# Eye movements in patients with glaucoma when viewing images of everyday scenes

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## Abstract

This study tests the hypothesis that patients with bilateral glaucoma exhibit different eye movements compared to normally-sighted people when viewing computer displayed photographs of everyday scenes. Thirty glaucomatous patients and 30 age-related controls with normal vision viewed images on a computer monitor whilst eye movements were simultaneously recorded using an eye tracking system. The patients demonstrated a significant reduction in the average number of saccades compared to controls (P=0.02; mean reduction of 7% [95% confidence interval [CI]: 3 to 11%]. There was no difference in average saccade amplitude between groups but there was between-person variability in patients. The average elliptical region scanned by the patients by a bivariate contour ellipse area (BCEA) analysis, was more restricted compared to controls (P=0.004; mean reduction of 23% [95% CI: 11 to 35%]). A novel analysis mapping areas of interest in the images indicated a weak association between severity of functional deficit and a tendency to not view regions typically viewed by the controls. In conclusion, some eye movements in some patients with bilateral glaucomatous defects differ from normal-sighted people of a similar age when viewing images of everyday scenes, providing evidence for a potential new window for looking into the functional consequences of the disease.

**Keywords:** glaucoma, visual fields, eye-movements.

## Introduction

Glaucoma is a group of eye diseases that causes chronic and progressive loss of the visual field (VF) through death of retinal ganglion cells. Glaucoma is thought to affect over 60 million individuals around the globe. This figure is expected to rise further still in the coming years (Quigley and Broman, 2006), expressing a heightened need for an enhanced understanding into the nature and expression of the disease. Currently, the core understanding of visual function in glaucoma stems from standard clinical measurements such as automated VFs, which serve as the primary functional biomarker for the clinical identification and monitoring of the disease. However, less attention is paid to the manifestation of the disease outside of the clinic, such as how visual loss may impact performance in daily activities and the patient's quality of life. Studies using questionnaires and interview have revealed that glaucomatous patients feel their disease has an adverse impact on their ability to carry out everyday visual tasks such as driving, reading and searching for objects as well as their general ability to move from place to place (Green et al., 2002, Nelson et al., 1999, Hartmann and Rhee, 2006, Viswanathan et al., 1999). Studies of function in experiments have somewhat supported patient claims, demonstrating that individuals with glaucoma are impaired in several visual tasks compared with those with healthy vision of a similar age (Ramulu et al., 2009, Kotecha et al., 2009, Haymes et al., 2008, Smith et al., 2011). In addition, patients with even moderately damaged VFs are thought to be at an increased risk of falls and accidents, and in the case of certain more severe defects, glaucoma can ultimately lead to the removal of the patient's driving license (Haymes et al., 2007, White et al., 2006, Szlyk et al., 2005, Ramulu, 2009).

Whilst it appears that glaucoma will inevitably have some impact on the individual's daily life, little is known about the underlying mechanisms that influence how visual loss is perceived and managed by an individual. Research suggests that patients' perceptions of their glaucomatous loss are often distorted in that they are unaware of the true extent of their visual defect (Jampel et al., 2002, Shaw, 2005). The actions of compensatory brain processes that allow missing visual information to be 'filled in' (Safran and Landis, 1996, Komatsu, 2006) and for one eye to essentially compensate for the other inevitably complicate how both the patient and clinician perceive the impairment due to the disease. A useful educational outcome may therefore emerge from more research into potential underlying visual

functioning in these patients, beyond the VF test routinely administered in the clinical setting with automated perimetry.

One key area of insight for this may lie in patients' eye movements, and how they may differ from visually 'healthy' individuals. Efficient eye movements are a necessity for successful everyday visual functioning, by allowing the individual to scan available visual information before selecting the objects of most functional importance and bringing them onto the fovea for more detailed analysis. A finding that patients with eye disease move their eyes differently to visually healthy individuals could therefore help underpin the nature of the functional impairment. The link between eye movements and visual functioning has already received some clinical interest in amblyopia (Kanonidou et al., 2010), retinitis pigmentosa (Turano et al., 2001) and age-related macular degeneration (AMD) (Bullimore and Bailey, 1995). Individuals with contrast sensitivity loss were also found to produce abnormal eye movement behaviour as they manoeuvred complex routes through their natural environment (Vivekananda-Schmidt et al., 2004). Furthermore, the potential of such eye movement behaviour research with regards to the development of future low-vision rehabilitation strategies is beginning to be recognised (Seiple et al., 2005). With regards to glaucoma, a recent study by our group indicated that some patients produced different eye movement behaviour when viewing films of road and traffic scenes filmed from the perspective of a driver compared with visually healthy individuals of a similar age (Crabb et al., 2010). This result shed some light on the issue of determining the VF component of fitness to drive. Moreover, it provided some evidence to support previous research findings that individuals with more severe peripheral visual field defects tend to make more fixations, more errors and take longer to find objects than control subjects (Coeckelbergh et al., 2002). Little is known regarding the impact of glaucomatous visual field loss on eye movement behaviour in other visual scenarios. Evidence suggests that eye movements can be driven by the nature and complexity of the task (Andrews and Coppola, 1999, Rayner, 2009, Henderson, 2003, Bertera and Rayner, 2000) and as such, it is of worth to consider the eye movement behaviour produced in a variety of naturalistic situations in order to gain a wider insight into the functioning of those with visual field loss.

The principal aim of this study was to use eye tracking to examine the hypothesis that patients with bilateral glaucomatous field defects exhibit significant differences in eye movement characteristics as compared to age-related healthy control subjects when naturally viewing a series of images of everyday scenes. The results should provide further insight into

the nature of compensatory mechanisms used by glaucomatous patients when perceiving the world around them, which could in turn pave the way for additional educational and management strategies.

## Method

### *Participants*

Patients were recruited from Moorfields Eye Hospital Trust London and the Fight for Sight Optometry Clinic at City University London. All patients had received a clinical diagnosis of glaucomatous optic neuropathy (primary open angle or normal tension glaucoma) with resulting VF defects in both eyes. All patients took part in an eye examination prior to participation in the study consisting of corrected binocular visual acuity (VA) using an Early Treatment Diabetic Retinopathy Study (ETDRS) chart and contrast sensitivity (CS) using a Pelli-Robson (PR) chart. All patients were required to have a corrected VA of at least 6/12 in each eye, with no other ocular disease other than glaucoma. In addition, VFs (central SITA 24-2 and 10-2 on both eyes) were recorded on a Humphrey Visual Field Analyzer (HFA, Carl Zeiss Meditec, CA, USA). To be included in the study, patients were required to have 'overlapping' binocular defects as measured by an estimate of their integrated visual field (IVF) (Crabb and Viswanathan, 2005, Crabb et al., 2004): a simulated binocular visual field in which patients' best point-by-point monocular sensitivity is used (PROGRESSOR software: Moorfields Eye Hospital, London, UK / Medisoft Ltd., Leeds, UK). Specifically patients were required to have two or more IVF locations with sensitivities of less than 20dB. It should be noted that these are only estimates of binocular defects, and not truly measured with both eyes open using something like the Binocular Esterman Test. Visually healthy control subjects were recruited from University staff, centres for the elderly and the University optometry clinic. The binocular contrast sensitivity and visual acuity of all control subjects was also recorded. VFs (SITA 24-2 FAST HFA) were also measured to ensure the volunteer had no defects which would compromise their role as a control subject in the study. A corrected binocular VA of at least 6/12 was required for all recruited participants. Astigmatic error was less than ±2.5 Dioptres in all those recruited. All participants claimed to be in good general health during an interview and participants were not enrolled if they were on any significant medication other than that for their glaucoma. ('Significant medication' included anti-depressants or treatment for diabetes or significant use of β-blocker medication, all of which were deliberately mentioned). Recruitment of patients and controls was made simultaneously with the specific effort to age match participants.

The study was approved by the ethics committee of each of the participating institutions where subjects were recruited (Moorfields and Whittington Research Ethics Committee,

London; School of Community and Health Sciences Research and Ethics Committee, City University London). Written informed consent, according to the tenets of the Declaration of Helsinki, was obtained prior to examination from each subject. All the data, with patient identifiers removed, were transferred to a secure computer at the University.

