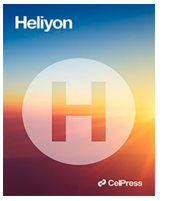
Heliyon 10 (2024) e41118

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon



Research article

Spontaneous and perturbation-based EEG cortical excitability markers are associated with plasma p-tau181 concentration in healthy middle-aged adults

Ruben Perellón-Alfonso [a,b,c,*], Kilian Abellaneda-Pérez [c,d,e], Indre Pileckyte [f], María Cabello-Toscano [a,b], Lídia Mulet-Pons [a,b], Lídia Vaqué-Alcázar [a,b,g], Gabriele Cattaneo [c,d,e], María Redondo-Camós [c,d,e], Goretti España-Irla [h,i,j], Selma Delgado-Gallen [c,d,e], Javier Solana Sánchez [c,d,e], Henrik Zetterberg [k,l,m,n,o,p], Jose M. Tormos [q], Nicolai Franzmeier [r,s,t], Alvaro Pascual-Leone [u,v,w,1], David Bartrés-Faz [a,b,c,**,1]

[a] *Department of Medicine, Faculty of Medicine and Health Sciences, and Institute of Neurosciences, University of Barcelona, Barcelona, Spain*
[b] *Institut d'Investigacions Biomèdiques August Pi i Sunyer (IDIBAPS), Barcelona, Spain*
[c] *Institut Guttmann, Institut Universitari de Neurorehabilitació adscrit a la Universitat Autònoma de Barcelona, Barcelona, Spain*
[d] *Universitat Autònoma de Barcelona, Bellaterra (Cerdanyola del Vallès), Spain*
[e] *Fundació Institut d'Investigació en Ciències de la Salut Germans Trias i Pujol, Badalona, Spain*
[f] *Center for Brain and Cognition, Pompeu Fabra University, Barcelona, Spain*
[g] *Sant Pau Memory Unit, Department of Neurology, Institut d'Investigacions Biomèdiques Sant Pau-Hospital de Sant Pau, Universitat Autònoma de Barcelona, Barcelona, Spain*
[h] *Department of Psychology, Northeastern University, Boston, MA, USA*
[i] *Center for Cognitive & Brain Health, Northeastern University, Boston, MA, USA*
[j] *Department of Physical Therapy, Movement, & Rehabilitation Sciences, Northeastern University, Boston, MA, USA*
[k] *Department of Psychiatry and Neurochemistry, Institute of Neuroscience and Physiology, the Sahlgrenska Academy at the University of Gothenburg, Mölndal, Sweden*
[l] *Clinical Neurochemistry Laboratory, Sahlgrenska University Hospital, Mölndal, Sweden*
[m] *Department of Neurodegenerative Disease, UCL Institute of Neurology, Queen Square, London, UK*
[n] *UK Dementia Research Institute at UCL, London, UK*
[o] *Hong Kong Center for Neurodegenerative Diseases, Clear Water Bay, Hong Kong, China*
[p] *Wisconsin Alzheimer's Disease Research Center, University of Wisconsin School of Medicine and Public Health, University of Wisconsin-Madison, Madison, WI, USA*
[q] *Centro de Investigación Translacional San Alberto Magno - Facultad Ciencias de la Salud - Universidad Católica de Valencia, Spain*
[r] *Institute for Stroke and Dementia Research (ISD), University Hospital, LMU, Munich, Germany*
[s] *Munich Cluster for Systems Neurology (SyNergy), Munich, Germany*
[t] *University of Gothenburg, The Sahlgrenska Academy, Institute of Neuroscience and Physiology, Department of Psychiatry and Neurochemistry, Sweden*
[u] *Hinda and Arthur Marcus Institute for Aging Research and Deanna and Sidney Wolk Center for Memory Health, Hebrew SeniorLife, Boston, MA, USA*
[v] *Department of Neurology, Harvard Medical School, Boston, MA, USA*
[w] *Linus Health, Inc., Boston, MA, USA*

* Corresponding author. Department of Medicine, Faculty of Medicine and Health Sciences, and Institute of Neurosciences, University of Barcelona, Barcelona, Spain.
** Corresponding author. Department of Medicine, Faculty of Medicine and Health Sciences, and Institute of Neurosciences, University of Barcelona, Barcelona, Spain
E-mail address: ruben.perellon@ub.edu (R. Perellón-Alfonso).
[1] Joint senior authors.

https://doi.org/10.1016/j.heliyon.2024.e41118
Received 1 July 2024; Received in revised form 8 December 2024; Accepted 9 December 2024
Available online 10 December 2024

ARTICLE INFO

Keywords:
Alzheimer's disease
Tau
EEG
Transcranial magnetic stimulation

ABSTRACT

In early-stage Alzheimer's disease (AD) amyloid-β (Aβ) deposition can induce neuronal hyperactivity, thereby potentially triggering activity-dependent neuronal secretion of phosphorylated tau (p-tau), ensuing tau aggregation and spread. Therefore, cortical excitability is a candidate biomarker for early AD detection. Moreover, lowering neuronal excitability could potentially complement strategies to reduce Aβ and tau buildup. There is, however, a lack of understanding of the relationship between cortical excitability and p-tau increase *in vivo*. Therefore, in a sample of 658 healthy middle-aged (between the ages of 40 and 65) participants of the Barcelona Brain Health Initiative cohort study, we examined the relation of blood-based tau, phosphorylated at amino acid 181 (p-tau181), reflecting neuronal p-tau secretion; neurofilament light chain (NfL), as a passively released control for p-tau181; and electroencephalography (EEG) markers of cortical excitability. A subsample of 47 participants also completed a controlled brain perturbation approach via transcranial magnetic stimulation (TMS) with concurrent EEG. Results show that both spontaneous (i.e., resting-state) and perturbation-based TMS-EEG markers, are associated with blood p-tau181, particularly in older individuals. The perturbation-based marker was a significantly more sensitive predictor of p-tau181 concentration than the spontaneous resting state EEG-based marker. The relationships observed are not present for the NfL control. These results show that relationships between p-tau181 and cortical excitability are present in healthy middle-aged subjects and that p-tau181 increases may reflect activity-dependent secretion.

1. Introduction

Hyperexcitability is a hallmark of early Alzheimer's disease (AD). Research on human patients and mouse models consistently shows hyperexcitability at the single neuron level, as well as in neural networks or entire brain systems [1,2]. Further evidence from research in mice indicates that extracellular soluble amyloid-beta (Aβ) induces neuronal hyperexcitability [3]. Subsequently, this hyperexcitability promotes the secretion of tau, which propagates through trans-synaptic transmission to affect distal brain regions [4], leading to progressive neuronal degeneration and cognitive decline. Consequently, emerging interventions for AD focus on clearing Aβ to reduce tau pathology and spread [5]. However, given that Aβ may trigger the release of phosphorylated tau by stimulating neuronal excitability, reducing excitability could further enhance the effects of these treatments [6].

