Binocular vision and fixational eye movements

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The aim of this study was to assess the relationship between binocular vision and fixation stability (FS). Across three experiments, we investigated (a) whether fixation was more stable during binocular versus monocular viewing across a range of stimulus contrasts in normal observers (n = 11), (b) whether binocular rivalry affected FS in normal observers (n = 14), and (c) whether FS was affected by interocular contrast differences in normal observers (n = 8) and patients with anisometric amblyopia (n = 5). FS was quantified using global bivariate contour ellipse area, and microsaccades were detected using an unsupervised cluster-detection method. In normal observers, binocular viewing showed more stable fixation at all stimulus contrasts, and binocular rivalry did not affect FS. When interocular contrast was manipulated under dichoptic viewing conditions, normal observers exhibited less stable fixation for an eye that viewed 0% contrast (no fixation target). In anisometric amblyopia, fixation was less stable in both eyes when the fellow eye viewed at 0% contrast. No effects were observed at other interocular contrast differences. Overall, binocular FS was impaired in both eyes in anisometric amblyopia compared to normal observers. We conclude that binocular vision influences FS in normal observers but in an all-or-nothing fashion, whereby the presence or absence of a binocular target is important rather than the relative contrast of the targets in each eye. In anisometric amblyopia, the fellow eye appears to control FS of both eyes under dichoptic viewing conditions.

Introduction

Small, involuntary eye movements (microsaccades, drifts, and tremors) occur during fixation of a stationary target (Helmholtz, 1925; Leigh & Zee, 1999; Martinez-Conde, Macknik, & Hubel, 2004; Steinman, Cushman, & Martins, 1982). The amplitude of these fixational eye movements determines fixation stability (FS)—a measure of ocular motion extent that occurs during fixation. FS is often quantified using bivariate contour ellipse area (BCEA), larger BCEA values indicate less stable fixation. Normal fixational eye movements may benefit spatial vision (Kagan, 2012; Martinez-Conde, 2006; Martinez-Conde et al., 2004; Rucci & Desbordes, 2003; Rucci & Poletti, 2015); however, abnormally large fixational eye movements can impair vision by moving the point of fixation away from the fovea and smearing the retinal image (Chung, Kumar, Li, & Levi, 2015; Chung & Bedell, 1995; Simmers, Gray, & Winn, 1999).

A number of studies have observed less stable fixation (increased amplitude of fixational eye movements) in amblyopic eyes (Chung et al., 2015; Ciuffreda, Kenyon, & Stark, 1991; González, Wong, Niechwiej-Szewdo, Tarita-Nistor, & Steinbach, 2012; Raveendran, Babu, Hess, & Bobier, 2014; Schor & Hallmark, 1978; Shaikh, Otero-Millan, Kumar, & Ghasia, 2016; Shi et al., 2012; Srebro, 1983; Subramanian, Jost, & Birch, 2013). Less stable fixation is associated with a greater interocular acuity difference (González et al., 2012) and poorer stereacuity (Subramanian et al., 2013) in amblyopia, indicating a possible association between impaired binocular vision and reduced FS. Supporting such an association is the finding that transient FS improvements in strabismic amblyopia occurred when fixation targets were bifo-veally aligned and interocular contrast was manipulated to overcome amlyopic eye suppression and promote binocular combination (Raveendran et al., 2014).

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Three studies have reported generally more stable fixational eye movements under binocular compared to monocular viewing conditions. The first involved two rhesus monkeys (Motter & Poggio, 1984), and the other two involved humans (González et al., 2012; Krauskopf, Cornsweet, & Riggs, 1960). Conceptually, binocular viewing might improve FS in observers with normal vision by increasing resolution of the fixation target through binocular summation (Campbell & Green, 1965). However, fixation targets are typically clearly visible, high-contrast images displayed for a prolonged period of time. These factors may minimize binocular summation effects (Bearse & Freeman, 1994). Alternatively, engagement of vergence and fusion mechanisms during binocular viewing may improve FS by activating interocular feedback mechanisms within oculomotor control pathways (Otero-Millan, Macknik, & Martinez-Conde, 2014; Schor, 1979). In strabismic amblyopia, reduced amblyopic eye FS appears to be associated with a loss of foveation and suppression (Raveendran et al., 2014), both of which degrade fusion and vergence.

