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## Microsaccadic eye movement orientations are equivocal in the presence of competing stimuli

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Dedicated to Prof Jay M Enoch

Will someone reflexively look towards a primed target or to a non-primed target, when no instructions are given? Knowing this could help design visual function tests without the need for instructions. Simply, a target could be presented for a "priming phase" followed by two targets one of which is the primed target and the other is not. We asked the question to which target will an obsever look. We studied this on normally-sighted adults. Eye movements were tracked using EyeLink1000 Plus eye tracker and microsaccades were analyzed. The targets presented were from LEA symbols that are commonly used in children's visual acuity chart. Target size (15', 20' or 25') and presentation duration (200, 400 or 600 ms) were randomized. No instructions were given to the participants beyond asking them to look at the computer monitor in experiment I, and instructions were given to specifically look towards the primed target in experiment II. Overall we found that no preference (proportion of microsaccades <50%) was observed either to the primed or to the novel target in either of the experiments. The presence of two competing stimuli abolishes the microsaccde orientation to a target of interest, even with explicit verbal instructions. © Anita Publications. All rights reserved.

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### **1** Introduction

When no instructions are given, and if a person was primed with a target and then presented with two targets, one of which is the primed and the other non-primed, where would one first look, when the task is mere passive viewing with no goals of any kind to achieve? Would one look at the familiar target (primed) or to the novel target (non-primed) ? The motivation for answering this question came from exploring clinical test paradigms with eye tracking and without the need for giving instructions. Such a situation may arise when testing infants and young children. Tests designed for these young patients must be rapid and robust. Professor Jay Enoch explored rapid tests to address few challenges in testing children [1].

While preferential looking tests (e.g. Teller Acuity Cards, Cardiff acuity card etc.) that rely on an examiner's judgement of the patient's looking behaviour, are often used without instructions, it is not apparent if the individual was actually able to resolve or recognize the target or are merely looking at a blurred patch. Perhaps for reasons like this, poor correlation has been found between these tests and letter acuity [2]. Although the motivation for studying the eye movements for primed targets stemmed from a need to test young children, we realized there was not much of an understanding in this looking behavior even in "typically normal" adults. While few studies have looked at eye movements without any specific

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instructions for activities of daily living like television viewing [3] or sandwich making [4], there still is an implicit goal to achieve in these tasks. Laboratory based psychophysical studies provide careful instructions about the experimental task, and usually practice trials are given to make sure the instructions are well understood. In the study reported here neither instructions were given nor practice trials were given. This is similar to studies done on infants where no instructions can be given. In the field of developmental psychology, the familiarization-novelty paradigm is common. In this paradigm, some trials are dedicated for familiarization before introducing the novel targets to the infants. However, this paradigm has given mixed results in infants [5]. Priming influences saccadic eve movements towards the primed targets, even if they are less salient than non-primed targets. This is because the memory representation of the primed target is more easily and quickly activated [6]. In the present study paradigm, an attempt was made to investigate the effect of priming/familiarity on the looking behaviour. Specifically microsaccades were examined for its orientation towards the primed or novel target. Cueing/priming has been shown to affect the direction of microsaccades, essentially orienting the microsaccades towards where a saccadic eve movement is planned [7,8]. While these studies showed only a single saccadic target after priming the individual, little is known about microsaccades when two simultaneous targets are presented after priming. The results from such a study could reveal the eye movement planning for a primed or novel target.

#### 2 Materials and Methods

The study protocol was approved by the Institutional Review Board of L V Prasad Eye Institute and followed the tenets of the Declaration of Helsinki. All participants gave written, informed consent to participate and were free to withdraw at any time. Two experiments were conducted.

#### **Experiment** I

#### Participants

Participants (age >18 years) were a convenient sample selected from the staff and trainees at the institute. All had normal or corrected to normal visual acuity (20/20, N6) with no ocular pathology or known systemic diseases. All were naïve to the purpose of the experiment. They were recruited by requesting to participate in a research study where they have to look at a computer monitor. They were also told that if they have any questions during the experiment, it will be answered at the end of the experiment.

