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The Pupillary Light Response Reflects Encoding, but Not Maintenance, in Visual Working Memory

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The pupillary light response has been shown not to be a purely reflexive mechanism but to be sensitive to higher order perceptual processes, such as covert visual attention. In the present study we examined whether the pupillary light response is modulated by stimuli that are not physically present but are maintained in visual working memory. In all conditions, displays contained both bright and dark stimuli. Participants were instructed to covertly attend and encode either the bright or the dark stimuli, which then had to be maintained in visual working memory for a subsequent change-detection task. The pupil was smaller in response to encoding bright stimuli compared to dark stimuli. However, this effect did not sustain during the maintenance phase. This was the case even when brightness was directly relevant for the working memory task. These results reveal that the encoding of task-relevant and physically present information in visual working memory is reflected in the pupil. In contrast, the pupil is not sensitive to the maintenance of task-relevant but no longer visible stimuli. One interpretation of our results is that the pupil optimizes its size for perception of stimuli during encoding; however, once stimuli are no longer visible (during maintenance), an "optimal" pupil size no longer serves a purpose, and the pupil may therefore cease to reflect the brightness of the memorized stimuli.

Keywords: pupillometry, visual working memory, encoding, maintenance

Traditionally, the pupillary light response (PLR) has been thought to respond to the amount of light entering the eye (Loewenfeld, 1993). However, recent studies have shown that the PLR is not just a reflexive process but is also affected by cognitive factors (as reviewed in Mathôt & Van der Stigchel, 2015). For instance, stimuli that subjectively appear bright (e.g., the sun) trigger a constriction of the pupil compared to equiluminant stimuli that appear less bright (e.g., an indoor scene; Binda, Pereverzeva, & Murray, 2013a; Laeng & Endestad, 2012; Naber & Nakayama, 2013). Furthermore, the PLR has been shown to reflect the percept rather than the actual stimulus during episodes of binocular rivalry; that is, when a bright stimulus is presented to one eye and a dark stimulus to the other eye, the pupil constricts when the brighter stimulus dominates awareness (Naber, Frässle, & Einhäuser, 2011).

Moreover, a recent study by Laeng and Sulutvedt (2014) has suggested that the PLR is sensitive to mental imagery. Participants were first presented with a triangle that varied in luminance. Next, while the display remained blank, participants were asked to imagine the same triangle. Results showed that the pupil dilated or constricted in response to, respectively, dark and bright imagined objects. This finding was confirmed in a subsequent experiment, in which participants had to imagine familiar scenarios instead of triangles while looking at a blank screen.

In the present study, we examined the PLR when stimuli were not physically present but were kept in visual working memory (VWM). Given that the PLR appears sensitive to higher order perceptual representations, we wondered whether it is also sensitive to remembered stimuli rather than actual stimuli. VWM refers to the temporary storage and manipulation of visual information (Baddeley, 1992; Blankenship, 1938; Smith & Jonides, 1997). Theories of VWM generally distinguish between an encoding phase and a maintenance phase. Encoding occurs when a to-be-remembered object is selected in order to store its features. Maintenance happens when the object is no longer visible and its

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features have to be mentally rehearsed in order to not be forgotten (Cohen, Sreenivasan, & D'Esposito, 2014; Woodman & Vogel, 2005). Encoding and maintenance processes in VWM have been claimed to be two distinct processes (Baddeley, 1992; Woodman & Vogel, 2005). Here we assessed whether the PLR differentially responds to these stages or is sensitive to both of them. For this purpose, participants were first presented with a cue indicating which of two types of stimuli they had to encode and maintain: dark or bright stimuli. This was then followed by a retention interval, in which participants were required to maintain the stimuli in VWM for a subsequent change-detection task.

If the PLR reflects the interaction with only the actual stimulus, the pupil should be smaller for the brighter stimuli during the encoding phase when the stimuli are presented on the screen. But if the PLR also reflects the content of VWM, this difference in pupil size should persist during the retention interval when the stimuli have to be maintained. To make a distinction between the encoding and the maintenance phase, we included a control condition in which the stimuli only had to be encoded but did not have to be maintained. In the control condition, participants were presented with the same dark and bright stimuli but had to indicate whether a target, which was presented before the stimulus array, was present. Here the response was given after a time period that matched the retention interval in the memory condition. Any maintenance-related effects on the pupil in the memory condition should thus arise after the encoding phase as indicated by the control condition.

Experiment 1

In Experiment 1, we examined modulations of the PLR by stimuli that are not physically present but are kept in visual working memory (VWM). With the inclusion of the control (attention) condition, we could distinguish the PLR induced by the interaction with the actual stimulus during the encoding phase from the PLR during the maintenance phase. If the content of VWM were reflected in the PLR, the pupil should be smaller for brighter stimuli during the retention interval in the experimental condition than in the control condition, in which merely the presence, rather than the characteristics, of the stimulus had to be maintained.

