Electrophysiological biomarkers of behavioral dimensions from cross-species paradigms

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INTRODUCTION

Many clinical treatment trials in psychiatry have failed at the cost of time, effort, money, and the hope of the patients tested. These translational failures are often attributed to either a lack of consistent quantification of the same neural processes across species [1, 2] or to the use of “fast and dirty” behavioral techniques that have little-to-no relevance to human testing [3]. In response, the National Institutes of Mental Health (NIMH) formed the Cognitive Neuroscience Treatment Research to Improve Cognition in Schizophrenia (CNTRICS) to identify cognitive systems and component processes that could be tested across species [1]. Continuing this theme, NIMH also initiated the Research Domain Criteria (RDoC) initiative [4, 5], promoting a focus on specific behavioral dimensions and related neurophysiological circuits instead of end phenotypes. A common theme across these new paradigms is the need for brain-based neural signals that are specifically linked to behavioral dimensions, that must be sensitive to systemic alterations due to mental health disorders, and that should ideally be translatable between the species. Ultimately, the availability of specific, sensitive, and translatable neural signals would increase the likelihood of positive animal trial results being translated to positive clinical trial results. Motivated by a specific UH2/3 funding mechanism from the NIMH, we aimed to test three candidate behavioral assays and assess the homology of concurrent neurophysiologic responses across species (UH2 phase), with future studies confirming pharmacologic sensitivity across species (UH3 phase).

Candidate domains that are deficient in psychiatric disorders include effortful motivation, reinforcement learning, and cognitive control. Effortful motivation is recognized as a core contributor to psychosocial impairments in psychiatric conditions, ranging from amotivation in people with schizophrenia and depression to increased goal-directed activity in mania. There are various methods for assessing effort-based decision making, each with associated deficits observed across psychiatric conditions [6–9]. Motivational deficits can also be measured across species, although techniques vary widely [10–12]. One method for measuring effortful motivation is the progressive ratio breakpoint task, linked to a single, well-defined action requirement. Motivation is measured by the point that the participant stops responding to gain a reward, is reduced in people with schizophrenia [13, 14], and accounts for 24% of the variance in their global cognitive functioning [15]. A reduced breakpoint is also observed in animal models relevant to schizophrenia [16], while an increased breakpoint is observed in animal models of mania [17]. Thus, effortful motivation can be measured in a manner consistent across species.

Another promising experimental domain is reinforcement learning, which requires an agent to learn stimulus-action pairings based on rewarding or punishing outcomes. These outcomes are often delivered probabilistically, requiring long-term integration of action values [18, 19]. Probabilistic reinforcement learning paradigms are naturally transferrable across vertebrates [20–23],

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and are thus an ideal candidate for domain consistency. Probabilistic learning deficits are observed in people with psychiatric conditions, such as schizophrenia [24, 25], bipolar disorder [26], and depression [27–29], bolstering the translational utility of findings. Reinforcement learning theory provides a quantification of abstract processes [30], facilitating an interpretation of neural signals by their confirmation to theorized parameters and computations.

Finally, cognitive control is a domain that is reliably associated with psychiatric distress. Cognitive control requires goal-driven action selection over prepotent tendencies [31, 32], and it can be elicited using several paradigms including various continuous performance tests (CPTs). Prior to the development of the five-choice (5C)-CPT [33], cognitive control and attention were not typically measurable in the same task in rodents. The 5C-CPT has since been reverse-translated for use in humans and used to provide evidence that cognitive control is deficient in schizophrenia [34] and bipolar disorder [35]. Cross-species pharmacological predictive validity has been demonstrated by the effects of amphetamine, which improves 5C-CPT performance in humans, rats, and mice [35, 36]. Importantly, for cognitive control, a measure of response inhibition (false alarm rate) is functionally separable from the more traditional impulsivity measure of premature responses, as evidenced by dopamine D4 receptor and 5-HT2C mechanism sensitivity, respectively [37].

Across these three task domains of effortful motivation, reinforcement learning, and cognitive control it is possible to assess behaviors with preserved consistency across species with outcomes that are sensitive to deficits in clinical populations. However, behavioral consistency has proven insufficient, and shared neural substrates of task engagement are necessary to increase confidence in any treatment translated across species. While there are numerous studies advancing candidate biomarkers of specific domains, many techniques are inherently ill-suited for translating behavioral or neurophysiology between species. Fixed-head techniques like fMRI in humans or calcium imaging in animals have limited translatability. Invasive recordings like depth electrophysiology are compelling but such studies are rare in humans. Electrophysiological recordings naturally encompass multiple scales of measurement in a hierarchical, integrated manner. For example, local fields couple to scalp-recorded EEG: regardless of scale (depth, dura, scalp, etc.), field activity is always measured [38]. Thus, electrophysiology is uniquely well-suited for addressing questions about translatable neural signal biomarkers.

Even with the methodologic promise of comparative electrophysiology, a major impediment toward improving this species gap has been the difficulty of developing paradigms that 1) can quantify EEG responses related to specific behaviors, 2) are impacted by mental health disorders, and 3) are suitable for both human and animal studies. Fortunately, the advent of touchscreen technology for rodents has greatly increased the sophistication of behavioral testing. Here, we detail RDoC-relevant behavioral domains impacted by mental health (effortful motivation, reward learning, and cognitive control) that can be quantified in similar tasks across humans and mice and that are associated with an a priori defined candidate spectral EEG biomarker (Fig. 1). Only some of these behavioral and neural signatures were successfully translated here—yet even failures yielded critical lessons for advancing this field.

