Does stimulus location prime the retrieval of information from memory? A pupillometry study.

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Abstract

The constriction or dilation of the pupil following changes in brightness is referred to as the pupil light response. In the past, the pupil response was believed to be based on an exclusively reflexive mechanism. However, recent research shows that it may also be affected by higher cognitive functions relating to visual working memory and attention. The current study tested whether the location of a stimulus can prime retrieval of information from memory, using pupillometry measures. The task consisted of a stimulus array, where a bright and a dark grated circle were presented on the screen. Following, the stimuli were removed from the screen and the participants made eye movements to the previous locations of the stimuli following a fixation point. The participants' pupil size was recorded when they made saccades to the dark and bright locations. The trial ended with a memory task, which kept the participants engaged in the experiment. Upon visual inspection of the data, it could be seen that for the first saccade the pupil constricted more when the movement was made to a bright location compared to a dark location. Tests for this effect did not reach significance, however. The trend observed in the pupil traces may still be meaningful, therefore more research in this area is suggested.

Keywords: pupil light response, pupil sides, saccades, visual working memory

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Instinctively, when one thinks about studying or testing memory, explicit report comes to mind. For example, giving the person a string of letters to remember, and having them say the string back to you, or showing them a series of pictures and later asking them which pictures they recognise. And that is true – memory is very often studied through verbal or written reports (directly asking someone whether or what they remember). This approach, however (similarly to any method which uses self-report), can be prone to bias. An example could be demand characteristics, a bias which refers to participants interpreting the goal of an experiment and adapting their behaviour to what they believe is desirable (Morling, 2015). An alternative approach that can mitigate this bias when testing memory is tracking eye movements, which has been found useful for studying memory without explicit report (Ryan et al., 2010). This can be studied by looking at scanning patterns of new and known images. For example, different movement patterns when scanning an identical and a changed version of an image seen before can reflect the consolidation of the changed detail into memory. This method shows that we can evaluate what has already been encoded in memory by means of tracking eye movements, but recent research also shows that eye movements may facilitate encoding items into memory.

A study by Hanning et al. (2016) examined the importance of task-relevance and oculomotor selection. In one task, locations were cued as either target locations or non-target locations. The participants were to make a saccade to the target location. It was found that the object that was encoded as a saccade target was successfully maintained in working memory, which did not happen for locations which were not saccade targets. An advantage was also found in situations when a saccade was prepared, but the eye movement was not performed. In the second task of the experiment, the location of stimuli was cued as task relevant, but no saccade was to be made to that location. In this task, no working memory advantage was found. The findings of this study indicate that oculomotor selection is related to working memory, where task relevance does not seem to provide a benefit for working memory encoding. Medimorec et al. (2021) also studied the role of eye movements in memory encoding, more specifically, they studied the mechanism behind implicit sequence learning. The study compared sequence learning in the absence and presence of motor responses during learning. They used serial reaction time (SRT) tasks, and the tasks were divided into O-SRT tasks, where no motor restrictions were imposed, and F-SRT tasks, where participants were restricted from making eye movements. The researchers made use of anticipation measures to evaluate performance on the tasks. It was found that performance on the SRT task was higher when there were no motor response restrictions imposed on the participants. The results suggest that eye movements seem to be necessary for sequence learning.

So far, the relevance of eye movements in regard to memory encoding was discussed. However, there is another type of eye movement that may be important in studying memory – changes in pupil dilation. Most people are familiar with the pupil light response, or PLR (when the pupil constricts in response to increased luminance, and dilates when it is darker) and the pupil near response, or PNR (the pupil constricting when looking at a near object, and dilating when looking at a far object). However, there is yet another pupil response – the psychosensory pupil response. The psychosensory reflex happens in response to an arousing stimulus, thought, or emotion. In response to such stimuli, the pupil tends to dilate. However, why the pupil dilates in response to such a broad variety of stimuli as, for instance, sounds is still unclear (Mathot et al., 2018).