Sixty participants were recruited for the study: 30 glaucomatous patients with a mean age of 68.5 (SD: 9.7) years and 30 visually healthy age-related controls also with a mean age of 68.4 years (SD: 9.5 years; means not significantly different; P=0.97. Variances not significantly different; P=0.92.). The HFA mean deviation (MD) is a standard clinical measure of the overall severity of a VF defect with more negative values indicating greater VF loss. The MD is calculated as the overall reduction in VF sensitivity relative to a group of healthy age-matched observers. Change in MD is conventionally used as a marker for VF defect severity both in the clinic and in clinical trials. The patients had a range of VF defect severity: average MD was –10.1 (SD: 7.5) dB, –8.2 (SD: 5.2) dB and –6.6 (SD: 4.9) dB in the right eye, left eye and best eye, respectively. Average Pelli-Robson contrast sensitivity values were significantly worse in the patients (mean: 1.80 [SD: 0.16] log units) compared to the control subjects (mean: 1.95 log units [SD: 0.04]) using a two-sample t-test ($P<0.001$; 95% confidence interval [CI] for the mean difference of 0.08, 0.20). Mean ETDRS corrected binocular LogMAR VA was 0.06 (SD: 0.12) and -0.04 (SD: 0.10) in the patients and controls respectively. The difference between these mean values was statistically significant ($P<0.001$; two-sample t-test) but the actual size of the average difference, 0.10, was small (95% CI for the mean difference of 0.05 to 0.16).

***Procedure***

A slide-show of 28 images was shown to each participant. The images were presented on a 22 inch CRT computer monitor (Iiyama Vision Master PRO 514, Iiyama Corporation, Tokyo, Japan) with the participant viewing distance set at 60cm from the monitor, subtending a half-angle of 20.3° by 14.9°. The images were photographs of natural or urban scenes taken using the same camera (Sony DSC-T1, Sony Corporation, Tokyo, Japan) and were presented in random order at a resolution of 1600 x 1200 with a refresh rate of 100Hz. Each image within the slide-show was displayed for exactly 3.2 seconds. Every participant was given the following instruction before starting the study: “Please view these images as you would a slide show of photographs, like if you are viewing a photo album. You can move your eyes around as much as you like”. They were invited to comment on the pictures during the breaks

between each image presentation, and were encouraged to offer an opinion about each photograph such as whether they found it pleasant or recognised the location at which the photograph was taken. This was to ensure that the participants were actually viewing the images, but still allowed for the existence of a 'passive' component in that they were able to view any part of the image that they wished and did not feel that they were being 'tested'. Examples of three of the images with superimposed eye movement data (scanpaths) for three patients and three controls are shown in Figure 1.

***Eye movement recording***

Eye movements were recorded using the head-mounted Eyelink II eyetracker (SR Research Ltd., Ontario, Canada). This instrument samples pupil position monocularly at 500 Hz, with an average accuracy of better than 0.5º and uses velocity and acceleration thresholds of 30°/s and 8000°/s$^2$ respectively, to identify saccades. These thresholds were also applied in other studies that used the Eyelink to investigate eye movements for natural scenes (Frey et al., 2008, Tatler et al., 2007, Foulsham et al., 2011). All saccades of amplitude less than 0.5º were removed so as to exclude noise within the eye movement data; 2.8% of the patient data and 2.7% of the control eye movement data on average were consequently removed. A chin rest was used to minimize head movements and patients were asked to keep their head as still as possible. However if any head movements did occur these were compensated for by the Eyelink II's head movement detection system which adjusts the point of regard accordingly. The Eyelink II proprietary algorithm was used to calibrate and verify the subject's point of regard in relation to the correct location on the display. A specified calibration accuracy of at least a "good" level was a prerequisite before each trial. Between each trial (image being displayed) a drift correction was performed, and in the case where a large drift was detected, a recalibration and verification performed.

All participants wore refractive correction suitable for the viewing distance of 60cm, and the same set of trial frames was worn by each participant (even if they did not need any correction) to ensure that any restriction to the field of view as a result of spectacle frames was equivalent for each person.

***Analysis***

The eye tracker automatically extracts and records a number of eye movement parameters. For this study, number of saccades per second per trial, average fixation duration (ms) per trial and average saccade amplitude (degrees) per trial were considered. These summary eye movement parameters were calculated for each image as viewed by each participant.

The experimental design meant all participants viewed the same 28 images, albeit in random order, so these were assumed to be repeat measures. A General Linear Model (GLM) was used to perform a mixed two-way analysis of variance (ANOVA) (accommodating missing data by the method of median imputation) to assess each of three different eye movement parameters (response variables) using the statistical software package SPSS 18 (IBM Corporation, Somers, NY, USA). In this two-way ANOVA arrangement, variation in the response variable (each eye movement parameter) was expected to be different across the images and across the participants, with the null hypothesis of real interest being no difference in the average value for the eye movement parameters between the patients and controls examined (F test on the main factor, participant group, from the ANOVA). Averages across the whole experiment (means for each eye movement parameter across all 28 images) were also calculated for each participant and were plotted to illustrate overall effects, including overall variability within groups.

In addition, univariate associations between the eye movement parameters against severity of visual field defect as measured by the Best Eye MD (the mean deviation of the better eye 24-2 visual field) and severity of contrast sensitivity (PR log CS) were explored using Spearman's correlation coefficient in the patient group alone. As a consequence of the 'real world' nature of the study, monocular visual fields for the patients were also merged to form a binocular integrated visual field (IVF) for every patient. This method has been previously used and validated in the context of disability produced by visual field loss (Crabb and Viswanathan, 2005, Crabb et al., 2004) and arguably helps represent the link between loss of vision and behaviour in an everyday environment without the use of additional binocular VF testing. Greyscale representations of these binocular integrated VF plots were compared to the eye movement parameters for each patient.

*Secondary analyses*

In order to assess the size of the average viewing area for an individual across the slide-show, all fixations (point of regards) were extracted from the software for each participant across all 28 images. Then, to quantify the spatial coincidence of the point of regard location for each participant, the best-fit bivariate contour ellipse (BCE) of the points was calculated. The BCE area has been previously used to quantify fixation eye movement stability in patients with macular degeneration (Crossland and Rubin, 2002, Bellmann et al., 2004, González et al., 2006) and to quantify viewing areas for subjects as they watch movies (Goldstein et al., 2007, Crabb et al., 2010). In short, the centre of the BCE represents the mean 'point of regard', with the spatial extent of the ellipse being one standard deviation from this centre along two principal axes, theoretically affording 'coverage' of approximately 68% of the 'point of regard' locations. The BCE area (BCEA in degrees squared) therefore provides a summary measure of the spatial extent of a participant's gaze over the entire set of images. These values are then compared between groups to examine the hypothesis that patients may or may not have a more restricted viewing area compared to controls. Univariate associations between BCEA and best eye MD, PR log CS and IVF were also explored.

BCEA provides a summary of the spatial extent of the observer's fixations over the entire set of images but it does not represent the regions of interest that the observer focussed on. The next secondary analyses attempted to determine if some of the patients failed to look at parts of the image that were typically viewed by the visually healthy controls. This required a novel analysis and the development of a bespoke computer program in C#. For each of the 28 images, every fixation made by the visually healthy control subjects was plotted on a grid of the same size as the image (1600 by 1200 pixels). Each fixation was assumed to subtend a diameter of 1.2 degrees (the approximate size of the foveola), and these were represented on the grid (note that in order to validate this choice of diameter the effect was also tested on other sizes [1° to 3°, at 0.2° intervals]). A 'Not seen' 'location' would mean that the pixel would never be covered by any of the controls subjects' (n=30) fixations. It should be noted that the map is not weighted according to how many fixations were made at a location; it is simply binary, divided into areas seen (by at least one control) or 'not seen'. The next step involved consideration of the fixations made by each of the individual patients in relation to this map. For each patient, the program records the percentage of fixations that fall within the regions of interest generated by the sample of control subjects. So, a relatively low value for this metric would indicate that the patient has tended to fixate in areas of the image that none

of the control subjects would have fixated on. To test if the percentage of fixations that the controls and patients make within the control region differ, a bootstrapping style variation of the above analysis was performed. This was done by generating a control map based upon 29 control subjects and comparing this map to the point of regard of the removed control. Conducting this for each image and control combination generated a metric estimating the percentage of fixations within the expected 'normal' viewing area for each control. In turn, every patient is compared to every combination of 29 controls per image and the average of this is used to generate the percentage of fixations spent within that region for each patient.