Method

Participants. Fifteen observers (eight female) participated in Experiment 1.[1] Participants were recruited from Utrecht University. All participants were between 18 and 23 years of age and reported normal or corrected-to-normal vision. All participants signed informed consent before participating and received monetary compensation.

Apparatus and software. The left eye was recorded with an Eyelink 1,000 (SR Research, Mississauga, Ontario, Canada), a video-based eye tracker sampling at 1,000 Hz. Stimuli were presented on a 24-in. LG 24MB65PM monitor (1,920 × 1,200 px, 60 Hz). Stimulus presentation was controlled with OpenSesame (Mathôt, Schreij, & Theeuwes, 2012) and PsychoPy (Peirce, 2007). A chin rest was used to fixate the participant's head relative to the camera.

Procedure and stimuli. Before each experiment, a 9-point eye-tracker calibration was performed. Before each trial, a 1-point drift check was completed. Participants were instructed to keep their gaze at the central fixation point throughout the entire trial. All experiments consisted of a *visual working memory* (VWM) condition and an *attention* (ATT) condition, which were blocked and switched halfway through the experiment. The order of the conditions was counterbalanced between participants. The trial sequences are depicted in Figure 1.

In the VWM condition, participants remembered targets and performed a change-detection task after a retention interval. The targets were part of a memory array. The memory array consisted of four red (4.2°, 28.7 cd/m^2) and four green (4.2°, 45.9 cd/m^2) placeholders, which were arranged in a circle with an eccentricity of 6.5° around the fixation point. The bright and dark targets were always presented in alternation (bright, dark, bright, dark, etc.) and were equally spaced in a ring around the central fixation point. Observers were instructed to remember the shapes inside a specific placeholder color, either red or green, as indicated by a red or a green cue (1.2°). The number of trials in which one placeholder color contained dark shapes and the other placeholder color contained bright shapes was the same. This varied randomly from trial to trial but was fixed per trial. Thus, on a particular trial, observers would be remembering either all dark or all bright shapes. We used a color cue instead of directly instructing participants to remember the bright or black stimuli in order to deemphasize brightness in the task instructions. The shapes that had to be remembered consisted of eight different basic figures: pentagon, triangle, star, rhombus, square, cross, circle, and quatrefoil. All shapes were matched on surface area. The bright shapes had a luminance of 98.0 cd/m^2, and the dark shapes had a luminance of 0.13 cd/m^2. The memory array was followed by a retention interval of 4,000 ms. During the retention interval, participants kept the targets in memory for the following change-detection task. In the subsequent change-detection task, participants indicated whether all the targets were the same (*left arrow* key) or one of the targets had changed (*right arrow* key). In 50% of the trials, all targets were the same as during the encoding phase. The trial ended once the participant had responded.

The color of the targets, the color of the placeholders, and same or different in the change-detection task were randomized within blocks. The VWM condition consisted of 96 experimental trials (six blocks) preceded by 16 practice trials.

In the ATT condition, participants searched for one specific element within a search array. The ATT condition was a control for the VWM condition, because the same stimuli had to be only encoded and not maintained in memory. The search array in the ATT condition was the same as the memory array in the VWM condition and was preceded by the presentation of the to-be-searched target. The target (2.5°) was presented in the gray color of the background on either a red or a green placeholder (4.2°) for 1,000 ms. The color of the placeholder indicated within which figures of the search array the target had to be searched. The search array was presented for 3,000 ms and followed by a retention interval of 4,000 ms. During the retention interval, participants withheld their response to the presence of the target. Therefore, the

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[1] Participant data, as well as experimental and analyses scripts, are available from https://bitbucket.org/smathot/p0014.3-pupil-size-and-working memory-same-different/