Fig. 1 Schematic electroencephalograph (EEG) recording in humans and mice. The present studies utilized EEG recordings in humans and mice while they performed tasks that probed RDoC-relevant domains of functioning, including effortful motivation, reward learning, and cognitive control. Humans used a joystick to respond to on-screen stimuli, while mice responded using a touchscreen. Scalp (human) and dura (mice) EEG recordings were recorded during the execution of these tasks. Time-frequency regions-of-interest were contrasted between task conditions to compare neural signatures of these RDoC domains.
METHODS AND MATERIALS

Human participants
The human portion of this study was conducted at the UCSD Medical Center, with approval from the UCSD Human Subject Institutional Review Board. Healthy men and women (18–35 years; n = 57) were recruited from the community and monetarily compensated for participation. First, subjects underwent phone screening to assess current and past medical and psychiatric history, medication and recreational drug use, and family history of psychosis. Following informed consent, participants completed an in-depth screening visit, including a physical examination, urine toxicology screen, and urine pregnancy test. All exclusion criteria consisted of 216 trials: 90 target and 18 nontarget stimuli for each of the three blocks of 80 trials. The target and nontarget stimuli were presented in a pseudorandom order (to ensure no more than three of the same trial types in a row), with a 1 sec response window available for all trials and a variable intertrial interval (ITI; 500, 1000, or 1500 ms). The full task consisted of 216 trials: 90 target and 18 nontarget stimuli for each of the difficult conditions. Composite metrics of task performance were used in the analysis of performance, including hit rate, false alarm rate (FAR), d' prime, and bias.

Human electrophysiological recording and preprocessing
Continuous electrophysiological (EEG) data were recorded using a BioSemi Active Two system. Data were recorded in DC mode from 64 scalp leads, four electrooculogram (EOG) leads recorded at the superior and inferior orbit of the left eye and outer canthi of each eye, and one nose and two mastoid electrodes for offline re-referencing. The electrode offsets were kept below 25 mV and all channels were referenced to the system’s internal loop (CMS/DRL electrodes). All data were collected using a 512 Hz sampling rate utilizing a first-order antialiasing filter. Custom Matlab scripts and EEGLab [39] functions were used for all data processing. Data were first epoched around the imperative stimuli and then average referenced. Bad channels and bad epochs were identified using a conjunction of the FASTER algorithm [40] and pop_rejchan from EEGLab and were subsequently interpolated and rejected, respectively. Eye blinks were removed following independent component analysis in EEGLab.

Animal subjects
Male and female C57BL/6J mice were obtained from The Jackson Laboratory (Bar Harbor, ME), housed in same-sex groupings of two per cage in a temperature- and humidity-controlled vivarium under a reverse 12 h light/dark cycle (lights off:0800 h) and tested during the dark phase. A total of 12 male and 12 female mice were used. All experimental procedures were performed in accordance with the National Institutes of Health Guide for Care and Use of Laboratory Animals and were approved by the University of New Mexico Health Sciences Center Institutional Animal Care and Use Committee. See Supplemental Materials for information on touchscreen pretraining. All rewarding outcomes included...
Mice were trained in the 5C-CPT as previously described [36] (see Supplemental Materials and Supplemental Figure S1). Target trials were indicated by illumination of a single stimulus window; nontarget trials consisted of illumination of all windows. Hits and correct rejections were rewarded. False alarms resulted in a 10 s timeout period. Mice were then tethered to the recording apparatus for two sessions of 2:1 difficulty conditions were included, with easy (3 s response window) and hard (1.5 s response window) trials across ten recording sessions.

Mouse probabilistic learning task (PLT)
Throughout each session of the PLT, mice were presented with three pairs of unique stimuli (fan/marble, honey/cave, spider/fan) in three separate 20-trial blocks. For the first block, one stimulus was rewarded 90% of the time and the other was rewarded 10% of the time. The breakpoint was the last ratio completed at the end of the 1-h session. Mice completed one session of PRBT.

Mouse five-choice continuous performance task (SC-CPT)
Mice were trained in the SC-CPT as previously described [36] (see Supplemental Materials and Supplemental Figure S1). Target trials were indicated by illumination of a single stimulus window; nontarget trials consisted of illumination of all five windows. Hits and correct rejections were rewarded. False alarms resulted in a 10 s timeout period. Mice were first trained on a 2:1 ratio (2 target trials to 1 nontarget) for five sessions. They were then tethered to the recording apparatus for two sessions of 2:1 to acclimate to the head stage, and then moved to a 5:1 ratio. Similar to the human SC-CPT, two different difficulty conditions were included, with easy (3 s response window) and hard (1.5 s response window) trials across ten recording sessions.

Human and mouse EEG processing
For the sake of descriptive simplicity, both the scalp-recorded signal in humans and the dura-recorded signal in mice are referred to as “EEG.” Time-frequency measures were computed by multiplying the fast Fourier transformed (FFT) power spectrum of single-trial EEG data with the FFT power spectrum of a set of complex Morlet wavelets defined as a Gaussian-windowed complex sine wave: $$e^{i2\pi ft}e^{-t^2/(2\sigma^2)}$$, where \( t \) is time, \( f \) is frequency (which increased from 1–50 Hz in 50 logarithmically spaced steps), and the width (or “cycles”) of each frequency band was set to increase from 3/(2f) to 10/(2f) as frequency increased. Then, the time series was recovered by computing the inverse FFT. The end result of this process is identical to time-domain signal convolution, and resulted in estimates of instantaneous power taken from the magnitude of the analytic signal. Each epoch was then cut in length (cues: –500 to +1000 ms; responses: –1000 to +500 ms).

Averaged power was normalized by conversion to a decibel (dB) scale (10*\text{log10}(power(t)/power(baseline))), allowing a direct comparison of effects across frequency bands. The baseline consisted of averaged power -300 to -200 ms before all task-specific stimuli, except the response-locked mouse SC-CPT trials, which benefitted from greater trial-specific clarity using a preresponse –800 to –700 ms window. A 100 ms duration is often used as an effective baseline, since pixel-wise time-frequency data points have already been resolved over smoothed temporal and frequency dimensions with the wavelets. For the PRBT, the entire duration of all epochs was used as the baseline.

**Statistical analysis**
Species were analyzed with separate mixed-effects models. For mice, individual sessions were concatenated and each mouse was treated as a random effect, similar to humans. The contrast conditions within each task were treated as fixed effects. For mouse data, only trials with at least 30 epochs were used in the SC-CPT or PLT (PRBT always used five trials at the beginning and five trials at the end). In the human dataset, there were clear a priori hypotheses and there was more level-2 data (more subjects), so a smaller threshold was used for level-1 rejection (trials). For the SC-CPT, this minimum was ten trials and for the PLT, the minimum was 20 trials. For the PRBT, 1-s epochs were averaged for the first 50 s and the last 50 s of the task.

