

Towards Finding A Link Between Neuronal Oscillations, Declarative Memory, and Viewing Behavior

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**Abstract**

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Lesion studies in humans have shown that the ability to acquire new declarative memories is a distinct cerebral function, separable from other perceptual and cognitive abilities, that relies on medial temporal lobe (MTL) structures such as the hippocampus and its surrounding cortex. For decades, looking behavior has been used as a covert measure of declarative memory. Importantly, not only do experience-dependent changes in viewing behavior often depend on the integrity of the MTL, but eye movements have also been shown to modulate neuronal activity within its structures. However, little is known about the neural mechanisms by which memory interacts with eye movements. While the MTL presumably plays a role in the consolidation of declarative memories via its extensive reciprocal connections with putative memory storage sites in neocortex, this ability is also thought to involve the synchronized activation of ensembles of neurons distributed throughout the brain. By combining intracranial electrophysiological recordings and infrared eye tracking in human epilepsy patients, this study aims to explore the hypothesis that oscillatory activity within the MTL, and between the MTL and neocortex, mediates the neural interactions underlying memory formation during free visual exploration.

## Introduction

The study of biological systems in more natural settings, to dissect the underlying operations of the nervous system, has garnered a lot of traction. Given primates' natural inclination toward visual exploration and their impressive ability for recalling visual stimuli viewed only for a couple of seconds, researchers have proposed the use of behavioral paradigms that allow both humans and monkeys to freely view images as a naturalistic approach for studying memory processes (Meister & Buffalo, 2016).

Measuring the novelty preference in looking behavior is a common metric for assessing memory during free viewing. Several studies based on this approach have uncovered a variety of effects of experience on viewing behavior. For example, in the visual paired-comparison task, human adults, infants, and monkeys exhibit a preference for looking at novel images when presented alongside repeated ones. Importantly, this behavior has been shown to depend on the integrity of MTL structures (McKee & Squire, 1993) and to correlate with aware memory (Manns, Stark, & Squire, 2000). Similarly, when human subjects look at an intermixed series of novel and repeated scenes, they tend to make fewer, and longer, fixations in response to the repeated images, and also sample less of its regions compared to novel scenes (Ryan, Althoff, Whitlow, & Cohen, 2000; Smith, Hopkins, & Squire, 2006; Smith & Squire, 2008). Taken together, these effects are commonly referred to as the "repetition effect".

Despite the converging evidence supporting the use of experience-dependent eye movements as a covert measure of memory, the exact nature and neural substrate of the memory indexed by eye movements remains under investigation. In particular, some studies have shown experience-dependent changes in viewing behavior, even when overt behavioral reports fail (Hannula & Greene, 2012). These findings have challenged the traditional view that hippocampal-dependent memory is accompanied by conscious knowledge of what has been learned, and suggest that eye movements may reveal the effects of experience independently of an individual's ability to consciously recall the experience in itself. Greene (Greene, 2007) has proposed that expectation may be an important variable when investigating aware (conscious) versus unaware (unconscious) memory, and that it may interfere with the expression of unaware memory via the repetition effect. Consistent with this observation, a recent study by Smith & Squire (Smith & Squire, 2017) found that the relationship between experience-dependent eye movements and awareness was dramatically different depending on whether participants knew that memory would be tested. Under the expectation of memory testing, the repetition effect reflected aware memory and was hippocampus-dependent. In contrast, when there was no expectation of memory testing, the repetition effect reflected unaware memory that was independent of the hippocampus. Although the latter finding is reminiscent of the phenomenon of priming (Schacter & Buckner, 1998), it would be difficult to determine whether the MTL contributes to implicit memory based on lesion evidence alone, as neuroanatomical boundaries between explicit and implicit memory may not be rigid (Dew & Cabeza, 2011). In fact, several studies have shown implicit hippocampal-related memory processing related to eye movement behavior during free viewing (Hannula & Ranganath, 2009; Olsen et al., 2016; Ryals, Wang, Polnaszek, & Voss, 2015).

Some researchers believe that recognition memory is supported by both a familiarity and a recollection process that depend on different regions of the MTL (Rudy, 2014), and currently it is not clear whether the repetition effect distinguishes between these two processes. Studies in humans have led to the proposal that the hippocampus is essential for recollection, but is not critical for simple recognition memory or judgments of familiarity (Brown & Aggleton, 2001). The "Remember/Know" procedure has

often been used to dissociate impairments in simple recognition from recall deficits (Holdstock et al., 2005), based on the assumption that “Remember” judgments reflect recollection, whereas “Know” judgments reflect familiarity. However, Squire *et al.* (Squire, Wixted, & Clark, 2007) have argued that “Remember” and “Know” judgments may instead reflect strong versus weak memories, respectively. In support of this idea, functional hippocampal activity has been associated with memory strength, even for familiarity-based or recognition memories (Kirwan, Wixted, & Squire, 2008). Testing for the source of learned information, such as knowledge of when or where something was previously experienced, has also been used for examining the contribution of MTL structures to recollection and familiarity. However, it has been suggested that source memory may be primarily dependent on the integrity of the frontal lobes (Buckner & Wheeler, 2001), as functional hippocampal activity has been found to support item and source memory judgments to a similar degree (Gold et al., 2006).