The computer program produces a visualization of the 'normal' observer or region of interest map. Examples of these are shown in figure 2. The darker, semi-transparent region represents the area that no control subjects viewed, and the fully transparent regions are where at least one of the control subjects fixated on something that was of interest. Fixations made by two glaucomatous patients for the two images in figure 2 (A, C) are displayed as green and red dots in the figure (B, D). Many of the fixations from these patients are not in the region of interest generated by the controls. A summary measure of failure to fixate on regions of interest from the control map was calculated across the entire image set for each patient. These secondary analyses would then allow a test of the hypothesis that this 'unusual' viewing pattern would be more apparent in patients with worse VF defect and worse CS. This was examined by looking at associations between the metric from the analysis and Best eye MD and CS.

## Results

Each of the eye movement parameters considered was assessed using a mixed two-way GLM ANOVA with the images (trials) representing repeated measures. The departure of the F statistic from 1 summarises the extent of a difference in the main factor (patients versus controls) and the P-value refers to the null hypothesis of no effect, or no difference, in that main factor. No obvious departure from Normality was observed in any of the average response variables as assessed by the Kolmogorov-Smirnov test. In the case where Mauchly's test indicated that the assumption of sphericity did not hold, degrees of freedom were corrected using the Greenhouse-Geisser method. There was statistical evidence that on average patients made fewer saccades per second when compared to controls ($F_{1,58}$=6.2; P=0.016) and there was no significant interaction term ($F_{16,931}$=1.6; P=0.07; groups by image) meaning that the effect was almost always consistent across all the 28 repeat measures (trials). Following this, as would be expected, there was also statistical evidence that patients had longer average fixation duration compared to controls ($F_{1,58}$=8.1; P=0.006) with again no significant interaction term ($F_{5,270}$=1.4; P=0.23) meaning that the effect was consistent irrespective of the image viewed. There was, however, no statistical evidence for a difference in the mean saccade amplitude between the groups ($F_{1,58}$=2.3; P=0.14).

Figure 3(a, b, c) illustrates the results by plotting overall mean values for the three eye movement summary measures for each participant in the experiment. The overall mean number of saccades/second per trial (each lasting 3.2 seconds) was 3.1 (SD: 0.4) and 3.3 (SD: 0.3) for patients and controls respectively; an average reduction in saccade numbers of 6.7% (95% CI: 2.6 to 10.8%). The overall mean fixation duration (ms) was 297 (SD: 52) and 265 (SD: 35) for patients and controls, giving, as expected, an equivalent percent difference between patients and controls. The overall mean saccade amplitude (in degrees) was 5.4 (SD: 1.5) and 5.8 (SD: 0.8) for patients and controls. Whilst the ANOVA indicated no significant difference in the average values for saccade amplitude it is clear from figure 3(c) that there was a greater spread and variation in the between-person saccade amplitude parameter for the patients compared to the controls; this effect was statistically significant (P=0.001; F-test on variances).

Results from the exploratory univariate analysis of association between the eye movement parameters and severity of visual field loss (best eye MD in dB) and severity of contrast sensitivity damage (PR Log CS) in the patient group (n=30) are shown in table 1. There is no

evidence of any statistically significant correlations, suggesting that level of visual field damage or level of contrast sensitivity impairment is not associated with the eye movement parameters.

## Secondary analyses

The BCEA summarises the magnitude of the overall viewing region for a participant across all the images. The mean BCEA was 317 (SD: 89) and 244 (SD: 100) degrees squared for the controls and the patients respectively. Therefore, the average BCEA for the patients was 23% smaller than the average BCEA for the controls (95% CI: 11 to 35%) and this difference was highly significant (P=0.004; two-sample t-test). Results for the glaucomatous patients and control subjects can be viewed in figure 4. When investigating univariate analysis of association between the BCEA and severity of visual field loss (best eye MD in dB) and severity of contrast sensitivity damage (PR Log CS) in the patient group (n=30) there is no evidence of any statistically significant correlations (shown in table 1). This suggests that the level of VF damage or contrast sensitivity impairment is not associated with the size of BCEA in this group of patients.

The bootstrapping techniques applied to our 'region of interest' analysis indicated that each control made, on average, 73.8% (SD: 6.6%) of their fixations within every combination of the other 29 controls in the group. This value was no different to the average in the glaucoma group (75.6% [SD: 9.4%]) although there was a significant difference in the spread of the values (variance) between the groups (F-Test on variances; P= 0.05). If the controls are ranked and a $90^{th}$ percentile cut off is applied to the lowest values in that group (i.e. three controls viewing the least "normal" area are removed), then four patients in the glaucomatous group are identified to be below this cut off (65.1%). This number of patients flagged is obviously not beyond what would be expected by chance but the individuals are marked, for interest, in figure 5 with a black circle to the bottom right of their IVF.

When investigating the patient group alone there appears to be some weak association with MD and a tendency to not view regions typically viewed by the controls which is close to statistical significance (Spearman's rho: 0.35; P=0.06). This association is stronger when considering contrast sensitivity (rho: 0.40; P<0.05) (this correlation remains statistically significant, or close to statistical significance, when a range of values are used as an

alternative to the 1.2° representing the foveola [1° to 3° with 0.2° intervals; median rho: 0.37, IQR: 0.34 to 0.40]). Simple linear regression lines have been added to the scatterplots in figure 6. $R^2$ values indicate that only a modest amount of variation of the percentage fixation within the control region can be attributed to severity of functional damage (BEMD $R^2$=21.8%; CS $R^2$=18.7%). Still, this analysis suggests that there is a tendency for patients with a worsening MD in the best eye and a decline in contrast sensitivity to view regions of the image differently than the controls.

| | **Number of Saccades** | **Saccade Amplitude** | **Fixation Duration** | **BCEA** | **Fixations within control region** |
|---|---|---|---|---|---|
| **BE MD** | 0.01 | 0.05 | 0.01 | -0.10 | 0.35 * |
| **PR Log CS** | 0.17 | -0.15 | -0.20 | -0.24 | 0.40 ** |

Table 1: Spearman's rank correlation coefficient (rho) for the three eye movement parameters, BCEA and fixations within control region against severity of visual field loss (best eye MD) and severity of contrast sensitivity damage (PR Log CS) in the n=30 patients. None of these associations were statistically significant apart from * (approaching statistical significance; P=0.06) and ** (P<0.05).

Figure 5 shows greyscale representations of each patient's central binocular IVF. These binocular VFs cover field of view of about 20º (being composed of monocular 24-2 HFA VFs). Summary measures for the eye movement parameters, BCEA and BEMD are given for each patient.

## Discussion

Results from this study provide some evidence to suggest that individuals with bilateral glaucoma produce different eye movements compared to visually healthy individuals of a similar age when passively viewing images of natural scenes. For instance, the patient group made fewer saccades and consequently fixated for longer, than the control subjects when viewing the images. Since new information can only be acquired during fixations (when the eyes hold still) (Matin, 1974), and it is not possible to develop a sufficiently detailed percept from one glance alone (Irwin, 1991); the finding that the patients made fewer saccades suggests they were subsequently unable to process as much of the visual scene as their visually healthy counterparts during the specified time period. An increased number of saccades (and fixations) will also normally compensate for the fact that the quality of information naturally falls off from the central point of regard (Henderson, 2003), and so the fact that less saccades were made in a condition characterised by peripheral loss further suggests that the perceptual quality of each scene was not as fruitful for some of the glaucomatous patients as compared to visually healthy people. Nevertheless, the production of fewer saccades by default enabled the participant to fixate for longer on selected information, and this finding may be the first indicator of the employment of an alternative strategy as a result of the reduction in visual field in an attempt to ensure the most important visual information is not missed. For instance, evidence suggests that the time spent looking at a particular area is related to how informative the individual believes that information to be (Antes, 1974, Henderson, 2003). Therefore, it may be that by spending longer focusing on a particular aspect of the image, the glaucomatous patient either consciously or subconsciously hopes to focus on only the more useful information. Moreover, the observation could, perhaps, simply reflect a difference in the temporal processing of information between glaucomatous patients and individuals with normal vision, but this should be the subject of further investigation. The finding of an increased fixation duration in the patient group also adds weight to the results in that it suggests that the result that less saccades were produced by the patients reflected a genuine change in eye movements and was not simply a fact of poor eye movement recording in one group or other personal factors such as whether patients tended to blink more throughout the task. In addition, this study also introduced investigation into more spatial aspects of eye movement behaviour in terms of the size and general location of the fixations made. According to the results of the BCEA analysis, fixations made by the patients when they viewed the images naturally (and therefore could view any part of the

image they liked) were restricted to narrower regions of the image on average compared with the controls, again implying that the patients were only able to gain a limited interpretation of the image in front of them. When perceiving visual scenes, it is believed that a much wider area of visual field is required to sufficiently gather information than in other visual tasks such as reading (Rayner, 2009). Loss of VF will therefore limit the amount of information that is able to be processed in a certain length of time; a finding supported by research showing that masking increasing areas of the field of view will largely impair an individual's ability to recognise features of a scene (Saida and Ikeda, 1979). Despite this result, there was surprisingly no significant difference when comparing the mean saccade amplitudes of the patients with the controls in this study. This is likely due to the large between-person variability in eye movements observed in the patients compared with the controls; for instance, some individual patients recorded average saccade amplitude around three times larger than other patients. This variability may still serve as an indicator that glaucoma may interfere with the strategies normally employed by healthy individuals, and force some patients to utilise alternatives that are very different to others. The strategy chosen could have important implications for real-life situations during which the rapid scanning of the visual field is required; for instance, for making judgements when crossing the road or during a driving manoeuvre when the immediate visual scene must be taken into consideration.

