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Neural underpinnings of visual awareness investigated with transcranial magnetic stimulation

Neuronalne podstawy świadomości wzrokowej badane za pomocą
przezczaszkowej stymulacji magnetycznej

PhD thesis

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Abbreviations

aMPFC, anterior medial prefrontal cortex;
aPFC, anterior prefrontal cortex;
cTBS, continuous theta burst stimulation;
dlPFC, dorsolateral prefrontal cortex;
EMG, electromyography;
iTBS, intermittent theta burst stimulation;
M1, primary motor cortex;
MEP(s), motor-evoked potential(s);
PAS, Perceptual Awareness Scale;
PFC, prefrontal cortex;
PMd, dorsal premotor cortex;
RMT, resting motor threshold;
RT(s), response time(s);
rTMS, repetitive transcranial magnetic stimulation;
spTMS, single-pulse transcranial magnetic stimulation;
TBS, theta burst stimulation;
TMS, transcranial magnetic stimulation.

1. Introduction

Understanding the neural processes involved in shaping awareness of visual stimuli is one of the major challenges in consciousness science. Despite significant efforts by consciousness researchers, the neural underpinnings of visual awareness (subjective experience of visual information) remain unclear. The scientific literature on consciousness presents divergent perspectives regarding the brain areas that correlate with visual awareness. One perspective posits the necessity of PFC for becoming aware of the content of visual perception, while another suggests the sufficiency of occipital and parietal brain areas (for a review, see Michel & Morales, 2020). This disagreement has become one of the most discussed issues in current consciousness research (Boly et al., 2017; Odegaard et al., 2017). Although there is empirical evidence which connects visual awareness to late, widespread frontoparietal activity, the notion has faced criticism due to studies associating it with early activation of the sensory cortex and/or associative areas (Koch et al., 2016). At the same time, PFC is typically linked with metacognition (Fleming & Dolan, 2012), which refers to the cognitive processes that oversee and evaluate other cognitive processes, along with the outcomes of this evaluation, which encompasses metacognitive regulation (Paulewicz, Siedlecka, & Koculak, 2020). While visual metacognition and visual awareness are considered to be closely related (Rahnev et al., 2022), the models of visual awareness formation typically emphasise the influence of visual stimuli while overlooking the potential impact of the later stages of information processing (Seth & Bayne, 2022). This latter influence could be considered twofold: later stages can be consequences, not the correlates of consciousness (e.g., neural correlates of responses), but additional information added at the later stages of processing may also enrich awareness (due to the accumulation of non-perceptual evidence). Action-related information can serve as an example. The motor system is closely intertwined with the visual system, and previous studies suggest bidirectional influences between these systems. Numerous studies have examined the action-perception loop, demonstrating the mutual influence between perceptual information and action in tasks, leading to enhanced task performance (Donnarumma et al., 2017; Hecht et al., 2001). Additionally, other sources of evidence, such as prior expectations (Snyder et al., 2015), attentional engagement (Fazekas & Overgaard, 2018), or previous behavioural responses (Rahnev et al., 2015), can influence reported awareness. Thus, it is hypothesised that action planning or execution may affect reported awareness, for example, through performance monitoring, integrating additional evidence, triggering attentional mechanisms, or restricting the number of response alternatives (Anzulewicz et al., 2019). This is because the formation of awareness may be considered in terms of evidence accumulation (Del Cul et al., 2009). Although the process of accumulating evidence is closely tied to perceptual stimulation, it is assumed to persist even after the stimulus vanishes, extending beyond stimulus-related behaviours and influencing judgements of visual awareness (Anzulewicz et al., 2019). Considerable efforts have been dedicated to investigating the neuroimaging correlates of consciousness and disentangling them from post-perceptual processes that are not related to awareness (for a review, see Koch et al., 2016).

Correlational neuroimaging evidence is insufficient to definitively establish whether the PFC is causally involved in visual awareness. Neuroscientists often encounter challenges when

drawing causal inferences based on neuroimaging methods which typically limit them to correlational inferences. Non-invasive brain stimulation methods, such as TMS, have been applied to address this limitation. TMS is based on electromagnetic induction. It enables the induction of an electric field that may influence the activity of targeted brain tissue. Originally, it was assumed that TMS would make it possible to establish the causal relations between the activity of specific brain areas, cognitive functions, and behaviours. However, with the development of neuroimaging techniques, it has become apparent that the brain cannot simply be parcelled into regions responsible for specific cognitive functions. The impact of brain lesions and non-invasive brain stimulation can extend beyond individual areas to affect brain networks (Beynel et al., 2020; Bolognini & Ro, 2010; Ruff et al., 2009; Ziemann, 2010). Currently, TMS is one of the methods used to investigate the neural underpinnings of awareness and was used for the same purpose in this thesis.

TMS applications can be categorised into online TMS and offline TMS procedures. Online TMS refers to the use of TMS while a participant is performing a task, whereas offline TMS refers to using TMS prior to a task. The physiological effects of spTMS (a single pulse) can be detected for over a dozen seconds. Alternatively, rTMS paradigms involve applying pulses with a specific frequency pattern during or before an experimental task, aiming to induce neuroplasticity-like effects. These rTMS effects can persist for dozens of minutes following a protocol application and can be assessed using behavioural procedures or neuroimaging techniques. Online spTMS protocols can be employed to investigate certain causal claims, for example, by perturbing brain activity and analysing the topographic pattern of TMS-induced changes using effective connectivity measures (Hallett et al., 2017). It is possible to evaluate the influence of the stimulated area on interconnected regions and examine the distinct functional contributions of multiple cortical areas within a network. However, the use of this type of TMS is limited to specific experimental designs. In comparison to online spTMS, the rTMS approach has limitations in revealing the causal relations between brain areas and cognitive functions. Online rTMS cannot be combined with neuroimaging techniques to record concurrent brain activity, while the effects of offline rTMS depend on neuroplasticity-like effects that may occur at various time points from the start of rTMS protocol (for a review of TMS protocols, see Bergmann et al., 2016). In the presented studies, a patterned type of offline rTMS protocol was used in the first study and an online spTMS protocol in the second.

The use of rTMS to study visual perception is not novel. Several research groups have used rTMS to manipulate awareness-related processes by stimulating the PFC in healthy participants (Bor et al., 2017; Rahnev et al., 2016; Rounis et al., 2010). One study indicated a decrease in reported visual awareness for correct responses following active cTBS to the bilateral dlPFC compared to sham (inactive) cTBS (Rounis et al., 2010). However, the replication attempt failed to reproduce this effect (Bor et al., 2017). Another study reported an increase in metacognitive ability after cTBS to the right aPFC and the right dlPFC, without observing differences in behavioural decision confidence ratings (Rahnev et al., 2016). Since awareness is often reported using subjective scales that require judgements, the PFC may be associated with metacognition but may not be essential for visual awareness of the stimulus.

PFC activity may be related to the judgements of the stimulus experience rather than the formation of the experience of the stimulus. However, awareness may be influenced not only by the early stages of visual processing but also by subsequent stages and potentially non-perceptual information. The precise involvement of the PFC in perceptual and post-perceptual processing remains an open issue. Post-perceptual processing stages and non-perceptual evidence likely play a significant role in awareness reports.

One example of such non-perceptual evidence is a behavioural response to a visual stimulus. Previous research has suggested that the behavioural response in identification tasks may play a role in visual awareness and confidence reports (Siedlecka et al., 2019; Siedlecka et al., 2020; Siedlecka, Koculak & Paulewicz, 2021). However, it is unclear whether the decision-making process or the actual movement is critical. In this vein, TMS has been used to study motor-related influences on metacognition. In an spTMS study conducted by Fleming et al. (2015), spTMS was employed before or after a two-alternative forced-choice task, followed by a confidence rating on the decision made. The researchers found that applying TMS to the PMd associated with the chosen response resulted in higher confidence in that response and greater metacognitive efficiency, i.e., the ability to accurately judge the sensitivity of one's own perception (Fleming & Lau, 2014), compared to TMS applied to the PMd associated with the unchosen response. However, there was no evidence of TMS influencing accuracy in identifying the visual stimuli. The effect of TMS on average confidence ratings was observed irrespective of whether it was applied before or after the identification response, suggesting the involvement of post-decision processes (regarding one's identification decision) in the participants' confidence. However, the application of TMS to M1 did not yield any of these effects. The researchers proposed that PMd activity, but not M1 activity, contributes to confidence ratings. No such TMS study has been done regarding the investigation of visual awareness prior to the one reported in this thesis.

To sum up, TMS is frequently used to investigate how short-term changes in the excitability of a stimulated brain area impact cognitive functions. However, this approach is sometimes associated with implicit assumptions that TMS can selectively influence the area of interest, thus helping to determine its role. Various publications have presented TMS-based conclusions without describing alternative explanations or distinguishing between direct and indirect causal effects of a TMS-induced change in activity within a specific area on a certain behaviour or brain process. In the first article of this thesis, I explore the different types of inferences that can be drawn from TMS studies, providing an overview of the method used in the studies conducted and how they can be interpreted. This theoretical analysis supported the way I interpreted the TMS data of previous studies and the TMS studies presented in this thesis. Due to the lack of definitive findings regarding the involvement of the PFC in visual awareness, the first study (second article) examines the effects of temporarily influencing PFC activity on both visual perception and metacognitive efficiency. Furthermore, considering previous behavioural findings that suggest a link between the behavioural response and visual awareness, along with the limited evidence from previous TMS studies on the impact of M1 stimulation on

metacognition, the second study (third article) included in this thesis focuses on stimulating M1 using TMS to investigate the role of motor-related information in visual awareness ratings.

2. Aims

The purpose of the studies was to use TMS to investigate whether changes in PFC and M1 activity impact visual awareness reports. The studies aimed to expand our understanding of the neural underpinnings of visual awareness. These studies also aimed to support theories of consciousness that connect awareness with metacognition, decision-making, and cognitive control, thereby contributing to the development of both research and theoretical understanding of consciousness. Additionally, the results of these studies may enhance researchers' comprehension of metacognitive deficits in neurological and mental disorders related to consciousness, perception, and metacognition. Therefore, in theory, their findings also have the potential to advance the development of therapeutic methods.

Overall, the main objective of the theoretical article was to critically evaluate the utility of TMS in scientific research and provide guidance for researchers aiming to make causal inferences based on TMS. That is, to analyse and organise TMS-based inferences at a theoretical level while also highlighting the methodological challenges that may weaken causal inferences in studies using rTMS. The specific goals of the article were twofold. Firstly, to examine methodological challenges related to TMS that may compromise the ability to establish potential causal relations between brain regions and cognitive functions. Secondly, to explore the added value of incorporating neuroimaging techniques into rTMS-based inferences and to propose recommendations for selecting suitable control conditions in TMS studies.

The first study examined the effects of TBS protocols applied to the PFC on metacognitive efficiency and visual perception, specifically exploring whether these effects were specific to the measures of identification task performance and awareness rating. The study had two primary aims. Firstly, to test whether cTBS to the left aMPFC can influence visual awareness. Secondly, to investigate (to the possible degree) whether the aMPFC activity plays a role in visual stimulus experience and/or metacognitive efficiency. Three different TBS protocols were used. The first hypothesis was that cTBS to the left aMPFC would influence visual awareness ratings (H1). The second hypothesis was that cTBS to the left aMPFC would influence metacognitive efficiency estimates (H2). Finally, the third hypothesis was that two distinct offline TBS protocols (cTBS and iTBS) would have opposite effects on visual awareness (H3).

The second study investigated the influence of TMS-induced motor information on awareness ratings. The study had two specific aims. Firstly, to determine whether this information influences visual awareness ratings. Secondly, to investigate whether the level of TMS-induced MEP amplitude (measured with EMG) allows for quantifying the level of perceptual evidence accumulated. The study hypothesised that applying spTMS to M1 would result in higher reported visual awareness compared to the control condition (H1). Additionally, the study hypothesised that there would be no difference in metacognitive efficiency between M1 and

the control TMS condition (H2) and that MEPs induced by TMS indirectly reflect the amount of accumulated evidence for stimulus perception (H3).

3. Methods

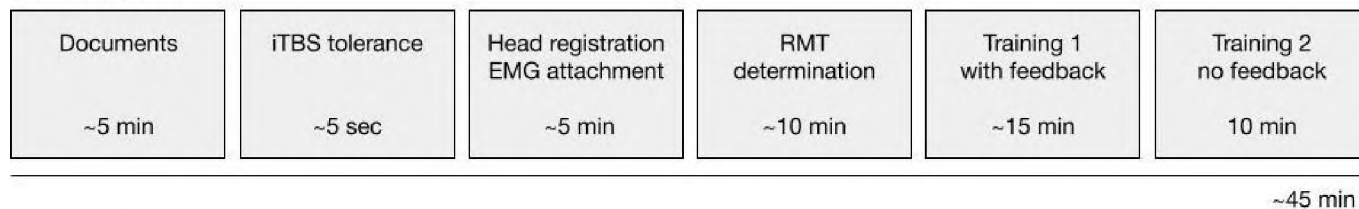
To conduct the studies, I programmed and used TMS protocols and computer procedures specific to each study. The computer tasks involved both *objective* (identification task) and *subjective* (adapted PAS) measures of visual perception. A pilot test was conducted before each study to test and improve data quality. Both studies had within-group designs, included healthy volunteers, and adhered to relevant ethical and methodological standards. The studies recruited volunteers who met the criteria for participation, which included having no history of neurological disorders, psychiatric disorders, head injury, and having normal or corrected to normal vision, among many other criteria. Volunteers were aged between 18 and 40 years. Before participating, they were informed about the study procedures, risks, and benefits through written information brochures. Prior to TMS sessions, the participants completed a screening questionnaire about their health and safety concerning TMS and they provided written informed consent. The first study involved 21 participants, whereas the second study involved 46 participants. Participants received financial compensation for their participation.

During both studies, participants viewed a simple visual stimulus (a Gabor patch) embedded in white noise for a brief duration (near visual threshold presentation). The training and testing procedures included multiple trials comprising of the identification task and the adapted PAS (Ramsøy & Overgaard, 2004). In the identification task, participants had to decide whether the Gabor patch was oriented towards the left or right and answer by pressing a key on a keyboard. They indicated their visual awareness by providing a key press or a verbal report, which was then recorded. In both studies, the order of TMS protocols was counterbalanced. In both studies, participants' visual sensitivity was measured by identification task performance (stimulus contrast required to achieve a particular level of accuracy in the first study and accuracy in the second study), while visual awareness was measured by the adapted PAS. Additionally, RTs to both measures were collected. The association between identification task performance and awareness ratings was also investigated to estimate metacognitive efficiency. Specifically, the M-ratio was used to examine the relation between reported visual awareness and identification task performance, which is assumed to reflect the effectiveness of internal information processing (Fleming & Lau, 2014). The M-ratio (meta- d'/d') was calculated following the codes of Maniscalco and Lau (2014) and Fleming (2017). In the first study, a logistic regression model was used as the primary method to analyse metacognitive efficiency. In both studies, the data was analysed using mixed-effects regression models fitted in the R statistical environment (R Core Team, 2019). All parameters were compared between the TMS conditions. The data and scripts for analyses are provided on the Open Science Framework.

The first study was conducted at Aarhus University, using three TBS protocols applied to the left PFC, guided by individual magnetic resonance images. Specifically, the left aMPFC was stimulated, based on its proposed involvement in metacognitive efficiency (McCurdy et al., 2013; Molenberghs et al., 2016). The TBS protocols comprised bursts of high-frequency

stimulation repeated at regular intervals continuously or with breaks for less than 4 minutes (Huang et al., 2005). Two active TBS protocols preceded the computer task and aimed to induce short-term neuroplasticity-like effects, while the third served as a control protocol. The experimental flow of the first study is illustrated in Figure 1.

Procedure | Day 1



Procedure | Day 2-4

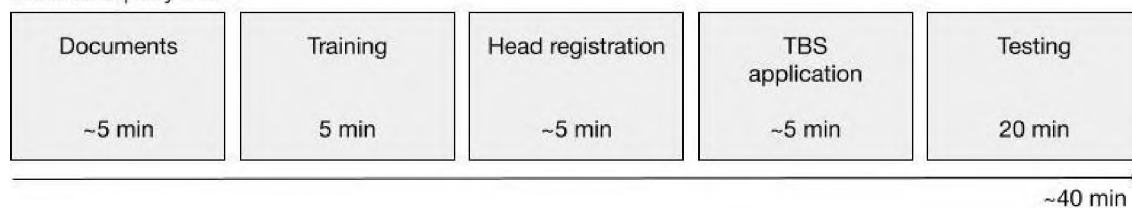


Fig. 1 – Schematic illustration of the first study’s procedure. The study consisted of four sessions conducted over four days, with each session separated by a minimum of four days to reduce the impact of task learning. Participants performed the sessions at the same time of the day to minimise any time-of-day effects. On the first day, participants received a few burst pulses to their aPFC to provide them with sufficient information to make an informed decision about their continued participation in the study. After that, EMG electrodes were attached, and the RMT was determined. Participants then received behavioural training with the computer task to become familiar with the experimental procedure. This training enabled the establishment of stimulus contrasts that were used as initial contrasts in the staircases during the testing phase. On each of the following days, participants completed a 5-minute training task and then performed the experimental task shortly after receiving one of three different TBS protocols. The testing time for TBS effects was 20 minutes and was divided into four blocks per condition, separated by 20-s breaks.

The second study was conducted at Jagiellonian University Hospital. The study involved employing spTMS guided by neuronavigation using the average brain template. The study involved administering TMS in combination with a computer procedure under two conditions: left M1 stimulation and a control condition (interhemispheric cleft stimulation). The experimental flow of the second study is illustrated in Figure 2.

Procedure

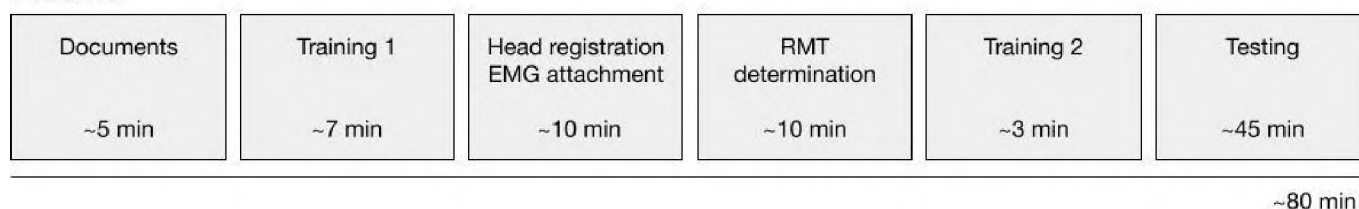


Fig. 2 – Schematic illustration of the second study’s procedure, which comprised a single session. At the start of the session, participants completed the documentation and received training in the computer task. During this phase, stimulus contrasts were established based on the task performance to be used in the subsequent procedures. Following this, EMG electrodes were attached, and RMT was determined. Participants then underwent a short training period before completing the experimental task, during which they received spTMS to the left M1 and a control area. The testing phase lasted approximately 45 minutes.

4. Articles with co-authorship declarations

4.1. Article 1 with co-authorship declarations

Hobot, J., Klincewicz, M., Sandberg, K. & Wierzchoń, M. (2021). **Causal inferences in repetitive transcranial magnetic stimulation research: Challenges and perspectives.** *Frontiers in Human Neuroscience*, 14, 574.



Causal Inferences in Repetitive Transcranial Magnetic Stimulation Research: Challenges and Perspectives

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Transcranial magnetic stimulation (TMS) is used to make inferences about relationships between brain areas and their functions because, in contrast to neuroimaging tools, it modulates neuronal activity. The central aim of this article is to critically evaluate to what extent it is possible to draw causal inferences from repetitive TMS (rTMS) data. To that end, we describe the logical limitations of inferences based on rTMS experiments. The presented analysis suggests that rTMS alone does not provide the sort of premises that are sufficient to warrant strong inferences about the direct causal properties of targeted brain structures. Overcoming these limitations demands a close look at the designs of rTMS studies, especially the methodological and theoretical conditions which are necessary for the functional decomposition of the relations between brain areas and cognitive functions. The main points of this article are that TMS-based inferences are limited in that stimulation-related causal effects are not equivalent to structure-related causal effects due to TMS side effects, the electric field distribution, and the sensitivity of neuroimaging and behavioral methods in detecting structure-related effects and disentangling them from confounds. Moreover, the postulated causal effects can be based on indirect (network) effects. A few suggestions on how to manage some of these limitations are presented. We discuss the benefits of combining rTMS with neuroimaging in experimental reasoning and we address the restrictions and requirements of rTMS control conditions. The use of neuroimaging and control conditions allows stronger inferences to be gained, but the strength of the inferences that can be drawn depends on the individual experiment's designs. Moreover, in some cases, TMS might not be an appropriate method of answering causality-related questions or the hypotheses have to account for the limitations of this technique. We hope this summary and formalization of the

Abbreviations: A_{1C} , a change in A_1 activity is present; BOLD, blood oxygen level-dependent; H, P_X takes place in A_1 ; P_X , process underlying cognitive function X; P_Y , process underlying cognitive function Y; I_1 , inference 1; I_2 , inference 2; I_3 , inference 3; I_4 , inference 4; I_5 , inference 5; rTMS, repetitive Transcranial magnetic stimulation (TMS); S_0 , a sham rTMS protocol; rTMS₁, an active rTMS protocol 1; S_{1A} , rTMS₁ is applied to A_1 ; rTMS₂, an active rTMS protocol 2; TMS, transcranial magnetic stimulation; T_X , task X; T_{XD} , an observed difference in T_X performance; T_Y , task Y.

reasoning behind rTMS research can be of use not only for scientists and clinicians who intend to interpret rTMS results causally but also for philosophers interested in causal inferences based on brain stimulation research.

Keywords: causal inferences, brain plasticity, brain excitability, repetitive TMS, TMS-neuroimaging

INTRODUCTION

A fundamental issue in human neuroscience is how to make causal inferences based on research data. Traditional use of neuroimaging methods limits experimental conclusions to correlational inferences (though, the methods of effective connectivity are used to postulate causal inferences; see Valdes-Sosa et al., 2011). Following their introduction, brain stimulation methods, especially TMS, started to be considered as a remedy for this limitation. TMS was developed over thirty years ago and is based on electromagnetic induction (Barker et al., 1985). A TMS coil induces an electric field which might influence the activity of brain tissue. It was originally thought that TMS would make it possible to conclude the causal relations between brain activity, cognitive functions, and behaviors. However, it has since become clearer that the brain cannot simply be parceled into regions responsible for certain functions, and the impact of brain lesions and non-invasive brain stimulation is not necessarily limited to a single area but extends to networks. Currently, TMS is often used to test hypotheses about how short-term changes in the excitability of a stimulated brain area affect cognitive functions. In online TMS paradigms, electromagnetic pulses are applied concurrently with the experimental measurement. The physiological consequences of a single electromagnetic pulse can be detected for over a dozen seconds (Furubayashi et al., 2013). In repetitive (rTMS) paradigms, pulses with a particular frequency pattern are applied during or before experimental measurement because they often lead to neuroplasticity-like changes (Chung et al., 2015). The neuromodulatory rTMS effect can be assessed with standard experimental procedures or neuroimaging techniques (for a review of combined TMS-EEG studies, see Thut and Pascual-Leone, 2010); it can be observed even for up to 45 min after a single protocol application (Huang et al., 2005), or it can last for months after multiple protocol applications over repeated TMS sessions in longitudinal studies (Speer et al., 2000, 2009; Li et al., 2004; Choi et al., 2014, 2019; Kang et al., 2016). Thus, TMS is often considered to be an extension of neuroimaging, which (due to its influence on brain activity) allows causal relations to be tested.

TMS is frequently used to decompose the functional organization of the brain. Multiple scientific articles contain statements that TMS can be used to draw both causal brain-behavior inferences (Sack, 2006; Śliwińska et al., 2014) and causal relationships between brain structure and function (Schutter et al., 2004; Bolognini and Ro, 2010; Hartwigsen, 2015; Veniero et al., 2019). In research practice, this often leads to implicit assumptions that TMS can selectively influence the area of interest, therefore its role can be established. Consequently, multiple studies have presented rTMS-based

conclusions on the causal role of certain brain areas in certain cognitive functions (e.g., Carmel et al., 2010; Philiastides et al., 2011; Zanto et al., 2011; Bourgeois et al., 2013; Izuma et al., 2015; Schaal et al., 2015; Siuda-Krzywicka et al., 2016; Montefinese et al., 2017), often without describing alternative explanations or making a distinction between direct and indirect causal effects of an rTMS-induced change in activity in a certain area on a certain behavior or brain process.

Employing chronometry (tracking the time course of functional relevance), online single-pulse, double-pulse, or short-burst TMS protocols (including double-coil approaches) allow investigation of the causal relations between the activity of certain brain areas and behaviors or cognitive functions especially when effective connectivity measures are also employed (e.g., de Graaf et al., 2009). These protocol types might be used to influence cognitive functions or perturb brain activity to track the signal propagation and analyze the topographic pattern of TMS-evoked changes in brain activity. This allows researchers to: (1) identify the brain areas involved in certain behavior; (2) assess the impact of the stimulated brain area upon interconnected areas *via* direct connections or intermediate areas, including inter-hemispheric interactions (Blankenburg et al., 2008); (3) reveal bottom-up and top-down influences between brain areas; and (4) dissect the specific functional contributions of different cortical areas of an investigated network. Crucially, the propagation of TMS-evoked activity can depend on the degree of wakefulness (Massimini et al., 2005), which in some studies may act as a confound but in others may allow the state-dependence of interactions among remote and interconnected brain regions to be investigated. However, this use of TMS is limited to specific experimental designs, and some TMS effects (as in the case of all active TMS protocols) may be side effects of the stimulation procedure (Holmes and Meteyard, 2018; for a review, see Bestmann et al., 2008a).

The rTMS approach is more limited than single-pulse, double-pulse, and burst-pulse TMS in terms of helping to understand the causal relationships between brain areas and cognitive functions (however, in certain designs rTMS can be used for chronometry, see Rossi et al., 2011). Online rTMS does not allow concurrent brain activity registration using neuroimaging techniques, while offline rTMS effects depend on neuroplasticity-like changes which might occur at various time points after the start or the end of rTMS. Thus, rTMS does not allow tracking of the direct influence of perturbation to determine the time point at which an area makes a critical contribution to a given behavior or to investigate effective connectivity between brain areas. Although most non-invasive stimulation methods share the same limitations

as rTMS, for purposes of clarity we narrow the scope of the discussion below to rTMS. Most of the issues, that are mentioned below, related to the pitfalls of TMS have already been selectively discussed (e.g., Siebner and Rothwell, 2003; Robertson et al., 2003; Thickbroom, 2007; Bestmann et al., 2008a; Siebner et al., 2009). The current article aims to combine, organize, and analyze these insights at the theoretical level and indicate their possible consequences for inferences based on rTMS evidence. Below, we first analyze several known methodological issues that can invalidate inferences about direct causal relations between brain areas, brain processes, and cognitive functions investigated with TMS. Second, we discuss the special role that neuroimaging plays in rTMS-based inferences and approaches to creating TMS control conditions.

INFERENCES BASED ON CONDITIONAL STATEMENTS

Causal inference, and specifically inference based on interventions in the operation of a complex system such as the brain, fall within the theoretical framework of the general theory of causality that was developed by Pearl (2000). We use a small part of Pearl's Structural Causal Model. This is because unlike causal frameworks such as Bradford Hill's criteria (Hill, 1965), Pearl's framework is resistant to counterexamples and makes sense of probabilistic causal inferences about specific mechanisms that are parts of complex systems. In this view, to characterize a relationship between event A and event B as causal is to say that a selective intervention on A might lead to a change in the distribution of B. We assume a causal influence of one event on another is direct if none of the variables included in a given causal model mediates this effect; otherwise, it is indirect. In a setting such as a TMS experiment, where intervention is randomized, we compare the intervention-related distribution of variables with a control distribution and expect to find suitable neuronal candidates that cause the response. For clarity purposes, we address TMS-related inferences with the use of conditional logic.