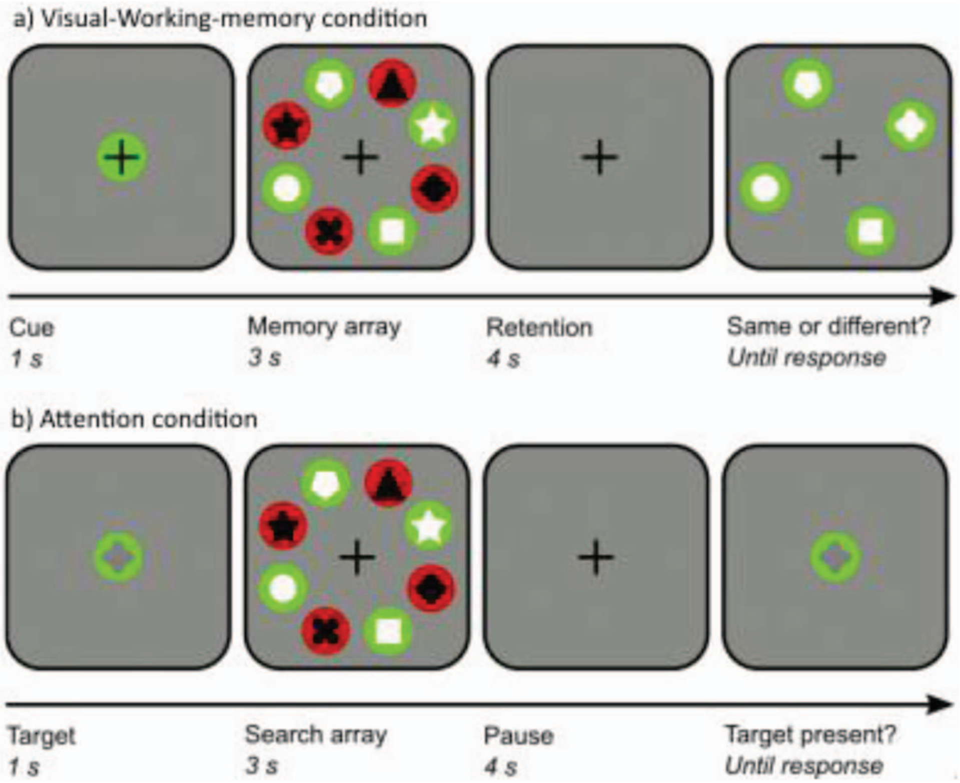

Figure 1. The trial sequence as presented in (a) the visual working memory (VWM) condition and (b) the attention (ATT) condition. In the VWM condition, participants were instructed to remember the shapes presented in the cued color. In the ATT condition, participants had to search for the target. See the online article for the color version of this figure.

trial progression of the ATT condition was similar to that of the VWM condition. The response was given during the second presentation of the target after the retention interval. The trial ended once the participant had indicated whether the target had been present (*left arrow* key) or absent (*right arrow* key).

The color of the targets, the color of the placeholders, and the presence or absence of the target in the search array were randomized within blocks. The ATT condition consisted of 96 experimental trials (six blocks) preceded by 16 practice trials.

Data analysis.

Significance and trial-exclusion criteria. For the linear mixed-effects (LME) analyses we used $t > 2$, which is comparable to $p < .05$ (Baayen, Davidson, & Bates, 2008). However, explicit p values have been omitted due to recent concerns about p value estimation for LME models. For the pupil-trace analysis, only sequences of at least 200 ms for which $t > 2$ were considered to be significant (Mathôt, Van der Linden, Grainger, & Vitu, 2013). Our analysis is similar to that in previous studies investigating modulations of the PLR (e.g., Mathôt, Dalmaijer, Grainger, & Van der Stigchel, 2014). Trials were excluded when, at any point after cue onset and before the response, participants fixated more than 2.4°[2] in a radius from the fixation dot. No other filtering criteria were applied.

Pupil-trace analyses. We analyzed pupil surface throughout the trial relative to a baseline period of 100 ms prior to the cue onset (cf. Mathôt et al., 2013). Cubic-spline interpolation was used for the reconstruction of blinks (Mathôt, 2013). To test for effects of target brightness (dark, bright) in each condition (visual working memory, attention), we conducted an LME with target brightness as fixed effect for each condition separately. Periods during which there was a significant pupil effect were based on these models. To test whether the effect of target brightness differed between conditions, we conducted an LME with target brightness, condition, and their interaction as fixed effects. In all cases, models included by-participant random intercepts and slopes for all fixed effects, used pupil size as dependent measure, and were conducted for each 10-ms window separately.

As can be seen in Figure 2, both the presentation of the cue and the search display triggered a pupillary constriction; these are visual responses to a change of visual input. In addition, there was a slow dilation throughout the trial, reflecting steadily increasing arousal. This pupil dilation relative to the start of the trial is noninformative because it indicates only that the participant paid attention but does not provide information about the content of VWM (Mathôt et al., 2014). Therefore, we focused on a constriction of the pupil on target-color-bright compared to target-color-dark trials, which from here on are referred to as the pupil effect.

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[2] A stricter criterion of, for example, a 1.2° threshold did not substantially change the results of the experiments.