Analysis of Variance (ANOVAs) and t tests were used to test hypotheses about condition-specific differences within each task, separately for each species. All tests were two-tailed. We also determined whether sex moderated these effects, although there were no specific hypotheses about the role of sex. Test statistics are shown in Tables 1 & 2. Simple effects contrasts are shown in Table 3 along with the time and frequency periods.
frequency ranges for each tf-ROI. All effect sizes are presented as partial eta-squared ($\eta^2$) or Cohen’s $d$ (mean difference divided by the pooled standard deviation).

RESULTS
Statistical differentiation followed an a priori approach, where each task had a predicted spatial, temporal, and frequency range for the contrast of interest. These time-frequency regions-of-interest (tf-ROIs) were broadly defined based on well-replicated findings from the human EEG literature (detailed for each task below). In the discussion, we note how the exact tf-ROIs discovered here will be used in future pharmacologic studies, providing a chance for direct replication and theoretical extension of the candidate biomarkers. Each figure shows the tf-ROI in magenta, as well as topographic plots highlighting the target electrode.

Predictions: PRBT
This task required subjects to engage in active behavior to gain a reward at each level. In humans, levels increased after rotating the joystick, while in mice, levels increased after sufficient touches to the screen. In both cases, the number of actions required for the next reward progressively increased. The point at which the subject stopped responding was identified as their breakpoint and was used as an index of effortful motivation. Previous EEG studies have implicated alpha power as a concomitant of effortful behavior in humans [41–43], including changes due to physical and mental fatigue [44, 45]. Here, we examined if this relationship was present during the PRBT and if it was common between species. The alpha-band was defined as 8–12 Hz, and electrode POz was selected to be within the mass of broad posterior alpha. Epochs were locked to the first 50 and last 50 s at electrode POz in humans, and to the first five and last five rewarded responses in the posterior lead in rodents. Since this alpha-band effect was expected to be relatively consistent across events, the time window was arbitrarily set from 0–200 ms postevent. It was hypothesized that alpha power at this posterior lead would be larger at the end of the task, as indicated for physical vs. cognitive effort [46].

Outcomes: PRBT
In humans, the breakpoint was around 7 (Fig. 2C). In mice, the breakpoint was around 4 (Fig. 2D). There were no sex differences

Fig. 4 The five-choice continuous performance task (5C-CPT) had two levels of difficulty. A–B In humans, difficulty was manipulated with easy (unmasked) vs. hard (masked) visual contrast conditions. Difficulty altered d prime but not bias. C–D In mice, difficulty was modulated with easy (3 s delay) vs. hard (1.5 s delay) conditions. Task demand did not change d prime or bias in mice. E–F Time-frequency plots of response-locked data at FCz in humans or the anterior lead in mice. Since a correct nontarget (nogo) condition does not require a response, these epochs were time-locked to the end of the delay period. The magenta boxes show the theta-band tf-ROI. G–H EEG tf-ROI quantification of the go easy vs. go hard difference in prerresponse theta power. Green asterisks indicate statistically significant (p < 0.05) within-subject differences.
in either the number of trials completed or the breakpoint (human $t < 0.01$, mouse $t < 0.12$). Following minimum epoch count requirements, and due to two technical problems in human EEG, there were $n = 52$ humans ($M = 24$, $F = 28$) and $n = 20$ mice ($M = 11$, $F = 9$). Both the humans and mice had a significant late > early alpha power contrast (Table 1). There were no main or interactive effects with sex for either species.

Unlike the other experiments in this report, and to the best of our knowledge, the hypothesis of an alpha-band marker of breakpoint-related effort had not been tested. This alpha difference (last minus first) was proposed to scale with greater motivation loss, and it was indeed negatively correlated with the breakpoint in humans ($p = 0.046$; Supplemental Figure S2). Notably, time-on-task, as measured by the number of seconds on the PRBT did not correlate with breakpoint (rho $= 0.18$). This outcome highlights the fact that participants achieved a higher breakpoint through effort, which correlated with alpha-band difference, not time. A stepwise regression verified this specific relationship, where seconds did not correlate with the alpha difference ($F < 1$), yet the addition of the breakpoint in the next level led to a significant $F$ change ($F(2, 49) = 4.03$, $p < 0.001$).

| Table 1. Test statistics for two (sex) * two (condition) ANOVAs for EEG time-frequency regions-of-interest (tf-ROIs). |

| TF-ROI | df | Main: time | Main: sex | Diff: sex |
|-------|----|------------|-----------|-----------|
| PRBT  | 6  | $F = 0.45, p = 0.59$ | $F = 0.21, p = 0.63$ | $F = 0.00, p = 0.90$ |
| Mouse | 6  | $F = 0.35, p = 0.71$ | $F = 0.01, p = 0.90$ | $F = 0.00, p = 0.90$ |
| 5C-CPT| 6  | $F = 0.75, p = 0.38$ | $F = 0.01, p = 0.90$ | $F = 0.00, p = 0.90$ |