To consider a simple type of TMS-based inference, assume that a researcher is interested in cognitive function X. To investigate the process (P_X) that underlies this function, the researcher aims to determine whether brain area 1 (A_1), which is typically associated with P_X , is engaged during a task that is assumed to engage cognitive function X (T_X). For example, one may investigate the involvement of the dorsolateral prefrontal cortex in decision confidence by measuring the effect of rTMS on confidence ratings. In such a case, the hypothesis (H) often states that P_X takes place in A_1 and is tested with the application of an active rTMS protocol 1 (rTMS₁) to A_1 . We can formally represent this pattern of reasoning in the following way (the logic symbol \wedge represents the logical conjunction, i.e., "and," and the \rightarrow represents implication, i.e., "if <antecedent> then <consequent>"):

H – P_X takes place in A_1
 S_{1A} – rTMS₁ is applied to A_1

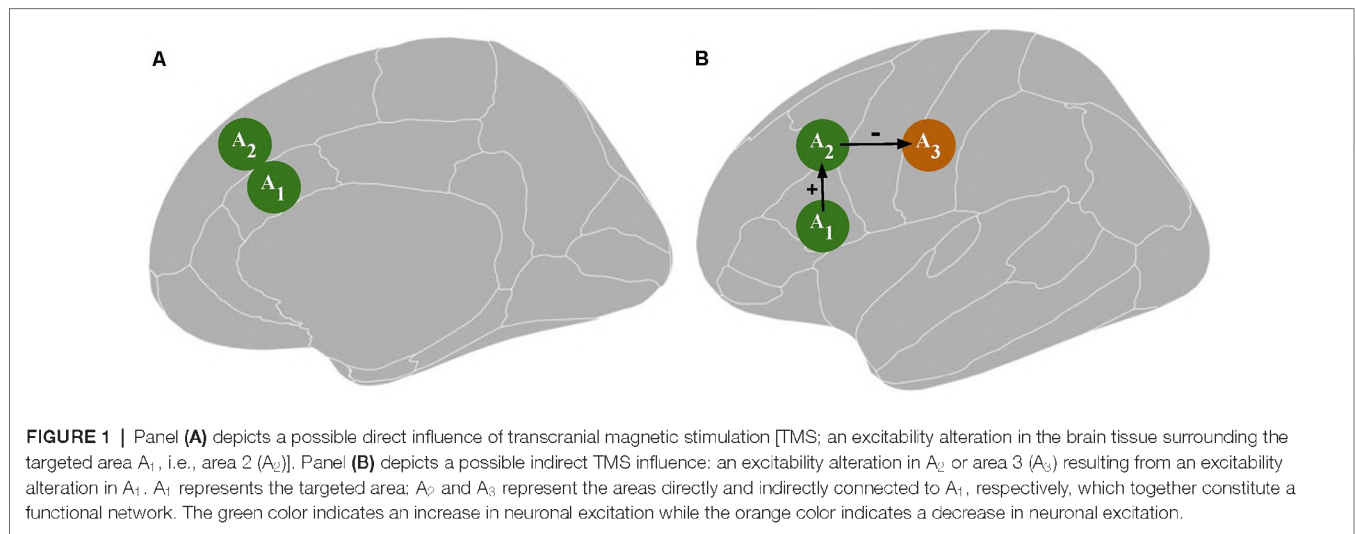
T_{XD} – a difference in T_X performance is observed (as compared to a control condition)

$$I_1(((H \wedge S_{1A}) \rightarrow T_{XD}) \wedge (S_{1A} \wedge T_{XD})) \rightarrow H$$

Inference 1 (I_1) states that the statement that P_X takes place in A_1 is true if the following two premises are true: (1) if P_X takes place in A_1 and rTMS₁ is applied to A_1 then a difference in T_X performance is observed; and (2) rTMS₁ is applied to A_1 and a difference in T_X performance is observed.

I_1 depicts the basic form of reasoning used in rTMS research. However, like any inductive inference, this form of reasoning does not always lead to true conclusions. For example, the occurrence of the difference in T_X performance may be unrelated to rTMS₁, in which case, two independent factors contribute to falsely interpreting the consequent of the condition as true. Thus, causal reasoning based on misuse of I_1 may lead to false conclusions. Possible overconfidence in I_1 -based inferences might also stem from overlooking both how TMS and brains work. First, the assumption that TMS selectively influences a targeted area is not always true. The strength of the induced electric field decreases together with the distance from the coil, so the brain areas above or adjacent to the targeted area are likely to be stimulated more than the intended one (Heller and van Hulsteyn, 1992). Second, applying TMS to one area can indirectly influence multiple brain areas that are structurally connected to it and lead to an alteration of the functional state of the targeted network, as pointed out in several reviews (Ruff et al., 2009; Bolognini and Ro, 2010; Ziemann, 2010; Beynel et al., 2020). In sum, TMS applied to a specific brain region can influence other regions directly (e.g., due to stimulation of an area above or adjacent to the area investigated) or indirectly *via* neural connections (e.g., indirect stimulation of an area that is connected to the investigated area or activity alteration in another area due to excitability alteration in the investigated area). These factors limit the strength of causal conclusions based on I_1 .

Accordingly, rTMS₁ may be responsible for a difference in T_X performance *via* unintended stimulation of an area other than A_1 . For example, assume that A_1 is structurally connected to brain area 2 (A_2). Then, there is a possibility that A_2 activity is influenced: (1) directly by rTMS₁ when A_1 is targeted (**Figure 1A**); or (2) indirectly by rTMS₁ *via* an alteration of A_1 activity. At the same time, A_2 is responsible or more important than A_1 for executing P_X (**Figure 1B**). Unintentional direct stimulation of A_2 may occur in several ways. First, the physical spread of an electrical field may reach areas adjacent to the targeted one. Second, since electrical current follows the path of least resistance, the electric field distribution is highly dependent on cerebrospinal fluid distribution and brain folding, thus the peak of the electric field can occur in gray matter regions located some distance from the electric field's expected peak, which is judged based on the location of the center of the (figure-of-eight) coil. This might result in greater stimulation of area/s other than the targeted one (Bijsterbosch et al., 2012). Third, it is challenging to distinguish



whether the rTMS effect stems from excitability alteration in the targeted area or an area above it that possibly has a distinct specialization. These concerns may be raised especially when deeper structures such as the anterior cingulate cortex (Hayward et al., 2007) or insula (Pollatos et al., 2016) are investigated. The vast majority of TMS studies target superficial structures; however, the rule that the strongest electrical field is generated within the outermost areas applies even if the distances (which might be the consequences of brain folding) are small. Because a large part of the cortex lies within sulci, targeted brain coordinates in numerous TMS studies have to be placed within sulci (Busan et al., 2009; Cappelletti et al., 2009; Salillas et al., 2009). Additionally, stimulation of deeper brain structures is obtained at the expense of inducing wider electrical field spread in the brain (Roth et al., 2007; Deng et al., 2013; Downar et al., 2016). For example, metabolic and physiological effects on the primary motor cortex and the primary somatosensory cortex can be observed after rTMS to premotor areas (Siebner et al., 2003). This may compound the difficulty in distinguishing the contribution of direct vs. indirect rTMS effects. The network effects may produce remote activity alteration in cortical areas via cortico-cortical routes and in subcortical structures via cortico-subcortical projections (Strafella et al., 2003; Lefaucheur et al., 2020). The extent of the network effects depends on rTMS protocol parameters (Bestmann et al., 2003). Additionally, the assumption that a difference in T_X performance is caused by an rTMS₁-induced change in A_1 activity may be misleading due to the occurrence of placebo and sensory side effects (Abler et al., 2005). Moreover, rTMS may influence areas related to general cognitive resources (e.g., regions engaged in attentional or working memory processing) or the observed effect may be specific to the T_X design (e.g., resulting from rTMS₁ influence on brain regions involved in response generation during T_X), which is not related to the influence on the investigated cognitive function. In sum, overconfidence in I_1 has multiple ways to lead researchers to overinterpret their data as evidence that P_X takes place in A_1 .

Since statements that follow I_1 cannot fully support the conclusion that P_X takes place in A_1 , can some other inference be used to show that P_X is not executed in A_1 ? This would provide independent evidence for excluding that region from the area of research interest. This way of reasoning is indeed found in TMS literature: based on the lack of an observed effect, some authors postulate a lack of rTMS influence on investigated cognitive functions (e.g., Ghabra et al., 1999; Poulet et al., 2004; Jung et al., 2010; Bor et al., 2017), which might suggest that an investigated area is not involved in the process underlying the investigated cognitive function. Consider then the inference of the following structure (the logic symbol \neg represents negation, i.e., “not”):

$H - P_X$ takes place in A_1
 $S_{1A} - rTMS_1$ is applied to A_1

T_{XD} —a difference in T_X performance is observed (as compared to a control condition)

$$I_2(((H \wedge S_{1A}) \rightarrow T_{XD}) \wedge (S_{1A} \wedge \neg T_{XD})) \rightarrow \neg H$$

Inference 2 (I_2) states that the statement that P_X is not executed in A_1 is true if the following two premises are also true: (1) a difference in T_X performance is observed if P_X takes place in A_1 and rTMS₁ is applied to A_1 ; (2) rTMS₁ is applied to A_1 and a difference in T_X performance is not observed.

In research practice, rTMS₁ does not always lead to a change in A_1 activity and/or a difference in T_X performance. rTMS₁ may have no factual effect because: (1) the rTMS₁ frequency pattern is inadequate for investigating P_X (e.g., theta burst stimulation is applied but P_X is independent of theta-gamma coupling; De Ridder et al., 2007); (2) rTMS₁ parameters are set too low (e.g., intensity or current direction) to influence P_X (Valero-Cabré et al., 2017); (3) brain-intrinsic factors such as neurochemical and neurophysiological properties of A_1 prevent an alteration in its excitability (e.g., it is impossible to facilitate or inhibit A_1 to

a greater extent than it is before rTMS₁ application; Karabanov et al., 2015); and (4) to influence A₁, rTMS₁ should be applied with greater precision (e.g., based on individual functional brain images; Hannula and Ilmoniemi, 2017). Altogether, this is enough evidence to assume that I₂ is not a stronger form of reasoning than I₁. I₁ and I₂ include a hidden assumption that rTMS₁ leads to an alteration in A₁ activity but not all active rTMS applications have neural effects. To claim that A₁ has changed, the assertion based on the inference presented below has to be true:

S_{1A}—rTMS₁ is applied to A₁

A_{1C}—a change in A₁ activity is present

$I_3((S_{1A} \rightarrow A_{1C}) \wedge S_{1A}) \rightarrow A_{1C}$

I₃ states that the statement that A₁ activity is changed if the following two premises are true: (1) a change in A₁ activity is present if rTMS₁ is applied to A₁; and (2) rTMS₁ is applied to A₁.

The issue of the impact of rTMS₁ on the activity of A₁ might be addressed with the use of neuroimaging.

TMS AND NEUROIMAGING

A way of strengthening TMS-based inferences is to combine TMS with neuroimaging, the advantages of which have already been exhaustively described (e.g., Sack, 2006; Bestmann et al., 2008b; Bergmann et al., 2016). Multiple studies have already successfully employed neuroimaging to determine whether a particular rTMS protocol leads to a change in A₁ activity (e.g., Bestmann et al., 2008c; Ruff et al., 2008; Capotosto et al., 2012). Despite the advantage of neuroimaging methods in allowing detection of a change in A₁ activity, confirmation that the change in A₁ activity accompanies TMS₁ cannot fully confirm H. Importantly, even if the change in A₁ activity can be confirmed with neuroimaging, it does not always lead to a difference in T_X performance (Reithler et al., 2011). TMS₁ may have no observable effect because: (1) TMS₁ could have additional consequences that hinder the original stimulation effect, such as the occurrence of compensatory effects that diminish the TMS-induced alteration in A₁ activity or that fulfill the function of A₁ (Andoh and Martinot, 2008); and (2) T_X may not provide an adequate measure of P_X because T_X or its performance level is not demanding enough to be influenced by TMS₁, or T_X is not sensitive enough to capture the impact of TMS₁. Nevertheless, this does not imply that null TMS results are not meaningful because they are crucial to proving the functional irrelevance of a brain region to performing a particular function (de Graaf and Sack, 2011).

Next, assume that the influence of TMS₁ on A₁ can be effectively measured by neuroimaging methods and T_X, and both a change in A₁ activity and a difference in T_X performance is observed. This leads to stronger reasoning than I₁ (inference 4; I₄):

H – P_X takes place in A₁

S_{1A}—rTMS₁ is applied to A₁

T_{XD}—a difference in T_X performance is observed (as compared to a control condition)

A_{1C}—a change in A₁ activity is present

$I_4(((H \wedge S_{1A}) \rightarrow T_{XD}) \wedge (S_{1A} \wedge T_{XD})) \wedge$
 $((S_{1A} \rightarrow A_{1C}) \wedge S_{1A}) \rightarrow T_{XD}) \rightarrow H$

I₄ states that the statement that P_X takes place in A₁ is true if the following two premises are true: (1) the antecedent of I₁; and (2) a difference in T_X performance is observed if the antecedent of I₃ is true (analogous reasoning including $\neg T_{XD}$ instead of T_{XD} can be used to infer about the lack of A₁ involvement in P_X).

Again, since the inference is inductive, I₄ is not immune to error and H might be false. Even if it is not, I₄ merely adds to I₁ that whenever rTMS₁ is applied to A₁, its activity is changed, and if this occurs then a difference in T_X performance is observed. However, this reasoning pattern does not guarantee the correctness of the conclusion that the change in A₁ activity is a cause of the difference in T_X performance, and therefore that P_X takes place in A₁. It may be the case that TMS₁ is a cause of both the change in A₁ activity and the difference in T_X performance, but the change in A₁ activity is not a cause of the difference in T_X performance. Thus, the causal inference between rTMS₁ to A₁ and the difference in T_X performance is stronger when the purported cause is brain stimulation but not when the purported cause is the change in brain activity, i.e., TMS causes are not analogs of neural causes. To strengthen I₄ inference one might additionally provide evidence that whenever the difference in T_X performance is observed the change in A₁ activity is present (inference 5; I₅):

H – P_X takes place in A₁

S_{1A}—rTMS₁ is applied to A₁

T_{XD}—a difference in T_X performance is observed (as compared to a control condition)

A_{1C}—a change in A₁ activity is present

$I_5((((H \wedge S_{1A}) \rightarrow T_{XD}) \wedge (S_{1A} \rightarrow T_{XD})) \wedge$
 $((S_{1A} \rightarrow A_{1C}) \wedge S_{1A}) \rightarrow T_{XD}) \wedge (T_{XD} \rightarrow A_{1C})) \rightarrow H$

I₅ states that the statement that P_X takes place in A₁ is true if the following two premises are true: (1) the antecedent of I₄; and (2) a change in A₁ activity is present if a difference in T_X performance is observed.

I₄ and I₅ are improvements over I₁, and I₂ and provide more confidence in TMS results. However, the limits of TMS-based conclusions also strongly depend on the complexity of the brain processes/cognitive functions investigated. The assumption that P_X takes place in A₁ may be simply inadequate because the complexity of P_X may require it to be executed by a network rather than a single area (Pessoa, 2014), i.e., a brain area determined with TMS to be “responsible” for a certain cognitive function may be necessary but not sufficient for the realization of this cognitive function. Thus, instead of focusing on the functional properties of a single brain area, often it is necessary to investigate the functional interactions between remote but interconnected brain regions (for a review of different

paradigms, see Romei et al., 2016). However, even though H might alternatively state that A_1 is partly (not fully) responsible for P_X , all the above issues related to the described inferences still hold.

In essence, the employment of neuroimaging may allow the following questions to be answered: (1) Does rTMS₁ applied to A_1 lead to a detectable change in A_1 activity (Siebner et al., 2000)?; (2) How big is the influence of rTMS₁ on areas adjacent to A_1 ?; (3) Which areas are functionally connected to A_1 , and are they involved in P_X and/or T_X (Bestmann et al., 2005)?; (4) How does rTMS₁ affect connectivity between certain brain areas or networks (Gratton et al., 2013)?; (5) What is the relation between the effects of rTMS₁ and the other brain activations that occur during T_X ?; (6) What is the relation between the effects of rTMS₁ and the difference in T_X performance?; and (7) Which kind of neuroplastic changes arise, and when (Poepl et al., 2018)? These investigations might be supported by the use of effective connectivity measures (Iwabuchi et al., 2019) based on the application of causal dynamic modeling, Granger causality (Friston et al., 2013), or graph theory (Farahani et al., 2019). Additionally, novel modeling approaches that can localize cortical TMS effects might be employed to determine whether the cortical area is effectively stimulated by TMS (Weise et al., 2020). At the same time, neuroimaging evidence can include confounding activations rather than clearly represent the network responsible for the cognitive function X because: (1) TMS₁ may serve as a common cause that has several transcranial and non-transcranial consequences (Conde et al., 2019), thus some of the brain activations (including compensatory mechanisms) may be unrelated to P_X ; and (2) engagement in T_X may activate processes unrelated to P_X (which can be addressed with appropriate control conditions). Therefore, determining whether observed changes in brain activity are associated more with activity change in A_1 or its adjacent areas and differentiating between network effects related to P_X and compensatory effects is both challenging. In sum, the above patterns of reasoning may still lead to false conclusions, especially if no adequate control condition is employed.

rTMS CONTROL CONDITIONS

TMS might result in various psychological, auditory, and somatosensory side effects that might trigger shifts of attention, influence alertness, or interact with elements of the experimental task. Factors like the placement of the TMS coil or the occurrence of a clicking sound can influence task performance. For example, Duecker et al. (2013) showed that lateralized sham TMS pulses caused automatic shifts of spatial attention towards the location of the TMS coil. The use of sham TMS is intended to account for the impact of active TMS's placebo and sensory side effects. The former is related to behavioral and cognitive changes (including certain expectations) that result from a person's belief that their brain is being stimulated, while the latter is related to somatosensory effects (e.g., muscle twitches), peripheral nerve stimulation, and auditory effects (perception of a clicking sound). The sham approach might induce placebo effects of different magnitude (Burke et al., 2019).

The mismatch between active TMS and the sensory effects of control TMS can form participants' beliefs about the effectiveness of brain stimulation. The sham approaches can to a certain degree reproduce the sensory effects of active TMS without meaningfully influencing brain activity. They are based on the employment of either regular but tilted TMS coils, in which case, the electric field can still be sufficiently strong to result in somatosensory effects and peripheral nerve stimulation (Loo et al., 2000; Lisanby et al., 2001) or purpose-built sham TMS coils which have a magnetic shield that attenuates the electromagnetic field and prevents stimulation of the brain concurrently limiting somatosensory and peripheral nerve stimulation effects (for a review, see Duecker and Sack, 2015). To mitigate the trade-off between invoking somatosensory effects and not stimulating the brain, Duecker and Sack (2015) recommend the use of surface electrodes for skin stimulation in combination with a sham TMS coil.

However, sham TMS approaches do not demonstrate area specificity. Thus, Duecker and Sack (2015) recommend it might be beneficial to use sham TMS over each brain area where active TMS is applied to ensure that all stimulation sites have a control condition for the sensory side effects of TMS. Proper choice of control condition/s involves taking into account the difference between clinical and experimental research as well as whether and how the investigated process can be influenced by participants' beliefs. While single-blinding seems to be feasible in between-subject designs, due to distinctive TMS effects, double-blinding is difficult to perform (Broadbent et al., 2011). However, it is practiced to use the sham and active TMS coils that are indistinguishable to the researcher carrying out the stimulation, and/or this researcher is not informed about the hypothesis of the study (Basil et al., 2005). One might also minimize the placebo effect-related issues by the employment of between-subject designs (on the cost of increasing interindividual variability). Despite the chosen design, the researcher might gather from participants information on blinding success or how the TMS was experienced in a form of a short questionnaire which can further inform the study results (Flanagan et al., 2019). An alternative to the control stimulations (including active and sham TMS control strategies) might be an investigation of interindividual differences in the response to TMS measured with neuroimaging techniques and correlating them with the chosen behavioral measure.

The probabilistic strength of inferences based on experimental studies largely depends on the type of control condition used. Below, we discuss how considerations regarding control condition/s apply to TMS research designs. In general, when investigating whether P_X underlies cognitive function X , the simplest study designs consist of investigating a difference in T_X performance between pre- and post-TMS conditions or between the application of TMS₁ and a sham rTMS protocol (rTMS₀) to the same area (Duecker and Sack, 2015).

Suppose that TMS₁ and rTMS₀ protocols were applied to A_1 . If a difference in T_X performance is observed between rTMS₁ and rTMS₀ conditions, besides explanations based on sensory and placebo TMS effects (Duecker and Sack, 2015) there are alternative explanations that should be taken into consideration

that is related to the direct and indirect influence of TMS on: (1) the areas surrounding A_1 ; (2) excitability of A_2 , which could be more important for executing P_X ; (3) processes responsible for general cognitive functions; and (4) processes not specific to cognitive function X but to T_X execution. Given this, eliminating these possible alternative explanations should guide the designs of TMS studies.

Protocol Control

Ideally, $rTMS_0$ should account for sensory and placebo effects of $rTMS_1$ but does not cause a change in A_1 activity (Duecker and Sack, 2015). Typically used $rTMS_0$ that attempts not to influence brain activity fail to control for all the effects that are not specific to the change in A_1 activity because we might assume the ideal control should influence areas which are stimulated when A_1 is targeted with TMS to separate the consequence of the change in A_1 activity from the consequences of influencing other brain areas. For example, if an area is embedded in brain folds or lies relatively deep in the brain, then distal cortical areas which are situated above that area are affected by the electrical field, most likely more strongly (Heller and van Hulsteyn, 1992). This issue (a direct stimulation influence on the areas surrounding A_1) can be partly addressed with a control condition by diminishing the intensity of the used protocol to account for the stimulation of the areas lying above A_1 , i.e., influencing cerebrospinal fluid distribution or superior areas while not reaching A_1 in a significant manner. However, it has to be taken into account that the relationship between TMS protocol intensity and its outcome might not be linear (e.g., Chung et al., 2018). Additionally, active protocols with certain frequency patterns are often classified in TMS literature as “inhibitory” or “excitatory”. Thus, sometimes the protocol patterns of $rTMS_1$ and another active rTMS protocol 2 ($rTMS_2$) differ and might be commonly conceived as being inhibitory and excitatory, respectively; thus, they are used to obtain a difference in T_X performance directly (e.g., Gann et al., 2020) or to prime cortex excitability before the application of other protocols (e.g., Todd et al., 2009). It is important to note that inhibitory and excitatory rTMS properties are extrinsic to the protocol pattern and may vary depending on, e.g., protocol length, current direction, intensity, genome, and the targeted area characteristics, including its tissue excitability history and tissue excitability before protocol application (Polanía et al., 2018). Therefore, applying TMS_1 and TMS_2 separately to A_1 cannot inform what change or difference in A_1 activity is represented by a difference in T_X performance unless it is previously known how the activity of A_1 is related to the difference in T_X performance, or the change in A_1 activity was recorded with neuroimaging methods that can differentiate between an increase or a decrease of A_1 activity.

Area Control

The following, previously mentioned, issues can be addressed with a control condition that includes a control area: (1) stimulation of areas next to A_1 ; (2) an indirect network effect on A_2 activity that is more important for executing P_X ; and (3) influence on processes responsible for more general cognitive functions than cognitive function X issue that undermine

the strength of TMS-based inferences. In TMS studies, it is often assumed that an adequate control condition employs a stimulation protocol that affects an area that has the lowest possibility of playing a role in P_X or does not influence the brain at all.

For a long time, the vertex was conceived to be such a site because it was presumed that its stimulation does not affect the brain at all. Nonetheless, several years ago it was shown that the blood oxygen level-dependent (BOLD) signal decreases in the default mode network after applying 1 Hz rTMS to the vertex, and this is not accompanied by any significant BOLD increases throughout the brain (Jung et al., 2016). The authors concluded that this supports the use of vertex stimulation as a control condition. However, such a conclusion is problematic for several reasons. First, it presumes that an increase in the BOLD signal, which determines which parts of the brain are most active, will be observed after the application of a protocol that predominantly acts in an inhibitory manner (Fitzgerald et al., 2002). Second, there is an assumption that a decrease in the BOLD signal cannot indicate a change in neuronal activity (which could represent an increase in the activity of inhibitory neurons). Also, distinctly increasing and decreasing neuronal activity in an area is not equivalent to improving and impairing a cognitive function that depends on this area. Some brain processes require a decrease in local brain activity, e.g., deactivation has often been observed in the hippocampus during encoding and retrieval tasks believed to recruit this brain structure (Axmacher et al., 2009). Third, there is an assumption that the adequate control area is the one with the lowest possibility of affecting P_X . Targeting A_2 (an area which is not anticipated to carry out P_X) does not confirm the specificity of A_1 for carrying out P_X , i.e., that P_X is carried out exclusively in A_1 . Since the evidence in favor of the specificity of A_1 is based on inductive reasoning, in theory, it would be required to effectively stimulate all brain areas to conclude that A_1 and only A_1 is responsible for P_X . Conceivably, an opposite approach should be adopted: adequate control for the site requires the selection of a control site that has a high probability of influencing P_X . However, this approach is challenged by consideration of possible indirect network influences on A_1 due to the possibility of the control site's involvement in processes interacting with P_X . Furthermore, assume that P_X requires activation in areas A_1 and A_2 . When a difference in T_X performance between the conditions with $rTMS_1$ to A_1 and $rTMS_1$ to A_2 is analyzed and $rTMS_1$ in the first condition resulted in impairment of T_X performance but in the second condition resulted in improvement of T_X performance, one might erroneously conclude that only one area is crucial for X . Similarly, if $rTMS_1$ in both conditions influenced T_X performance in the same manner, one might erroneously conclude that $rTMS_1$ was ineffective. Thus, limiting control conditions to area control might be not sufficient to adequately explain the TMS effect.

Task Control

The issues of influencing processes responsible for more general cognitive functions rather than cognitive function X and influencing processes specific to T_X but not to cognitive function X , both of which weaken the strength of TMS-based inferences,

can be addressed with task control. Dissociations may help reduce the probability of drawing erroneous conclusions on the neural bases of cognitive functions (Machery, 2012). To solve complex issues regarding certain cognitive functions or to include a task control condition in a study, e.g., to demonstrate that a certain brain area is selectively engaged in the execution of P_X but not in the execution of the neuronal process that underlies a different cognitive function Y (P_Y), rTMS can be employed to determine whether the neural underpinnings of cognitive functions X and Y differ. In this case, inferences can be based on a single dissociation that is observed whenever TMS influences T_X and influences T_Y to a lesser extent. This may lead to the conclusion that A_1 plays a role in P_X but not P_Y .

However, the results of studies employing task control may still be confounded by the confounds already mentioned. Additionally, the following confounds might be present: (1) a task that taps into one of two processes (T_X into P_X) might be less sensitive than a task that taps into another one (T_Y into P_Y); (2) due to its characteristics, P_X might be more difficult to measure than P_Y ; (3) the relative difficulties of T_X and T_Y are likely to require a different amount of available cognitive resources (e.g., memory, attention); (4) when cognitive resources are limited, different brain networks may be engaged in T_X or T_Y execution than when they are available; and (5) a discrepancy between how T_X and T_Y engage A_1 and A_2 can be observed, even when they recruit the same area or network, e.g., carrying out T_X may require a decrease in A_1 activity, while carrying out T_Y may require an increase in A_1 activity. In all the above circumstances, it would be erroneous to conclude with certainty that cognitive functions X and Y are based on two distinct brain substrates. The solution may consist of designs that combine different control approaches and allow double dissociation (Dunn and Kirsner, 2003), e.g., T_X but not T_Y performance is impaired when rTMS₀ application and rTMS₁ application outcomes are compared after stimulation to A_1 , while T_Y but not T_X performance is impaired when the rTMS₀ and rTMS₁ outcomes are compared after stimulation to A_2 . In the case of an uncrossed double dissociation, a difference in T_X performance and a difference in T_Y performance is observed when A_1 condition and A_2 condition are compared (when pre- and post- rTMS₁ or rTMS₁ and rTMS₀ are compared) but one condition is associated with higher performance in both tasks. A cross-over double dissociation is observed when rTMS₁ to A_1 influences T_X performance more than rTMS₁ to A_2 , and rTMS₁ to A_2 influences T_Y performance more than rTMS₁ to A_1 (for a summary of the solutions that aim to control for TMS confounds, see **Figure 2**).