The role of cortical excitability in the Aβ-tau axis has been primarily investigated in human patients and animal models of AD. However, recently developed blood-based biomarkers [7], enable research at any stage, including in preclinical healthy populations [8]. Additionally, advances in electroencephalography (EEG) now allow studying cortical excitability non-invasively. This can be achieved either indirectly by capturing spontaneous activity during the resting state [9] or directly through controlled brain perturbations using concurrent transcranial magnetic stimulation ([10]; TMS).

Here, we investigated the relationship between cortical excitability and tau secretion in a healthy middle-aged population, using data from the Barcelona Brain Health Initiative project [11]. We included 648 participants who completed resting-state EEG and provided blood samples to measure plasma phosphorylated tau at amino acid 181 (p-tau181) and neurofilament light (NfL). Additionally, a subsample of 47 participants also underwent single pulse TMS of the left prefrontal cortex and left inferior parietal lobule with concurrent EEG. Based on evidence indicating that excitability might stimulate the secretion and propagation of tau [3,4], we hypothesized that higher excitability would correlate with higher concentration of p-tau181 but not with NfL. Furthermore, we hypothesized that perturbation-based measures would be more sensitive predictors of p-tau181 concentration. This is because spontaneous excitation/inhibition balance reflects a more heterogeneous measure of complex excitatory and inhibitory interactions [12], which precludes a direct estimation of net excitability.

2. RESULTS

2.1. Perturbation-based cortical excitability is positively correlated with secreted p-tau181 in healthy middle-aged adults aged 61 and older

Single-pulse TMS produces a repeatable evoked EEG response, with its earliest and latest components (from 15 to 35 ms and 160–240 ms after the pulse, respectively) directly reflecting voltage-gated sodium channel (VGSC)-mediated excitability, as these can be directly inhibited by VGSC blockers such as carbamazepine [13,14]. Using this *perturbation-based-excitability* measure, we investigated the relationship between cortical excitability and p-tau181 in a healthy middle-aged population.

A linear regression model with a response variable *p-tau181*, and the predictors *age, perturbation-based-excitability*, and their interaction, $F(43, 4) = 5.86$, $p = .002$, $\eta^2 = .29$, revealed a trend level main effect of *perturbation-based-excitability*, $\beta = -.019$, $p = .061$, and a significant interaction between *perturbation-based-excitability* and *age*, β = 3.6e-4, p = .036. A Johnson-Neyman interaction analysis was used to further study this interaction, revealing that *perturbation-based-excitability* significantly predicts p-tau181 concentration starting at age 61 (Fig. 1).

The reported model incorporates the late evoked component only (i.e., 160–240 ms after the pulse), as an equivalent model

incorporating the early evoked component (i.e., 15–35 ms after the pulse) did not reach significance $p = .061$.

Finally, to show that the relationship is specific to potentially secreted p-tau protein rather than passively released proteins, we fitted a linear regression model with *NfL* as the response variable, $F(43, 4) = 6.32$ -value, $p = .001$, $\eta^2 = .31$. This model revealed no significant main effects of *age*, $p = .109$, or *perturbation-based excitability*, $p = .649$, nor a significant interaction between them, $p = .556$.

2.2. Spontaneous cortical excitation/inhibition balance is positively correlated with secreted p-tau181 in healthy middle-aged adults aged 54 and older

Recent studies have shown that the EEG power spectrum's $1/f$-like aperiodic activity partially reflects the overall cortical balance of excitation and inhibition [9,15]. Abnormalities in this measure have been consistently reported [16] and have been linked to cognitive function [17]. Therefore, we used this *spontaneous-excitation/inhibition* measure to further investigate the relationship between cortical excitability and p-tau181 concentration.

A general linear regression model with *p-tau181* as the response variable, and predictors including *age, spontaneous-excitation/inhibition*, and their interaction, $F(644, 4) = 5.07$, $p = .002$, $\eta^2 = .03$, revealed a trend level main effect of *spontaneous-excitation/inhibition*, $\beta = -.45$, $p = .060$, and a significant interaction between *spontaneous-excitation/inhibition* and *age*, $\beta = .009$, $p = .029$. A Johnson-Neyman interaction analysis was used to further study this interaction, revealing that *spontaneous-excitation/inhibition* significantly predicts p-tau181 concentration beginning at age 54 (Fig. 2).

To show that the relationship is specific to potentially secreted p-tau protein, rather than passively released proteins, we fitted a linear regression model with *NfL* as the response variable, $F(644, 4) = 62.9$ -value, $p < .001$, $\eta^2 = .22$. This model revealed a significant main effect of *age*, $\beta = -.021$ $p < .001$, but no significant main effect of *spontaneous-excitability*, $p = .339$, or interaction between *age* and *spontaneous-excitability*, $p = .311$.

2.3. Perturbation based excitability is a better predictor of p-tau181 concentration than spontaneous excitation/inhibition balance

To compare the predictive value of perturbation-based excitability against spontaneous excitation/inhibition balance, we fitted a full linear regression model with *p-tau181* as the response variable. Predictors included *age, perturbation-based-excitability*, their interaction, *spontaneous-excitation/inhibition*, and its interaction with age $F(41, 6) = 3.627$, $p = .008$, $\eta^2 = .31$. The model revealed a trend level main effect of *perturbation-based excitability*, $\beta = -.021$, $p = .051$, and a significant interaction between *perturbation-based excitability* and *age*, $\beta = 3.8\text{e-}04$ $p = .031$. However, there was no significant main effect of *spontaneous-excitation/inhibition*, $p = .834$, or its interaction with *age*, $p = .926$.

Model comparisons between the full model and the reduced model (which included only perturbation-based excitability, age, and their interaction as predictors) revealed that the reduced model provides a better fit for *p-tau181*. The full model did not significantly improve the fit compared to the reduced model, $p = .616$. Additionally, smaller Akaike Information Criterion (AIC) and Bayesian Information Criterion (BIC) values, along with a larger Bayes factor, further support the reduced model as the better fit (Table 1).

3. Discussion

We established the relationship between cortical excitability and secreted p-tau in a healthy middle-aged population using non-invasive and potentially scalable methods.

