Spontaneous Eye Blink Rate during the Working Memory Delay Period Predicts Task Performance

Jefferson Ortega[1], Chelsea Reichert Plaska[1,2], Bernard A. Gomes[3], Timothy M. Ellmore[1,2*]

[1]Department of Psychology, The City College of the City University of New York, New York, USA

[2]Behavioral and Cognitive Neuroscience Program, The Graduate Center of the City University of New York, New York, USA

[3]Department of Neurosurgery, Cedars-Sinai Medical Center, Los Angeles, California, USA

*Corresponding Author: Timothy M. Ellmore, Ph.D. tellmore@ccny.cuny.edu

Abstract

Spontaneous eye blink rate (sEBR) has been found to be a non-invasive indirect measure of striatal dopamine activity. Dopamine (DA) neurons project to the prefrontal cortex (PFC) through the mesocortical dopamine pathway and their activity is implicated in a range of cognitive functions, including attention and working memory (WM). The goal of the present study was to understand how fluctuations in sEBR during different phases of a working memory task relate to task performance. Across two experiments, with recordings of sEBR inside and outside of a magnetic resonance imaging bore, we observed sEBR to be positively correlated with WM performance during the WM delay period. Additionally we investigated the non-linear relationship between sEBR and WM performance, and modeled a proposed "Inverted-U-shape" relationship between DA and WM performance. We also investigated blink duration, which is proposed to be related to sustained attention, and found blink duration to be significantly shorter during the encoding and probe periods of the task. Taken together, these results provide support towards sEBR as an important correlate of working memory task performance. The relationship of sEBR to DA activity and the influence of DA on the PFC during WM maintenance is discussed.

Keywords: Spontaneous eye blink rate, working memory, dopamine, attention, blink duration

Introduction

The healthy human blinks around 15-20 times per minute[1], however the precorneal tear film, which lubricates the eye, only begins drying up approximately 25s after a blink ends[2]. This suggests that we blink more often than needed to maintain a lubricated precorneal tear film. Previous research has found task-related modulation of spontaneous eye blink rate (sEBR), which indicates that blinking could be reflective of cognitive factors[3,4]. For example, reading is accompanied by low levels of sEBR, while high levels of sEBR have been reported during conversation[5]. More recent studies have found sEBR can measure attentional load and fatigue[6], attentional control[7,8], track working memory updating and gating[9], and predict differences in exploration during reinforcement learning[10]. A growing body of literature continues to provide evidence supporting sEBR as an effective measure of striatal dopamine (DA) activity[11]. However, whether sEBR does indeed reflect DA activity is still in debate today[12,13].

The connection between sEBR and DA first came from observations of neurological and psychiatric disorders which found decreased sEBR in patients with Parkinson's[14,15], a neurodegenerative disorder that affects the dopaminergic system in the brain, causing symptoms like rigidity[16]. Schizophrenia has also been suggested to provide evidence towards the connection between sEBR and DA due to excessive DA activity in the striatum[17] and increased sEBR in schizophrenia patients[18,19]. Additionally, sEBR and DA has previously been investigated in pharmacological studies, which have observed an increase in sEBR after administration of DA agonists, while DA antagonists decreased sEBR in primates[20,21]. In one study, researchers found sEBR was correlated with dopamine levels specifically in the caudate nucleus in monkeys suggesting that DA could regulate blink rate[22]. This is further supported by another study that found sEBR to be closely related to *in vivo* and positron emission tomography

(PET) measures of striatal D2 receptor density in the ventral striatum and caudate nucleus of adult male vervet monkeys[23]. These findings provide valuable evidence towards sEBR being a viable measure of striatal DA activity and has led to many researchers to adopt sEBR as a measure of DA activity. Moreover, sEBR is an easy-to-record physiological measure that is non-invasive and affordable.

One cognitive process of interest that is closely related to DA activity is working memory (WM), which is the process of actively holding information online and manipulating it to meet task demands[24]. Human studies investigating DA in WM tasks have found both caudate dopamine activity during WM maintenance and DA synthesis capacity to be positively correlated with WM capacity[25,26]. Additionally, many studies have found supporting evidence for the importance of DA in WM and have specifically pointed to the involvement of the prefrontal cortex (PFC) in WM function[27-31], especially during WM maintenance[30-32]. Though it is widely accepted that the PFC plays an important role in WM function[33], many researchers still debate PFC's role in WM[34]. One model that attempts to elucidate the PFC's role in WM function is the prefrontal cortex basal ganglia WM model (PBWM)[35,36], a computational neural network model that suggests that WM requires robust maintenance and rapid selective updating. The PBWM states that the frontal cortex facilitates robust, active maintenance through recurrent excitation in frontal neurons, while the basal ganglia orchestrates a gating mechanism that controls the flow of information into WM[35]. Previous research has pointed towards DA being important for this sustained firing activity in the PFC during WM maintenance[37-39]. These studies have found that blocking dopaminergic inputs and D1 receptors in the PFC leads to worse performance on WM tasks suggesting that blocking dopaminergic inputs, and specifically D1 receptors, in the PFC possibly disrupts the active maintenance of information[40]. The relationship between DA and WM performance is believed to follow an inverted-U-shape, where too little or too much dopamine impairs performance, as seen in psychopharmacological studies[41]. In one study, the effects of administered dopaminergic drugs on PFC function depended on baseline levels of performance, whereas administration of bromocriptine, a dopamine agonist, impaired performance for individuals with higher working memory abilities while improving performance for individuals with lower working memory abilities[42].

Although eye blink rate has been used to investigate the basal ganglia's role in working memory updating and gating[9], few studies have investigated the relationship between sEBR and WM performance. Additionally, while the effects of sEBR on WM function have been investigated in previous studies[9,43-45], the impact of sEBR during the different phases of a WM task and how it relates to WM performance remains unclear. Modulation of sEBR across these different phases, such as encoding, maintenance, and retrieval are developed in infants as young as 10 months, indicating that sEBR can reflect dopamine function in early human development[45]. While no direct relationship between performance and sEBR was observed in this study, there was a relationship between the variability of sEBR and WM performance in which infants who had a greater change in sEBR across the task phases had better performance. This finding suggests that the variability of sEBR could reflect transient changes in DA which is important for creating or maintaining representations[45]. A related physiological measure that can provide insight into cognitive function is blink duration. Blink duration is longer when individuals solve word problems with insight rather than analytically[46]. Moreover, previous studies have found blink duration to decrease as visual workload increases, suggesting that shorter blinks could relate to prolonged sustained attention[47,48].

The goal of the current study was to investigate how fluctuations in sEBR during different phases of a WM task relate to performance. We hypothesized that sEBR would be positively correlated with WM performance during WM maintenance and that it would also have a non-linear relationship such that low and high sEBR would lead to worse performance. We also investigated blink duration during the different phases of the task. Since shorter blinks are believed to reflect sustained attention and visual workload[47,48], we hypothesized that blink duration would be fastest during the encoding period when participants are being shown images that they will have to remember.

Results

Experiment 1

In Experiment 1, we examined sEBR with task performance while the delay period interval was varied. First, because of the established non-linear relationship between DA and WM task performance[49], Spearman's rho correlation coefficient (r_s) was used to analyze the relationship between sEBR and task

performance. sEBR during the delay period of the task correlated positively with task performance (r_s = .617, p = .0019). There was no significant relationship between sEBR during the other phases of the task and task performance (Figure 2) There was also no significant relationship between sEBR and hours slept the night before nor between sEBR and reaction time (RT) across all periods of the task (Table 1). Second, we used a paired samples t-test to compare participants' blink duration during the encoding period to blink duration during the delay, probe, and post-probe scrambled stimulus periods. There was a significant difference between participants blink duration during the encoding period (mean blink duration = 192.14 ms, SD = 31.42) and delay period (mean blink duration = 217.86 ms, SD = 34.74), t(13) = -4.643, p < .001) (Figure 3). The difference between probe (mean blink duration = 199.64 ms, SD = 26.41) and delay blink duration was also significant, t(13) = 2.190, p = 0.047. There was also a significant difference between participants blink duration during the encoding period and scrambled period (mean blink duration = 224.86 ms, SD = 33.22), t(13) = -4.842, p < .001) and between probe and scrambled period, t(13) = -3.086, p = 0.009 (Figure 3). There was no significant difference between blink duration in the encoding period and blink duration in the probe period, t(13) = -1.304, p = .215) (Figure 3).