**Table 2.** Test statistics for 2 (sex) * 2 (condition) ANOVAs for behavioral performance on the PLT and 5C CPT.

| TF-ROI | df | Main: probability | Main: sex | Prob* sex |
|--------|----|------------------|-----------|-----------|
| Human: accuracy | 1.51 | $F = 54.40, p < 0.001$ | $F = 0.87, p = 0.36$ | $F = 0.02, p = 0.97$ |
| Mouse: accuracy | 1.18 | $F = 0.02, p = 0.90$ | $F = 2.24, p = 0.15$ | $F = 0.11, p = 0.75$ |
| 5C-CPT | df | Main: difficulty | Main: sex | Diff: sex |
| Human: hit rate | 1.53 | $F = 23.07, p < 0.001$ | $F = 1.41, p = 0.24$ | $F = 0.03, p = 0.87$ |
| Mouse: hit rate | 1.13 | $F = 0.58, p = 0.46$ | $F = 0.02, p = 0.89$ | $F = 0.17, p = 0.68$ |
| Human: FA | 1.53 | $F = 2.01, p = 0.16$ | $F = 0.97, p = 0.33$ | $F = 0.01, p = 0.94$ |
| Mouse: FA | 1.13 | $F = 4.33, p = 0.06$ | $F = 2.32, p = 0.15$ | $F = 0.14, p = 0.70$ |
| Human: d prime | 1.53 | $F = 28.78, p < 0.001$ | $F = 3.27, p = 0.08$ | $F = 0.01, p = 0.96$ |
| Mouse: d prime | 1.13 | $F = 0.91, p = 0.34$ | $F = 0.53, p = 0.48$ | $F = 0.13, p = 0.72$ |
| Human: bias | 1.53 | $F = 1.33, p = 0.25$ | $F = 1.72, p = 0.20$ | $F = 0.09, p = 0.99$ |
| Mouse: bias | 1.13 | $F = 3.48, p = 0.09$ | $F = 3.52, p = 0.08$ | $F = 0.13, p = 0.75$ |
| Human: hit RT | 1.53 | $F = 146.59, p < 0.001$ | $F = 1.76, p = 0.19$ | $F = 0.05, p = 0.93$ |
| Mouse: hit RT | 1.13 | $F = 9.29, p = 0.008$ | $F = 1.18, p = 0.30$ | $F = 0.15, p = 0.25$ |

Bold values represent $p$ values and effect sizes.
The analysis of mouse performance required some different operational definitions and statistical approaches, since they always had one hour to complete the task and most mice stopped at a breakpoint of “four” while a few stopped at “seven.” In mice, there was no relationship between alpha power and the number of epochs completed (\(\rho(22) = -0.09, p = 0.70\)), although this may be due to a reduced sample size. When analyzed as two groups, the mice with a breakpoint of “four” had a nonsignificantly higher alpha power than those with a breakpoint of “seven” (\(t(18) = 1.21, p = 0.24\)), supporting the premise that a higher sample size may have yielded the same correlation seen in humans. Predictions: PLT

Trials that resulted in correct feedbacks were used for all analyses. In mice, rewarded responses were immediately indicated by a 1 s, pure noise tone concomitant with the illumination of the magazine light and delivery of the reward. Comparisons were split based on the probabilistic aspect of the reward feedback, creating high probability (i.e., target response followed by reward) vs. low probability (i.e., nontarget response followed by reward) contrasts. While this contrast is ideal for comparing the same process without interference from different sensory or imperative events, it unfortunately conflicted with our strong epoch count requirements (see Methods and Materials). These criteria led to the necessity of splitting the data based on the exact same number (first cohort: mean difference = 0.65 dB, CI = 0.14, 1.15; second cohort mean difference = 0.65 dB, CI = −0.97, 2.27). Although not included in the a priori hypotheses, analyses for EEG time-frequency region of interests for punishment-related theta with statistical analyses (Supplemental Tables S1 & S2), with corresponding theta power representation (Supplemental Figure S3), are described, in addition to correlations to mouse accuracy related to reward- and punishment-associated delta power differences (Supplemental Figure S4).

Predictions: SC-CPT

Only hits on target trials and correct rejections on nontarget trials were used for EEG analysis. This novel SC-CPT also introduced two varying difficulty levels using backward masks. In humans, these were easy (standard, unmasked) and hard (masked) visual contrast conditions. In rodents, we utilized supposedly easy (3 s delay) and hard (1.5 s delay) conditions. In mice, rewards were immediately indicated by a 1 s, pure noise tone concomitant with the illumination of the magazine light and delivery of reward. These rewards were locked to the response on hits and the end of the delay period on correct rejections. The nontarget vs. target contrasts were expected to elicit frontal midline theta power, which is a reliable indicator of cognitive conflict [54, 55]. However, it was not possible to verify that cues were visually attended to by the mice, so response-locked epochs were used for both species. Epochs were locked to responses at electrode FCz in humans—where the reward positivity ERP component is maximal [47–49]—and at the frontal lead in rodents. We hypothesized that low vs. high probability rewards would elicit a frontal midline delta-band power burst [47, 50]. While this reward-locked delta burst is reliably observed in humans, the timing and frequency varies between the published studies [47, 49–51]. Here, the temporal window was defined from 250 to 550 ms post-feedback; however, the frequency window was 1.3–2 Hz for humans and 1–1.4 Hz for mice.

Outcomes: PLT

For humans, overall PLT accuracy was greater than chance, with no difference between the sexes (Table 2). For mice, overall accuracy did not differ from chance. However, many mice were excluded from subsequent analysis due to a low number of epochs; the accuracy of the cohort used in EEG analysis was significantly higher than chance (\(t(13) = 2.26, p = 0.04, d = 0.60\)), with no difference between sexes (\(r < 1\)). Following these minimum epoch requirements for high and low probability events, the sample sizes of EEG analyses were reduced (human: \(M = 7, F = 11\); mouse: \(M = 5, F = 8\)). Both the humans and mice had a significant low > high probability delta-band contrast, with a significant main effect of sex in humans (males > females), (Table 1). While this carefully contrasted delta-band effect in mice is compelling, it was disappointing that the mice performed so indiscriminately during EEG assessment. To test the reliability of this delta-band contrast, a separate cohort (\(N = 12; M = 6, F = 6\)) was tested over 6 days on a single pair of stimuli that had 100 vs. 50% probabilities of reward. All mice performed at around 80% accuracy (i.e., they selected the 100% rewarding option 80% of the time: \(t(11) = 20.90, p < 0.001, d = 6.03\)), suggesting a high level of intrinsic exploration (Fig. 3I). Critically, time-frequency contrasts revealed a surprise-evoked delta-band burst in the same tf-ROI (Fig. 3J-K). Although this cohort did not reveal a significant statistical differentiation between conditions (\(t(11) = 0.89, p = 0.39, d = 0.18\)), this may still be expected from a true effect. The \(p\)-value alone is a poor metric for assessing replicability; effect sizes and confidence intervals are more useful for assessing the utility of an experimental outcome [52, 53]. Here, we observed that the mean difference between conditions were in fact the exact same number (first cohort: mean difference = 0.65 dB, CI = 0.14, 1.15; second cohort mean difference = 0.65 dB, CI = −0.97, 2.27). Although not included in the a priori hypotheses, analyses for EEG time-frequency region of interests for punishment-related theta with statistical analyses (Supplemental Tables S1 & S2), with corresponding theta power representation (Supplemental Figure S3), are described, in addition to correlations to mouse accuracy related to reward- and punishment-associated delta power differences (Supplemental Figure S4).