As hypothesized, the more sensitive and direct marker of cortical excitability, induced by TMS perturbation, indicates that higher cortical excitability is associated with higher p-tau181 concentration. This finding aligns with the recently shown synergic relationship between Aβ and tau [18], where the presence of Aβ renders tau's inhibitory effect on neuronal activity ineffective [19]. However, the absence of a selective Aβ biomarker in this study prevents us from directly testing this hypothesis. Nonetheless, it is reasonable to

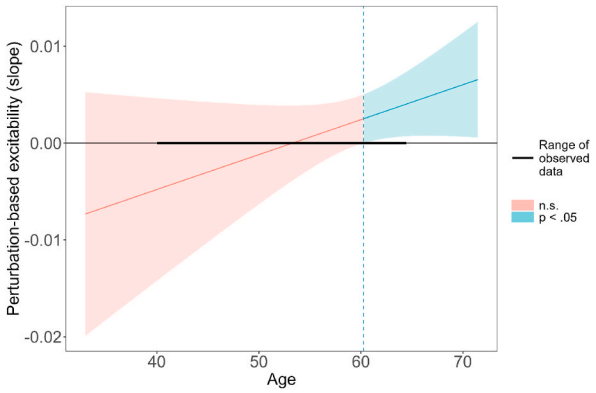


Fig. 1. Johnson-Neyman interval plot illustrating the age interval at which the relationship between perturbation-based excitability and p-tau181 is significant (i.e., 61–65 years of age). n.s. = non-significant.

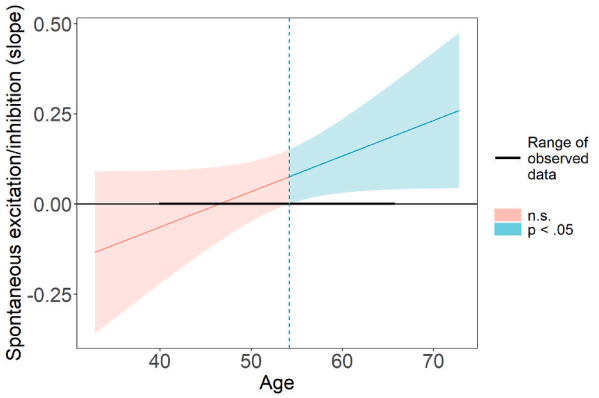


Fig. 2. Johnson-Neyman interval plot illustrating the age interval at which the relationship between spontaneous excitability and p-tau181 is significant (i.e., 54–65 years of age). n.s. = non-significant.

Table 1
Model comparison results of the full (spontaneous + perturbation) Vs reduced (perturbation only) model.

Regression Model	*AIC*	*BIC*	*Bayes Factor*	*p*	η^2
Spontaneous + Perturbation	181.8	194.7	.03	.616	.31
Perturbation only	178.9	188.1	26.9		.29

expect that increases in p-tau181 are accompanied by increases in Aβ concentration, as higher blood p-tau181 levels have been consistently correlated with Aβ accumulation in cerebrospinal fluid and positron emission tomography [20–22]

Conversely, the results also show that cortical excitation/inhibition balance is positively correlated with p-tau181 concentration. Computational modeling and evidence from research in mice and macaques [9], suggest that steeper slopes in the aperiodic component of the power spectrum might indicate a shift in balance toward inhibition. In this context, our results suggest that a greater shift toward inhibition (i.e., steeper slopes) is associated with higher p-tau181 concentration.

While this may appear to contrast with the findings obtained from the perturbation-based model, it is important to note key differences between the two measures. The perturbation-based marker is location specific and, more importantly, specific to VGSC excitability, more directly reflecting the net excitability of pyramidal neurons at the stimulation site [13,14]. In contrast, the spontaneous marker is an heterogeneous, global measure likely capturing the complex interplay of multiple excitatory and inhibitory neurotransmission pathways across the cortex [12,23]. As such, it lacks the spatial and neurophysiological specificity of the perturbation-based metric. Therefore, the relationship observed between spontaneous and perturbation-based metrics is not directly comparable. The spontaneous measure may not be sensitive enough to detect the localized increases in excitability that the perturbation-based measure reveals as associated with higher p-tau181 concentration.

Interestingly, our results show that relationship between excitability and p-tau depends on age, becoming significant in participants aged over 54 and 61 years for spontaneous and perturbation-based excitability, respectively. There is ample evidence that in normal aging there is measurable structural and functional brain deterioration of the brain [24,25]. encompassing biochemical, metabolic, cellular, and molecular changes [26]. Therefore, a possible explanation for the non-linear associations we report, is that, by late middle age, normal aging-related brain deterioration may reach a critical threshold where aberrant interactions between Aβ and p-tau begin to manifest. These interactions may partly present as changes in cortical excitability. Supporting this notion, recent research in the same BBHI cohort also shows that even in the absence of pathology, subtle declines in dual-task gait performance become apparent only after 54 years of age [27].

We have also shown that the relationships observed between cortical excitability markers and p-tau do not hold for NfL. This highlights that the presented markers of cortical excitability are probably related to secreted protein passing to the blood stream when dissolved, rather than being passively released as a byproduct of axonal damage and cell death. Although both NfL and tau can be either passively released or secreted into the extracellular space, tau secretion has been observed in a regulated manner in healthy and pathological conditions [28], while passive release of tau has been mostly observed as a byproduct of cell death or injury after an acute stroke [29]. Conversely, NfL concentration have been shown to increase in CSF and blood proportionally to the degree of axonal damage, both in normal aging and in neurodegenerative diseases [30], suggesting that NfL is mostly passively released as a byproduct of cell injury.

3.1. Limitations and future directions

This study has limitations, including the absence of a selective Aβ biomarker and the exclusive focus on middle-aged subjects. These constraints prevent us from exploring how the described relationships progress into older ages and pathological stages. Nevertheless,

we have demonstrated, for the first time, that cortical excitability changes are related to p-tau concentration beginning in late middle age, even in the absence of obvious amyloid and tau pathology or cognitive decline. Future research should incorporate selective Aβ biomarkers and longitudinal designs to investigate how the observed relationships evolve with aging and during the pathological stages of neurodegenerative diseases.

Research in preclinical healthy populations is essential for identifying individuals at risk of developing neurodegenerative diseases and for implementing early interventions to delay or prevent disease progression. Such interventions could include modifiable lifestyle changes or targeted therapies aimed at reducing Aβ and p-tau buildup. In this regard, we have shown that readily available blood-based biomarkers and non-invasive electrophysiology are valuable tools for studying associations between cortical excitability and proteins implicated in neurodegenerative pathophysiology. This is important not only for early disease detection but also for providing neurophysiological insights into the relationship between p-tau and cortical excitability.