Experiment 2

In Experiment 2, the task design was identical to Experiment 1. First, we examined the relationship between sEBR during each WM task phase and performance. sEBR during the delay period of the task was correlated positively with task performance (r_s = .446, p = .006), with no significant relationship observed between sEBR in the other periods of the task and task performance (Figure 4). We then looked at the relationship between sEBR during different delay lengths (i.e., the entire 2, 5, or 9 sec delay interval) and task performance. We found sEBR during the five second delay period to have a significant positive correlation with task accuracy (r_s = .334, p = .047) (Figure 5). The relationship between sEBR and task accuracy during the nine second delay was not significant (r_s = .271, p = .110) (Figure 5). Finally, in order to investigate if sEBR can track DAs nonlinear relationship, we then computed a quadratic polynomial regression model on sEBR during the different delay period lengths of

the task and performance. There was no significant polynomial regression equation found between task accuracy and the two second delay ($\beta = -2.407$, $p = 0.238$), task accuracy and the five second delay ($\beta = -1.595$, $p = 0.119$), nor task accuracy and the nine second delay ($\beta = -1.838$, $p = 0.186$) (Figure 5). There was no significant relationship between sEBR and hours slept the night before nor between sEBR and RT across all periods of the task (Table 2).

Discussion

In the present study, we investigated the temporal fluctuations in sEBR across a WM paradigm and its relation to WM performance in two experiments, inside and outside an MRI scanner. Using the same Sternberg working memory paradigm, we observed a strong positive relationship between sEBR and task performance only during the WM task delay in both experiments. We also modelled a proposed "inverted-U-shape" relationship between DA and WM task performance, considering sEBR as an indirect measure of DA[11,49]. Additionally, in Experiment 1, we observed faster blink durations during the task encoding and probe period compared to the delay and post-probe scrambled periods, which is believed to reflect sustained attention during stimulus encoding[48]. Overall, our findings suggest that sEBR during the WM delay period may reflect active maintenance of information, which may involve sustained firing activity in the PFC and that shorter blink duration can reflect sustained attention during a WM task's encoding period.

The density of D2 receptors in the striatum and the frontal cortex is 1:11, respectively[50]. It is believed that resting sEBR more strongly reflects D2 dopamine receptor (D2) activity than D1 dopamine receptor[23] (D1) activity, mostly because of the increased sensitivity of D2 receptors to DA than D1[51] receptors in the striatum. However, D1 receptors are more abundant in the PFC than D2 receptors[52], suggesting the possibility that sEBR may be able to reflect D1 receptor activity in the PFC. In this study, we found that increased sEBR during the delay period of a Sternberg scene WM task was positively correlated with WM performance. Additionally, the positive relationship between sEBR and WM performance was only significantly related during the delay period of the task when participants were maintaining information in WM, also when the PFC is likely to be recruited[25,53,54]. Therefore, our results

suggest that sEBR may reflect D1 receptor activity in the PFC. Moreover, our findings provide support towards previous research that has found D1 receptor activity in the PFC to correlate with WM performance[55,56]. Additionally, these results provide evidence in support of computational models that state that D1 receptor activity modulates sustained activity of PFC neurons during working memory tasks[54]. Interestingly, the positive relationship between sEBR and task performance was present in both the short and medium interval delay periods, but not the long delay period (Figure 5). This could be due to a lack of statistical power, as the long delay had the least number of trials included in the analysis. This is because the longer delays were shortened and combined with the shorter delays for the analysis (see Methods for details). Thus, the long delay interval had a maximum of 54 trials per participants while the medium and short delay interval had a maximum of 108 and 162 trials per participant, respectively.

The PFC is implicated in working memory[27-31] and is believed to maintain representation of task-relevant information by sustained neuronal firing during the delay period[35,36]. However, recent findings counter that idea and instead suggest that delay period activity in the PFC could actually be a top-down signal that influences other areas, such as the posterior sensory areas, which is believed to be the site where actual memory representations are being maintained[57]. Additionally, researchers have found that delay period activity in the PFC does not reflect the encoding of stimulus information[58,59]. Although PFC is active during WM maintenance, it may not actually hold information about a stimulus. Instead, it is proposed that the PFC and posterior areas share information through coupling of oscillatory activity in both areas[60-62]. Moreover, oscillatory activity in the frontal and posterior areas has been observed to increase during the delay period of WM tasks[63]. It is possible that sEBR was correlated with higher performance not due to D1 receptor modulation of sustained activity in PFC neurons, but instead due to long-range coupling of oscillatory activity between the PFC and posterior sensory areas[57].

Dopamine levels are believed to follow an inverted U-shape during working memory tasks[49]. Our results suggest an inverted U-shape relationship between sEBR during the delay period and WM performance, but the significance of the fit was marginal. We believe that failure to achieve non-linear model significance was due to lack of extreme (sub- and supraoptimal) sEBRs observed in the pool of

participants. Previous research has found both low and high levels of D1 receptor activity in the PFC to decrease performance in working memory tasks[64,65]. Thus, it is possible that if participants had sub- and supraoptimal sEBRs, then our polynomial regression model may have been significant. Future research should investigate sEBR with healthy subjects and with subjects that have been observed to have extreme sEBR in order to have a wider variety of sEBRs and to better understand its connection with DA and the PFC. Additionally, other methods of DA measures could be used to investigate DA during the delay period such as correlations with neuromelanin-sensitive MRI which can detect neuromelanin, a product of dopamine metabolism[66].

Finally, we observed shorter blink durations during the encoding period of the task compared to the delay and scrambled period, but not the probe period. In previous studies, blink duration has been found to decrease with higher visual workloads[47,67,68], indicating that shorter blink durations could be associated with an increased demand on attention. Due to visual working memory's limited capacity, which studies have suggested is limited to maintaining 3-4 items simultaneously[69,70], goal-directed processing must control which items occupy this limited space. Attention is believed to act as this "gatekeeper" for working memory; it selects the items that are most relevant for the current goals[71]. Therefore, shorter blink duration during the encoding period of the task could be reflecting this "gatekeeping" role of attention, which may be less important during the delay and scrambled periods of the task. Additionally, we observed no difference between blink duration during the encoding period and the probe period, which could suggest that attention was also engaged similarly during the probe as selective attention has been found to facilitate retrieval functions in WM tasks[72,73].

To conclude, we investigated temporal changes of sEBR during different phases of a WM task and its relation to WM performance. We observed a significant positive correlation between sEBR and WM task performance during the delay period, but not during the other phases of the task. Additionally, we found evidence for an association of blink duration during both stimulus encoding and WM probe

retrieval, potential reflecting attention modulation. To the best of our knowledge, this is the first study to investigate sEBR during different phases of a WM task using complex visual scenes. Future studies should continue to investigate sEBRs connection to measures of cortical (especially PFC) and subcortical dopamine and assess linear and non-linear relationships to task performance in healthy and diseased populations (e.g., schizophrenia and Parkison's disease).

Methods

Participants

Participants selected for Experiment 1 were part of a larger study. Nineteen healthy participants (8 males; M = 23.79; S.D. = 7.72) were recruited for Experiment 1. In Experiment 1, sEBR was measured inside a 3 tesla Siemens Prisma MRI scanner. In Experiment 2, sEBR was recorded in a sound attenuated EEG booth during acquisition of EEG data while participants sat upright. Fifty-three healthy participants (29 males; M = 23.58; S.D. = 5.79) were recruited for Experiment 2. All participants were recruited via flyers, which were posted throughout [1]the City College of New York campus and all participants had normal or corrected-to-normal vision with no reported neurological or psychiatric disorders. Four participants were removed from Experiment 1 due to noisy data collection, and 17 participants were removed from Experiment 2 due to multiple reasons (11 participants were removed due to bad EOG channel quality, 4 participants were removed due to stimulus marker malfunction, and 2 participants were removed for failing to adhere to protocol). Participants were either compensated $15 or received one psychology course credit per hour of participation in the study. Informed consent was obtained from all participants in the study. The protocol was approved by the Institutional Review Board of the City College of New York. The final sample for the analysis in Experiment 1 was fifteen subjects, and for Experiment 2 was thirty-six subjects.

Task and Procedure

Participants completed three runs, each run containing 54 trials, of a modified version of the Sternberg WM task[74]. Naturalistic scenes were used as stimuli and were sampled from the SUN database[75]. The task consisted of an encoding period, delay period, probe period, and post-probe

scrambled stimulus period (which served as a visual baseline and to signal end of trial). During the encoding period, participants were shown three subsequent novel scenes for 1400 ms each. During the delay period, a black fixation cross was shown on a grey background for varied lengths (either 2, 5, or 9 seconds long). Each run of the task had 18 trials of each delay duration with order of presentation randomized. The probe was presented for 1400 ms after the delay period and consisted of a new image (one that has not been presented yet) or an old image (one that was shown during encoding). The chance of receiving a new probe was 50%. Participants indicated whether the image presented was either a new or an old image with a button press. Then, after the probe, a Fourier phase-scrambled scene was shown for 1000 ms, indicating the end of the current trial followed by a period of blank screen jitter. Participants also completed the St. Mary's Hospital sleep questionnaire prior to the start of the experiment[76], a self-reported assessed of the duration and quality of sleep in the previous 24-hour period.

sEBR and Blink Duration Recording

Participants were not given instructions about when to blink during the experiment. Previous studies have found blink rate to be stable between 10 am and 5 pm[77,78]. For both Experiment 1 and Experiment 2, sEBR was recorded between 10:00 am and 3:00 pm. During Experiment 1, sEBR was recorded inside a 3 tesla Siemens Prisma MRI scanner using an MRI compatible EyeLink 1000 Eye Tracker (SR Research) and was recorded at 500 Hz. In Experiment 2, eye blinks were recorded using an electrooculogram (EOG) that was recorded during 64-channel scalp electroencephalogram (EEG) using a Brain Products cap. EOGs were placed above the left eye and below the right eye to track blinking. Blink detection was performed using MNE Python via the function "*find_eog_events*"[79]. In Experiment 1 and 2, sEBR was computed as the average number of blinks per task period. The first two seconds of all delay periods were used in the analysis of the short delay interval. Additionally, the first five seconds of the nine second delay was used, in addition the 5 second delay period, in the analysis of the medium delay interval. Finally, blink duration was measured using an MRI compatible EyeLink 1000 Eye Tracker (SR Research) only in Experiment 1. Since blinks are typically 100-400 ms in duration[80], blink durations that were shorter or longer than this were removed from the analysis.