If the MTL were critical for recognition memory performance, the evoked activity of its neurons should reflect whether a given stimulus is novel or familiar (Jutras & Buffalo, 2010). Such modulations have been described among single neurons in the MTL. In both humans and monkeys, for example, hippocampal neurons modulate their activity in response to repeated scenes, and the magnitude of this response correlates with memory strength for the familiar stimuli (Jutras & Buffalo, 2010; Rutishauser, Mamelak, & Schuman, 2006). In contrast, neural signals in the rhinal cortex often show significant firing rate changes related to the repetition of specific stimuli (Suzuki, Miller, & Desimone, 1997). This pattern of activation may support recognition memory through the combination of stimulus-selective information from the perirhinal and entorhinal cortices, and a more general, abstract signal of novelty or familiarity (Jutras & Buffalo, 2010).

Since neuronal spiking in multiple brain regions, including the prefrontal cortex, is phase-locked to the hippocampal theta rhythm in rodents (Sirota et al., 2008), hippocampal theta oscillations are thought to be important for organizing interactions between disparate brain regions. In monkeys, theta-band power in hippocampal local field potentials (LFPs) for well-remembered images is significantly higher immediately preceding stimulus onset, and this effect has been shown to extend beyond the early stages of stimulus presentation (Jutras, Fries, & Buffalo, 2013). Moreover, the phase of hippocampal theta has been shown to modulate the amplitude of locally-generated gamma-band oscillations (Colgin et al., 2009), which could be relevant for memory given that gamma synchrony, within and between MTL regions, is enhanced during successful memory task performance (Fell et al., 2001; Montgomery & Buzsáki, 2007). In monkeys, hippocampal spike-gamma-band field coherence during the encoding of complex images is positively correlated with subsequent recognition memory performance (Jutras, Fries, & Buffalo, 2009). This synchronization of neuronal spiking to the local network oscillation may represent a mechanism by which hippocampal neurons, sharing similar response properties, undergo functional coupling during memory formation (Canolty et al., 2010). Furthermore, it could contribute to spike-timing dependent long-term potentiation (Bi & Poo, 1998), allow for a representation of behaviorally-relevant stimuli to be held in working memory (Pesaran et al., 2002), and/or enhance the impact of hippocampal neurons on output targets in the entorhinal cortex. In both humans and monkeys, a positive correlation between hippocampal gamma-band power during stimulus encoding and recognition memory performance has also been observed (Jutras et al., 2009; Sederberg et al., 2007). However, in contrast to gamma-band coherence, gamma-band power also increases in proportion to image looking time (Jutras et al., 2009). Therefore, the amplitude of the LFP may reflect both attention and memory on a trial-by-trial basis, which is consistent with studies associating specific

ERP deflections (*e.g.*, the P3 wave) with attentional processing (Kok, 2001), and with hippocampal-dependent processing of novel stimuli (Knight, 1996).

Finally, eye movements during stimulus encoding have been associated with modulations of oscillatory hippocampal activity that predict memory formation. For example, research in monkeys has shown that the phase of theta oscillations during stimulus encoding is reset upon saccade offset, and this phase reset is more consistent during the viewing of images that are later well-remembered. Interestingly, this effect is specific to saccades made in anticipation of an exploratory state (Jutras et al., 2013), and is not present when saccades are made in the dark (Hoffman et al., 2013), suggesting that it may arise from the timing of potentially relevant visual input into the hippocampus. Since long-term potentiation in the hippocampus is preferential for specific theta phases (Hölscher, Anwyl, & Rowan, 1997), phase resetting may represent a mechanism that ensures that the hippocampus is in an ideal encoding state for integrating incoming sensory information into memory via synaptic plasticity (Jutras et al., 2013). It has also been suggested that the theta cycle provides a reference in which multiple items are represented at different phases of the cycle, which may have important implications for the maintenance of working memory and for long-term memory retrieval (Fell & Axmacher, 2011).

By gaining additional knowledge about changes in large-scale network activity within the MTL, and between the MTL and neocortex during free viewing, this project aims to expand our understanding of the dynamic neural correlates of memory indexed by eye movements.

## Methods

Electrocorticography (ECoG) and stereoelectroencephalography (sEEG) signals were recorded from ten subjects being monitored for intractable medial temporal lobe epilepsy at Harborview Medical Center (Seattle, WA, USA). The number, type, orientation, and size of the electrode arrays was determined based on each individual's anatomy and clinical considerations. All participants were provided with informed consent in accordance with the University of Washington's Institutional Review Board. Electrode positions were identified from structural MRI and CT scans using iElectrodes (Blenkman et al., 2017). Neural signals from 6 of the participants were recorded at approximately 2 kHz using a PZ5 NeuroDigitizer and an RZ5D BioAmp Base Processor (Tucker-Davis Technologies). However, due to a filtering issue in one of the digital signal processors, a copy of the data (sampled at 1 kHz) had to be recovered from the Xltek EMU128FS clinical brain monitoring system (Natus Medical Incorporated). Channels with interictal epileptiform discharges (IEDs) were used to estimate clock drift between these two electrophysiology systems, and to align the clinical neural recordings with the experimental TTL pulses. An ATLAS Neurophysiology System (Neuralynx) was used to record the neural signals from the remaining 4 participants at approximately 4 kHz. All neural recordings were resampled to 1 kHz and rereferenced using a Laplacian scheme. Channels with excessive artifacts or power line noise, or those lying around the seizure onset zone, were discarded. In addition, following visual inspection, all trials with IEDs and all channels with more than 10% of trials containing IEDs, were removed. However, these criteria led to almost all channels within the MTL being rejected. Therefore, future work on the development of automated methods for dissecting the underlying irritative networks (see Janca et al., 2018), quantifying their properties, and measuring their effect on memory performance and eye movement behavior is required.