Outcomes: SC-CPT

In humans, the difficulty manipulation (masking), caused a significantly lower hit rate, longer RTs, and lower \(d'\) prime, indicative of worse attention but no change to false alarms (response inhibition) or, importantly, bias of responding.

Table 3. Summary of simple effects.

| Test        | Low freq | High freq | Start time | End time | \(t\)  | df | \(p\)     | \(d\) | Match? |
|-------------|----------|-----------|------------|----------|-------|----|-----------|------|--------|
| PRBT alpha  | Human    | 8         | 12         | 0        | 200   | 6.14| 51        | <0.001| 0.92   | Yes    |
|             | Mouse    | 8         | 12         | 0        | 200   | 2.15| 19        | 0.04  | 0.66   |        |
| PLT delta   | Human    | 1.3       | 2          | 250      | 550   | 2.44| 17        | 0.03  | 0.56   | Yes    |
|             | Mouse    | 1         | 1.4        | 250      | 550   | 2.78| 12        | 0.02  | 0.40   |        |
| SC-CPT theta| Human    | 4         | 8          | −500     | 0     | 5.58| 54        | <0.001| 1.06   | No     |
|             | Mouse    | 4         | 8          | −500     | 0     | 0.68| 10        | 0.51  | 0.26   |        |

Time and frequency ranges for event-related tf-ROIs and simple effects statistical contrasts (paired \(t\) statistic, Cohen’s \(d\)) within each task. For the PRBT, the contrasts are early > late trials. For PLT, contrasts are low > high reward probability. For the SC-CPT, the simple effect is the go hard > go easy condition.
DISCUSSION

Here, we report that consistent behaviors and related neural signatures can be elicited across various tasks and domains in humans and mice. These candidate EEG responses displayed remarkable temporal, spatial, and frequency consistency between species, largely consistent with our a priori hypotheses. Specifically, the PRBT (effortful motivation) and PLT (reward learning) revealed consistent neural signatures of posterior alpha and reward delta respectively, seen in both humans and mice while performing these tasks. Additionally, the SC-CPT revealed consistent target-locked theta across species.

Effortful motivation: PRBT

The behavioral performance of humans and mice in the PRBT was consistent with earlier reports [15, 16, 58]. Previous EEG studies have implicated alpha power with effortful behavior in humans [41–43], including changes due to physical and mental fatigue [44, 45]. More recently, diminished alpha power was described in mice lacking metabotropic glutamate receptor 5 [59], and rats lacking the Fmr1 gene [60], although it is not clear if this was tied to motivational state since it was simply in awake rodents. Our present data, therefore, add to human literature showing a duration-specific decline in posterior alpha power in humans, confirming this same effect in mice performing the PRBT, thereby enabling assessment of both patient populations and their rodent models. The scale of this alpha power decline correlated with the breakpoint in humans, but evidence for a similar relationship in mice was uncertain, likely due to lower sample sizes. Some evidence in support of the relationship emerged when comparing the alpha power of animals with differing breakpoints and requires future study. Given that posterior alpha is the single most dominant background rhythm in humans, these data support the idea that some common neural architecture is preserved across mammalian species that is stimulated during the performance of the same task. Future studies will have to confirm that this neural correlate of effortful performance is altered across clinical populations and in animals manipulated to be relevant to the population, and whether it is sensitive to pharmacologic agents.

Reward learning: PLT

While humans were predictably effective at performing this task, mice performed just above chance, unless the task was simplified. Despite these addressable difficulties in training and performance, the similarities between tasks facilitates comparison of EEG responses during task completion. The analytic contrasts were able to be well-controlled within each species, facilitating a comparison of the underlying process (e.g., low vs. high probability corresponding to high vs. low reinforcement prediction error), without interference from different sensory or imperative stimuli. The prediction of a delta-band enhancement to reward surprise was borne out in both species. An additional study with easier discriminability replicated the observation of the delta-band effect with consistent confidence intervals, albeit not the statistical differentiation. This spectral representation of the reward positivity ERP component has been described in humans, particularly its sensitivity to formal estimates of reward prediction error [50]. These findings are the first demonstration of this same spectral response in dura-recording from rodents, although a similar slow cingulate-recorded ERP response in this same time range was observed in the difference between the reward and punishment trials in rats [61]. Mice are prey species and are more sensitive to punishment [62, 63] than rats in similar paradigms [64]. Although not specified by our a priori predictions, we also investigated punishment surprise-evoked theta power (Supplemental Figure S4). However, this response was not significantly modulated in mice.

Cognitive control: SC-CPT

The SC-CPT assesses cognitive control and is sensitive to deficits in clinical populations and modulations by pharmacologic agents. Although humans easily maintain focus on the screen between stimuli (enabling EEG assessment locked to stimulus presentation), such assessment is much more difficult in mice given their need to turn around toward the food delivery area, thereby increasing misses to the moment of stimulus presentation, limiting stimulus-locked EEG events. Without aggressive implementation changes, such as head-fixing, mice are unlikely to reliably visually attend to the screen during the ITI, driving stimulus-locked EEG events, unlike humans. The addition of different auditory tones for target and nontarget trials may be needed for effective stimulus-locked manipulation for future trials, though the need for trial-and-error parameterization will likely delay the utilization of this task. The response-locked differentiation of EEG signals to target and nontarget trials presented here is technically a misnomer because correct rejections to nontarget trials do not include a response.

Response-locked theta was strongly enhanced in more difficult hit trials in humans. While response-locked theta was seen in mice, no effect of difficulty was observed on performance or this EEG response in mice. This difference likely reflects the ineffectiveness of manipulating trial difficulty based on stimulus durations by trial type in mice—shorter delays make target trials more difficult but makes withholding from nontarget trials easier. Ultimately, more work is required for manipulation of spatial attention and parameterization of difficulty in mice (e.g., a similar backward mask used in humans), although the addition of discriminant auditory tones may be able to address multiple issues. A wealth of prior findings suggests that it is too early to rule out frontal theta as a viable candidate for cross-species translation. Posterior cingulate theta power enhancement has been observed in humans and rats [65], as has a cue-locked dopamine-dependent theta signal [66]. These data, therefore, provide support but require further work.