Statistical analysis

The results reported in this study were computed using JASP (Version 0.14.1). We investigated the relationship between sEBR and various other variables (performance and hours slept) using either Spearman's rho (strictly for sEBR and performance) or Pearson's r correlation (used for comparison of reaction time and hours slept with sEBR). Spearman's rho was used to measure the relationship between sEBR and WM performance due to the established idea that DA follows an "inverted U-shape" with WM performance. We hypothesized that the relationship between sEBR and WM performance would represent a monotonic function, and thus used Spearman's rho to be the more appropriate measure. For all other correlations, we were only interested in investigating their linear relationships. To assess how blink duration changed across the different phases of the study, we used a paired sample t-test on blink duration during the different phases of the task. Finally, we investigated the non-linear relationship between sEBR during the delay period of the task and performance by computing a polynomial regression model (2nd order).

Figures and Tables

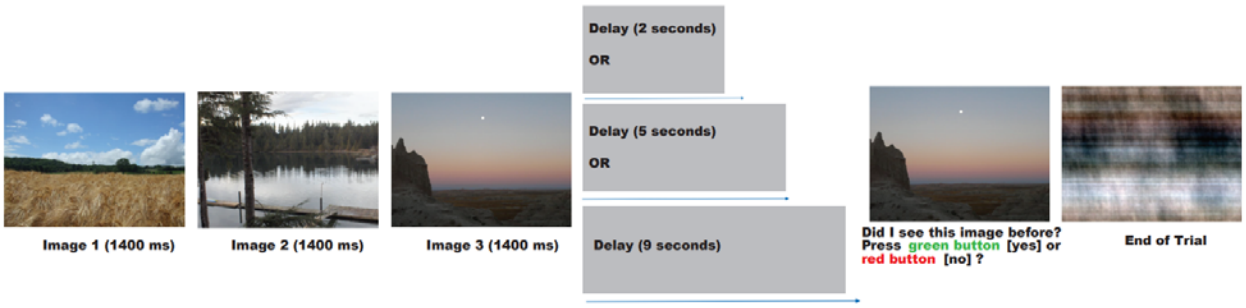


Figure 1. Task design. Each trial began with an encoding period where three novel complex scenes were presented for 1400 ms each. The encoding period was followed by a delay period where a fixation cross was presented on a grey background for a varied amount of time (2 s, 5s, or 9 s). After the delay period, the probe was presented for 1400 ms and participants had to identify whether the image was a new image or one of the previously presented images with a button press. After the probe, a scrambled image was presented which indicated the end of the trial.

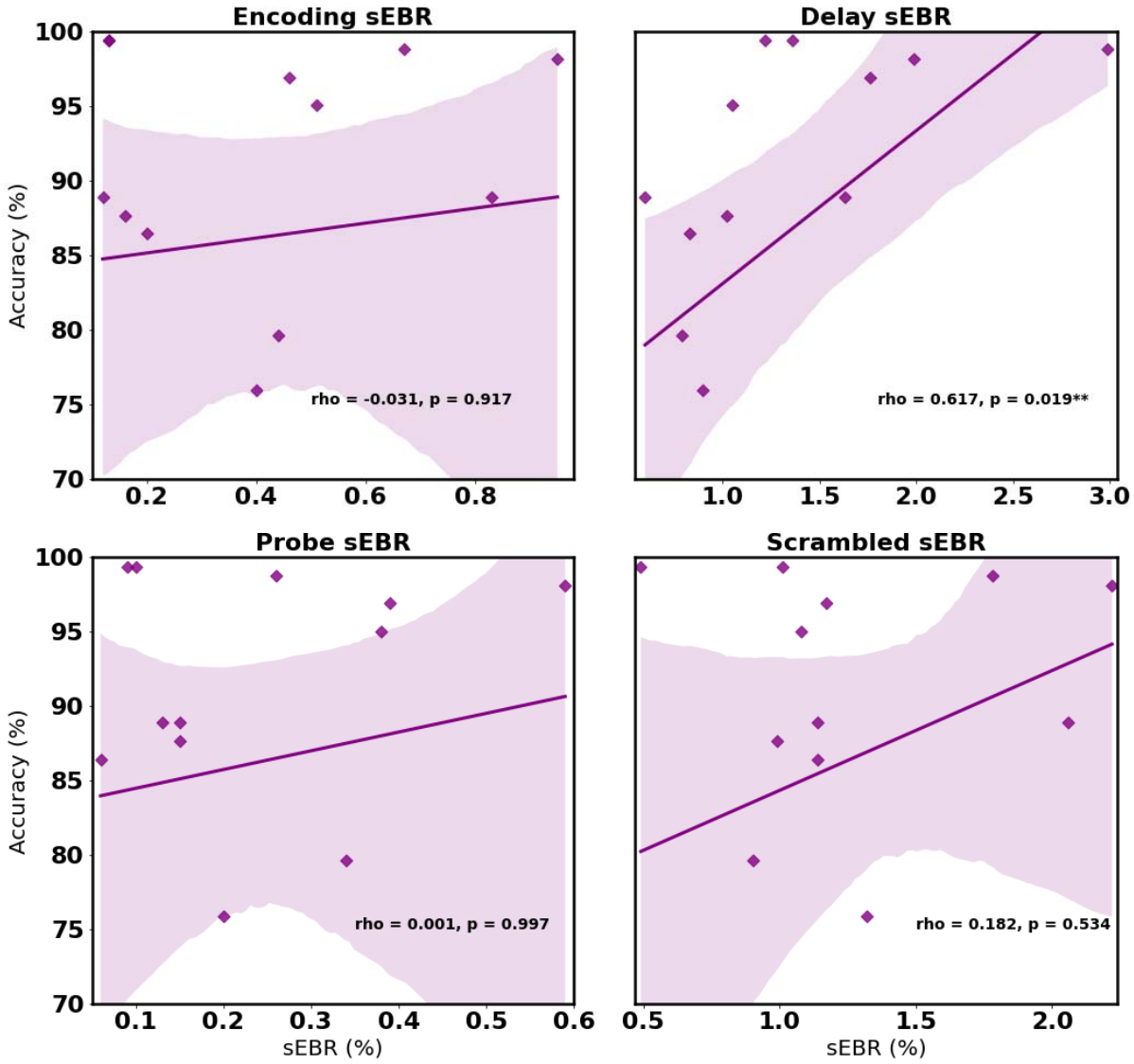


Figure 2. Correlation between sEBR and task accuracy in Experiment 1. Delay period sEBR was significantly positively correlated with task accuracy (spearman's rho = 0.617, p = 0.0019) compared to the other parts of the task. Fitted line represents linear regression model fit. Shaded region depicts 95% confidence interval. * p < .05, ** p < .01, *** p < .001.