At the beginning of the experiment, participants were informed that they would be viewing a

series of scenes and that the objective of the study was to learn how people visually explore the world. They were instructed to look at each scene for as long as it was displayed on the screen, while remaining alert and keeping their heads still. Stimuli were randomly selected from a large collection of natural scenes from National Geographic (including landscapes, cityscapes, wildlife, etc.), scaled to approximately  $20 \times 15^\circ$ , and displayed on a V176LBM LCD monitor (Acer) positioned approximately 52 cm away from the participant's face. Stimulus delivery and experimental control were achieved using Presentation software (Neurobehavioral Systems). Synchronization pulses were sent from the display computer to both the eye tracking and electrophysiological recording systems in order to align stimuli presentation with the gaze and neural recordings, respectively. In addition, a photoresistor was attached to the display monitor to control for latency/jitter in stimulus timing.

The experimental task was divided into four blocks (Fig. 1). In Block 1, 60 scenes were presented, 40 from Set A and 20 from Set B. In Block 2, 40 repeated scenes (Set A) were presented, intermixed with 60 novel scenes (40 from Set C and 20 from Set D). During Blocks 1 and 2, eye movements were recorded while participants simply viewed the scenes without expectation of memory testing. In Block 3, memory was tested for a subset of the repeated and novel scenes from Block 2 (40 from Set A and 40 from Set C, respectively), and participants made old/new recognition judgments using a slider scale. Note that participants' responses in Block 3 were used as an estimate of whether the repeated scenes from Block 2 could have been recognized as familiar if memory had been tested after each scene presentation during Block 2, resembling a source memory judgement. In Block 4, 40 repeated scenes (20 from Set B and 20 from Set D) were presented, intermixed with 40 novel scenes (Set E), and memory judgements were made immediately following each scene. Furthermore, source memory was tested by asking participants whether the images they identified as familiar in Block 4 were first shown during Blocks 1 or 2. In contrast to Block 3, during Block 4 eye movements were recorded while participants viewed the scenes under expectation of memory testing. In Blocks 1, 2, and 4 a fixation target was shown for a random duration of 1 – 1.5 s before each scene, and scenes were presented for 3 s. Conversely, in Block 3 each scene remained on the screen until the participant clicked on the slider scale, and a blank screen was shown for 200 ms between scenes. Old/new and source memory judgements were discretized offline into a six-point likert scale (1 = definitely X, 2 = probably X, 3 = maybe X, 4 = maybe Y, 5 = probably Y, and 6 = definitely Y, where X and Y represent the two choices) adjusted to account for response bias.

Eye movements were recorded at 1 kHz using an infrared EyeLink Portable Duo eye tracker (SR Research). To precisely map the participants' gaze onto the computer screen pixels, the eye tracker was calibrated prior to experimental Blocks 1 and 4 by having the participants fixate on various targets within a 13-point array covering up 90% of the screen's area. Instantaneous eye velocity and acceleration were computed via the EyeLink's Online Parser System, which implements a moving filter that is optimal for the detection of microsaccades, minimizes the extension of saccade durations, and preserves saccadic peak velocities. Furthermore, eye movements were automatically parsed into fixations, saccades, and blinks using a novel unsupervised clustering method (Fig. 2A) in MATLAB (MathWorks). Briefly, the algorithm first identifies blink periods as data segments with missing pupil information or with very fast changes in pupil area. To remove eye movement artifacts around the blinks caused by partial pupil occlusion, the end points of each blink period are extended until the eye velocity and acceleration measurements fall, for at least 20 ms, below thresholds of 22  $^\circ/s$  and 3800  $^\circ/s^2$ , respectively. These threshold values are typically used by the EyeLink's Online Parser System for detecting saccades with

amplitudes as small as  $0.3^\circ$ . The algorithm then searches for velocity local maxima, separated by at least 40 ms to avoid dynamic overshoots, and selects the largest peaks across a 1 s sliding window to obtain an average saccade rate of 5 Hz (Fig. 2B). Since saccade rates are typically lower than 4 Hz, this set of candidate events contains both true saccades and an undetermined, but upper bounded, number of non-saccadic eye movements (Otero-Millan et al., 2014). Next, the 2-D components underlying these peaks (Fig. 2C) are converted into polar coordinates, and the radial coordinates (log-transformed, standardized, and winsorized) are separated into 2 groups using a hierarchical clustering algorithm (Fig. 2D); the group with the largest average peak velocity is selected as the true saccade cluster. To identify fixation events, the 2-D components (standardized and winsorized) underlying all other local velocity maxima are transformed via PCA, and this new set of observations is analyzed using a velocity-based algorithm adapted from (Engbert & Kliegl, 2003). Specifically, all observations lying within an ellipse with major and minor axes equal to three times the standard deviation of the principal components are identified as fixations, and observations beyond the confines of this ellipse are added to the saccade cluster (Fig. 2E). Finally, these fixation and saccade events are used within a k-nearest neighbors algorithm (with  $K = 3$  neighbors) to classify all remaining gaze samples as belonging to either a fixation or saccadic eye movement (Fig. 2F). To remove false positives, any saccades at least 10 ms long and with velocity peaks less than 40 ms apart, are grouped into smaller clusters, and all events except for the leading saccade in each cluster are reclassified as fixations. Similarly, saccades with amplitude less than or equal to  $0.3^\circ$  are reclassified as fixations, and any remaining saccades with end points within 20 ms from each other are merged into a single event. Finally, since the blink process may truncate or produce spurious fixations, all fixation events shorter than 100 ms immediately preceding or following a blink are removed.