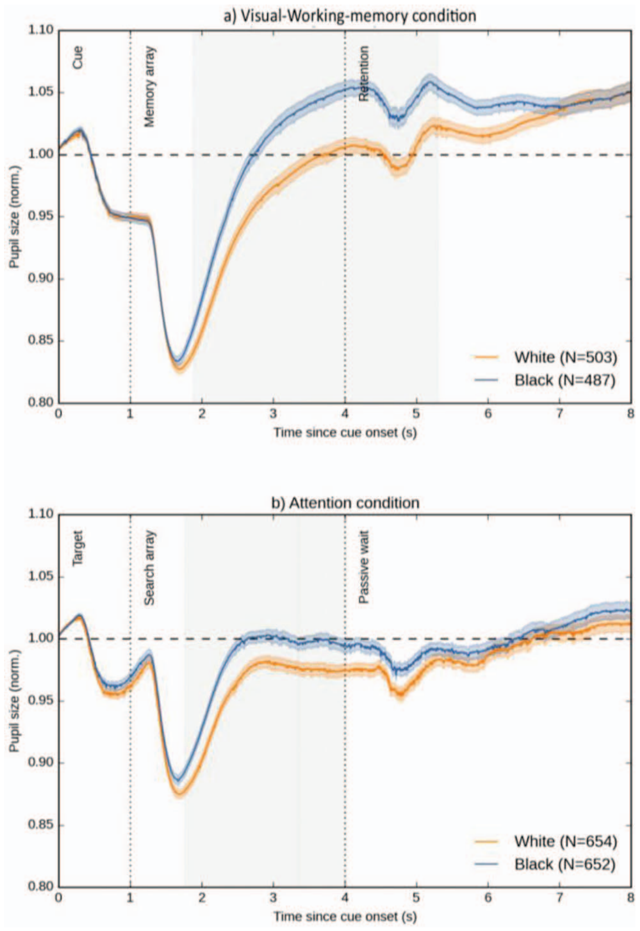

Figure 2. Pupil size as a function of stimulus color and time since cue onset for (a) the visual working memory condition and b) the attention condition. The lower of the two solid lines in each panel (orange line) portrays pupil size for bright stimuli, and the upper of the two solid lines in each panel (blue line) for dark stimuli. Data reflect the unsmoothed grand mean signal. The horizontal dashed lines indicate the baseline pupil size. Error shadings indicate standard errors. norm. = normalized. See the online article for the color version of this figure.

Phrased differently, we restrict our analyses to differences between conditions.

Results and Discussion

No participants were excluded, and after selection, 2,524 trials (87.6%) were entered into the analyses.

Behavioral data. In the VWM condition, the mean accuracy was 83% (*SD* 9%). In the ATT condition, the mean accuracy was 99% (*SD* 1%).

A paired-samples t test showed no significant difference in accuracy between bright and dark targets in the ATT condition, $t(14) = 0.225$, $p = .825$, or in the VWM condition, $t(14) = 0.245$, $p = .81$.

Pupil traces. The pupil responses for the VWM and ATT condition are shown in Figure 2. In the VWM condition, there was a significant pupil effect, such that the pupil was smaller for bright targets than dark targets, between 1,880 and 5,299 ms (i.e., all LMEs conducted within this window yielded a reliable effect of target brightness). In the ATT condition, the same pupil effect was evident during the interval 1,760–4,019 ms, with the exception of a brief period between 3,339 and 3,350 ms. There was no interaction between condition and target brightness (i.e., LMEs did not yield a reliable Target Brightness × Condition interaction for at least 200 consecutive milliseconds).

The results of Experiment 1 show that the pupil was smaller when bright rather than dark stimuli were encoded into working memory; however, this effect dissipated during VWM maintenance and was no longer present during the final part of the delay period. Although the pupil effect qualitatively persisted for some time during VWM maintenance, this was likely a lingering response stemming from encoding; that is, it took some time for the pupil effect to disappear. This interpretation is supported by the observation that the pupillary results of the VWM condition were similar to those of the ATT condition: In the ATT condition, participants had to remember the presence of the target, not the visual objects. Because there was also still a pupil effect at the start of the retention interval in the ATT condition, this effect cannot be explained by the content of VWM but is probably the aftermath of the encoding phase.

In Experiment 1, the stimuli were all canonical figures, and hence the stimuli were probably easy to verbalize. Therefore, the loss of the pupil effect during the maintenance phase could possibly be explained by the verbalization of the stimuli. In order for stimuli to be stored in VWM, they should not be verbalized. Within working memory, the phonological loop and the visuospatial sketchpad entail two different systems (Baddeley, 2003). If a stimulus is therefore verbalized, it will not be maintained in VWM. In Experiment 2 we tried to avoid the verbalization of stimuli.

Experiment 2

In Experiment 2 we used stimuli that were harder to verbalize. The task-relevant stimulus's feature for the change-detection task was no longer the shape, as in Experiment 1, but the orientation of a single shape. The orientation of the stimuli varied only slightly, which made verbalization harder, and hence we tried to ensure that visual (as opposed to verbal) working memory was used. The rest of the design stayed the same.

Method

Participants. Sixteen new observers (11 female) participated in Experiment 2.