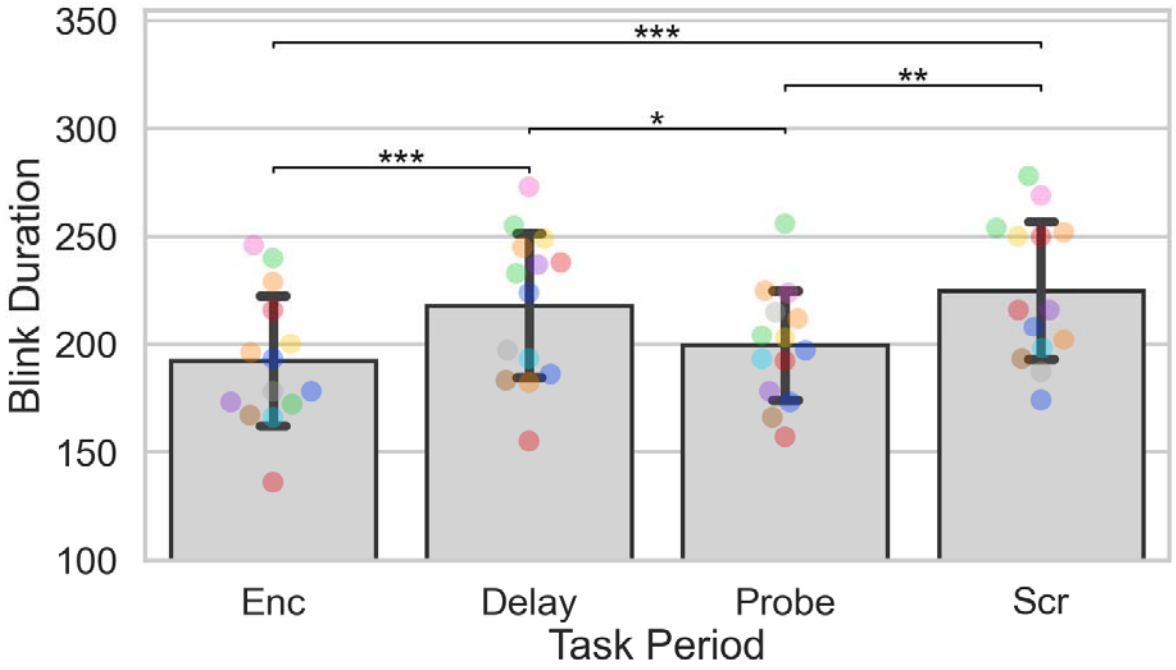


Figure 3. Paired samples t-test of blink duration across task periods. Encoding and probe period blink duration was significantly lower than scrambled (Scr) and delay period blink duration. Each individual color represents a single participant. Note: some colors may be presented twice in one bar due to lack of primary colors available to display. * $p < .05$, ** $p < .01$, *** $p < .001$.

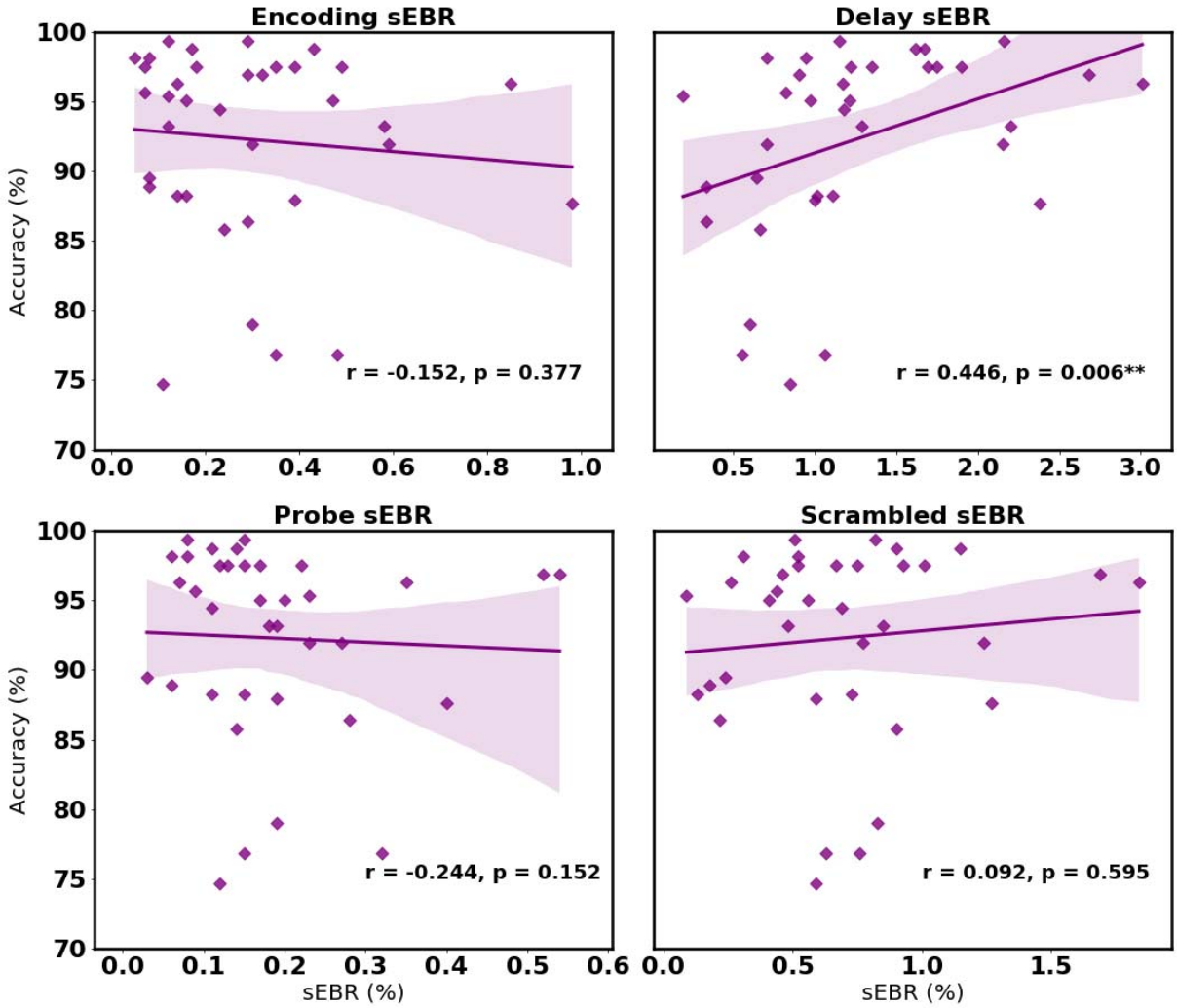


Figure 4. Correlation between sEBR and task accuracy in Experiment 2. Delay period sEBR was significantly positively correlated with task accuracy (spearman's rho =0.446, p = 0.006) compared to the other parts of the task. Fitted line represents linear regression model fit. Shaded region depicts 95% confidence interval. * p < .05, ** p < .01, *** p < .001.

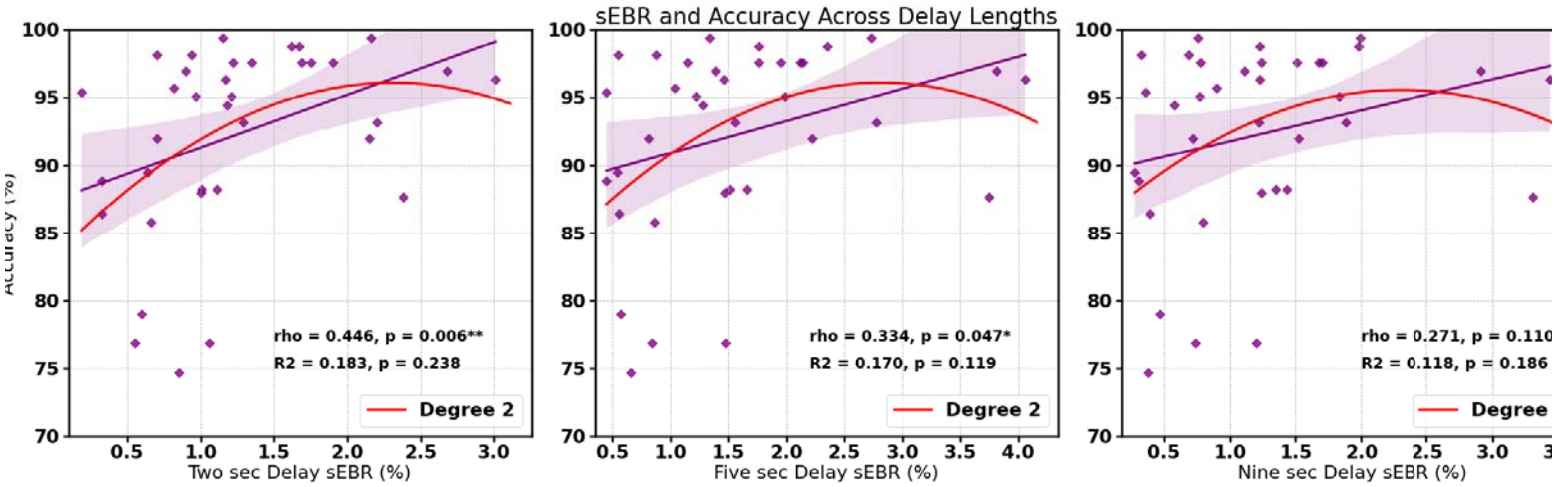


Figure 5. Correlation and polynomial regression model between task accuracy and sEBR across different delay lengths. The relationship between delay period sEBR and task performance was significantly positively correlated during the short and medium delay, but not the long delay. Fitted red line represents polynomial regression model fit. The non-linear relationship between sEBR and WM performance reveals an inverted "U" shape, however the model is not significant. Shaded region depicts 95% confidence interval. * p < .05, ** p < .01, *** p < .001.

