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Using Pupillometry to Assess Visual Working Memory for Temporal Order

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Abstract

What factors influence our ability to maintain the temporal order of sequences in visual working memory? We used taskevoked pupillometry to investigate how the semantic content and spatial positions of images influence memory for temporal order. We found that memory for temporal order was stronger for sequences consisting of semantically meaningful images compared to abstract images, but there was no memory benefit for presenting sequences from left-to-right compared to centrally or from right-to-left. In addition, we found that pupil dilation was greater for sequences of semantic images compared to abstract images and particularly for items later in the sequence and in sequences presented from left-to-right. These results highlight the utility of using pupillometry to study working memory processes and provide new insights into how the nature of the items to be remembered and the spatial positions of those items influence visual working memory for temporal order.

Keywords: pupillometry; working memory; spatiotemporal associations; temporal memory; serial order

Introduction

Working memory is the cognitive system that enables us to maintain information in mind once sensory traces of that information are no longer present (Baddeley, 2012). Classically, working memory capacity has been measured using explicit behavioral responses, including verbal recall and same/different judgments. More recently, phasic changes in pupil dilation have been used to assess working memory processes. In addition to responding to changes in light levels, pupils are responsive to changes in arousal and cognitive activity (Eckstein et al., 2017; Mathôt, 2018). Across a variety of cognitive tasks, pupil dilation has been shown to track with difficulty and engagement, such that more difficult tasks and greater cognitive effort is associated with increased pupil dilation relative to easier tasks that require less cognitive effort. Because changes in pupil dilation occur spontaneously, pupillometry is a powerful implicit method for studying working memory processes.

In working memory tasks, pupil dilation tracks with memory load, such that increasing memory load results in increasing pupil dilation (Granholm et al., 1996; Kahneman & Beatty, 1966). In addition, pupil dilation also tracks with the success of working memory processes. For example, in visual working memory tasks with arrays of items presented simultaneously, task-evoked pupil dilation during the delay period following stimulus presentation is greater for arrays that will be successfully remembered compared to arrays that will be forgotten (Robison & Unsworth, 2019; Unsworth & Robison, 2018). In aural and visual digit span tasks in which the items are presented sequentially, pupil dilation is greater for longer sequences compared to shorter sequences (Kahneman & Beatty, 1966; Klingner et al., 2011). Further, item-level analyses of pupil dilation reveal that individual and age-related differences in the number of items for which the pupil continues to dilate are related to differences in working memory capacity (Johnson et al., 2014). Pupil dilation increases for items in a sequence that are below or approaching an individual's maximum capacity, and then plateaus or declines when items continue to be presented beyond that maximum capacity. Taken together, these studies demonstrate that fluctuations in pupil dilation not only track the overall memory load, but also the degree to which individuals remain engaged with the task and continue to exert effort to encode new items into working memory.

In the present study, we used task-evoked changes in pupil dilation to assess memory for temporal order in visual working memory. In two experiments, we assessed how stimulus type and spatial presentation affect working memory for the order of visual sequences. Working memory is thought to be capacity limited (Cowan, 2001; Luck & Vogel, 2013), but that capacity is not constant within an individual. Instead, the capacity of working memory fluctuates depending on the nature of the to-be remembered items. Visual working memory, for example, is influenced by the type of image being maintained: a greater number of simple stimuli can be remembered compared to more complex stimuli, and semantically meaningful items are more easily remembered than abstract items (Alvarez & Cavanagh, 2004; Brady et al., 2016; Starr et al., 2020). In Experiment 1, we build off of these prior findings by asking whether the benefits found for semantically meaningful arrays of items also extend to memory for the temporal order of those items when they are presented sequentially.