Procedure and stimuli. In Experiment 2, the stimuli were squares that had been rotated in 11.25° steps, making a total of eight possible stimuli. Furthermore, to keep overall accuracy at around the same level as in Experiment 1, we had the memory array contain six instead of eight stimuli. Therefore, three stimuli had to be kept in VWM in the VWM condition. The memory and search array for Experiment 2 can be found in Figure 3.

Results and Discussion

No participants were excluded. After selection (using the same criteria as for Experiment 1), 2,320 trials (75.5%) were entered into the analyses.

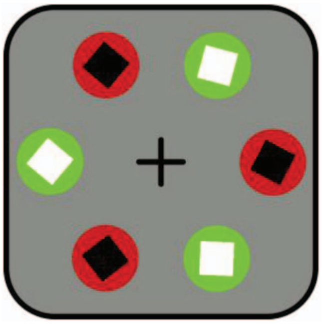

Figure 3. The memory array as presented in the visual working memory condition and the search array as presented in the attention condition consisted of a total of six squares that had been rotated in 11.25° steps. See the online article for the color version of this figure.

Behavioral data. In the VWM condition the mean accuracy was 66% (*SD* 7%). In the ATT condition the mean accuracy was 73% (*SD* 8%).

A paired-samples *t* test showed no significant difference in accuracy between bright and dark targets in the ATT condition, $t(15) = .554$, $p = .588$, nor in the VWM condition, $t(15) = .990$, $p = .338$.

Pupil traces. The results are qualitatively similar to those of Experiment 1. There was no significant effect of Target Brightness in the VWM condition. In the ATT condition there was a brief interval (2,220–2,419 ms) showing a significant effect of Target Brightness as shown in Figure 4. There was no interaction between Condition and Target Brightness.

In Experiment 2, we found no significant effect of stimulus brightness on the PLR in the VWM condition. Because there was also no pupil effect during the encoding phase, it is possible that the low amount of valid trials, especially in the VWM condition, left us with less statistical power to detect differences. But because the pupil traces of Experiment 2 resembled the pupil traces of Experiment 1, we tentatively concluded that it was not the verbalization of the stimuli that prevented a pupil effect during the maintenance phase. In Experiment 3 we examined the possibility that luminance has to be the task-relevant feature in order to evoke a PLR during the maintenance phase.

Experiment 3

In both Experiments 1 and 2, the brightness of the stimuli was not the task-relevant feature. Participants had to perform a change-detection task regarding the shape or orientation of the items and not their brightness. It has been suggested that VWM stores only those visual features that are relevant for subsequent behavior (Dehaene, Kerszberg, & Changeux, 1998). Because the brightness was irrelevant for subsequent behavior, it is possible that the brightness information was lost shortly after encoding and, with that loss, the pupil effect. In Experiment 3, the experimental design was adapted such that we used stimuli of different luminances. The participant's task was now to remember the specific luminance rather than the shape or orientation.

Method

Participants. Sixteen new observers (12 female, one author [Tessel Blom]) participated in Experiment 3.

Procedure and stimuli. In Experiment 3, the change-detection task involved the luminance of the stimuli. The memory and search array consisted of four circular stimuli. Two of these were more-or-less white (the bright targets) but differed slightly in their exact brightness; the other two were more-or-less black (the dark targets) but also differed slightly in brightness. There were three possible variations of white, two of which were randomly selected on each trial. Similarly, there were three possible variations of black, two of which were selected on each trial. In the VWM condition, participants had to keep the luminance of two stimuli in VWM and perform a change-detection task where in 50% of the trials one of the luminances had changed. The memory and search array for Experiment 3 are shown in Figure 5.