Pearson's Correlations

Variable		Reaction Time	Hours Slept
1. Enc sEBR	Pearson's r	-0.148	-0.251
	p-value	0.613	0.386
4. Delay sEBR (2s)	Pearson's r	-0.421	0.099
	p-value	0.133	0.735
2. Probe sEBR	Pearson's r	0.082	-0.096
	p-value	0.780	0.743
3. Scr sEBR	Pearson's r	-0.185	0.032
	p-value	0.526	0.913

* p < .05, ** p < .01, *** p < .001

Table 1. Correlations between sEBR and reaction time and sEBR and hours slept in Experiment 1. sEBR was not correlated with neither reaction time nor hours slept the night before. * p < .05, ** p < .01, *** p < .001.

Pearson's Correlations

Variable		Reaction Time	Hours Slept
1. Enc sEBR	Pearson's r	0.075	-0.112
	p-value	0.664	0.517
4. Delay sEBR (2s)	Pearson's r	-0.030	0.008
	p-value	0.861	0.965
2. Probe sEBR	Pearson's r	0.016	-0.007
	p-value	0.927	0.969
3. Scr sEBR	Pearson's r	0.238	-0.092
	p-value	0.163	0.593

* p < .05, ** p < .01, *** p < .001

Table 2. Correlations between sEBR and reaction time and sEBR and hours slept in Experiment 2. sEBR was not correlated with neither reaction time nor hours slept the night before. * p < .05, ** p < .01, *** p < .001.

Acknowledgments

Research reported in this publication was supported by the National Institute of Mental Health of the National Institutes of Health under Award Number R56MH116007 (T.M.E.).

Author contributions

T.M.E designed the study. J.O, C.R, and B.G performed the research. J.O analyzed and interpreted the data, prepared the figures, and wrote the final manuscript. C.R, B.G, and T.M.E edited and reviewed the manuscript.

Competing interests

The authors declare no competing interests.

References

1 Tsubota, K. *et al.* Quantitative videographic analysis of blinking in normal subjects and patients with dry eye. *Arch Ophthalmol* **114**, 715-720, doi:10.1001/archopht.1996.01100130707012 (1996).

2 Norn, M. J. A. o. Desiccation of the precorneal film: I. Corneal wetting time. **47**, 865-880 (1969).

3 Oh, J., Jeong, S. Y. & Jeong, J. The timing and temporal patterns of eye blinking are dynamically modulated by attention. *Hum Mov Sci* **31**, 1353-1365, doi:10.1016/j.humov.2012.06.003 (2012).

4 Siegle, G. J., Ichikawa, N. & Steinhauer, S. Blink before and after you think: Blinks occur prior to and following cognitive load indexed by pupillary responses. *Psychophysiology* **45**, 679-687, doi:10.1111/j.1469-8986.2008.00681.x (2008).

5 Bentivoglio, A. R. *et al.* Analysis of blink rate patterns in normal subjects. *Mov Disord* **12**, 1028-1034, doi:10.1002/mds.870120629 (1997).

6 Maffei, A. & Angrilli, A. Spontaneous eye blink rate: An index of dopaminergic component of sustained attention and fatigue. *Int J Psychophysiol* **123**, 58-63, doi:10.1016/j.ijpsycho.2017.11.009 (2018).

7 Colzato, L. S., van den Wildenberg, W. P., van Wouwe, N. C., Pannebakker, M. M. & Hommel, B. Dopamine and inhibitory action control: evidence from spontaneous eye blink rates. *Exp Brain Res* **196**, 467-474, doi:10.1007/s00221-009-1862-x (2009).

8 Unsworth, N., Miller, A. L. & Robison, M. K. Individual differences in encoding strategies and free recall dynamics. *Q J Exp Psychol (Hove)* **72**, 2495-2508, doi:10.1177/1747021819847441 (2019).

9 Rac-Lubashevsky, R., Slagter, H. A. & Kessler, Y. Tracking Real-Time Changes in Working Memory Updating and Gating with the Event-Based Eye-Blink Rate. *Sci Rep* **7**, 2547, doi:10.1038/s41598-017-02942-3 (2017).

10 Van Slooten, J. C., Jahfari, S. & Theeuwes, J. Spontaneous eye blink rate predicts individual differences in exploration and exploitation during reinforcement learning. *Sci Rep* **9**, 17436, doi:10.1038/s41598-019-53805-y (2019).

11 Jongkees, B. J. & Colzato, L. S. Spontaneous eye blink rate as predictor of dopamine-related cognitive function-A review. *Neurosci Biobehav Rev* **71**, 58-82, doi:10.1016/j.neubiorev.2016.08.020 (2016).

12 Dang, L. C. *et al.* Spontaneous Eye Blink Rate (EBR) Is Uncorrelated with Dopamine D2 Receptor Availability and Unmodulated by Dopamine Agonism in Healthy Adults. *eNeuro* **4**, doi:10.1523/ENEURO.0211-17.2017 (2017).

13 Sescousse, G. *et al.* Spontaneous eye blink rate and dopamine synthesis capacity: preliminary evidence for an absence of positive correlation. *Eur J Neurosci* **47**, 1081-1086, doi:10.1111/ejn.13895 (2018).

14 Hall, A. The Origin and Purposes of Blinking. *Br J Ophthalmol* **29**, 445-467, doi:10.1136/bjo.29.9.445 (1945).

15 Reddy, V. C., Patel, S. V., Hodge, D. O. & Leavitt, J. A. Corneal sensitivity, blink rate, and corneal nerve density in progressive supranuclear palsy and Parkinson disease. *Cornea* **32**, 631-635, doi:10.1097/ICO.0b013e3182574ade (2013).

16 Dauer, W. & Przedborski, S. Parkinson's disease: mechanisms and models. *Neuron* **39**, 889-909, doi:10.1016/s0896-6273(03)00568-3 (2003).

17 Howes, O., McCutcheon, R. & Stone, J. Glutamate and dopamine in schizophrenia: an update for the 21st century. *J Psychopharmacol* **29**, 97-115, doi:10.1177/0269881114563634 (2015).

18 Swarztrauber, K. & Fujikawa, D. G. An electroencephalographic study comparing maximum blink rates in schizophrenic and nonschizophrenic psychiatric patients and nonpsychiatric control subjects. *Biol Psychiatry* **43**, 282-287, doi:10.1016/S0006-3223(97)00028-0 (1998).
19 Adamson, T. A. Changes in blink rates of Nigerian schizophrenics treated with chlorpromazine. *West Afr J Med* **14**, 194-197 (1995).
20 Elsworth, J. D. *et al.* D1 and D2 dopamine receptors independently regulate spontaneous blink rate in the vervet monkey. *J Pharmacol Exp Ther* **259**, 595-600 (1991).
21 Jutkiewicz, E. M. & Bergman, J. Effects of dopamine D1 ligands on eye blinking in monkeys: efficacy, antagonism, and D1/D2 interactions. *J Pharmacol Exp Ther* **311**, 1008-1015, doi:10.1124/jpet.104.071092 (2004).
22 Taylor, J. *et al.* Spontaneous blink rates correlate with dopamine levels in the caudate nucleus of MPTP-treated monkeys. **158**, 214-220 (1999).
23 Groman, S. M. *et al.* In the blink of an eye: relating positive-feedback sensitivity to striatal dopamine D2-like receptors through blink rate. *J Neurosci* **34**, 14443-14454, doi:10.1523/JNEUROSCI.3037-14.2014 (2014).
24 Baddeley, A. Working memory. *Science* **255**, 556-559, doi:10.1126/science.1736359 (1992).
25 Landau, S. M., Lal, R., O'Neil, J. P., Baker, S. & Jagust, W. J. Striatal dopamine and working memory. *Cereb Cortex* **19**, 445-454, doi:10.1093/cercor/bhn095 (2009).
26 Cools, R., Gibbs, S. E., Miyakawa, A., Jagust, W. & D'Esposito, M. Working memory capacity predicts dopamine synthesis capacity in the human striatum. *J Neurosci* **28**, 1208-1212, doi:10.1523/JNEUROSCI.4475-07.2008 (2008).
27 Cools, R. & Robbins, T. W. Chemistry of the adaptive mind. *Philos Trans A Math Phys Eng Sci* **362**, 2871-2888, doi:10.1098/rsta.2004.1468 (2004).
28 Courtney, S. M., Petit, L., Maisog, J. M., Ungerleider, L. G. & Haxby, J. V. An area specialized for spatial working memory in human frontal cortex. *Science* **279**, 1347-1351, doi:10.1126/science.279.5355.1347 (1998).
29 Wager, T. D. & Smith, E. E. Neuroimaging studies of working memory: a meta-analysis. *Cogn Affect Behav Neurosci* **3**, 255-274, doi:10.3758/cabn.3.4.255 (2003).
30 Fuster, J. M. & Alexander, G. E. Neuron activity related to short-term memory. *Science* **173**, 652-654, doi:10.1126/science.173.3997.652 (1971).
31 Funahashi, S., Bruce, C. J. & Goldman-Rakic, P. S. Mnemonic coding of visual space in the monkey's dorsolateral prefrontal cortex. *J Neurophysiol* **61**, 331-349, doi:10.1152/jn.1989.61.2.331 (1989).
32 Constantinidis, C. *et al.* Persistent spiking activity underlies working memory. **38**, 7020-7028 (2018).
33 Roberts, A. C., Robbins, T. W. & Weiskrantz, L. E. *The prefrontal cortex: executive and cognitive functions*. (Oxford University Press, 1998).
34 Seamans, J. K. & Yang, C. R. The principal features and mechanisms of dopamine modulation in the prefrontal cortex. *Prog Neurobiol* **74**, 1-58, doi:10.1016/j.pneurobio.2004.05.006 (2004).
35 Frank, M. J., Loughry, B. & O'Reilly, R. C. Interactions between frontal cortex and basal ganglia in working memory: a computational model. *Cogn Affect Behav Neurosci* **1**, 137-160, doi:10.3758/cabn.1.2.137 (2001).
36 Hazy, T. E., Frank, M. J. & O'Reilly, R. C. Banishing the homunculus: making working memory work. *Neuroscience* **139**, 105-118, doi:10.1016/j.neuroscience.2005.04.067 (2006).
37 Sawaguchi, T. J. N. r. The effects of dopamine and its antagonists on directional delay-period activity of prefrontal neurons in monkeys during an oculomotor delayed-response task. **41**, 115-128 (2001).
38 de Frias, C. M. *et al.* Influence of COMT gene polymorphism on fMRI-assessed sustained and transient activity during a working memory task. **22**, 1614-1622 (2010).