Cortical excitability represents a potentially modifiable and complementary target for early intervention, as it can be effectively reduced using pharmacological [31] or non-invasive brain stimulation interventions [32,33]. Exploring the efficacy of such targeted interventions to disrupt the Aβ, p-tau and excitability axis in at-risk preclinical populations could provide promising strategies for early prevention and mitigation.

4. Materials and methods

4.1. Study participants

Participants were recruited from the Barcelona Brain Health Initiative project (BBHI; [11]) an ongoing longitudinal study that investigates brain health determinants in middle-aged adults and started recruiting participants in 2017. All participants included in the BBHI were free from any self-reported neurological or psychiatric diagnosis at the time of recruitment. For the present study we selected all participants who completed both blood extraction and resting-state EEG, and whose data survived preprocessing. The final sample included consists of 648 subjects aged 40–65 years, $M = 52.3$ $SD = 7.2$, 307 female. Additionally, a subsample of 47 participants (aged 40–64 years, $M = 54.8$ $SD = 7.1$, 16 female) completed TMS with concurrent EEG (from now on TMS-EEG). In addition, participants' age and biological sex were also collected and used as covariates in statistical analysis. The BBHI cohort, including details on inclusion and exclusion criteria, is described in greater detail elsewhere [11].

4.2. Ethics and consent

The study protocols were approved by the ethics committee: Comitè Ètic d'Investigació (CEIm) de la Fundació Unió Catalana d'Hospitals (ref. CEIC 17/06) and all participants gave their informed consent to participate in accordance with the declaration of Helsinki.

4.3. Blood-based biomarkers

Ethylenediaminetetraacetic acid plasma samples were collected through venipuncture.

Plasma concentrations of phosphorylated p-tau181 and NfL were measured using Single molecule array (Simoa) methods on an HD-X instrument (Quanterix, Billerica, MA, USA), as previously described [34,35].

4.4. TMS-EEG recording and preprocessing

TMS was applied over the left dorsolateral prefrontal cortex (L-PFC) and the left inferior parietal lobule (L-IPL). Stimulation was guided by a BrainSight neuronavigation system (RogueResearch, Inc., Canada). The targets were determined based on either anatomy –for subjects recorded before 2019 ($n = 27$)– or the functional cortical parcellation by Yeo and colleagues [36] –for subjects studied after 2019 ($n = 20$). See supplementary materials for MRI acquisition parameters and detailed target determination procedures. Stimulation was set to 120 % of resting motor threshold, determined as the lowest intensity needed to produce motor evoked potentials of no less than 50 μV peak-to-peak in the first dorsal interosseous muscle on the relaxed right hand, achieving this in a minimum of 3 out of 6 attempts [37]. For each designated target, a series of 120 single biphasic pulses were administered using an MCF-B65 butterfly coil attached to a MagPro X100 stimulator (Magventure, Inc., Denmark). Pulses were spaced randomly, between four to 6 s apart. The sequence in which targets were stimulated was shuffled for every participant. To diminish the auditory responses triggered by the click from the TMS coil, participants were equipped with earplug-earbuds emitting white noise at their maximum tolerated volume. EEG was recorded concurrently using a TMS-compatible ActiChamp 64-channel amplifier system, paired with an ActiCap Slim featuring active electrodes (from BrainProducts, GmbH., Germany). Electrode impedance was consistently monitored and maintained below 5kΩ throughout the recording. EEG data was captured from DC to 500Hz and converted into a digital format at a rate of 1 KHz.

Data was preprocessed using functions from the EEGLAB toolbox [38] and the TESA plugin [39]. Briefly, data was first segmented around the TMS pulse (−1000 to 1000 ms from the pulse) and baseline corrected (−900 ms to −100 ms from the pulse). Then the direct electrical pulse artifact (between −2 ms and 14 ms from pulse) was zero-padded. Bad channels were then identified via visual inspection and removed. Bad epochs were visually tagged and then rejected with the aid of voltage thresholding (>100 μV), probability and kurtosis. A first round of fast independent component analysis (ICA) was used to reject the initial large amplitude muscular artifact. The zero-padded pulse artifact was then linearly interpolated, data was re-referenced to the average of all channels and

previously removed channels were spherically interpolated. Finally, a second round of ICA was used to reject any other remaining artifacts (e.g., muscle, eye-movements, heartbeat and others), as well as the somatosensory and auditory potentials evoked by transcutaneous scalp nerve excitation and coil firing sounds and vibrations. The preprocessed TMS-evoked potential data was then used for source localization (see supplementary materials for source localization procedures).

4.5. Resting-state EEG recording and preprocessing

EEG was recorded for 10 min at rest (i.e., 5 min eyes closed, 5 min eyes open) using an Enobio 32 channel system (Neuroelectrics, Spain) at a sampling rate of 512 Hz with a 50Hz notch filter of order 1. Electrode impedance for all channels was kept below 25 kΩ. We included only eyes closed data in the analysis, as it contains less eye and muscle related artifacts, and hence guarantees a higher survival rate and quality during automatic data preprocessing.

Data was preprocessed using a fully automated pipeline developed in house (see supplementary materials for pipeline script and details) and consisting of EEGLAB [38] and custom-made MATLAB functions (The MathWorks INC. USA). The pipeline ran through all the 748 EEG resting-state datasets and logged the number of bad channels and epochs. For the current analysis we kept only those datasets that retained at least 22 out of 32 channels and 100 artifact free epochs, resulting in 648 individual clean recordings for subsequent statistical analysis.

4.6. TMS-EEG perturbation based cortical excitability

To quantify VGSC mediated excitability at each stimulation target location we first defined a region of interest (ROI) of 100 vertices around each subject's stimulation target coordinate, which corresponds to a cortex surface area of approximately 10 cm^2. To allow group level statistics, the TMS-evoked potential (TEP) time-series in source space of each vertex within the target ROI were rectified, averaged together, and then normalized via z-score transformation:

$$z=(TEP-\mu)/\sigma$$

Where μ is the average of the pre-stimulus baseline (from −500 ms to −3 ms relative to the TMS pulse) and σ is the standard deviation of the baseline. Finally, we computed the trapezoidal integration (i.e., area under the curve) of the normalized TEP for the early and late components (i.e., from 15 to 35 ms and from 160 to 240 ms after the TMS pulse (Fig. 3A and B).