Three eye movement measures assessed how participants viewed the novel and repeated scenes during Blocks 2 and 4: (a) the number of complete fixations (*i.e.*, fixations that started and ended within the 3 s viewing period); (b) the average fixation (geometric mean) duration for all complete fixations during the 3 s viewing period; and (c) the number of regions sampled, that is, the number of different regions within a  $22 \times 17$  grid of  $1 \times 1^\circ$  squares tiling up the scene (plus  $1^\circ$  margins) in which a complete fixation was detected. Eye movements in Blocks 1 and 3 were not analyzed. Moreover, trials where participants made 3 or fewer complete fixations and/or complete saccades (amounting to at least one fixation/saccade per second) were removed from further analysis. The differential viewing of novel and repeated scenes (*i.e.*, the repetition effect) was tested using two-tailed, Wilcoxon signed rank tests. Data from one of the participants, who had previously undergone a temporal lobectomy and exhibited both abnormal eye movement activity and subpar memory performance, was excluded from all analyses.

## Results

**Memory accuracy:** Participants were slightly less accurate ( $P = .078$ ) in their memory judgements during Block 3 ( $72.3 \pm 8.9\%$ ,  $d' = 2.8 \pm 1.8$ ) than Block 4 ( $78.5 \pm 6.2\%$ ,  $d' = 1.9 \pm 1.6$ ). Considering that none of the scenes in Block 3 were truly novel, it was surprising to find similar false alarm rates (*i.e.*, identifying novel scenes as repeated) between Blocks 3 and 4 ( $19.4 \pm 11.0\%$  vs.  $11.9 \pm 15.7\%$ ,  $P = .188$ ). Memory scores for the repeated scenes were also similar ( $64.1 \pm 13.1\%$  and  $69.0 \pm 10.6\%$  for Blocks 3 and 4, respectively,  $P = .188$ ). Hence, the ability of participants to identify familiar scenes was not affected by

having to make memory judgements in Block 3 for repeated images presented during Block 2. Consequently, the following two analyses were performed for Blocks 2 and 4: eye movement measures from all novel scenes were compared with eye movement measures from repeated scenes that were both correctly (hits), and incorrectly (misses), identified. In particular, the hits were used to measure eye movements in association with aware memory, and the misses were used to measure eye movements related to unaware memory.

**Eye movements (Block 2):** During Block 2, under no expectation of memory testing, participants viewed novel (Set C) and repeated scenes (Set A) differently; *i.e.*, they exhibited the repetition effect. Specifically, participants made fewer fixations ( $7.9 \pm 1.8$  vs.  $8.4 \pm 1.8$ ), sampled less regions ( $7.3 \pm 1.5$  vs.  $7.7 \pm 1.5$ ), and made longer fixations ( $255.9 \pm 39.7$  vs.  $244.4 \pm 36.6$  ms) when viewing repeated versus novel scenes (Fig. 3). Importantly, participants exhibited the repetition effect independently of both the accuracy of their memory judgements, and their confidence level in them (Fig. 4). These analyses were based on all participants that had at least one hit and one miss, and at least one high-confidence and one low-confidence judgment associated with a repeated scene, respectively. High-confidence responses were chosen as confidence ratings of 1 or 6 (*i.e.*, definitely sure), and low-confidence responses as confidence ratings from 2 – 5 (*i.e.*, maybe sure or probably sure).

**Eye movements (Block 4):** Participants also exhibited the repetition effect when they expected that memory would be tested (Fig. 5). Specifically, when viewing repeated scenes (Sets B and D) compared to the novel scenes (Set E), participants made fewer fixations ( $8.4 \pm 2.0$  vs.  $8.8 \pm 2.1$ ), sampled less regions ( $7.4 \pm 1.6$  vs.  $8.2 \pm 1.7$ ), and made longer fixations ( $254.4 \pm 56.0$  vs.  $251.8 \pm 46.5$  ms). However, in contrast to the findings from Block 2, the repetition effect was primarily observed when participants identified repeated scenes correctly, or when they had high confidence in their decisions (Fig. 6). These analyses were based on all participants that had at least one hit and one miss, and at least one high-confidence and one low-confidence judgment associated with a repeated scene, respectively. High-confidence responses were chosen as confidence ratings of 1 or 6 (*i.e.*, definitely sure), and low-confidence responses as confidence ratings from 2 – 5 (*i.e.*, maybe sure or probably sure).

## Discussion

Patients with medial temporal lobe epilepsy view previously presented scenes differently than novel scenes (the repetition effect). Specifically, when viewing familiar scenes, patients make fewer fixations, sample fewer regions, and make longer fixations compared to novel scenes (Fig. 3 & Fig. 5). When simply viewing the scenes with no expectation of memory testing, the repetition effect occurred regardless of whether the patients recognized the old/new status of the scenes, as well as whether their memory judgements were made with high or low confidence (Fig. 4). Although the sample size and number of trials were not large enough to account for the small effect sizes, as previously reported in (Smith & Squire, 2017), the general trends (*i.e.*, the skewed distributions of data points around the reference line  $y = x + b$ ) are consistent with their previous findings in both healthy control subjects and in patients with bilateral lesions limited to the hippocampus. Consequently, when there is no expectation of memory testing, the repetition effect seems to reflect unaware memory that has been shown to be

independent of the hippocampus. In contrast, when patients expected that memory would be tested, the repetition effect was mainly observed when they were aware of which scenes were old and which were new, and when they had high confidence in their memory judgements (Fig. 6). This is consistent with previous findings that showed that, in the context of memory testing, the repetition effect not only reflects aware memory, but is also hippocampus-dependent (Smith & Squire, 2008).