39 Durstewitz, D. & Seamans, J. K. J. B. p. The dual-state theory of prefrontal cortex dopamine function with relevance to catechol-o-methyltransferase genotypes and schizophrenia. **64**, 739-749 (2008).
40 Seamans, J. K., Floresco, S. B. & Phillips, A. G. J. J. o. n. D1 receptor modulation of hippocampal–prefrontal cortical circuits integrating spatial memory with executive functions in the rat. **18**, 1613-1621 (1998).
41 Stewart, C. V. & Plenz, D. Inverted-U profile of dopamine-NMDA-mediated spontaneous avalanche recurrence in superficial layers of rat prefrontal cortex. *J Neurosci* **26**, 8148-8159, doi:10.1523/JNEUROSCI.0723-06.2006 (2006).
42 Kimberg, D. Y., D'Esposito, M. & Farah, M. J. Effects of bromocriptine on human subjects depend on working memory capacity. *Neuroreport* **8**, 3581-3585, doi:10.1097/00001756-199711100-00032 (1997).
43 Irwin, D. E. Short-term memory across eye blinks. *Memory* **22**, 898-906, doi:10.1080/09658211.2013.847959 (2014).
44 Ladas, A., Frantzidis, C., Bamidis, P. & Vivas, A. B. Eye Blink Rate as a biological marker of Mild Cognitive Impairment. *Int J Psychophysiol* **93**, 12-16, doi:10.1016/j.ijpsycho.2013.07.010 (2014).
45 Bacher, L. F., Retz, S., Lindon, C. & Bell, M. A. J. I. Intraindividual and interindividual differences in spontaneous eye blinking: relationships to working memory performance and frontal EEG asymmetry. **22**, 150-170 (2017).
46 Salvi, C., Bricolo, E., Franconeri, S. L., Kounios, J. & Beeman, M. Sudden insight is associated with shutting out visual inputs. *Psychonomic Bulletin & Review* **22**, 1814-1819, doi:10.3758/s13423-015-0845-0 (2015).
47 Ahlstrom, U. & Friedman-Berg, F. J. Using eye movement activity as a correlate of cognitive workload. *Int J Ind Ergonom* **36**, 623-636, doi:10.1016/j.ergon.2006.04.002 (2006).
48 Veltman, J. A. & Gaillard, A. W. Physiological indices of workload in a simulated flight task. *Biol Psychol* **42**, 323-342, doi:10.1016/0301-0511(95)05165-1 (1996).
49 Cools, R. & D'Esposito, M. J. B. p. Inverted-U–shaped dopamine actions on human working memory and cognitive control. **69**, e113-e125 (2011).
50 Camps, M., Cortes, R., Gueye, B., Probst, A. & Palacios, J. M. Dopamine receptors in human brain: autoradiographic distribution of D2 sites. *Neuroscience* **28**, 275-290, doi:10.1016/0306-4522(89)90179-6 (1989).
51 Frank, M. J. & O'Reilly, R. C. A mechanistic account of striatal dopamine function in human cognition: psychopharmacological studies with cabergoline and haloperidol. *Behav Neurosci* **120**, 497-517, doi:10.1037/0735-7044.120.3.497 (2006).
52 Lidow, M. S., Goldman-Rakic, P. S., Gallager, D. W. & Rakic, P. Distribution of dopaminergic receptors in the primate cerebral cortex: quantitative autoradiographic analysis using [3H]raclopride, [3H]spiperone and [3H]SCH23390. *Neuroscience* **40**, 657-671, doi:10.1016/0306-4522(91)90003-7 (1991).
53 Postle, B. R. Delay-period activity in the prefrontal cortex: one function is sensory gating. *Journal of cognitive neuroscience* **17**, 1679-1690 (2005).
54 Durstewitz, D. & Seamans, J. K. The computational role of dopamine D1 receptors in working memory. *Neural Netw* **15**, 561-572, doi:10.1016/s0893-6080(02)00049-7 (2002).
55 Sawaguchi, T. & Goldman-Rakic, P. S. The role of D1-dopamine receptor in working memory: local injections of dopamine antagonists into the prefrontal cortex of rhesus monkeys performing an oculomotor delayed-response task. *J Neurophysiol* **71**, 515-528, doi:10.1152/jn.1994.71.2.515 (1994).
56 Sawaguchi, T. & Goldman-Rakic, P. S. D1 dopamine receptors in prefrontal cortex: involvement in working memory. *Science* **251**, 947-950, doi:10.1126/science.1825731 (1991).
57 Lara, A. H. & Wallis, J. D. The Role of Prefrontal Cortex in Working Memory: A Mini Review. *Front Syst Neurosci* **9**, 173, doi:10.3389/fnsys.2015.00173 (2015).

58 Curtis, C. E. & D'Esposito, M. Persistent activity in the prefrontal cortex during working memory. *Trends Cogn Sci* **7**, 415-423, doi:10.1016/s1364-6613(03)00197-9 (2003).
59 Riggall, A. C. & Postle, B. R. The relationship between working memory storage and elevated activity as measured with functional magnetic resonance imaging. *Journal of Neuroscience* **32**, 12990-12998 (2012).
60 Engel, A. K., Fries, P. & Singer, W. Dynamic predictions: oscillations and synchrony in top–down processing. *Nature Reviews Neuroscience* **2**, 704-716 (2001).
61 Fries, P. Neuronal gamma-band synchronization as a fundamental process in cortical computation. *Annual review of neuroscience* **32**, 209-224 (2009).
62 Canolty, R. T. & Knight, R. T. The functional role of cross-frequency coupling. *Trends in cognitive sciences* **14**, 506-515 (2010).
63 Roux, F. & Uhlhaas, P. J. Working memory and neural oscillations: alpha–gamma versus theta–gamma codes for distinct WM information? *Trends in cognitive sciences* **18**, 16-25 (2014).
64 Bezu, M. *et al.* Spatial Working Memory in Male Rats: Pre-Experience and Task Dependent Roles of Dopamine D1- and D2-Like Receptors. *Front Behav Neurosci* **11**, 196, doi:10.3389/fnbeh.2017.00196 (2017).
65 Arnsten, A. F. Catecholamine influences on dorsolateral prefrontal cortical networks. *Biol Psychiatry* **69**, e89-99, doi:10.1016/j.biopsych.2011.01.027 (2011).
66 Neuromelanin-sensitive MRI as a noninvasive proxy measure of dopamine function in the human brain
67 Van Orden, K. F., Limbert, W., Makeig, S. & Jung, T. P. Eye activity correlates of workload during a visuospatial memory task. *Hum Factors* **43**, 111-121, doi:10.1518/001872001775992570 (2001).
68 Benedetto, S. *et al.* Driver workload and eye blink duration. *Transportation research part F: traffic psychology and behaviour* **14**, 199-208 (2011).
69 Luck, S. J. & Vogel, E. K. The capacity of visual working memory for features and conjunctions. *Nature* **390**, 279-281 (1997).
70 Vogel, E. K., Woodman, G. F. & Luck, S. J. Storage of features, conjunctions, and objects in visual working memory. *Journal of experimental psychology: human perception and performance* **27**, 92 (2001).
71 Awh, E., Vogel, E. K. & Oh, S.-H. Interactions between attention and working memory. *Neuroscience* **139**, 201-208 (2006).
72 Kuo, B.-C., Rao, A., Lepsien, J. & Nobre, A. C. Searching for targets within the spatial layout of visual short-term memory. *Journal of Neuroscience* **29**, 8032-8038 (2009).
73 Leszczyński, M., Myers, N. E., Akyürek, E. G. & Schubö, A. Recoding between two types of STM representation revealed by the dynamics of memory search. *Journal of Cognitive Neuroscience* **24**, 653-663 (2012).
74 Sternberg, S. High-speed scanning in human memory. *Science* **153**, 652-654 (1966).
75 Xiao, J., Hays, J., Ehinger, K. A., Oliva, A. & Torralba, A. Sun database: Large-scale scene recognition from abbey to zoo. *IEEE computer society conference on computer vision and pattern recognition.* 3485-3492 (2010).
76 Ellis, B. W. *et al.* The St. Mary's Hospital sleep questionnaire: a study of reliability. *Sleep* **4(1)**, 93-97 (1981).
77 Barbato, G. *et al.* Diurnal variation in spontaneous eye-blink rate. *Psychiatry research* **93**, 145-151 (2000).
78 Doughty, M. J. & Naase, T. Further analysis of the human spontaneous eye blink rate by a cluster analysis-based approach to categorize individuals with ‘normal’versus ‘frequent’eye blink activity. *Eye & contact lens* **32**, 294-299 (2006).
79 Gramfort, A. *et al.* MEG and EEG data analysis with MNE-Python. *Front Neurosci* **7**, 267, doi:10.3389/fnins.2013.00267 (2013).