The overall aim of this study was to further investigate the interaction between binocular vision and FS. In Experiment 1, we measured FS for each eye of normal observers during monocular and binocular viewing of fixation stimuli presented within a contrast range of 100% to 0%. If improved perception of the fixation target due to binocular summation is responsible for improved FS during binocular viewing, we would expect a greater advantage of binocular viewing at lower contrasts where binocular summation for suprathreshold stimuli is more pronounced (Bearse & Freeman, 1994). Alternatively, if improved FS under binocular viewing is due to the activation of vergence and fusion mechanisms, binocular viewing should provide a relatively constant improvement in FS relative to monocular viewing across the whole contrast range.

In Experiment 2, we tested whether interocular suppression affected FS in normal observers using binocular rivalry, which causes alternating suppression of each eye. Binocular rivalry has been found to increase the rate of microsaccades (Sabrin & Kertesz, 1980, 1983), which may make fixation less stable. Reduced FS during binocular rivalry would indicate that interocular suppression can influence FS.

Finally, in Experiment 3, we measured FS across a range of interocular contrast differences in normal observers and participants with anisometropic amblyopia. We hypothesized that interocular contrasts favoring the amblyopic eye would improve amblyopic eye FS by reducing interocular suppression as has been reported for strabismic amblyopia (Raveendran et al., 2014). Conversely, we hypothesized that large interocular contrast differences would impair FS in normal observers due to disrupted binocular combination (Pardhan & Gilchrist, 1990, 1992).

**General methods**

**Apparatus**

Fixational eye movements were measured under nondichoptic and dichoptic viewing conditions. During nondichoptic viewing, participants viewed a gamma corrected 7-in. LCD monitor (Lilliput, CA, http://lilliputweb.net/non-touch-screen-monitors/7-inch-monitors/619gl-70np-c.html) at 40 cm. Dichoptic viewing was achieved using a haploscope constructed from two cold mirrors that transmitted infrared light (Edmund Optics, NJ, http://www.edmundoptics.com/optics/optical-mirrors/specialty-mirrors/cold-mirrors/1900) placed 15 cm from the eyes with the head steadied using a chin rest. Two 7-in. gamma-corrected, luminance-matched LCD monitors (one visible to each eye) were placed laterally along each arm of the haploscope and set 25 cm from the mirrors, creating accommodation and vergence demands of 2.5D and 2.5MA, respectively, which were the same as the nondichoptic viewing condition. Therefore, in both nondichoptic and dichoptic viewing conditions, the planes of accommodation and vergence were fixed at 40 cm. Stimuli were presented using a MacBook, (Apple, Inc., Cupertino, CA) connected to a multidisplay adapter (Dual-Head2Go, Matrox Graphics Inc., Quebec, Canada, http://www.matrox.com/graphics/en/products/gxm/dh2go/analog).

An infrared eye tracker (EyeLink-II, 500 Hz, SR Research, Osgoode, Canada, http://www.sr-research.com/EL_II.html) was used to record fixational eye movements. Nine-point calibrations were completed for each eye separately.

**Participants**

All participants provided informed, written consent, and the study was approved by the office of research ethics, University of Waterloo. All the procedures involved adhered to the Declaration of Helsinki. Normal observers had best corrected visual acuity of 20/20 or better in each eye, stereoaucity of <60 s of arc (Randot test) and no strabismus. Eye dominance was assessed using the Porta sighting test. Participants with anisometropic amblyopia had an interocular VA difference of at least 2 logMAR lines and a fellow eye visual acuity ≤ 0.02 logMAR. Anisometropia was defined as an interocular refractive error difference of ≥1.50 diopter spherical equivalent.
Data analysis