Our secondary analyses did not yield any direct evidence that the patient group, as a whole, were viewing different regions of interest in the images when compared to visually healthy people. Yet, there was some evidence of worsening VF defect severity (as measured by the MD in the best eye) and worsening contrast sensitivity (as measured by PR CS) in the patients being weakly associated with a tendency to fixate in areas of the image not typically viewed by the controls with healthy vision. This might suggest that there is something specific to glaucomatous VF defects that cause some patients to perceive the image differently to the controls, although more data would be needed to fully support this claim. Indeed the secondary analyses could be further refined when marking those 'regions of interest' by, for example, giving more weight to those areas viewed more frequently by the visually healthy people or thinking more carefully about the limitation of the 'hard' edges, and the size of the fixation 'circle' that was employed. Peripheral information is thought to be used to help filter visual information so that the eyes are directed to more useful items (Mackworth and Morandi, 1967), and it may be the case that the nature of the typically peripheral defects seen in glaucoma somewhat compromises this ability, thus decreasing the

likelihood that the patient will fixate on the same areas as the controls. Moreover, low level visual properties including contrast levels have been shown to influence fixation, with a higher proportion of fixations being made on regions of increased luminance-contrast (Reinagel and Zador, 1999, Krieger et al., 2000, Einhauser and Konig, 2003). Diminished contrast sensitivity – which often coincides with glaucomatous visual field loss (Hawkins et al., 2003, Stamper, 1989) - is therefore likely to impede ability to detect these regions and thus drive the individual to fixate on other, perhaps less salient areas.

Explaining the differences in eye movements observed between groups in terms of the basic parameters and BCEA is more complex, as unlike when considering the spatial locations observed, there was no evidence of a direct association between these variables and MD in the best eye or PR CS. The patients and controls were all elderly and carefully selected to have similar age so this cannot explain the differences in the eye movements. All participants were required to have a corrected VA of at least 6/12 in each eye but average binocular ETDRS VA was slightly better in the controls. VF defects are very difficult to quantify and although summary measures like MD are used clinically they are only a 'blunt' measure of the VF and tell us little about the topography or spatial nature of glaucomatous VF defects. Despite the patients performing differently to the controls on the whole, it should be reiterated that there was still a large amount of variation in eye movement behaviour across the patients. Closer examination of individual cases can help to decipher what additional factors relating to vision may be driving the results. Particular cases of note are patients A and B (Figure 5), who despite having similar MD values in their best eye VF recorded the largest and smallest BCEA respectively. Closer examination of the VFs of the aforementioned patients hints that defect location may also be an important factor in determining the development of compensatory patterns; for instance, it can be seen that patient A's VF defect is more peripheral than that of patient B, who has severe central loss in the upper right hemisphere. Perhaps patient A is less aware of their defect than patient B during everyday functioning, and has therefore not adopted compensatory eye movement strategies within a central 20º. Figure 7 further shows the viewing patterns of patients A and B differ considerably when viewing the same image. Defect awareness is likely to encompass a wide range of characteristics including years since diagnosis, rate of progression and the stage of disease at which the individual was diagnosed, in addition to personal expectations and experiences. A limitation of this study was that this type of data was not collected.

Investigation of the spatial relationship between gaze patterns and VF defects thus awaits further investigation.

Of course there may still have been some additional demand characteristics in existence in this study; for example, since the photographs used in the current study depicted a variety of different scenes taken on different dates, weather types and lighting conditions, it was not possible to control for factors such as saliency, contrast and pattern features of objects which are thought to influence saccadic behaviour (Itti and Koch, 2000, Krieger et al., 2000, Acik et al., 2009, Harding and Bloj, 2010). Nevertheless, the sample of images could be said to be reflective of natural scenes, and since there was found to be no statistically significant interaction term in the eye movements between images, it appears that the nature of the task was still relatively consistent throughout the duration of the study. An additional anomaly relates to whether the participants were actually paying sufficient attention to the images. Care was taken by the experimenter to ask the participants informal questions about the images between presentations but there was not an explicit performance measurement. We wanted the participant to view any part of the image that they wished; perhaps as they would when encountering a visual scene for the first time so as to capture 'natural' eye movements removed from increased task demand. Nevertheless it may be have been that some observed eye movements were in response to visual scenes generated during mind-wandering as opposed to those that were being driven by the real-life scene (Brandt and Stark, 1997).

Some of the findings presented in the current study were orthogonal to results previously published by this group regarding eye movement behaviour when viewing a dynamic driving scene. In the latter, it was reported that there was no difference in the size of viewing area between patients and controls but that patients produced more, and thus shorter, fixations than the controls when searching for hazards in the Hazard Perception Test (HPT) (Crabb et al., 2010). It has previously been reported that participants will alter their eye movements in situations where the quantity of visual information is heightened and the task becomes more cognitively demanding (Greene and Rayner, 2001, Bertera and Rayner, 2000, Vlaskamp and Hooge, 2006). In the current task, cognitive demands were minimal in that participants were presented with static, consistent information whereby any object could be chosen for fixation during the known time period. The participants were in effect tasked to simply 'enjoy' the images. Conversely the HPT was dynamic and as such the viewer was faced with a huge array of changing information every second. Consequently, the participants in the HPT were under pressure to not only find a target object, but to do so during a very limited time period.

We therefore suggest that these inconsistencies may be related to the task complexity and quantity of visual information that was available to the participants.

This study provides some new evidence about how patients with bilateral glaucoma view pictures of natural and everyday scenes. Further studies into the eye movement patterns of patients with glaucoma when carrying out other more cognitively demanding visual tasks, such as reading and searching for objects, are undoubtedly needed before an accurate understanding of the functional deficits experienced by patients with glaucoma can be gained. For instance, it has been shown previously that glaucomatous patients were slower to locate objects in images of natural scenes than age-related visually healthy people (Smith et al., 2011). It may be that the functional difficulties observed in the patient group in this task were related to the employment of alternative eye movement patterns by the glaucomatous individuals as a result of their visual loss. The variability in patient response in the data presented in this study suggest that other experiments consider whether fixation patterns may be adaptive, in the sense that there could be increased probability that scan paths will include fixations in damaged parts of the visual field. Such studies require innovative quantitative approaches of the eye movement patterns beyond, for example, simply counting the number of saccades and the size of the saccades. Examining whether patients with unilateral glaucoma produce different eye movement behaviour in their diseased eye may also be helpful. The idea that some patients make different eye movements to visually healthy control people opens up the idea of thinking how this information might be used in rehabilitation of visual impairment. This has received attention in other retinal diseases such as age-related macular degeneration (AMD) where some eye movement research rehabilitative techniques for visual impairment, with evidence to suggest that reading ability can improve following training focusing on eye positioning and movements (Seiple et al., 2005). Providing auditory feedback about eye movements is also thought to help improve task performance (Hall, 2001). Identifying optimal strategies for patients with glaucoma may help improve performance in other visual tasks (Vukicevic, 2009), and to serve as a way to educate the patient about their defect. We hope that the results from this study may stimulate future investigations which may also make such interventions a reality for glaucomatous patients in an attempt to improve their understanding of their condition and their ability to independently go about their daily activities.

## Conclusions

This exploratory study provides some new evidence that patients with bilateral glaucoma exhibit different eye movement behaviour compared to visually healthy subjects of a similar age when viewing images of everyday scenes. These results should stimulate further eye movement studies in glaucoma and these might provide a new 'window' into understanding the functional deficits of the disease. We speculate that understanding such 'real world' visual function deficits in glaucoma is perhaps a first step towards designing appropriate strategies for patient education about the impact of a visual field defect and potential rehabilitation. Further studies in eye movements and glaucoma are already underway in our laboratory.