Limitations and future directions

While the mere concept of comparing cross-species brain responses deserves a critical appraisal, there is good reason to theorize that some electrophysiological activities remain preserved across species. Although classic EEG frequencies are non-specifically
related to cognitive constructs and are likely to simply reflect the intrinsic computations of the generative cortex, event-related local field oscillations are closely linked to any neural mechanism that implements neural computations [67–70]. There is a marked preservation of temporal activity across vertebrate brains, likely due to architectural adjustments that evolved to prioritize retention of temporal coding schemes [71]. Increasing evidence also confirms neurodevelopmental CNS synchronization in EEG responses between humans and rodents, as well as the consistent impact of alcohol and auditory stimuli on these event-related oscillations [65, 72–74]. These theoretical justifications and empirical outcomes are compelling, and they dovetail with the potential for assessing electrophysiology in each species.

Statistical effects reported here were modest. As noted earlier, modulation of these exact t-ROIs will be tested in future studies as a continuation of the novel UH funding mechanism via an overall “learn-confirm” design strategy. This report serves to convey a crystallized set of parameters that will be used in future tests of pharmacologic modulation. With additional experiments and increased sample sizes in mice (comparable to that of humans), the degree of test-retest reliability will be established and further consistencies may be revealed across species. We included both males and females of both species and, while sex differences in learning have been reported [75–77], we largely have not seen such sex differences. These future studies will add to our current knowledge.

These data only compared findings from a single electrode in humans with a single dura lead in mice. While this theory-driven reduction of spatial dimensionality is appropriate with our a priori hypotheses and the preliminary goals of this study, it offers only a fraction of assessable EEG activities in each species. Any conclusion of translational similarity is also based on a qualitative assessment of common within-species statistical effects. While this simplicity is beneficial here, future comparative studies could utilize data normalization, computational modeling, and covariance statistics for quantitative assessments of common neural signatures between species. Notably, these data-driven strategies require a large amount of data, and thus they are not likely to be undertaken unless they follow compelling findings from small-scale hypothesis-driven studies, as presented here.

CONCLUSION

The failure of preclinical models based on behavioral measures alone is well-established. If we are to understand the complex neural mechanisms underlying cognitive deficits in psychiatric disorders, novel approaches linked to neural outcomes must be taken. This field is most likely to advance by investigating similar bio-signals between species. The comparison of mouse and human event-related EEG responses is, therefore, an appropriate next step, based not only on the methodological advantages but also the theoretical similarities between potentially preserved neural mechanisms. Here, we present three tasks that are for the first time revealing a common translational event-related EEG responses between humans and mice.

Importantly, the PRBT revealed that arousal-related posterior alpha appears common between species, and it should be easy to assess the generalizability of this effect within a variety of other tasks. From the PLT, we reveal a very compelling similarity within mid-frontal delta-band power. These two successful paradigms—PLT and PRBT—are both currently being assessed with pharmacologic manipulations across species. While the 5C-CPT presented potential consistencies with target-locked theta seen across species, more work is required for parametric confirmation in mice. The candidate biomarkers advanced here will soon be further evaluated as electrophysiological signatures of behavioral dimensions from cross-species paradigms.

CODE AVAILABILITY

All data and Matlab codes are available on Openneuro.org. accession #d003638.