FS was quantified using global BCEA \((\text{González et al., 2012; Steinman et al., 1982; Timberlake et al., 2005})\) defined using the following equation:

\[
BCEA = \pi \chi^2 \sigma_x \sigma_y \sqrt{1 - \rho^2},
\]

where \(\chi^2\) is the chi-square value (two degrees of freedom) corresponding to a probability value of 0.682 \((\text{i.e.}, \pm 1 \text{ SD})\); \(\sigma_x, \sigma_y\) correspond to standard deviations of horizontal and vertical eye positions, respectively; and \(\rho\) corresponds to the Pearson correlation coefficient between horizontal and vertical eye positions. BCEA provides the area of the ellipse that encompasses 68\% of eye positions within a trial. Therefore, larger BCEA values indicate less stable fixation.

In addition, microsaccades were detected using an unsupervised cluster-detection method \((\text{Otero-Millan, Castro, Macknik, & Martinez-Conde, 2014})\). BCEA and microsaccadic amplitude data were log transformed and analyzed using ANOVA. Tukey and \(t\) tests were used to explore simple effects. All statistical tests had a critical alpha value of 0.05.

Experiment 1

Methods

Eleven normal observers \((27 \pm 4 \text{ years})\) participated in this experiment. The fixation stimulus (Figure 1) consisted of an 8.1° frame and a 1° fixation cross presented on a gray background \((50 \text{ cd/m}^2)\). The contrast of the cross and the light portions of the frame were presented at 0\%, 5\%, 10\%, 20\%, 40\%, 80\%, and 100\% Weber contrast levels in relation to the gray, mean luminance background. The dark portions of the target maintained a constant high contrast in all trials and provided a peripheral fusion lock that was essential for dichoptic stimulus presentation.

There were three viewing conditions: (a) monocular fixation with the dominant eye (DE), (b) monocular fixation with the nondominant eye (NDE), and (c) binocular (nondichoptic) viewing. Nonviewing eyes were occluded with an opaque, tight-fitting eyepatch. Each combination of stimulus contrast and viewing condition was measured four times for each participant in a random order. Each trial lasted 30 s.

Results

Data were analyzed using repeated-measures ANOVA with factors of viewing condition \((\text{monocular vs. binocular viewing})\), eye \((\text{DE vs. NDE})\), and contrast \((\text{seven levels})\). BCEA values were significantly smaller \((\text{more stable fixation})\) for binocular than monocular viewing: main effect of viewing condition, \(F(1, 20) = 7.97, p = 0.02\) (Figure 2). The advantage of binocular viewing was consistent across all stimulus contrasts: no viewing condition by contrast interaction, \(F(1, 20) = 1.21, p = 0.42\). However, there was a significant main effect of contrast, \(F(6, 120) = 13.90, p < 0.001\), that was due to larger BCEA values at 0\% contrast compared to all other contrast levels \((\text{Tukey honestly significant difference [HSD]}, p < 0.001)\). The nonzero contrast levels did not vary significantly from one another \((p < 0.05)\). There was no main effect of eye, \(F(1, 20) = 0.02, p = 0.89\), and no other two-way interactions. There was a three-way interaction, \(F(1, 20) = 2.27, p = 0.04\), possibly due to higher variability in the NDE data as post hoc testing did not reveal the source of this interaction.

Unlike BCEA, microsaccadic amplitudes did not differ between monocular and binocular viewing \((\text{Figure 2})\), \(F(1, 20) = 0.2, p = 0.67\). However, in agreement with the BCEA data, there was a significant main effect of contrast \((\text{Figure 2})\), \(F(6, 90) = 9.128, p < 0.001\), that was due to larger amplitudes for the 0\% contrast condition than all other conditions except the 5\% condition \((\text{Tukey HSD}, p < 0.01)\). This implies that the absence of a central fixation target significantly increased microsaccadic amplitudes. There was no main effect of eye, \(F(1, 20) = 0.03, p = 0.87\), and no interactions. Microsaccadic frequency did not vary significantly between binocular and monocular viewing conditions for both DE, \(F(1, 9) = 0.002, p = 0.962\), and NDE, \(F(1, 9) = 0.001, p = 0.98\) (refer to Supplementary Table S1 for mean and \(SEM\)).
Summary of Experiment 1

Fixation was more stable during binocular fixation compared to monocular fixation across all stimulus contrast levels (0% to 100%). However, microsaccadic amplitude did not differ between monocular and binocular fixation. Fixation became less stable, and microsaccadic amplitude increased at 0% stimulus contrast.