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### Author Contributions

Conceived and designed the experiments: NDS DPC DFGH. Performed the Experiments: NDS FCG RB. Analysed the Data: NDS DPC. Wrote the paper: NDS DPC FCG.

## References


ACIK, A., ONAT, S., SCHUMANN, F., EINHAUSER, W. & KONIG, P. 2009. Effects of luminance contrast and its modifications on fixation behavior during free viewing of images from different categories. *Vision Res,* 49**,** 1541-53.

ANDREWS, T. J. & COPPOLA, D. M. 1999. Idiosyncratic characteristics of saccadic eye movements when viewing different visual environments. *Vision Research,* 39**,** 2947-2953.

ANTES, J. R. 1974. The time course of picture viewing. *Journal of Experimental Psychology,* 103**,** 62-70.

BELLMANN, C., FEELY, M., CROSSLAND, M. D., KABANAROU, S. A. & RUBIN, G. S. 2004. Fixation stability using central and pericentral fixation targets in patients with age-related macular degeneration. *Ophthalmology,* 111**,** 2265-2270.

BERTERA, J. & RAYNER, K. 2000. Eye movements and the span of the effective stimulus in visual search. *Attention, Perception, & Psychophysics,* 62**,** 576-585.

BRANDT, S. A. & STARK, L. W. 1997. Spontaneous Eye Movements During Visual Imagery Reflect the Content of the Visual Scene. *Journal of Cognitive Neuroscience,* 9**,** 27-38.

BULLIMORE, M. A. & BAILEY, I. L. 1995. Reading and Eye Movements in Age-Related Maculopathy. *Optometry & Vision Science,* 72**,** 125-138.

COECKELBERGH, T. R., CORNELISSEN, F. W., BROUWER, W. H. & KOOIJMAN, A. C. 2002. The effect of visual field defects on eye movements and practical fitness to drive. *Vision Res,* 42**,** 669-77.

CRABB, D. P., FITZKE, F. W., HITCHINGS, R. A. & VISWANATHAN, A. C. 2004. A practical approach to measuring the visual field component of fitness to drive. *Br J Ophthalmol,* 88**,** 1191-1196.

CRABB, D. P., SMITH, N. D., RAUSCHER, F. G., CHISHOLM, C. M., BARBUR, J. L., EDGAR, D. F. & GARWAY-HEATH, D. F. 2010. Exploring eye movements in patients with glaucoma when viewing a driving scene. *PLoS One,* 5**,** e9710.

CRABB, D. P. & VISWANATHAN, A. C. 2005. Integrated visual fields: a new approach to measuring the binocular field of view and visual disability. *Graefe's Archive for Clinical and Experimental Ophthalmology,* 243**,** 210-216.

CROSSLAND, M. D. & RUBIN, G. S. 2002. The use of an infrared eyetracker to measure fixation stability. *Optometry and vision science : official publication of the American Academy of Optometry,* 79**,** 735-739.

EINHAUSER, W. & KONIG, P. 2003. Does luminance-contrast contribute to a saliency map for overt visual attention? *Eur J Neurosci.,* 17**,** 1089-97.

FOULSHAM, T., TESZKA, R. & KINGSTONE, A. 2011. Saccade control in natural images is shaped by the information visible at fixation: evidence from asymmetric gaze-contingent windows. *Atten Percept Psychophys.,* 73**,** 266-83.

FREY, H.-P., HONEY, C. & KÖNIG, P. 2008. What's color got to do with it? The influence of color on visual attention in different categories. *Journal of Vision,* 8.

GOLDSTEIN, R. B., WOODS, R. L. & PELI, E. 2007. Where people look when watching movies: Do all viewers look at the same place? *Computers in Biology and Medicine,* 37**,** 957-964.

GONZÁLEZ, E., TEICHMAN, J., LILLAKAS, L., MARKOWITZ, S. & STEINBACH, M. 2006. Fixation stability using radial gratings in patients with age-related macular degeneration. *Can J Ophthalmol,* 41**,** 333-339.

GREEN, J., SIDDALL, H. & MURDOCH, I. 2002. Learning to live with glaucoma: a qualitative study of diagnosis and the impact of sight loss. *Soc Sci Med,* 55**,** 257-67.

GREENE, H. H. & RAYNER, K. 2001. Eye-movement control in direction-coded visual search. *Perception,* 30**,** 147-57.

HALL, E. C., CLUFFREDA, K.J. 2001. Eccentric viewing training in macular degeneration using auditory ocular motor biofeedback. *Journal of Behavioral Optometry,* 12**,** 87-93.

HARDING, G. & BLOJ, M. 2010. Real and predicted influence of image manipulations on eye movements during scene recognition. *J Vis,* 10**,** 8 1-17.

HARTMANN, C. W. & RHEE, D. J. 2006. The patient's journey: glaucoma. *Bmj,* 333**,** 738-9.

HAWKINS, A. S., SZLYK, J. P., ARDICKAS, Z., ALEXANDER, K. R. & WILENSKY, J. T. 2003. Comparison of contrast sensitivity, visual acuity, and Humphrey visual field testing in patients with glaucoma. *J Glaucoma,* 12**,** 134-8.

HAYMES, S. A., LEBLANC, R. P., NICOLELA, M. T., CHIASSON, L. A. & CHAUHAN, B. C. 2007. Risk of Falls and Motor Vehicle Collisions in Glaucoma. *Invest. Ophthalmol. Vis. Sci.,* 48**,** 1149-1155.

HAYMES, S. A., LEBLANC, R. P., NICOLELA, M. T., CHIASSON, L. A. & CHAUHAN, B. C. 2008. Glaucoma and On-Road Driving Performance. *Invest. Ophthalmol. Vis. Sci.,* 49**,** 3035-3041.

HENDERSON, J. M. 2003. Human gaze control during real-world scene perception. *Trends in Cognitive Sciences,* 7**,** 498-504.

IRWIN, D. E. 1991. Information integration across saccadic eye movements. *Cognitive Psychology,* 23**,** 420-456.

ITTI, L. & KOCH, C. 2000. A saliency-based search mechanism for overt and covert shifts of visual attention. *Vision Research,* 40**,** 1489-1506.

JAMPEL, H. D., SCHWARTZ, A., POLLACK, I., ABRAMS, D., WEISS, H. & MILLER, R. 2002. Glaucoma patients' assessment of their visual function and quality of life. *J Glaucoma,* 11**,** 154-63.

KANONIDOU, E., PROUDLOCK, F. A. & GOTTLOB, I. 2010. Reading strategies in mild to moderate strabismic amblyopia: an eye movement investigation. *Invest Ophthalmol Vis Sci,* 51**,** 3502-8.

KOMATSU, H. 2006. The neural mechanisms of perceptual filling-in. *Nat Rev Neurosci,* 7**,** 220-231.

KOTECHA, A., O'LEARY, N., MELMOTH, D., GRANT, S. & CRABB, D. P. 2009. The Functional Consequences of Glaucoma for Eye-Hand Coordination. *Invest. Ophthalmol. Vis. Sci.,* 50**,** 203-213.

KRIEGER, G., RENTSCHLER, I., HAUSKE, G., SCHILL, K. & ZETZSCHE, C. 2000. Object and scene analysis by saccadic eye-movements: an investigation with higher-order statistics. *Spat Vis,* 13**,** 201-14.

MACKWORTH, N. H. & MORANDI, A. J. 1967. The gaze selects informative details within pictures. *Perception and Psychophysics,* 2**,** 547-552.

MATIN, E. 1974. Saccadic suppression: A review and an analysis. *Psychological Bulletin,* 81**,** 899-917.

NELSON, P., ASPINALL, P. & O'BRIEN, C. 1999. Patients' perception of visual impairment in glaucoma: a pilot study. *Br J Ophthalmol,* 83**,** 546-52.

QUIGLEY, H. A. & BROMAN, A. T. 2006. The number of people with glaucoma worldwide in 2010 and 2020. *Br J Ophthalmol,* 90**,** 262-7.

RAMULU, P. 2009. Glaucoma and disability: which tasks are affected, and at what stage of disease? *Current Opinion in Ophthalmology,* 20**,** 92-98.

RAMULU, P. Y., WEST, S. K., MUNOZ, B., JAMPEL, H. D. & FRIEDMAN, D. S. 2009. Glaucoma and Reading Speed: The Salisbury Eye Evaluation Project. *Arch Ophthalmol,* 127**,** 82-87.

RAYNER, K. 2009. Eye movements and attention in reading, scene perception, and visual search. *The Quarterly Journal of Experimental Psychology*.