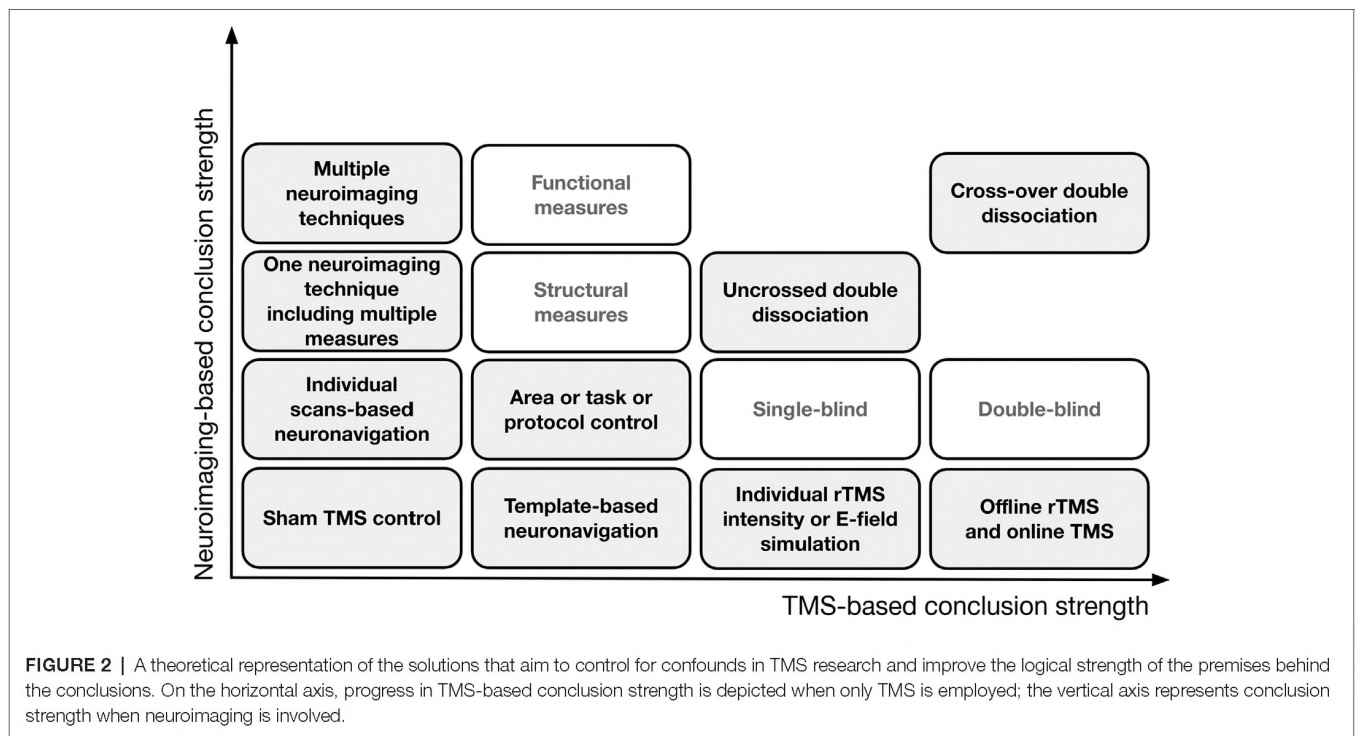
Can it then be concluded that P_X takes place in A_1 while P_Y takes place in A_2 ? Unfortunately, most of the mentioned confounds also apply to double dissociations (e.g., rTMS₁ to A_1 reduces the available cognitive resources to T_X , while S_2 to A_2 reduces them to T_Y). In the case of uncrossed double dissociations, the additional confound may be that the task demand function for A_1 increases monotonically, while the task demand function for A_2 is U-shaped: A_2 is more active when a task requires fewer or more cognitive resources. In such circumstances, if T_X and T_Y recruit a single process whose neural correlate includes A_1 and A_2 , for A_1 the greater task demands

may correspond to the increase in its activity, while for A_2 the greater task demands can correspond to its inactivation. Such an issue can be avoided when a cross-over double dissociation is observed, but the following confounds may still be present: (1) neuroplasticity-like effects occur at a different rate in A_1 and A_2 (e.g., depending on the type of brain cells affected by the stimulation); (2) rTMS₁ and rTMS₂ protocols applied to different areas may differently influence excitability in these areas; (3) an increase in A_1 excitability results in a decrease in A_2 activity, which is necessary to perform T_Y , while an increase in A_2 excitability results in inactivation of A_1 , which is the area necessary to perform T_X ; (4) the execution of P_X may correspond to A_1 activity increase while the execution of P_Y may correspond to A_1 inactivation; and (5) both A_1 and A_2 are recruited depending on the available cognitive resources, and the processes recruited when the amount of available resources is greater differ from the processes recruited when fewer resources are available. In all the above circumstances, it would be premature to conclude with certainty that cognitive functions X and Y are based on two distinct brain substrates.

In certain types of research (mostly preclinical and clinical studies), rTMS effects might be studied using longitudinal designs. The effect of longitudinal rTMS studies can be long-lasting, thus they can be used to investigate stable neuroplastic changes and determine whether the observed rTMS effect consistently arises over the time course of a study (Auriat et al., 2015). They also reduce the erroneous identification of side effect-associated changes as the brain stimulation effect, and they enable the employment of multiple testing measures. Similar to single-session rTMS effects, the rTMS effects in longitudinal studies might be related to individual excitability of brain areas, but they are less prone to the influence of day-to-day fluctuations in cortex excitability (Huber et al., 2013). However, there is still a possibility that the long-term effects of neuroplasticity in longitudinal studies might be related to placebo effects or be influenced by confounding factors that occur over the time course of the study.

CONCLUSIONS

TMS has traditionally been used to provide evidence for functional brain specialization. Nevertheless—as has been getting clearer over the past two decades—the application of rTMS alone does not allow causal inferences to be drawn on neural causes without additional assumptions. A change in the execution of an experimental task might be a consequence of rTMS but at the same time not a consequence of a change in the excitability of a targeted area. However, this might be avoided when: (1) the research question is grounded in previous research and accounts for the complexity of the investigated cognitive function; (2) neuroimaging/neurophysiological techniques are employed to monitor the direct and indirect influence of rTMS; and (3) more than one control condition is employed in a single experiment to reduce the number of possible interpretations. On one hand, functional neuroimaging could make it possible to determine whether the process responsible for the investigated



cognitive function has local or network characteristics and can be used to study the spread of TMS effects throughout the brain networks. On the other hand, confounding factors of neuronal correlates of investigated cognitive processes need to be addressed within each TMS-neuroimaging study. Although TMS has been proven to be a very effective brain stimulation method, its characteristic features have to be considered in reasoning based on its employment. In this article, we have clarified the difference between the causal effects of TMS and structure-related causal effects, and we have pointed out that the latter can be divided into direct and network effects. We have also outlined issues related to TMS-based inferences. Taking them into account requires limiting the extent of TMS-based reasoning but at the same time may support analysis of possible confounds and improve research designs to alleviate these confounds. Although the aforementioned issues are often addressed by experts in the field of non-invasive brain stimulation, we hope that the presented summary and theoretical analysis will help researchers who are developing the field of human-neuroscience based on TMS-based inferences. Even though rTMS without neuroimaging cannot unequivocally prove structure-related causal claims concerning direct relations between brain processes carried out in certain areas and certain behaviors/cognitive functions, it might be used for probabilistic statements about causal influences if its limitations are kept in mind. The fact that combining rTMS with neuroimaging techniques allows stronger inferences to be made does not imply that one should use rTMS only in combination with neuroimaging or/and multiple control conditions. The need for neuroimaging or/and multiple control conditions depends on the research question guiding the study and how its results

are intended to be interpreted. There is a trade-off between the inferential limit and experimental feasibility; therefore, when feasible, combining rTMS with neuroimaging, multiple control conditions, and/or perturbational TMS is recommended and might provide further support for conclusions regarding experimental outcomes.

AUTHOR CONTRIBUTIONS

JH drafted the manuscript. MK, KS, and MW suggested changes and provided comments on the manuscript. JH improved the manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Tilburg, 29.04.23

Dr. Michał Klincewicz, Assistant Professor, Tilburg University

Co-authorship statement

As a co-author of the publication: **Causal inferences in repetitive transcranial magnetic stimulation research: Challenges and perspectives, *Frontiers in Human Neuroscience*, 14, 2021, 574**, I declare that my own substantial contribution to this publication consists of:

- support in developing the article concept,
- providing comments and suggestions on the draft of the manuscript,
- providing comments and suggestions on the reviewed version of the manuscript,
- providing comments and suggestions on the responses to reviews,
- proofreading of the manuscript and the responses to reviews.

Hereby, I agree to submit the above-mentioned publication by MSc Justyna Hobot as part of a doctoral dissertation in the form of a thematically coherent collection of articles published in scientific journals.

I declare that an independent and identifiable part of the abovementioned publication shows individual contribution of MSc Justyna Hobot consisting of:

- developing the article concept,
- drafting the manuscript,
- correcting the manuscript,
- managing the publication process.



.....
(co-author's signature)

Copenhagen, 13.04.23

Dr. Kristian Sandberg, Associate Professor, Aarhus University

Co-authorship statement

As a co-author of the publication: **Causal inferences in repetitive transcranial magnetic stimulation research: Challenges and perspectives, *Frontiers in Human Neuroscience*, 14, 2021, 574**, I declare that my own substantial contribution to this publication consists of:

- providing comments on the manuscript.

Hereby, I agree to submit the above-mentioned publication by MSc Justyna Hobot as part of a doctoral dissertation in the form of a thematically coherent collection of articles published in scientific journals.

I declare that an independent and identifiable part of the abovementioned publication shows individual contribution of MSc Justyna Hobot consisting of:

- developing the article concept,
- drafting the manuscript,
- correcting the manuscript,
- managing the publication process.


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Krakow, 13.04.23

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Co-authorship statement

As a co-author of the publication: **Causal inferences in repetitive transcranial magnetic stimulation research: Challenges and perspectives, *Frontiers in Human Neuroscience*, 14, 2021, 574**, I declare that my own substantial contribution to this publication consists of:

- providing comments on the manuscript.

Hereby, I agree to submit the above-mentioned publication by MSc Justyna Hobot as part of a doctoral dissertation in the form of a thematically coherent collection of articles published in scientific journals.

I declare that an independent and identifiable part of the abovementioned publication shows individual contribution of MSc Justyna Hobot consisting of:

- developing the article concept,
- drafting the manuscript,
- correcting the manuscript,
- managing the publication process.

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4.2. Article 2 with co-authorship declarations

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Continuous Theta Burst Stimulation to the left anterior medial prefrontal cortex influences metacognitive efficiency

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ABSTRACT

The contribution of the prefrontal areas to visual awareness is critical for the Global Neuronal Workspace Theory and higher-order theories of consciousness. The goal of the present study was to test the potential engagement of the anterior medial prefrontal cortex (amPFC) in visual awareness judgements. We aimed to temporarily influence the neuronal dynamics of the left amPFC via neuroplasticity-like mechanisms. We used different Theta Burst Stimulation (TBS) protocols in combination with a visual identification task and visual awareness ratings. Either continuous TBS (cTBS), intermittent TBS (iTBS), or sham TBS was applied prior to the experimental paradigm in a within-participant design. Compared with sham TBS, we observed an increase in participants' ability to judge their perception adequately (metacognitive efficiency) following cTBS but not iTBS. The effect was accompanied by lower visual awareness ratings in incorrect responses. No significant differences in the identification task performance were observed. We interpret these results as evidence of the involvement of PFC in the brain network that underlies metacognition. Further, we discuss whether the results of TMS studies on perceptual metacognition can be taken as evidence for PFC involvement in awareness itself.

1. Introduction

One of the commonly discussed issues in consciousness research is whether activity of the posterior part of the brain is sufficient for visual awareness, or whether PFC involvement is also necessary (for the review, see Boly et al., 2017; Koch et al., 2016; Owen and Guta, 2019; Sandberg et al., 2016). There is evidence relating perceptual awareness to late long-range frontoparietal activity. This evidence comes from multiple fMRI studies which registered activity in frontal regions when participants reported being conscious of certain visual stimuli (e.g., Beck et al., 2001; Binder et al., 2017; Lumer et al., 1998; Imamoglu et al., 2014; Lau and Passingham, 2006; for the review, see Dehaene and Changeux, 2011) and some EEG studies (e.g., Salti et al., 2015). The activity of the PFC, including dorsolateral and medial PFC areas, has been suggested to constitute NCC related to conscious access to the con-

tent of perception (Dehaene and Naccache, 2001; Haynes et al., 2005; Imamoglu et al., 2014). Moreover, recent evidence shows that the inferior frontal cortex contributes to resolution of perceptual ambiguities (Weilhammer et al., 2021). PFC is claimed to be involved in maintaining and broadcasting specific perceptual contents (for a review, see Mashour et al., 2020; Michel and Morales, 2020). Proponents of the Global Neuronal Workspace Theory (Dehaene and Changeux, 2011) and higher-order theories of consciousness (Brown, 2015, 2019; Lau and Rosenthal, 2011; LeDoux and Brown, 2017) predominantly claim that NCC include PFC areas (Michel and Morales, 2020).

Concurrently, the threshold for access to consciousness and introspective ability have been reported to relate to structural features of the aPFC (Allen et al., 2017; Del Cul et al., 2009; Fleming et al., 2010). Similarly, a few fMRI studies have provided evidence of aPFC engagement in metacognition, i.e., knowledge about one's own cognitive processes.

Abbreviations: amPFC, anterior medial prefrontal cortex; AP, anterior-posterior; aPFC, anterior prefrontal cortex; CI, confidence intervals; cTBS, continuous theta burst stimulation; dlPFC, dorsolateral prefrontal cortex; fMRI, functional magnetic resonance imaging; sham, intermediate-sham theta burst stimulation; iTBS, intermittent theta burst stimulation; M1, primary motor cortex; MNI, Montreal Neurological Institute; MRI, magnetic resonance imaging; MEG, magnetoencephalography; MSO, maximal stimulator output; NCC, neural correlates of consciousness; NS, not significant; PA, posterior-anterior; PAS, Perceptual Awareness Scale; RMT, resting motor threshold; RT, reaction time; rTMS, repetitive transcranial magnetic stimulation; SD, standard deviation; TBS, theta burst stimulation; TMS, transcranial magnetic stimulation.

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The metacognition measured with perceptual confidence ratings correlates with grey matter volume and myelination in the aPFC. However, different studies link it to functionally lateralized lateral or medial areas (Allen et al., 2017; McCurdy et al., 2013; Fleming et al., 2010, 2012). Higher metacognition in a multiple choice question task followed by confidence rating has been associated with decreased left aMPFC activity (Molenberghs et al., 2016) and greater right medial aPFC functional connectivity with other brain regions (Baird et al., 2013). Moreover, the right aPFC has been reported to be functionally involved in the metacognitive aspects of decision-making (Fleming et al., 2012). Higher right aPFC activity has also been linked to better metacognition about short-term recognition memory (Yokoyama et al., 2010), which shows that the aPFC may play a role in metacognition in general, possibly thanks to the functional diversity of its sub-areas (Gilbert et al., 2010).

1.1. Controversy on the role of PFC for visual awareness

While some research showed that the right dlPFC and the left aPFC lesions shift the threshold for reporting stimulus awareness (Colás et al., 2019; Del Cul et al., 2009), other lesion studies on PFC (likely because of different lesion extent) did not (Eslinger and Damasio, 1985; Kozuch, 2014; Tononi and Laureys, 2009; for the review see Boly et al., 2017). Nevertheless, multiple studies investigating visual NCC did not find PFC activity (Grill-Spector et al., 2000; Tse et al., 2005; Williams et al., 2008) to be associated with visual awareness. They claim that visual awareness is associated either with the early activity of the occipital cortex and/or association areas beyond the PFC (for a review, see Koch et al., 2016; Storm et al., 2017; Tononi et al., 2016). The relationship between the P3 wave (believed to have a partly frontal origin) and awareness in some early EEG studies (e.g., Sergent et al., 2005) have been taken as evidence for the role of the PFC in awareness, yet later MEG studies have shown that stimulus awareness is more reliably decoded from occipital than from frontal sources (Andersen et al., 2016; Sandberg et al., 2013). Taken together, these studies thus do not provide strong evidence that the PFC is part of the NCC (for a review, see Förster et al., 2020).

At the same time, neural activity in the PFC might be related to subjective reports but not to awareness of perceptual content. It is claimed that some modulations of PFC activity represent processes that co-occur with or follow stimulus awareness (for a review, see Aru et al., 2012; De Graaf et al., 2012). Therefore, some researchers suggest that results supporting the role of PFC in visual awareness in fact reflect higher-order post-perceptual aspects of visual awareness, like perceptual information maintenance (especially in identification tasks), or planning and execution responses, e.g., report encoding (Andersen et al., 2016; Brascamp et al., 2015; Frassle et al., 2014; Grill-Spector et al., 2000; Pitts et al., 2014; for the review, see Storm et al., 2017; Tsuchiya et al., 2015, 2016). However, when defending PFC involvement in awareness formation, researchers point to studies where PFC activity is registered even if a report is not required (Noy et al., 2015; Vidal et al., 2015). A couple of studies that recorded neuronal ensemble activity from the macaque ventrolateral PFC provide similar support (Bellet et al., 2022; Kapoor et al., 2022; see also Panagiotaropoulos et al., 2020). All the aforementioned studies led us to consider whether the PFC is involved in visual awareness per se or in some forms of metacognition or introspection.

1.2. TMS to PFC and visual awareness

As illustrated in the previous sections, neuroimaging has not provided conclusive evidence of whether the PFC has a causal role in stimulus awareness. Therefore, TMS has been used to manipulate processes related to perceptual judgements by targeting the dlPFC. The first evidence that activity in the dlPFC is causally related to changes in conscious perception was provided by Turatto et al. (2004), who employed

10-Hz rTMS while participants were performing a visual change detection task. To date, according to our knowledge, only one offline rTMS study has reported the engagement of the PFC in the formation of visual awareness judgements. This was shown by applying cTBS to the bilateral middle frontal gyrus and testing visual stimuli perception with an identification task combined with visual awareness judgements (Rounis et al., 2010). Compared to the sham, the cTBS resulted in an overall decrease in metacognitive efficiency, i.e., the ability to judge one's own perception accurately (Fleming and Lau, 2014; in studies on consciousness, metacognitive efficiency is typically estimated as a relation between awareness rating and task performance). This effect was accompanied by lower visual awareness ratings in correct responses in the post-cTBS versus the pre-cTBS condition. A replication study did not reproduce these effects (Bor et al., 2017). Interestingly, Rahnev et al. (2016), contrary to their expectations, reported an increase in metacognitive efficiency after cTBS to the right aPFC as compared to the primary somatosensory cortex. Moreover, this effect was also present after cTBS to the right dlPFC. A similar effect was observed after bilateral cTBS to the aPFC and was interpreted as being related to improved associative recognition memory awareness (Ryals et al., 2015). However, a recent study presented a disruption in prospective memory awareness judgements after applying cTBS to the left PFC (Brodmann area 9/10; Carbajal et al., 2019). Taken together, previous studies have not provided consistent evidence to determine the role of the PFC (and especially the aPFC) in the formation of visual awareness judgements.

1.3. Measurement of awareness and metacognition

Where does the inconsistency between the results investigating the role of the PFC in visual awareness judgements come from? Partly, it might result from the measures of awareness employed. Awareness cannot be measured directly, and multiple empirical awareness measures have been proposed so far (Overgaard, 2017). It is considered that objectively detectable stimuli can remain subjectively invisible (at least to some degree). Thus, awareness measures mostly include subjective scales: sometimes perceptual awareness is measured only with a scale (e.g., Christensen et al., 2006), while sometimes it is measured with a combination of an objective task and a scale (e.g., Schwiedrzik et al., 2009). When researchers to measure visual experience use an objective task only, they most often use the term 'visibility' (e.g., Imamoglu et al., 2014). The association between an objective task and a scale is often analysed (Sandberg et al., 2010; Wierzchoń et al., 2014). In this paper, we operationalize visual awareness judgements as reports of stimulus visibility (Lyyra, 2019; Overgaard et al., 2006), which depend on both the processes responsible for generating visual awareness and the processes related to metacognitive efficiency. Following Overgaard and Sandberg, 2012, we consider the latter to be equivalent to introspection.

All variants of higher-order theories state that a higher-order mechanism is necessary for awareness to occur (Michel and Morales, 2020), while some theoretical approaches frame awareness as a first-order mental state (Overgaard and Sandberg, 2012). At the same time, awareness reports are considered to represent a specific type of metacognitive decision, namely an internal decision about the perceptual content (Fleming, 2020; Overgaard and Sandberg, 2012). For these reasons, metacognitive tasks are often used to study perceptual awareness (Bor et al., 2017; Norman and Price, 2015; Rounis et al., 2010). Since the term 'metacognition' refers to cognitive processes that concern other cognitive processes, it can be conceived in two ways in research on visual awareness: as a metacognitive process where a higher-order process represents or operates on a lower-order process, thus allowing some content of visual perception to become consciously perceived (Brown, 2015; Lau, 2019; Lau and Rosenthal, 2011); or as a metacognitive process (judgement of conscious experience) which operates on conscious representation (process responsible for conscious experience, irrespective of whether this representation is considered to be of a higher-order; Fleming, 2020; Overgaard and Sandberg, 2012). Since metacognition is

typically considered a process that we are conscious of engaging in, by referring to metacognitive efficiency in this paper, we refer to a process of metacognitive judgement we are conscious of engaging in and that concerns the process of visual perception (without postulating or negating the presence of representational levels).

To examine the involvement of the aPFC in visual awareness, we conducted a study in which we administered two active and one sham TBS protocols; we expected that the active protocols may exert the opposite influence on cortex excitability level as has been observed for the primary motor cortex (Huang et al., 2005). We stimulated a left aMPFC area that is proposed to be related to metacognitive efficiency (McCurdy et al., 2013; Molenberghs et al., 2016). We then (1) measured the extent to which participants are objectively sensitive to visual information, which is represented by an estimate of performance in an identification task (the contrast of visual stimuli which were used in adaptive staircases); (2) measured visual awareness judgements represented by PAS ratings. PAS is a categorical scale of stimulus awareness measurement introduced by Ramsøy & Overgaard (2004); (3) measured metacognitive efficiency, i.e., participants' ability to differentiate the content of their visual perception, which is represented by a logistic regression estimate, as used in our previous research (Łukowska et al., 2018; Sandberg et al., 2010; Wierchoń et al., 2014, 2019), and supplemented with meta-d'/d' (M-ratio; Maniscalco and Lau, 2012). M-ratio is a common measure of metacognitive efficiency, in which metacognitive efficiency (operationalized with meta-d') is corrected for objective task sensitivity (operationalized with d'; Fleming and Lau, 2014).

In summary, the goal of this study was to determine whether influencing left aPFC excitability affects reported visual awareness and, if so, whether it is associated with an overall shift in awareness judgements or/and an impact on metacognitive efficiency. Considering the diversity in previous research outcomes, our hypotheses were non-directional. We assumed that a difference in identification task performance should simply indicate a change (not necessarily conscious) in stimulus recognition. Impairment of the processes responsible for visual stimulus experience should lead to lower awareness ratings for both correct and incorrect identification responses. Therefore, (in principle) such impairment would not influence the metacognitive efficiency measure. However, a decrease in metacognitive efficiency should lead to misclassification of the perceptual content, i.e., lower PAS ratings in correct responses and/or higher PAS ratings in incorrect responses. Thus, a difference in the regression estimate and M-ratio that is not accompanied by a difference in identification task performance or overall PAS ratings would imply a difference in metacognitive efficiency.

2. Materials and methods

2.1. Participants

Twenty-four native Danish volunteers with normal or corrected-to-normal vision who fulfilled the criteria for participation in TMS studies (no history of neurological disorders, psychiatric disorders, or head injury etc., as assessed by a safety screening questionnaire) were recruited from the volunteers' database at the center of Functionally Integrative Neuroscience, Aarhus University, Denmark. Prior to the experiment, participants were asked to complete a screening questionnaire regarding their health and safety aspects of TMS and to sign a written informed consent form. Two participants discontinued the study after the first session. One of them reported that the peripheral facial nerves impacted via TMS caused minor pain; another felt weak after a couple of TBS burst pulses applied to the PFC. Another participant reported a headache after receiving the cTBS protocol and quit the study based on medical advice. Twenty-one right-handed participants completed the study (10 males, 7 left-eye dominant, age mean = 23, SD = 2.7, range = 18–27). We determined the sample size based on the number of participants used by Rounis et al. (2010), where the sample was 20 participants (to counterbalance the three TBS protocols, we included 21 participants).

All participants received financial compensation for taking part in the study. The study was approved by the local ethics committee, *De Videnskabetiske Komitéer for Region Midtjylland*. It was carried out in accordance with the approved guidelines for TMS research (Rossi et al., 2009; Rossini et al., 2015) and adhered to the tenets of the Declaration of Helsinki (Holm, 2013). Before each application of TBS, participants were reminded that they could quit the study at any time without providing a reason.

2.2. Session sequence

The experiment lasted for four days and included four sessions, each performed at the same time of the day and separated by at least four days to reduce the influence of task learning. On the first day, participants received a couple of burst pulses to the aPFC so they could make informed decisions about further participation in the study. Afterwards, individual RMTs were estimated. Next, they received behavioural training to become familiar with the experimental task and provide us with approximate estimates of their performance level. In each of the following three sessions, participants performed a 5-minute block of task training; subsequently, they received one of three different TBS protocols (TBS order was counterbalanced across participants) and completed four 5-minute blocks of the experimental task.

2.3. Behavioral procedure

The task was run on a PC using PsychoPy software (Peirce, 2007). A chinrest and an LCD monitor (1920×1080 resolution, 60-Hz refresh rate) placed 60 cm away from the participants' eyes were used. The task was performed under constant dim lighting conditions. The experiment was conducted in English (but the PAS was presented in Danish).

Participants performed a visual identification task (Fig. 1). At the onset of each trial, a fixation dot was displayed for 500 ms followed by a target Gabor patch presented for 33 ms, tilted left or right. Next, they reported the tilting of the Gabor patch using accordingly labelled 'Z' and 'X' keyboard keys ('L' and 'R'). Finally, participants reported their visual awareness using the PAS as shown in Fig. 2A. They were instructed to report their stimulus awareness according to the PAS description provided in Fig. 2B and to give their response with one of four keys labelled 1–4 and to position the right-hand index finger over the key '1'. The response times for the identification response and the PAS response were unlimited, but participants were instructed to respond as quickly and accurately as possible. Participants could signal finger slips on each trial by pressing the space key (slip trials were excluded from the analysis).

By using two tilt-specific 3-down/1-up unlimited staircases, we kept the identification task performance at 79% throughout the training and experimental sessions, following the assumption that quantifying differences in metacognitive processing requires holding the behavioural performance constant and dissociating the effect of TBS on metacognitive measures from the identification task performance (Rounis et al., 2010). With a step size equal to 0.5%, the stimulus contrast was decreased after three consecutive correct responses, or it was increased after one incorrect response.

The first-day training session started with a displayed instruction and comprised fifteen trials with high stimulus contrast and accuracy feedback (right/wrong), followed by a 10-minute training session with the experimental task, including verbal feedback from the experimenter, and another 10-minute individual training session with no feedback. The experimental sessions started from individual stimulus contrasts, which were established based on the training session's performance, determined for the left and right Gabor patch separately. Similarly to Rounis et al. (2010) and Bor et al. (2017), the staircases were identical for all sessions. Outside of the staircase, after every 15, 20 or 25 trials, a trial with the same high-contrast stimulus was run to prevent participants from changing awareness judgement criteria during the period

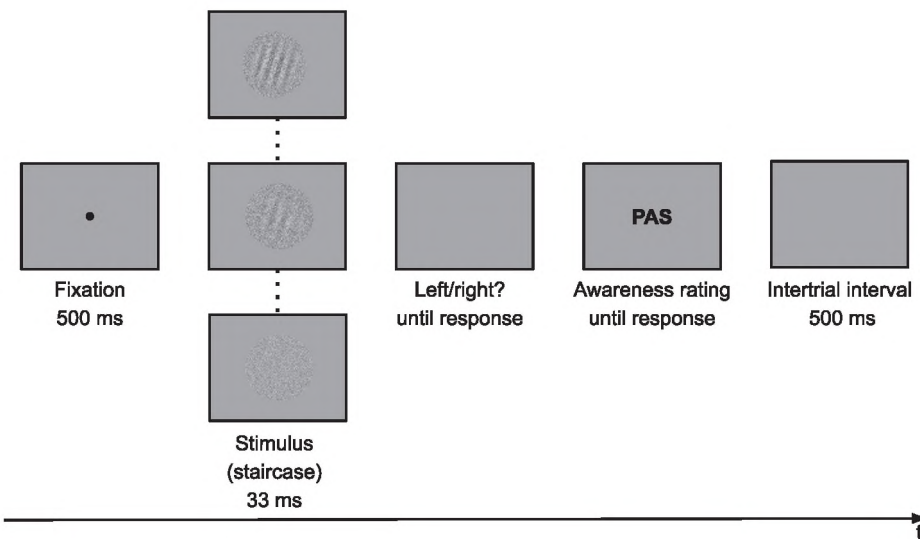


Fig. 1. Schematic illustration of the experimental task. A circular fixation dot (visual angle $\sim 0.5^\circ$, presentation time = 500 ms) was presented at the screen centre, followed by a Gabor patch of variable contrast (size = 128x128 pix, visual angle $\sim 3^\circ$, spatial frequency ~ 4 cycles per degree, a standard deviation of the Gaussian envelope = 5 pixels, presentation time = 33 ms, tilted -45° or 45° rotation from vertical angle respectively) embedded in a same-sized circular visual white noise against a grey background, with equal probability and in random order. Participants were required to perform an identification task, i.e., to determine the tilting of the Gabor patch. After each identification response, participants provided a visual awareness rating of stimulus tilting using the PAS. The trials were separated by 500-ms intervals.