Experiment 2: Effect of binocular rivalry on fixation stability

Methods

Fifteen normal observers (28 ± 5 years) participated. Visual stimuli were 100% contrast, circular sinusoidal gratings (3.6° diameter, 1.1 c/°) with a central 0.5° fixation target (Figure 3). Three dichoptic conditions were presented: (a) dichoptic fusion, identically oriented gratings presented dichotically to both eyes; (b) binocular rivalry, orthogonal dichoptic gratings; and (c) monocular stimulation, a sinusoidal grating was presented to the left eye and a mean luminance (gray) blank screen was presented to the right eye. The stimuli were presented so that the right-eye stimulus varied across conditions, whereas the left-eye stimulus remained constant (Figure 3). Therefore, any change in left-eye FS could only be due to changing binocular interactions. We did not vary presentation conditions according to eye dominance because Experiment 1 did not identify any consistent effects of eye dominance on FS in observers with normal binocular vision. During binocular rivalry, participants indicated horizontal grating, vertical grating, or piecemeal percepts using buttons on a gamepad (Sidewinder, Microsoft). A baseline nondichoptic fusion condition was also presented whereby a single sinusoidal grating was viewed binocularly without the haploscope. Grating orientation was changed every 4 s in the dichoptic fusion, monocular stimulation, and nondichoptic fusion conditions. Conditions were presented in a random order. Each condition lasted 40 s and was repeated six times.

Results

Data were analyzed using repeated-measures ANOVA. Left eye (the eye with a constant stimulus; Figure 4) BCEA varied significantly across the four experimental conditions, $F(3, 33) = 8.34$, $p < 0.001$. However, this main effect was due to significantly...
smaller BCEA values in the nondichoptic fusion condition compared to all other conditions (rivalry vs. nondichoptic fusion, \( p = 0.02 \); dichoptic vs. nondichoptic fusion, \( p = 0.001 \)). The rivalry, monocular, and dichoptic fusion conditions did not differ significantly (\( p > 0.05 \); Figure 4) indicating that interocular suppression during binocular rivalry did not increase BCEA. An exploratory analysis revealed that BCEA values did not differ between periods of suppression and dominance during rivalry for a particular eye. The mean BCEA values of the left eye during periods of suppression and dominance were 0.17 ± 0.11 deg\(^2\) and 0.18 ± 0.12 deg\(^2\) (\( p = 0.62 \)) and the mean BCEA values of the right eye during periods of suppression and dominance were 0.17 ± 0.13 deg\(^2\) and 0.17 ± 0.12 deg\(^2\) (\( p = 0.95 \)). Intriguingly, fixation was less stable during periods of piecemeal compared to the periods of suppression/dominance (Figure 5).

A comparison of BCEA for the left and right eyes revealed a significant interaction between condition and eye, \( F(3, 39) = 5.16, p = 0.004 \). Post hoc pairwise analyses (Tukey HSD) revealed a significant difference in BCEA between the two eyes for the monocular viewing condition during which the right eyes (viewing a blank screen) had larger BCEAs than the left eyes (viewing a high-contrast grating), \( p = 0.03 \). The eyes did not differ for any other condition. Unlike BCEA, a comparison of microsaccadic amplitude between the two eyes revealed no main effect of eye, \( F(1, 11) = 0.6, p = 0.81 \), and no condition by eye interaction, \( F(3, 33) = 0.56, p = 0.65 \), indicating no differences between the eyes for any condition. A comparison of microsaccadic frequency showed a significant main effect of viewing condition, \( F(3, 33) = 16.76, p < 0.001 \), and Tukey HSD revealed a significant lower frequency in the nondichoptic viewing condition compared to all three dichoptic viewing conditions (\( p < 0.001 \); refer to Supplementary Table S1 for mean and SEM).