Recent findings show that there is more complexity to the PLR than previously believed. For example, a study by Mathot et al. (2015) found that the preparation of the PLR happens simultaneously to the preparation of a saccade toward a stimulus. It was found that even before the participants moved their eyes, the pupil already responded with a constriction when the participants' attention was directed to the bright side of the screen. The results show that the PLR can be induced even when a stimulus is only covertly attended, indicating that the PLR may reflect visual awareness, not only stimulus brightness. The findings suggest that the pupil already accommodates to the target location brightness, even before the saccade is made. Another study that examined the PLR in the context of spatial attention was one by Binda and colleagues (Binda et al., 2015). In this study the researchers measured the pupil size of participants while manipulating the brightness of the display and the location of attention. It was found that pupil size changes were dependent on where the attention was oriented. The findings of this study indicate that changes in pupil size do not only reflect reflexive functions, but on top of that, the changes may be related to more complex processes such as attention.

Further research of the PLR, studying patients with Parinaud's syndrome, found that the PLR in response to actual changes in the luminance of the visual field and the PLR related to visual attention may have different underlying pathways (Binda et al., 2017). Patients with Parinaud's syndrome have an impaired PLR, while other pupil responses remain intact. In the experiment, both patients and a control group were shown a bright and a dark circle presented on either the left or right side of a fixation dot. The participants were cued to covertly attend to either the left or right stimulus. Strikingly, both in the patient group and the control group, the pupil was more constricted when the participants attended to the brighter stimulus. The results show that there are different pathways that underlie the PLR, a main pathway (which is often depleted in patients with Parinaud's syndrome), and a secondary pathway which avoids the depleted area and still influences pupil size.

Recent literature shows that the PLR may also be related to visual memory. Husta et al. (2019) used the PLR to study the association between the sensory brain areas and visual working memory. In their study, participants covertly attended to, and were asked to memorize either bright or dark stimuli. The researchers found a smaller pupil size when a cue presented before the stimulus instructed the participants to memorise the upcoming bright stimulus. Moreover, they found that visual working memory content was reflected in the PLR during encoding as well as during maintenance in memory. The results of this study indicate that the PLR is connected to higher cognitive processes and is more than just a reflexive response. Hoffing et al. (2015) also studied memory from a pupillometry perspective. They used a learning task which included targets and distractors. They found that the participants performed better in recognising stimuli that were paired with targets compared to the distractors. In connection, they found that the pupil size showed greater changes for the stimulus paired with the target than with the distractor. Those results indicate that changes in pupil size are related to increased encoding of images into memory. Another recent study examined fluctuations in working memory task performance and the related changes in pupil size (Robinson et al., 2018). The results showed that changes in pupil size can also reflect fluctuations in working memory performance tasks.

Taken together, the recent literature shows that both saccadic eye movements and pupillary responses appear to be related in some way to memory processes. In the current study pupil size is measured as the participant makes eye movements to the locations where the stimuli were previously presented to investigate, from a pupillometry perspective, whether the location of a stimulus can prime retrieval of information about that stimulus from memory.

Methods

Participants

The participants of this study were first-year Psychology students at the University of Groningen. The study included 35 participants, 68% of which were female and 32% were male. All participants had normal or corrected-to-normal vision. Participants were recruited using SONA Systems, an on-line participant management tool. Every participant gave written informed consent to their participation. Completing the experiment was compensated with 2.4 SONA credits.

The study was approved by the Ethics Committee of the Faculty of Behavioural and Social Sciences of the University of Groningen (PSY-2122-S-0270).

Materials

The experiment was a behavioural task created in OpenSesame (Mathôt et al., 2012). Apart from behavioural data, we also collected pupil size data using the GP3 Eye-Tracker (Gazepoint, 2021). The experiment was presented on an iiyama MA203DT-D 22" monitor. The participants were seated at about 60 cm from the screen.