A second aim of the present study was to investigate whether visual working memory capacity for the temporal order of items in a sequence can be enhanced through the use of spatial cues. Specifically, we presented items in spatial sequences that were either congruent or incongruent with the mental timeline. The mental timeline is mental model of time based off of a linear reference frame (Bender & Beller, 2014; Bonato et al., 2012). In Western cultures, for example, time is mapped onto a horizontal axis, frequently with a left-toright orientation such that earlier events are represented on the left and later events are represented on the right. This spatial organization of temporal order is automatically activated during both verbal and visual working memory span tasks (Guida et al., 2016, 2018; van Dijck & Fias, 2011). For example, van Dijck and Fias (2011) had participants remember lists of numbers or foods and then use their right and left hands to make category judgments (parity for numbers and fruit versus vegetable for foods) about the list items. Participants were faster when the left was used to respond to items presented earlier in the list and the right hand was used to respond to items later in the last, in comparison to when the opposite hand was used (e.g., responding with the left hand for items later in the list). This effect, which is similar to the classic SNARC effect for spatializing numerical magnitudes (Dehaene et al., 1993), suggests that individuals are spontaneously mapping items presented earlier in the list to the left side of space and items presented later in the list to the right side of space. However, these studies have focused specifically on the spatial-positional association of the response codes themselves - in other words, how temporal position within the sequence affects the speed with which responses are made with the left versus right hand. If these effects are not specific to the response codes but instead reflect a spontaneous mental organization of the items in the sequence from left to right, then we might expect that presenting the sequence of items in spatial positions congruent with the mental organization of temporal order could strengthen memory for temporal order.

In line with this hypothesis, recent work suggests that adults draw on the mental timeline to remember the temporal order of events in episodic memory. Pathman et al. (2018) found that there is a strong memory advantage for temporal order when the items are presented in a left-to-right spatial pattern compared to a right-to-left or nonlinear pattern. Specifically, participants were better able to remember the temporal order in which items were presented when those items were presented spatially from left-to-right. Pathman et al. theorize that temporal memory is facilitated when the spatial locations and temporal order of the items within a sequence are congruent with the mental timeline and negatively affected when the locations and order are incongruent. The paradigm used by Pathman et al. (2018) tested the effects of spatial positioning on episodic memory for temporal order, and it remains to be seen whether a similar benefit is also present for working memory.

The present study had three primary goals. The first was to assess the use of pupillometry as a measure of visual working memory processes in a visual span task. The second goal was to assess how the semantic content of images affects memory for temporal order. In Experiment 1, participants were shown a sequence of images one at a time, presented centrally, and then shown all of the images and asked to reorder them to match the temporal order of presentation. We manipulated the content of the images such that half of the images were semantically meaningful (line drawings of common objects), and half of the objects lacked semantic content (abstract line drawings). In this task, participants did not need to remember the identity of the items presented in the sequence, only the order in which they were presented. If pupil dilation tracks with the exertion of mental effort to encode new items into working memory, then we predicted that pupil dilation would increase for each sequentially presented image that is being attended to and would then flatten off once capacity is

reached (Granholm et al., 1996; Johnson et al., 2014). Because visual working memory capacity is greater for images with semantic content compared to without semantic content, we also predicted that pupil dilation would continue to increase for items later in the sequence for the semantic image trials compared to the abstract image trials. The third goal was to assess whether working memory capacity for temporal order can be increased by presenting the sequence of images in spatial positions consistent with the mental timeline. In Experiment 2, we compared memory for temporal order when sequences were presented from left-toright compared to when the sequences were presented from right-to-left. Because left-to-right sequences are privileged in episodic memory (Pathman et al., 2018), we predicted both better memory and greater pupil dilation for sequences presented from left-to-right compared to right-to-left.

Experiment 1

In Experiment 1, we tested participants' visual working memory spans for sequences of semantic and abstract images while measuring task-evoked pupil dilation.

Method

Participants Data from 20 participants were included in the final analyses. Data from one additional participant were excluded due to insufficient pupillometry data (see Pupil Data Analyses). Participants were recruited from the psychology research pool at the University of Washington and were compensated with course extra credit. All participants provided informed consent to a protocol approved by the local IRB.

Task Participants were told that they would view a sequence of images and would then be asked to recreate the temporal order of the images. The experiment began with a practice trial with a sequence of four images presented in the center of the screen. After viewing the sequence, participants were handed an iPad that showed the images in a random order in a vertical array and were told to rearrange the images by dragging and dropping them into the order they had been shown on the screen, with the first image on top and the last image on the bottom. After the practice trial, participants completed eight test trials with sequences of nine images. Four trials contained semantic images and four contained abstract images (Figure 1), and the different trial types were presented in an alternating order. The semantic images consisted of common objects and animals, and the abstract images consisted of random line arrangements. All images consisted of white lines on a black background.

