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“Evaluation of Fixations during reading”

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This study was submitted as part of the obligations for the conferment of the Master in Science certification of the MSc Program “Optics and Vision” and was presented at the three-member committee constituted by:

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ABSTRACT

Purpose

The main purpose of this study was to evaluate fixations recorded during reading in two different luminance conditions, with an emphasis on their duration and spatial distribution (fixation stability). Other characteristics of fixations, such as number of microsaccades and their characteristics have been also analyzed.

Methods

In this study a group of 14 people participated with a mean age of 30,6 years (25-36), native Greek speakers, with habitual refractive correction, VA better than 0.0 logMAR in each eye. They have been asked to read a paragraph, consisted of 141 words, in high and low luminance conditions. Eye movement data have been recorded under two conditions of luminance using a video eyetracker (EyeLink II by SR Research Ltd.) at a sampling rate of 500 Hz at a distance of 40 cm. We have developed an algorithm to analyze the recorded data, to detect forward and regressive saccades and obtain information about the characteristics of fixations, such as fixation duration, 68% BCEA and also to detect and analyse microsaccades.

Results

There was not any statistical significant difference in the number of fixations in high and low luminance conditions. Luminance had an effect on the percentage distribution of saccades, as the percentage of regressive fixations was lower and the percentage of forward fixations was higher in low luminance. We also showed that the median fixation duration and median 68 % BCEA were higher in low luminance conditions. For both forward and regressive fixations, we found a weak positive statistical significant correlation between 68% BCEA and fixation duration, in high and low luminance conditions. The average number of microsaccades per fixation was higher in low luminance conditions and the average microsaccades rate (N/sec) was higher in high luminance conditions for both forward and regressive fixations. We also showed that the average number of microsaccades increases as a function of fixation duration, while the average microsaccades rate decreases. The majority of the detected microsaccades are oriented to the opposite direction of the previous saccade.

Conclusions

Ex-Gaussian analysis was proved a useful tool to understand the effect of luminance in the distributions of fixation duration, 68% BCEA, and the microsaccades' characteristics (peak velocity and amplitude). The separation of forward and regressive fixations lead to a better understanding of their characteristics during reading. The analysis presented in this study could be helpful for future studies to unveil the effect of other factors on fixations, such as contrast, print size, personality characteristics and other cognitive factors.

ΠΕΡΙΛΗΨΗ

Σκοπός

Ο βασικός σκοπός της συγκεκριμένης μελέτης ήταν η αξιολόγηση των σημείων προσήλωσης που καταγράφηκαν κατά την διάρκεια της ανάγνωσης (σε δύο διαφορετικές συνθήκες φωτεινότητας), με έμφαση στην διάρκεια και την χωρική κατανομή (σταθερότητα) τους. Επίσης αναλύθηκαν άλλα χαρακτηριστικά των σημείων προσήλωσης, όπως ο αριθμός και τα χαρακτηριστικά των μικροσακκαδικών κινήσεων.

Μέθοδοι

Στην μελέτη συμμετείχε μια ομάδα 14 ανθρώπων, με μέση ηλικία τα 30,6 χρόνια (25-36), με οπτική οξύτητα καλύτερη από 0.0 logMAR σε κάθε οφθαλμό. Από τους συμμετέχοντες ζητήθηκε να διαβάσουν μια παράγραφο, αποτελούμενη από 141 λέξεις, σε συνθήκες υψηλής και χαμηλής φωτεινότητας. Η καταγραφή των οφθαλμικών κινήσεων πραγματοποιήθηκε στις δύο συνθήκες φωτεινότητας, χρησιμοποιώντας τον ανιχνευτή οφθαλμικών κινήσεων EyeLink II της SR Research Ltd. με συχνότητα δειγματοληψίας τα 500 Hz σε απόσταση 40 cm. Για την ανάλυση των καταγραφών, αναπτύξαμε έναν αλγόριθμο με βασικές λειτουργίες την ταξινόμηση των σημείων προσήλωσης σε forward και regressive σημεία προσήλωσης, τον υπολογισμό των χαρακτηριστικών τους (διάρκεια προσήλωσης, 68% BCEA, ανίχνευση και υπολογισμός χαρακτηριστικών των μικροσακκαδικών κινήσεων).