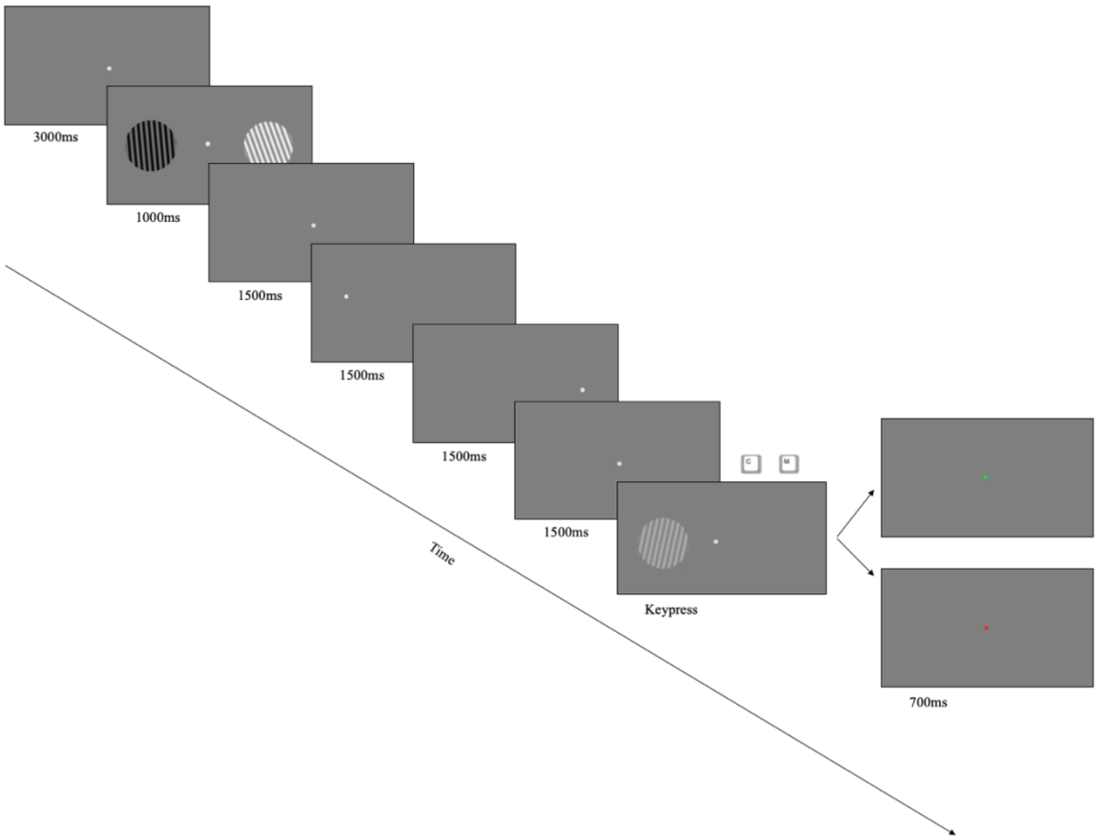
The stimuli used in the experiment were grated circles of different brightnesses: bright and dark grated circles (with a size of 96 pixels) were used as the initial stimuli and a grey grated circle was used as the test stimulus (for an example of the stimuli please refer to the visualisation of the trial sequence in Figure 1.). The circles also differed in the angle of the grating. The angle of the bright circle was randomly chosen from the range of -40 to 40 degrees. The angle of dark circle gratings differed from the bright circle's angle with a randomly selected number ranging from -20 to 20 degrees. The angle of the grey circle (and the difficulty of the task) depended on the participant's accuracy on previous trials. This was achieved using the two-down one-up staircase procedure. This method was used in order to make the difficulty of the task equal across participants.