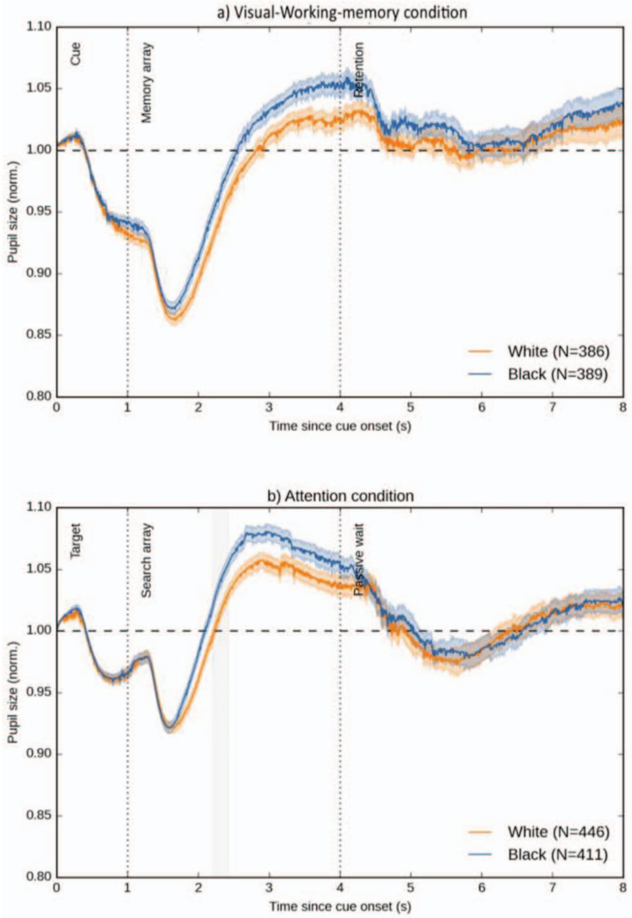

Figure 4. Pupil size as a function of stimulus color and time since cue onset for (a) the visual working memory condition and (b) the attention condition in Experiment 2. The lower of the two solid lines in each panel (orange line) portrays pupil size for bright stimuli, and the lower of the two solid lines in each panel (blue line) for dark stimuli. Data reflect the unsmoothed grand mean signal. The horizontal dashed lines indicate the baseline pupil size. Error shadings indicate standard errors. norm. = normalized. See the online article for the color version of this figure.

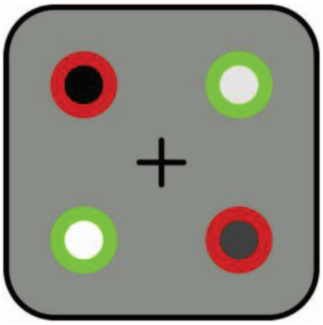

Figure 5. The memory array as presented in the visual working memory condition and the search array as presented in the attention condition, consisting of four circles with two light and two dark circles. See the online article for the color version of this figure.

Results and Discussion

No participants were excluded, and after selection, 2,411 trials (78.4%) were entered into the analyses.

Behavioral data. In the VWM condition the mean accuracy was 79% (*SD* 5%). In the ATT condition the mean accuracy was 74% (*SD* 10%).

A paired-samples *t* test showed a significant difference in accuracy for bright ($M = .75$, $SD = .055$) and dark ($M = .82$, $SD = .072$) targets in the VWM condition, $t(15) = 3.594$, $p = .003$, and a significant difference in accuracy for bright ($M = .69$, $SD = .12$) and dark ($M = .8$, $SD = .12$) targets in the ATT condition, $t(15) = 3.377$, $p = .004$. However, given the close similarity of the pupillary results of Experiment 3 (see the next section) to those of Experiments 1 and 2, we believe that the difference in accuracy for bright and dark targets did not strongly affect pupillary responses.

Pupil traces. Figure 6 shows the pupil responses for the VWM condition and the ATT condition. In the VWM condition, a significant pupil effect was found between 1,880 and 4,799 ms and between 4,820 and 5,069 ms and in the ATT condition between 310 and 4,659 ms and between 6,180 and 6,569 ms. In the ATT condition, the pupil effect already emerged when the target was presented, because the target contained a luminance to which the PLR responded (this was not the case in Experiments 1 and 2). This induced a strong interaction between condition and stimulus color. Crucially, this interaction was driven by the first 2 or 3 s during encoding and fully disappeared before the retention interval (4–8 s); again, we found no difference (i.e., no Target Brightness × Condition interaction) between the VWM and ATT conditions during this interval.

In Experiment 3, we again found that the pupil was smaller when bright, compared to dark, stimuli were encoded into working memory but that this effect dissipated during VWM maintenance. We concluded that the content of VWM was not reflected in the PLR.

Crossexperimental Analyses

Relationship Between the Pupil Effect and Behavioral Performance

We assumed that the pupil effect during encoding reflected how well stimuli were encoded into visual working memory. If so, then participants who showed a strong pupil effect should also have done well on the change-detection task. To test this, we determined the following per-participant measures: accuracy on the change-detection task and the pupil effect during the last 100 ms of the encoding interval in the VWM condition (during which the overall pupil effect was largest). The three experiments differed in their overall accuracy and pupil effect, which could have driven a correlation between both measures. To account for this, we conducted a multiple linear regression with pupil size as dependent measure and accuracy and experiment as predictors. This analysis showed a clear relationship between accuracy and pupil size, $t(43) = 2.8$, $p = .028$ (see Figure 7), that was not (fully) driven by overall differences between our three experiments. This confirms our assumption that the pupil effect during encoding reflects encoding of information into visual working memory.