In general, under no expectation of memory testing, the repetition effect was larger (*i.e.*, smaller *p*-values) when memory judgements were associated with low confidence compared to high confidence, or when patients failed to recognize repeated scenes as old (misses) compared to when they successfully recognized them (hits). The latter is consistent with previous findings from Smith & Squire (Smith & Squire, 2017), and could be related to how strongly the patient senses that a scene has been previously encountered. In other words, a belief that a scene was presented earlier might lead to eye movements intended to confirm such an impression, and these eye movements might differ from those that scan an apparently novel scene.

The finding that expectation of memory testing can affect eye movements is reminiscent of seminal work by Yarbus (Yarbus, 1967), in which participants sampled different regions of a scene depending on the questions that they were asked about it. However, the fact that experience-dependent eye movements can reflect aware or unaware memory depends not only on task instructions, or an expectation of memory testing, but also on the type of task. For instance, the phenomenon whereby participants preferentially explore the altered portion of a repeated scene reflects aware, hippocampus-dependent memory, regardless of whether memory testing is expected (Smith, Hopkins, & Squire, 2006; Smith & Squire, 2008). In addition, the preferential viewing effect typically observed in visual paired-comparison tasks, for which participants do not expect memory testing, has been associated with aware, hippocampus-dependent memory. Therefore, to fully characterize the interaction between awareness and the MTL in experience-dependent eye movements, it is critical to determine (1) whether the effect reflects aware or unaware memory; (2) whether the effect depends on the MTL; and (3) whether task instructions or an expectation of memory testing alter the relationship between the effect and aware/unaware memory.

### **Future Research**

Having characterized the repetition effect in this patient population, despite the high incidence of IEDs within their MTLs, future efforts should focus on identifying electrophysiological biomarkers of memory performance (measured through both experience-dependent changes in viewing behavior and recognition memory tests) during the free viewing of complex scenes. Naturally, one would ask whether successful stimuli encoding and/or recall is associated with differential modulations in network-level activity, such as power/phase fluctuations in the LFP and changes in cross-frequency coupling within the MTL, and between the MTL and neocortex. Furthermore, one could examine the effect that expectation of memory testing may have on MTL activity, and its relationship with eye movements.

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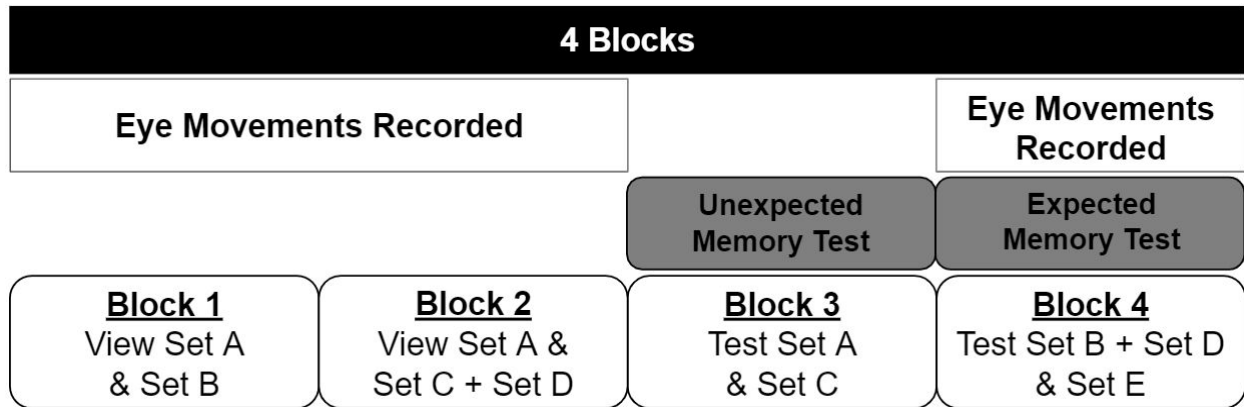
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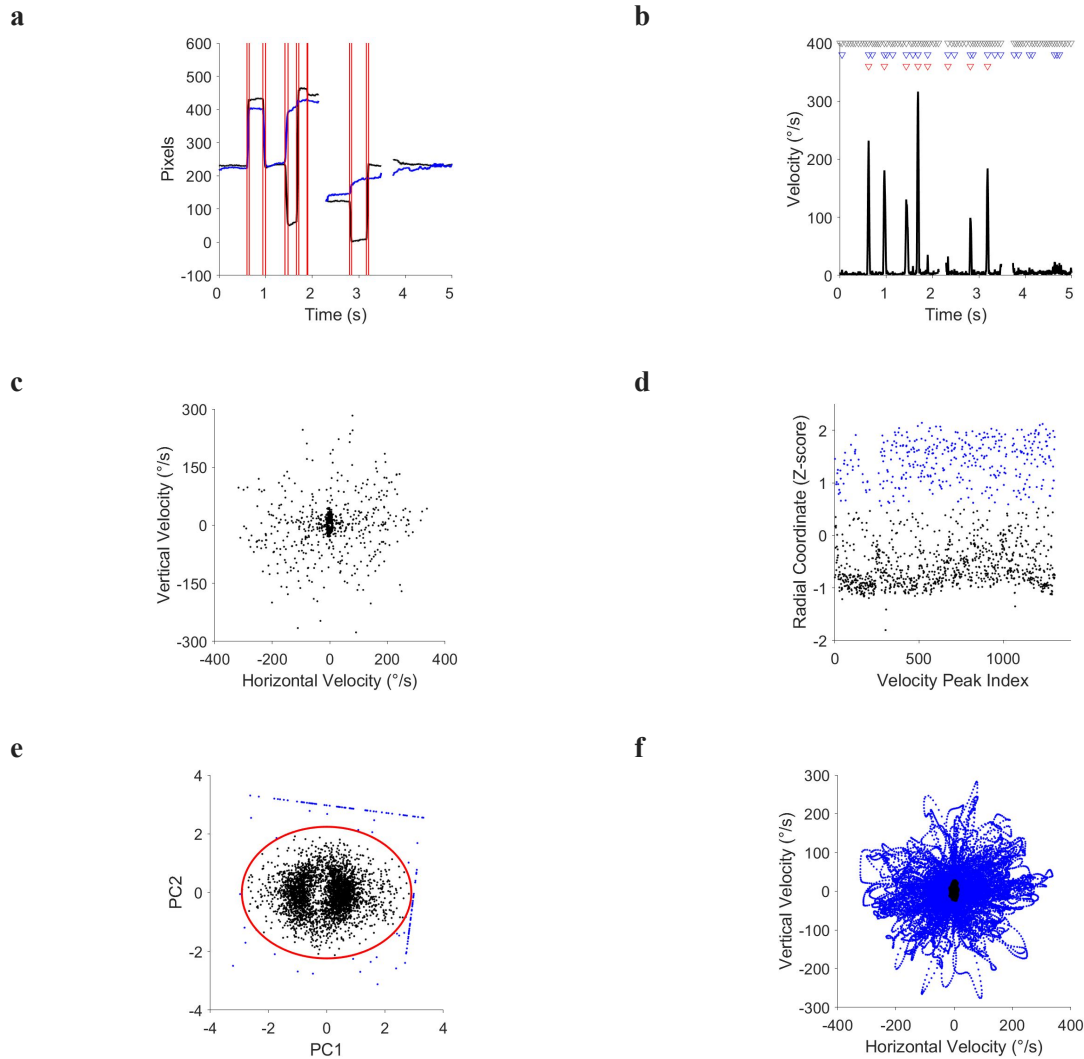
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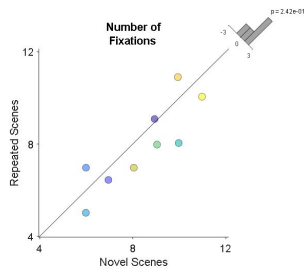
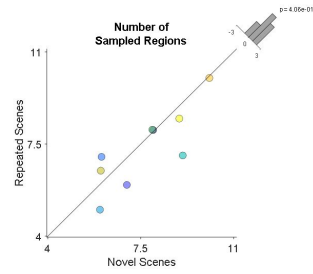
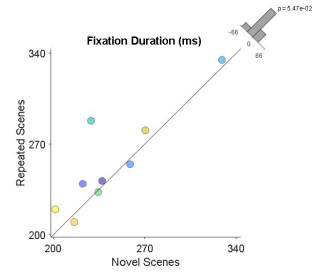
## Figures