Procedure

The participants began by familiarizing themselves with the instructions and study information. After having the chance to ask any questions regarding the study, the participants were asked to provide written informed consent. After giving informed consent, the experiment began with an instruction screen. This was followed by a set of 16 practice trials, which allowed the participants to familiarise themselves with the task. Upon completion of the practice trials, the experimental phase commenced, consisting of 16 blocks of 16 trials. The trials began with a fixation point located at the centre of the screen, which was presented for 3000ms. The purpose of this step was to collect information about the participants' baseline pupil size. The central fixation was followed by the presentation of a bright and a dark grated circle on the right and left side of the fixation point, lasting 1000ms. After the stimulus presentation, the retention interval began, and the participants were asked to make eye movements following a fixation point. After the retention interval, a grey grated circle was presented on either the right or left side of the fixation point. The participants were asked to determine whether the angle of the gratings on the circle is the same (by pressing the 'C' key) or different (by pressing the 'M' key) than the stimulus previously presented in that location. After giving a response, the participants were provided with brief visual feedback in the form of a fixation dot presented in the centre of the screen. A green fixation point indicated that the given response was correct, whereas a red fixation point indicated a wrong response. The feedback was presented for 700ms before the beginning of the next trial. A visual representation of the trial sequence can be found in Figure 1.

Figure 1

A visualisation of the trial sequence



The participants were given a break in the experiment after every block of 16 experimental trials. During the break, they were presented with their average accuracy and response time and had the chance to rest before the next block of trials. Following the completion of the experiment, the participants were provided with more details regarding the purpose of the study (upon request) and thanked for their participation.

Results

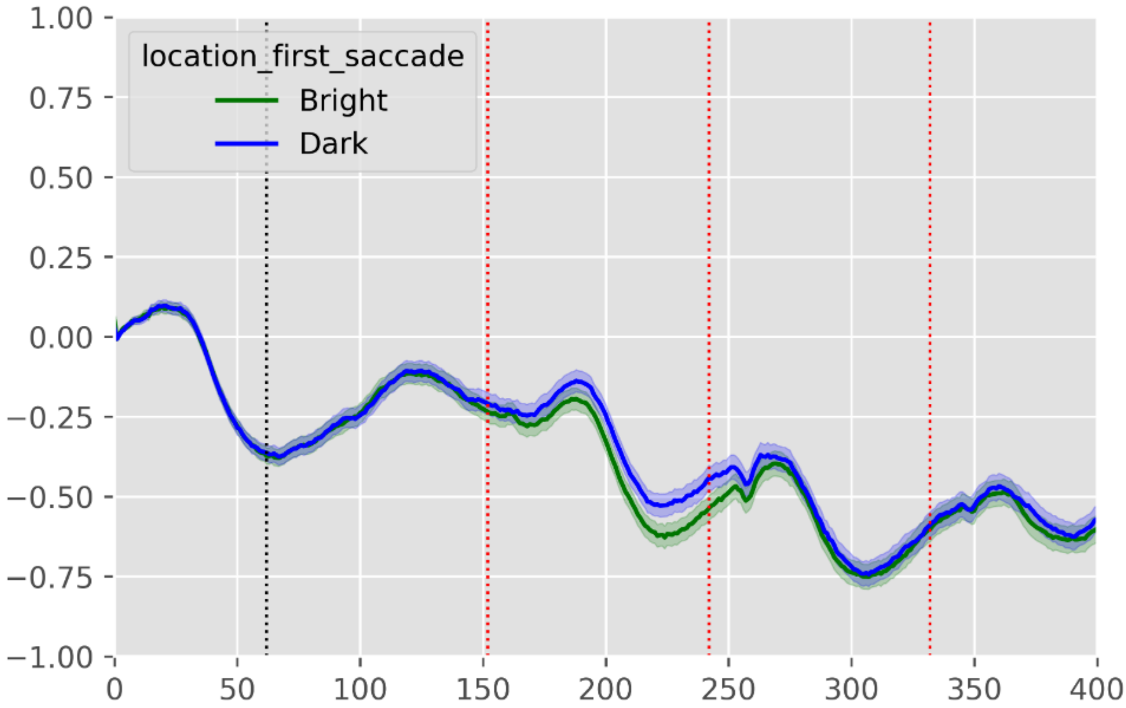
The aim of the study was to research, from a pupillometry perspective, whether the location of a previously presented stimulus can prime retrieval of information about that item from memory. Therefore, pupil size was used as the dependent variable, whereas the location of the fixation (location of a previously presented dark or bright stimulus) was used as the independent variable. Based on the findings of Husta et al. (2019), we expected the pupil to constrict more when the participants make a saccade to the location of a previously presented bright stimulus, and an inverse effect is assumed for the location of a dark stimulus. This was assessed by means of a time series test. In this test, the time series data is split into 4 subsets. The test selects one of the subsets, referred to as the test set. Then, a linear mixed effects model is conducted on each sample of the remainder of data, referred to as the training set. Ultimately, the sample in the training set which has the highest absolute *z* value will be tested against the test set (Mathôt & Vilotijević, 2022).

