Transcranial ultrasonic stimulation of central thalamus reduces arousal in healthy volunteers

Amber R. Hopkins[1]* (co-first), Joshua A. Cain[1] (co-first), Elizabeth White[1], Stephanie Abakians[1], Martin M. Monti[1,2]

[1]Department of Psychology, University of California Los Angeles, Los Angeles, United States
[2]Brain Injury Research Center, Department of Neurosurgery, University of California Los Angeles, Los Angeles, CA, United States

*Corresponding author: Amber R. Hopkins (arhopkins@ucla.edu

Abstract

We present the first causal evidence in healthy humans linking central thalamus to vigilance and pulvinar to visuospatial attention using low-intensity transcranial focused ultrasound stimulation (tFUS). In a within-subjects, counterbalanced design, 27 healthy volunteers completed the Psychomotor Vigilance Task (PVT), a measure of vigilance, and the Egly Driver Task (EDT), which assesses visuospatial attention, before and after central thalamus, pulvinar, and sham sonication with tFUS. Central thalamic sonication significantly impaired performance on both tasks in a manner consistent with decreases in arousal. Pulvinar sonication resulted in more subtle effects consistent with a selective disruption of visuospatial attention. Specifically, participants were less responsive to visual stimuli presented in the visual field contralateral to the targeted pulvinar. These results demonstrate that tFUS can elicit measurable behavioral changes in healthy volunteers and underscores its potential as a high-resolution tool for noninvasive brain mapping, capable of differentiating functional contributions of adjacent thalamic nuclei only millimeters apart.

Keywords: *thalamus*, *attention, vigilance, arousal, consciousness, transcranial ultrasound stimulation, neuromodulation*

Introduction

The thalamus and its interactions with the cortex play a central role in perception, attention, arousal, and consciousness. However, the exact contributions of individual thalamic nuclei to healthy human cognition remain imperfectly understood because current knowledge is based on correlational neuroimaging work with healthy volunteers and lesion or invasive neurostimulation research with non-human animal models and human patients populations [1-5]. The emerging technique of low-intensity focused transcranial ultrasound stimulation (tFUS) addresses this gap, allowing for the safe [6-8] and non-invasive modulation of deep brain tissue in healthy volunteers with high spatial precision [9]. In this work, we leverage tFUS to dissociate the roles of two parts of the thalamus in healthy human cognition, specifically the central thalamus and the pulvinar.

A growing body of research implicates the central thalamus, which includes the intralaminar thalamic nuclei and adjacent tissue, in arousal and consciousness [1, 10]. The central thalamus not only bridges the brainstem and cortex as part of the ascending reticular activating system [11-13] but also shows widespread and reciprocal connections with the cortex that could put it in position to facilitate cortical communication and synchrony [1, 10, 14-17]. Invasive electrical stimulation of the central thalamus in awake non-human primates can increase arousal [18] or induce lapses in consciousness [19], depending on the stimulation parameters but has also been shown to increase behavioral and electrophysiological markers of arousal in anesthetized rodents [3, 4, 20] and non-human primates [1, 21-23]. In addition, application of invasive deep brain stimulation and tFUS to the central thalamus in human patients with disorders of consciousness (DOC) can bring some improvements in their behavioral responsiveness [5, 24-26].

In contrast, the pulvinar, which subsumes the posterior third of the thalamus, shows interactions with the cortex consistent with a role in visual processing and visuospatial attention. The pulvinar is reciprocally connected with visual cortices but also higher-order frontal and parietal areas [27-32]. Pulvinar activity shifts with visual attention. Visual responses modulated by attention can be recorded from the pulvinar [33-35] and neuroimaging studies report enhanced pulvinar activation with attention [36, 37]. Other work, however, suggests that the pulvinar plays a central, coordinating role in visual attention. The pulvinar has been shown to synchronize cortical activity following attention, regulating the transmission of information between cortical areas according to attentional demands [38]. In addition, neural activity propagates from the pulvinar to cortex during states of engaged visual attention [39]. Moreover, lesions and deactivation of the pulvinar have been associated with deficits of visuospatial attention in humans [40-43] and non-human primates [2, 44-46], especially for the contralateral visual field and when stimuli in multiple locations compete for attention [2, 40, 43].

In this work, we leverage the tFUS to modulate subcortical tissue to test the causal roles of the central thalamus and the pulvinar in healthy human cognition. Using a within-subjects, sham-controlled, and counterbalanced design, 27 healthy volunteers completed the Psychomotor Vigilance Task (PVT), a measure of vigilance [47], and the Egly Driver Task (EDT), which assesses visuospatial attention [48], before and after central thalamus, pulvinar, and sham sonication with tFUS. Targeting the central thalamus with tFUS impaired performance on both tasks in a manner consistent with a reduction in arousal level. Pulvinar sonication resulted in more subtle deficits in

visuospatial attention. Specifically, participants were less responsive to visual stimuli presented in the visual field contralateral to the targeted pulvinar. These results constitute the first causal evidence in the healthy brain for the role of the central thalamus and pulvinar and underscores the potential of tFUS as a high-resolution tool for causal brain mapping of deep brain regions in healthy volunteers.

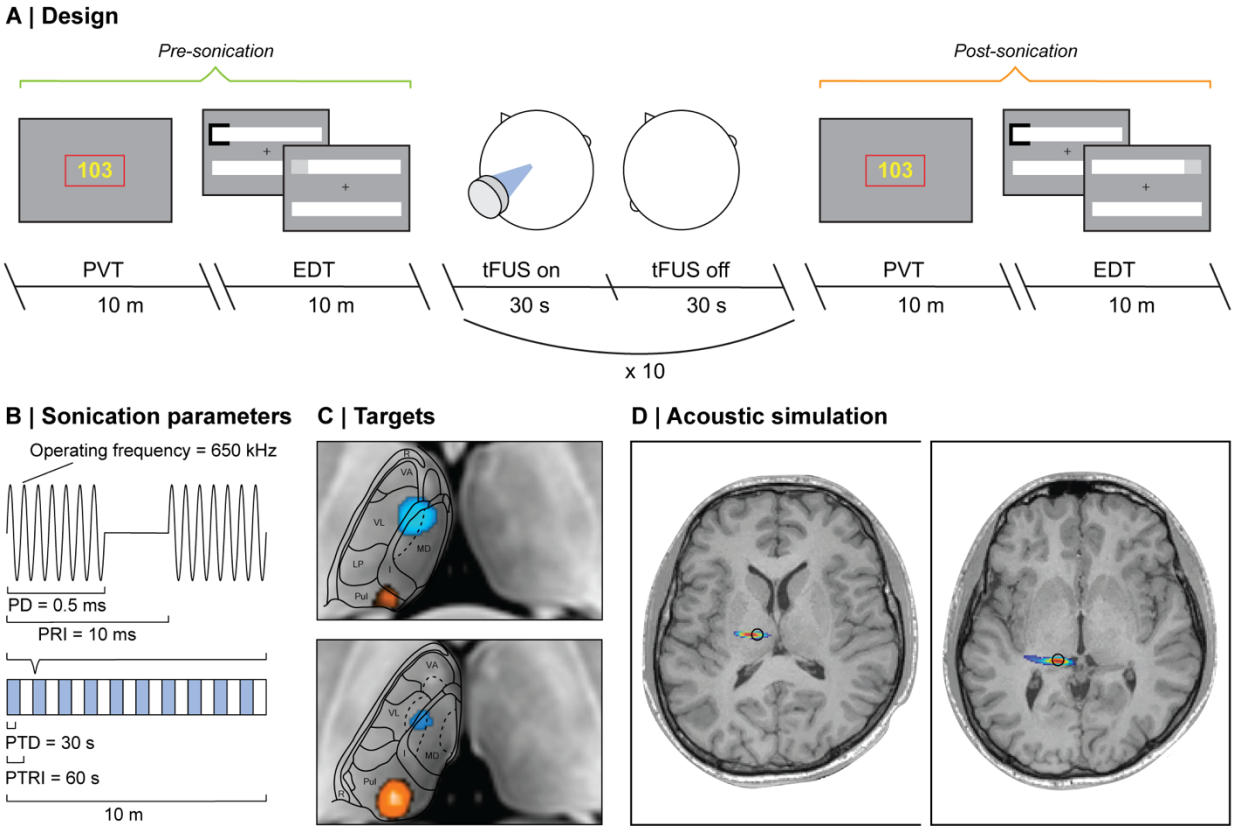

Figure 1: *Methods. (A): Session schematic. Participants completed the PVT and EDT before (pre-sonication) and after (post-sonication) sham, pulvinar, and central thalamic sonication (in separate sessions). Session and task order were counterbalanced across participants. During the PVT, participants maintained central fixation and pressed a button as quickly as possible when a cue (yellow millisecond counter) appeared. During the EDT, participants maintained central fixation and pressed a button when they detected a visual target (change in contrast at the end of a bar). On main trials, a spatial cue (black square) indicated where the target was most likely to appear (with 75% cue validity). On catch trials, no target followed the cue. (B): Sonication parameters. 10 blocks of 30 seconds of sonification (blue) and 30 seconds of rest (white), corresponding to a 30-second pulse train duration (PTD) and 60-second pulse train repetition interval (PTRI). We used a 0.5 ms pulse duration (PD) and 10 ms pulse repetition interval (PRI), resulting in a 5% (PD/PRI) duty cycle (DC) and a 100 Hz (1/PRI) pulse repetition frequency (PRF). The $I_{spta.3}$ was ≤ 720 mW/cm^2 and the $I_{sppa.3}$ was ≤ 14.40 W/cm^2. (C): tFUS targets. Heatmap showing tFUS aiming masks across participants in standard space for the central thalamus (blue) and pulvinar (orange). 5-mm spheres were drawn in standard space in each target region and then warped onto the T1 image of each participant to create a tFUS aiming mask. (D): Simulated acoustic effects when targeting left central thalamus (left) and pulvinar (right) for an example subject with a 80-mm fixed focal length transducer. The ultrasound focus is colored by the amount of acoustic pressure achieved, from 50% of the maximum pressure (blue) to the maximum pressure (red). The left central thalamus (left) and pulvinar (right) are outlined in black based on the participant's tFUS aiming mask and to scale.*

Results

Psychomotor Vigilance Task (PVT)

Mixed-effects models testing the three-way interaction between the effects of ultrasound condition (sham, pulvinar, and central thalamus sonication), block (pre-sonication and post-sonication), and time into the task (minutes) were used to examine whether targeting the central thalamus or the pulvinar with tFUS impacted behavioral markers of vigilance from the PVT [47], including response time, response speed, and lapses in attention. Descriptive statistics for the data used in the models are presented in Fig. 2A, D, and G. Additional results are described in the Supplemental Material.