Bayesian Analysis

Visual inspection of the results of Experiments 1 (see Figure 2), 2 (see Figure 4), and 3 (see Figure 6) reveals two key points: In the

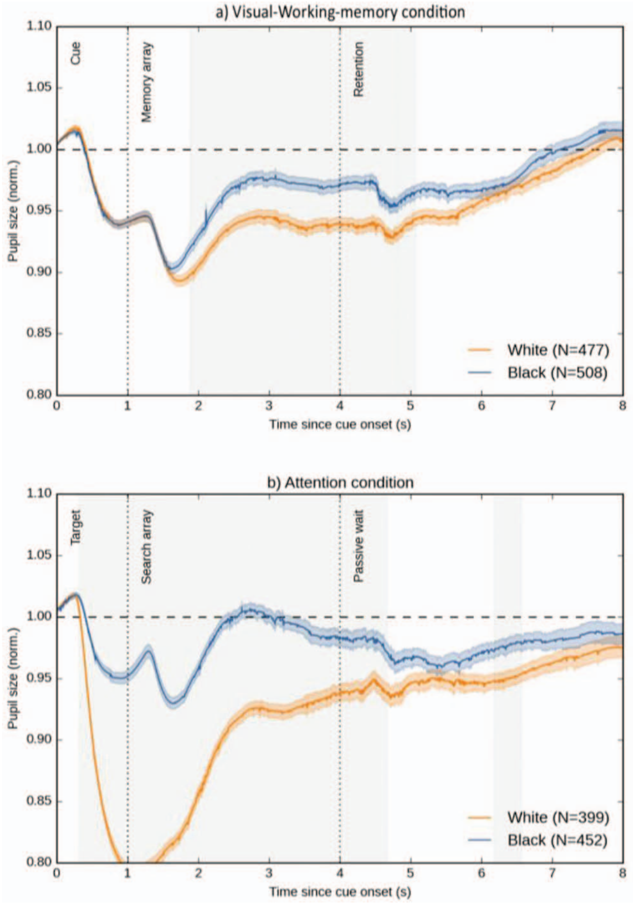

Figure 6. Pupil size as a function of stimulus color and time since cue onset for (a) the visual working memory condition and (b) the attention condition in Experiment 3. The lower of the two solid lines in each panel (orange line) portrays pupil size for bright stimuli, and the upper of the two solid lines in each panel (blue line) for dark stimuli. Data reflect the unsmoothed grand mean signal. The horizontal dashed lines indicate the baseline pupil size. Error shadings indicate standard errors. norm. = normalized. See the online article for the color version of this figure.

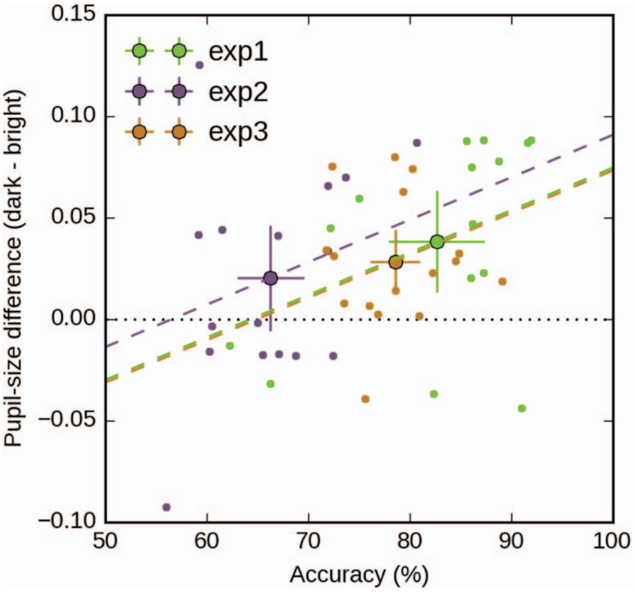

Figure 7. Accuracy on the change-detection task predicted the pupil effect during the last 100 ms of the encoding interval of the VWM condition. Small dots indicate individual participants. Large dots indicate estimated experiment means, with 95% confidence intervals (from left to right: Exp 2, Exp 3, Exp 1). Dashed (colored) lines indicate estimated regression slopes for the three experiments. (Estimates are parallel because our analysis did not include an Experiment × Accuracy interaction.) exp = experiment. See the online article for the color version of this figure.

VWM condition, the pupil effect built up during the encoding interval and dissipated during the maintenance interval; the time course of the pupil effect was similar between the VWM and ATT conditions (although overall pupil size differed somewhat). This suggests that encoding bright or dark stimuli affects pupil size, but maintaining these stimuli in working memory does not. To quantify the evidence for this conclusion, we conducted a Bayesian analysis using the following logic.

If maintaining bright or dark stimuli in working memory has no effect on pupil size, then the pupil effect in the VWM condition should be short-lived and thus similar to that in the ATT condition. Therefore, the pupil effect should be the same during the last 100 ms of the maintenance interval in the VWM condition (7.9–8.0 s) and the last 100 ms of the passive-wait interval in the ATT condition (7.9–8.0 s). To test this, we determined the per-participant pupil effect during these two intervals and compared them using a Bayesian paired-samples *T* test. This revealed moderate evidence against a difference (Bf = 0.169); that is, at the end of the trial, there was no difference in pupil effect between the VWM and ATT conditions.