80 Stern, J. A., Walrath, L. C. & Goldstein, R. The endogenous eyeblink. *Psychophysiology* **21**, 22-33 (1984).

Image 1 (1400 ms)

Image 2 (1400 ms)

Image 3 (1400 ms)

Delay (2 seconds)

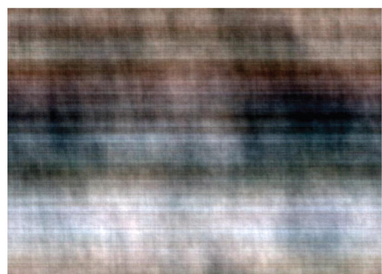
OR

Delay (5 seconds)

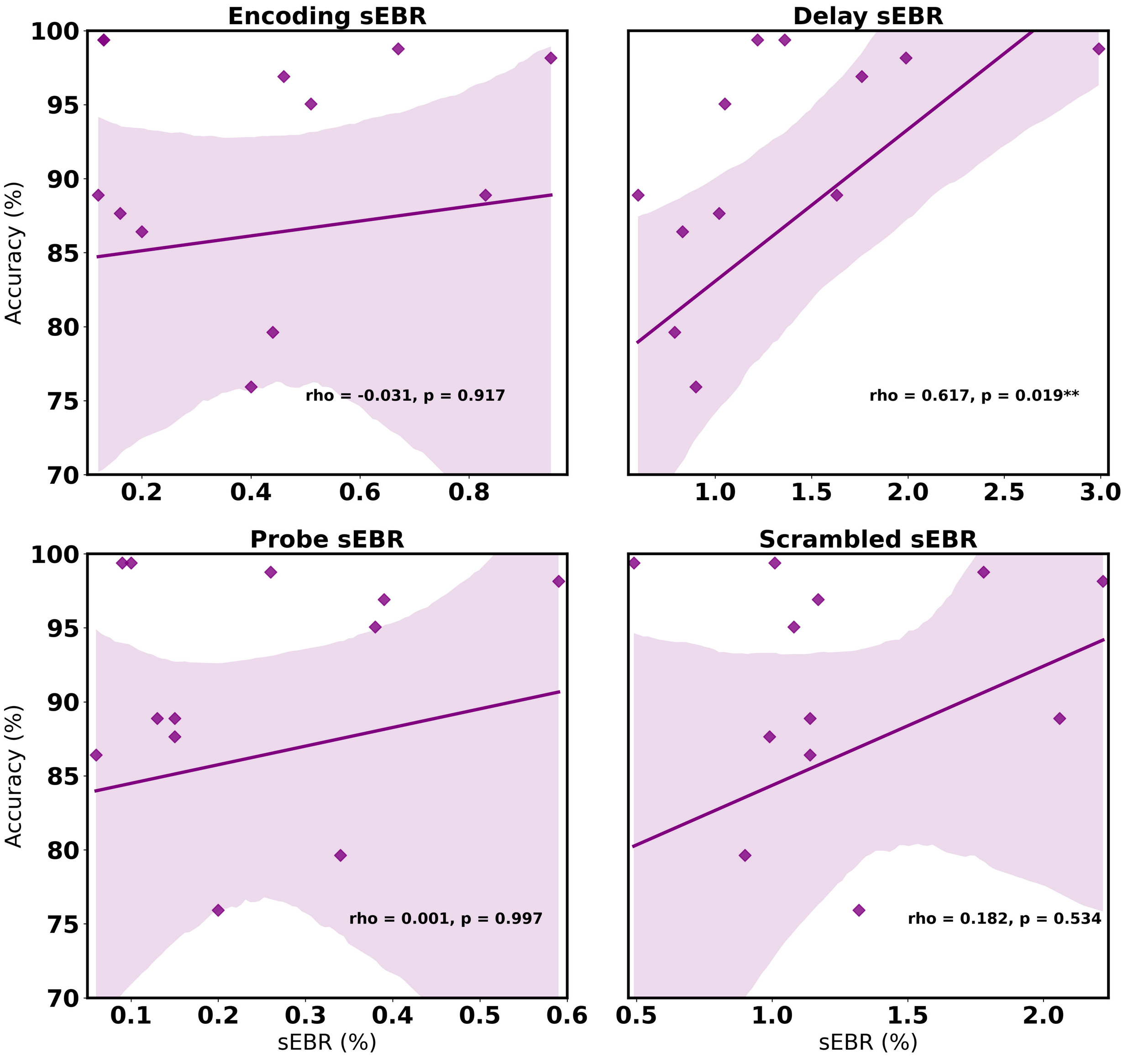
OR

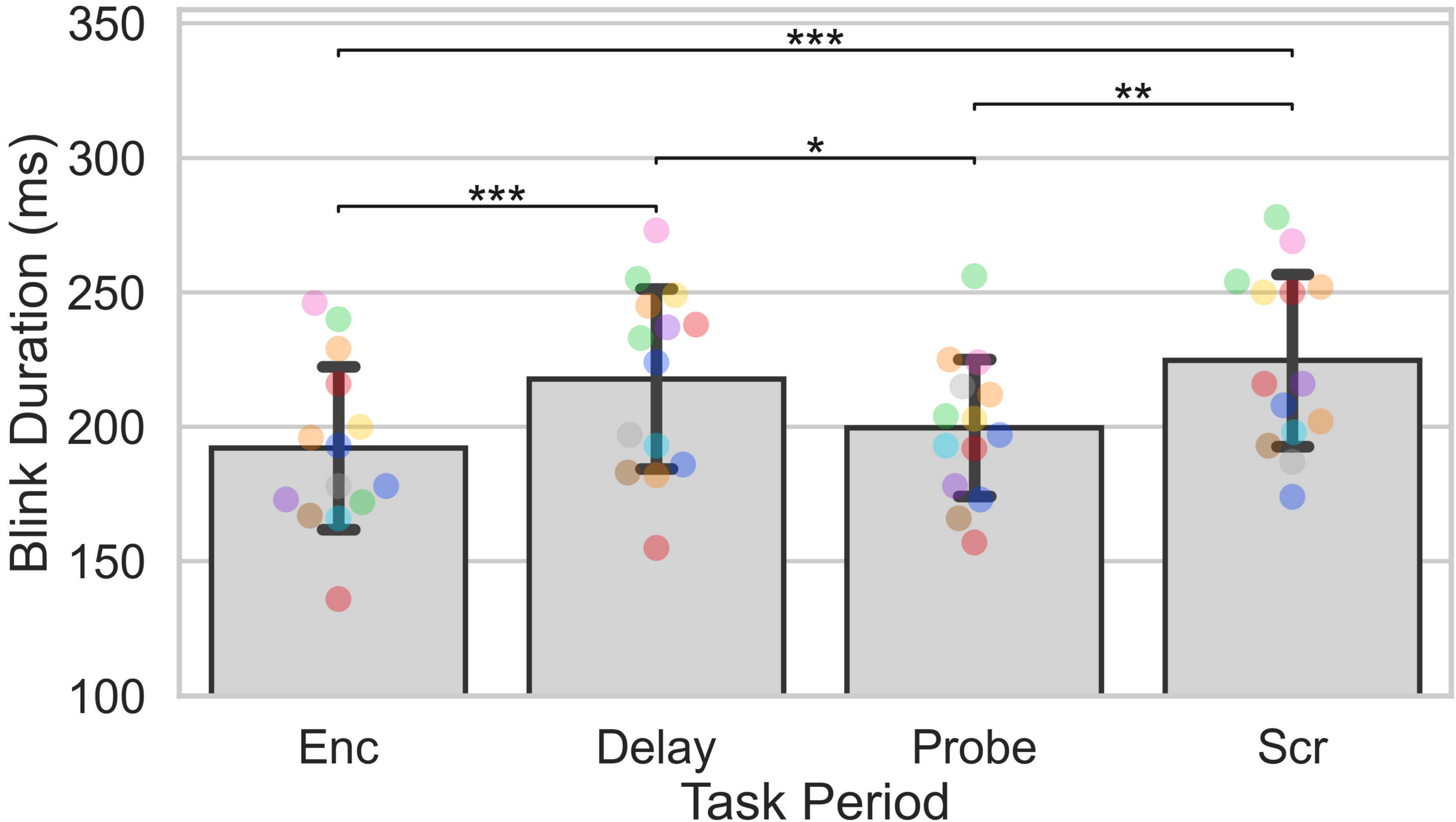
Delay (9 seconds)