Αποτελέσματα

Δεν βρέθηκε στατιστικά σημαντική διαφορά στον αριθμό των σημείων προσήλωσης και στις δύο συνθήκες φωτεινότητας. Η φωτεινότητα είχε στατιστικά σημαντική επίδραση στην ποσοστιαία κατανομή των διαφορετικών τύπων σημείων προσήλωσης, καθώς το ποσοστό των regressive σημείων προσήλωσης ήταν χαμηλότερο και το ποσοστό των forward σημείων προσήλωσης ήταν υψηλότερο σε συνθήκες χαμηλής φωτεινότητας. Η διάμεσος της κατανομής της διάρκειας των σημείων προσήλωσης αλλά και της κατανομής των 68% BCEA ήταν μεγαλύτερη σε συνθήκες χαμηλής φωτεινότητας. Βρέθηκε ασθενής στατιστικά σημαντική συσχέτιση μεταξύ της διάρκειας των σημείων προσήλωσης και του 68% BCEA, για τους δύο τύπους σημείων προσήλωσης και στις δύο συνθήκες φωτεινότητας. Ο μέσος αριθμός μικροσακκαδικών κινήσεων ανά σημείο προσήλωσης ήταν υψηλότερος σε συνθήκες χαμηλής φωτεινότητας, ενώ η μέση αναλογία μικροσακκαδικών ανά δευτερόλεπτο ήταν μικρότερη σε συνθήκες χαμηλής φωτεινότητας. Επίσης ο μέσος αριθμός μικροσακκαδικών ανά σημείο προσήλωσης αυξάνει ως συνάρτηση της διάρκειας των σημείων προσήλωσης, ενώ η μέση αναλογία μικροσακκαδικών ανά δευτερόλεπτο φθίνει. Οι περισσότερες μικροσακκαδικές που ανιχνεύθηκαν είχαν προσανατολισμό αντίθετο από αυτόν της προηγούμενης σακκαδικής κίνησης.

Συμπεράσματα

Η χρήση της ανάλυσης ex-Gaussian αποδείχθηκε ιδιαίτερα χρήσιμη για την κατανόηση της επίδρασης της φωτεινότητας στις κατανομές της διάρκειας των σημείων προσήλωσης, του 68% BCEA όπως επίσης και των χαρακτηριστικών των ανιχνευθέντων μικροσακκαδικών κινήσεων (μέγιστη ταχύτητα και πλάτος). Ο διαχωρισμός των σημείων προσήλωσης σε forward και regressive, ήταν ιδιαίτερα σημαντικός ώστε να κατανοήσουμε τις διαφορές τους με βάση τα χαρακτηριστικά που αναλύθηκαν.

Η ανάλυση που παρουσιάσαμε στην συγκεκριμένη εργασία, θα μπορούσε να αποτελέσει σημαντική βοήθεια σε επόμενες μελέτες των οφθαλμικών κινήσεων κατά την ανάγνωση, ώστε να αναδειχθεί η επίδραση άλλων παραγόντων στα σημεία προσήλωσης, όπως η αντίθεση φωτεινότητας και το μέγεθος των γραμμάτων, προσωπικά χαρακτηριστικά και συμπεριφοριστικοί παράγοντες.

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PART 1. INTRODUCTION

1. Visual System and Visual Processing

1.1 Visual System

Visual system is one the most important parts of sensory system. It serves as the basis for our perception of the outside world. The ability to perceive, interact and obtain information from the surrounding environment, is derived from the functions of the visual system.

Visual perception begins in retina where rays of light are focused by the optical elements of the visual system and there light is converted to electrical signal, which through the optic nerves is sent to higher centers of the brain for further processing.