## Task

Participants were seated in a dimly illuminated room (17 lux) at a distance of 71 cm from a computer monitor (HP W1972a, size: 412×232 mm, resolution: 1280×1024 pixels, screen refresh rate: 60 Hz). Head movements were stabilized using chin and forehead rest. Desktop mounted EyeLink 1000 Plus eye tracker (SR Research Ltd., Ottawa, Canada) was used to measure the eye positions (data sampled at 500 Hz). A nine-point calibration procedure was done prior to the start of the experiment. The participants were explicitly told to look at the dots for the calibration routine. Post the calibration, the participants were not given any instructions beyond asking them to look at the computer screen. All the tests were performed binocularly. *Stimuli* 

The LEA symbols (circle, apple, house, and square) were selected as the visual stimuli, as it is commonly used in testing visual acuity for children. Also, all the symbols give a percept of a blurred circle at detection threshold [9]. These symbols were displayed in white color (R,G,B = 255, 255, 255) on a black background (R,G,B = 0, 0, 0) on the computer monitor using MATLAB R2014b (MathWorks, MA, USA) with psychoolbox (version 3.0.13) [10,11]. At the start of a trial, a single white LEA symbol (visual angle 50', 20/200 size) was displayed for 2000 ms (Fig 1). This was considered as the priming phase. Post this, two LEA symbols "targets" were displayed, one of which was the same as the previous symbol shown earlier (primed target) and the other (non-primed target) was from one

of the remaining three LEA symbols. Simultaneous paradigm [12], was adopted in this study, i.e. the disappearance of the primed target and the appearance of the other two targets occurred simultaneously.





The target size (15', 20' or 25') and target duration (200, 400 or 600 ms) for the primed and nonprimed targets were randomized. The choice of the target duration was to permit only one saccadic eye movement at the lower limit (200 ms). The choice of the target size was at suprathreshold for detection, with the smallest target size being at the size of 20/60, the cut-off visual acuity limit for low vision definition (moderate visual impairment) [13]. After the presentation of the two LEA symbols, a black screen appeared for a duration of 1000 ms after which the next trial began with the priming phase. The primed and nonprimed targets were displayed at 0.5° eccentricity on either side of the priming symbol. The direction of the side (left/right) for the primed and non-primed targets was chosen randomly for each trial. The choice of target eccentricity was to allow visibility of both the targets. Since the LEA symbol used in the priming phase was larger than the target sizes, a motion drift artifact was avoided. It is also known that priming is not affected by the size variation between the primed target and the later viewed target [14].

A total of 36 unique trials (4 LEA symbols  $\times$  3 different target sizes  $\times$  3 different target durations were repeated 3 times, resulting in a total of 108 trials per participant. A drift correction [15] was conducted after every ninth trial and then a break was given if desired by the participant. At the end of the experiment, participants were asked an open-ended question to give written feedback about their experience and observations made from the task. This was done to understand if the participants figured out the experimental paradigm. The total duration of the experiment was about 10-12 minutes per participant.

#### Experiment II

Experiment II was similar to Experiment I, except that the participants were given explicit instructions to look towards the primed target. A demo trial was shown for these participants. These participants were aware of the study purpose. A different group of participants, with similar demographics profile as in Experiment I, were enrolled for this experiment.

#### Eye movement analysis

Given that the targets were shown only at 0.5° eccentricity in this study, it will be more appropriate to investigate for microsaccades. It is known that microsaccades could indicate covert attention [7]. Therefore,

we examined the microsaccades direction using an algorithm developed by Engbert & Kleigl [7] and by Engbert & Mergenthaler [16]. According to this algorithm, microsaccades were defined by a velocity criteria (combined for horizontal and vertical positions) that exceeds a relative velocity threshold (i.e. 5 standard deviations above the median velocity) for a minimum duration of 6 ms (or 3 samples in our set up). Additionally, the magnitude of the microsaccade should be less than or equal to 1° and should occur after 25 ms from the previous microsaccade. It has been found that the majority of the microsaccades tend to orient in the direction in which an eventual saccadic eye movement will be made [7,17,18]. Hence, we analyzed the direction of the majority of the microsaccades made during the examined time interval. Trials with loss of eye position by the tracker, and inaccurate fixation (not at the center of the screen) were discarded. The proportion of microsaccadic eye movement was made within the examined time interval was also computed. The proportion of trials in which no eye movement was made within the examined time interval was documented as well.