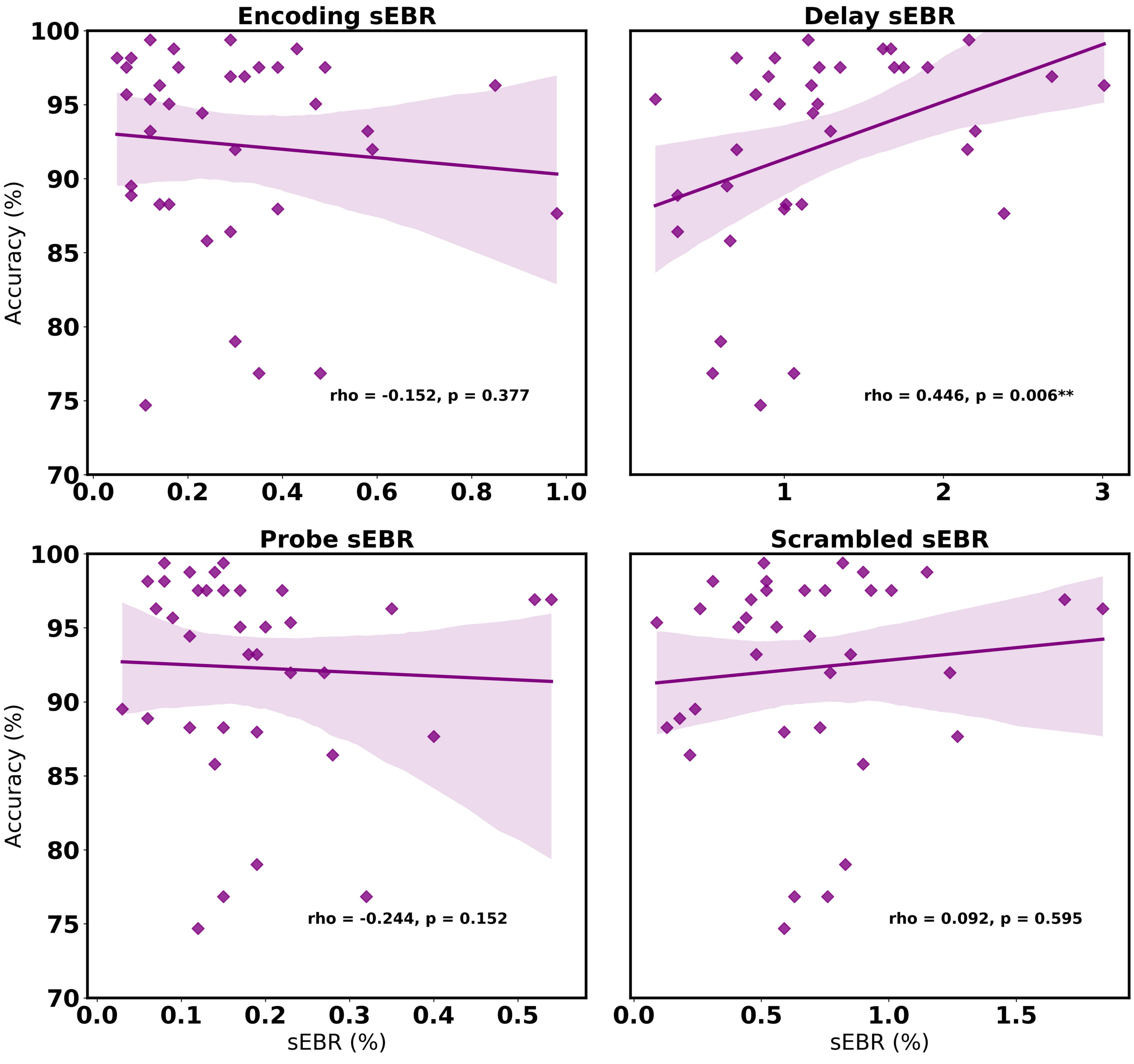
Did I see this image before?
Press green button [yes] or
red button [no] ?

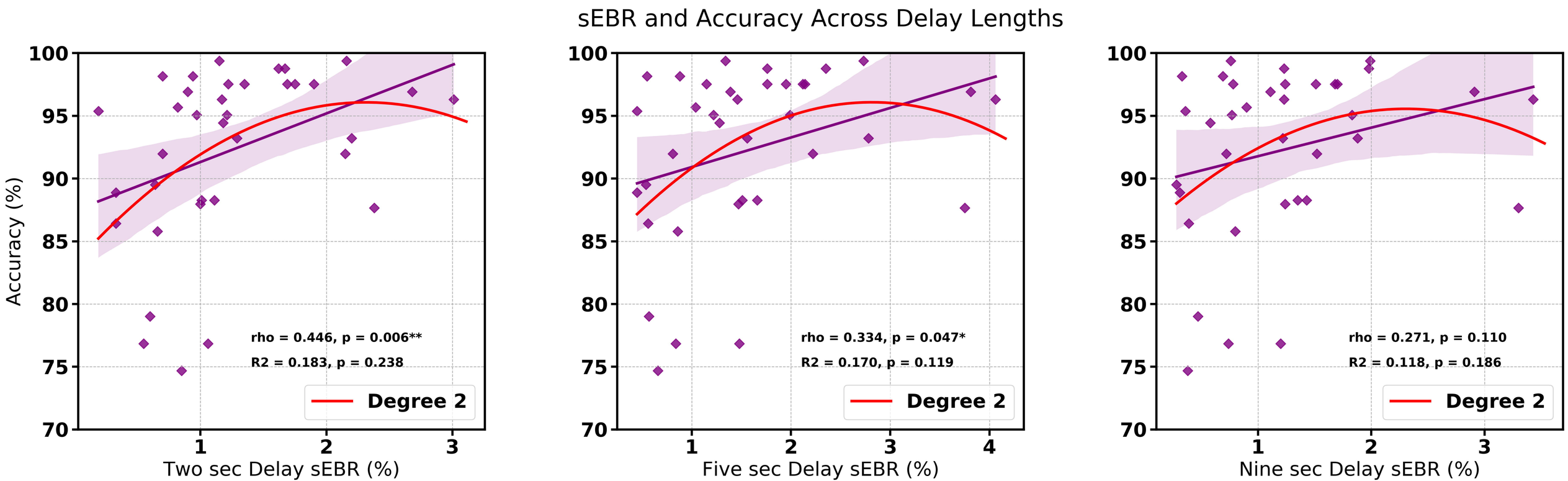


End of Trial


Encoding sEBR
Delay sEBR
Probe sEBR
Scrambled sEBR
Accuracy (%)
sEBR (%)
rho = -0.031, p = 0.917
rho = 0.617, p = 0.019**
rho = 0.001, p = 0.997
rho = 0.182, p = 0.534
100
95
90
85
80
75
70
0.2
0.4
0.6
0.8
1.0
1.5
2.0
2.5
3.0
0.1
0.3
0.5


350
300
250
200
150
100
Blink Duration (ms)
***
**
*
***
Enc
Delay
Probe
Scr
Task Period


Encoding sEBR
Delay sEBR
Probe sEBR
Scrambled sEBR
Accuracy (%)
sEBR (%)
rho = -0.152, p = 0.377
rho = 0.446, p = 0.006**
rho = -0.244, p = 0.152
rho = 0.092, p = 0.595
100
95
90
85
80
75
70
0.0
0.2
0.4
0.6
0.8
1.0
1
2
3
0.1
0.3
0.5
1.5


sEBR and Accuracy Across Delay Lengths
Accuracy (%)
Two sec Delay sEBR (%)
rho = 0.446, p = 0.006**
R2 = 0.183, p = 0.238
Degree 2
Five sec Delay sEBR (%)
rho = 0.334, p = 0.047*
R2 = 0.170, p = 0.119
Degree 2
Nine sec Delay sEBR (%)
rho = 0.271, p = 0.110
R2 = 0.118, p = 0.186
Degree 2