The following figure (Fig. 1) shows the fundamental structures of the human eye and the way of light, which enters the eye from the cornea and as it is focused by the cornea and the lens, it passes the vitreous humor and finally reaches retina. The absorption of light and its transduction into electrical signals is carried out by the photoreceptors. There, visual information is transferred to the ganglion cells via the bipolar cells from the receiving photoreceptors, and the information is transduced via a three-stage biochemical cascade. The ganglion cells are capable of not only detecting weak contrasts and rapid changes in light intensity, but are also attributed to the processing of visual aspects such as movement, fine spatial detail, or color. At this point, the electrical signals travel to the lateral geniculate nucleus (LGN) of the thalamus in the brain, and from there, to the primary visual cortex (which is also referred to as visual area 1, V₁) at the rear of the brain. Visual information is crossed-images from the right side are seen by the left retina and transmitted to the right thalamus, and vice versa (Sashank Prasad, 2011) .

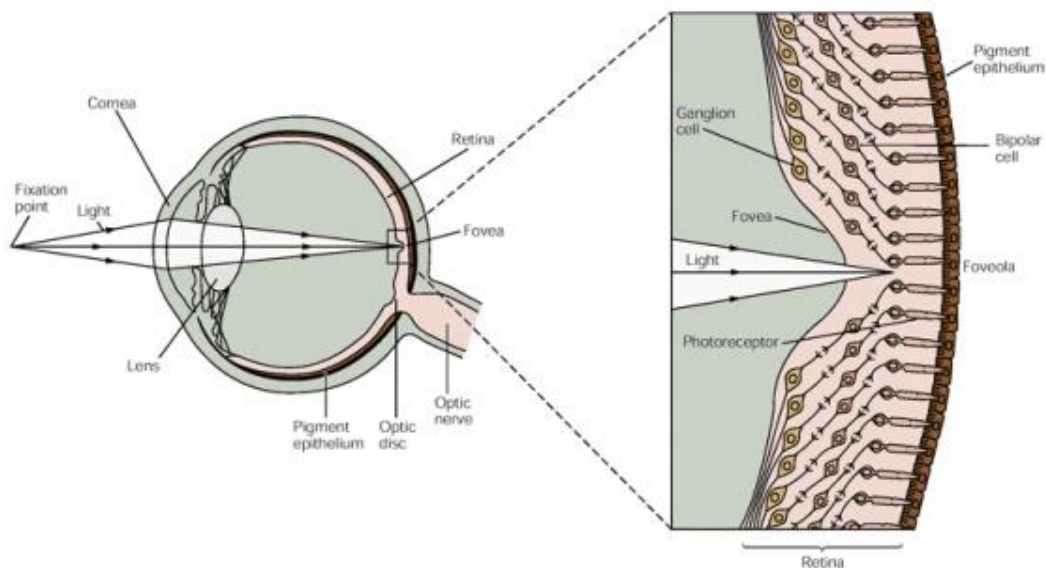


Figure 1 Vertical sagittal section of the human eye and in the right side a schematic enlargement of the retina. Light enters through the cornea, going through the vitreous humor and reaches back of the retina at the fovea(Kandel, Schwartz, & Jessell, 2000).

1.2 The Retina- Distribution of Photoreceptors

After light passes through the cornea, the aqueous humor, the lens, and the vitreous humor, it is focused onto the retinal photoreceptors. The light must pass through a number of retinal layers of nerve fibers, nerve cells, and blood vessels before striking the receptors. These retinal layers (aside from blood vessels) are transparent because of the small size of the elements and the tight packing arrangement (Levin & Adler, 2011).

The retina is the third and inner coat of the eye which is a light sensitive layer of tissue and it lies on the retinal pigment epithelium, a supportive layer of cells that, in turn, rests on the choroidal vascular layer. The inner surface of the retina is in contact with the vitreous. Nutrition is derived to the retina from both the retinal blood vessels, and from the choroid. The retinal blood vessels nourish the inner layers of the retina, while the choroid primarily supplies nutrients (via the retinal pigment epithelium) to the photoreceptor layer.

The retina has three distinctive areas (Fig. 2), the macular lutea, the fovea and the optic disc.