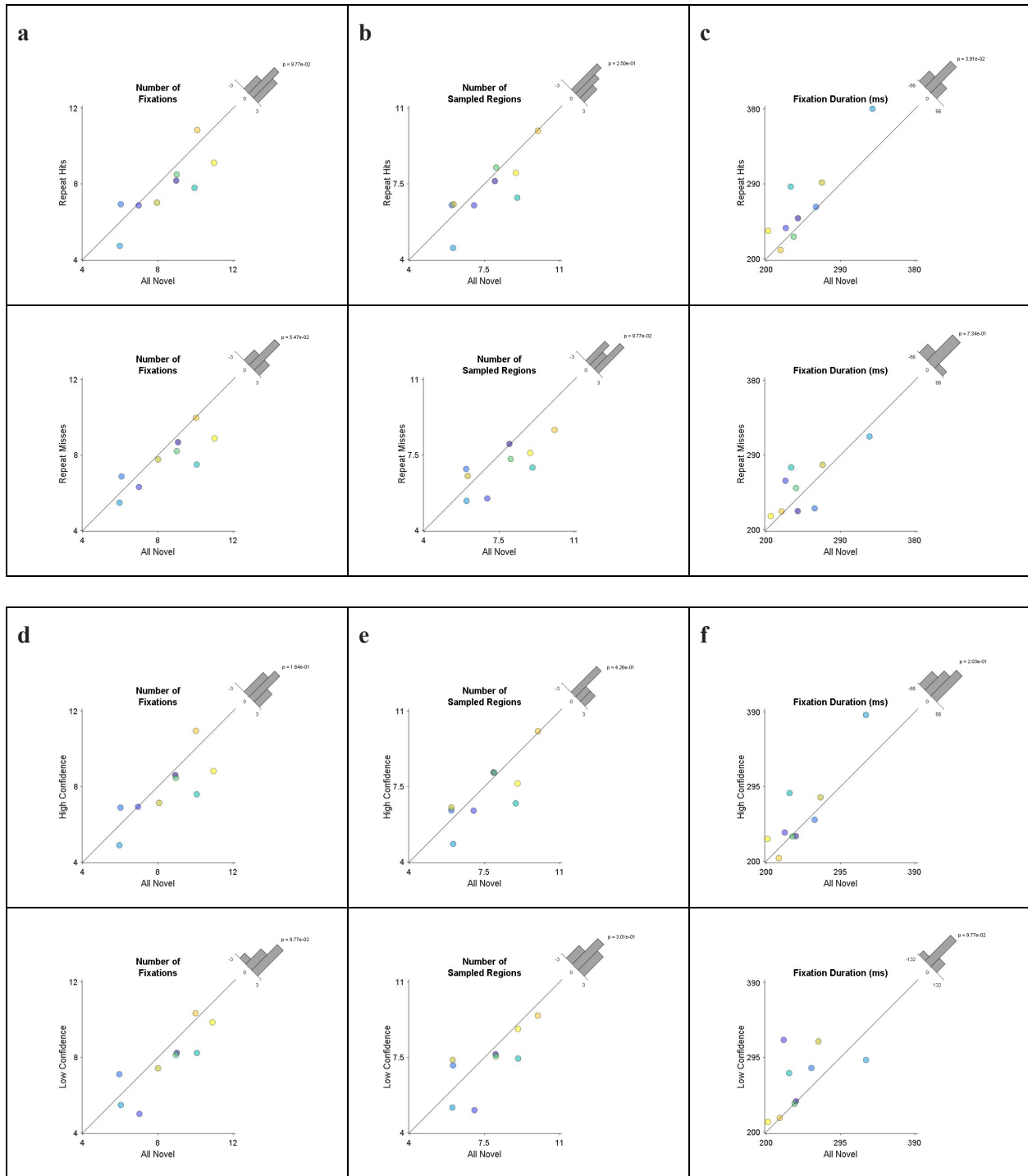
**Figure 1.** The four experimental blocks in the recognition memory task. In Block 1, participants viewed 60 natural scenes (40 from Set A and 20 from Set B). In Block 2, participants again viewed the 40 scenes from Set A, intermixed with 60 novel scenes (40 from Set C and 20 from Set D). During Blocks 1 and 2, eye movements were recorded in response to novel and repeated scenes while participants simply viewed the scenes with no expectation of memory testing. In Block 3, 80 scenes from Block 2 were presented again (40 from Set A and 40 from Set C), and participants used a slider scale to make judgements on whether the scenes had been old/new during Block 2. These memory scores allowed eye movements in Block 2 to be separately analyzed depending on whether they could have been recognized as familiar if memory had been tested following each scene presentation. In Block 4, participants viewed 40 repeated scenes (20 from Set B and 20 from Set D), intermixed with 40 novel scenes (Set E), and made old/new recognition judgments after each scene. Thus, Block 4 provided concurrent measures of eye movements and memory in response to novel and repeated scenes.