Response time. On average, response times decreased by 3.22 ms (95% CI = [-5.13, -1.30], z = -3.29, p = 0.001, p_{adj} = 0.004) and 3.20 ms (95% CI = [-5.12, -1.28], z = -3.27, p = 0.001, p_{adj} = 0.004) after sham and pulvinar sonication, respectively, but increased by 5.34 ms (95% CI = [3.38, 7.30], z = 5.35, p < 0.0001, p_{adj} < 0.0001) after central thalamic sonication (see Fig. 2B). The difference in average response times between the post- and pre-sonication blocks was 8.56 ms (95% CI = [5.82, 11.30], z = 6.13, p < 0.0001, p_{adj} < 0.0001) and 8.55 ms (95% CI = [5.80, 11.29], z = 6.11, p < 0.0001, p_{adj} < 0.0001) greater for the central thalamus condition than the sham and pulvinar conditions, respectively (see Fig. 2B). Response times at task onset (0 minutes into the task) did not change after sham or pulvinar sonication but were 5.80 ms (95% CI = [1.93, 9.68], z = 2.94, p = 0.003, p_{adj} = 0.009) higher after central thalamus sonication (see Fig. 2C). The difference in response times at task onset between the post- and pre-sonication blocks was 7.09 ms (95% CI = [1.67, 12.51], z = 2.57, p = 0.010, p_{adj} = 0.023) and 4.69 ms (95% CI = [-0.73, 10.11], z = 1.70, p = 0.089, p_{adj} = 0.18) greater for the central thalamus condition than the sham and pulvinar conditions, respectively (see Fig. 2C).

Response speed. On average, response speeds increased by 0.04 ms (95% CI = [0.02, 0.06], z = 4.21, p < 0.0001, p_{adj} < 0.0001) and 0.03 ms (95% CI = [0.01, 0.05], z = 3.35, p = 0.0008, p_{adj} = 0.004) after sham and pulvinar sonication, respectively, but decreased by 0.03 ms (95% CI = [-0.05, 0.01], z = -3.04, p = 0.002, p_{adj} = 0.009) after central thalamic sonication (see Fig. 2E). The difference in average response speeds between the post- and pre-sonication blocks was 0.07 ms (95% CI = [-0.10, -0.04], z = -5.12, p < 0.0001, p_{adj} < 0.0001) and 0.06 ms (95% CI = [-0.09, -0.04], z = -4.51, p < 0.0001, p_{adj} = 0.0001) lower for the central thalamus condition than the sham and pulvinar conditions, respectively, on average (see Fig. 2E). Response speeds at task onset (0 minutes into the task) did not change with sham or pulvinar sonication but were 0.04 ms (SE = 0.02, 95% CI = [-0.08, 0.00], z = -2.02, p = 0.043, p_{adj} = 0.115) lower after central thalamic sonication (see Fig. 2F). The difference in response speeds at task onset between the post- and pre-sonication blocks was 0.06 ms (SE = 0.03, 95% CI = [-0.11, 0.00], z = -2.01, p = 0.045, p_{adj} = 0.115) lower in the central thalamic condition than the sham condition (see Fig. 2F).

Lapses. On average, participants were 0.66 (95% CI = [0.56, 0.78], z = -4.99, p < 0.0001, p_{adj} < 0.0001) and 0.70 (95% CI = [0.59, 0.83], z = -4.04, p = 0.0001, p_{adj} = 0.0005) times as likely to show a lapse in attention after sham and pulvinar sonication, respectively, but were no more or less likely after central thalamic sonication (odds ratio = 1.14, 95% CI = [0.97, 1.34], z = 1.56, p = 0.12, p_{adj} = 0.24; see Fig. 1H). The odds ratio for lapses between the blocks was 1.73 (95% CI = [1.37, 2.18], z = 4.65, p <

0.0001, p_{adj} < 0.0001) and 1.62 (95% CI = [1.28, 2.06], z = 4.01, p = 0.0001, p_{adj} = 0005) times higher for the central thalamus condition than the sham and pulvinar conditions, respectively (see Fig. 1H). Participants were 0.69 (95% CI = [0.49, 0.98], z = -2.10, p = 0.035, p_{adj} = 0.079) and 0.67 (95% CI = [0.47, 0.96], z = -2.19, p = 0.029, p_{adj} = 0.079) times as likely to show a lapse at task onset (0 minutes into the task) after sham and pulvinar sonication, respectively, but were no more or less likely after central thalamic sonication (odds ratio = 1.16, 95% CI = [0.84, 1.61], z = 0.89, p = 0.37, p_{adj} = 0.67; see Fig. 1I). The odds ratio for lapses between the blocks at task onset was 1.67 (95% CI = [1.04, 2.67], z = 2.67, p = 0.033, p_{adj} = 0.079) and 1.72 (95% CI = [1.06, 2.78], z = 2.21, p = 0.027, p_{adj} = 0.079) times higher for the central thalamus condition than the sham and pulvinar conditions, respectively (see Fig. 1I).

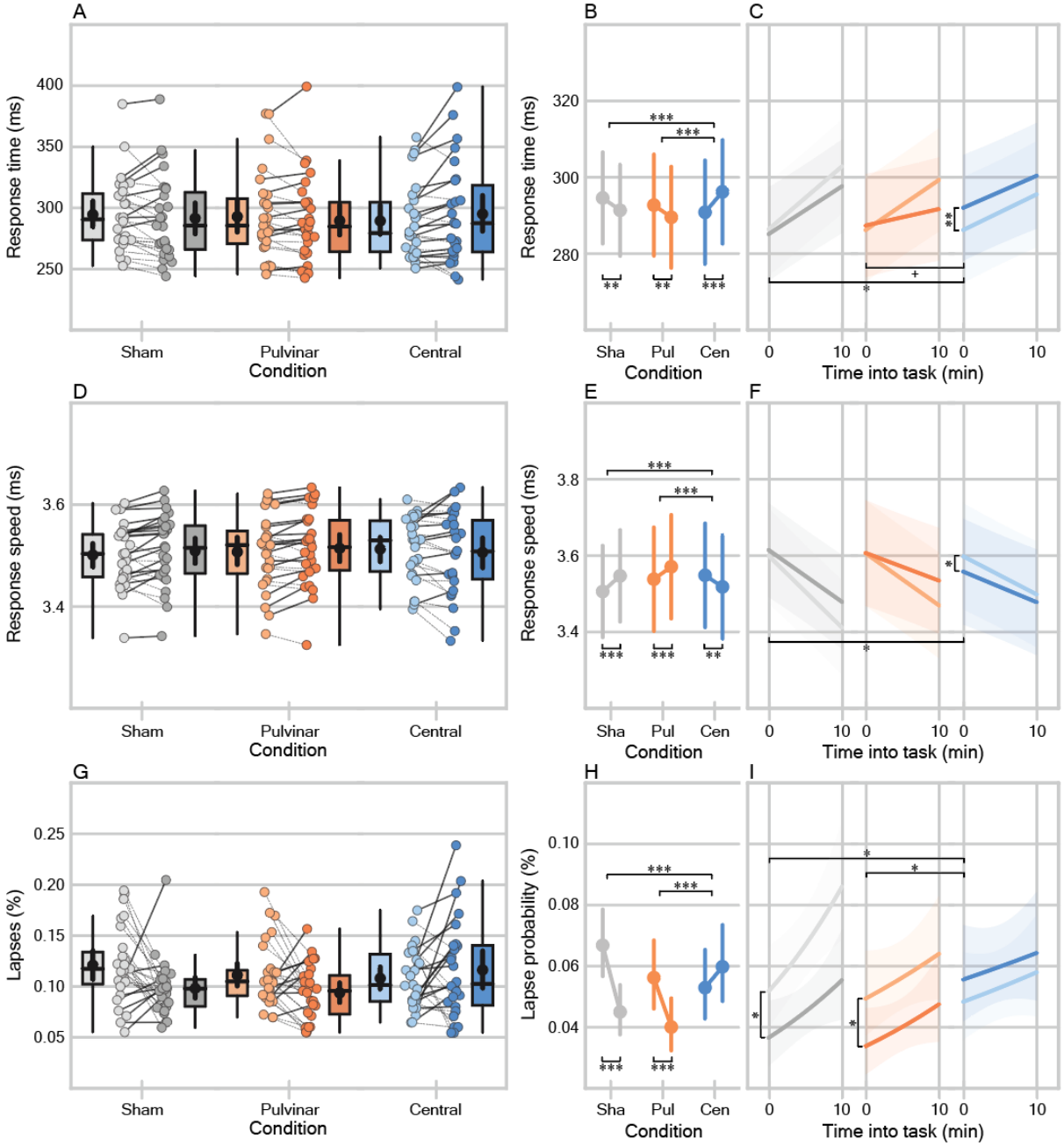

Figure 2: *Central thalamic sonication impaired vigilance, on average. (A, D & G): Raw data descriptive statistics. Box, whisker, and strip plots showing the distribution of response time (A), response speed (D), and lapses (G) during each block (pre- and post-sonication) for the sham (left), pulvinar (middle) and central thalamus (right) conditions. Boxes show first quartile, median, and third quartile and whiskers span the datapoints within 1.5 × the IQR from the lower and upper hinges. Black points and bars show inside each box show mean across participants and 95% confidence intervals. Individual points are participant means. Lines connecting participants across the blocks are dashed or solid if the participant showed a higher value of the outcome variable before or after sonication, respectively. (B-C, E-F, & H-I): Mixed-effects modeling. Brackets and asterisks indicate significant differences. (B, E, & H): Estimated marginal means for response time (C), response speed (D), and predicted probability of lapsing (E) for each block and ultrasound condition. Pre- and post-sonication appear left to right for each condition. Bars show 95% confidence intervals. (C, F, & I): Predicted response time (C), response speed (D), and probability of lapsing (E) for each block and ultrasound condition over time into the task (minutes). Bands show 95% confidence intervals. Pre- and post-sonication are lighter and darker, respectively. (Abbreviations: 'sha': sham, 'pul': pulvinar, 'cen': central)*

Egly Driver Task (EDT)

Mixed-effects models were used to examine how pulvinar and central thalamic sonication affected behavioral markers of visuospatial attention during EDT [48], specifically accuracy and response time. Descriptive statistics for the data used in the models and additional results are presented in the Supplemental Material.