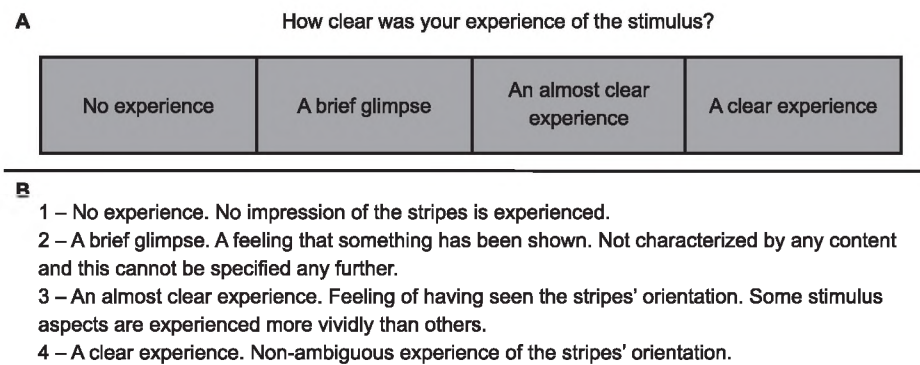


Fig. 2. A) The PAS as presented in every trial. B) The PAS description – an adapted version of the original PAS, published by Ramsøy and Overgaard (2004). This description was provided in the instructions for the pre-training and pre-testing sessions. Following the theoretical position of the authors of the original scale, we treated the PAS as the measure of visual experience, thus we excluded the fragments of the original PAS point definition that referred to confidence.

of the experiment. In each testing session, a 4 min 40-s training block preceded TBS to remind participants of how to perform the tasks. After termination of the stimulation, the participants moved to an adjacent room and commenced the behavioural testing without further delay. Testing thus typically commenced shortly after stimulation and lasted 20 min per condition.

2.4. TMS protocols

During TMS, participants wore earplugs for noise protection. The TBS protocols were delivered with a MagPro X100 stimulator using an MC-B70 Butterfly Coil (with the maximal initial dB/dt of 31 kT/s at the coil surface) for the active stimulation; an MC-Placebo-B70 Butterfly Coil was used for the sham protocol. The TBS protocols had conventional patterns and durations (Huang et al., 2005). The protocols were delivered at 75% of the individual RMT and the average intensity equalled 28% ($SD = 3.9$) of MSO. The estimation of individual RMT started from applying 40% of MSO single-pulse TMS to the left M1 and adjusting stimulation intensity. It was established at what spot the suprathreshold TMS induced the maximal twitch of the right first dorsal interosseous hand muscle. Then, TMS was delivered until the lowest intensity that resulted in motor-evoked potentials larger than 50 μV peak-to-peak amplitude on five out of ten consecutive trials was reached. The site of stimulation was determined with the Nexstim eXimia NBS 3.2 neuronavigation system using individual structural MRI images with a previously predefined region of interest. The stimulation coordinates were in the anterior medial PFC [$-9, 54, 18$] in the standard MNI space (Molenberghs et al., 2016); the transformation to individual participant brain space was done using a custom-made MATLAB script. The estimated mean peeling depth

equalled 2.6 cm ($SD = 0.15$) from the scalp. Throughout the RMT determination procedure and the subsequent application of cTBS, the main axis of the coil was orientated at 45° offset from the PA direction. The TMS pulses were biphasic ($\sim 280 \mu s$). The current in the brain was PA-AP at the M1 and AP-PA at the aPFC. The coil was kept tangentially to the scalp, which was ensured by using the neuronavigation system. The TBS protocols were counterbalanced between participants and included three protocols, under which 3-pulse bursts at 50 Hz were applied at 5 Hz. We administered two active TBS protocols: cTBS, in which burst trains were applied continuously for 40 s; and iTBS in which burst trains were applied for 2 s and repeated every 10 s for 192 s. These protocols are conceived to have opposite effects on cortical excitability (Huang et al., 2005). For the sham stimulation, we used an imTBS protocol in which burst trains were applied for 5 s and repeated every 15 s for 110 s. Participants kept their eyes closed during the application of the protocols.

2.5. Calculation

During each post-TBS session, participants completed an average of 452 trials over four blocks (1: 59–152, 2: 73–160, 3: 77–168, 4: 72–165; excluding slips, high stimulus contrast trials from outside of the staircase procedure, and trials for which the contrast estimated by the staircase was 0). The staircase procedure decreased the differences in accuracy between participants to the point where any mixed-effects model with accuracy as a dependant variable did not converge. To confirm that the staircase procedure worked as intended and to compare accuracy between conditions, we fitted a non-hierarchical logistic regression model and used the *glm* function with the binomial family. Due to employment

of the staircase procedure, the identification task performance was estimated with a change in stimulus contrast and identification task RT. To test stimulus contrast distributions, we regressed contrast on the TBS condition and random participant-specific intercepts and random TBS condition effects. To test the differences in rating distributions, we fitted a cumulative link mixed-effects model with the TBS condition as fixed effects, random participant-specific intercepts and random TBS condition effects using the Laplace approximation. To test the differences in RT in both tasks in which the response time was unlimited, we removed trials which exceeded the upper limit of 9 SD. Then we fitted linear mixed-effects regression models with TBS conditions as fixed effects, random participant-specific intercepts and random TBS condition effects. To analyse the metacognitive efficiency, we used a logistic regression model, which is assumed to be the correct model for predicting binary outcomes such as accuracy (Norman and Price, 2015), in which higher estimates indicate higher metacognitive efficiency. The metacognitive efficiency model was based on task accuracy, predicted by the interaction between the TBS condition and PAS rating with participant-specific PAS rating intercepts. To employ this model, the PAS ratings were rescaled to the 0–3 range. To further test the differences in rating distributions depending on the identification task accuracy, we fitted a cumulative link mixed-effects model with fixed and random effects of accuracy, TBS condition and their interaction. The primary analysis of metacognitive efficiency is supplemented by M-ratio analyses. To calculate M-ratios, we used both the code of Maniscalco & Lau (2014) and the HMeta-d model of Fleming (2017). The meta-d' parameter was estimated by finding the d' value that would produce the observed subjective ratings' hit and false alarm rates under the assumption of a metacognitively perfect observer. M-ratio indicates the amount of evidence available for metacognitive judgement relative to the amount of evidence available for an objective (identification task) decision, e.g., an M-ratio value of 0.7 shows that 30% of the sensory evidence available for the objective decisions is lost when metacognitive judgements are made, while a value of 1.3 suggests that more evidence is available for metacognitive judgements than for objective decisions. M-ratio values higher than 1 are considered to be due to processing of stimulus information that follows an identification decision or due to gaining non-perceptual information (Fleming, 2017; Skóra and Wierchoń, 2016; van den Berg et al., 2016); they might also be due to processing of information that is parallel in time to the identification task decision. We estimated the M-ratios for each TBS condition and each participant separately. Next, we fitted linear models to estimate the difference in each parameter between the sham TMS condition and the active TBS conditions. We calculated M-ratio to be able to compare our results with the results of previous cTBS studies (Bor et al., 2017; Rounis et al., 2010). All presented models were fitted using the R statistical environment (R Core Team, 2019). The mixed-effects regression models were fitted using the *lme4* package (Bates et al., 2015), and the cumulative link mixed-effects models were fitted with the *ordinal* package (Christensen, 2019). For the pairwise comparisons, we used the *emmeans* package, and the p values for pairwise comparisons were adjusted using the Tukey method (Lenth, 2021). The p values were estimated with the *lmerTest* package (Kuznetsova et al., 2017). The generalised linear model was fitted with the *stats* package, which is included in R. We have reported all conditions used and the data exclusions. Perhaps different measures of metacognitive efficiency can be estimated using our data. The data and the script for data cleaning are provided on the Open Science Framework (www.osf.io/3yb2g).

3. Results

All the models that estimated the differences between the conditions for each parameter of interest converged. Consistently with the predictions, the accuracy analysis of the identification task did not reveal any difference between cTBS vs sham, iTBS vs sham, and cTBS vs iTBS (Fig. 3A, Table 1A); this shows that the stimuli contrast-based staircases

Table 1

Between-conditions comparison of regression coefficients for the regression models; cTBS and iTBS compared to sham and cTBS compared to iTBS. A) Results summary of the logistic regression model for the identification task accuracy, with TBS condition as a fixed effect. B) Results summary of the linear mixed-effects regression model for the stimulus contrast, with TBS condition as a fixed effect and random participant-specific intercepts and random TBS condition effects. C) Results summary of the linear mixed-effects model for the identification task reaction time, with TBS condition as a fixed effect; participant-specific TBS condition effect and intercept were used as random effects. D) Results summary of cumulative link mixed-effects model for the PAS ratings with TBS condition as fixed effect; participant-specific TBS condition effect and intercept were used as random effects. E) Results summary of the linear model for the PAS reaction time with TBS condition as a fixed effect; participant-specific TBS condition effect and intercept were used as random effects.

A Identification task accuracy					
Predictor	Estimate	Est. Error		z value	Pr (> z)
cTBS - sham	-0.04	0.04		-1.14	.254
iTBS - sham	-0.03	0.04		-0.87	.387
cTBS - iTBS	-0.01	0.04		-0.27	.787
B Stimulus contrast					
Predictor	Estimate	Std. Error	df	t value	Pr (> t)
cTBS - sham	0.08	0.10	19.85	0.85	.406
iTBS - sham	0.19	0.10	19.98	1.96	.065
cTBS - iTBS	-0.11	0.09	19.99	-1.24	.229
C Identification task RT (ms)					
Predictor	Estimate	Std. Error	df	t value	Pr (> t)
cTBS - sham	-50.56	41.59	19.84	-1.22	.238
iTBS - sham	-42.42	49.10	19.91	-0.86	.398
cTBS - iTBS	-8.14	38.56	19.82	-0.21	.835
D PAS rating					
Predictor	Estimate	Std. Error		z value	Pr (> z)
cTBS - sham	-0.23	0.12		-1.86	.063
iTBS - sham	-0.04	0.09		-0.47	.635
cTBS - iTBS	-0.18	0.12		-1.51	.132
E PAS rating RT (ms)					
Predictor	Estimate	Std. Error	df	t value	Pr (> t)
cTBS - sham	-16.78	18.31	19.72	-0.92	.371
iTBS - sham	15.60	30.70	19.92	0.51	.617
cTBS - iTBS	-32.38	27.28	19.91	-1.19	.249

Significance code: . p < .1.

on accuracy fulfilled their role. For stimulus contrast, no significant effect was observed for any TBS comparison (Fig. 3B, Table 1B). There was close to significantly higher contrast in the iTBS condition as compared to the sham condition.

Similarly, the analysis of the identification task RT did not show any difference in all the TBS comparisons (Table 1C). The mean identification task RT was 772 ms (SD = 497).

Regarding distribution of PAS ratings, we found that the overall ratings in the cTBS condition were close to significantly lower relative to the sham TBS condition. No difference was observed for the other comparisons (Fig. 4, Table 1D).

The mean PAS rating RT was 398 ms (SD = 393). The analysis of PAS ratings RT did not reveal any difference between the conditions (Table 1E).

Higher metacognitive efficiency was observed in the cTBS compared to the sham TBS, but no difference was found between the iTBS compared to the sham TBS or the cTBS compared to the iTBS (Fig. 5, Table 2A).

We observed a significant interaction between cTBS and accuracy (i.e., correct and incorrect responses; Table 2B).

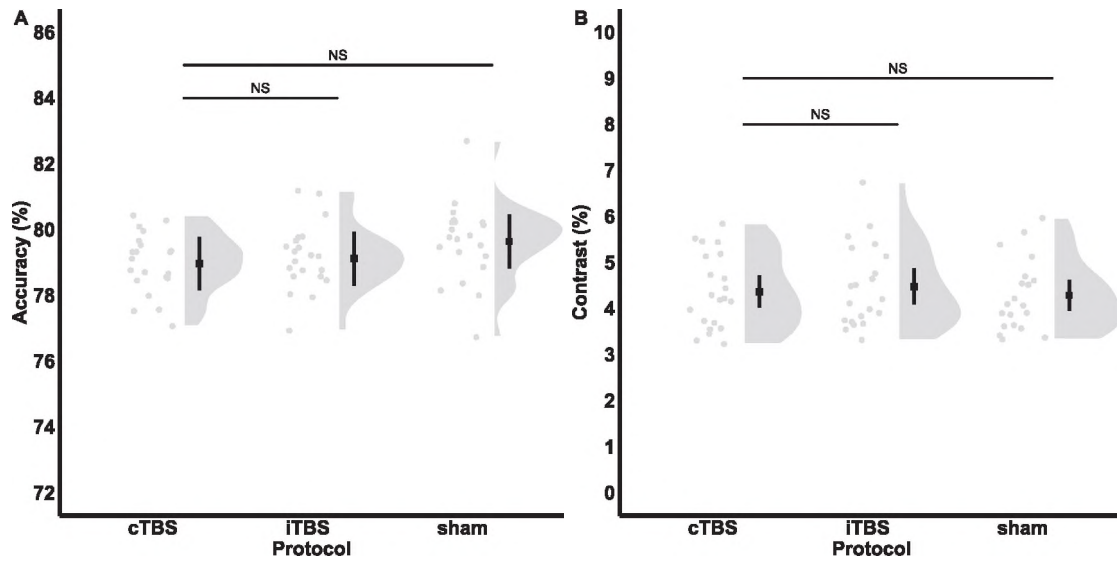


Fig. 3. A) Identification task accuracy (the percent of correct responses) depending on the TBS protocol. B) Stimulus contrast (the percent of computer screen maximum) depending on the TBS protocol. The grey drawings represent individual means and their distributions. The black squares represent estimated means, and the error bars represent 95% CI derived from the models.

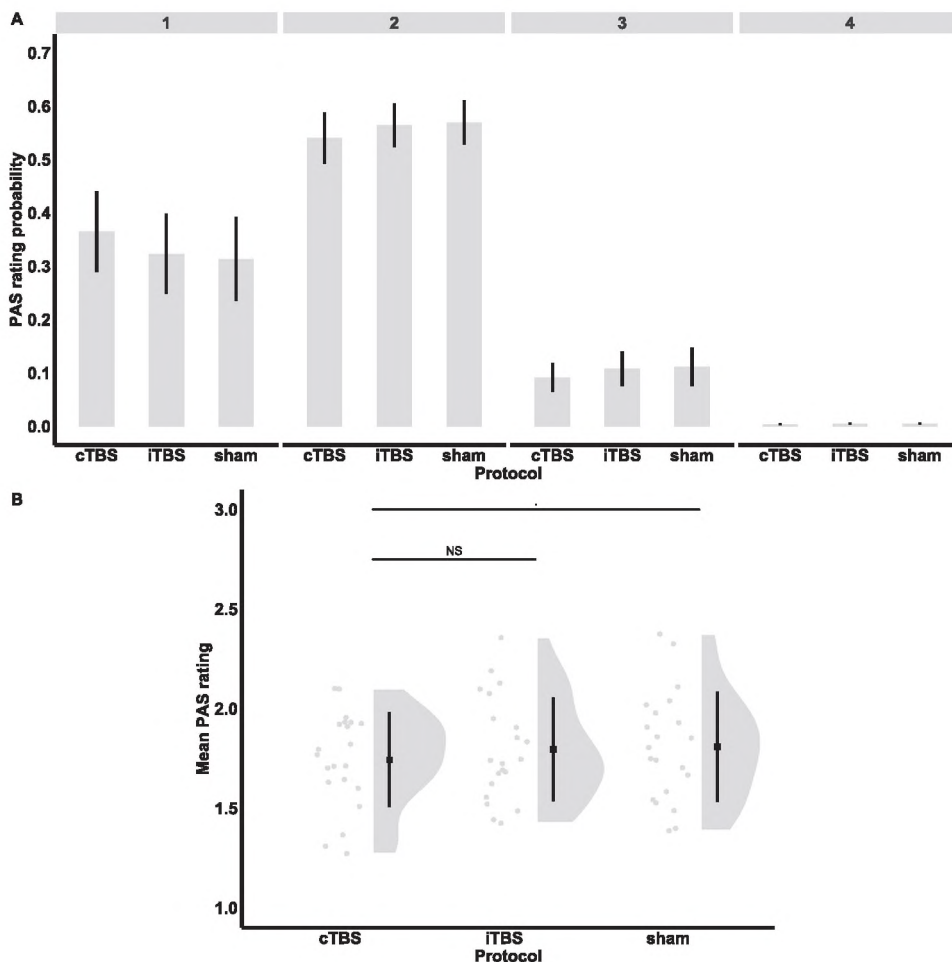


Fig. 4. A) PAS rating probability as depending on the TBS protocol. The grey bars represent estimated means and the error bars represent 95% CI derived from the model. B) Mean PAS rating depending on the TBS protocol. The black squares represent means, and the error bars represent \pm SD. Significance code: . $p < .1$.

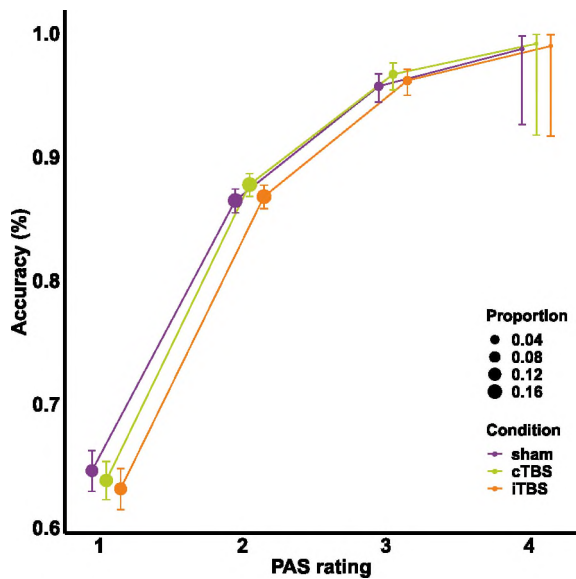


Fig. 5. Model fit for the relationship between decision accuracy and PAS ratings in each TBS condition. The position of filled circles represents the estimated average accuracy for each scale point. The bars represent 95% CI. The size of the dot describes the proportion of each PAS rating, taking into account the total number of trials.

Table 2

A) Results summary of the generalised linear mixed-effects model for the identification task accuracy with an interaction between the fixed effects of TBS condition and PAS rating and a random effect of PAS rating. B) Results summary of the cumulative link mixed-effects model for PAS rating with fixed and random effects of accuracy, TBS condition and their interaction. The basic accuracy condition was incorrect trials. Thus, the intercept concerns PAS rating in incorrect trials.

A Metacognitive efficiency				
Predictor	Estimate	Std. Error	z value	Pr (> z)
cTBS - sham	-0.03	0.05	-0.69	.488
iTBS - sham	-0.06	0.05	-1.26	.208
cTBS - iTBS	-0.03	0.05	0.60	.551
cTBS - sham:rating	0.15	0.06	2.38	.017*
iTBS - sham:rating	0.09	0.06	1.48	.139
cTBS - iTBS:rating	0.06	0.06	0.94	.350
B PAS rating: Interaction of TBS condition and identification task accuracy				
Predictor	Estimate	Std. Error	z value	Pr (> z)
cTBS - sham	-0.46	0.16	-2.93	.003**
iTBS - sham	-0.11	0.13	-0.80	.424
cTBS - iTBS	-0.35	0.18	-1.98	.048*
cTBS - sham:acc1	0.27	0.14	1.99	.047*
iTBS - sham:acc1	0.09	0.09	0.97	.332
cTBS - iTBS:acc1	0.18	0.14	1.28	.200

Significance code: * $p < .05$, ** $p < .01$.

Regarding the PAS ratings for incorrect responses, using pairwise comparisons we observed more conservative thus more accurate ratings in the cTBS condition as compared to the sham TBS ($z = -2.93, p = .040$; Fig. 6A and C), but not as compared to iTBS ($z = -1.98, p = .355$). No effect was observed in the correct responses ($z = -1.42, p = .715$ and $z = -1.30, p = .786$ respectively; Fig. 6B and D). Taken together, these findings thus indicate that cTBS resulted in a slight increase in metacognitive efficiency compared to sham TBS, and this effect was driven by lower visibility ratings in incorrect trials.

The results of M-ratio comparisons led us to inferences that were comparable to those based on the regression approach. When M-ratio

Table 3

A) Results summary of the M-ratio model fitted with the lmer function to estimate group-level differences. Linear mixed-effects model included TBS protocol as a fixed effect and intercept as a random effect. The M-ratios were calculated using Brian Maniscalco's code. Random effects ($\sigma^2 = 0.03, \tau_{00} = 0.05, N = 21, \text{Observations } 63$). B) Results summary of the M-ratio model fitted with the lmer function to estimate group-level differences. Linear mixed-effects model included TBS protocol as a fixed effect and intercept as a random effect. The M-ratios were calculated using the HMeta-d model. Random effects ($\sigma^2 = 0.02, \tau_{00} = 0.03, N = 21 \text{ ID}, \text{Observations } 63$).

M-ratio						
	Predictors	Estimate	Sdt. Error	df	t value	Pr (> t)
A	cTBS - sham	0.11	0.05	40.00	2.14	0.039*
	iTBS - sham	0.05	0.05	40.00	0.99	0.327
	cTBS - iTBS	0.06	0.05	40.00	1.15	0.259
B	cTBS - sham	0.09	0.05	40.00	1.83	0.075.
	iTBS - sham	0.07	0.05	40.00	1.34	0.187
	cTBS - iTBS	0.02	0.05	40.00	0.48	0.632

Significance code: . $p < .1$, * $p < .05$.

was estimated with Maniscalco's code (Maniscalco and Lau, 2012), M-ratio was higher in the cTBS condition than in the sham TBS condition ($p = .039$) and no statistically significant difference was found in the other comparisons between conditions (Fig. 7.A, Table 3.A). When M-ratio was estimated based on the HMeta-d model (Fleming, 2017), the difference between the cTBS and sham TBS conditions did not reach statistical significance ($p = .075$; Fig. 7.B, Table 3.B).

4. Discussion

We observed a higher metacognitive efficiency estimate in the cTBS condition as compared to the sham TBS condition, which suggests left aMPFC engagement in processes responsible for metacognitive efficiency. This result is further supported by the observed interaction between the TBS conditions and accuracy related to lower awareness ratings in the incorrect but not in the correct identification task responses in the cTBS condition compared to the sham TBS condition. No evidence was found for differences in the identification task performance, as indicated by the physical stimulus contrast and the identification task RT across TBS conditions. Also, no evidence was found for differences in the PAS RT across TBS conditions. Numerically, both active TBS protocols, when compared to the sham TBS, altered all measures (besides the PAS RT) in the same direction but with different strengths.

Observing an interaction between identification task accuracy and PAS ratings leads us to hypothesise that the detected cTBS effect is related to metacognitive judgement, which relies on performance monitoring. Importantly, the results do not indicate a change in the ability to rate awareness as higher in the correct responses in the cTBS condition. The observation that the cTBS effect on the PAS ratings was limited to incorrect responses suggests it might be attributed to improved error monitoring or integration of error-related information (including metacognitive judgements about the absence of particular stimuli).

Other potential interpretation of our results could be that disruption of particular top-down influences, e.g., disruption of the influence of expectations on perception, could lead to lower PAS ratings in the cTBS condition, specifically on incorrect trials, where bottom-up sensory processing is likely noisy. In this sense, cTBS might have reduced some illusory experiences of the stimulus that was not presented. Another interpretation could be that disruption of typical processing might assign a greater role to expectations. An example of such a case is when a participant expects a stimulus that is different than the one presented and the participant answers incorrectly. The participant might to some degree detect the error and thus rate awareness lower.

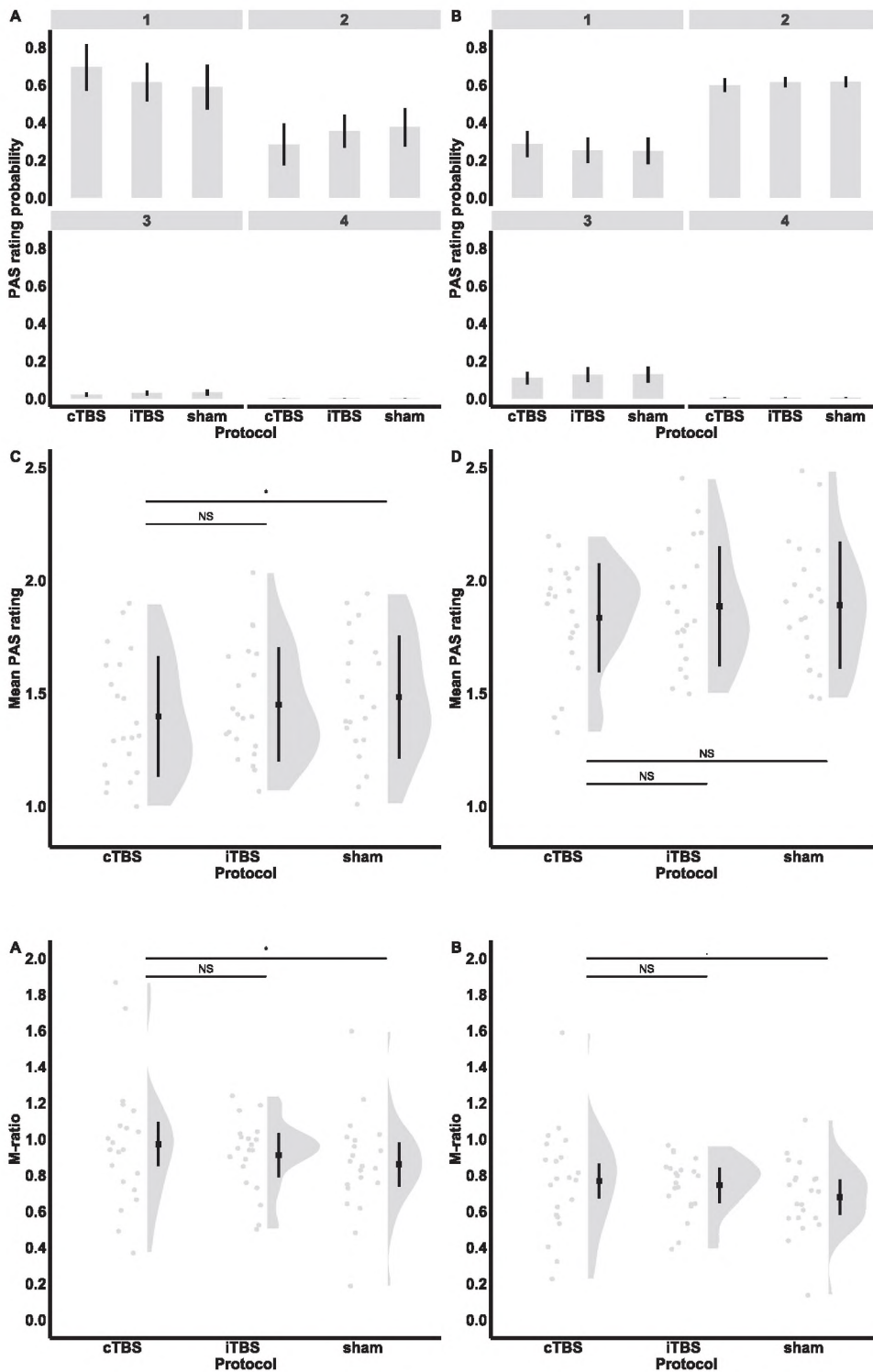


Fig. 6. A) PAS rating probability depending on the TBS protocol on the incorrect responses. The grey bars represent estimated means and the error bars represent 95% CI derived from the model. B) PAS rating probability, depending on the TBS protocol on the correct responses. The grey bars represent estimated means, and the error bars represent 95% CI derived from the model. C) Mean PAS rating depending on the TBS protocol on the incorrect responses. The black squares represent means, and the error bars represent \pm SD. D) Mean PAS rating, depending on the TBS protocol on the correct responses. The black squares represent means, and the error bars represent \pm SD. Significance code: * $p < .05$.

Fig. 7. A) M-ratio depending on the TBS protocol. The M-ratios were calculated using Brian Maniscalco's code. B) M-ratio depending on the TBS protocol. The M-ratios were calculated using the HMeta-d model. The grey drawings represent individual means and their distribution. The error bars represent estimated means with 95% CI derived from the models. Significance code: . $p < .1$, * $p < .05$.

Previous evidence shows that response accuracy influences metacognitive efficiency estimates even when not response confidence but visual awareness judgements are made (Siedlecka et al., 2020; Wokke et al., 2020). While the engagement of posterior brain areas might be necessary to gain awareness about events occurring in an environment, the PFC might particularly specialise in awareness of internal processes, such as the detection of errors, thus influencing awareness ratings. Observing the increase in metacognitive efficiency estimates not accompanied by an increase in PAS ratings in correct responses might suggest there is no unitary process responsible for metacognitive efficiency, but

metacognitive efficiency relies on multiple processes, one of which could be the error detection process, which might be influenced individually.

4.1. Comparison to previous research and theoretical considerations

Unlike previous studies, there could be a potential shift (yet not statistically significant) towards a higher stimulus contrast in the iTBS condition as compared to the sham TBS condition; this could occur because of modulation of low-level visual processing or the impairment of decision-making processing that is required in the identification task.