REINAGEL, P. & ZADOR, A. M. 1999. Natural scene statistics at the centre of gaze. *Network: Computation in Neural Systems,* 10**,** 341-350.

SAFRAN, A. B. & LANDIS, T. 1996. Plasticity in the adult visual cortex: implications for the diagnosis of visual field defects and visual rehabilitation. *Current Opinion in Ophthalmology,* 7**,** 53-64.

SAIDA, S. & IKEDA, M. 1979. Useful visual field size for pattern perception. *Percept Psychophys,* 25**,** 119-25.

SEIPLE, W., SZLYK, J. P., MCMAHON, T., PULIDO, J. & FISHMAN, G. A. 2005. Eye-Movement Training for Reading in Patients with Age-Related Macular Degeneration. *Investigative Ophthalmology & Visual Science,* 46**,** 2886-2896.

SHAW, M. E. 2005. Increasing compliance with glaucoma therapy: "so, convince me I have something wrong with my eyes". *Insight,* 30**,** 7-9.

SMITH, N. D., CRABB, D. P. & GARWAY-HEATH, D. F. 2011. An exploratory study of visual search performance in glaucoma. *Ophthalmic and Physiological Optics,* 31**,** 225-232.

STAMPER, R. L. 1989. Psychophysical changes in glaucoma. *Surv Ophthalmol,* 33 Suppl**,** 309-18.

SZLYK, J. P., MAHLER, C. L., SEIPLE, W., EDWARD, D. P. & WILENSKY, J. T. 2005. Driving Performance of Glaucoma Patients Correlates With Peripheral Visual Field Loss. *Journal of Glaucoma,* 14**,** 145-150.

TATLER, B. W., WADE, N. J. & KAULARD, K. 2007. Examining art: dissociating pattern and perceptual influences on oculomotor behaviour. *Spatial Vision,* 21**,** 165-184.

TURANO, K. A., GERUSCHAT, D. R., BAKER, F. H., STAHL, J. W. & SHAPIRO, M. D. 2001. Direction of Gaze while Walking a Simple Route: Persons with Normal Vision and Persons with Retinitis Pigmentosa. *Optometry & Vision Science,* 78**,** 667-675.

VISWANATHAN, A. C., MCNAUGHT, A. I., POINOOSAWMY, D., FONTANA, L., CRABB, D. P., FITZKE, F. W. & HITCHINGS, R. A. 1999. Severity and Stability of Glaucoma: Patient Perception Compared With Objective Measurement. *Arch Ophthalmol,* 117**,** 450-454.

VIVEKANANDA-SCHMIDT, P., ANDERSON, R. S., REINHARDT-RUTLAND, A. H. & SHIELDS, T. J. 2004. Simulated Impairment of Contrast Sensitivity: Performance and Gaze Behavior during Locomotion through a Built Environment. *Optometry & Vision Science,* 81**,** 844-852.

VLASKAMP, B. N. & HOOGE, I. T. 2006. Crowding degrades saccadic search performance. *Vision Res,* 46**,** 417-25.

VUKICEVIC, M., FITZMAURICE, K. 2009. Eccentric viewing training in the home environment: Can it improve the performance of activities of daily living? *Journal of Visual Impairment and Blindness,* 103**,** 277-290.

WHITE, S. C., ATCHISON, K. A., GORNBEIN, J. A., NATTIV, A., PAGANINI-HILL, A. & SERVICE, S. K. 2006. Risk factors for fractures in older men and women: The Leisure World Cohort Study. *Gender Medicine,* 3**,** 110-123.

## Figure Legends

Figure 1: Images A, C and E show three of the images (out of 28) with superimposed scanpaths of eye movements from three of the glaucoma patients. Images B, D, F are the same images showing different control subjects viewing the same images. The dots represent the location of fixations, with the size of the dot representative of the fixation duration. The saccades are represented by the connecting lines and the bivariate contour ellipse (BCE) is shown encompassing approximately 68% of the individual's viewing area calculated, in this instance, for the single image.

Figure 2: Images A and C show original images and B and D show the control 'region of interest' (generated by the fixations of the controls) superimposed as the transparent regions of the image. If no control views a region the area is darkened. The fixations from two different patients are represented by the red and green dots are displayed in image B and D: many of the fixations made by these patients fall in areas of the image that were never viewed by any of the 30 age-matched controls. (Note the size of the symbol is related to the length of fixation duration)

Figure 3: Boxplots showing average number of saccades (A), fixation duration (B) and saccade amplitude (C) for the controls and patients (whiskers on plots are set at the minimum and maximum values). The average number of saccades is decreased in the patient group. Although the average saccade amplitude is similar in both groups, the between-person variability (illustrated by the width of the boxes) is significantly larger in the patients compared to the controls.

Figure 4: Boxplots showing the distribution of bivariate contour ellipse area (BCEA in degrees squared) values for the patients and the controls, with the extreme points of the whiskers indicating the minimum and maximum values. The patients tend to view a more restricted region of the images when compared to the controls.

Figure 5: Greyscales of binocular integrated visual fields (IVF) for all the patients. Overall BCE area (BCEA) is given in degrees squared and the patients are ranked in order of this value. The average percentage of the control area per image viewed by each patient is also stated (CA). Each patient's best eye MD (BE MD) is specified in dB. The average number of saccades (S) and saccade amplitude (SA) in degrees per image is stated for each of greyscales. The IVFs with a black circular mark to the bottom right indicate patients recording values for the control 'region of interest' analysis that fell outside the $90^{th}$ percentile of the normative range. In other words these patients exhibited unusual viewing patterns.

Figure 6: Scatter plots with a regression line showing the relationship between the percentage of fixations made by the patients within the control 'region of interest' against best eye MD and contrast sensitivity.

Figure 7: The left column shows four unmodified images that were displayed to the participants. Each image in the right column shows the corresponding control 'region of interest' overlaid with the BCE and fixation locations of example patients A (red) and B (green); the visual fields for these patients are shown in Figure 5.