Accuracy. Central thalamic sonication impaired visual target detection on main trials. Participants were 1.32 (95% CI = [1.20, 1.45], z = 5.59, p < 0.0001) and 1.16 (95% CI = [1.05, 1.27], z = 3.05, p = 0.002) times more likely to respond correctly on main trials ('hits') after sham and pulvinar sonication, respectively, but were no more or less likely after central thalamic sonication (odds ratio = 0.98, 95% CI = [0.89, 1.07], z = -0.49, p = 0.63), on average (see Fig. 3A). The odds ratio for being correct between the blocks on average was 0.74 (95% CI = [0.65, 0.85], z = -4.39, p < 0.0001) and 0.84 (95% CI = [0.74, 0.96], z = -2.53, p = 0.011) times lower for the central thalamus condition compared to the sham and pulvinar conditions, respectively. Participants responded correctly on main trials more often when the spatial cue was valid after sham and pulvinar sonication but not central thalamic sonication (see Fig. 3D). On ipsilateral valid-cue main trials, the odds ratio for accuracy between the blocks was 0.71 (95% CI = [0.57, 0.88], z = -3.05, p = 0.002) and 0.78 (95% CI = [0.63, 0.97], z = -2.21, p = 0.027) times lower for the central thalamus condition than the sham and pulvinar conditions, respectively. On contralateral valid-cue main trials, the odds ratio for accuracy between the blocks was 0.70 (95% CI = [0.57, 0.86], z = -3.43, p = 0.0006) and 0.82 (95% CI = [0.67, 1.00], z = -1.95, p = 0.051) times lower for the central thalamus condition than the sham and pulvinar conditions, respectively.

Pulvinar sonication impaired visual target detection but only on main trials where the spatial cue directed attention to the ipsilateral visual field and the visual target appeared in the contralateral visual field (see Fig. 3D). The odds ratio for being correct between the blocks was 1.33 (95% CI = [0.89, 1.99], z = 1.37, p = 0.17) and 1.32 (95% CI = [0.88, 1.98], z = 1.32, p = 0.18) times higher for valid-cue ipsilateral and valid-contralateral main trials compared to invalid-cue contralateral main tris for the pulvinar sonication condition, respectively. The odds ratio for being correct between the blocks was 1.89 (95% CI = [0.93, 3.86], z = 1.75, p = 0.08) times higher for valid-cue ipsilateral main trials than for invalid-cue contralateral main trials for the pulvinar sonication condition compared to the sham condition.

Pulvinar sonication reduced reactivity to spatial cues on catch trials compared to sham and central thalamus sonication, on average (see Fig. 3B). Participants were 0.65 (95% CI = [0.40, 1.08], z = -1.66, p = 0.097) and 0.62 (95% CI = [0.37, 1.03], z = -1.84, p = 0.066) times as likely to respond correctly ('correct rejections') after sham and pulvinar sonication, respectively, but were no more or less likely in the pulvinar condition (odds ratio = 0.83, 95% CI = [0.49, 1.41], z = -0.68, p = 0.50), on average. Participants responded correctly on catch trials ('correct rejections') with contralateral spatial cues more often after pulvinar sonication but not after sham or central thalamus sonication (see Fig. 3D). The odds ratio for correct responses catch trials with contralateral spatial cues between the blocks was 2.60 (95% CI = [0.82, 8.26], z = 1.63, p = 0.10) times higher for the pulvinar condition than for the sham condition and 0.27 (95% CI = [0.08, 0.88], z = -2.18, p = 0.03) times lower for the central thalamus condition than for the pulvinar condition. The odds ratio for correct responses between the blocks was 2.74 (95% CI = [0.89, 8.50], z = 1.75, p = 0.080) times higher for catch trials with contralateral spatial cues than catch trials with ipsilateral spatial cues in the pulvinar condition. The odds ratio for correct responses between the blocks was 2.83 (95% CI = [0.57, 13.90], z = 1.28, p = 0.20) times higher for right-cue catch trials than left-cue catch trials for the pulvinar condition compared sham and 0.15 (95% CI = [0.03, 0.75], z = -2.13, p = 0.021) times lower on right-cue catch trials than left-cue catch trials for the central thalamus condition compared to the pulvinar condition.

Response time. Participants took longer to respond to the visual target on main trials when the spatial cue was ipsilateral, but the visual target was contralateral after pulvinar sonication but not after sham or central thalamus sonication (see Fig. 3E). Specifically, response times decreased by 8.25 ms (95% CI = [-18.04, 1.54], z = -1.65, p = 0.098) and 9.32 ms (95% CI = [-19.13, 0.5048 z = -1.86, p = 0.062) after sham and central thalamus sonication but did not change after pulvinar sonication (estimate = 4.54 ms, 95% CI = [-5.76, 14.83], z = 0.86, p = 0.39) on main trials with ipsilateral spatial cues but contralateral visual targets. The difference in response times between post- and pre-sonication on these trials was 12.79 ms (95% CI = [-1.42, 27.00], z = 1.76, p = 0.078) higher for the pulvinar condition compared to the sham condition and 13.86 ms (95% CI = [-28.08, 0.36], z = -1.91, p = 0.056) lower for the central thalamus condition compared to the pulvinar condition. The change in response time across the blocks was 13.79 ms (95% CI = [-2.83, 23.36], z = 2.23, p = 0.014) higher on invalid trials with the contralateral visual targets compared to valid contralateral trials in the pulvinar condition. Additionally, the change in response time across the blocks was 11.27 ms (95% CI = [-2.53, 25.07], z = 1.60, p = 0.10) higher on invalid contralateral target trials than invalid ipsilateral trials in the pulvinar condition.

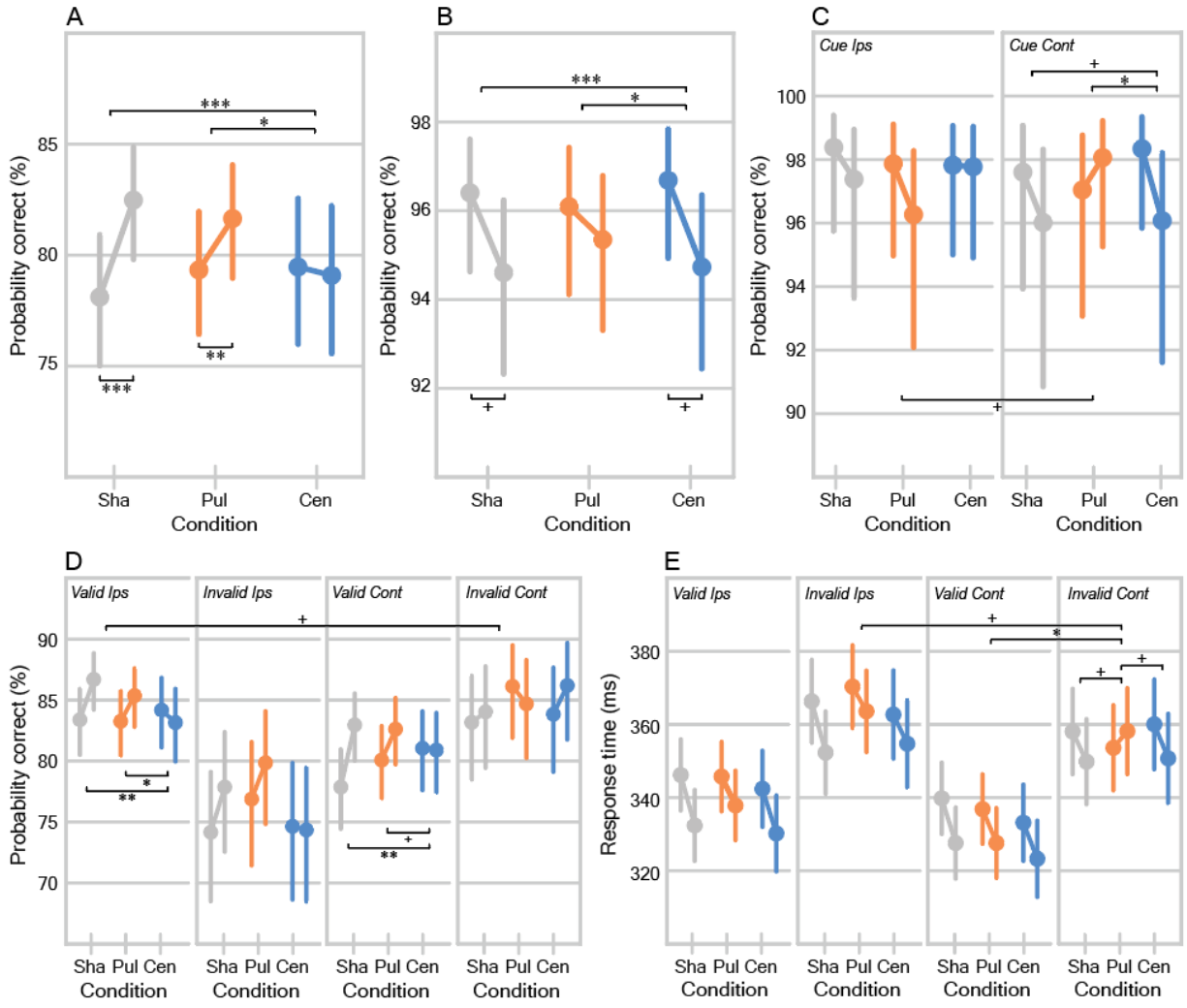

Figure 3: *Mixed-effects modeling results from the Edgly Driver Task (EDT). Brackets and asterisks indicate significant differences. Bars show 95% confidence intervals. (A & B): Estimated marginal means for the predicted probability of being correct on main trials (A) and catch trials (B) for each condition at pre-sonication and post-sonication (left to right), on average. (C): Estimated marginal means for the predicted probability of being correct on catch trials with ipsilateral spatial cues (left) and contralateral spatial cues (right) for each condition at pre-sonication and post-sonication (left to right). (D & E): Estimated marginal means in the mixed-effects models for the predicted probability of being correct (B) as well as response time (D) for each ultrasound at pre-sonication and post-sonication (left to right) on valid-cue ipsilateral (left), invalid-cue ipsilateral (middle left), valid-cue contralateral (middle right), and invalid-cue contralateral (right) main trials. (Abbreviations: 'sha': sham, 'pul': pulvinar, 'cen': central, 'ips': ipsilateral, 'con': contralateral)*

Discussion

Targeting the central thalamus with tFUS slowed responses and increased lapses during the PVT but also reduced visual target detection rates and slowed responses during the EDT overall, which is consistent with a reduction in arousal. Pulvinar sonication, however, resulted in less pronounced but more specific deficits in visuospatial attention. Specifically, participants were less reactive to spatial cues when they appeared in the visual field contralateral to the targeted pulvinar but also failed to

detect, and took longer to respond to, contralateral visual targets when the spatial cue directed their attention to the ipsilateral visual field, competing for attentional resources. These results not only constitute the first causal evidence in the healthy human brain that the central thalamus and the pulvinar are important for arousal and visuospatial attention, respectively, but also demonstrates that tFUS can modulate behavior, over a period of at least 30 minutes, and is spatially precise enough to have different effects when applied to deep brain tissues only millimeters apart.