The participants also completed a memory task (a dummy task to keep them engaged in the experiment), but it would also be interesting to investigate the task performance. For exploratory purposes, the accuracy on the memory task was evaluated and compared across dark and bright stimuli using a paired t-test. The significance level of all tests was set to α = .05. The data analysis was carried out in Python 3 (Van Rossum & Drake, 2009). The Gazepoint Parser package was used for parsing the eye tracker data, and the Time Series Test package (Mathôt & Vilotijević, 2022) was used for the analysis.

Blinks and time points surrounding the blinks were detected in the pupil trace, and those observations were interpolated. After the exclusions, two timeframes of interest were selected: 154 samples – 242 samples for the first saccade, and 254 samples – 332 samples for the second saccade. 12 samples (or 200ms) at the beginning of each saccade were excluded from the intervals, since there is usually a latency of 200ms between when a target appears and when the participants begin to move their eyes (Purves et al., 2012).

Figure 2

The Pupil Traces for the First Saccade Made to the Bright and Dark Location



Note. The unit of the X axis is samples. Since the Gazepoint eye tracker is 60Hz, here 60 samples represent 1 second. The two grated circles were presented for the first 1 second (60 samples). The black dotted line indicates the moment when the stimuli were removed from the screen. The red dotted lines represent the moments when the participants began to make saccadic eye movements.

Figure 2 shows the pupil traces for the first saccade made to the bright and dark location. It can be seen that the traces overlap up until the point when the first eye movement is made (around 150 samples, or 2.5 seconds). When the first eye movement is made, the pupil seems to constrict more when the movement is to a bright location than to a dark location. This effect seems to diminish around the second saccade. It appears that a certain trend can be seen during the first saccade. However, when the statistical test was conducted using the time series test, the difference between pupil size when making a saccade to a previously bright and dark location was not statistically significant for neither the first saccade (z = 1.57, p = 0.12) nor the second saccade (z = 1.22, p = 0.22).

For exploratory purposes, we also tested the accuracy on the dummy task depending on whether the test appeared in the bright or dark location. However, the t-test revealed that there is no significant difference in accuracy depending on whether the bright or dark location was tested (t = 1.01, p = 0.31).

Discussion

Findings

The current study tested whether stimulus location can prime retrieval of information from memory. It was hypothesised that when the participants make an eye movement to the location of a previously presented bright stimulus, their pupils would constrict more compared to when making a saccade to the location of a dark stimulus. The study did not find a statistically significant difference in pupil size between the bright and dark location. This might be due to the fact that the participants were instructed to remember the angle of the grating on the circle and were informed that the colour of the stimuli is insignificant to the task. Ultimately, the participants may have only encoded the relevant aspect of the stimulus. Perhaps that is why the brightness was not encoded and therefore no effect was found. In the study by Husta et al. (2019) participants were just instructed to covertly attend to the bright and dark circles as a whole, whereas in the current study participants were asked to memorise only a certain attribute of the stimulus.