Although performance monitoring may affect response strategy and result in response bias, it cannot be excluded that performing identification tasks as such requires some form of metacognition. Our major result is consistent with that reported by [Rahnev et al. \(2016\)](#), who observed increased metacognitive ability after cTBS to the right aPFC as compared to the primary somatosensory cortex. In contrast to our result, [Rounis et al. \(2010\)](#) observed a decrease in metacognitive efficiency following cTBS to the dlPFC. The effect in the latter study was related to a lower reported level of visual awareness in correct rather than incorrect responses in the post-cTBS condition compared to the pre-cTBS condition (a comparison between the active cTBS and sham cTBS conditions was not reported). This is not in agreement with the results of [Rahnev et al. \(2016\)](#) study, where it was observed that cTBS to the right dlPFC increased metacognitive abilities. The most straightforward explanations of this discrepancy are that aPFC and dlPFC play qualitatively different roles in metacognitive processing, and/or their overall patterns of activity associated with metacognitive efficiency are different. The alternative explanation could be that cTBS to PFC decreases awareness ratings, but the experimental protocols used to date do not make it possible to detect the effect independently of the accuracy. This assumption is inconsistent with the lack of evidence for the cTBS influence on overall confidence ratings reported by [Bor et al. \(2017\)](#) and [Rahnev et al. \(2016\)](#); however, these studies used relatively small samples, i.e., fewer than 20 participants per condition, which is likely not enough to investigate group-level cTBS effects.

Regarding the inconsistency of the results of TBS studies, it is important to note that TBS after-effects may vary depending on the stimulated brain area ([Martin et al., 2006](#)), its prior pattern of activity ([Gentner et al., 2008](#)), and stimulation parameters, such as current direction ([Talelli et al., 2007](#)), intensity ([Bohning et al., 1999](#)), and protocol length ([Gamboa et al., 2010](#); [Gentner et al., 2008](#)), all of which were different in the mentioned studies, possibly leading to different cTBS outcomes at the neuronal level. Moreover, all cTBS studies included different objective tasks and subjective assessment scales. Using different types of metacognitive ratings can result in the investigation of phenomena with overlapping but distinct qualities (see e.g., [Overgaard and Sandberg, 2012](#)). While [Rahnev et al. \(2016\)](#) and [Bor et al. \(2017\)](#) collected response confidence ratings that are to some extent based on non-perceptual information, like action-specific feedback ([Fleming et al., 2015](#)) or error monitoring ([Yeung and Summerfield, 2014, 2012](#)), [Rounis et al. \(2010\)](#) used visual awareness ratings in their study. On the one hand, if an investigated effect concerns an impact on subjective visibility, awareness ratings measure this effect with more sensitivity than confidence ratings, which suggests that cTBS in previous studies could influence subjective visibility rather than confidence and thus could impact metacognitive ability estimates. On the other hand, cTBS in our study could lead participants to consider awareness judgement to resemble response confidence judgement, therefore the incorrect responses were more often accompanied by lower metacognitive ratings.

Based on the reported pattern of results, we consider it more probable that the cTBS effect on metacognitive efficiency estimates in our study stems from impairment in the ability to judge one's own visual experience, rather than subjective visibility impairment. However, it should be also noted that the observed TMS effects on metacognitive efficiency estimates in cTBS studies may be consequences of the direct or indirect influence of cTBS on processes that directly influence metacognitive judgement, such as criterion setting, overall confidence or impulsivity or other cognitive processes, including expectations, attention, working memory, retrospective memory, and self-related processing, which might in turn influence metacognitive ratings.

Research shows that PFC lesions may affect metacognitive efficiency when judging response confidence ([Fleming et al., 2014](#)), but there is also support for the presence of a cognitive control organisation gradient in PFC ([Azuar et al., 2014](#); [Badre et al., 2009](#)). [Rahnev and colleagues](#) proposed that the caudal, middle and rostral areas of PFC contribute

differently to perceptual decision-making, which reflects on the progressively later stages of this process, i.e., selection, criterion setting, and evaluation, respectively ([Rahnev et al., 2016](#); [Shekhar and Rahnev, 2018](#)). It is assumed that dlPFC evaluates how much information is available for stimulus identification decisions when a participant performs the objective task, while aPFC plays an integrative role and its activity increases along with the reliability of participant's confidence judgements ([Morales et al., 2018](#); [Fleming et al., 2012](#); [Yokoyama et al., 2010](#)).

The lower awareness ratings might be interpreted as decreased conscious access to lower levels of perceptual processing; however, in principle they could also be related to a change in participants' definition of what constitutes a particular level of perceptual awareness (i.e., depend on an introspective decision criterion rather than a perceptual change) and/or relate to the overall confidence level. Although neuronal representation of confidence in the perceptual decision has been linked to the ventral striatum ([Hebart et al., 2016](#)), dlPFC activity correlates with reported confidence ([Fleck et al., 2006](#); [Morales et al., 2018](#)).

Targeting the dlPFC with TMS has been reported to influence the level of confidence and/or metacognitive efficiency ([Chiang et al., 2014](#); [Rahnev et al., 2016](#); [Rounis et al., 2010](#); [Shekhar and Rahnev, 2018](#)). In a study by [Shekhar and Rahnev \(2018\)](#), single-pulse TMS to the dlPFC decreased reported confidence, whereas when applied to the aPFC it increased metacognitive efficiency (these effects were observed for the second half of the experimental trials only). Therefore, it might be that the effects of cTBS on visual awareness ratings in [Rounis et al. \(2010\)](#) and our study stem from influences on confidence, which played a role in the observed cTBS effects on metacognitive efficiency estimates via lowering the metacognitive ratings in either correct or incorrect trials. Future studies should address whether the effects rely on changes in metacognitive efficiency or the use of more conservative criteria (metacognitive bias).

Theoretically, a difference in visual awareness or in the criteria for reporting certain levels of awareness may result in a difference being observed in metacognitive efficiency measures ([Rausch and Zehetleitner, 2017](#)). For example, in our study it could be a shift in the criterion for reporting a brief glimpse of a stimulus, resulting in lower PAS ratings (because the cTBS effect on PAS ratings was limited to incorrect trials, and low visibility experience accompanies the incorrect responses more than the correct responses). While we do not exclude that criteria shifts may be reasons for reporting lower metacognitive ability in [Rounis et al. \(2010\)](#) and/or observing the cTBS effect on metacognitive efficiency in our study, this explanation might be less likely than the change in metacognitive efficiency. An argument for the latter is that neither [Bor et al. \(2017\)](#) nor [Rahnev et al. \(2016\)](#), who both employed cTBS, found an effect on confidence ratings, whereas an impact on metacognitive efficiency was found in both [Rounis et al. \(2010\)](#) and [Rahnev et al. \(2016\)](#) despite the different types of the metacognitive judgements used. We further partly supported our conclusion by presenting the results of analyses of M-ratio, which is considered to be one of the most bias-independent methods for measuring metacognition ([Fleming et al., 2014](#)).

Since unilateral PFC lesions can cause top-down attention and memory deficits, another issue concerns disentangling perceptual awareness and metacognition from these processes ([Voytek et al., 2010](#)). Attention shares neural underpinnings with processes often defined as metacognition or awareness ([Fernandez-Duque et al., 2000](#); for the review, see [Lamme, 2020](#)). On the one hand, participants in the studies of [Rounis et al. \(2010\)](#) and [Bor et al. \(2017\)](#) had to attend selectively to one of two peripheral stimuli, therefore the effects of dlPFC stimulation might have been associated with a disturbance of attention that mediated the influence on metacognitive efficacy. Moreover, due to the adjacent location of the dlPFC and the aPFC and the anatomical and functional connections between these areas ([Azuar et al., 2014](#); [Badre et al., 2009](#)), some additional direct or indirect TMS influences might be considered. At the same time, it is known that cTBS to dlPFC can influ-

ence working memory (Schickanz et al., 2015; Vékony et al., 2018). Given the fact that the identification decision and visual awareness judgement were provided simultaneously and were included a relative judgement in Rounis et al. (2010) study (and for this reason, required a high level of working memory resources), it may be hypothesised that cTBS decreased working memory capacity rather than metacognitive efficiency. However, in our study, stimuli were presented singly in the centre of the screen, and objective and subjective responses were separated; therefore, we can consider this explanation less likely. While our study attempted to account for individual differences in seeing left- and right-orientated Gabor patches, we cannot exclude that including separate staircases for both Gabor patches impacted response strategies. In sum, we observed an effect that is consistent with some of the previous research; however, because of the issues discussed above, the results of studies investigating the impact of TBS to PFC on visual awareness should be interpreted with caution and followed up in larger samples.

4.2. Implications for theories of consciousness

One might infer that the results of our study provide support for the Higher-Order Thought Theory (Lau and Rosenthal, 2011) or the Global Neuronal Workspace Theory (Dehaene and Naccache, 2001; Dehaene, 2014), but our results do not support theories such as the Recurrent Processing Theory, which does not hold the assumption that NCC includes PFC (Lamme, 2020), or the Integrated Information Theory, which argues that PFC connectivity patterns are not suited to integrating information and thus are not important for awareness formation (Tononi et al., 2016). However, we are reluctant to make such far-reaching conclusions. The observation that PFC activity influences metacognitive efficiency does not imply that PFC is necessary for certain conscious content to occur. A recent synthesis of research analysing outcomes of studies in which intracranial electrical stimulation to the PFC was employed suggests no evidence for reliable alternations in perceptual awareness following PFC stimulation (Raccach et al., 2021). This is inconsistent with the predictions of some higher-order theories and some developments of Global Workspace Theory. On the one hand, this does not imply that stimulation to PFC cannot influence perceptual awareness in a way that is not noticeable or reportable by participants or can be observed only under specific task conditions (for the commentary articles to Raccach et al., 2021, see Baars et al., 2021 and Naccache et al., 2021). On the other hand, participants' ability to differentiate spontaneous fluctuations in conscious experience from other effects elicited by neuromodulation techniques could limit the reliability of some brain stimulation research (Fox and Parvizi, 2021).

The conflicting conclusions concerning the role of PFC in consciousness found in the scientific literature might stem from different understandings of the terms 'visual consciousness' or 'visual awareness', e.g., whether confidence or working memory-related processes are assumed to constitute visual awareness. Although perceptual content has been decoded above chance level from PFC activity in various experiments (for a review, see Odegaard et al., 2017), awareness-related PFC activity might not represent any specific perceptual content. The PFC's role in consciousness might be limited to processes that influence overall conscious experience or to stimulus awareness judgement. Currently, no method allows response criteria or overall confidence to be disentangled from stimulus awareness itself. Thus, we cannot exclude that differences observed in awareness ratings are caused by judgement bias. However, the confidence with which we perceive a stimulus is often considered to be a component of the conscious experience of this stimulus. Some researchers argue that representations in the global workspace always carry with them an estimate of confidence (Kouider et al., 2010; Shea and Frith, 2019) or involve a feeling of knowing (Baars et al., 2021). Thus, confidence might be treated as a component of visual awareness (Lau and Rosenthal, 2011). Currently, explaining the relationship between perceptual metacognition and perceptual awareness

is considered one of the key goals for the field of visual metacognition research (Rahnev et al., 2021).

4.3. Methodological considerations and future directions

Even though we demonstrated that cTBS to the aMPFC increases estimates of metacognitive efficiency, which supports the role of the PFC in the assessment of subjective experience, there is not yet conclusive evidence that this effect stems from a change in PFC excitability. There are two probable but overlooked sources of confounds in TMS studies that investigate near-threshold perception. The first is peripheral nerve stimulation, which might influence vigilance and in consequence influence factors related to metacognitive judgement. The second source is the potential change in retinal activity, which might influence visual perception. An alternative explanation for lower PAS ratings in incorrect responses in the cTBS condition is that stimulation to the PFC results in an electric current passing through the eyeballs, thus influencing retinal activity and, in consequence, impairing visual processing. This might not be specific to aPFC stimulation; for example, Webster & Ro (2017) suggest that phosphenes (simple visual sensations, often in the form of light spots) that are perceived as a result of TMS to areas as far from the retina as the vertex or parietal cortex may arise from retinal stimulation. However, if influencing vigilance level or retinal stimulation were the case, likely it would cause a difference in identification task performance, but we did not observe this in the cTBS or the sham comparison. Nonetheless, objective tasks are less sensitive to detecting subtle changes in visual awareness compared to subjective awareness ratings (Sandberg et al., 2010), and performing an objective task is more automatic than providing subjective assessment, thus it might not require the engagement of the aPFC. Nevertheless, in this context, it is worth noting that the psychometric function for PAS has a shallower slope than that for accuracy (Sandberg et al., 2011), which suggests that identification task performance should be more affected than PAS ratings by the same external influence on visual processing.

It is important to note that cTBS was never significantly different from iTBS in all analyses. The reason for this could be related to suboptimal iTBS parameters, iTBS inefficacy in a particular area, or participants' individual differences, which affected the overall iTBS efficacy. Previous research that included multiple measures of cognitive functions reported only a minor impact of iTBS to PFC areas (Grossheinrich et al., 2009). Another explanation is that iTBS's impact on the discrimination task was high enough to balance the potential iTBS impact on PAS ratings by (insignificantly from the statistical perspective) increasing the contrast of presented stimuli. Also, similarly to some previous TBS studies, we do not find support for conceiving cTBS and iTBS as protocols that always have the opposite behavioural consequences (Grossheinrich et al., 2009; Hamada et al., 2013; Viejo-Sobera et al., 2017). This is in line with evidence that the typical excitatory and inhibitory outcomes of different TBS protocols applied to M1 might not be transferable to PFC. While some studies have provided evidence for the efficiency of cTBS to PFC in modulating cognitive performance, a couple of studies involving both cTBS and iTBS to PFC showed no differences between cTBS and iTBS effects, or they observed differences only in certain tasks (for the meta-analysis of PFC TBS influence on executive functions, see Lowe et al., 2018). Molenberghs et al. (2016) observations that metacognitive ability was inversely related to aMPFC activity, and that cTBS in our study led to higher metacognitive efficiency estimates, point towards the overall inhibitory influence of cTBS in our study. Future studies employing cTBS to influence awareness could combine it with neuroimaging to examine cTBS effects at the brain level.

Future research might disentangle the extent of PFC contribution to the content of consciousness from its contribution to the level of wakefulness; it might also clarify the role of different subcomponents of PFC in shaping awareness judgement. Especially, there is a need to distinguish between the correlates of content directly related to stimulus, non-perceptual content, and post-perceptual content of awareness,

as well as to consider potential PFC involvement in decisions about the absence of stimulus (Anzulewicz et al., 2019; Fleming, 2020). These additional steps could focus on dissociating stimulus awareness from other cognitive processes, and on determining to what degree metacognitive processes are lateralized. Future studies may also address the problem of whether TMS primarily affects confidence or visual awareness by employing both measures at once and/or including above-threshold stimuli combined with confidence ratings. There is also a need to clarify whether alterations in metacognitive efficiency accompanying brain stimulation to aMPFC does not result from the direct (due to the spread of electric field) or indirect (via network effects) influence on the activity of other brain areas, like posterior medial frontal cortex, which is likely involved in error monitoring (Dehaene et al., 1994; Gehring et al., 1993). Since MPFC receives visual information from the superior temporal cortex (Kondo, Saleem & Price, 2003, 2005), it also seems compelling to investigate communication between PFC and the temporal lobe.

Importantly, the issue concerning the difference between metacognition and awareness is closely related to how conscious experience is operationalised within different theories of consciousness. It may further be noted that the observed effect sizes were not large, thus caution is required when interpreting the results. Considering the differences in findings across all studies to date on metacognition that included TBS to PFC, PFC engagement in perceptual awareness needs further investigation, and we look forward to future attempts to investigate this issue with the use of better stimulation techniques, larger samples, and behavioural paradigms allowing better differentiation between metacognitive efficiency and visual awareness to address the concerns that have been raised in this article.

5. Conclusion

Summing up, our study indicates that the left aMPFC is involved in processes related to metacognitive efficiency, but its involvement in the overall level of reported visual awareness remains inconclusive. While the results of our study specifically support the claim that PFC activity affects the assessment of visual awareness, it does not explicitly support or exclude the possibility that PFC is necessary for the conscious experience of stimulus. The cTBS effect was related to a decrease of awareness in incorrect trials, which points towards an effect on metacognition effect rather than stimulus awareness. Additionally, we did not find evidence supporting the hypothesis that cTBS and iTBS exert opposite effects.

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Declaration of Competing Interest

none

Credit authorship contribution statement

Justyna Hobot: Conceptualization, Methodology, Software, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Visualization, Project administration. **Zuzanna Skóra:** Methodology, Software, Resources, Writing – review & editing. **Michał Wierchoń:** Conceptualization, Methodology, Resources, Writing – review & editing, Supervision, Project administration, Funding acquisition. **Kristian Sandberg:** Conceptualization, Methodology, Resources, Writing – review & editing, Supervision, Project administration, Funding acquisition.

Data availability

The data and scripts for running the statistical analysis are publicly available at www.osf.io/3yb2g.

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Co-authorship statement

As a co-author of the publication: **Continuous Theta Burst Stimulation to the left anterior medial prefrontal cortex influences metacognitive efficiency**, *NeuroImage*, **272, 2023, 119991**, I declare that my own substantial contribution to this publication consists of:

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Krakow, 13.04.23

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As a co-author of the publication: **Continuous Theta Burst Stimulation to the left anterior medial prefrontal cortex influences metacognitive efficiency, NeuroImage, 272, 2023, 119991**, I declare that my own substantial contribution to this publication consists of:

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4.3. Article 3 with co-authorship declarations

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Transcranial Magnetic Stimulation-Induced Motor Cortex Activity Influences Visual Awareness Judgments

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The influence of non-visual information on visual awareness judgments has recently gained substantial interest. Using single-pulse transcranial magnetic stimulation (TMS), we investigate the potential contribution of evidence from the motor system to judgment of visual awareness. We hypothesized that TMS-induced activity in the primary motor cortex (M1) would increase reported visual awareness as compared to the control condition. Additionally, we investigated whether TMS-induced motor-evoked potential (MEP) could measure accumulated evidence for stimulus perception. Following stimulus presentation and TMS, participants first rated their visual awareness verbally using the Perceptual Awareness Scale (PAS), after which they responded manually to a Gabor orientation identification task. Delivering TMS to M1 resulted in higher average awareness ratings as compared to the control condition, in both correct and incorrect identification task response trials, when the hand with which participants responded was contralateral to the stimulated hemisphere (TMS-response-congruent trials). This effect was accompanied by longer PAS response times (RTs), irrespective of the congruence between TMS and identification response. Moreover, longer identification RTs were observed in TMS-response-congruent trials in the M1 condition as compared to the control condition. Additionally, the amplitudes of MEPs were related to the awareness ratings when response congruence was taken into account. We argue that MEP can serve as an indirect measure of evidence accumulated for stimulus perception and that longer PAS RTs and higher amplitudes of MEPs in the M1 condition reflect integration of additional evidence with visual awareness judgment. In conclusion, we advocate that motor activity influences perceptual awareness judgments.

Keywords: awareness scale, identification task, motor cortex, motor-evoked potential, transcranial magnetic stimulation, visual perception

Abbreviations: 2AFC, two-alternative forced choice; dlPFC, dorsolateral prefrontal cortex; FDI, first dorsal interosseous; M1, primary motor cortex; MEP, motor-evoked potential; MSO, maximal stimulator output; PAS, Perceptual Awareness Scale; PFC, prefrontal cortex; PMd, dorsal premotor cortex; PPC, posterior parietal cortex; RMT, resting motor threshold; RTs, response times; S1, primary somatosensory cortex; SD, standard deviation; SEs, standard errors; TMS, transcranial magnetic stimulation.

INTRODUCTION

Uncovering the neural processes that shape conscious content is considered a central problem in consciousness science (Faivre et al., 2017). Access to conscious content is based on the accumulation of stimulus-based evidence, prior knowledge, and biases (Dehaene, 2008; Lau, 2008; Dehaene and Changeux, 2011; Overgaard, 2018). We consider conscious access to be a non-dichotomous phenomenon (Overgaard et al., 2006; Kouider et al., 2010; Fazekas and Overgaard, 2016, 2018a; Jonkisz et al., 2017; Lyyra, 2019, for alternative explanations see: Sergent and Dehaene, 2004; Del Cul et al., 2007) that is reflected in awareness judgments (Overgaard et al., 2006; Anzulewicz et al., 2019). Therefore, we operationalize conscious access with perceptual awareness ratings. This approach is consistent with several major views on consciousness, including the hierarchical view (Lau and Rosenthal, 2011), the Partial Awareness Hypothesis (Kouider et al., 2010), and some current understandings of conscious access, e.g., the Multi-Factor Account of Degrees of Awareness (Fazekas and Overgaard, 2016, 2018a). Perceptual awareness judgments—like decision confidence judgments—are a type of metacognitive judgment (Lau and Rosenthal, 2011; Fleming, 2020) and can be measured on multiple scales, such as continuous visual analog scales (Hayes and Patterson, 1921; Sergent and Dehaene, 2004) and the PAS (Ramsøy and Overgaard, 2004). The latter requires participants to rate stimulus awareness with ratings ranging between “no experience” and “a clear experience.” PAS is considered a sensitive and exhaustive measure of stimulus awareness (Sandberg et al., 2010) and is widely used in consciousness research (Sandberg and Overgaard, 2015).

Multiple theories frame conscious access (more or less explicitly) in the context of stimulus evidence accumulation (Dehaene et al., 2003; Dehaene, 2008; Lamme, 2010; Block, 2011; Mudrik et al., 2016). This has bound research to experimental paradigms that manipulate stimuli characteristics; however, the physical qualities of stimuli do not fully explain the qualities of conscious access, which implies the presence of additional sources of evidence (Anzulewicz and Wierzchoń, 2018; Tagliabue et al., 2019). Several such sources have been proposed, e.g., prior expectations (Snyder et al., 2015), previous responses (Rahnev et al., 2015), or attentional engagement (Fazekas and Overgaard, 2018b). Nevertheless, these sources are associated with the early stages of awareness-related processing (e.g., Dehaene et al., 2014). Here, we present an investigation of motor-related information influence that occurs at the later stages of awareness-related processing on stimulus awareness judgment.

Out of many possible contributors, the motor system seems especially related to perception. Numerous studies have explored the action–perception loop and have shown that in tasks requiring coordination of perceptual information and action, both systems influence each other and enhance task performance (Hecht et al., 2001; Donnarumma et al., 2017). Similar conclusions have come from experiments in which coupling between perception and action was more superficial than in action–perception loop procedures (e.g., linking certain stimuli to particular response keys: Siedlecka et al., 2019, 2020a). A recent study showed that visual awareness judgments are

sensitive to accuracy feedback in a stimulus identification task (Siedlecka et al., 2020b). Participants reported lower awareness after an incorrect response in the previous trial, and the effect was strengthened by trial-by-trial accuracy feedback.

Nevertheless, a couple of studies have presented a more immediate effect that shapes the experience of just-presented stimuli. Several studies have shown a consistent effect of identification tasks and rating response order on the association between metacognitive ratings and identification task accuracy (Wierzchoń et al., 2014; Siedlecka et al., 2016; Wokke et al., 2020). These studies' authors suggested that carrying out a behavioral response acts as an additional source of evidence for metacognitive judgments. Following this, Anzulewicz et al. (2019) listed four possible mechanisms through which action planning or execution could influence reported awareness. They point to possible (1) indirect effects that stem from motor cortex activity that affects cognitive processing, (2) perceptual evidence accumulation being influenced by attentional engagement, (3) enhancement of performance monitoring, and (4) integration of additional (including post-perceptual) evidence with the evidence accumulation process.

It has been shown that the evidence accumulation process is strongly coupled with the presence of perceptual stimulation, but it continues even after its disappearance and might persist after stimulus-related decision to inform metacognitive judgments (Yeung and Summerfield, 2012; Murphy et al., 2015; Navajas et al., 2016; Fleming and Daw, 2017; Wokke et al., 2020); this is what we refer to as post-perceptual evidence accumulation. In the current study, unlike in speed 2AFC tasks oftentimes used to measure perceptual evidence accumulation, we focus on post-perceptual evidence accumulation for formation of metacognitive judgment. The hypothesis that post-perceptual information can concurrently influence metacognitive judgment is supported by Gajdos et al. (2019). The authors showed that higher confidence ratings were observed in trials in which an identification response to a stimulus was preceded by partial muscular activation. They argued that such muscle activity could contribute to participants' confidence in their identification response to a stimulus, but it could not influence the identification task itself. However, these studies do not provide sufficient evidence to prove that partial muscular activations influence confidence judgment and not the opposite.

This issue of causal relation can be partially resolved by experimentally introducing additional M1 activity that is unrelated to the main task. In Siedlecka et al. (2019), this was achieved by including an irrelevant task that participants performed between stimulus presentation and PAS rating. Performing an additional motor response congruent with the response scheme of the identification task led to higher awareness ratings than when performing an incongruent one. At the same time, the congruence between the additional motor response and the identification task response was not related. Siedlecka et al.'s experiment provides arguments for the influence of motor system activity on visual awareness judgment. Although it cannot be ruled out that in this task the additional response itself or the visual information from the additional task cue were responsible for the observed effect, two other recent studies show an effect of

action on confidence ratings. Faivre et al. (2020) have shown that sensorimotor conflicts might decrease metacognitive efficiency (a quantitative measure of participants' level of metacognitive ability, given a certain level of 2AFC task performance), and Filevich et al. (2020) have presented that continuous report paired to stimulus presentation leads to higher confidence ratings; however, there was no evidence that absolute confidence judgments or metacognitive efficiency varies with the presence or absence of overt responses.

Assessment of the selective effect of motor information on visual awareness judgments requires directly altering motor cortex activity. Fleming et al. (2015) attempted this by applying single-pulse TMS either before or after a 2AFC task followed by decision confidence rating. They showed that TMS applied to the PMd that was associated with the chosen response was associated with higher response confidence and consequently higher metacognitive efficiency than TMS associated with the unchosen response, while no evidence for the influence of TMS on identification accuracy was found. The TMS effect on mean confidence rating was observed for TMS applied both before and after the identification response, thus suggesting the contribution of post-decision processes to confidence in one's identification decision. None of these effects was observed for TMS applied to M1. Fleming et al. suggested that PMd but not M1 activity contributes to confidence ratings. However, both TMS intensity and the number of participants taking part in the experiment were relatively low, thus encouraging the collection of more evidence on this matter.

Considering the limitations of the previous research, we investigated whether externally introduced motor-related information can be integrated into judgment of visual awareness. To achieve this, we delivered twitch-causing TMS to an M1 representation of the index finger involved in providing identification responses to mimic the influence of identification response on metacognitive judgment. Moreover, we used verbally reported PAS to separate the TMS and the identification response to minimize TMS-induced motor activity's interference with the activity that resulted from the intentional identification task decision. Based on Fleming et al.'s (2015) results, we did not expect to observe the influence of TMS on identification decision performance, including its RT. Based on Siedlecka et al.'s (2019) findings, we expected to observe higher awareness ratings in the M1 condition compared to the control condition (TMS to the interhemispheric cleft). In addition, we calculated response-specific metacognitive efficiency measures. Since in our experiment, the scale response preceded the identification response and we asked for perceptual awareness judgments (not confidence judgments) in identification task decisions, we did not expect to observe any difference in metacognitive efficiency between M1 and the control TMS condition.

To actively monitor the precision of TMS delivery, we recorded MEP amplitudes on the response finger that was contralateral to the stimulation side. It has been established that imagined unilateral movements increase the excitability of contralateral M1 (Jeannerod, 1995; Facchini et al., 2002; Fourkas et al., 2006). Previous research on MEP has shown that its amplitude can reflect the level of M1 excitability

(Fitzgerald et al., 2002). For these reasons, we expected M1 excitability to be influenced by the preparatory motor plan for the subsequent identification response proportionally to the accumulated evidence for the identification decision. This should lead to a correlation between MEP amplitudes and PAS ratings as well as a correlation between PAS ratings and their RTs, thus representing accumulated evidence for visual awareness judgment.

MATERIALS AND METHODS

The experiment was carried out in the TMS Laboratory at the Neurology Clinic of Jagiellonian University Hospital. The study was approved by the Ethics Committee of the Institute of Psychology at Jagiellonian University and was carried out in accordance with the guidelines for TMS research (Rossi et al., 2009; Rossini et al., 2015) and the Declaration of Helsinki (Holm, 2019).

Participants

Healthy volunteers meeting the criteria for participation in TMS studies (no history of neurological disorders, psychiatric disorders, head injury, etc., as assessed by a safety screening questionnaire) and with normal or corrected-to-normal vision were recruited using advertisements on social media. One participant dropped out due to TMS-induced headache, while 46 participants (one reported left-handedness, 11 males, 35 females, $M_{\text{age}} = 23.2$, range = 19–37) completed the study. The general purpose of the experiment was explained to participants, and they were informed that they could withdraw at any time without giving a reason. Prior to the experiment, the participants completed safety screening questionnaires and signed informed consent forms. After the experiment, they received monetary compensation (160 PLN).