4.7. Spontaneous resting state EEG cortical excitation/inhibition balance

To estimate excitation/inhibition balance, first the EEG power spectrum was computed via the Welch's method, whereby the fast Fourier transform of each 2 s epoch is computed and then all epochs' spectra are averaged together. The aperiodic component of the EEG spectrum (i.e., $1/f$ -like activity) was estimated using the *fooof* toolbox [15], and then the average (i.e., across all channels) exponent (i.e., slope) of the aperiodic fit line was taken as the estimate of $1/f$ -like activity and, hence, as a proxy of cortical excitation/inhibition balance [9], with steeper slopes (i.e., higher values) indicating a shift in the balance towards inhibition and vice-versa

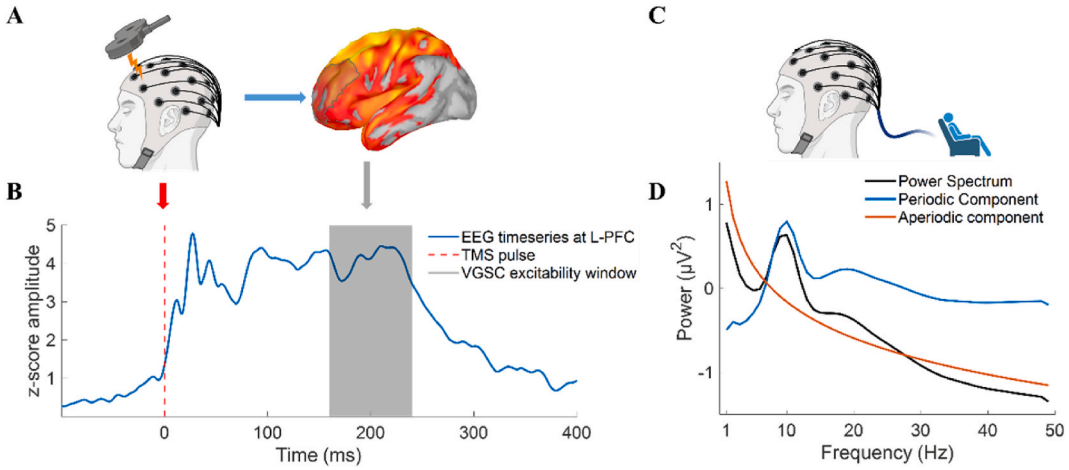


Fig. 3. Illustration of EEG cortical excitability markers. A) illustrates the TMS-evoked perturbation of the EEG and the spatial region of interest in source space (grey transparent patch over L-PFC) and, B) the time-window taken from the TMS-evoked EEG timeseries to compute the perturbation-based marker of VGSC excitability (grey transparency spanning from 160 to 240 ms after TMS pulse). C) illustrates the recording of resting state EEG and, D) the decomposition of the power spectrum at rest to isolate the aperiodic component, the slope of which we take here as the spontaneous marker for excitation/inhibition balance. In both B and D panels, grand average waveforms for all participants included in the study are shown. EEG, electroencephalography; L-PFC, left prefrontal cortex; VGSC, voltage-gated sodium channel.

(Fig. 3C and D).

4.8. Statistical analysis

All statistical analyses were performed in R version 4.2.3 (R Foundation for Statistical Computing, Vienna, Austria).

To determine the relation of perturbation-based VGSC mediated excitability and p-tau181 in the TMS-EEG subsample ($n = 47$), we used stepwise general linear modeling –for both the early and late perturbation-based response (i.e., from 15 to 35 ms and 160–240 ms after the pulse, respectively)– to determine the best fitting model and discard irrelevant predictor terms. The criterion for removing terms was the models' chi-squared test of the change in the deviance that results from removing the term. The starting model included all main effects and interactions of perturbation-based excitability (at both L-IPL and L-PFC targets), age, biological sex and TMS targeting method (i.e., functional or anatomical). The final, best-fitting model is reported in the results section. It includes the main effects of age and late VGSC excitability at the L-PFC target, as well as their interaction.

To determine the relationship between spontaneous excitation/inhibition balance during resting-state EEG and p-tau181 concentration in the full sample (n = 648), we used stepwise general linear modeling in a similar manner. The starting model included all possible main effects and interactions of spontaneous excitability, age, and biological sex. The final model reported in the results section included the main effects of age and spontaneous excitability, as well as their interaction. Due to the gamma-like distribution of p-tau181 values in this sample, models were fitted with a gamma distribution and a "log" link function.

To compare the predictive value of spontaneous excitation/inhibition balance and perturbation-based excitability, we used an additional model including the predictors from both models.

Finally, to demonstrate that cortical excitability markers are more likely associated with secreted proteins entering the bloodstream rather than passively released proteins, we contrasted these models by using NfL as the response variable in place of p-tau181.

CRediT authorship contribution statement

Ruben Perellón-Alfonso: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Methodology, Formal analysis, Data curation, Conceptualization. **Kilian Abellaneda-Pérez:** Writing – review & editing, Methodology, Conceptualization. **Indre Pileckyte:** Writing – review & editing, Methodology, Conceptualization. **María Cabello-Toscano:** Writing – review & editing, Formal analysis, Data curation. **Lídia Mulet-Pons:** Writing – review & editing, Data curation. **Lídia Vaqué-Alcázar:** Writing – review & editing, Data curation. **Gabriele Cattaneo:** Writing – review & editing, Conceptualization. **María Redondo-Camós:** Methodology, Investigation. **Goretti España-Irla:** Methodology, Investigation. **Selma Delgado-Gallen:** Methodology, Investigation. **Javier Solana Sánchez:** Writing – review & editing, Funding acquisition, Conceptualization. **Henrik Zetterberg:** Writing – review & editing, Methodology. **Jose M. Tormos:** Project administration, Funding acquisition, Conceptualization. **Nicolai Franzmeier:** Writing – review & editing, Visualization, Validation, Methodology. **Alvaro Pascual-Leone:** Writing – review & editing, Project administration, Funding acquisition, Conceptualization. **David Bartrés-Faz:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Funding acquisition, Conceptualization.

Data availability

The script used to preprocess the resting state EEG data is available as a supplementary material. The data that support the findings of this study are available from the corresponding author, upon reasonable request.

Funding

R.P-A. was supported by a fellowship from "la Caixa" Foundation (ID 100010434; Fellowship code: LCF/BQ/DI19/11730050).

K.A.-P. was financially supported by a Juan de la Cierva research grant (FJC2021-047380-I) of the Spanish Ministry of Science and Innovation.

I.P. was supported by a fellowship from "la Caixa" Foundation (ID 100010434; fellowship code LCF/BQ/DI18/11660026).

L.V.-A. was supported by a Margarita Salas junior postdoctoral fellowship from NextGenerationUE.