### Summary of Experiment 2

Interocular suppression during binocular rivalry in observers with normal binocular vision did not influence FS. However, during the monocular stimulation condition, the eye viewing a blank screen had less stable fixation than the eye viewing a target.

### Experiment 3

#### Methods

Eight normal observers and five participants with anisometropic amblyopia (Table 1) were recruited. Amblyopia was defined as an interocular visual acuity difference of at least 2 logMAR lines and a fellow fixing eye visual acuity ≤ 0.02 logMAR. Anisometropia was defined as an interocular refractive error difference of ≥ 1.50D (Gao et al., 2018; Guo et al., 2016). The visual stimuli were the same as in Experiment 1 and were presented dichoptically. Contrast to one eye was fixed at 100%, and contrast to the other eye was presented at 0%, 5%, 10%, 20%, 40%, 80%, and 100%. Normal observers completed two viewing conditions (Figure 6): (a) DE contrast was varied and NDE contrast was fixed and (b) NDE contrast was varied and DE contrast was fixed. In the amblyopia group, fellow eye contrast was varied, and amblyopic eye contrast was fixed at 100%. In addition, a condition with amblyopic eye contrast at 0% and fellow eye contrast at 100% was also presented. Stimulus presentation order was randomized, and each eye and contrast combination was presented four times. Each trial lasted 30 s.

The rationale behind recruiting only the observers with anisometropic amblyopia was as follows. A previous study (Raveendran et al., 2014) showed that a lack of foveal fixation was the major factor for impaired fixation stability in the amblyopic eye of individuals with strabismic amblyopia during dichoptic viewing. In this study, our aim was to understand the role of interocular suppression on fixation stability, not a lack of foveal fixation.
Results

In normal observers (Figure 7), BCEA values for the two viewing conditions (NDE fixed/DE varied and NDE varied/DE fixed) were analyzed separately using repeated measures ANOVAs. The ANOVA models had factors of contrast (seven levels of interocular contrast difference) and eye (DE vs. NDE) and revealed significant contrast by eye interactions for both the NDE fixed/DE varied, $F(6, 42) = 3.78$, $p =$
0.004, and NDE varied/DE fixed, $F(6, 42) = 3.298$, $p = 0.009$, conditions. In both cases, this was due to a significant difference in BCEA between the eyes for the 0% contrast condition, whereby the eye viewing 0% contrast exhibited greater BCEA than the eye viewing 100% contrast ($p < 0.001$). The two eyes did not differ in BCEA for any other interocular contrast ratios.

Normal observer microsaccadic amplitudes exhibited a similar pattern of results (Figure 7). The interaction between condition and eye was not significant for the NDE fixed/DE varied condition, $F(6, 36) = 1.52$, $p = 0.20$, but was significant for the NDE varied/DE fixed condition, $F(6, 36) = 2.40$, $p = 0.047$, where there was a significant difference between eyes for the 0% contrast condition only ($p = 0.01$). However, there was no significant effect on microsaccadic frequency for both viewing conditions: NDE fixed/DE varied, $F(6, 36) = 0.59$, $p = 0.735$, and NDE varied/DE fixed, $F(6, 36) = 0.57$, $p = 0.752$ (refer to Supplementary Table S1 for mean and $SEM$).