Reduced vigilance under diminished states of arousal like sleep deprivation [49] is reliably accompanied by slower responses [50, 51] and more frequent lapses [50, 52] during the PVT. We achieved similar effects by targeting the central thalamus with tFUS. Specifically, participants responded more slowly and showed lapses more often during the PVT after central thalamic sonication but not after sham or pulvinar sonication (see Fig. 2). These effects seem consistent with a suppression of the central thalamus. For instance, reduced thalamic activation during lapses in the PVT and elevated thalamic activation during non-lapse periods for sleep deprived volunteers has been reported [53]. Indeed, a recent study from our group targeted anterior parts of the left thalamus in awake healthy volunteers with tFUS at the same parameters we used here and observed decreases in blood-oxygen level dependent (BOLD) signals during sonication and blood perfusion after sonication in the targeted thalamic regions but also in connected cortical areas [54].

Thalamic activity during vigilance tasks under sleep deprivation reflects a complex interplay between the effects of sleep loss, which dampens arousal, and engagement in the task, which increases arousal [55]. At rest, thalamic activity is suppressed under sleep deprivation when compared to rested wakefulness [56-59]. During sustained vigilance tasks like the PVT, however, the thalamus shows greater activation under sleep deprivation than rested wakefulness, which suggests that the thalamus becomes more active to compensate for the dampening effects of sleep deprivation on task performance [55, 57, 60]. These effects might be more central parts of the thalamus in particular [55, 56]. If targeting central thalamus with tFUS in our experiment emulates the effects of sleep deprivation, we might expect thalamic activity during the PVT to be greater after central thalamic sonication compared to before. Future work could administer our experimental design in the MRI environment to examine whether targeting the central thalamus with tFUS disrupts or amplifies the interplay of thalamic activity between decreased arousal and task engagement reported for sleep deprivation.

Where the effects we observe from targeting the central thalamus with tFUS differ from reported with sleep deprivation involves the time-on-task effect (also called the vigilance decrement), whereby performance on a vigilance task worsens over the duration of the task from fatigue or boredom [61-64]. For the PVT, the time-on-task effect refers to a slowing of responses, an increase in lapses, and an increase in response time variability over the duration of the task, which is present during rested wakefulness and exacerbated by states like sleep deprivation [50, 63-66]. Our participants exhibit the time-on-task effect during the PVT, as their responses slowed, and they showed lapses more often over time into the task (see Fig. 2). However, while central thalamic sonication was associated with slower response and more lapse, on average, it did not change this time-on-task effect. This could indicate that targeting the

central thalamus with tFUS reduced arousal more generally but did not disrupt the capacity to maintain vigilance over the duration of the task. Interestingly, there is evidence that maintaining vigilance over uninterrupted periods recruits a predominantly right-lateralized network of cortical and subcortical structures, including right fronto-parietal cortex and right thalamus [67, 68]. Activity in these regions increases over longer durations of uninterrupted attentional demands [67] and patients with damage to relevant structures in their right hemisphere show stronger deficits in sustained attention than patients with left-hemisphere damage [69-73]. This might explain why we do not observe an alteration of the time-on-task effect with central thalamic sonication. If maintaining vigilance over uninterrupted periods recruits thalamic and cortical areas in the right hemisphere mostly, then targeting the left central thalamus with tFUS in our experiment might not change the time-on-task effect during the PVT.

Lesions and deactivation of the pulvinar have been associated with deficits of visuospatial attention in humans [40-43] and non-human primates [2, 44-46], especially for the contralateral visual field. Behavioral impairments in patients with pulvinar damage are particularly strong when distractor stimuli compete for attentional resources [40, 43]. In addition, [2] used muscimol to inactivate the pulvinar in behaving non-human primates and observed diminished exploration of the contralesional visual field, especially when stimuli appeared in both visual fields. We observed similar alterations of visuospatial attention by targeting the pulvinar with tFUS. Participants were less reactive to contralateral spatial cues during the EDT after pulvinar sonication, which is reflected by the increase in accuracy on catch trials that had contralateral spatial cues arising from fewer responses (‘false alarms’). In addition, participants responded correctly less often, and took longer to respond to, contralateral visual targets but only when an ipsilateral spatial cue had appeared beforehand, competing for attention. While our results are consistent with the literature, they are statistically weak due to the very low number of catch trials and invalid-cue main trials included in the abbreviated EDT protocol that we administered here that severely degraded statistical power for these comparisons. Future work administering the full EDT protocol, before and after pulvinar sonication, will be necessary to assess this issue with the appropriate statistical power.

By selecting cue-to-target intervals from a continuous range of values between 300 and 1100 ms during the EDT, fluctuations in spatial and object attention as a function of cue-to-target interval have been mapped, demonstrating that visuospatial attention is discontinuous and samples the environment in theta-rhythmic cycles (3–8 Hz) [39, 74-76]. These cycles may organize neural activity into alternating attentional states of engagement (heightened sensitivity to stimuli that appear in the attended location) and disengagement (decreased sensitivity to stimuli that appear in the attended location and increased probability of shifting attention to another location) [75]. While theta-rhythmic cycles in attention have been linked to theta oscillations in frontal and parietal cortices [75, 77], the pulvinar seems to play a coordinating role. [39] assessed pulvino-cortical interactions during theta-rhythmic sampling by recording frontal eye fields, lateral intraparietal area, and the pulvinar in awake non-human primates completing the EDT. They report that neural activity propagates from the pulvinar to the cortex during engagement but from the cortex to the pulvinar during disengagement. Suppressing the pulvinar with tFUS may thus disrupt the theta-rhythmic cycles of attention. However, because we used an abbreviated version of the EDT with

only short (300 ms) and long (1100 ms) cue-to-target intervals to keep task duration similar between the EDT and PVT, we cannot examine the effects of pulvinar sonication on the theta-rhythmic cycles of attention. Future work should administer the entire EDT, selecting cue-to-target intervals from the continuous range of values between 300 and 1100 ms to test whether targeting the pulvinar with tFUS disrupts the theta-rhythmic alternation of attentional states.

This work draws several important methodological insights for tFUS. We were able to induce distinct behavioral changes by targeting different parts of the thalamus with tFUS. Targeting the central thalamus led to changes in behavior consistent with a decrease in arousal level but targeting the pulvinar was associated with more specific deficits in visuospatial attention. These results have several important methodological implications. First, tFUS at the parameters that we apply here is sufficient to alter behavior for at least 20 minutes without dissipating. Each task was 10 minutes long and task order was counterbalanced across participants, but we did not observe any differences in the effects of sonication on task performance that could be explained by the order in which participants completed the task in (see Supplemental Material). In addition, these results demonstrate that tFUS can map the healthy human brain with unprecedented spatial precision. Targeting parts of the thalamus that are only millimeters apart had distinct effects on behavior. The ability of tFUS to selectively modulate subcortical structures with such high spatial precision and non-invasively makes way for causal inferences in the study of subcortical networks as well as the treatment of neurological conditions.

Methods

All data analysis scripts, pre-processing pipelines, and anonymized raw data sets have been uploaded to the Open Science Framework (OSF) repository associated with this article.

Experimental model and study participant details

Participants

27 right-handed and English-speaking adults (18 years of age or older) with normal or corrected-to-normal vision were recruited following procedures approved by the Institutional Review Board at the University of California, Los Angeles (UCLA). individuals were excluded from the experiment if they self-disclosed current or recent use of any psychoactive medications or recreational drugs (e.g., antidepressants or stimulants), a counter-indication for entering the magnetic resonance imaging (MRI) environment (e.g., MRI-incompatible implants), possible or planned pregnancy (in the short-term), or had hair longer than a ¼ of an inch over their left temporal region and were unwilling to shave it. Participants provided written informed consent and were compensated at a rate of $40 per hour for their participation. If participants voluntarily withdrew before completing the full protocol (e.g., loss to follow-up), then a new participant was recruited to replace them.

Method details

Design

A 3×2 within-within design was used to test whether targeting the pulvinar or the central thalamus with tFUS altered behavioral markers of vigilance during the Psychomotor Vigilance Task (PVT) [47] and visuospatial attention during an Egly-Driver Task (EDT) [48]. Participants underwent an MRI scan followed by three sonication sessions, including one per ultrasound condition: sham, pulvinar, and central thalamus sonication. Session order was counterbalanced across participants. In each sonication session, the PVT and EDT were administered before and after sonication. The order in which the tasks were administered was counterbalanced across the participants. Sonication sessions were held on different days at least 24 hours apart to allow for potential lasting effects of sonication to fade between sessions. However, sessions were held around the same time of day for each participant.

Magnetic resonance imaging (MRI)

MR data were acquired with a 3T Siemens Prisma fit MRI scanner at the UCLA Staglin IMHRO Center for Cognitive Neuroscience. For every participant, we acquired an anatomical image (T1-weighted) image for use with a frameless stereotactic neuronavigational system using a T1-weighted MPRAGE sequence with the following parameters: TR = 2,400 ms, TI = 1,060 ms, TE = 2.12 ms, flip angle = 88, voxel size = 1×1×1 mm isotropic, matrix size = 256×256, 192 slices. The raw T1-weighted data was converted from DICOM to NIFTI format using *dcm2niix* [78].

tFUS aiming mask

We targeted the central thalamus because of its association with arousal and consciousness [1] and the pulvinar because of its coordinating role in visuospatial attention [28]. We targeted structures on the left side of the head for all participants.