A.P-L. was partly supported by the Barcelona Brain Health Initiative, National Institutes of Health (R01AG076708, R01AG059089, R03AG072233), and the Bright Focus Foundation.

H.Z. is a Wallenberg Scholar supported by grants from the Swedish Research Council (#2023-00356; #2022-01018 and #2019–02397), the European Union's Horizon Europe research and innovation programme under grant agreement No 101053962, Swedish State Support for Clinical Research (#ALFGBG-71320), the Alzheimer Drug Discovery Foundation, USA (#201809–2016862), the AD Strategic Fund and the Alzheimer's Association (#ADSF-21-831376-C, #ADSF-21-831381-C, #ADSF-21-831377-C, and #ADSF-24-1284328-C), the Bluefield Project, Cure Alzheimer's Fund, the Olav Thon Foundation, the Erling-Persson Family Foundation, Stiftelsen för Gamla Tjänarinnor, Hjärnfonden, Sweden (#FO2022-0270), the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 860197 (MIRIADE), the European Union Joint Programme – Neurodegenerative Disease Research (JPND2021-00694), the National Institute for Health and Care Research University College London Hospitals Biomedical Research Centre, and the UK Dementia Research Institute at UCL (UKDRI-1003).

This project has received funding from the European Union's Horizon 2020 Research and Innovation Programme under the Marie Skłodowska-Curie grant agreement No. 713673.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Alvaro Pascual-Leone reports a relationship with Neuroelectrics that includes: consulting or advisory. Alvaro Pascual-Leone reports a relationship with Magstim Inc. that includes: consulting or advisory. Alvaro Pascual-Leone reports a relationship with TetraNeuron that includes: consulting or advisory. Alvaro Pascual-Leone reports a relationship with Skin2Neuron that includes: consulting or advisory. Alvaro Pascual-Leone reports a relationship with MedRhythms that includes: consulting or advisory. Alvaro Pascual-Leone reports a relationship with Hearts-Radiant that includes: consulting or advisory. Henrik Zetterberg reports a relationship with AbbVie that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Acumen that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Alector that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Alzinova that includes: consulting or advisory. Henrik Zetterberg reports a relationship with ALZPath that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Annexon that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Apellis that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Artery Therapeutics that includes: consulting or advisory. Henrik Zetterberg reports a relationship with AZTherapies that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Cognito Therapeutics that includes: consulting or advisory. Henrik Zetterberg reports a relationship with CogRx that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Denali that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Eisai that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Merry Life that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Nervgen that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Novo Nordisk that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Optoceutics that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Passage Bio, Inc. that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Pinteon Therapeutics Inc that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Prothena that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Red Abbey Labs that includes: consulting or advisory. Henrik Zetterberg reports a relationship with reMYND that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Roche that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Samumed that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Siemens Healthineers that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Triplet Therapeutics Inc that includes: consulting or advisory. Henrik Zetterberg reports a relationship with Wave that includes: consulting or advisory. David Bartres-Faz reports a relationship with Linus Health that includes: consulting or advisory. A.P-L is co-founder of TI solutions and co-founder and chief medical officer of Linus Health where he has shares and share-options. A.P-L is listed as an inventor on several issued and pending patents on the real-time integration of transcranial magnetic stimulation with electroencephalography and magnetic resonance imaging, and applications of noninvasive brain stimulation in various neurological disorders; as well as digital biomarkers of cognition and digital assessments for early diagnosis of dementia.

H.Z. has given lectures in symposia sponsored by Alzecure, Biogen, Cellectricon, Fujirebio, Lilly, and Roche, and is a co-founder of Brain Biomarker Solutions in Gothenburg AB (BBS), which is a part of the GU Ventures Incubator Program (outside submitted work). If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

Authors thank the Barcelona Brain Health Initiative project participants, whose invaluable contribution made this research possible.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e41118.

References

[1] H. Targa Dias Anastacio, N. Matosin, L. Ooi, Neuronal hyperexcitability in Alzheimer's disease: what are the drivers behind this aberrant phenotype? Transl. Psychiatry 12 (1) (2022) https://doi.org/10.1038/s41398-022-02024-7.

[2] S. Zadey, S.S. Buss, K. McDonald, D.Z. Press, A. Pascual-Leone, P.J. Fried, Higher motor cortical excitability linked to greater cognitive dysfunction in Alzheimer's disease: results from two independent cohorts, Neurobiol. Aging 108 (2021) 24–33, https://doi.org/10.1016/j.neurobiolaging.2021.06.007.

[3] M.A. Busche, X. Chen, H.A. Henning, J. Reichwald, M. Staufenbiel, B. Sakmann, A. Konnerth, Critical role of soluble amyloid-β for early hippocampal hyperactivity in a mouse model of Alzheimer's disease, Proceedings of the National Academy of Sciences of the United States of America 109 (22) (2012) 8740–8745, https://doi.org/10.1073/pnas.1206171109.

[4] M.K. Schultz, R. Gentzel, M. Usenovic, C. Gretzula, C. Ware, S. Parmentier-Batteur, J.B. Schachter, H.A. Zariwala, Pharmacogenetic neuronal stimulation increases human tau pathology and trans-synaptic spread of tau to distal brain regions in mice, Neurobiol. Dis. 118 (July) (2018) 161–176, https://doi.org/10.1016/j.nbd.2018.07.003.

[5] Y. Zhang, H. Chen, R. Li, K. Sterling, W. Song, Amyloid β-based therapy for Alzheimer's disease: challenges, successes and future, Signal Transduct. Targeted Ther. 8 (1) (2023) 1–26, https://doi.org/10.1038/s41392-023-01484-7.

[6] A. Kondo, K. Shahpasand, R. Mannix, J. Qiu, J. Moncaster, C.H. Chen, Y. Yao, Y.M. Lin, J.A. Driver, Y. Sun, S. Wei, M.L. Luo, O. Albayram, P. Huang, A. Rotenberg, A. Ryo, L.E. Goldstein, A. Pascual-Leone, A.C. McKee, K.P. Lu, Antibody against early driver of neurodegeneration cis P-tau blocks brain injury and tauopathy, Nature 523 (7561) (2015) 431–436, https://doi.org/10.1038/nature14658.

[7] C.E. Teunissen, I.M.W. Verberk, E.H. Thijssen, L. Vermunt, O. Hansson, H. Zetterberg, W.M. van der Flier, M.M. Mielke, M. Del Campo, Blood-based biomarkers for Alzheimer's disease: towards clinical implementation, Lancet Neurol. 21 (1) (2022) 66–77.