In summary, a Bayesian analysis confirmed that our results best fit a model in which pupil size is driven by the brightness of stimuli during encoding, but not maintenance, of visual working memory.

General Discussion

In the present set of experiments, we examined whether the content of VWM is reflected in the pupillary light response (PLR). Because the PLR has been shown to be sensitive to higher order perceptual representations (Binda, Pereverzeva, & Murray, 2013b; Laeng & Endestad, 2012; Naber & Nakayama, 2013), we examined modulations of the PLR by stimuli that were not physically present but had to be kept in VWM. Participants covertly attended and encoded bright and dark stimuli, which had to be maintained in VWM for a subsequent change-detection task. As expected, the pupil was smaller when the bright stimuli had to be encoded compared to when the dark stimuli had to be encoded. This indicates that the encoding of information into visual working memory is reflected in the PLR. Interestingly, we observed a correlation between the accuracy on the change-detection task and the difference in pupil size between bright stimulus trials and dark stimulus trails during the encoding phase, where a larger pupil effect correlated with a higher accuracy. Many researchers have shown that an item must first be attended before it can be encoded into VWM (e.g., Mack & Rock, 1998), and because it has been previously suggested that the PLR can be used to track the focus of attention (Mathôt et al., 2014, 2013), our results are in line with these findings.

We further assessed whether the PLR differentially responds to encoding and maintenance of visual information. The pupil effect that emerged during the encoding phase did not sustain during the maintenance phase. This was consistent across all three experiments: whether it was the shape (Experiment 1), orientation (Experiment 2), or luminance (Experiment 3) of the stimulus that was relevant for subsequent behavior, the maintenance of the stimuli was not reflected in the PLR. A subsequent Bayesian analysis showed that pupil size was likely driven by the brightness of stimuli during encoding, but not maintenance, of visual working memory. We therefore conclude that the content of VWM is not reflected in the PLR.

Because the relation between working memory encoding and the PLR is strictly correlational, it cannot be determined whether the observed effect on the PLR is caused by the encoding in VWM or whether being encoded into VWM is the result of a modulation of the PLR. Indeed, when considering a possible explanation for the PRL modulation, it becomes evident that the PLR is not simply an epiphenomenon of encoding information in VWM. Because the optimal size of the pupil depends on how much light is available, we argue that the observed effects on the pupillary light response may serve to optimize the pupil size specifically for objects that need to be encoded in VWM. The pupil size is therefore tuned to the brightness of the to-be-encoded information, making the link between the pupillary light response and memory encoding beneficial. However, once stimuli are no longer visible (during maintenance), an "optimal" pupil size no longer serves a purpose, and the pupil may therefore cease to reflect the brightness of the memorized stimuli.

At first sight, the present paradigm is quite similar to paradigms used to investigate mental imagery: Both during imagery and during the maintenance of an object in visual working memory, the relevant object is not present on the screen and is represented only internally. Indeed, the two processes have been found to be related: Keogh and Pearson (2011) found that performance in visual working memory can predict the strength of mental imagery as assessed with binocular rivalry. Furthermore, individuals with strong mental imagery seem to use mental imagery as a mnemonic strategy for visual working memory tasks (Keogh & Pearson, 2014). The present findings suggest a possible dissociation between mental imagery and visual working memory with respect to the effect on the PRL: Whereas the PLR does appear to reflect the content of mental imagery (Laeng & Sulutvedt, 2014), the PLR is not modulated by the content of visual working memory. There are, however, many possible reasons for this difference. First, it might be that the timing of the two processes is different. The present interval was relatively short (i.e., 4 s), whereas mental imagery is generally assessed using longer intervals (e.g., Keogh & Pearson, 2014; Laeng & Sulutvedt, 2014). Although there was no such hint in our data, it could be that the effects of visual memory become apparent only with a longer interval. Second, it might be that the internal operations differ in the amount of mental effort that is required: Whereas visual memory does not require any computations on the internal representation, mental imagery is perhaps a more active process, requiring additional mental resources. Whatever the explanation, our results are reminiscent of the findings by Binda, Pereverzeva, and Murray (2014), who showed that knowledge that a task-relevant bright stimulus will appear was found be insufficient to cause pupil constriction, which occurs only when the stimulus is displayed and it is attended. Future studies could compare the effects of maintenance in visual working memory and imagery on the pupillary light response to directly investigate this apparent dissociation.

To conclude, the present set of experiments showed that encoding information into VWM is reflected in the PLR: The encoding of a bright stimulus, compared to the encoding of a dark stimulus, leads to a pupil constriction. The maintenance of said stimuli in VWM, however, is not reflected in the PLR. Our results therefore suggest that the pupil size is tuned to the brightness of the to-be-encoded information, allowing for an optimal encoding of visual information.

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