REFERENCES

1. Barch DM, Carter CS. Measurement issues in the use of cognitive neuroscience tasks in drug development for impaired cognition in schizophrenia: a report of the second consensus building conference of the CNTRICS initiative. Schizophr Bull. 2008;34:613–8.
2. Young JW, Light GA. Cross-species neurophysiological biomarkers of attentional dysfunction in Schizophrenia: bridging the translational gap. Neuropsychopharmacology. 2018;43:230–1.
3. Sarter M. Animal cognition: defining the issues. Neurosci Biobehav Rev. 2004;28:645–50.
4. Insel T, Cuthbert B, Garvey M, Heinssen R, Pine DS, Quin K, et al. Research domain criteria (RDoC): toward a new classification framework for research on mental disorders. Am J Psychiatry. 2010;167:748–51.
5. Cuthbert BN, Insel TR. Toward new approaches to psychotic disorders: the NIMH research domain criteria project. Schizophr Bull. 2010;36:1061–2.
6. Bryce CA, Florreso SB. Perturbations in effort-related decision-making driven by acute stress and corticotropin-releasing factor. Neuropsychopharmacology. 2016;41:2147–59.
7. Cocker PJ, Hisking JG, Benoit J, Winstanley CA. Sensitivity to cognitive effort mediates psychostimulant effects on a novel rodent cost/benefit decision-making task. Neuropsychopharmacology. 2012;37:1825–37.
8. Horan WP, Reddy LF, Barch DM, Buchanan RW, Dunayevich E, Gold JM, et al. Effort-based decision-making paradigms for clinical trials in schizophrenia: part 2 - External validity and correlates. Schizophr Bull. 2015;41:1055–65.
9. Reddy LF, Horan WP, Barch DM, Buchanan RW, Dunayevich E, Gold JM, et al. Effort-based decision-making paradigms for clinical trials in schizophrenia: part 1 - psychometric characteristics of 5 paradigms. Schizophr Bull. 2015;41:1045–54.
10. Florreso SB, Ghods-Sharifi S. Amygdala-prefrontal cortical circuitry regulates effort-based decision-making. Cereb Cortex. 2007;17:251–60.
11. Hisking JG, Florreso SB, Winstanley CA. Dopamine antagonism decreases willingness to expend physical, but not cognitive, effort: a comparison of two rodent cost/benefit decision-making tasks. Neuropsychopharmacology. 2015;40:1005–15.
12. Salamone JD, Corea M, Farrar AM, Nunes EJ, Pardo M. Dopamine, behavioral economics, and effort. Front Behav Neurosci. 2009;3:13.
13. Wolf DH, Satterthwaite TD, Kantrowitz JJ, Katchmar N, Vandelak E, Elliott MA, et al. Amotivation in schizophrenia: integrated assessment with behavioral, clinical, and imaging measures. Schizophr Bull. 2014;40:1328–37.
14. Strauss GP, Whearty KM, Monra LF, Sullivan SK, Ossenfort KL, Frost KH. Avolition in schizophrenia is associated with reduced willingness to expend effort for reward on a Progressive Ratio task. Schizophr Res. 2016;170:198–204.
15. Bismark AW, Thomas ML, Tarasenko M, Shiliuk AL, Rackelmann SY, Young JW, et al. Relationship between effortful motivation and neurocognition in schizo- phrenia. Schizophr Res. 2018;193:69–76.
16. Young JW, Markou A. Translational rodent paradigms to investigate neuro-mechanisms underlying behaviors relevant to amotivation and altered reward processing in schizophrenia. Schizophr Bull. 2015;41:1024–34.
17. Young JW, Geyer MA, Halberstadt AL, van Enkhuizen J, Minassian A, Khan A, et al. Convergent neural substrates of inattention in bipolar disorder patients and dopamine transporter-deficient mice using the 5-choice CPT. Bipolar Disord. 2020;22:46–58.
18. Frank MJ, Seeberger LC, O’Reilly RC. By carrot or by stick: cognitive reinforcement learning in parkinsonism. Science (80-). 2004;306:1940–3.
19. Ragland JD, Cohen NJ, Cools R, Frank MJ, Hannula DE, Ranganath C. CNTRICS imaging biomarkers final task selection: Long-term memory and reinforcement learning. Schizophr Bull. 2012;38:62–72.
20. Amital N, Young JW, Higa K, Sharf AF, Geyer MA, Powell SB. Isolation rearing effects on probabilistic learning and cognitive flexibility in rats. Cogn Affect Behav Neurosci. 2014;14:388–406.
21. Arnone DA, Jones LH, Sweeney JA, Ragozzino ME. Differences in BTBR T+/Ifj and C57BL/6j mice on probabilistic reversal learning and stereotypy behaviors. Behav Brain Res. 2012;227:64–72.
22. Bari A, Theobald DE, Caprioli D, Mar AF, Aciód-Micah A, Dalley JW, et al. Serotonin modulates sensitivity to reward and negative feedback in a probabilistic reversal learning task in rats. Neuropsychopharmacology. 2010;35:1290–301.
23. Hyman JM, Holroyd CB, Seamas JK. A novel neural prediction error found in anterior cingulate cortex ensembles. Neuron. 2017;95:447–56.e3.
24. Reddy LF, Waltz JA, Green MF, Wynk JW, Horan WP. Probabilistic reversal learning in schizophrenia: stability of deficits and potential causal mechanisms. Schizophr Bull. 2016;42:942–51.

25. Waltz JA, Frank MJ, Robinson BM, Gold JM. Selective reinforcement learning deficits in schizophrenia support predictions from computational models of striatal-cortical dysfunction. Biol Psychiatry. 2007;62:756–64.

26. Ryu H, Ha RJ, Lee SJ, Ha K, Cho HS. Behavioral and electrophysiological alterations or reinforcement learning in manic and euthymic patients with bipolar disorder. CNS Neurosci Ther. 2017;23:248–56.

27. Bakic J, Pourtois G, Jepma M, Duprat R, De Raedt R, Baeken C. Spared internal but impaired external reward prediction error signals in major depressive disorder during reinforcement learning. Depress Anxiety. 2017;34:89–96.

28. Konstanty Y, Okamoto Y, Ueda K, Okoda K, Okada G, Yoshimura S, et al. Effects of depression on reward-based decision making and variability of action in probabilistic learning. J Behav Ther Exp Psychol. 2012;43:1088–94.

29. Pizzagalli DA, Jahn AL, O'Shea JP. Toward an objective characterization of an anhedonic phenotype: a signal-detection approach. Biol Psychiatry. 2005;57:319–27.

30. Sutton RS, Barto AG. Reinforcement learning: an introduction. Cambridge: MIT Press; 1998.

31. Luck SJ, Ford JM, Sarter M, Lustig C. CNTRICS.

32. Lustig C, Kozak R, Sarter M, Young JW, Robbins TW. CNTRICS.

33. Buzsáki G, Anastassiou CA, Koch C. The origin of extracellular EEG activity. Neuron. 2012;72:942–58.

34. Battle LR, Minassian A, Kuzmin VN, Maier MG, Dagalakis VJ, Myers J, et al. The effect of reduced dopamine D4 receptor expression in the 5-choice continuous performance task: separating response inhibition from premature responding. Behav Brain Res. 2011;222:183–92.

35. Nolan H, Whelan R, Reilly RB. FASTER: fully automated statistical thresholding for EEG artifact rejection. J Neurosci Methods. 2010;192:152–60.

36. Barwick F, Arnett P, Slobounov S. EEG correlates of fatigue during administration of a history of adolescent alcohol exposure. Addict Biol. 2020;25:1–7.

37. Young JW, Powell SB, Scott CN, Zhou X, Geyer MA. The effect of reduced dopamine D4 receptor expression in the 5-choice continuous performance task: separating response inhibition from premature responding. Behav Brain Res. 2011;222:183–92.

38. Buzsáki G, Anastassiou CA, Koch C. The origin of extracellular fields and currents — EEG, ECoG, LFP and spikes. Nat Rev Neurosci. 2012;13:407–20.

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

40. Nolan H, Whelan R, Reilly RB. FASTER: fully automated statistical thresholding for EEG artifact rejection. J Neurosci Methods. 2010;192:152–60.

41. Kardon O, Adam K, Mance I, Churchill NW, Vogel EK, Berman MG. Distinguishing cognitive effort and working memory load using scale-invariance and alpha suppression in EEG. Neuroimage. 2020;211:116622.

42. Pathania A, Leiker AM, Euler M, Miller MW, Lohse KR. Challenge, motivation, and EEG response in mGLuR5 knockout mice. J Neurophysiol. 2020;123:22–32.

43. Sei

44. Barwick F, Arnett P, Slobounov S. EEG correlates of fatigue during administration of a history of adolescent alcohol exposure. Addict Biol. 2020;25:1–7.