Session Overview

The experiment was conducted using a within-participant design in a single session. Participants practiced (15 trials, ~2 min) the procedure, with the identification task preceding the PAS rating within each trial (Ramsøy and Overgaard, 2004; Sandberg et al., 2010). Then a 1-up-3-down staircase was used to estimate the stimulus contrast (100 trials; step sizes from 0.9 to 0.5%, starting with 12% of the maximal contrast of the monitor) that would lead to approximately 79% correct responses. The median stimulus contrasts for each PAS rating were calculated based on all trials acquired in the staircase procedure (~5 min) for use in the following experimental procedure, in which four fixed contrasts were used in a random manner and with equal probability (PAS1: mean = 10.22%, SD = 1.90; PAS2: mean = 10.65%, SD = 1.87; PAS3: mean = 11.21%, SD = 1.93; PAS4: mean = 12.29%, SD = 1.96). The same contrasts were used for M1 and control conditions.

Subsequently, individual RMTs for TMS were determined, and participants completed a 32-trial training session that was identical to the experimental procedure: TMS pulses were applied to the left M1, and the PAS rating was followed by

the identification task response. Finally, they completed the experimental task, which consisted of two conditions in four counterbalanced blocks (two blocks of TMS to M1, and two blocks of TMS to interhemispheric cleft, alternately). Each block consisted of 100 trials, which summed up to 400 trials that took about 45 min to complete.

Stimuli and Procedure

The task was coded in PsychoPy software (Peirce, 2007) and was run on a PC. Participants placed their heads on a chinrest, 60 cm away from an LCD monitor (1920 × 1080 resolution, 60 Hz refresh rate). A microphone was attached to the chinrest for the purpose of PAS verbal responses recording. In the experimental task at first, a fixation dot was presented for 500 ms. A Gabor patch masked with white noise was then displayed for 33 ms. Gabor patches were tilted left or right (−45° or 45° of rotation from vertical angle, respectively; 128 × 128 pixels, which translated to ~3° of visual angle, spatial frequency ~4 cycles per degree, embedded in the same size white noise against the gray background) presented centrally on the screen. A white noise patch of constant contrast was presented with the stimulus to reduce its visibility. Stimulus presentation was followed by an empty screen displayed for 450 ms. Subsequently (i.e., 483 ms from the stimulus onset), a TMS pulse was administered and a screen prompting the PAS rating (with the points of the scale defined as 0 = no experience; 1 = a brief glimpse; 2 = an almost clear experience; 3 = a clear experience) was displayed for 3 s dedicated to provide a verbal response. Irrespective of whether a verbal response was provided or not, PAS was followed by a screen prompting a behavioral response to the identification task that was displayed until a keyboard button was pressed up to 3 s (either “Z” with the left index finger or “M” with the right one). Participants did not receive any feedback about their performance. **Figure 1** outlines the temporal organization of an experimental trial.

TMS Parameters

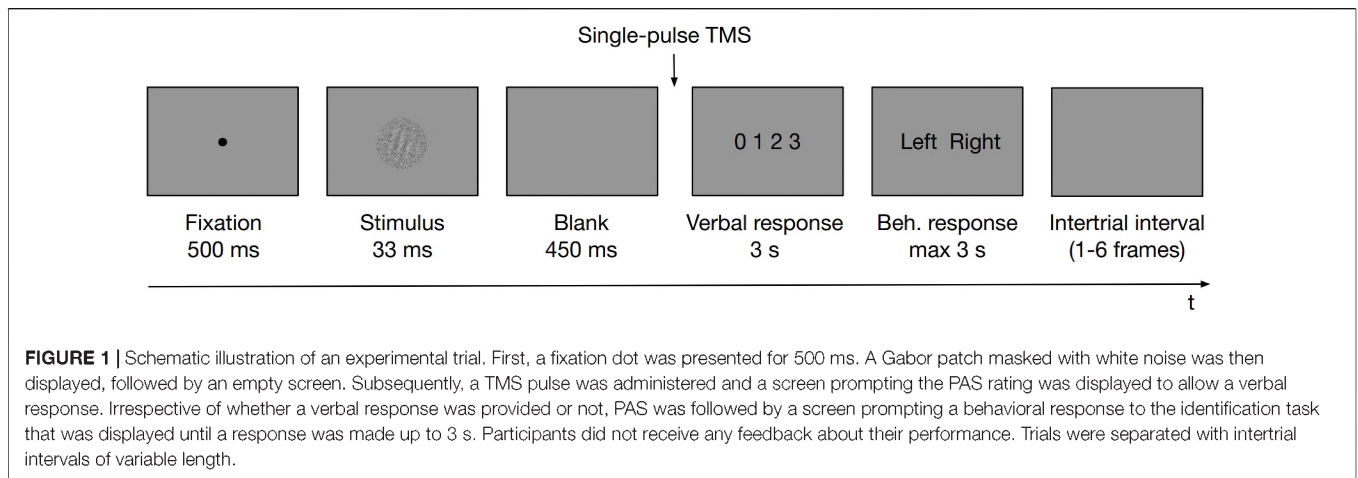
Biphasic TMS was delivered with a Magstim Super Rapid² Plus¹ stimulator using a 70 mm Double Air Film Coil at 110% of the individual RMT (average intensity = 65.87% of the MSO, SD = 10.67). The electromyographic signal was recorded from the FDI muscle of the right index finger throughout the whole experimental procedure. The individual RMT estimation started with applying TMS at 50% of MSO to the left M1. Then, by varying the stimulation intensity, the site where suprathreshold TMS induced the maximal twitch in the right index FDI muscle was established. Afterward, the lowest intensity that resulted in an MEP of more than 50 μV peak-to-peak amplitude in five out of 10 consecutive trials was determined. In the control condition, TMS was applied to the interhemispheric cleft between the superior parietal lobules, with the coil handle pointing backward. The site of stimulation and the tangential position of the coil in relation to the scalp were monitored using the average brain template in theBrainsight 2.3 neuronavigation system. For the M1 stimulation, the main axis of the coil was oriented at 45° offset from the posterior–anterior (PA) direction, but it remained untilted for the control condition. The current induced in the brain was PA–AP.

Participants wore earplugs for noise protection throughout the duration of TMS.

Data Analysis

No statistical analyses were conducted before the completion of the experiment and no participants who completed the experiment were excluded from the analysis. Trials with no PAS response and identification response were removed; the remaining data (17,969 trials, 97.7%) were analyzed using the R statistical environment (R Core Team, 2019). We used mixed-effects regression models fitted with the lme4 package (Bates et al., 2015) and compared the models of different complexity with ANOVA to determine which models describe the data the most accurately. The R notations of the models presented in the results section can be found in the data analysis scripts¹. To obtain approximate *p*-values via Satterthwaite’s method, we used the lmerTest package (Kuznetsova et al., 2017). Additionally, we used the phia (De Rosario-Martinez, 2015) and emmeans packages (Lenth, 2019) for pairwise comparisons, employing Tukey’s method for family-wise error rate correction. We used code provided in an implementation of response-specific meta-*d*’ (Maniscalco and Lau, 2014) to calculate (1) identification task sensitivity index *d*’, (2) type 1 criterion indicating identification response bias, and (3) M-ratio (meta-*d*’/*d*’), which is a measure of metacognitive efficiency in which metacognitive sensitivity (operationalized with meta-*d*’) is corrected for objective task sensitivity (operationalized with *d*’; Fleming and Lau, 2014). The M-ratio indicates the amount of evidence available for metacognitive judgment relative to the amount of evidence available for objective decision, e.g., the M-ratio value of 0.8 indicates that 20% of the sensory evidence available for the objective decision is lost when making metacognitive judgments, while M-ratio value of 1.2 suggests that more evidence is available for metacognitive judgments than for objective decision that can be due to further processing of stimulus information or gaining non-perceptual information (Fleming, 2017). For MEP-related calculations, for every trial, the highest peak-to-peak amplitude was determined in the 75 ms after the TMS pulse, irrespective of the condition. We intended to present results from the full dataset, therefore we did not limit analysis to trials in which the MEP amplitude exceeded 50 μV, as is commonly done (Anderson and George, 2009). In order to convert verbal recordings with PAS ratings into a machine-readable format, we used Python’s Speech Recognition package (Zhang, 2017); we calculated speech onset with Chronset and trials for which the algorithm failed were corrected manually. RTs were measured either from the PAS screen or the onset of the identification task response cue. We use congruence between TMS and identification response as a fixed factor. Although no TMS-induced movement was present in the control condition, we used congruence to refer to right-hand responses. Because TMS was limited to the left hemisphere, all responses provided with the right index finger were TMS-response congruent (congruent trials, *n* = 8,933), while all those provided with the left index finger were TMS-response

¹<https://osf.io/29n6j>



incongruent (incongruent trials, $n = 9,036$). We used non-directional tests with α level set at 5%.

RESULTS

Identification Task

Identification task accuracy data were analyzed using a logistic mixed-effects regression model with condition and congruence as fixed effects. All effects were taken as random at the participant level. As expected, no significant differences in accuracy were found (see **Table 1** for the model summary and **Figure 2**): neither between the control and M1 conditions within congruent trials ($z = 0.87$, $p = 0.384$), nor between incongruent and congruent trials within the M1 condition ($z = -0.07$, $p = 0.944$). No interaction between condition and congruence was observed ($z = -0.68$, $p = 0.497$). Taken together, no evidence was thus found for a general effect of the M1 condition on the accuracy, despite the high number of trials and participants.

Moreover, we calculated d' and the type 1 criterion for every participant for both TMS conditions separately; we fitted a linear mixed-effects model with condition as a fixed factor, and a participant-specific intercept as a random effect. No difference in d' [$t(45.0) = -0.86$, $p = 0.394$] and the type 1 criterion [$t(45.0) = -1.107$, $p = 0.274$] between the M1 condition and

the control condition was observed. The analysis thus did not find evidence for a difference in the identification ability and the response criterion in the identification task across conditions.

To investigate identification RTs, we fitted a linear mixed-effects regression model with interactions between condition, congruence, and PAS rating as fixed effects. All effects were taken as random at the participant level. We found that the RTs in the M1 condition were significantly longer than in the control condition within congruent trials [$t(86.78) = 2.30$, $p = 0.024$]; also, in the M1 condition, congruent trials took longer than incongruent ones [$t(99.99) = 3.05$, $p = 0.003$]. Additionally, we were interested in how these RT differences manifested across PAS ratings. Conditional pairwise comparisons revealed that identification RTs for the middle ratings were significantly longer

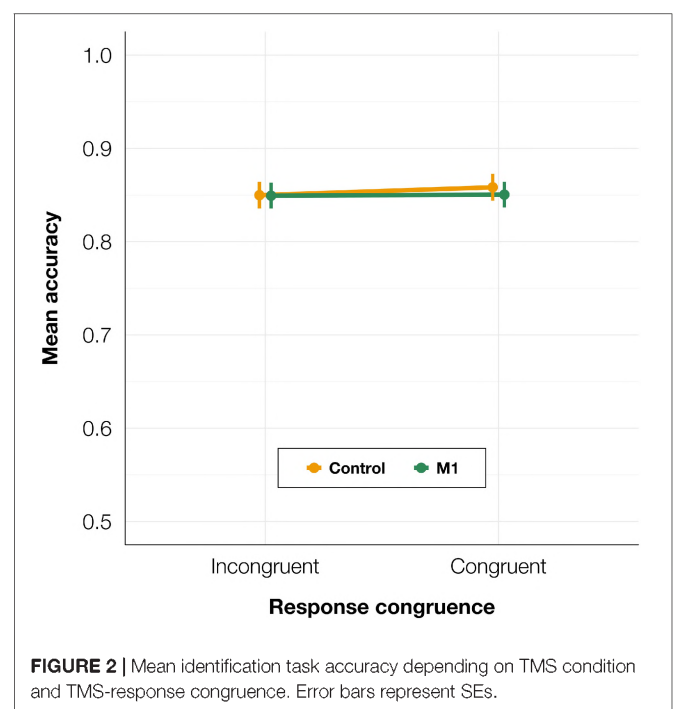


TABLE 1 | Results summary of the mixed-effects logistic regression model for accuracy with TMS condition and TMS-response congruence as fixed effects; participant-specific condition effect, congruence effect, and intercept were used as random effects.

	Estimate	SE	z	p
(Intercept)	1.738	0.11	15.58	<0.001***
TMS condition	0.063	0.07	0.87	0.384
TMS-response congruence	-0.009	0.13	-0.07	0.944
TMS	-0.059	0.09	-0.68	0.497
condition × TMS-response congruence				

Significance code: *** $p < 0.001$.

TABLE 2 | Pairwise comparisons of identification RTs' regression coefficients for the linear mixed-effects regression model with interactions between condition, congruence, and PAS rating as fixed effects, and with participant-specific condition effect, congruence effect, and intercept as random effects. *P* values adjusted with Tukey correction method. **(A)** Comparisons of estimates for each PAS rating between M1 and control condition in congruent trials. **(B)** Comparisons of estimates for each PAS rating between congruent and incongruent trials in the M1 condition.

	Estimate	SE	z Ratio	<i>p</i> Adjusted
(A) Congruent, M1—Control				
PAS 0	19.58	10.54	1.86	0.247
PAS 1	22.37	8.19	2.73	0.032*
PAS 2	25.49	8.95	2.85	0.023*
PAS 3	5.68	13.64	0.42	0.976
(B) M1, Congruent—Incongruent				
PAS 0	16.08	10.13	1.59	0.386
PAS 1	27.92	7.65	3.65	0.002**
PAS 2	25.94	8.57	3.03	0.013*
PAS 3	4.33	13.16	0.33	0.988

Significance code: ***p* < 0.01, **p* < 0.05.

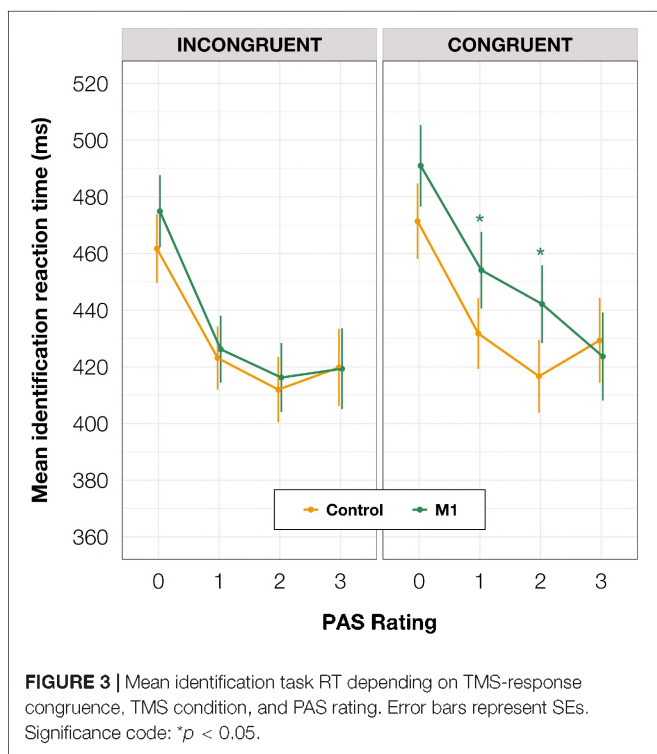


FIGURE 3 | Mean identification task RT depending on TMS-response congruence, TMS condition, and PAS rating. Error bars represent SEs. Significance code: **p* < 0.05.

in the M1 condition compared to the control condition within congruent trials. The same was observed for congruent trials as compared to incongruent trials within the M1 condition (see **Table 2** and **Figure 3**). In sum, identification responses were slower when the TMS influenced the muscle activity of the hand with which participants responded to stimuli which were detected but not seen clearly, thus indicating an extended evaluation process in these cases.

TABLE 3 | Results summary of the linear mixed-effects model for PAS ratings with condition and TMS-response congruence as fixed effects; participant-specific condition effect, congruence effect, and intercept were used as random effects.

	Estimate	SE	t (df)	<i>p</i>
(Intercept)	1.344	0.074	18.20 (45.7)	<0.00***
TMS condition	-0.050	0.028	-1.76 (66.5)	0.083
TMS-response congruence	-0.057	0.043	-1.34 (52.6)	0.185
TMS condition × TMS-response congruence	-0.054	0.024	-2.30 (17829.3)	0.021*

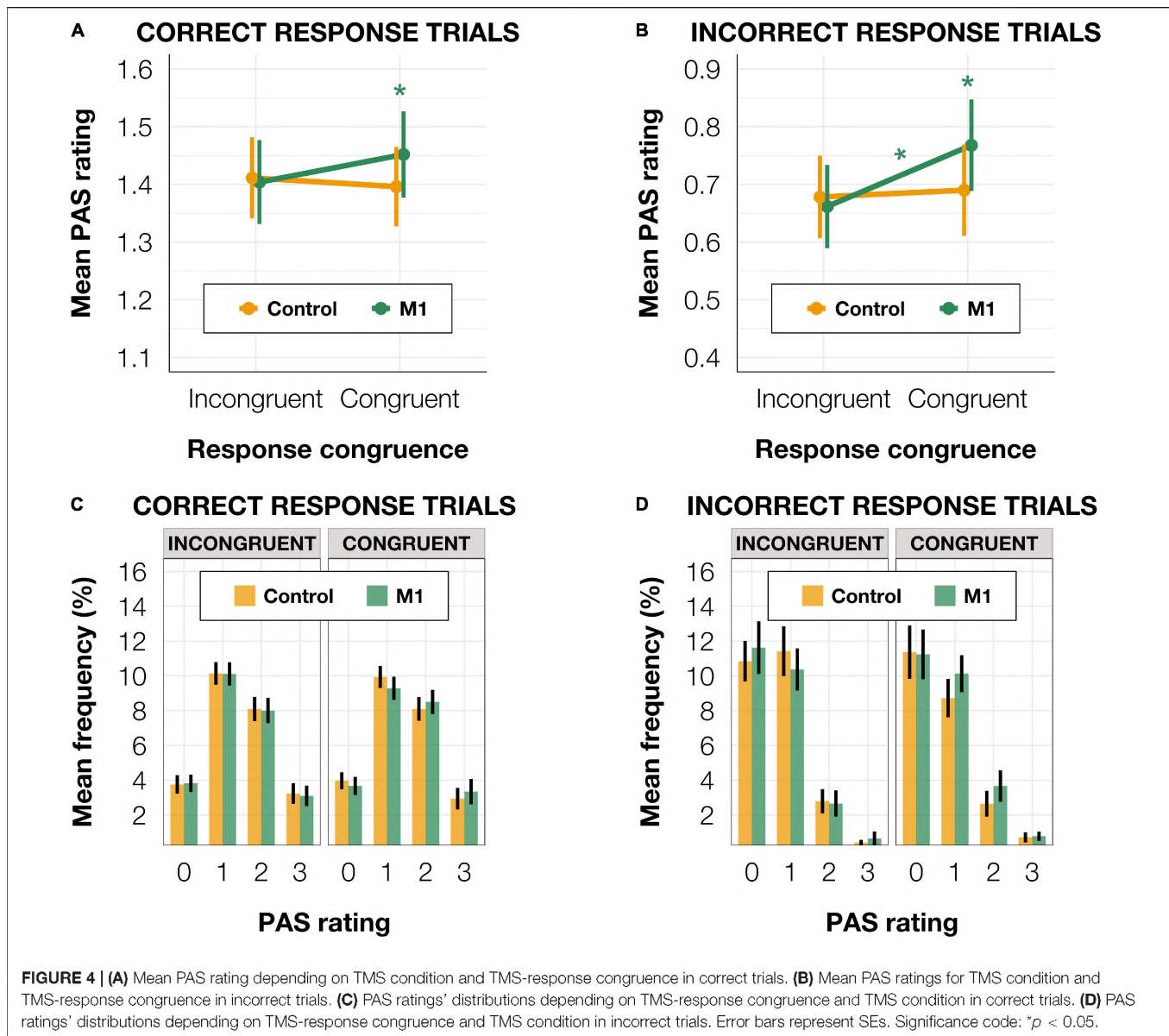
Significance code: ****p* < 0.001, **p* < 0.05, .*p* < 0.1.

PAS Ratings

To test the impact of TMS on PAS ratings, we fitted a linear mixed-effects model with interaction between condition and congruence as fixed effects. All effects were taken as random at the participant level. We used linear modeling to achieve comparability with the previous study of Fleming et al. (2015) and because the available implementations of ordinal models do not allow random effects in individual thresholds (Bürkner and Vuorre, 2019). We observed a significant interaction between condition and congruence [$t(17,829.20) = -2.30$, $p = 0.021$, see **Table 3** for regression model summary]. We reparameterized the model to check the effect of the TMS condition that was nested in the TMS-response congruence effect and observed a numerically higher mean PAS rating in M1 compared to the control condition in congruent [$t(66.46) = 1.76$, $p = 0.083$] but not incongruent trials [$t(65.84) = -0.16$, $p = 0.876$].

Since Fleming et al. (2015) observed a similar effect in correct trials and a reversed pattern (higher confidence in incongruent than congruent trials) in incorrect trials, we ran the model separately for subsets of correct ($n = 14,841$) and incorrect ($n = 3,128$) identification response trials. The results pattern did not depend on accuracy. For correct trials, we observed a significant effect of interaction between condition and congruence [$t(14,751.0) = -2.54$, $p = 0.011$] and a significantly higher mean PAS rating in M1 compared to the control condition in congruent [$t(70.93) = 1.20$, $p = 0.050$] but not in incongruent trials [$t(70.33) = 0.16$, $p = 0.795$; **Figures 4A,C**]. In incorrect trials, a significant interaction between condition and congruence [$t(1,650.52) = -2.01$, $p = 0.044$] was also present. There was a significantly higher mean PAS rating in M1 compared to the control condition within congruent [$t(90.90) = 2.04$, $p = 0.044$] but not incongruent trials [$t(88.23) = 0.44$, $p = 0.659$; **Figures 4B,D**]. The reparameterization of the model did not show an effect of congruence [$t(50.71) = 0.94$, $p = 0.354$] in M1 correct trials, but it revealed a significant difference between congruent and incongruent trials [$t(74.70) = 2.05$, $p = 0.044$] in incorrect M1 trials (see **Figures 4A,B**).

Additionally, to compare RTs of PAS ratings, we fitted a mixed-effects linear regression model with interactions between condition, congruence, and PAS ratings as fixed effects. All effects were taken as random at the participant level. This analysis revealed that the PAS rating RTs in the M1 condition were



significantly longer than in the control condition [$t(59.38) = 3.58$, $p < 0.001$] within congruent trials. Since no interaction between condition and congruence was observed [$t(17,749.88) = 0.31$, $p = 0.754$], the effect was observed for both congruent and incongruent trials. The pairwise comparisons revealed evidence that the effect applied to the two lowest ratings' RTs (see **Table 4** and **Figure 5** for pairwise comparisons).

M-Ratio

To test whether there was a difference in metacognitive efficiency between the M1 and control conditions, we calculated M-ratios for every participant for TMS and congruence conditions separately. We fitted a linear mixed-effects model with condition and congruence as fixed factors, and with participant-specific condition effect and intercept as random effects. We found no significant effect of condition [$t(91.6) = -0.15$,

$p = 0.882$] or congruence [$t(90.0) = 0.98$, $p = 0.332$], and no interaction between condition and congruence was observed [$t(90.0) = -0.48$, $p = 0.630$; see **Figure 6**]. It should be noted that due to the greater analysis complexity, these tests may have lower statistical power than the other analyses presented in the paper (Kristensen et al., 2020). However, because there was an increase in PAS ratings for both correct and incorrect trials, we did not expect to observe a difference in M-ratio.

MEP Amplitudes

To test differences in MEP amplitudes, we fitted a linear mixed-effects model with interaction between condition, congruence, and PAS rating as fixed effects, and with participant-specific condition effects, congruence effect, and intercept as random effects. Since only M1 TMS was supposed to influence the motor cortex, a significant difference between MEP amplitudes

TABLE 4 | Pairwise comparisons of PAS RTs' regression coefficients of the linear mixed-effects regression model with interactions between condition, congruence, and PAS ratings as fixed effects, and participant-specific condition effect, congruence effect, PAS rating effect, and intercept as random effects.

	Estimate	SE	z Ratio	p Adjusted
(A) Incongruent, M1—Control				
PAS 0	79.75	18.8	4.23	<0.001***
PAS 1	69.67	16.0	4.36	<0.001***
PAS 2	38.06	17.2	2.21	0.119
PAS 3	33.98	23.0	1.48	0.452
(B) Congruent, M1—Control				
PAS 0	69.44	19.0	3.66	0.001**
PAS 1	80.44	16.2	4.99	<0.001***
PAS 2	39.48	17.0	2.32	0.094
PAS 3	20.24	23.0	0.88	0.815

P values adjusted with Tukey correction method. (A) Comparisons of estimates for each PAS rating between M1 and control condition in incongruent trials. (B) Comparisons of estimates for each PAS rating between M1 and control condition in congruent trials. Significance code: ****p* < 0.001, ***p* < 0.01, •*p* < 0.1.

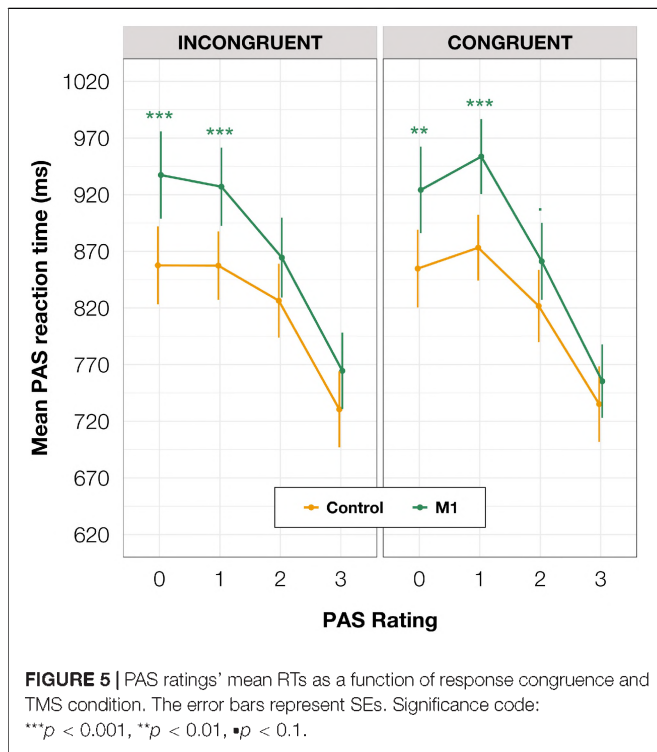


FIGURE 5 | PAS ratings' mean RTs as a function of response congruence and TMS condition. The error bars represent SEs. Significance code: ****p* < 0.001, ***p* < 0.01, •*p* < 0.1.

in the M1 condition compared to the control condition was observed [$t(45.81) = 10.61, p < 0.001$]. Interestingly, there was a significant interaction between congruence and condition [$t(17465.41) = 5.70, p < 0.001$], and the results of the model reparameterization showed significantly higher MEP amplitudes in congruent trials [$t(116.09) = 6.55, p < 0.001$]. Additionally, we were interested in determining whether this difference was related to PAS ratings. Therefore, we performed pairwise comparisons of MEP amplitudes for each PAS rating

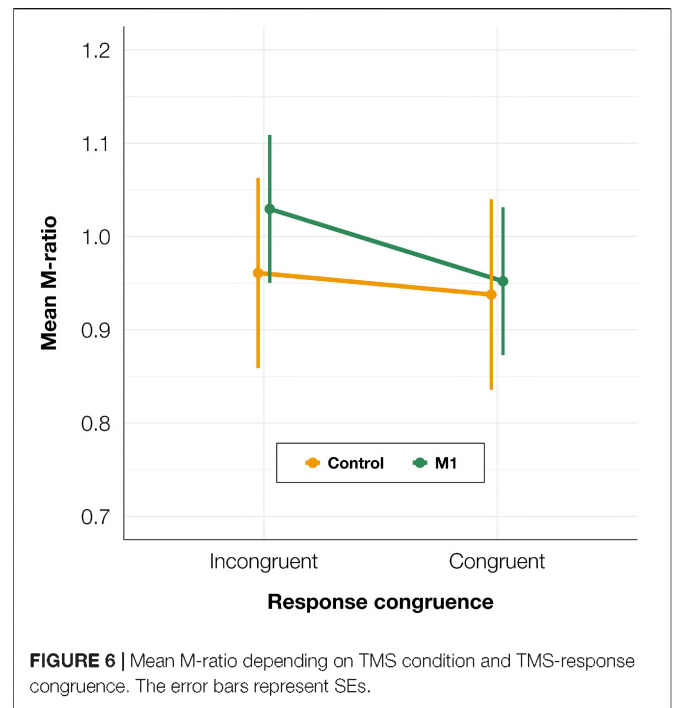


FIGURE 6 | Mean M-ratio depending on TMS condition and TMS-response congruence. The error bars represent SEs.

TABLE 5 | Pairwise comparisons of mean MEP amplitude regression coefficients of the linear mixed-effects regression model with interactions between condition, congruence, and PAS ratings as fixed effects, and with participant-specific condition effect, congruence effect, and intercept as random effects.