[8] N. Mattsson-Carlgren, G. Salvadó, N.J. Ashton, P. Tideman, E. Stomrud, H. Zetterberg, R. Ossenkoppele, T.J. Betthauser, K.A. Cody, E.M. Jonaitis, R. Langhough, S. Palmqvist, K. Blennow, S. Janelidze, S.C. Johnson, O. Hansson, Prediction of longitudinal cognitive decline in preclinical alzheimer disease using plasma biomarkers, JAMA Neurol. 80 (4) (2023) 360–369, https://doi.org/10.1001/jamaneurol.2022.5272.

[9] R. Gao, E.J. Peterson, B. Voytek, Inferring synaptic excitation/inhibition balance from field potentials, Neuroimage 158 (March) (2017) 70–78, https://doi.org/10.1016/j.neuroimage.2017.06.078.

[10] G. Darmani, U. Ziemann, Pharmacophysiology of TMS-evoked EEG potentials: a mini-review, Brain Stimul. 12 (3) (2019) 829–831, https://doi.org/10.1016/j.brs.2019.02.021.

[11] G. Cattaneo, D. Bartrés-Faz, T.P. Morris, J.S. Sánchez, D. Macià, C. Tarrero, J.M. Tormos, A. Pascual-Leone, The Barcelona brain health initiative: a cohort study to define and promote determinants of brain health, Front. Aging Neurosci. 10 (OCT) (2018), https://doi.org/10.3389/fnagi.2018.00321.

[12] J. Ahmad, C. Ellis, R. Leech, B. Voytek, P. Garces, E. Jones, J. Buitelaar, E. Loth, F.P. dos Santos, A.F. Amil, P.F.M.J. Verschure, D. Murphy, G. McAlonan, From mechanisms to markers: novel noninvasive EEG proxy markers of the neural excitation and inhibition system in humans, Transl. Psychiatry 12 (1) (2022), https://doi.org/10.1038/s41398-022-02218-z.

[13] G. Darmani, T.O. Bergmann, C. Zipser, D. Baur, F. Müller-Dahlhaus, U. Ziemann, Effects of antiepileptic drugs on cortical excitability in humans: a TMS-EMG and TMS-EEG study, Hum. Brain Mapp. 40 (4) (2019) 1276–1289, https://doi.org/10.1002/hbm.24448.

[14] I. Premoli, A. Biondi, S. Carlesso, D. Rivolta, M.P. Richardson, Lamotrigine and levetiracetam exert a similar modulation of TMS-evoked EEG potentials, Epilepsia 58 (1) (2017) 42–50, https://doi.org/10.1111/epi.13599.

[15] T. Donoghue, M. Haller, E.J. Peterson, P. Varma, P. Sebastian, R. Gao, T. Noto, A.H. Lara, J.D. Wallis, R.T. Knight, A. Shestyuk, B. Voytek, Parameterizing neural power spectra into periodic and aperiodic components, Nat. Neurosci. 23 (12) (2020) 1655–1665, https://doi.org/10.1038/s41593-020-00744-x.

[16] M. Kopčanová, L. Tait, T. Donoghue, G. Stothart, L. Smith, A.A. Flores Sandoval, P. Davila-Perez, S. Buss, M.M. Shafi, A. Pascual-Leone, P.J. Fried, C.S. Y. Benwell, Resting-state EEG signatures of Alzheimer's disease are driven by periodic but not aperiodic changes, bioRxiv : The Preprint Server for Biology (2023) 190, https://doi.org/10.1101/2023.06.11.544491. July 2023).

[17] C.S.Y. Benwell, P. Davila-Pérez, P.J. Fried, R.N. Jones, T.G. Travison, E. Santarnecchi, A. Pascual-Leone, M.M. Shafi, EEG spectral power abnormalities and their relationship with cognitive dysfunction in patients with Alzheimer's disease and type 2 diabetes, Neurobiol. Aging 85 (2020) 83–95, https://doi.org/10.1016/j.neurobiolaging.2019.10.004.

[18] M.A. Busche, B.T. Hyman, Synergy between amyloid-β and tau in Alzheimer's disease, Nat. Neurosci. 23 (10) (2020) 1183–1193, https://doi.org/10.1038/s41593-020-0687-6.

[19] S.L. DeVos, B.T. Corjuc, C. Commins, S. Dujardin, R.N. Bannon, D. Corjuc, B.D. Moore, R.E. Bennett, M. Jorfi, J.A. Gonzales, P.M. Dooley, A.D. Roe, R. Pitstick, D. Irimia, M.P. Frosch, G.A. Carlson, B.T. Hyman, Tau reduction in the presence of amyloid-β prevents tau pathology and neuronal death in vivo, Brain 141 (7) (2018) 2194–2212, https://doi.org/10.1093/brain/awy117.

[20] E.R. McGrath, A.S. Beiser, A. O'Donnell, Q. Yang, S. Ghosh, M.M. Gonzales, J.J. Himali, C.L. Satizabal, K.A. Johnson, R.P. Tracy, S. Seshadri, Blood phosphorylated tau 181 as a biomarker for amyloid burden on brain PET in cognitively healthy adults, J. Alzheim. Dis. 87 (4) (2022) 1517–1526, https://doi.org/10.3233/JAD-215639.

[21] X.N. Shen, Y.Y. Huang, S.D. Chen, Y. Guo, L. Tan, Q. Dong, J.T. Yu, M.W. Weiner, P. Aisen, R. Petersen, C.R. Jack, W. Jagust, J.Q. Trojanowki, A.W. Toga, L. Beckett, R.C. Green, A.J. Saykin, J.C. Morris, R.J. Perrin, M. Zmuda, Plasma phosphorylated-tau181 as a predictive biomarker for Alzheimer's amyloid, tau and FDG PET status, Transl. Psychiatry 11 (1) (2021) 1–10, https://doi.org/10.1038/s41398-021-01709-9.

[22] J. Therriault, M. Vermeiren, S. Servaes, C. Tissot, N.J. Ashton, A.L. Benedet, T.K. Karikari, J. Lantero-Rodriguez, W.S. Brum, F.Z. Lussier, G. Bezgin, J. Stevenson, N. Rahmouni, P. Kunach, Y.T. Wang, J. Fernandez-Arias, K.Q. Socualaya, A.C. Macedo, J.P. Ferrari-Souza, P. Rosa-Neto, Association of phosphorylated tau biomarkers with amyloid positron emission tomography vs tau positron emission tomography, JAMA Neurol. 80 (2) (2023) 188–199, https://doi.org/10.1001/jamaneurol.2022.4485.