**Observers with amblyopia**

In the amblyopia group (Figure 8), BCEA exhibited no interaction between contrast and eye $F(6, 24) = 0.570$, $p = 0.750$, and there was no main effect of eye, $F(1, 4) = 0.230$, $p = 0.656$, indicating that the eyes did not differ in BCEA for any condition. There was a main effect of contrast, $F(6, 24) = 2.843$, $p = 0.031$, whereby BCEA for both eyes was larger for the 0% contrast to the fellow eye/100% contrast to the amblyopic eye condition than any of the other conditions ($p < 0.01$). This suggested that BCEA was

![Figure 5. Fixation stability during binocular rivalry. Mean fixation stability values of the left and right eyes during periods of dominance (horizontal stripes), suppression (blank), and piecemeal (checkered). Error bars represent ±1 SE.](image)

<table>
<thead>
<tr>
<th>Participant</th>
<th>Age</th>
<th>Gender</th>
<th>Refractive error</th>
<th>VA (distance)</th>
<th>VA (near)</th>
<th>Sensory status</th>
<th>W4DT</th>
<th>Stereoacuity</th>
</tr>
</thead>
<tbody>
<tr>
<td>S1</td>
<td>38</td>
<td>F</td>
<td>AME: +3.50/−1.00DC × 135</td>
<td>AME: 0.5</td>
<td>AME: 0.66</td>
<td>D: Fusion</td>
<td>60</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>FFE: plano</td>
<td>FFE: −0.3</td>
<td>FFE: −0.1</td>
<td>N: Fusion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S2</td>
<td>48</td>
<td>F</td>
<td>AME: +4.00</td>
<td>AME: 0.46</td>
<td>AME: 0.46</td>
<td>D: Fusion</td>
<td>400</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>FFE: plano</td>
<td>FFE: −0.10</td>
<td>FFE: −0.04</td>
<td>N: Fusion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S3</td>
<td>26</td>
<td>M</td>
<td>AME: +3.50</td>
<td>AME: 0.3</td>
<td>AME: 0.4</td>
<td>D: Fusion</td>
<td>&gt;800</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>FFE: plano</td>
<td>FFE: 0.0</td>
<td>FFE: 0.0</td>
<td>N: Fusion</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S4</td>
<td>42</td>
<td>M</td>
<td>AME: +1.50/−3.25 × 170</td>
<td>AME: 0.4</td>
<td>AME: 0.9</td>
<td>D: Suppression</td>
<td>&gt;800</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>FFE: plano</td>
<td>FFE: 0.02</td>
<td>FFE: 0.0</td>
<td>N: Diplopia</td>
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</tr>
<tr>
<td>S5</td>
<td>47</td>
<td>M</td>
<td>AME: +1.75DS</td>
<td>AME: 0.3</td>
<td>AME: 0.4</td>
<td>D: Fusion</td>
<td>200</td>
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<tr>
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<td></td>
<td></td>
<td>FFE: plano</td>
<td>FFE: 0.0</td>
<td>FFE: 0.0</td>
<td>N: Fusion</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 1. Clinical details of observers with anisometropic amblyopia. Notes: AME = amblyopic eye; FFE = fellow fixing eye. Worth four dot test (W4DT) was measured at 6 m (distance = D) and 40 cm (near = N). Stereoacuity was measured using preschool Randot stereo test, and the values are in arcseconds.
determined entirely by the stimulus presented to the fellow eye. To test this theory, BCEA values for two conditions (amblyopic eye contrast at 100% and fellow eye contrast at 0% vs. amblyopic eye contrast at 100% and fellow eye contrast at 0%) were compared (Figure 9). Amblyopic-eye BCEA was smaller (more stable fixation) when the amblyopic eye viewed 0% contrast and the fellow eye viewed 100% contrast than the opposite configuration ($p = 0.03$). This supports the theory that amblyopic eye BCEA is determined by the fellow eye under binocular viewing conditions.

Microsaccadic amplitudes (Figure 8) also did not exhibit an interaction between contrast and eye, $F(6, 24) = 0.885, p = 0.521$, or a main effect of eye, $F(1, 4) = 2.137, p = 0.218$. There was a main effect of contrast, $F(6, 24) = 2.843, p = 0.031$, but this appeared to be due to variability across the contrast range rather than larger amplitudes only for the 0% contrast condition. Moreover, there was no effect of contrast on microsaccadic frequency, $F(6, 24) = 0.858, p = 0.54$ (refer to Supplementary Table S1 for mean and SEM).