Standard space masks for each target region were generated in MNI space by creating 5 mm spheres over the pre-determined target voxels (see Fig. 1C). A tFUS aiming mask was created for each participant by warping the target region masks from standard (MNI) space onto their T1 image using *applywarp* from the FMRIB Software Library (FSL; https://fsl.fmrib.ox.ac.uk; [79]), which would be overlayed the individual T1 in a neuronavigational system to aim the tFUS transducer. Linear and non-linear registrations between native structural (T1) and standard structural (MNI) images were performed using *flirt* and *fnirt* in FSL [80]. All steps were visually inspected and adjusted as needed to ensure anatomical plausibility.

Transcranial focused ultrasound stimulation (tFUS)

Low-intensity tFUS was administered using a BXPulsar 1002 drive system and an accompanying fixed-focal-length, single-element, and air-backed spherical transducer with a fixed focal length of 80 mm, aperture of 61 mm, and operating frequency of 650 kHz (Brainsonix, Corp., USA) [81]. A Brainsight neuronavigation system (Rogue Research, Inc., Canada) was retrofitted for concurrent use with tFUS. Optical trackers placed on the participant's head and a custom transducer holder were co-registered with the individual T1 image in the Brainsight system, enabling us to visualize transducer position relative to the participant's brain in real time. Sham sonication was achieved by placing a pigmented gel pad that absorbs sound (Brainsonix Corp., USA) between the surface of the transducer and the scalp, preventing ultrasound transmission to the head [82]. Acoustic coupling gel pads (Brainsonix Corp., USA) that promote ultrasound transmission [81] were used during real sonication, ensuring that set up was identical between the sham and real sonication sessions. During the sham sessions, the transducer was aimed at either the pulvinar or the central thalamus in a counterbalanced order across the participants.

Sonication regime

Ultrasound was delivered in an on/off block design consisting of 10 blocks of 30 seconds of sonication and 30 seconds of rest. We used a 0.5 ms pulse duration (PD), 60 s pulse repetition interval (PRI), 5% duty cycle (DC), 100 Hz pulse repetition frequency (PRF), < 720 mW/cm^2 $I_{spta.3}$, and < 14.40 W/cm^2 $I_{sppa.3}$ based on previous work [54]. These intensities comply with the United States Food and Drug Administration (FDA) guidelines for diagnostic ultrasound [83] and have no known adverse thermal bioeffects [84, 85].

Procedure

The individual T1 image and tFUS aiming mask were opened in the Brainsight neuronavigational system and a marker was generated in the center of the target region assigned to the session. The participant's head was secured in a comfortable position using chin and head rests and they were instructed to remain as still as possible to minimize movement during tFUS aiming and throughout sonication. The transducer was first placed over the left temple (approximately 1/3 of the distance from the corner of the left eye to the left tragus and superior by 2 cm), which is the thinnest part of the temporal bone and ideal cranial entry-point for minimizing ultrasound disruption by the skull. Deviations in transducer position were made as needed until the projected trajectory of the ultrasound beam passed through the target region. The transducer was kept as flat against the side of the head as possible to deliver ultrasound perpendicular to the skull surface, minimizing the scattering, reflection, or refraction of the ultrasound

[86]. Once an adequate transducer position was achieved, aqueous ultrasound gel (Aquasonic Clear Ultrasound Transmission Gel; Parker Laboratories, Inc., USA) was applied to the region subsuming the diameter of the transducer such that no hair permeated the gel layer and visible air bubbles were pressed out [87, 88]. A thin layer of gel was also applied between the surface of the transducer and the gel pad with air bubbles smoothed out. If reaching the target region required tilting the transducer, then an angled gel pad that filled the expanding gap between the transducer and the scalp was used. After placing the transducer, additional gel was applied to fill any remaining concavities between the transducer and the scalp. Transducer position was verified continuously throughout sonication and adjustments were made as needed to ensure that the projected trajectory of the ultrasound passed through the target region.

Apparatus and stimuli

Stimuli and tasks were created and administered using PsychoPy (Version 2021.2.3; [89]). Participants were seated in upright position 150 cm from the monitor with a button box placed in their right hand for the duration of each task. Earplugs were provided to reduce auditory distractions during the tasks.

Psychomotor vigilance task (PVT)

The PVT is a response time task that assesses the capacity to maintain vigilance (also called sustained attention) over uninterrupted periods of time [47]. Participants maintained central fixation and pressed a button as quickly as possible when they saw a visual cue for 10 minutes without any breaks. The visual cue was a yellow millisecond counter that appeared at the center of the monitor inside of a red rectangle at random 2-10 second inter-trial intervals. Response time was recorded on every trial. The counter stopped when the participant pressed the button but remained on the screen for one second before the trial ended, displaying the response time recorded on that trial. If the participant pressed the button before the visual cue appeared or too quickly after it appeared (response time $\leq$ 100 ms), then a false start message appeared on the monitor. If the participant did not press the button within 30 seconds of the visual cue being presented, then the trial timed out and a beep was played to alert the participant.

Many outcomes can be derived from the response time during the PVT, such as mean or median response time, lapses in attention (unusually slow responses traditionally defined as response times ≥ 500 ms or twice the mean response time), and response speed (1/response time) [50, 90]. A time-on-task effect (also called a vigilance decrement) can be observed with the PVT, whereby responses slow, lapses become more frequent, and response times become more variable over the duration of the task. Reduced vigilance arising from, for example, sleep deprivation, is associated with slower responses and more frequent lapses during the PVT overall but also exacerbates the time-on-task effects [51, 91-96].

Egly-Driver Task (EDT)

The EDT is a visuospatial attention task [48]. Participants maintained central fixation and pressed a button as quickly as possible when they detected a visual target stimulus over 10 minutes with a 30-second break every 2 minutes in two kinds of trials, including main trials and catch trials. Trial onset was marked by the presentation of two horizontal white bars, including one above the fixation cross and another below it. After a variable delay of 400-800 ms, a spatial cue (black square) appeared at the end of one of the horizontal bars to direct attention to that location. On main trials (90% of the trials), a

visual target appeared after either a short (300-350 ms) or long (1000-1050 ms) interval from the spatial cue. The visual target was a change in contrast at the end of one of the horizontal bars in either the cued location or another one. The spatial cue indicated the location the target was most likely to appear in (with 75% cue validity). The trial ended after a response window of 150-700 ms from the target. On catch trials (10% of trials), no target appeared, and the trial ended 500-1800 ms after the cue. Participants were either correct or not on each trial. The correct response on a main trial was to press the button ('hit') and the incorrect response was not to ('miss'). The correct response on a catch trial was to not press the button ('correct rejection') and the incorrect response was to press the button ('false alarm'). Response time was recorded whenever there was a button press. On a main trial, if the participant pressed the button before the visual target appeared or too soon after it appeared (response time $\leq$ 100 ms), the response was considered a false start and treated as incorrect.

The contrast of the visual target relative to the horizontal white bars was adjusted for each participant using a staircase procedure at the start of their first sonication session. The staircase trials were identical to the EDT trials described above, except that that the contrast of the visual target changed on every trial. The staircase followed a two-down, one-up rule where successful trials were followed by more difficult ones, decreasing the contrast of the visual target relative to the white bars until the participant failed to detect the visual target, and then adjusting the contrast accordingly. The staircase ended when we identified the minimum contrast with which the participant could correctly detect the visual target in 50% of the trials that it was presented. This final contrast value was saved and used for all EDT administrations for that participant.

Spatial attention leads to enhanced sensory processing and behavioral responses for stimuli presented in the attended region [97]. In the EDT, participants detect the visual target more often (and respond to it more quickly) when the spatial cue correctly indicated the location the visual target appeared in compared to when the spatial cue was invalid, demonstrating the behavioral benefit of visual attention as well as the behavioral cost of needing to shift attention to other spatial locations [48]. By varying cue validity as well as the visual field that the spatial cue and visual target appear in, the EDT can also be used to examine hemispheric differences in the behavioral benefits and costs associated with visual attention. Previous applications of the EDT used cue-to-target intervals that were randomly selected from a continuous range of values between 300 and 1100 ms [39, 74]. However, we administered an abbreviated version with only short (300-350 ms) and long (1000-1050 ms) cue-to-target intervals to ensure that task duration would be consistent between the PVT and the EDT but we would still be able to determine whether targeting pulvinar or central thalamus with tFUS affected accuracy and response time during the EDT, and if this depended on cue validity and the visual field that the stimuli appeared in.

Quantification and statistical analyses

Mixed-effects modeling was used to examine whether sonication in any of the ultrasound conditions affected behavioral markers of vigilance from the PVT and visuospatial attention from the EDT. We used mixed-effects models because of the hierarchical and repeated-measures structure of our data but also because they allow for the specification of random effects. Mixed-effect modeling was performed with the

lme4 (Version 1.1; [98]) and lmerTest [99] packages in R (Version 3.1; R Core Team [100]). Data cleaning and visualization was completed in Python (Version 3.8).

Random effects for participant and task order were included in every mixed-effects model. Since participants underwent the ultrasound conditions in separate sessions on different days, we specified a random intercept for participant with a random slope for condition (1 + condition | participant). The random slope for condition captures how the effect of condition varies for each participant when the other fixed effects are held at their reference levels (including block at pre-sonication), accounting for any individual differences in task performance before sonication between the sessions. Including the random intercept for participant also accounts for having multiple observations per participant, since we use unaggregated data from a repeated-measures design in the models. A random intercept for task order (1 | task order) was included to account for differences in task performance arising from the order in which participants completed the tasks. Task order was counterbalanced across participants, separating the participants into two groups (PVT then EDT and EDT then PVT). The random intercept for task order allows each group to have their own baseline values of the outcome variables in the models. Specifying these random effects better isolates the effects of targeting the pulvinar and the central thalamus with tFUS.