Each trial began with a 3000 ms fixation cross presented in white on a black background. Each image within a trial was presented for 500 ms and was followed by a 3000 ms intertrial interval with a fixation cross. After the final image in each sequence, participants were shown a vertical array of the images in a random order and were asked to reorder the images to match the presentation order from top to bottom, with the first image in the sequence on the top and the final image on the bottom. No feedback was provided.



Figure 1: Example stimuli used in the semantic (top) and abstract (bottom) image conditions.

Procedure Participants were tested individually in a quiet, dim room and were seated approximately 65 cm from the monitor with their heads unrestrained. Pupil data were continuously measured using an EyeLink 1000 eye tracker (SR Research) at a frequency of 500 Hz. Prior to the practice phase of the task, participants completed a five-point calibration, which was occasionally repeated between test trials if participants moved out of the field of view of the eye tracker. The entire experiment took approximately 30 minutes.

Pupil Data Analysis Pupil data were cleaned and analyzed in R using the PupillometryR package (Forbes, 2020) and self-developed analysis scripts. Pupil data were first downsampled to 50 ms time bins, outlier data points (e.g., those corresponding to blinks) were filtered out using a hanning filter, and resulting gaps were linearly interpolated. Trials that were missing more than 33% of the data were removed (n = 9), and participants missing more than 50% of trials were removed (n = 1). Our primary time period of interest for measuring pupil dilation was the intertrial fixation period following each image. For each trial and each participant, we calculated a baseline measure of pupil dilation that corresponded to the average pupil dilation during the final 200 ms of the fixation period prior to the first image in the sequence. Trials with baseline pupil dilations more than two standard deviations away from that individual participant's mean baseline pupil dilation were excluded (n = 2). We then calculated the relative change in pupil dilation during the fixation period after each image relative to the initial baseline fixation period by subtracting the baseline pupil dilation from the measured pupil dilation for each timepoint within the 3000 ms intertrial fixation period (Mathôt & Vilotijević, 2022).

Results

Memory accuracy We analyzed the number of images per trial that participants placed in the correct temporal position using a mixed effects model with trial type (abstract or semantic) as a fixed effect and participant as a random effect. This model revealed a significant effect of trial type ($\beta = 3.34$, p < .001). Participants remembered the position of significantly more images in the semantic condition compared to the abstract condition ($M_{SEM} = 6.55$, $SEM_{SEM} = .22$; $M_{ABS} = 3.21$, $SEM_{ABS} = .22$).

Pupil dilation To assess how pupil dilation changed in response to each image in the sequences, we conducted a mixed effects model with the maximum relative pupil dilation as the dependent variable, trial type, image number, and their interaction as fixed effects, and participant as a random effect. This model revealed significant main effects of both trial type and image number ($\beta_{\text{TRIALTYPE}} = 25.65, p <$.001; $\beta_{\text{IMAGE}} = 12.31$, p < .001), as well as a significant interaction ($\beta = -7.12$, p < .001; Figure 2). Post-hoc paired ttests controlling for multiple comparisons indicated no significant difference in pupil dilation between the semantic and abstract conditions for images 1-5 (ts (83,85) < .847, ps > .398), but pupil dilation was significantly greater for semantic compared to abstract images for images 6-9 (ts (83,85) > 2.02, ps < .045). These results indicate that when a greater number of images can be held in working memory, as in the case of the semantic images, maximum pupil dilation is greater overall compared to when fewer images can be maintained in working memory, as in the case of the abstract images. In addition, when the temporal position of a greater number of images can be successfully remembered, pupil dilation continues to increase for images later in the sequence.



Figure 2: Maximum pupil dilation during the fixation period following each image. Error bars indicate SEM.

Experiment 2

In Experiment 2, we explored the effects of presenting images from left-to-right compared to right-to-left. If participants are activating the mental timeline to support visual working memory, then we would expect participants to correctly remember the order of more images in the leftto-right compared to right-to-left presentation condition.

Method

Participants Data from 40 participants were included in the analyses (left-to-right condition: n = 20, right-to-left condition: n = 20), none of whom participated in Experiment 1. Data from an additional three participants were excluded due to insufficient pupil data (see Pupil Data Analysis). Participants were recruited from the psychology research pool at the University of Washington and were compensated with course extra credit. All participants provided informed consent to a protocol approved by the local IRB.