**Controls versus anisometropic amblyopia**

The NDE fixed/DE varied data for normal observers were compared to the amblyopia data (amblyopic eye fixed/fellow eye varied) to assess whether FS differed between the two groups (Figure 9) using an ANOVA with factors of group (controls vs. amblyopia), eye (fellow eye/DE vs. amblyopic eye/NDE) and contrast level (seven levels). As shown in Figure 10, BCEA was larger in both eyes of the amblyopia group compared to normal observers, $F(1, 11) = 7.86, p = 0.01$. The difference between the two eyes varied significantly with contrast: significant contrast by eye interaction, $F(6, 66) = 3.18, p = 0.008$. Specifically, there was a greater BCEA difference between the control and amblyopia groups at low contrasts. Similar trends were present for microsaccadic amplitudes although only a significant main effect of contrast, $F(6, 60) = 6.38, p < 0.001$, was revealed by the mixed ANOVA model (see Supplementary Table S2 for full statistical details). However, there was no significant main effect of contrast, $F(6, 60) = 1.24, p = 0.29$; main effect of group (controls vs. amblyopia), $F(1, 10) = 1.88, p = 0.20$; and no significant interaction (contrast by group), $F(6, 60) = 0.513, p = 0.79$, for microsaccadic frequency.

**Summary of Experiment 3**

FS was not affected by interocular contrast in control participants except when one eye viewed 100% contrast and the other viewed 0% contrast. In this situation, the eye viewing 0% contrast had less stable fixation in agreement with the results of Experiment 2. In the amblyopia group, amblyopic eye FS appeared to be consensually controlled by the fellow eye. Fixation was less stable in both eyes of participants with amblyopia compared to controls.

**Discussion**

The objective of this study was to investigate the role of binocular vision in FS. To meet this objective, we conducted three experiments investigating the impact of binocular versus monocular viewing, binocular rivalry, and interocular contrast difference on FS.
Monocular versus binocular fixation

In agreement with previous studies (González et al., 2012; Motter & Poggio, 1984), we observed significantly more stable fixation under binocular compared to monocular viewing conditions. We extended this previous work to show that the binocular FS advantage occurs across a wide range of fixation stimulus contrasts. The improved FS under binocular viewing conditions was not due to changes in microsaccadic amplitude, as has previously been reported (González et al., 2012; Nallour Raveendran, 2013; Schulz, 1984). Notably, the binocular FS advantage was present for the 0% contrast condition, i.e., the absence of a central fixation target, indicating that the binocular summation of contrast for the central fixation target is not responsible for improving FS.

Apart from the 0% contrast condition, in which fixation became less stable for both binocular and monocular viewing conditions, we found no effect of fixation target contrast on FS. This is consistent with a previous study that varied fixation stimulus contrast (Ukwade & Bedell, 1993). Our results are also in agreement with reports of reduced FS (Cherici, Kuang, Poletti, & Rucci, 2012; González et al., 2012; Raveendran et al., 2014) and increased microsaccadic amplitude (McCamy, Najafian Jazi, Otero-Millan, Macknik, & Martinez-Conde, 2013) in the absence of a fixation target.

Overall, our results demonstrate a clear binocular advantage for FS that is independent of fixation stimulus contrast and also occurs in the absence of a fixation stimulus. Given this pattern of results, the engagement of vergence mechanisms may account for improved FS under binocular viewing (peripheral fusion locks were present for all stimulus contrasts).

Binocular interaction and FS

Less stable fixation correlates with interocular acuity difference (González et al., 2012) and stereoacuity (Subramanian et al., 2013) in amblyopia, indicating a possible association between impaired binocular vision and impaired FS. Therefore, we hypothesized that manipulations of binocular interaction would affect FS. We tested this hypothesis in Experiments 2 and 3.