Mixed-effects logistic regressions were used for binary outcome variables, specifically lapses during the PVT and accuracy during the EDT. Fixed effects and estimated marginal mean contrast outputs for the mixed-effects logistic regressions are reported in odds ratios, which indicate how much more or less likely an event is to occur in one group compared to another. An odds ratio of 1 reflects equal likelihood of the event occurring in each group while an odds ratio less than or greater than 1 reflects less or more likelihood of the event occurring in one group than the other, respectively. Linear mixed-effects models were used for continuous outcomes, including response time and response speed during the PVT and response time during the EDT. The fixed effects and estimated martin mean contrast outputs for the linear mixed-effects models are reported in the units of the dependent (outcome) variable of the model.

Psychomotor vigilance task (PVT)

Mixed-effects models were used to examine whether sonication in any of the conditions affected behavioral markers of visuospatial attention from the PVT, specifically response time, response speed, and lapses. Response speed is the reciprocal of response time, which is derived by dividing the response time on every trial by 1000 and then reciprocally transforming it [50]. Lapses refer to lapses in attention and are unusually delayed responses to the cue. Response times greater than 500 ms or twice the mean response time for an individual participant are usually considered lapses [4]. However, these criteria did not work for our data. Our participants never or rarely showed response times greater than 500 ms or twice their mean response time, even before data cleaning (see Supplemental Material). Instead, we combined the data from all PVT blocks for each participant and considered trials with response times greater than the median response time plus two standard deviations for that participant to be lapses. Follow-up estimated marginal means contrasts were used to make specific comparisons in the models and adjusted for multiple comparisons using the amini-Hochberg method [101]. Key results are presented in the main text. Additional results are described in the Supplemental Material.

Data cleaning. Individual trials were discarded if the response time on that trial was a false start (≤ 100 ms) or a definite outlier compared to the other trials for that task block and participant based on Tukey's method (less than the first quartile minus 3 × the interquartile range (IQR) or greater than the third quartile plus 3 × the IQR) [102, 103]. Whole sessions from an individual participant were excluded from the mixed-effects models if the participant was a definite outlier in their change in response time from pre- to post-sonication relative to the other participants for that session, also based on Tukey's. All other sessions were included in the mixed-effects models to retain as much data as possible and improve estimates of the random effects in the mixed-effects models. Because all PVT outcomes are derived from the response time, the data cleaned based on response time was used in all mixed-effects models.

Analysis. Mixed-effects models were used to examine whether sonication in any of the conditions affected behavioral markers of vigilance from the PVT, specifically response time, response speed, and lapses in attention. Three separate models were run, including one per outcome. Linear mixed-effects models were used for response time and response speed, which were both continuous variables in the unaggregated data. A mixed-effects logistic regression was used for lapses because an individual trial was either a lapse or not, making lapse a binary variable in the unaggregated data. All models shared the following specification: outcome ~ condition × block × time into the task block + (1 + condition | participant) + (1 | task order) where the outcome was response time, response speed, or lapses. This model specification includes categorical fixed effects for ultrasound condition (sham, pulvinar, and central thalamus sonication) and block (pre-sonication and post-sonication) but a continuous fixed effect for time into the task (in minutes). Testing the three-way interaction between the effects of condition, block, and time into the task allows us to examine how each outcome changed after sonication on average but also over the duration of the task. Descriptive statistics for the cleaned, unaggregated PVT data used in the model are presented in Fig. 2A, D, and G.

Follow-up estimated marginal means contrasts were used to make more specific comparisons in the models and were adjusted for multiple comparisons using the Benjamini-Hochberg method [101]. The same set of contrasts were run for each outcome (response time, response speed, and lapses). To test whether sonication in any of the conditions affected an outcome on average, we contrasted the estimated marginal means for the outcome from post-sonication to pre-sonication for each condition, averaging over time into the task. To test whether the effect of sonication on the outcome differed between conditions on average, we contrasted the estimated marginal means for the difference in the outcomes between post- and pre-sonication between the conditions, averaging over time into the task. To test whether sonication in any condition affected how an outcome changed over the duration of the task, we contrasted estimated marginal means for the average change in the outcome per minute in the block from post-sonication to pre-sonication for each condition. To test whether the effect of sonication on how an outcome changed over the duration of the task differed between the conditions, we contrasted estimated marginal means for the difference in the average change in the outcome per minute in the block between post-sonication and pre-sonication between the conditions. To test whether sonication in any condition affected an outcome immediately after sonication (at task block onset), we contrasted the estimated marginal means for the outcome at 0 minutes into the task

from post-sonication to pre-sonication for each condition. To test whether the effect of sonication on an outcome at task onset differed between the conditions, we contrasted the estimated marginal means for difference in the outcome at 0 minutes into the task between post- and pre-sonication between the conditions.

Egly driver task (EDT)

Mixed-effects models were used to examine whether sonication in any of the conditions affected behavioral markers of visuospatial attention from the EDT, specifically accuracy and response time. Follow-up estimated marginal means contrasts were used to make more specific comparisons in the models and adjusted for multiple comparisons using the Benjamini-Hochberg method [101]. Key results are presented in the main text. Additional results are described in the Supplemental Material.

Data cleaning. Individual trials with a recorded response time ≤ 100 ms were considered false starts and treated as no button presses. Since accuracy and response time were independent (accuracy was recorded every trial but response time was only recorded when there was a button press), we cleaned the data separately for each outcome variable. Whole sessions from a participant were left out of the mixed-effects model for an outcome if that participant was a definite outlier in their change in that outcome from pre- to post-sonication relative to other participants for that session based on Tukey's method (less than the first quartile minus 3 × the IQR or greater than the third quartile plus 3 × the IQR) [102, 103]. All other blocks and sessions were included in the mixed-effects models to retain as much data as possible but also to improve estimates of the random effects.

Accuracy. Participants could be either correct ('hits' on main trials and 'correct rejections' on catch trials) or incorrect ('misses' on main trials and 'false alarms' on catch trials) on an individual trial, making accuracy a binary variable in the unaggregated data. Three mixed-effects logistic regressions were used to explore the effects of sonication in the different conditions on accuracy during the EDT.

To examine whether sonication in any of the ultrasound conditions affected accuracy during the EDT, and whether this differed for catch and main trials, we fit a mixed-effects logistic regression testing the three-way interaction between the effects of condition (sham, pulvinar, and central thalamus sonication), block (pre- and post-sonication), and trial type (main and catch) on accuracy with the following specification: accuracy ~ condition × block × trial type + (1 + condition | participant) + (1 | task order). To test whether sonication in any condition affected accuracy on main trials, we contrasted estimated marginal means for accuracy on main trials from post- to pre-sonication for each condition. To test whether the effect of sonication on accuracy during main trials differed between the conditions, we contrasted the estimated marginal means for the difference in accuracy on main trials at post- to pre-sonication between the conditions. The same contrasts were performed for catch trials.

To determine whether the effect of sonication in any of the conditions on accuracy depended on whether the visual cue appeared in the left (ipsilesional) or right (contralesional) hemifield, we fit a mixed-effects logistic regression testing the three-way interaction between condition (pulvinar, central thalamus, and sham sonication), block (pre- and post-sonication), and visual cue hemifield (left or right) on accuracy. The model took the following specification: accuracy ~ condition × block × cue hemifield + (1 + condition | participant) + (1 | task order). To test whether sonication in any condition

affected accuracy on catch trials when the cue appeared in the right (contralesional) hemifield, we contrasted estimated marginal means for accuracy on right-cue catch trials from post- to pre-sonication for each condition. To test whether the effect of sonication on accuracy during right-cue catch trials differed between the conditions, we contrasted estimated marginal means for accuracy on these trials from post- to pre-sonication between the conditions. The same contrasts were performed for catch trials with visual cues in the left (ipsilesional) hemifield.

Lastly, to determine whether sonication in any condition affected accuracy during main trials, and if this depended on whether the cue was valid or which visual field the target appeared in, we fit a mixed-effect logistic regression testing the three-way interaction between the effects of ultrasound (sham, pulvinar, central thalamus sonication), block (pre- and post-sonication), and cue-target location on response time with the following specification: accuracy ~ condition × block × cue-target location + (1 + condition | participant) + (1 | task order). We considered all valid-cue trials but limited invalid-cue trials to those in which the cue and target appeared in different visual fields to better isolate any hemispheric effects. Cue-target location was thus a categorical variable that had four levels, including valid-cue ipsilateral trials, valid-cue contralateral trials, cue ipsilateral but target contralateral (invalid-contralateral), and cue contralateral but target ipsilateral (invalid-ipsilateral) trials.

To test whether sonication in any condition affected accuracy for any level of cue-target location, we contrasted the estimated marginal means for accuracy from post- to pre-sonication for each condition at each level of cue-target location. To test whether the effect of sonication on accuracy differed between conditions with any cue-target location, we contrasted the estimated marginal means for accuracy from post- to pre-sonication between conditions for each cue-target location. To test whether sonication in any condition on accuracy differed between the cue-target locations, we contrasted the estimated marginal means for accuracy from post- to pre-sonication between cue-target locations for each condition. To test whether the difference in the effects of sonication between cue-target locations differed between conditions, we contrasted the estimated marginal means for accuracy from post- to pre-sonication between the cue-target locations and between the conditions.

Response time. To determine whether sonication in any condition affected response time on main trials, and if this depended on whether the cue was valid or which visual field the target appeared in, we fit a mixed-effect logistic regression testing the three-way interaction between the effects of ultrasound (sham, pulvinar, central thalamus sonication), block (pre- and post-sonication), and cue-target location on response time with the following specification: response time ~ condition × block × cue-target location + (1 + condition | participant) + (1 | task order). We considered all valid-cue trials but limited invalid-cue trials to those in which the cue and target appeared in different visual fields to better isolate any hemispheric effects. Cue-target location was thus a categorical variable that had four levels, including valid-cue ipsilateral trials, valid-cue contralateral trials, cue ipsilateral but target contralateral (invalid-contralateral), and cue contralateral but target ipsilateral (invalid-ipsilateral) trials.

To test whether sonication in any condition affected response time for any level of cue-target location, we contrasted the estimated marginal means for response time from post- to pre-sonication for each condition at each level of cue-target location. To

test whether the effect of sonication on response time differed between the conditions for any level of cue-target location, we contrasted the estimated marginal means for response time from post- to pre-sonication between the conditions for each level of cue-target location. To test whether sonication in any condition on response time differed between the levels of cue-target location, we contrasted the estimated marginal means for response time from post- to pre-sonication between the levels of cue-target location for each condition. To test whether the difference in the effects of sonication between the cue-target locations differed between the conditions, we contrasted the estimated marginal means for response time from post- to pre-sonication between the levels of cue-target location and between the conditions.

