: A Journal of Neurology.* 2015;138(5):1160-1166.

98. Naccache L, King JR, Sitt J, et al. Neural detection of complex sound sequences or of statistical regularities in the absence of consciousness? *Brain : a journal of neurology.* 2015;138(Pt 12):e395.

99. Tzovara A, Simonin A, Oddo M, Rossetti AO, De Lucia M. Reply: Neural detection of complex sound sequences or of statistical regularities in the absence of consciousness? *Brain : a journal of neurology.* 2015;138(Pt 12):e396.

LIST OF SUPPLEMENTAL DIGITAL CONTENT

Supplemental Digital Content 1: Full review protocol, PDF.

Supplemental Digital Content 2: PRISMA checklist, PDF.

Supplemental Digital Content 3: Full search strategy Medline, PDF.

Supplemental Digital Content 4: QUADAS-2 worksheet with signaling questions, PDF.