Task The task was the same as in Experiment 1 with the exception that participants were randomly assigned to the left-to-right linear presentation condition (the first image appeared on the left of the screen and following images were presented left-to-right across the screen), or the right-to-left linear condition (the first image appeared on the right side of the screen and following images were presented right-to-left).

Procedure The procedure was the same as in Experiment 1.

Pupil Data Analysis Pupil data were cleaned and analyzed using the same procedures as in Experiment 1. Twenty-two trials were excluded because they were missing more than 33% of the data, two trials were excluded for having baseline pupil dilations more than two standard deviations from that individual participant's mean baseline pupil dilation, and three participants were excluded for not having valid pupil data for at least 50% of trials.

Results

Memory accuracy We analyzed the number of images per trial that participants placed in the correct temporal order using a mixed effects model with direction (left-to-right or right-to-left), trial type (abstract or semantic), and their interaction as fixed effects, and participant as a random effect. This model revealed a significant effect of trial type $(\beta_{\text{TRIALTYPE}} = 2.43, p < .001)$. Neither the effect of direction $(\beta_{\text{DIRECTION}} = 0.30, p = .56)$ nor the interaction term $(\beta_{\text{INTERACTION}} = -0.23, p = .57)$ were significant. Participants remembered the position of significantly more images in the semantic condition compared to the abstract condition (M_{SEM} = 6.34, SEM_{SEM} = .14; M_{ABS} = 3.66, SEM_{ABS} = .14), but not significantly more images in left-to-right condition compared to the right-to-left condition ($M_{LR} = 5.00$, $SEM_{LR} = .19$; M_{RL} = 5.19, SEM_{RL} = .20). In contrast to our predictions, presenting the images from left-to-right did not improve participants' memory for the order of the images relative to presenting the images from right-to-left (Figure 3).

Pupil dilation We conducted a mixed effects model with the maximum relative pupil dilation as the dependent variable, direction, trial type, image number, and their interactions as fixed effects, and participant as a random effect. This model revealed significant effects of trial type and image number ($\beta_{\text{TRIALTYPE}} = 38.58 \ p < .001$; $\beta_{\text{IMAGE}} = 16.62, \ p < .001$), but

no significant effect of direction ($\beta_{\text{DIRECTION}} = 62.03, p = .211$; Figure 4). In addition, the two-way and three-way interaction terms were all significant (all ($\beta_s > 6.07, ps < .001$).



trialtype 🖨 abstract 😫 semantic







In response to the significant three-way interaction between direction, trial type, and image number, we performed follow-up comparisons of the effects of presentation direction on pupil dilation for each image in the sequence separately for semantic and abstract image trials. For trials with semantic images, there was no significant difference in pupil dilation between the left-to-right and right-to-left conditions for images 1-6. Beginning with image 7, the maximum pupil dilation in the left-to-right condition was larger than in the right-to-left condition (*ts* (75,72) > 1.99, ps = .054). For trials with abstract images, however, there was no significant difference in pupil dilation between the conditions for any of the images (*ts* (77,75) < 1.29, ps > .20). Therefore, presentation direction affected maximum pupil dilation in response to the semantic image trials, but not for the abstract image trials.

General Discussion

In the present study, we used task-evoked pupil dilation to assess visual working memory for the temporal order of visual sequences. Across two experiments, we found that pupil dilation increased following the sequential presentation of additional items to be maintained in working memory. In addition, we found that this increase in pupil dilation varied as a function of the type and spatial position of the visual information to be remembered.

In both Experiment 1 and Experiment 2, we found that participants were able to correctly remember the temporal order of more items when those items were semantically meaningful compared to when they were abstract line drawings devoid of semantic content. This finding is consistent with prior work demonstrating that visual working memory capacity is greater for items for which individuals associate semantic meaning (Alvarez & Cavanagh, 2004; Asp et al., 2021; Brady et al., 2016; Starr et al., 2020). Notably, the present findings demonstrate that the mnemonic benefit for semantically meaningful objects extends beyond remembering the identity of those objects to additionally remember the temporal order in which those objects were presented.