Acknowledgements

This work was made possible by funding from the Tiny Blue Dot Foundation.

Author contributions

A.R.H.: Methodology, Software, Validation, Formal Analysis, Investigation, Data Curation, Writing – Original Draft, Writing – Reviewing & Editing, Visualization, Supervision, Project Administration. J.A.C.: Conceptualization, Methodology, Software, Investigation, Data Curation, Writing – Review & Editing, Supervision, Project Administration. E.W.: Investigation, Data Curation, Writing – Original Draft, Writing - Review & Editing. S.A.: Investigation, Data Curation, Writing - Review & Editing. M.M.M.: Conceptualization, Methodology, Resources, Writing – Review & Editing, Supervision, Project Administration, Funding Acquisition.

Competing interests

The authors declare no competing interests.

References

1. Redinbaugh, M.J., et al., *Thalamus Modulates Consciousness via Layer-Specific Control of Cortex.* Neuron, 2020. **106**(1): p. 66-75.e12.
2. Wilke, M., I. Kagan, and R.A. Andersen, *Effects of pulvinar inactivation on spatial decision-making between equal and asymmetric reward options.* Journal of cognitive neuroscience, 2013. **25**(8): p. 1270-1283.
3. Alkire, M.T., et al., *Thalamic microinjection of nicotine reverses sevoflurane-induced loss of righting reflex in the rat.* The Journal of the American Society of Anesthesiologists, 2007. **107**(2): p. 264-272.
4. Alkire, M.T., et al. *Thalamic microinfusion of antibody to a voltage-gated potassium channel restores consciousness during anesthesia*. in *The Journal of the American Society of Anesthesiologists*. 2009. The American Society of Anesthesiologists.
5. Schiff, N.D., et al., *Behavioural improvements with thalamic stimulation after severe traumatic brain injury.* Nature, 2007. **448**(7153): p. 600-603.
6. Legon, W., et al., *A retrospective qualitative report of symptoms and safety from transcranial focused ultrasound for neuromodulation in humans.* Scientific Reports, 2020. **10**(1): p. 5573.
7. Blackmore, J., et al., *Ultrasound neuromodulation: a review of results, mechanisms and safety.* Ultrasound in Medicine & Biology, 2019. **45**(7): p. 1509-1536.
8. Sarica, C., et al., *Human Studies of Transcranial Ultrasound neuromodulation: A systematic review of effectiveness and safety.* Brain Stimulation, 2022. **15**(3): p. 737-746.
9. Darmani, G., et al., *Non-invasive transcranial ultrasound stimulation for neuromodulation.* Clinical Neurophysiology, 2022. **135**: p. 51-73.
10. Saalmann, Y.B., *Intralaminar and medial thalamic influence on cortical synchrony, information transmission and cognition.* Frontiers in Systems Neuroscience, 2014. **8**.
11. Jones, B.E. and T.Z. Yang, *The efferent projections from the reticular formation and the locus coeruleus studied by anterograde and retrograde axonal transport in the rat.* Journal of Comparative Neurology, 1985. **242**(1): p. 56-92.
12. Krout, K.E., R.E. Belzer, and A.D. Loewy, *Brainstem projections to midline and intralaminar thalamic nuclei of the rat.* Journal of Comparative Neurology, 2002. **448**(1): p. 53-101.
13. Moruzzi, G. and H.W. Magoun, *Brain stem reticular formation and activation of the EEG.* Electroencephalography and clinical neurophysiology, 1949. **1**(1-4): p. 455-473.
14. Berendse, H.W. and H.J. Groenewegen, *Restricted cortical termination fields of the midline and intralaminar thalamic nuclei in the rat.* Neuroscience, 1991. **42**(1): p. 73-102.
15. Sanchez-Vives, M.V. and D.A. McCormick, *Cellular and network mechanisms of rhythmic recurrent activity in neocortex.* Nature neuroscience, 2000. **3**(10): p. 1027-1034.
16. Bharioke, A., et al., *General anesthesia globally synchronizes activity selectively in layer 5 cortical pyramidal neurons.* Neuron, 2022. **110**(12): p. 2024-2040.e10.
17. Suzuki, M. and M.E. Larkum, *General anesthesia decouples cortical pyramidal neurons.* Cell, 2020. **180**(4): p. 666-676. e13.
18. Baker, J.L., et al., *Robust modulation of arousal regulation, performance, and frontostriatal activity through central thalamic deep brain stimulation in healthy nonhuman primates.* Journal of Neurophysiology, 2016. **116**(5): p. 2383-2404.

19. Redinbaugh, M.J., et al., *Thalamic deep brain stimulation paradigm to reduce consciousness: Cortico-striatal dynamics implicated in mechanisms of consciousness.* PLOS Computational Biology, 2022. **18**(7): p. e1010294.
20. Lioudyno, M.I., et al., *Shaker-related potassium channels in the central medial nucleus of the thalamus are important molecular targets for arousal suppression by volatile general anesthetics.* Journal of Neuroscience, 2013. **33**(41): p. 16310-16322.
21. Bastos, A.M., et al., *Neural effects of propofol-induced unconsciousness and its reversal using thalamic stimulation.* Elife, 2021. **10**: p. e60824.
22. Tasserie, J., et al., *Deep brain stimulation of the thalamus restores signatures of consciousness in a nonhuman primate model.* Science advances, 2022. **8**(11): p. eabl5547.
23. Luppi, A.I., et al., *Local orchestration of distributed functional patterns supporting loss and restoration of consciousness in the primate brain.* Nature Communications, 2024. **15**(1): p. 2171.
24. Cain, J.A., et al., *Ultrasonic thalamic stimulation in chronic disorders of consciousness.* Brain Stimulation, 2021. **14**(2): p. 301-303.
25. Cain, J.A., et al., *Ultrasonic Deep Brain Neuromodulation in Acute Disorders of Consciousness: A Proof-of-Concept.* Brain Sciences, 2022. **12**(4): p. 428.
26. Hopkins, A.R., et al., *Emerging Treatment for Patients with Disorders of Consciousness: The Field of Neuromodulation*, in *Coma and Disorders of Consciousness*. 2024, Springer. p. 147-208.
27. Adams, M.M., et al., *Visual cortical projections and chemoarchitecture of macaque monkey pulvinar.* Journal of Comparative Neurology, 2000. **419**(3): p. 377-393.
28. Arcaro, M.J., M.A. Pinsk, and S. Kastner, *The anatomical and functional organization of the human visual pulvinar.* Journal of Neuroscience, 2015. **35**(27): p. 9848-9871.
29. Gutierrez, C., et al., *Neurochemical and connectional organization of the dorsal pulvinar complex in monkeys.* Journal of Comparative Neurology, 2000. **419**(1): p. 61-86.
30. Kaas, J.H. and D.C. Lyon, *Pulvinar contributions to the dorsal and ventral streams of visual processing in primates.* Brain research reviews, 2007. **55**(2): p. 285-296.
31. Shipp, S., *The functional logic of cortico–pulvinar connections.* Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences, 2003. **358**(1438): p. 1605-1624.
32. Soares, J.G., et al., *Connectional and neurochemical subdivisions of the pulvinar in Cebus monkeys.* Visual Neuroscience, 2001. **18**(1): p. 25-41.
33. Wilke, M., K.-M. Mueller, and D.A. Leopold, *Neural activity in the visual thalamus reflects perceptual suppression.* Proceedings of the National Academy of Sciences, 2009. **106**(23): p. 9465-9470.
34. Petersen, S.E., D.L. Robinson, and W. Keys, *Pulvinar nuclei of the behaving rhesus monkey: visual responses and their modulation.* Journal of neurophysiology, 1985. **54**(4): p. 867-886.
35. Bender, D. and M. Youakim, *Effect of attentive fixation in macaque thalamus and cortex.* Journal of neurophysiology, 2001. **85**(1): p. 219-234.
36. Kastner, S., et al., *Functional imaging of the human lateral geniculate nucleus and pulvinar.* Journal of neurophysiology, 2004. **91**(1): p. 438-448.
37. Smith, A.T., et al., *Dissociating vision and visual attention in the human pulvinar.* Journal of neurophysiology, 2009. **101**(2): p. 917-925.

38. Saalmann, Y.B., et al., *The pulvinar regulates information transmission between cortical areas based on attention demands.* science, 2012. **337**(6095): p. 753-756.
39. Fiebelkorn, I.C., M.A. Pinsk, and S. Kastner, *The mediodorsal pulvinar coordinates the macaque fronto-parietal network during rhythmic spatial attention.* Nature Communications, 2019. **10**(1): p. 215.
40. Snow, J.C., et al., *Impaired attentional selection following lesions to human pulvinar: evidence for homology between human and monkey.* Proceedings of the National Academy of Sciences, 2009. **106**(10): p. 4054-4059.
41. Rafal, R.D. and M.I. Posner, *Deficits in human visual spatial attention following thalamic lesions.* Proceedings of the National Academy of Sciences, 1987. **84**(20): p. 7349-7353.
42. Karnath, H.O., M. Himmelbach, and C. Rorden, *The subcortical anatomy of human spatial neglect: putamen, caudate nucleus and pulvinar.* Brain, 2002. **125**(2): p. 350-360.
43. Danziger, S., et al., *The effects of unilateral pulvinar damage in humans on reflexive orienting and filtering of irrelevant information.* Behavioural Neurology, 1999. **13**(3-4): p. 95-104.
44. Petersen, S.E., D.L. Robinson, and J.D. Morris, *Contributions of the pulvinar to visual spatial attention.* Neuropsychologia, 1987. **25**(1): p. 97-105.
45. Wilke, M., et al., *Pulvinar inactivation disrupts selection of movement plans.* Journal of Neuroscience, 2010. **30**(25): p. 8650-8659.
46. Zhou, H., Robert J. Schafer, and R. Desimone, *Pulvinar-Cortex Interactions in Vision and Attention.* Neuron, 2016. **89**(1): p. 209-220.
47. Dinges, D.F. and J.W. Powell, *Microcomputer analyses of performance on a portable, simple visual RT task during sustained operations.* Behavior Research Methods, Instruments, & Computers, 1985. **17**(6): p. 652-655.
48. Egly, R., J. Driver, and R.D. Rafal, *Shifting visual attention between objects and locations: evidence from normal and parietal lesion subjects.* J Exp Psychol Gen, 1994. **123**(2): p. 161-77.
49. Lim, J. and D.F. Dinges, *A meta-analysis of the impact of short-term sleep deprivation on cognitive variables.* Psychological bulletin, 2010. **136**(3): p. 375.
50. Basner, M. and D.F. Dinges, *Maximizing sensitivity of the psychomotor vigilance test (PVT) to sleep loss.* Sleep, 2011. **34**(5): p. 581-91.
51. Doran, S.M., H.P. Van Dongen, and D.F. Dinges, *Sustained attention performance during sleep deprivation: evidence of state instability.* Archives italiennes de biologie, 2001. **139**(3): p. 253-267.
52. Van Dongen, H.P., et al., *The cumulative cost of additional wakefulness: dose-response effects on neurobehavioral functions and sleep physiology from chronic sleep restriction and total sleep deprivation.* Sleep, 2003. **26**(2): p. 117-26.
53. Chee, M.W.L., et al., *Lapsing during Sleep Deprivation Is Associated with Distributed Changes in Brain Activation.* The Journal of Neuroscience, 2008. **28**(21): p. 5519-5528.
54. Cain, J.A., et al., *Real time and delayed effects of subcortical low intensity focused ultrasound.* Scientific reports, 2021. **11**(1): p. 1-14.
55. Ma, N., et al., *How acute total sleep loss affects the attending brain: a meta-analysis of neuroimaging studies.* Sleep, 2015. **38**(2): p. 233-240.
56. Elvsåshagen, T., et al., *Cerebral blood flow changes after a day of wake, sleep, and sleep deprivation.* NeuroImage, 2019. **186**: p. 497-509.