Figure 1

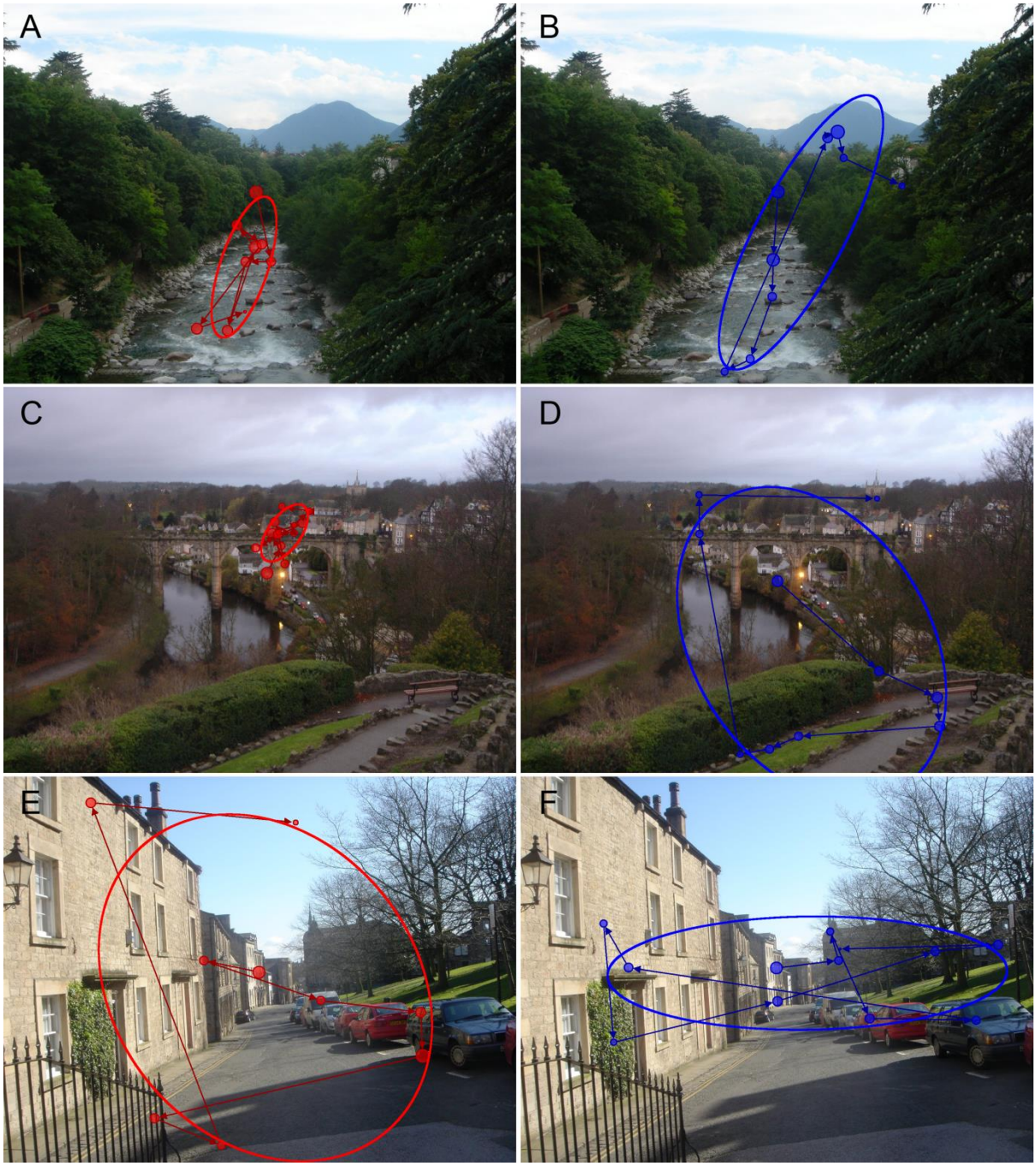


Figure 2

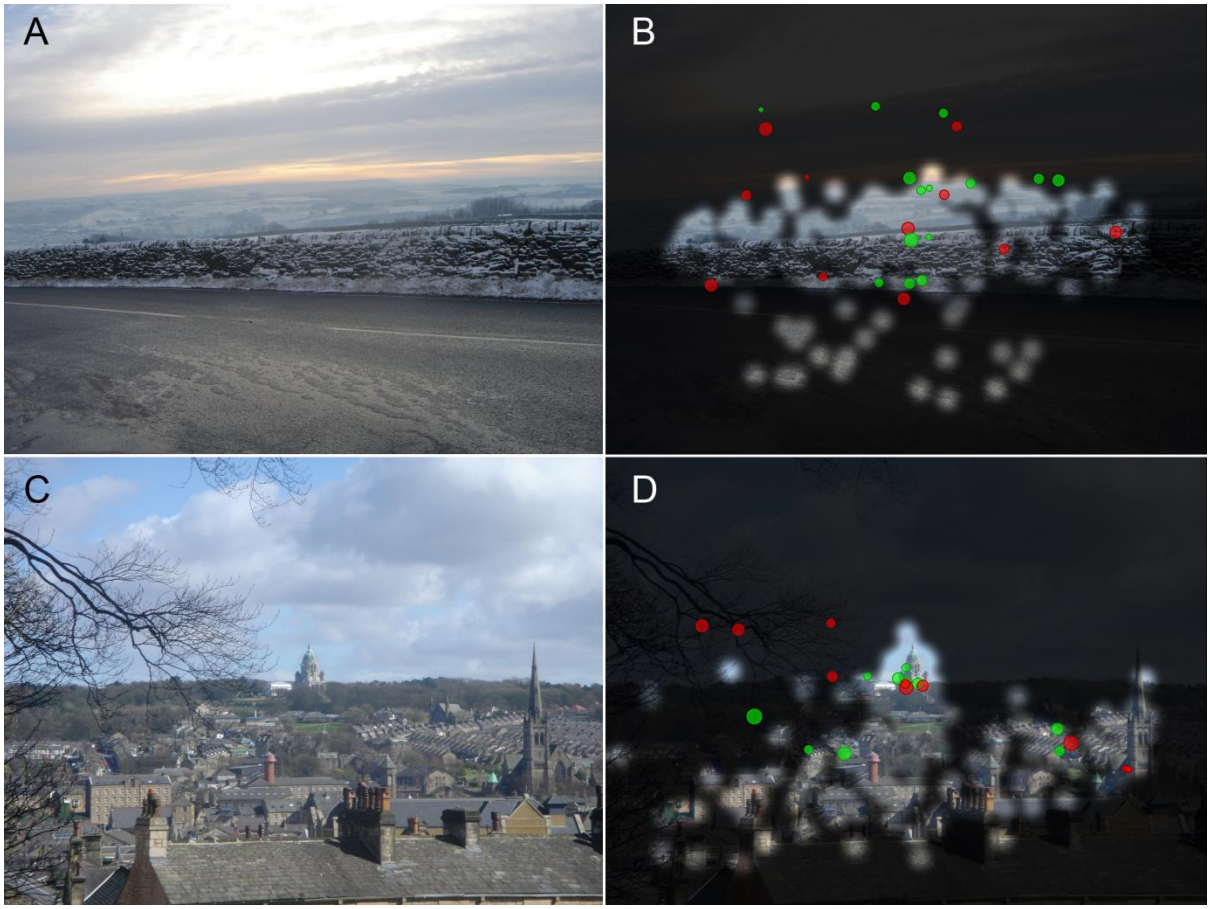
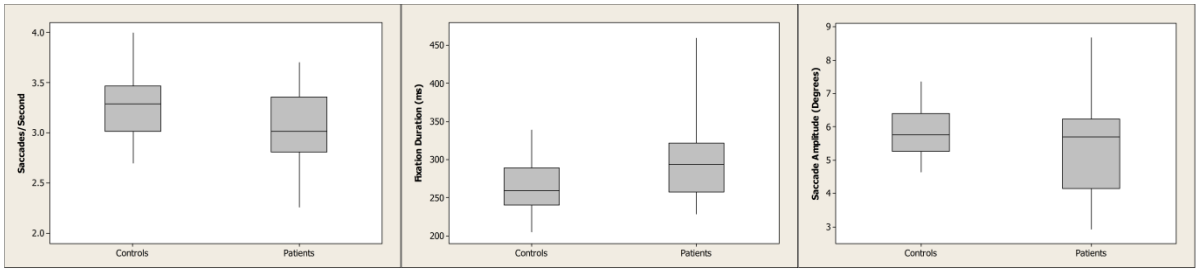


Figure 3


Saccades/Second
4.0
3.5
3.0
2.5
2.0
Controls
Patients
Fixation Duration (ms)
450
400
350
300
250
200
Controls
Patients
Saccade Amplitude (Degrees)
9
8
7
6
5
4
3
Controls
Patients