In parallel with the behavioral memory performance, in Experiment 1 we found that maximum pupil dilation was greater in response to semantic images compared to abstract images. This effect was driven by images later in the sequence - there was no significant difference in pupil dilation between the image types for images in the beginning of the sequence. Whereas pupil dilation began to flatten out around image five in sequences of abstract images, it continued to increase for sequences of semantic images. This pattern of results suggests that participants remained engaged and were willing to expend greater cognitive effort for sequences of semantic images, which may have contributed to the memory benefit for semantic versus abstract images. In addition, this pattern of results is consistent with previous findings that demonstrate that pupil dilation tracks with engagement while sequentially encoding items into working memory (Johnson et al., 2014; Klingner et al., 2011). However, in previous studies that used pupillometry to assess working memory for simultaneously presented arrays, pupil dilation has been influenced by both the number of items and the type of item being maintained in working memory. Specifically, greater mental effort, as indexed by greater pupil dilation, is required for larger arrays and for arrays of ambiguous stimuli in comparison to smaller arrays and arrays of stimuli that can be easily categorized (i.e., ambiguous

versus prototypical colors; Zhou et al., 2022). Additional work is therefore needed to clarify how pupil dilation is affected by stimulus type and memory load in simultaneous versus sequentially presented working memory tasks.

In Experiment 2, we tested whether the presentation of sequences of images from left-to-right, in spatial positions consistent with the mental timeline, could improve working memory capacity for temporal order. In contrast to our predictions, we found no difference in memory performance for sequences presented left-to-right compared to sequences presented right-to-left or to sequences presented centrally. We also found no interaction between presentation direction and image type, which indicates that even the more difficult abstract image sequences did not benefit from being presented from left-to-right.

This pattern of results contrasts with findings from a recent study that found improved episodic memory for temporal order when sequences were presented from left-toright (Pathman et al., 2018). Although one interpretation of the present results is that participants do not spontaneously activate the mental timeline when encoding the temporal order of sequences into working memory, it is also possible that differences in task structure beyond the type of memory assessed contributed to these conflicting findings. In the Pathman et al. (2018) study, when participants were prompted to remember the temporal position of the items, the response options were presented from left-to-right. This means that for items initially presented from left-to-right, the temporal order options were congruent with the initial presentation order. In the present study, we eliminated this confound by prompting participants to order the images from top-to-bottom, such that neither of the presentation directions was consistent with the temporal ordering task. In addition, image sequences in the Pathman et al. (2018) study consisted of just three items, which may have prompted participants to encode the ordinal position of the items (e.g., first, second, third). However, the present study used sequences of nine images, a span length that may exceed visual working memory capacity, which may have discouraged participants from encoding the ordinal position and instead encouraged participants to engage in alternative mnemonic strategies (e.g., attempting to create a story to link the semantic images). Furthermore. our memory prompt was not to recall the ordinal position but to recreate the temporal order in a simultaneously presented visual array. This prompt isolates memory for temporal order and eliminates the need to recall the identity of the images themselves. It is possible that we would have observed different patterns of memory performance if we had asked participants to recall the specific ordinal location of the images or required them to recall the identity of the images in addition to their temporal order. Additional work is therefore needed to better understand the situations when presenting information in a spatial manner congruent with the mental timeline does and does not benefit memory.

Although we found no effects of presentation direction on participants' explicit memory responses, pupil dilation did exhibit some sensitivity to this manipulation. Specifically, maximum pupil dilation was greater for semantic images compared to abstract images, and this main effect was driven by an interaction with presentation direction such that pupil dilation was greater for semantic images presented left-toright compared to semantic images presented right-to-left. In other words, patterns of pupil dilation differed by image type when the sequences were presented centrally or from left-toright, but not when the images were presented from right-toleft. As in Experiment 1, this pattern was driven by differences in pupil dilation for images towards the end of the sequence. This pattern of results suggests that participants in the left-to-right condition may have remained engaged for images in the semantic sequences even once they had reached their working memory capacity, whereas participants in the right-to-left condition disengaged earlier in the sequence. Unfortunately, our study may have been underpowered to detect true differences in either behavior or pupil dilation, leading to null effects that are difficult to interpret. However, the mismatch between the behavioral and pupillary responses suggests that pupil dilation may be sensitive to subtle differences in working memory processes across conditions that are not apparent in behavioral responses.

Taken together, the present results highlight the utility of using pupillometry for studying working memory span processes using stimuli other than digits. In particular, pupillometry may be a beneficial tool for investigating working memory processes in young children and infants who are unable to provide explicit behavioral responses. Our results also provide new insights into how the nature of the items to be remembered and the spatial positions of those items influence visual working memory for temporal order.

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