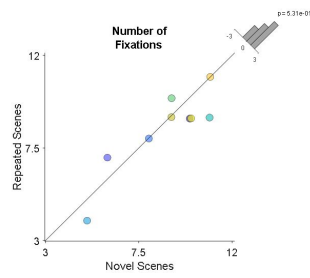
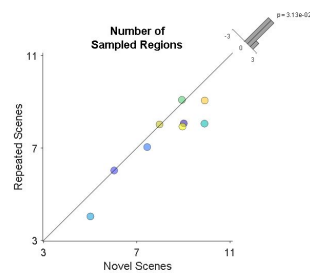
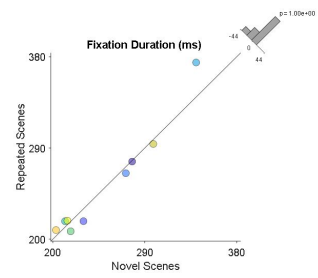
**Figure 2.** New unsupervised clustering algorithm for microsaccade detection. **(a)** Five second recording of horizontal (black trace) and vertical (blue trace) eye position. The gaps represent blink periods, and the vertical red lines represent the end points of the saccades detected by the algorithm. **(b)** Instantaneous eye velocity during this 5 s viewing period. Gray triangles represent local velocity maxima separated by at least 40 ms to avoid dynamic overshoots, blue triangles represent the largest of these velocity peaks within a 1 s sliding window to obtain an average saccade rate of 5 Hz, and the red triangles represent the final saccadic eye movement events detected by the algorithm. **(c)** Two-dimensional components underlying the velocity peaks marked by the blue triangles in (b). **(d)** Radial coordinates (log-transformed, standardized, and winsorized) resulting from the conversion of the 2-D velocities to polar coordinates. This new set of observations is separated into two groups using a hierarchical clustering algorithm, and the group with the largest average peak velocity (blue dots) is selected as the true saccade cluster. **(e)** The 2-D components (standardized and winsorized) of all other velocity peaks, marked by the gray triangles in (b), are transformed via PCA. All observations lying within an ellipse with major and minor axes equal to three times the standard deviation of the principal components (red curve) are classified as fixations (black dots), and any observations lying outside of this ellipse (blue dots) are added to the saccades cluster. **(f)** Finally, these fixation and saccade events are used within a KNN algorithm to classify all remaining gaze samples as belonging to either a fixation (black dots) or saccadic eye movement (blue dots).