In Experiment 2, we tested whether interocular suppression affected FS in normal observers. Suppression was induced using binocular rivalry. However,
rivalry had no effect on FS or microsaccadic amplitude. Interestingly, fixation was less stable during the periods of piecemeal rivalry compared to the periods of dominance/suppression. This effect requires further exploration in future work. It could be argued that the duration of binocular rivalry was too short to affect FS. Also, the chronic suppression that occurs in amblyopia is different from the alternating suppression that occurs in binocular rivalry. In Experiment 3, therefore, the effect of binocular interaction on FS was tested by varying interocular contrast in observers with normal binocular vision and those with amblyopia. In amblyopia, reducing the contrast of stimuli presented to the fellow eye can balance monocular inputs from the amblyopic eye and the fellow eye and overcome suppression of amblyopic eye signals (Baker, Meese, &
Therefore, we expected amblyopic eye FS to be improved when suppression was reduced by interocular contrast balancing and the FS of controls to be impaired by unequal interocular contrast. Contrary to our expectations, interocular contrast difference had no effect on FS or microsaccadic amplitude for either group with the exception of the 0% contrast conditions. For controls, this result demonstrates that simply having a visible target in each eye is sufficient for optimal FS in agreement with Experiment 2. For the amblyopia group, FS and microsaccadic amplitude appears to be controlled by the fellow eye across the whole interocular contrast range with the amblyopic eye exhibiting consensual responses. This conclusion is supported by the observation that amblyopic eye fixation is more stable when the fellow eye views the target and the amblyopic eye views a 0% contrast stimulus than vice versa (Figure 8). The consensual responses of the amblyopic eye appears to supersede any effect of reduced interocular suppression on amblyopic eye FS.

Our observation that FS was impaired in both eyes of the amblyopia group compared to controls is consistent with one previous study (Shaikh et al., 2016) in which the fellow eyes of the amblyopia group were found to have abnormal FS. However fellow eye FS was normal in all other previous studies of FS in amblyopia (Chung et al., 2015; González et al., 2012; Raveendran et al., 2014; Shi et al., 2012; Subramanian et al., 2013). The majority of these previous studies used monocular viewing conditions, whereas the current study used dichoptic stimulus presentation. It is possible that providing a stimulus to the amblyopic eye had a detrimental effect on fellow eye FS. Raveendran et al. (2014) also used dichoptic presentation but only recruited participants with strabismic amblyopia. Differences between anisometropic and strabismic amblyopia may account for the differences between the fellow eye results in the current study and those of Raveendran et al. Additional studies are required to test these possibilities.

In addition to showing that FS is not affected by the manipulation of binocular interactions, our data also demonstrate that FS can be independent between the two eyes. For control observers in Experiments 2 and 3, having a visible fixation stimulus to one eye and a blank or 0% stimulus contrast to the other eye led to more stable fixation in the eye with the visible stimulus although microsaccadic amplitude did not differ between the eyes. This interocular difference in FS cannot be accounted for by vergence mechanisms because both eyes saw fusion locks. The finding that FS has eye-specific components is consistent with neuro-
physiological studies demonstrating eye-specific oculo-motor control (Cullen & Van Horn, 2011; Van Horn, Waitzman, & Cullen, 2013). In addition, the lack of an effect on microsaccadic amplitude supports the theory that microsaccades are binocular and conjugate (Martinez-Conde et al., 2004; Martinez-Conde, Macknik, Troncoso, & Hubel, 2009; Rolfs, 2009). Therefore, it is likely that ocular drifts account for this pattern of results as they are a component for fixational eye movements that are independent between the two eyes (Krauskopf et al., 1960).

**Summary and conclusion**

In controls, our results suggest that two components contribute to FS: a general benefit of binocular versus
monocular viewing that may be linked to oculomotor mechanisms engaged by binocular viewing and eye-specific mechanisms that require visual feedback. In amblyopia, amblyopic eye FS appears to be consensual to the fellow eye, suggesting that eye-specific mechanisms are superseded by more accurate information from the fellow eye. Moreover, FS was worse in both eyes for observers with anisometropic amblyopia. This could be due to noisy monocular signals and/or a disruption of oculomotor mechanisms (e.g., abnormal inputs to superior colliculus; Shi et al., 2012).

Keywords: fixational eye movements, binocular interaction, microsaccades, BCEA, binocular rivalry, fixation stability, amblyopia

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