#### Data Analysis

Proportions of the microsaccades directed towards either the primed or the non-primed targets were analyzed for each of the independent variables: target size and target duration. Majority of the microsaccadic eye movements direction was considered for this analysis. Depending on the data being normally distributed or not, a parametric or non-parametric test was used. All the statistical analyses were performed with SPSS (version 20.0; IBM Corp., New York, NY). As indicated, in earlier studies [17,18], majority of the microsaccadic directions of eye movements were considered for this analysis.

#### **3** Results

#### Experiment I

Thirty-one (15 males) normally sighted adults participated in the study. The mean age  $\pm$  standard deviation (SD) of the participants was 24.7 $\pm$ 4.6 years. The feedback from the participants showed that most of them were unaware of the primed target. Only one participant reported the presence of the primed target. This participant's data, however, did not appear different from the average performance of the group and was included in the analysis.

A total of 3,348 trials were recorded; of these, the trials that were presented immediately after the drift correction had to be discarded for all participants because of a program glitch. This resulted in 12 discarded trials and 96 valid trials from each participant. Thus, a total of 2976 trials were analyzed. The data was found to be normally distributed (one-sample Kolmogorov-Smirnov test, p > 0.639) and, therefore, parametric tests were used. A total of 4% trials were discarded due to blinks.

#### Proportion of microsaccades

Overall, the proportion of microsaccades towards the primed target (mean: 36%, SD:  $\pm$ 9) was not significantly different (p > 0.29, paired *t*-test) with that towards the non-primed targets (34%, SD:  $\pm$ 8). In 24% (SD:  $\pm$ 14) of the trials, no microsaccadic eye movements were observed (Fig 2).

The difference in proportion between the two targets i.e. primed minus non-primed was plotted for individual participants (Fig 3). A stricter threshold criterion was applied to investigate how many participants "meaningfully" looked in the direction of the primed or non-primed target. For this calculation, the average of the standard deviations from the primed (9%) and non-primed (8%) targets was considered. Thus, a threshold difference greater than 8.5% between the primed and non-primed targets was considered as a true response. With such a criteria, our data showed that 24% (n = 7) of the participants were orienting in the direction of the primed targets, and only 7% (n = 2) of the participants looked to the non-primed targets. The remaining large majority of the participants had an equivocal preference (i.e. the difference in proportions did not exceed 8.5% between the primed and non-primed targets).



Fig 2. Distribution of the mean proportion of microsaccadic eye movements of all the participants is plotted along with the group average and standard deviation error bar towards primed and non-primed targets. Trials with no microsaccades are also plotted here, with its standard deviation (the negative value of standard deviation is of no practical value and hence the y-axis was trimmed to zero).



Fig 3. The difference between the proportion of microsaccades eye movements for primed and non-primed targets (y-axis) is shown for each participant (x-axis). The dotted line shows the cut-off threshold (8.5%), beyond which the microsaccade orientation is considered as meaningful (see text for explanation). The majority of the participants did not have a strong direction orientation.

#### Target duration and target size

Repeated measures of ANOVA showed the proportion of microsaccadic eye movements made towards the primed or non-primed was not influenced by the target size or by the target duration (Fig 4), (p = 0.22, repeated measures of ANOVA was done as the data was normally distributed).

#### Experiment II

A total of 11 participants (5 males) completed this experiment. The mean  $age \pm SD$  of the participants was  $24.5\pm1.1$  years. The results of this experiment were similar to the results of Experiment I. Essentially, the proportion of microsaccadic eye movements towards the primed and non-primed targets were comparable (Fig 5). The proportion of trials with no microsaccades were lesser in this cohort (11.9%) as compared to the earlier cohort (24%) who were not instructed to look in the direction of the primed target. Effect of the target size or the target duration on microsaccades were also similar to that observed in Experiment I.