Thayer et al. (2022) studied whether certain attributes of an object or stimulus are maintained in memory as a whole object, or as separate features. In this study participants were instructed to memorise two objects which were different colour-shape combinations. The participants completed a search task during a retention interval. They were to find a target amongst distractors that were superimposed over certain colour-shape combinations. The study consisted of two conditions: in the first one, one of the search items was identical to one of the remembered stimuli in both colour and shape; in the second condition, one of the search items had the colour of one of the memorised stimuli and the shape of the other stimulus. Guidance of attention based on visual working memory was observed in both conditions.

However, no advantage was found for the first condition, where both features came from one object. The findings of Thayer et al. (2022) suggest that objects may be maintained in visual working memory as separate features rather than complete objects.

Another reason behind the current findings may be that there is a different mechanism underlying saccade planning and working memory. Wang et al. (2018) studied the difference in the effects of eye movement planning and working memory. In this study, participants were instructed to either make a saccade towards a visual target location or a memorised target location. During a delay period, a bright and a dark stimulus were presented. The researchers found that the pupil constricted more when the location of bright stimulus (compared to the dark stimulus) corresponded with the target location. However, the effect was significantly reduced when there was no dependence between the stimuli and the target locations. The findings of this study indicate that there might be different (although similar) mechanisms underlying eye movement preparation and working memory.

One more explanation for why no effect was found in the current study may be spontaneous pupil size fluctuations. Lowenstein et al. (1963) studied the relation between pupil size and the participants' alertness. They found that the pupil size of alert and awake participants was large and stable. However, as the participants became fatigued, their pupil size decreased and became less stable. Since the experiment used in the current study lasted up to 80 minutes, most participants became fatigued throughout, which might have affected pupil size.

One other reason for the lack of significant effect in this study may be power. As we saw in the pupil trace, there was a visible difference in the traces for the bright and dark location during the first saccade despite the results not being significant. Perhaps if the power of the study was increased (possibly by increasing sample size), significant results could be achieved.

Overall, there are many factors that may influence pupil size: eye movements, changing display, cognitive load on the memory task, fatigue, and tiredness. Taken together, all these factors may have been the reason for an insignificant effect. However, despite the effect not being statistically significant, a certain trend can still be seen in Figure 2, when the participants make the first saccade. The traces appear to overlap, but they diverge at the moment of the saccade. Based on that, we cannot say that there definitely is no difference. This taken together with previous studies by Husta et al. (2019) and Mathot et al. (2015) suggests that the PLR can be invoked without looking at a brighter or darker stimulus, and is related to attention and visual working memory. Therefore, further research in this area is needed.

Limitations and Future Research

The most significant drawback of the current study was the duration of the experiment. Eye tracking tasks tend to be tiring and dull to the participants. Researchers often use a dummy task, similar to the one used in this experiment, in order to keep the participants engaged. However, our experiment lasted about 80 minutes, which was met with reluctance from some subjects. Most participants left the lab very fatigued and tired, and had complaints about the duration and monotony of the experiment. One solution to this could be to limit the number of trials in the experiment, or present breaks more often. Another improvement could be changing the dummy task. Participants could use the mouse to change the grating instead of using keys on a keyboard to determine whether the grating was the same or different. Perhaps this more engaging task would work better.

Alternatively, if we suspect that no effect was found due to the fact that the participants selectively memorised only a certain aspect of the stimulus, the task could be updated. The gratings of the circles could be removed, the participants could be asked to explicitly remember the brightness, and then make eye movements.

Conclusions

In the current study we were interested in whether the location of an object or stimulus can prime the retrieval of information about that stimulus from memory. We measured participants' pupil sizes while they made eye movements to the locations of previously presented dark and bright circles. Although somewhat of a difference in pupil size between the bright and dark location could be observed in the pupil trace, the test revealed no statistical difference between pupil sizes. There are a few factors, such as attention or fatigue, which were not accounted for and may have influenced pupil size in this experiment. However, further research could reveal interesting new findings in this area.

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