**a****b****c**

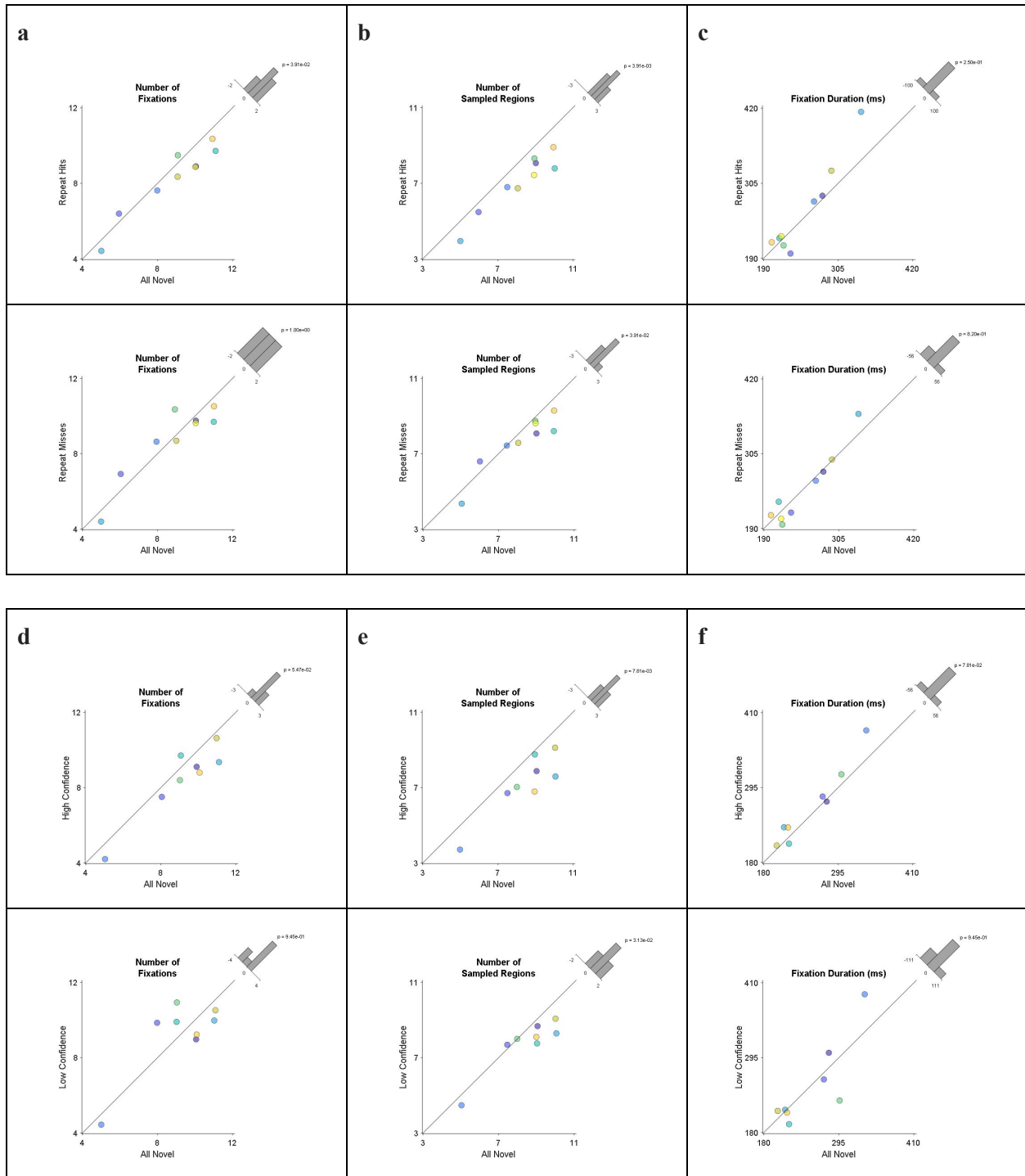
**Figure 3.** Measures of eye movements in response to 40 repeated scenes (Set A) and 40 novel scenes (Set C) while participants simply viewed the scenes without expectation of memory testing (Block 2). Recognition memory judgments for the same scenes were obtained directly after eye movements were recorded (Block 3). Data points represent the median measures for each participant computed across all trials where they made more than 1 fixation/saccade per second. Participants made **(a)** fewer fixations, **(b)** sampled less regions, and **(c)** made longer fixations in response to repeated scenes compared to novel scenes.



**Figure 4.** Participants viewed the repeated and novel scenes in Block 2 differently (*i.e.*, exhibited the repetition effect) irrespective of (a-c) whether their old/new judgements for the repeated scenes were correct (hits) or incorrect (misses), or (d-f) whether the latter were associated with high or low confidence. These analyses were based on all participants that had at least one hit and one miss, and at least one high-confidence and one low-confidence judgment associated with a repeated scene, respectively. High-confidence responses were chosen as confidence ratings of 1 or 6 (*i.e.*, definitely sure), and low-confidence responses as confidence ratings from 2 – 5 (*i.e.*, maybe sure or probably sure). Data points represent the average measures (geometric mean) for each participant computed across all trials where they made more than 1 fixation/saccade per second.

**a****b****c**

**Figure 5.** Measures of eye movements in response to 40 repeated scenes (20 from Set B and 20 from Set D) and 40 novel scenes (Set E) when memory was tested concurrently (Block 4). As in Block 2, participants made **(a)** fewer fixations, **(b)** sampled less regions, and **(c)** made longer fixations when viewing repeated scenes compared to novel scenes. Data points represent the median measures for each participant computed across all trials where they made more than 1 fixation/saccade per second.



**Figure 6.** In contrast to Block 2, participants exhibited the repetition effect only (a-c) when their memory judgements were correct, or (d-f) when their memory judgments were associated with high confidence. These analyses were based on all participants that had at least one hit and one miss, and at least one high-confidence and one low-confidence judgment associated with a repeated scene, respectively. High-confidence responses were chosen as confidence ratings of 1 or 6 (*i.e.*, definitely sure), and low-confidence responses as confidence ratings from 2 – 5 (*i.e.*, maybe sure or probably sure). Data points represent the average measures (geometric mean) for each participant computed across all trials where they made more than 1 fixation/saccade per second.