A B C

Figure 4

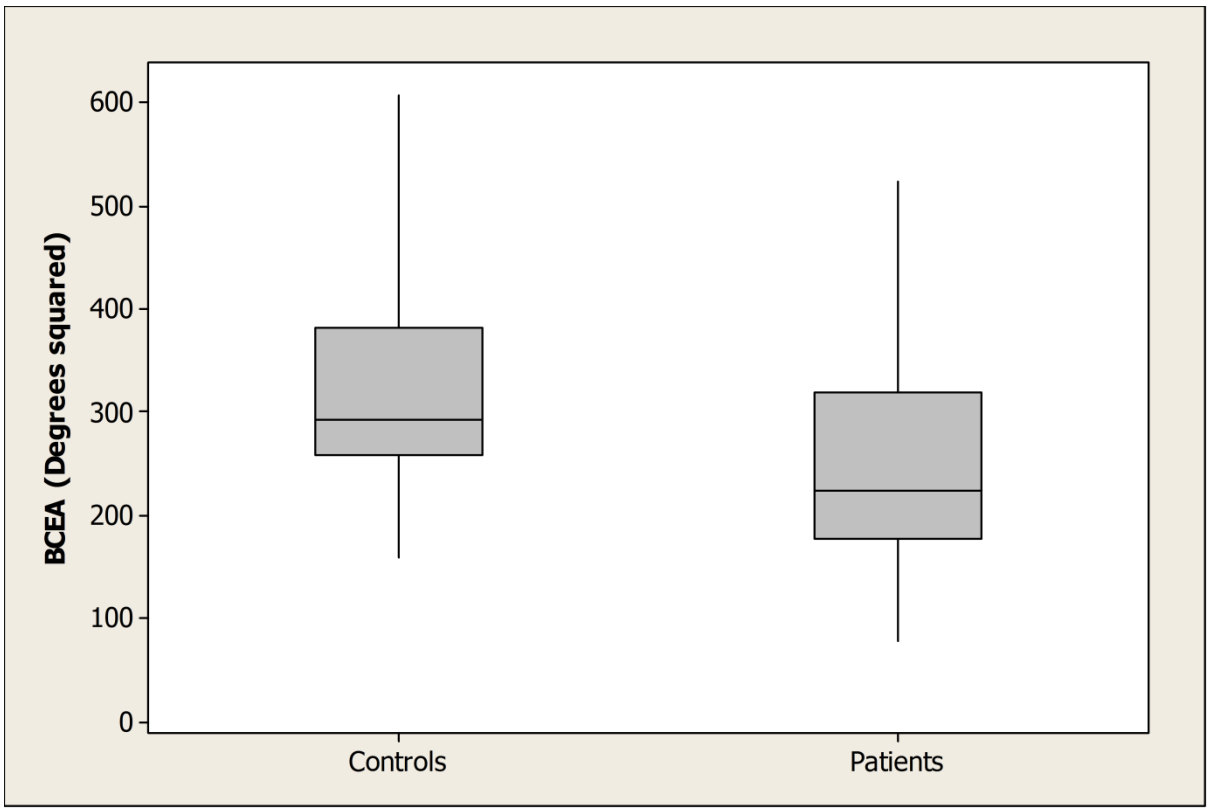

BCEA (Degrees squared)
600
500
400
300
200
100
0
Controls
Patients

Figure 5

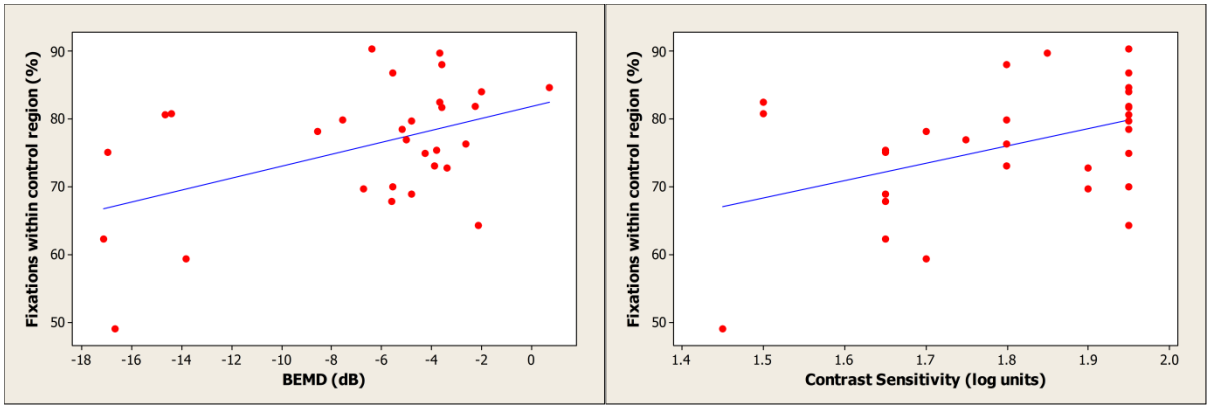

Fixations within control region (%)
90
80
70
60
50
-18
-16
-14
-12
-10
-8
-6
-4
-2
0
BEMD (dB)
Fixations within control region (%)
90
80
70
60
50
1.4
1.5
1.6
1.7
1.8
1.9
2.0
Contrast Sensitivity (log units)

Figure 6

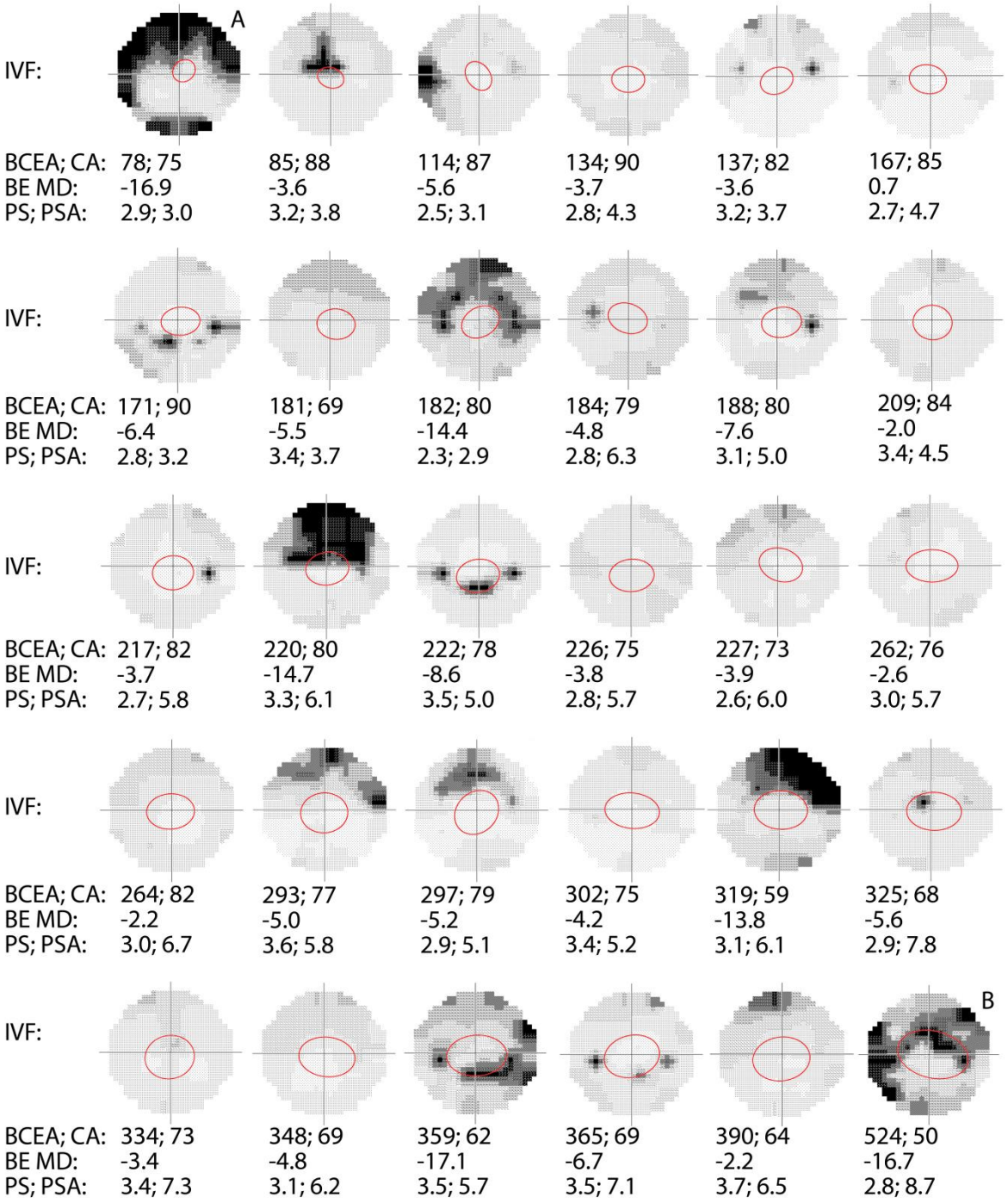

A
IVF:
BCEA; CA: 78; 75 85; 88 114; 87 134; 90 137; 82 167; 85
BE MD: -16.9 -3.6 -5.6 -3.7 -3.6 0.7
PS; PSA: 2.9; 3.0 3.2; 3.8 2.5; 3.1 2.8; 4.3 3.2; 3.7 2.7; 4.7
IVF:
BCEA; CA: 171; 90 181; 69 182; 80 184; 79 188; 80 209; 84
BE MD: -6.4 -5.5 -14.4 -4.8 -7.6 -2.0
PS; PSA: 2.8; 3.2 3.4; 3.7 2.3; 2.9 2.8; 6.3 3.1; 5.0 3.4; 4.5
IVF:
BCEA; CA: 217; 82 220; 80 222; 78 226; 75 227; 73 262; 76
BE MD: -3.7 -14.7 -8.6 -3.8 -3.9 -2.6
PS; PSA: 2.7; 5.8 3.3; 6.1 3.5; 5.0 2.8; 5.7 2.6; 6.0 3.0; 5.7
IVF:
BCEA; CA: 264; 82 293; 77 297; 79 302; 75 319; 59 325; 68
BE MD: -2.2 -5.0 -5.2 -4.2 -13.8 -5.6
PS; PSA: 3.0; 6.7 3.6; 5.8 2.9; 5.1 3.4; 5.2 3.1; 6.1 2.9; 7.8
B
IVF:
BCEA; CA: 334; 73 348; 69 359; 62 365; 69 390; 64 524; 50
BE MD: -3.4 -4.8 -17.1 -6.7 -2.2 -16.7
PS; PSA: 3.4; 7.3 3.1; 6.2 3.5; 5.7 3.5; 7.1 3.7; 6.5 2.8; 8.7

Figure 7