Fig 4. Proportion of eye movements towards primed and non-primed targets for (A) different target sizes and for (B) different target durations (ms = milliseconds). Error bars show the standard error of the mean.



Fig 5. Distribution of the mean proportion of microsaccadic eye movements of the 11 participants (similar to Fig 2).

#### 4 Discussion

The main focus of this study was to investigate microsaccadic eye movements when two targets were presented simultaneously, one of which is primed and the other is not. Specifically, we examined to see if the microsaccdes would be oriented towards the primed (familiar) or to the non-primed target (novel). Our investigation showed that such an orientation in microsaccdes was equivocal. This was true regradless of whether the instructions on where to look was given or not (Fig 2 and 5).

For a free viewing condition of a complex scene for a long duration of time (3 minutes), eye movement characteristics differed, depending on the given instructions [19]. In a passive TV viewing task upto 7 minutes, patients with age-related neurodegenrative eye disorders could be separated out based on scan paths of the eye [3]. In the present study, with only a brief exposure to simplistic targets no pattern of orientation emerged in young healthy adults. The present study's paradigm can be compared to the "choice trials" of Brascamp *et al* [20]. In their paradigm, participants were asked to indicate the direction of a notch in a diamond-shaped target (goal-oriented task). They had pop-out trials in which

one out of 3 targets differed by color; and in choice trials only two targets with different colors were shown. The observer was free to respond at any one of the targets, as all the diamond targets had a notch. It was found that the preference was for the pop-out target and that the choice trial was also biased by the preceding pop-out color. Colors can be perceived even without attention, and hence can be a strong priming agent [21]. In the present study, we did not use color targets for priming because we were eventually interested to study the resolution/recognition of optotypes, a task that requires visual attention.

In Experiment I, participants could not decipher the paradigm or were unaware of the primed targets. Lack of awareness is not uncommon. In an attention study [22] examining reflexive eye movements, participants reported that their eye movement was not affected by a novel distractor, even though the objective eye movement measure showed otherwise. It is noted that it takes about 400 ms to process visual information for conscious decision processing [23]. While one of the target duration in our study was greater than this (600 ms), the microsaccade orientation to look towards the primed target for the longer duration was also not significant. Such a trend was observed even with explicit instructions. This could indicate that it perhaps takes longer than 400 ms (or 600 ms) to make a conscious decision especially when two targets are presented simultaneously, to recognize the direction of the desired target and then to plan the eye movement.

Earlier studies on microsaccades indicate that the number of microsaccades decreases when a saccadic eye movement is to happen [24]. It is, therefore, possible that the microsaccades could have been suppressed, as the subjects planned for a saccadic eye movement. Participants in Experiment II, were instructed to look in the direction of the primed target. Hence, it can be speculated that they would have suppressed their microsaccades to plan their saccades. However, it was observed that the proportion of "No microsaccades" in this cohort was lesser than the cohort in Experiment I (Figs 2 and 5), hinting that microsaccades were not suppressed. The contribution of microsaccades in covert attention tasks has been well established [7,25]. It is possible that covert attention and role of microsaccades are more obvious in a goal-oriented task, to facilitate spatial selection for attention [7]. In Experiment I, goal-oriented task was not given. In Experiment II, participants were instructed to make a saccade. While the purpose in both experiments were different, no obvious orientation of microsaccade towards one or the other target was observed. The results of this study are similar to the bi-directional cue condition of an earlier study [7]. In that study, the bi-directional cue indicated the target could appear either to left or to the right. Under such a condition no definite directional orientation of microsaccade was observed.

In summary, experimental paradigm of presenting two targets after a primed target, does not elicit a strong directional predilection in microsaccadic eye movements. This result differs from experiments of single saccadic targets, where microsaccades are oriented towards the cued target. Testing visual acuity with short exposure durations, and with two alternate-choices using objective measures of microsaccades, therefore, may not be a feasible model. Further experimental paradigms needs to be developed to objectively use eye tracking to determine resolution acuity without instructions.

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