[23] B. Voytek, R.T. Knight, Dynamic network communication as a unifying neural basis for cognition, development, aging, and disease, Biol. Psychiatr. 77 (12) (2015) 1089–1097, https://doi.org/10.1016/j.biopsych.2015.04.016.

[24] A.M. Fjell, K.B. Walhovd, Structural brain changes in aging: courses, causes and cognitive consequences, Rev. Neurosci. 21 (3) (2010) 187–221, https://doi.org/10.1515/REVNEURO.2010.21.3.187.

[25] D. Tomasi, N.D. Volkow, Aging and functional brain networks, Mol. Psychiatr. 17 (5) (2012) 549–558, https://doi.org/10.1038/mp.2011.81.

[26] J. Lee, H.J. Kim, Normal aging induces changes in the brain and neurodegeneration progress: review of the structural, biochemical, metabolic, cellular, and molecular changes, Front. Aging Neurosci. 14 (June) (2022) 1–15, https://doi.org/10.3389/fnagi.2022.931536.

[27] J. Zhou, G. Cattaneo, W. Yu, O.Y. Lo, N.A. Gouskova, S. Delgado-Gallén, M. Redondo-Camós, G. España-Irla, J. Solana-Sánchez, J.M. Tormos, L.A. Lipsitz, D. Bartrés-Faz, A. Pascual-Leone, B. Manor, The age-related contribution of cognitive function to dual-task gait in middle-aged adults in Spain: observations from a population-based study, The Lancet Healthy Longevity 4 (3) (2023) e98–e106, https://doi.org/10.1016/S2666-7568(23)00009-0.

[28] M. Merezhko, R. Uronen, H.J. Huttunen, The Cell Biology of Tau Secretion 13 (September) (2020) 1–20, https://doi.org/10.3389/fnmol.2020.569818.

[29] H. Hampel, K. Blennow, L.M. Shaw, Y.C. Hoessler, H. Zetterberg, J.Q. Trojanowski, Total and phosphorylated tau protein as biological markers of Alzheimer ' s disease, Exp. Gerontol. 45 (1) (2010) 30–40, https://doi.org/10.1016/j.exger.2009.10.010.

[30] L. Gaetani, K. Blennow, P. Calabresi, M. Di Filippo, L. Parnetti, H. Zetterberg, Neurofilament light chain as a biomarker in neurological disorders, Journal of Neurology, Neurosurgery and Psychiatry (2019) 870–881, https://doi.org/10.1136/jnnp-2018-320106.

[31] C. Meisel, A. Schulze-Bonhage, D. Freestone, M.J. Cook, P. Achermann, D. Plenz, Intrinsic excitability measures track antiepileptic drug action and uncover increasing/decreasing excitability over the wake/sleep cycle, Proceedings of the National Academy of Sciences of the United States of America 112 (47) (2015) 14694–14699, https://doi.org/10.1073/pnas.1513716112.

[32] E. Houdayer, A. Degardin, F. Cassim, P. Bocquillon, P. Derambure, H. Devanne, The effects of low- and high-frequency repetitive TMS on the input/output properties of the human corticospinal pathway, Exp. Brain Res. 187 (2) (2008) 207–217, https://doi.org/10.1007/s00221-008-1294-z.

[33] Y.Z. Huang, M.J. Edwards, E. Rounis, K.P. Bhatia, J.C. Rothwell, Theta burst stimulation of the human motor cortex, Neuron 45 (2) (2005) 201–206, https://doi.org/10.1016/j.neuron.2004.12.033.

[34] M. Gisslén, R.W. Price, U. Andreasson, N. Norgren, S. Nilsson, L. Hagberg, D. Fuchs, S. Spudich, K. Blennow, H. Zetterberg, Plasma concentration of the neurofilament light protein (nfl) is a biomarker of CNS injury in HIV infection: a cross-sectional study, EBioMedicine 3 (2016) 135–140, https://doi.org/10.1016/j.ebiom.2015.11.036.

[35] T.K. Karikari, T.A. Pascoal, N.J. Ashton, S. Janelidze, A.L. Benedet, J.L. Rodriguez, M. Chamoun, M. Savard, M.S. Kang, J. Therriault, M. Schöll, G. Massarweh, J. P. Soucy, K. Höglund, G. Brinkmalm, N. Mattsson, S. Palmqvist, S. Gauthier, E. Stomrud, K. Blennow, Blood phosphorylated tau 181 as a biomarker for Alzheimer's disease: a diagnostic performance and prediction modelling study using data from four prospective cohorts, Lancet Neurol. 19 (5) (2020) 422–433, https://doi.org/10.1016/S1474-4422(20)30071-5.

[36] B.T. Thomas Yeo, F.M. Krienen, J. Sepulcre, M.R. Sabuncu, D. Lashkari, M. Hollinshead, J.L. Roffman, J.W. Smoller, L. Zöllei, J.R. Polimeni, B. Fisch, H. Liu, R. L. Buckner, The organization of the human cerebral cortex estimated by intrinsic functional connectivity, J. Neurophysiol. 106 (3) (2011) 1125–1165, https://doi.org/10.1152/jn.00338.2011.

[37] P.M. Rossini, D. Burke, R. Chen, L.G. Cohen, Z. Daskalakis, R. Di Iorio, V. Di Lazzaro, F. Ferreri, P.B. Fitzgerald, M.S. George, M. Hallett, J.P. Lefaucheur, B. Langguth, H. Matsumoto, C. Miniussi, M.A. Nitsche, A. Pascual-leone, W. Paulus, S. Rossi, H.R. Siebner, Clinical Neurophysiology Non-invasive electrical and

magnetic stimulation of the brain , spinal cord , roots and peripheral nerves : basic principles and procedures for routine clinical and research application . An updated report from an, Clin. Neurophysiol. 126 (6) (2015) 1071–1107, https://doi.org/10.1016/j.clinph.2015.02.001.

[38] A. Delorme, S. Makeig, EEGLAB: an open source toolbox for analysis of single-trial EEG dynamics including independent component analysis, J. Neurosci. Methods 134 (1) (2004) 9–21, https://doi.org/10.1016/j.jneumeth.2003.10.009.

[39] N.C. Rogasch, C. Sullivan, R.H. Thomson, N.S. Rose, N.W. Bailey, P.B. Fitzgerald, F. Farzan, J.C. Hernandez-pavon, NeuroImage Analysing concurrent transcranial magnetic stimulation and electroencephalographic data : a review and introduction to the open-source TESA software, Neuroimage 147 (June 2016) (2017) 934–951, https://doi.org/10.1016/j.neuroimage.2016.10.031.