M1, Congruent—Incongruent	Estimate	SE	z Ratio	p Adjusted
PAS 0	44.46	16.16	2.75	0.030*
PAS 1	33.46	11.97	2.80	0.026*
PAS 2	50.43	13.56	3.72	0.001**
PAS 3	114.23	21.20	5.39	<0.001***

P values adjusted with Tukey correction method. Comparisons of estimates for each PAS rating between congruent and incongruent trials in the M1 condition. Significance code: ****p* < 0.001, ***p* < 0.01, **p* < 0.05.

between congruent and incongruent trials in the M1 condition. Comparing amplitudes of MEPs across PAS ratings gradually yielded significant differences (see Table 5 and Figure 7 for detailed results).

To determine if it is possible to discern PAS rating from the preceding MEP, we compared amplitudes from trials with different PAS ratings. Only trials with rating 3 (a clear experience) were significantly different from the others, irrespective of congruence (see Table 6 for all pairwise comparisons).

DISCUSSION

In the present study, we aimed to gain insight into the influence of the motor system on perceptual awareness judgments. We determined whether TMS-induced activity that was delivered to M1 following stimulus presentation altered participants' judgments of stimulus awareness, as indexed by PAS ratings. Our results show that TMS congruent to participants' responses

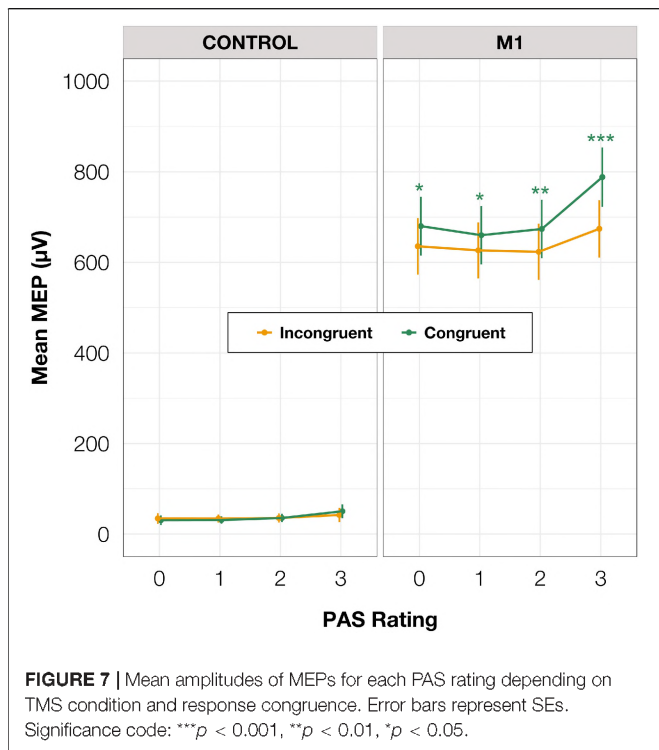


TABLE 6 | Pairwise comparisons of mean MEP amplitude regression coefficients of the linear mixed-effects regression model with interactions between condition, congruence, and PAS ratings as fixed effects, and with participant-specific condition effect, congruence effect, and intercept as random effects.

M1	PAS	Estimate	SE	z Ratio	p Adjusted
Incongruent	0–1	8.94	13.03	0.68	0.902
	0–2	12.20	14.49	0.84	0.834
	0–3	-38.64	19.19	-2.01	0.183
	1–2	3.26	11.83	0.29	0.993
	1–3	-47.58	17.39	-2.74	0.031*
	2–3	-50.85	17.53	-2.90	0.020*
Congruent	0–1	19.94	13.60	1.47	0.458
	0–2	6.23	14.62	0.43	0.973
	0–3	-108.41	19.16	-5.66	<0.001***
	1–2	-13.71	11.76	-1.17	0.648
	1–3	-128.35	17.11	-7.50	<0.001***
	2–3	-114.63	17.05	-6.72	<0.001***

P values adjusted with Tukey correction method. Comparisons of estimates between PAS ratings for congruent and incongruent trials in the M1 condition. Significance code: *** $p < 0.001$, * $p < 0.05$.

increased the reported stimulus awareness, but there was no evidence for altering the extent to which they are objectively sensitive to visual information. Moreover, no identification task bias was observed. We observed longer awareness rating RTs in the M1 condition in trials accompanied by stimulus awareness that was vaguer than an almost clear experience. Despite using a procedure with a delayed identification task, we observed longer identification RTs in the M1 condition, in which the TMS effect on RTs was limited to TMS-response-congruent trials.

Additionally, we attempted to determine whether TMS had an influence on the metacognitive efficiency measure, but we found no evidence to support such a claim. Finally, MEP amplitudes were related to PAS ratings and response congruence.

Based on the presented data, we suggest that the externally induced activity in M1 served as additional non-diagnostic evidence for the evidence-accumulation processes underlying visual awareness judgment and stimulus identification decisions. In response to TMS, we observed prolonged activity of these processes, predominantly when the perceptual evidence was not fully decisive. Moreover, the process of perceptual evidence accumulation also seems to be reflected in M1 excitability, which is implicated by MEP amplitude.

Motor Information Influences Visual Awareness Judgments

Our primary goal was to determine whether the activity of the motor system can contribute to perceptual judgments. The work of Siedlecka et al. (2019) has already shown that irrelevant motor responses that share a response scheme with a visual stimulus identification task increase reported stimulus awareness. However, their design did not allow the exclusion of confounding factors such as the introduction of additional visual information or attentional engagement, because participants were explicitly instructed and cued to perform an additional response (but see: Siedlecka et al., 2020a). In this study, we expanded upon their paradigm by applying TMS to M1 to reduce the influence of these confounds. Nevertheless, our conclusions reinforce those of Siedlecka et al. (2019): additional motor system activity can be incorporated in perceptual awareness judgment.

Unlike Siedlecka et al. (2019), we only found indications for post-perceptual evidence accumulation in response congruent trials, whereas their results showed an increase in PAS ratings irrespective of response congruence. This could be a consequence of the fact that participants in their study performed an intentional response before providing a rating. The additional task was very simple so that participants could have both motor plans prepared in each trial before responding to a visual cue, which would increase motor cortex activity in both hemispheres. In our study, TMS-induced motor activity could be weaker than that related to actual movement, which might explain its specific effect. Our results provide confirmatory evidence that the findings reported by Siedlecka et al. (2019) were a result of motor activity. Additionally, Filevich et al. (2020) conclude that key presses in continuous report conditions served as an additional source of evidence available for both the metacognitive judgment and biased participants toward more liberal confidence responses. A similar conclusion comes from the research on perception of voluntary action where both active and passive movements produced overconfidence (Charles et al., 2020).

A similar approach was undertaken by Fleming et al. (2015), who used single-pulse TMS either before or immediately after the 2AFC task response. In separate experiments, TMS was applied to either PMd or M1. The results of PMd TMS revealed higher confidence in TMS-response-incongruent (as compared to congruent) response trials in error trials within

the pre-response TMS condition. Moreover, for correct trials in the pre-response TMS condition, a tendency toward lower confidence in trials with TMS-incongruent as compared to TMS-congruent responses was observed. This difference was significant in the post-response TMS condition, but no M1 TMS effect was observed. An explanation of the discrepancies between their and our M1 TMS results could be the different timing of TMS, its intensity, or the substantial difference in sample size (23 vs. 46). Our conclusions suggest that M1 TMS might influence metacognitive judgment (although not necessarily metacognitive sensitivity). Besides, it might also suggest that the PMd TMS effect observed in Fleming's study could actually have partly been a result of an increase in confidence in TMS-response-congruent trials, as indicated by their results, especially in the post-response TMS condition. Providing additional activity to the motor cortex might strengthen ongoing activity and increase confidence in an already chosen response. In this interpretation, PMd activity would reinforce the motor plan related to the TMS congruent response that would compete with the plan in the hemisphere related to the TMS incongruent response. Thus, not only activity in M1 congruent to TMS would increase, but this increase would cause inhibition of M1 in the other hemisphere (Reis et al., 2008); as a consequence, activity in M1 would not only increase confidence in congruent responses but would also decrease it in incongruent ones. However, since all our participants except one declared to be right-handed the presented conclusions may not be generalizable to the left-handed population. Future research could address the issue related to the individual traits such as degree of handedness or plasticity history related to manual training, which could influence the interhemispheric communication (Davidson and Tremblay, 2013; Kuo et al., 2019; Loprinzi et al., 2020).

In addition, as might be supported by the analyses of metacognitive efficiency measures, PMd and M1 might differently impact metacognitive processes. Fleming et al. (2015) reported lower metacognitive efficiency (measured with M-ratio) in incongruent trials. Specifically, this was observed only for the PMd TMS that was delivered prior to the identification response. These results might further support the assumption that information related to PMd activity is incorporated into the evaluation of action performance. This could happen through increased activity of M1 related to the alternative motor scheme, which represents evidence against the chosen response. The facilitation of the alternative response could impact confidence and consequently lead to decreased M-ratio in TMS-response-incongruent trials. This seems in line with research showing temporarily increased excitability in M1 in response to PMd stimulation (Koch et al., 2006). Importantly, stimulation of the left PMd is often reported as affecting M1 in both hemispheres (Fujiyama et al., 2016). In our study, additional M1 activity would increase the amount of evidence for the congruent response (correct or not), resulting in no change in metacognitive efficiency measures while still impacting overall metacognitive ratings.

In the study of Fleming et al. (2015), the only RT effect observed was related to longer discrimination task and

confidence rating RTs in the PMd and M1 conditions when TMS was applied after the discrimination response as compared to before it. This effect was not replicated in the second experiment reported in their paper. In comparison, our results show longer PAS RTs in M1 condition compared to the control condition when stimulus awareness was absent or unclear. The slower PAS RT in M1 condition could result from a TMS influence on the motor areas responsible for generating oral movements (Möttönen et al., 2014); however, they were not observed for all PAS ratings (as would be expected from the oral movements impairment), only for the two lowest ones. Thus, we consider these results to be a consequence of additional evidence contribution to shape metacognitive judgment. We assume that when the stimuli experience is not clear, the evidence accumulation process operates for longer and/or with a lesser amount of evidence. This could make TMS-induced activity appear to be incorporated "on time" in a metacognitive judgment but also cause the post-perceptual evidence to be of higher importance. It might be the case that this was possible partially due to the inclusion of the identification response at the end of the trial so the accumulation processes could last longer.

The nature of TMS experiments often makes the manipulation apparent to participants. In our study, due to finger movements, it might have been clear to them which condition was the experimental one, thus triggering observer-expectancy effects. However, this should result in differences between conditions in identification accuracy or increase PAS ratings for both congruent and incongruent trials, but these effects were not present in the data. For the observer-expectancy effect to be the case, participants would have to hold a specific belief about the experimenters' expectation of higher awareness in the M1 condition and TMS-response congruent trials. They would have to remember to rate awareness as higher only when TMS is congruent with the identification task response, or when the Gabor is tilted to the right. The first would require the relatively difficult task of making a comparison with a response that follows an awareness rating. The second can be ruled out because PAS ratings for right-oriented Gabor stimuli provided with the left hand (incongruent trials in incongruent condition) were not higher as compared to the control. An alternative possible interpretations of our results are that TMS in TMS-response congruent condition triggered a distraction leading to attentional capture or influenced participants expectations, both resulting in that the participants paid greater attention to right-oriented Gabors in the M1 condition. If any of these was the case, then we should have observed a difference in the identification task performance and/or bias between the experimental conditions, but this was not the case. Therefore, although the design of the study cannot fully rule out attention or expectation effects, in our view their influence in this study is negligible.

Possible Mechanisms of Post-Perceptual Evidence Integration

Our experiment provides evidence for a distinct path in a complex system that integrates information between perception,

metacognition, and action. However, the structure of this system and the nature of the interactions between its parts is still largely unknown. This leaves an open question about the neuronal mechanism that leads to the effects observed in this study.

There is a growing body of evidence which shows that the PFC, especially the dlPFC, can be considered a key structure in integrating the information that is necessary for metacognitive judgments (Rounis et al., 2010; Fleming and Dolan, 2012). Assuming the central role of the dlPFC in awareness judgments, there are strong presumptions to treat the TMS effects observed in our study as an indirect influence because there are no direct connections between M1 and dlPFC (Passingham, 1993). This implies that the most probable route for the integration of information from M1 is through the S1. Nevertheless, there is ample evidence for the reciprocal connections between M1 and S1 (Gandolla et al., 2014), through which M1 activity would influence S1 activity. Such S1 influence could resemble feedback information about the muscle activation of the response finger.

Information transfer between M1 and S1 through muscle activation could explain why our experiment, in which TMS intensity was above participants' RMT, and the study by Siedlecka et al. (2019), in which participants performed an additional behavioral response, resulted in a significant influence of experimental manipulation on awareness ratings. Both explanations seem consistent with the results of Fleming et al. (2015), who used TMS intensity that was below the threshold of overt motor activity and thus limited the possibility of (1) sufficient direct influence from M1 on S1 or (2) sufficient muscle activity to cause somatosensory feedback. Similarly, Gajdos et al. (2019) suggest that pre-response partial muscle activation alters the somatosensory readout, which is later integrated into metacognitive judgment.

However, the PPC would also likely be involved in integrating somatosensory information with perceptual evidence from other modalities. In normal circumstances, information from the sensorimotor feedback loop would be used to compare executed behavior with the motor plan that requires the engagement of frontal areas (e.g., dlPFC and PM). The more pronounced the mismatch, the lower one's confidence in the accuracy of one's action would be. This might be why the procedural manipulation of Fleming et al. (2015) resulted in a difference in metacognitive efficiency in the pre-response TMS condition. Their stimulation of PMd possibly altered the 2AFC task response execution, thus creating a mismatch that was caught by the error monitoring processes. However, TMS in our experiment was delivered early enough before the identification response to be integrated as post-perceptual, additional evidence before a motor plan was fully formed. This would selectively increase the evidence for a stimulus associated with a particular motor plan, thus allowing participants to give higher metacognitive ratings in TMS-response-congruent trials. Crucially, early integration of this motor information would not create a mismatch between the planned and the performed response, so it did not lead to a change in metacognitive efficiency. The observation that TMS-related evidence interplayed with the selected motor plan

suggests that either higher PAS ratings and longer identification RTs in M1 TMS have a common cause, or PAS response provides additional evidence for identification task decisions.

MEP as a Measure of Accumulated Perceptual Evidence

Our additional hypotheses concerned the possibility of using the MEP to quantify the neuronal correlate of perceptual evidence accumulation. MEP amplitude is frequently used as a read-out of M1 excitability state (Bestmann and Krakauer, 2015). Cognitive manipulation of spatial attention (Mars et al., 2007), values assigned to different responses (Klein-Flügge and Bestmann, 2012), or contextual uncertainty (Bestmann et al., 2008) can all influence M1 excitability. Crucially, M1 can be treated as a recipient of a decision process initiated in other brain areas that modulates its excitability (Klein-Flügge and Bestmann, 2012; Klein-Flügge et al., 2013). When a relation with a particular response is present, MEP amplitudes for chosen versus unchosen actions distinguish the forthcoming choice before completion of the decision process (Klein-Flügge and Bestmann, 2012).

Our electromyographic results go along with these findings. We found an effect of M1 TMS-response congruence: congruent trials were characterized by higher MEP amplitudes. This effect was observed predominantly when participants reported high stimulus awareness. These results seem to be complementary to the dynamics of perceptual and post-perceptual evidence accumulation reflected in identification RTs. Taken together, they suggest that for clearly visible stimuli, when the necessary evidence has already been accumulated, the motor plan has been selected prior to TMS, thus increasing M1 excitability in preparation for execution of the response. Alternatively, no motor decision has been made, but the perceptual information about the stimulus has been passed from the visual cortex to M1, bypassing the PFC (Goodale, 2011). Presumably, within such conditions, additional evidence from the TMS does not play a crucial role in awareness judgment. Contrarily, while stimulus awareness is low, accumulation of evidence is still ongoing, thus allowing TMS to play a noticeable role.

Finally, changes in MEP amplitudes might reflect accumulation of stimulus-related evidence since the TMS-induced movement and the identification response were separated by several seconds, long before motor response execution. This seems possible based on the presence of connections from PPC to M1 (Gharbawie et al., 2011). There is a substantive body of evidence that PPC serves a multisensory integration function (Goodale and Milner, 1992; Koch et al., 2007; Kaas and Stepniewska, 2016) and plays an important role in performing voluntary movements, especially if they require visual input (Vingerhoets, 2014). In recent years, there has been growing evidence that PPC has direct reciprocal connections to M1 (Schulz et al., 2015; Isayama et al., 2019). These connections could serve as a potential pathway for perceptual evidence accumulated in PPC to directly influence M1 excitability in situations in which motor plans are simple or are executed automatically (as in our experiment). This could explain the differences in the excitability of M1 that were observed in our

experiment in trials with high PAS ratings, as more information would be transferred from PPC to M1.

CONCLUSION

Overall, our results shed new light on the relation between action and perceptual awareness by providing evidence that the motor system can be incorporated into metacognitive processes. Combined with previous results, these findings broaden our understanding of the interactions between action and conscious access that allow humans to dynamically adjust and re-evaluate their interactions with the environment. The significance of the influence of motor information on awareness judgments calls for broader theories of conscious access that primarily focus on processing sensory input.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are publicly available. These data can be found here: <https://osf.io/29n6j>.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Ethics Committee of the Institute of Psychology at Jagiellonian University. The participants provided their written informed consent to participate in this study.

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AUTHOR CONTRIBUTIONS

JH, MW, and KS designed the study. JH programmed and performed the study and drafted and corrected the manuscript. JH, MK, and BP were involved in the data analysis. MK provided changes to the manuscript. MW, KS, and BP provided comments on the manuscript. All authors contributed to the article and approved the submitted version.

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The reviewer JS declared a past co-authorship with several of the authors, MK, BP, and MW, to the handling editor.

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Krakow, 13.04.23

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Co-authorship statement

As a co-author of the publication: **Transcranial Magnetic Stimulation-Induced Motor Cortex Activity Influences Visual Awareness Judgments, *Frontiers in Neuroscience*, 14, 2020, 580712**, I declare that my own substantial contribution to this publication consists of:

- involvement in analysis of the results,
- involvement in interpretation of the results,
- involvement in visualisation of the results,
- providing comments and suggestions on the manuscript.

Hereby, I agree to submit the above-mentioned publication by MSc Justyna Hobot as part of a doctoral dissertation in the form of a thematically coherent collection of articles published in scientific journals.

I declare that an independent and identifiable part of the abovementioned publication shows individual contribution of MSc Justyna Hobot consisting of:

- major involvement in designing the study,
- programming the computer procedures,
- recruiting the research participants,
- performing all the measurements,
- analysing the results (with supervision),
- visualising the results,
- interpreting the results (with supervision),
- drafting the manuscript,
- correcting the manuscript,
- managing the publication process.



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- acquiring funding,
- involvement in designing the study,
- supervising analysis of the results,
- involvement in interpreting the results,
- providing comments on the manuscript.

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- major involvement in designing the study,
- programming the computer procedures,
- recruiting the research participants,
- performing all the measurements,
- analysing the results (with supervision),
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- drafting the manuscript,
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5. Outcomes

The first article included in the current thesis discussed the applicability of causal inferences in TMS research. While previous publications have addressed the issues of TMS-based inferences, this article provides a comprehensive summary and proposes new arguments upon existing knowledge. Firstly, the article clarified the distinction between the causal effects of TMS as an intervention and the causal effects related to the brain, which can be divided into direct and network effects. Secondly, it outlined the issues associated with TMS-based inferences. Addressing these issues in practice may require limiting the extent of TMS-based reasoning, but it can also aid the analysis of possible mediating and confounding variables and contribute to the improvement of TMS research designs. To address these limitations, it is recommended to ground research questions in outcomes of previous research, consider the complexity of the investigated cognitive function, use more than one control condition in a single study, and employ neuroimaging or neurophysiological techniques if possible. Neuroimaging enables examination of the direct and indirect influence of TMS. Thus, it can be used to study the spread of TMS effects throughout brain networks and often aids in determining whether the biological process under investigation has local or network characteristics. Although rTMS alone cannot definitively reveal structure-related causal claims regarding direct relations between brain processes carried out in certain areas and certain behaviours or cognitive functions, it can be employed to make probabilistic statements about causal influences if its limitations are acknowledged. While combining rTMS with neuroimaging techniques strengthens the validity of inferences, it does not imply that rTMS should be used exclusively in conjunction with neuroimaging. The need for including neuroimaging or multiple control conditions depends on the research question guiding the study and the extent to which the results are intended to be generalised. There is a trade-off between the limitations of inference and the feasibility of a study. Therefore, when workable, it is recommended to combine rTMS with neuroimaging, multiple control conditions, and/or spTMS, as it can offer additional support for research conclusions. While experts in non-invasive brain stimulation commonly address these issues, the theoretical analysis provided in the first article can assist researchers with less expertise who aim to design TMS studies or interpret TMS data.

In the first study, described in the second article, a higher metacognitive efficiency estimate was observed in the cTBS condition compared to the sham TBS condition, suggesting the involvement of the left aMPFC in accurate metacognitive judgements. This finding was further supported by an observed interaction between the TBS conditions and accuracy, related to lower awareness ratings in the incorrect identification task responses but not in the correct responses in the cTBS condition compared to the sham TBS condition. However, the role of left aMPFC in the overall level of reported visual awareness remains inconclusive. Although the results of this study support the claim that PFC activity affects the assessment of visual awareness, they do not explicitly support or rule out the possibility that the PFC is necessary for the conscious experience of a visual stimulus. No evidence was found for differences in the identification task performance, in the identification task RT, and in the PAS RT across the TBS conditions. Additionally, no evidence supported the hypothesis that cTBS and iTBS have opposite effects. Numerically, as compared to the sham TBS protocol, both active TBS

protocols altered all measures (except the PAS RT) in the same direction, albeit with varying magnitudes. These results suggest that the observed cTBS effect is related to metacognitive judgement and may be associated with performance monitoring. The observation that the cTBS effect on PAS ratings was limited to incorrect responses may suggest that it could be attributed to improved error monitoring or integration of error-related information. Another possible interpretation of these results could be the disruption of top-down influences, such as the influence of expectations on perception. Considering the issues related to rTMS-based inferences outlined in the first article, alternative explanations unrelated to the stimulation of the aMPFC, such as the peripheral impact of TBS on the retina, could be explored. In conclusion, our study offers limited support for the involvement of the PFC in the neural processes related to the assessment of visual awareness. However, it remains unclear whether the results concerning visual metacognitive efficiency can be considered evidence for PFC involvement in stimulus awareness itself.

In the study described in the third article, it was found that delivering TMS to M1 resulted in higher average awareness ratings in both correct and incorrect identification task response trials when the hand used to respond was opposite to the hemisphere stimulated in the M1 condition, i.e., TMS-response-congruent trials (congruent trials), compared to the control condition. This effect was accompanied by longer PAS RTs, regardless of the congruence between TMS and identification response. These results suggest that motor information, i.e., information that is not directly related to the content of visual perception, may affect the assessment of visual awareness. Additionally, longer identification task RTs were observed in congruent trials in the M1 condition compared to the control condition. There was no significant difference observed between conditions regarding metacognitive efficiency. The amplitudes of MEPs were associated with awareness ratings when considering response congruence, having higher values in congruent trials. Thus, MEP may serve as an indirect measure of perceptual evidence accumulation, while longer PAS RTs and higher amplitudes of MEPs within the M1 condition perhaps reflect the integration of additional evidence with visual awareness judgements. It is thus possible to indirectly infer (to a certain degree) the visual experiences of participants without reports. In summary, these findings suggest that motor cortex activity, in particular contexts, plays a role in visual awareness judgements, offering new insights into the relation between action and awareness. Thus, theories of consciousness should take into account the importance of non-visual factors, such as motor information, in shaping visual awareness judgements.

In summary, the findings of this thesis indicate that the PFC plays a role in shaping our subjective assessment of visual awareness and that factors unrelated to the content of visual perception may influence the evaluation of stimulus awareness. These results provide support to the Higher-Order Thought Theory (Brown et al., 2019; Lau & Rosenthal, 2011) or the Global Neuronal Workspace Theory (Dehaene, 2014; Dehaene & Naccache, 2001), but do not align with the Recurrent Processing Theory (Lamme, 2020) or some interpretations of the Integrated Information Theory (Koch et al., 2016; Tononi et al., 2016). However, it is important to note that the influence of PFC activity on metacognitive efficiency does not necessarily imply the

indispensability of the PFC for specific conscious content. The involvement of the PFC in consciousness may be restricted to processes that impact overall conscious experience or the judgement of stimulus awareness. Consequently, observed differences in awareness ratings could be influenced by judgement-related biases, including confidence. Nevertheless, certain researchers postulate that confidence estimates or a sense of knowing are intrinsic to representations within the Global Workspace (Baars et al., 2021; Kouider et al., 2010; Shea & Frith, 2019). Thus, depending on the chosen definition, confidence could be considered a feature of visual awareness (Lau & Rosenthal, 2011). Both visual awareness ratings and confidence ratings are regarded as metacognitive judgements (Fleming & Lau, 2014). Increasing evidence suggests that the PFC, particularly the dlPFC, plays a crucial role in integrating the information necessary for metacognitive judgements (Fleming & Dolan, 2012; Rounis et al., 2010). Given the presumed central role of the dlPFC in awareness judgements, it is likely that TMS to M1 leads to the integration of information in the PFC, however, the specific mechanisms involved in this integration remain unknown. Nonetheless, the results contribute new evidence to models of consciousness that conceptualise it within the framework of stimulus evidence accumulation (e.g., Block, 2011; Dehaene, 2008; Dehaene et al., 2003; Lamme, 2010).

6. Future aspects

As certain scientific inquiries are closer to being resolved, fresh ones emerge. There are several areas within TMS and consciousness research, including the studies presented, that may need further investigation and improvement.

6.1. Employment of TMS

One assumption prevalent in TMS literature, specifically in relation to the second article, is that a specific rTMS protocol influences neuronal activity in a certain direction. For instance, it is sometimes assumed that cTBS leads to decreased cortical excitability. In previous studies investigating the contribution of the PFC to subjective reports on visual awareness or visual metacognition, the effect of cTBS was described as follows: 'Physiological studies have shown that this produces a decrease in corticospinal excitability which lasts for about 20 min (Huang et al., 2005), when applied to the primary motor cortex, M1' (Rounis et al., 2010), 'The specific protocol used in our study is thought to suppress cortical excitability for up to 20 minutes' (Bor et al., 2017), and 'cTBS has been shown to produce a decrease in the excitation level in the stimulated cortex' (Rahnev et al., 2016). These publications refer to the same study on cTBS to M1 by Huang et al. (2005). However, subsequent studies indicate significant individual variability in the effects of cTBS on M1, suggesting that cTBS should not be universally regarded as an inhibitory protocol. Even when the same rTMS protocol is employed to stimulate the same brain area in different participants or studies, the direction of the effect is not always consistent (Caparelli et al., 2012; Hamada et al., 2013). Additionally, TMS-induced changes in neuronal activity in one brain area may not apply to other areas and other cognitive functions (Lowe et al., 2018). For these reasons, the effects of cTBS on M1 observed in the study by Huang et al. (2005) may not generalise to PFC. In studies by Rounis et al. (2010) and

Bor et al. (2017) where two brain areas (the dlPFC in both hemispheres) were targeted in a single experimental intervention, functional lateralization becomes a challenging aspect. For example, it has been observed that right-hemispheric dlPFC cTBS may lead to a more reward-guided performance, while left-hemispheric dlPFC cTBS may induce more avoidance-guided behaviour (Kaminski et al., 2011). When stimulating two areas within one study, it is difficult to determine which area should be stimulated first and what the consequences of the interaction between changes in brain activity in both areas will be, including potential compensatory effects (Lee & D'Esposito, 2012). To avoid this problem, I limited the rTMS in the first study to one brain area. Further research may benefit from including post-rTMS neuroimaging, which was not employed due to feasibility constraints. The utilisation of neuroimaging data can also enhance the accuracy of targeting the specific brain area of interest (as in the first study presented in this thesis) and enable the investigation of predictive factors that help to achieve the desired outcomes.

In addition, it is important to consider that the effects of rTMS are influenced by participants' activity before, during, and after rTMS, as well as various other individual factors (Hamada & Rothwell, 2016; Ridding & Ziemann, 2010). For example, factors such as the presence of task training immediately prior to applying a specific TMS protocol can alter cortical excitability and impact the expected outcomes. The magnitude and direction of the TMS effect depend on many factors, including the intensity of the stimulation protocol (Modugno et al., 2001), the baseline excitability of the stimulated brain area (Caparelli et al., 2012; Siebner et al., 2004; Weisz et al., 2012), the phase of the brain waves (Baur et al., 2020), the direction of the current flow through the TMS coil (Sommer et al., 2013; Talelli et al., 2007), and the duration of the TMS protocol (Gamboa et al., 2010). rTMS protocols can be applied at different durations, as exemplified by the mentioned cTBS studies, where a short version of the cTBS protocol (due to targeting both hemispheres) was used in the studies by Rounis et al. (2010) and Bor et al. (2017). A typical version was used in the study by Rahnev et al. (2016) and the first study presented in this thesis. Due to the combined influence of multiple factors, it is possible to observe neuroplasticity-like effects that contradict the expected outcomes. Additionally, determining the optimal values for protocol parameters, such as coil orientation for targeting specific areas, remains challenging. Typically, rTMS protocols adopt parameters based on previous studies that initially applied a given protocol in human studies. However, these parameters are often chosen arbitrarily and may not be suitable for specific studies targeting different brain areas or involving specific participant populations (e.g., individuals trained in particular behaviours). Therefore, protocol parameters, individual differences among study participants, and the effects of TMS on brain networks are crucial factors for future studies to consider.