57. Thomas, M., et al., *Neural basis of alertness and cognitive performance impairments during sleepiness. I. Effects of 24 h of sleep deprivation on waking human regional brain activity.* Journal of sleep research, 2000. **9**(4): p. 335-352.
58. Wu, J.C., et al., *The effect of sleep deprivation on cerebral glucose metabolic rate in normal humans assessed with positron emission tomography.* Sleep, 1991. **14**(2): p. 155-62.
59. Wu, J.C., et al., *Frontal lobe metabolic decreases with sleep deprivation not totally reversed by recovery sleep.* Neuropsychopharmacology, 2006. **31**(12): p. 2783-92.
60. Coull, J., et al., *The neural correlates of the noradrenergic modulation of human attention, arousal and learning.* European Journal of Neuroscience, 1997. **9**(3): p. 589-598.
61. Mackworth, J.F., *Performance decrement in vigilance, threshold, and high-speed perceptual motor tasks.* Canadian Journal of Psychology/Revue canadienne de psychologie, 1964. **18**(3): p. 209.
62. Pattyn, N., et al., *Psychophysiological investigation of vigilance decrement: boredom or cognitive fatigue?* Physiology & behavior, 2008. **93**(1-2): p. 369-378.
63. Hudson, A.N., H.P.A. Van Dongen, and K.A. Honn, *Sleep deprivation, vigilant attention, and brain function: a review.* Neuropsychopharmacology, 2020. **45**(1): p. 21-30.
64. Lim, J., et al., *Imaging brain fatigue from sustained mental workload: an ASL perfusion study of the time-on-task effect.* Neuroimage, 2010. **49**(4): p. 3426-3435.
65. Dinges, D.F. and N.B. Kribbs, *Performing while sleepy: Effects of experimentally-induced sleepiness*, in *Sleep, sleepiness and performance.* 1991, John Wiley & Sons: Oxford, England. p. 97-128.
66. Lim, J., et al., *Imaging brain fatigue from sustained mental workload: an ASL perfusion study of the time-on-task effect.* Neuroimage, 2010. **49**(4): p. 3426-35.
67. Langner, R. and S.B. Eickhoff, *Sustaining attention to simple tasks: a meta-analytic review of the neural mechanisms of vigilant attention.* Psychological bulletin, 2013. **139**(4): p. 870.
68. Cabeza, R. and L. Nyberg, *Imaging cognition II: An empirical review of 275 PET and fMRI studies.* Journal of cognitive neuroscience, 2000. **12**(1): p. 1-47.
69. Rueckert, L. and J. Grafman, *Sustained attention deficits in pat ients with right frontal lesions.* Neuropsychologia, 1996. **34**(10): p. 953-963.
70. Malhotra, P., E.J. Coulthard, and M. Husain, *Role of right posterior parietal cortex in maintaining attention to spatial locations over time.* Brain, 2009. **132**(3): p. 645-660.
71. Rueckert, L. and J. Grafman, *Sustained attention deficits in patients with lesions of posterior cortex.* Neuropsychologia, 1998. **36**(7): p. 653-660.
72. Coslett, H.B., D. Bowers, and K.M. Heilman, *Reduction in cerebral activation after right hemisphere stroke.* Neurology, 1987. **37**(6): p. 957-957.
73. Husain, M. and C. Rorden, *Non-spatially lateralized mechanisms in hemispatial neglect.* Nature Reviews Neuroscience, 2003. **4**(1): p. 26-36.
74. Fiebelkorn, I.C., Y.B. Saalmann, and S. Kastner, *Rhythmic sampling within and between objects despite sustained attention at a cued location.* Current biology, 2013. **23**(24): p. 2553-2558.
75. Fiebelkorn, I.C., M.A. Pinsk, and S. Kastner, *A Dynamic Interplay within the Frontoparietal Network Underlies Rhythmic Spatial Attention.* Neuron, 2018. **99**(4): p. 842-853.e8.

76. Helfrich, R.F., et al., *Neural Mechanisms of Sustained Attention Are Rhythmic.* Neuron, 2018. **99**(4): p. 854-865.e5.
77. Helfrich, R.F., et al., *Neural mechanisms of sustained attention are rhythmic.* Neuron, 2018. **99**(4): p. 854-865. e5.
78. Li, X., et al., *The first step for neuroimaging data analysis: DICOM to NIfTI conversion.* Journal of neuroscience methods, 2016. **264**: p. 47-56.
79. Jenkinson, M., et al., *Fsl.* Neuroimage, 2012. **62**(2): p. 782-790.
80. Jenkinson, M., et al., *Improved optimization for the robust and accurate linear registration and motion correction of brain images.* Neuroimage, 2002. **17**(2): p. 825-841.
81. Schafer, M.E., et al., *Design, Development, and Operation of a Low-Intensity Focused Ultrasound Pulsation (LIFUP) System for Clinical Use.* IEEE Trans Ultrason Ferroelectr Freq Control, 2021. **68**(1): p. 54-64.
82. Schafer, S.F., *Acoustic Coupling Pads for the Control of Ultrasound Neuromodulation Exposure*. 2023, Drexel University: United States -- Pennsylvania. p. 84.
83. Administration, U.S.F.a.D., *Marketing Clearance of Diagnostic Ultrasound Systems and Transducers, Guidance for Industry and Food and Drug Administration Staff*. 2019.
84. Medicine, A.I.o.U.i., *Statement on Mammalian Biological Effects of Ultrasound In Vivo*. 2015.
85. Bystritsky, A., et al., *A review of low-intensity focused ultrasound pulsation.* Brain stimulation, 2011. **4**(3): p. 125-136.
86. Duck, F.A., *Medical and non-medical protection standards for ultrasound and infrasound.* Progress in Biophysics and Molecular Biology, 2007. **93**(1): p. 176-191.
87. Eames, M.D.C., et al., *Trans-cranial focused ultrasound without hair shaving: feasibility study in an ex vivo cadaver model.* Journal of Therapeutic Ultrasound, 2014. **1**(1): p. 24.
88. Raymond, S.B. and K. Hynynen, *Acoustic transmission losses and field alterations due to human scalp hair.* IEEE transactions on ultrasonics, ferroelectrics, and frequency control, 2005. **52**(8): p. 1415-1419.
89. Peirce, J., et al., *PsychoPy2: Experiments in behavior made easy.* Behavior Research Methods, 2019. **51**(1): p. 195-203.
90. Dorrian, J., N.L. Rogers, and D.F. Dinges, *Psychomotor vigilance performance: Neurocognitive assay sensitive to sleep loss.* Sleep deprivation, 2004: p. 39-70.
91. Lim, J. and D.F. Dinges, *Sleep deprivation and vigilant attention.* Annals of the New York Academy of Sciences, 2008. **1129**(1): p. 305-322.
92. Jewett, M.E., et al., *Dose-response relationship between sleep duration and human psychomotor vigilance and subjective alertness.* Sleep, 1999. **22**(2): p. 171-179.
93. Van Dongen, H.P. and D.F. Dinges, *Investigating the interaction between the homeostatic and circadian processes of sleep–wake regulation for the prediction of waking neurobehavioural performance.* Journal of sleep research, 2003. **12**(3): p. 181-187.
94. Basner, M. and D.F. Dinges, *Maximizing Sensitivity of the Psychomotor Vigilance Test (PVT) to Sleep Loss.* Sleep, 2011. **34**(5): p. 581-591.
95. Graw, P., et al., *Circadian and wake-dependent modulation of fastest and slowest reaction times during the psychomotor vigilance task.* Physiology & Behavior, 2004. **80**(5): p. 695-701.

96. Drummond, S.P., et al., *The neural basis of the psychomotor vigilance task.* Sleep, 2005. **28**(9): p. 1059-1068.
97. Posner, M.I., *Orienting of attention.* Quarterly journal of experimental psychology, 1980. **32**(1): p. 3-25.
98. Bates, D., et al., *Fitting linear mixed-effects models using lme4.* arXiv preprint arXiv:1406.5823, 2014.
99. Kuznetsova, A., P.B. Brockhoff, and R.H. Christensen, *lmerTest package: tests in linear mixed effects models.* Journal of statistical software, 2017. **82**: p. 1-26.
100. R Core Team, R., *R: A language and environment for statistical computing.* 2023.
101. Benjamini, Y. and Y. Hochberg, *Controlling the False Discovery Rate: A Practical and Powerful Approach to Multiple Testing.* Journal of the Royal Statistical Society: Series B (Methodological), 1995. **57**(1): p. 289-300.
102. Tukey, J., *Exploratory Data Analysis Pearson.* London, United Kingdom.[Google Scholar], 1977.
103. Data, M.C., et al., *Noise versus outliers.* Secondary analysis of electronic health records, 2016: p. 163-183.