45. Moraes H, Deslandes A, Silveira H, Ribeiro P, Cagy M, Piedade R, et al. The effect of adolescent alcohol exposure on the reward system: a signal detection approach. Biol Psychiatry. 2005;57:319–27.

46. Nolan H, Whelan R, Reilly RB. FASTER: fully automated statistical thresholding for EEG artifact rejection. J Neurosci Methods. 2010;192:152–60.

47. Cavanagh JF, Bismark AW, Frank MJ, Allen JB. Multiple dissociations between corticomedial depression and anxiety in humans and rodents. Nat Neurosci. 2013;16:1–10.

48. Parker KL, Chen KH, Kingston JR, Cavanagh JF, Narayanan NS. Medial frontal —4 Hz activity in humans and rodents is attenuated in PD patients and in rodents with cortical dopamine depletion. J Neuropsychol. 2015;114:131–20.

49. Bastos AM, Ussery WM, Adams RA, Mangun GR, Fries P, Friston KJ. Canonical microcircuits for predictive coding. Neuron. 2012;76:695–711.

50. Friston K. A theory of cortical responses. Philos Trans R Soc Lond B Biol Sci. 2005;360:815–36.

51. Siegel M, Donner TH, Engel AK. Spectral fingerprints of large-scale neuronal interactions. Nat Rev Neurosci. 2012;13:21–34.

52. Woemeldorf T, Valiente TA, Sahin NT, Miller KJ, Tiesinga P. Dynamic circuit motifs underlying rhythm gain control, gating and integration. Nat Neurosci. 2014;17:1031–39.

53. Birth C, Perry W, Brigham J, Handler P. Cortical frequency discrimination in mnemonic tasks. J Neurosci. 2010;30:12329–38.

54. Ehlers CL, Wills DN, Desikan A, Phillips E, Havstad J. Decreases in energy and cognitive control in humans and rodents. Nat Neurosci. 2017;20:1–9.

55. Ehlers CL, Phillips E, Wills D, Benedict J, Sanchez-Alavez M. Phase locking of event-related oscillations is decreased in both young adult humans and rats with a history of adolescent alcohol exposure. Addict Biol. 2020;25:1–12.

56. Chen CS, et al. Divergent strategies for learning in males and females. Curr Biol. 2020;30:1–11.

57. Marquardt K, Sigdel R, Caldwell K, Brimijoin SL. Prenatal ethanol exposure impairs executive function in mice into adulthood. Alcohol Clin Exp Res. 2014;38:2962–6.

58. Cavanagh JF, Frank MJ, Klein TJ, Allen JB. Frontal theta links prediction errors to encoding activity in the 5C-CPT irrespective of concurrent low–4 Hz activity in humans and rodents. Nat Neurosci. 2013;16:1–10.

59. Cavanagh JF, Frank MJ, Laubach M. Common medial frontal mechanisms of adaptive control in humans and rodents. Nat Neurosci. 2013;16:1–10.

60. Ehlers CL, Wills DN, Desikan A, Phillips E, Havstad J. Decreases in energy and cognitive control in humans and rodents. Nat Neurosci. 2017;20:1–9.

61. Ehlers CL, Phillips E, Wills D, Benedict J, Sanchez-Alavez M. Phase locking of event-related oscillations is decreased in both young adult humans and rats with a history of adolescent alcohol exposure. Addict Biol. 2020;25:1–12.

62. Chen CS, et al. Divergent strategies for learning in males and females. Curr Biol. 2020;30:1–11.

63. Marquardt K, Sigdel R, Caldwell K, Brimijoin SL. Prenatal ethanol exposure impairs executive function in mice into adulthood. Alcohol Clin Exp Res. 2014;38:2962–6.

64. Cavanagh JF, Frank MJ, Klein TJ, Allen JB. Frontal theta links prediction errors to encoding activity in the 5C-CPT irrespective of concurrent low–4 Hz activity in humans and rodents. Nat Neurosci. 2013;16:1–10.

65. Ehlers CL, Wills DN, Desikan A, Phillips E, Havstad J. Decreases in energy and neural and behavioral correlates of self-control during practice. Front Hum Neurosci. 2012;6:229.

66. Cavanagh JF, Frank MJ. Frontal theta as a mechanism for cognitive control. Trends Cogn Sci. 2014;18:1–8.

67. Cohen MX, Cavanagh JF. Single-trial regression elucidates the role of prefrontal theta oscillations in response success. Front Psychol. 2011;2:30.

68. Cavanagh JF, Zambrano-Vazquez L, Allen JB. Theta lingua franca: a common mid-frontal substrate for action monitoring processes. Psychophysiology. 2012;49:220–38.

69. Carter RJ, Lione LA, Zhou X, Geyer MA, et al. Characterization of progressive motor deficits in mice transgenic for the human Huntington’s disease mutation. J Neuropsychol. 1999;19:3248–57.

70. Womelsdorf T, Valiante TA, Wallace T, Hutcheson DM. Consideration of species differences in developing novel molecules as cognition enhancers. Neurosci Biobehav Rev. 2013;37:2181–93.

71. Zeeb FD, Robbins TW, Winstanley CA. Serotonicergic and dopaminergic modulation of gambling behavior as assessed using a novel rat gambling task. Neuropharmacology. 2009;57:2239–49.
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J.F.C.: Conceptualization, Methodology, Software, Formal analysis, Writing—Original Draft, Funding acquisition. D.G.: Investigation. G.A.L.: Conceptualization, Methodology, Resources, Writing—Review & Editing, Funding acquisition. S.O.: Investigation. R.F.S.: Investigation, Data curation, Supervision, Project Administration. A.W.B.: Investigation, Data curation, Supervision, Project Administration. S.G.B.: Conceptualization, Methodology, Writing—Review & Editing, Funding acquisition. N.R.S.: Conceptualization, Methodology, Writing—Review & Editing, Investigation, Data curation, Supervision, Project Administration, Funding acquisition. J.L.B.: Conceptualization, Methodology, Writing—Review & Editing, Supervision, Project Administration, Funding acquisition. J.W.Y.: Conceptualization, Methodology, Writing—Original Draft, Funding acquisition.

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