6.2. TMS in visual awareness research

The second article demonstrated that applying cTBS to the aMPFC was associated with higher estimates of metacognitive efficiency than sham TBS. This result suggests the involvement of the PFC in the accuracy of visual awareness judgements. However, it does not provide definitive evidence that the effect is caused by changes in the PFC excitability. Given that the

effects of rTMS therapies largely rely on non-specific factors and the placebo effect (Malhi et al., 2021; Malhi & Bell, 2021), these factors may also play a role in rTMS studies conducted on healthy populations. In TMS studies, peripheral sensations may influence attention, arousal, and impulsivity, and subsequently affect awareness-related measures. Without explicitly addressing these issues, it is difficult to determine whether TMS does not affect awareness ratings due to its influence, for example, on arousal, which can occur through sensations resulting from TMS affecting the scalp muscles or its direct impact on the PFC. However, this is unlikely to be the case in our study, as the iTBS protocol, which (based on the author's experience) is more painful, did not show any significant difference compared to the sham TBS protocol. Also, the participants rather did not expect the effect in any particular direction. Future studies could investigate the extent to which the PFC contributes to visual awareness compared to its contribution to the level of arousal, along with examining the role of different subcomponents of the PFC in awareness judgements.

Further, in the first study, no significant differences between cTBS and iTBS were observed, potentially due to either suboptimal iTBS parameters, the overall limited effectiveness of iTBS, or variations in the timing and duration of after-effects across different TBS protocols. cTBS and iTBS likely have different optimal parameters, and their peak effects may occur at different time points (McCalley et al., 2021). Moreover, TBS protocols are specifically designed to target theta-gamma coupling (Vékony et al., 2018). While various brain processes, including those in the PFC, depend on theta-gamma coupling, it may not be the most crucial form of cross-frequency coupling for the processes related to visual awareness (although there are studies suggesting its relevance, such as Köster et al., 2018). In fact, when studying awareness, researchers can utilise multiple TMS protocols and target various areas within the PFC. Additionally, researchers may investigate whether alterations in metacognitive efficiency accompanying brain stimulation to the PFC are due to direct or indirect influences on the activity of other brain areas. For example, typically, the brain area that is closer to the scalp is stimulated more because of the shortest distance between that area and the TMS coil, making it challenging to disentangle the effects resulting from stimulation to the superficial area(s) from those resulting from the targeted area, if that area is at the deeper level in the brain (Deng et al., 2013). Consequently, researchers often cannot be certain about which areas are influenced and which processes are affected. Moreover, improvements in one cognitive domain following TMS may come at the expense of deteriorations in other domains (Kennedy et al., 2018), and the experimental task(s) employed may fail to capture any potentially undesirable effects. Therefore, future studies could incorporate control conditions for tasks that aim to measure cognitive processes other than visual awareness. This could help determine whether the observed effects are specific to visual awareness.

In relation to the third article, it is important to recognise that the motor response is not the sole factor influencing visual awareness ratings, and there may be multiple factors at play that have not been investigated yet. Numerous processes can occur between consciously perceiving a stimulus and rating one's awareness, and not all factors impacting participants' responses necessarily affect their awareness of stimuli. Therefore, it would be beneficial to more effectively distinguish between influences on visual awareness ratings and influences on

awareness of the stimulus itself. Additionally, it is important to note that the TMS-induced motor response reflects behavioural responses in specific experimental conditions that may not necessarily reflect natural settings. In many real-life situations, awareness of stimuli does not lead to motor responses. Moreover, when TMS induces movements, participants' sense of agency may be reduced. Therefore, this area holds some promise for further investigation by those interested in exploring the effects of TMS on processes related to visual awareness and a sense of agency.

Finally, to ensure robust results and account for high individual variability, it is recommended to have large sample sizes in TMS studies. In my research which was not included in this thesis, I addressed my concerns related to sample sizes and individual variability, with a specific focus on the occipital cortex. I conducted two subsequent TMS studies. The first study involved over 70 participants, where data was collected to assess individual resting motor thresholds and phosphene thresholds. These measures were correlated with neuroimaging data, questionnaire responses, and behavioural measures to investigate variations in conscious processing among healthy individuals (details available at <https://osf.io/g42xt/>). The second study was a two-day cTBS study involving 40 participants (details available at <https://osf.io/ty3mq/>). The manuscript based on this latter study is currently being finalised but cannot be included in this thesis due to the legal regulations on the expected PhD thesis format determined by Polish law.

6.3. Theory in visual awareness research

In the literature on consciousness, it is commonly assumed that the processes underlying responses to *objective* and *subjective* measures of visual perception differ (e.g., Maniscalco & Lau, 2016). However, at the level of brain processes, there may not be a substantial difference between processes targeted by *objective* and *subjective* measures of visual perception. For instance, a detection response is closely related to reporting visual awareness using a dichotomous scale with *seen* and *unseen* responses. The assumption that the neural mechanisms underlying responses in *objective* and *subjective* measures of visual perception are distinct may require closer evaluation. Also, the potential overlap between processes probed by tasks intended to measure different cognitive functions raises questions about distinguishing between stimulus awareness and other cognitive processes. The brain processes underlying what is defined as visual awareness can overlap with processes described as metacognition, confidence, working memory, or retrospective memory and similar, as participants often assess awareness of stimuli that are no longer present. Also, the term *visual awareness* can be interpreted in various ways. For some researchers, it may relate solely to the visual experience of a stimulus, while for others it may include a feeling of confidence. Distinguishing between visual metacognition and visual awareness requires further clarification, and some progress has already been made in this regard (Rahnev et al., 2022). This issue is also related to whether PAS measures visual awareness or judgements about visual awareness. Although PAS is considered a measure of visual awareness (Timmermans et al., 2010), it could be argued that PAS rating criteria, such as *a clear experience*, are, in fact, judgements. This is especially true considering that introspection within consciousness studies tends to be defined as a higher-order process (Ramsøy & Overgaard, 2004). Further

investigation is needed to explore the relation between models of neural correlates of consciousness, which primarily emphasise the transition between unconscious and conscious content (e.g., Crick & Koch, 1995; Dehaene, Sergent, & Changeux, 2003), and the potential inclusion of metacognition as a significant factor. Additionally, the understanding of metacognition can be approached in two ways: as a process that facilitates the emergence of stimulus awareness (Lau & Rosenthal, 2011; Rounis et al., 2010) or as a subsequent process which operates on conscious representation (Fleming, 2020; Overgaard & Sandberg, 2012). These issues necessitate further exploration and clarification.

In summary, the effectiveness of rTMS relies on various factors that should be considered in order to achieve satisfactory results. Drawing conclusions from TMS studies can be highly complex. Therefore, similar to any other area of research, it is essential to carefully consider meta-scientific and theoretical reflections on the questions posed, processes investigated, and the broader implications of specific study results. This approach will aid in balancing the scientific costs and benefits associated with the use of TMS in consciousness studies. I believe there are several elements present in this work that other researchers can build upon.

7. References

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8. Approvals of the ethics committees

The experimental procedures that are presented in the articles have obtained ethical approvals from two committees: the Jagiellonian University Institute of Psychology Research Ethics Committee and the *De Videnskabsetiske Komitéer for Region Midtjylland*. The opinions of these committees are included on the following pages.

KE/05/012017A

Kraków 22.08.2017 r.



UNIWERSYTET
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W KRAKOWIE

Wydział Filozoficzny

Instytut Psychologii

Opinia

Komisja ds. Etyki Badań Naukowych przy Instytucie Psychologii UJ akceptuje przedstawiony przez **mgr Justynę Hobot** projekt badań pt. ***Neuronalne korelaty świadomości metapoznawczej – badania TMS*** i stwierdza, że nie narusza on standardów etycznego prowadzenia badań naukowych.

Dr hab. Aleksandra Gruszka-Gosiewska

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**Projekt: Identification of sensory and cognitive characteristics from magnetic resonance (MR) data.**

De Videnskabetiske Komiteer for Region Midtjylland, Komité I, har behandlet projektet på sit møde den 20. april 2016, og du har efterfølgende indsendt revideret materiale. Komitéen har på den baggrund truffet følgende afgørelse.

Dato 06-06-2016

Sagsbehandler Anne-Marie Eybye

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Tel. +4578410184

Sagsnr. 1-10-72-69-16

Side 1

Afgørelse:

Projektet godkendes i henhold til lov nr. 593 af 14. juni 2011 om videnskabetisk behandling af sundhedsvidenskabelige forskningsprojekter.

Godkendelsen gælder for de anmeldte forsøgssteder, den anmeldte forsøgsansvarlige i Danmark samt for den angivne forsøgsperiode.

Godkendelsen gælder til den 31. marts 2022 og omfatter følgende dokumenter:

- Forsøgsprotokol, dateret 20. maj 2016, fil navngivet ver3_160520, fremsendt med mail af 20. maj 2016.
- Rekrutteringsmateriale, ikke dateret, fil navngivet ver1_160309, fremsendt med mail af 9. marts 2016.
- Deltagerinformation, ikke dateret, fil navngivet ver2_160329, fremsendt med mail af 5. april 2016.
- Samtykkeerklæring, ikke dateret, fil navngivet ver2_160329, fremsendt med mail af 29. marts 2016.

Godkendelsen omfatter tilladelse til, at der kan videregives oplysninger fra patientjournalen til forsker i henhold til sundhedsloven § 46, stk. 1. Tilladelsen omfatter videregivelse af de oplysninger, der er oplyst i protokollen.

Iværksættelse af projektet i strid med godkendelsen kan straffes med bøde eller fængsel, jf. komitélovens § 41.

Ændringer:

Foretages der væsentlige ændringer i protokolmateriale under gennemførelsen af projektet, skal disse anmeldes til komitéen i form af tillægsprotokoller. Ændringerne må først iværksættes efter godkendelse fra komitéen, jf. komitélovens § 27, stk. 1.

Anmeldelse af tillægsprotokoller skal ske elektronisk på www.drvc.dk med det allerede tildelte anmeldelsesnummer og adgangskode.

Væsentlige ændringer er bl.a. ændringer, der kan få betydning for forsøgspersonernes sikkerhed, fortolkning af den videnskabelige dokumentation, som projektet bygger på samt gennemførelsen eller ledelsen af projektet. Det kan fx være ændringer i in- og eksklusionskriterier, forsøgsdesign, antal forsøgspersoner, forsøgsprocedurer, behandlingsvarighed, effektparametre, ændringer om de forsøgsansvarlige eller forsøgssteder samt indholdsmæssige ændringer i det skriftlige informationsmateriale til forsøgspersonerne.

Hvor nye oplysninger betyder, at forskeren overvejer at ændre proceduren eller stoppe forsøget, skal komitéen orienteres om det.

Bivirkninger og hændelser:

Løbende indberetning

Komitéen skal omgående underrettes, hvis der under projektet optræder formodet alvorlige, uventede bivirkninger eller alvorlige hændelser, jf. komitélovens § 30, stk. 1.

Indberetningen skal ledsages af kommentarer om eventuelle konsekvenser for forsøget. Det er kun bivirkninger og hændelser forekommet i Danmark, der skal indberettes. Underretning skal ske senest 7 dage efter, at sponsor eller den forsøgsansvarlige har fået kendskab til tilfældet.

Ved indberetning kan anvendes et skema, der findes på www.drvc.dk. Skemaet med evt. bilag skal indsendes elektronisk i pdf-format til komite@rm.dk.

Årlig indberetning

En gang årligt i hele forsøgsperioden skal komitéen have tilsendt en liste over alle formodet alvorlige (ventede og uventede) bivirkninger og alvorlige hændelser, som er indtruffet i forsøgsperioden sammen med en rapport om forsøgspersonernes sikkerhed, jf. komitélovens § 30, stk. 2.

Ved indberetning kan anvendes et skema, der findes på www.dnvk.dk. Skemaet med evt. bilag skal indsendes elektronisk i pdf-format til komite@rm.dk.

Afslutning:

Den forsøgsansvarlige skal senest 90 dage efter afslutningen af projektet underrette komitéen herom, jf. komitélovens § 31, stk. 1. Projektet regnes som afsluttet, når sidste forsøgsperson er afsluttet.

Afbrydes projektet tidligere end planlagt, skal en begrundelse herfor sendes til komitéen senest 15 dage efter, at beslutningen er truffet, jf. komitélovens § 31, stk. 2.

Hvis projektet ikke påbegyndes, skal dette samt årsagen hertil meddeles komitéen.

Komitéen beder om kopi af den afsluttende forskningsrapport eller publikation, jf. komitélovens § 28, stk. 2. Vi skal i den forbindelse gøre opmærksom på, at der er pligt til at offentliggøre både negative, positive og inkonklusive forsøgsresultater, jf. komitélovens § 20, stk. 1, nr. 8.

Tilsyn:

Komitéen fører tilsyn med, at projektet udføres i overensstemmelse med godkendelsen, jf. komitélovens § 28 og § 29.

Følgende komitémedlemmer deltog i mødebehandlingen:

- Birgitte Mahler (formand)
- Helene Nørrelund
- Nete Hornung
- Rolf Dall

Lægpersoner

- Carl Johan Rasmussen (næstformand)
- Aleksander Aagaard
- Allan Clifford Christensen
- Jens Ove Kjeldsen
- Johs. Poulsen
- Mogens Bedsted

Venlig hilsen



Anne-Marie Eybye
Sekretær

midt
regionmidtjylland

Side 4

Kopi til:

- Neuropsykolog Kristian Sandberg, Aarhus Universitet / Aarhus Universitetshospital

9. Summaries

9.1. English summary

Neural underpinnings of visual awareness investigated with transcranial magnetic stimulation

The studies included in this thesis employed Transcranial Magnetic Stimulation (TMS) to investigate brain areas assumed to be involved in the formation of visual awareness judgements. TMS is a non-invasive brain stimulation method that temporarily influences neuronal activity in the targeted brain area. The thesis includes three published scientific articles. The first, theoretical article, discusses the constraints of TMS-based inferences and offers recommendations on how to address these limitations, specifically in the context of repetitive TMS (rTMS). The second and third articles present within-participant design experimental studies where TMS was applied to different brain areas of healthy individuals to examine its impact on visual awareness judgements and metacognitive efficiency, that is the ability to accurately judge one's own perception.

Unlike neuroimaging methods, TMS can modulate neuronal activity, which makes it a valuable tool for drawing causal inferences. The first article discusses the use of TMS to deduce the causal relations between brain areas and their functions. It critically assesses the extent to which causal inferences can be derived from rTMS data. It emphasises that relying solely on rTMS data does not provide sufficient grounds for strong inferences about the direct causal properties of targeted brain areas. The article proposes strategies for mitigating the limitations of rTMS, such as combining it with neuroimaging techniques and incorporating appropriate control conditions. It concludes that the strength of inferences drawn from rTMS studies largely depends on the experimental design, and rTMS may not always be suitable for answering questions related to causality. This article is intended for researchers aiming to design rTMS studies or interpret the results of rTMS studies causally.

The second article investigates the involvement of the PFC in the processes underlying visual awareness. While some theories of consciousness highlight the importance of the PFC as the neural underpinning of visual awareness (e.g., higher-order theories of consciousness), a number of researchers argue that PFC activity is not essential. It has also been hypothesised that the PFC is closely associated with metacognition. Therefore, the first study investigated the role of the PFC in visual awareness judgements by comparing three different rTMS protocols applied to the left anterior medial prefrontal cortex (aMPFC), a brain area associated with metacognition, to induce plasticity-like effects. The study employed three distinct Theta Burst Stimulation (TBS) protocols, namely continuous TBS (cTBS), intermittent TBS (iTBS), and sham TBS. The TBS protocol was applied prior to the behavioural testing involving a computer-based procedure with visual identification task and visual awareness ratings. The results indicated that cTBS led to higher estimates of metacognitive efficiency than the sham TBS. This effect was associated with lower visual awareness ratings for incorrect responses.

No significant differences were observed between the TBS conditions in terms of identification task performance or response times (RTs).

The third article addresses the ongoing discussion in the consciousness research community concerning the impact of non-visual information on visual awareness judgements. It describes a study that examined whether single-pulse TMS (spTMS) applied to the primary motor cortex (M1) approximately half a second after a stimulus presentation can serve as a piece of non-visual evidence and consequently influence visual awareness ratings. The study also investigated whether spTMS-induced motor-evoked potential (MEP) amplitude can reflect the amount of accumulated perceptual evidence. The study hypothesised that applying spTMS to M1 results in higher visual awareness ratings compared to a control condition. It also assumed that MEPs amplitudes would correspond to the amount of accumulated evidence. The experimental setup consisted of spTMS with a computer-based procedure involving visual awareness ratings and a visual identification task. The results revealed that in congruent trials (where the response hand used in the identification task matched the one stimulated in the M1 condition), there were higher visual awareness ratings and longer identification task RTs in the M1 condition compared to the control condition. Additionally, longer RTs for visual awareness ratings were observed in the M1 condition compared to the control condition, regardless of congruence, potentially suggesting the incorporation of additional evidence into visual awareness judgements. No significant difference was observed between conditions in terms of metacognitive efficiency. Furthermore, the amplitudes of MEPs were associated with the visual awareness ratings and exhibited higher values in congruent trials, implying that MEP could serve as an indirect measure of accumulated evidence.

Overall, the findings suggest that both the aMPFC and M1 can contribute to the formation of visual awareness judgements. However, it is crucial to recognise the limitations of the employed methods, especially TMS, and the intricate nature of the processes being studied.

9.2. Danish summary

Neurale faktorer bag visuel bevidsthed undersøgt med transkraniel magnetisk stimulation

Forskningen i denne afhandling anvendte Transkraniel Magnetisk Stimulation (TMS) til at undersøge hjerneregioner, der menes at være involveret i vurderinger af visuel bevidsthed. TMS er en ikke-invasiv metode til hjernestimulation, der midlertidigt påvirker neuronerne i det givne område. Afhandlingen indeholder tre videnskabelige artikler. Den første artikel er teoretisk og diskuterer begrænsningerne ved konklusioner baseret på TMS-resultater og giver anbefalinger til, hvordan disse begrænsninger kan håndteres, især i forbindelse med repetitiv TMS (rTMS). Anden og tredje artikel præsenterer eksperimentelle studier, hvor TMS blev anvendt på forskellige hjerneregioner hos raske forsøgsdeltagere for at undersøge effekten på vurderinger af visuel bevidsthed og metakognition, dvs. evnen til præcist at vurdere ens egen opfattelse.

I modsætning til hjernescanning kan TMS ændre på neuronal aktivitet, hvorfor der i et vist omfang kan drages kausale slutninger. Den første artikel diskuterer kritisk brugen af TMS til at drage kausale relationer mellem hjerneregioner og deres funktioner. Artiklen vurderer, at man ikke ud fra rTMS-data alene kan drage stærke konklusioner om bestemte hjerneområders kausale egenskaber. Artiklen foreslår strategier til at afhjælpe begrænsningerne ved rTMS, såsom at kombinere det med neurobilleddannelsesmetoder og inkludere passende kontroller. Det konkluderes, at styrken af slutninger, der drages fra rTMS-studier, i høj grad afhænger af det eksperimentelle setup, og at rTMS ikke altid er egnet til at besvare spørgsmål vedrørende kausalitet. Artiklen henvender sig til forskere, der ønsker at designe rTMS-studier eller fortolke resultaterne kausalt.

Den anden artikel fokuserer på involveringen af det præfrontale cortex (PFC) i processerne, der ligger til grund for visuel bevidsthed. Mens nogle teorier fremhæver betydningen af PFC som det neurale fundament for visuel bevidsthed, hævder mange forskere, at PFC-aktivitet ikke er afgørende. Derudover antages det, at PFC er tæt forbundet med metakognition. I denne henseende undersøgte den anden artikel rollen af PFC i vurderinger af visuel bevidsthed ved at sammenligne tre forskellige rTMS-protokoller, der blev anvendt på det venstre anteriore mediale præfrontale cortex (aMPFC), et område i hjernen forbundet med metakognition. Der blev anvendt tre forskellige Theta Burst Stimulation (TBS) protokoller: såkaldt "continuous TBS" (cTBS), "intermittent TBS" (iTBS) og "sham TBS". TBS-protokollen blev administreret umiddelbart før en computerbaseret visuel identifikationsopgave hvor forsøgspersonerne rapporterede deres visuelle bevidsthed af forskellige stimuli. Resultaterne indikerede, at cTBS førte til bedre metakognition end sham. Effekten var relateret til lavere vurderinger af visuel bevidsthed for forkerte svar. Der blev ikke observeret signifikante forskelle mellem TBS-betingelserne.

Den tredje artikel adresserer diskussion i bevidsthedsforskningsmiljøet om indvirkningen af ikke-visuel information på vurderinger af visuel bevidsthed. Den beskriver en undersøgelse af,

hvorvidt "enkelt-puls TMS" (spTMS) af den primære motoriske cortex (M1) cirka et halvt sekund efter stimuluspræsentation, kunne fungere som ikke-visuel evidens og dermed påvirke vurderinger af visuel bevidsthed. Det undersøgte også, om amplituden af det motorisk fremkaldte potentiale (MEP), induceret af spTMS, kunne afspejle mængden af akkumuleret evidens for stimulusopfattelse. Magnetstimulationen blev foretaget mens forsøgspersoner udførte en computerbaseret test, hvor de udførte en visuel identifikationsopgave som inkluderede rapport af bevidst oplevelse. Resultaterne viste, at stimulation af M1 ledte til, at visuelle oplevelser blev rapporteret som klarere end i en kontrolsituation. Der blev ikke observeret en signifikant effekt på forsøgspersonernes metakognitive evner. Derudover var MEP-amplituder forbundet med vurderinger af visuel bevidsthed, hvilket tyder på, at MEP kan fungere som en indikator for evidensakkumulering.

Samlet set tyder resultaterne på, at både aMPFC og M1 kan bidrage til vurdering af visuel bevidsthed. Det er dog vigtigt at erkende begrænsningerne ved de anvendte metoder, især TMS, og den komplekse karakter af de undersøgte processer.

9.3. Polish summary

Neuronalne podstawy świadomości wzrokowej badane za pomocą przeczaszkowej stymulacji magnetycznej

W pracach zawartych w niniejszej rozprawie wykorzystano przeczaszkową stymulację magnetyczną (TMS) do badania zaangażowania wybranych obszarów mózgu w proces powstawania sądów dotyczących świadomości wzrokowej. TMS to nieinwazyjna metoda stymulacji mózgu, za pomocą której można tymczasowo wpłynąć na aktywność wybranego obszaru mózgu. Rozprawa składa się z trzech opublikowanych artykułów naukowych. Pierwszy, teoretyczny artykuł, omawia ograniczenia wnioskowania przyczynowego opartego na zastosowaniu TMS i zawiera wskazówki dotyczące sposobu radzenia sobie z tymi ograniczeniami, szczególnie w kontekście repetytywnej TMS (rTMS). Artykuły drugi i trzeci przedstawiają badania eksperymentalne przeprowadzone w schemacie wewnątrzgrupowym. W ich trakcie stymulowano różne obszary mózgu u zdrowych osób w celu zbadania wpływu TMS na sądy dotyczące świadomości wzrokowej i trafność metapoznawczą: zdolność do adekwatnej oceny własnej percepcji.

W przeciwieństwie do metod neuroobrazowania, TMS może wpływać na aktywność komórek nerwowych, co czyni go cennym narzędziem wnioskowania przyczynowego. W pierwszym artykule omówiono możliwości wykorzystania TMS do analizy związków przyczynowych między obszarami mózgu a ich funkcjami. Artykuł przedstawia krytyczną ocenę zakresu, w jakim można dokonywać wnioskowania przyczynowego na podstawie danych z rTMS. Podkreśla, że poleganie wyłącznie na danych rTMS nie zapewnia wystarczających podstaw do formułowania silnych wniosków na temat przyczynowych własności zmiany pobudliwości stymulowanych obszarów mózgu. W artykule zaproponowano strategię radzenia sobie z ograniczeniami rTMS, takie jak łączenie stymulacji z technikami neuroobrazowania i uwzględnienie odpowiednich warunków kontrolnych. Stwierdzono, że konkluzyjność wniosków wyciąganych z badań rTMS w dużej mierze zależy od planu eksperymentalnego, a rTMS nie zawsze może być odpowiednią metodą poszukiwania odpowiedzi na pytania dotyczące powiązań przyczynowo-skutkowych. Artykuł ten jest przeznaczony dla osób, które chcą zaprojektować badania rTMS lub na ich podstawie prowadzić wnioskowanie przyczynowe.

Drugi artykuł dotyczy zaangażowania kory przedczołowej (PFC) w procesy leżące u podstaw świadomości wzrokowej. Podczas gdy niektóre teorie świadomości podkreślają znaczenie PFC dla powstawania świadomości wzrokowej (np. teorie świadomości wyższego rzędu), wielu badaczy twierdzi, że aktywność PFC nie jest niezbędna. Przypuszcza się również, że PFC jest ściśle związana z metapoznaniem. Z tego powodu w pierwszym badaniu zbadano rolę PFC w powstawaniu sądów dotyczących świadomości wzrokowej, porównując trzy różne protokoły rTMS zastosowane do lewej przedniej przyśrodkowej kory przedczołowej (aMPFC) – obszaru mózgu związanego z metapoznaniem, w celu wywołania efektów przypominających neuroplastyczność. W badaniu wykorzystano trzy różne protokoły stymulacji theta-burst (TBS),

a mianowicie ciągłą TBS (cTBS), przerywaną TBS (iTBS) i kontrolną TBS. Każdy protokół TBS stosowano przed testami behawioralnymi obejmującymi procedurę komputerową z zadaniem identyfikacji bodźców wzrokowych i oceną świadomości wzrokowej. Wyniki wskazują, że zastosowanie cTBS prowadzi do wyższej trafności metapoznawczej niż kontrolna TBS. Efekt ten wiązał się z niższymi ocenami świadomości wzrokowej w przypadku nieprawidłowych odpowiedzi. Nie zaobserwowano istotnych różnic między warunkami TBS pod względem poprawności wykonywania zadania identyfikacji ani czasów odpowiedzi (RT).

Trzeci artykuł dotyczy toczącej się w środowisku badaczy świadomości dyskusji na temat wpływu informacji pozapercepcyjnych na sądy dotyczące świadomości wzrokowej. Przedstawia badanie, w którym sprawdzano, czy stymulacja pierwszorzędowej kory ruchowej (M1) około pół sekundy po prezentacji bodźca z wykorzystaniem techniki stymulacji pojedynczymi impulsami TMS (spTMS) może służyć jako dowód pozapercepcyjny w procesie akumulacji dowodów i w konsekwencji wpływać na sądy dotyczące świadomości wzrokowej. W badaniu testowano również, czy amplituda ruchowego potencjału wywołanego (MEP) spTMS może odzwierciedlać ilość zgromadzonych dowodów percepcyjnych. W badaniu postawiono hipotezę, zgodnie z którą zastosowanie spTMS do M1 prowadzi do wyższych ocen świadomości wzrokowej w porównaniu z warunkiem kontrolnym. Założono również, że amplitudy MEP będą reprezentować ilość zgromadzonych dowodów. Procedura eksperymentalna składała się z spTMS oraz procedury komputerowej, obejmującej ocenę świadomości wzrokowej i zadanie identyfikacji bodźców wzrokowych. Wyniki wykazały, że w próbach zgodnych (w których dłoń, z której wykorzystaniem udzielano odpowiedzi w zadaniu identyfikacji, odpowiadała dłoni, której reprezentację stymulowano w warunku M1) zaobserwowano wyższe oceny świadomości wzrokowej oraz dłuższe RT w zadaniu identyfikacji. Dodatkowo niezależnie od zgodności, zaobserwowano dłuższe RT dla ocen świadomości wzrokowej w warunku M1 w porównaniu z warunkiem kontrolnym, co potencjalnie wskazuje na włączanie dodatkowych dowodów w procesie akumulacji dowodów. Nie zaobserwowano istotnych różnic między warunkami w odniesieniu do trafności metapoznawczej. Ponadto amplitudy MEP były powiązane z ocenami świadomości wzrokowej i wykazywały wyższe wartości w próbach zgodnych, co sugeruje, że MEP może służyć jako pośrednia miara procesu akumulacji dowodów.

Podsumowując, rezultaty niniejszych badań wskazują, że zarówno aMPFC, jak i M1 mogą przyczynić się do formowania oceny świadomości wzrokowej. Kluczowe jest jednak uwzględnienie ograniczeń stosowanych metod, zwłaszcza TMS, oraz złożonej natury badanych procesów.