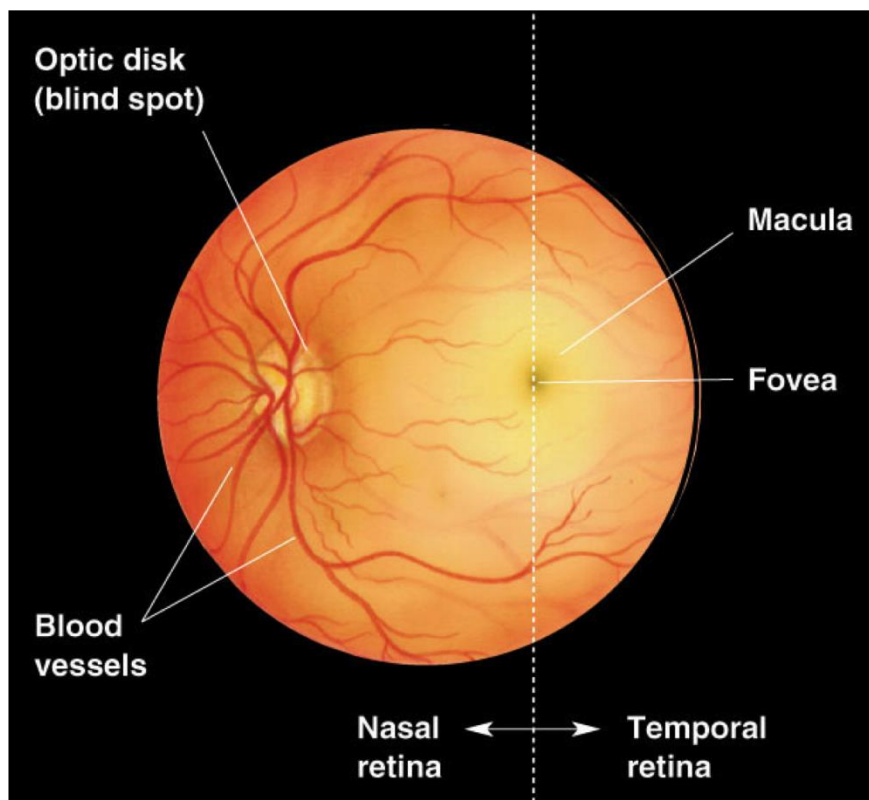


Figure 2 Fundusoscopic view of the human retina. Macula is in the center of the image, and the optic disk is located in the left side.

The macular lutea or area centralis is around 5.5 mm in diameter, centered on the fovea, and located between the superior and inferior temporal arcades. This area corresponds approximately to 17-20 degrees of visual angle.

The fovea is located in the center of the macular lutea and represents the retinal region of greatest visual acuity(Oyster, 1999). It has a diameter of about 1.5 mm, which corresponds to 5 degrees of visual angle. In the center of this area lies foveola which is a small (~1 degree) and also the thinnest (about 150µm) region of the retina, where the cone cells are attached to ganglion cells, providing the sharpest central vision in the retina.

The optic disc, is the point of exit for ganglion cell axons leaving the eye. The optic disc is placed 3 to 4 mm to the nasal side of the fovea. It is a vertical oval, with average dimensions of 1.76mm horizontally by 1.92mm vertically(Duane, Tasman, & Jaeger, 2013). The optic disc is also the entry point for the major blood vessels that supply the retina. There are no photoreceptors in this disc, which creates a blind spot in this area.

The retina consists of five types of neurons, photoreceptors, bipolar cells, ganglion cells, horizontal cells and amacrine cells. The cell bodies and processes of these neurons are stacked in five alternating layers, with the cell bodies located in the inner nuclear, outer nuclear, and ganglion cell layers, and the processes and synaptic contacts located in the inner plexiform and outer plexiform layers (Fig. 3). A direct three-neuron chain—photoreceptor cell to bipolar cell to ganglion cell—is the major route of information flow from photoreceptors to the optic nerve(Purves, 2012).

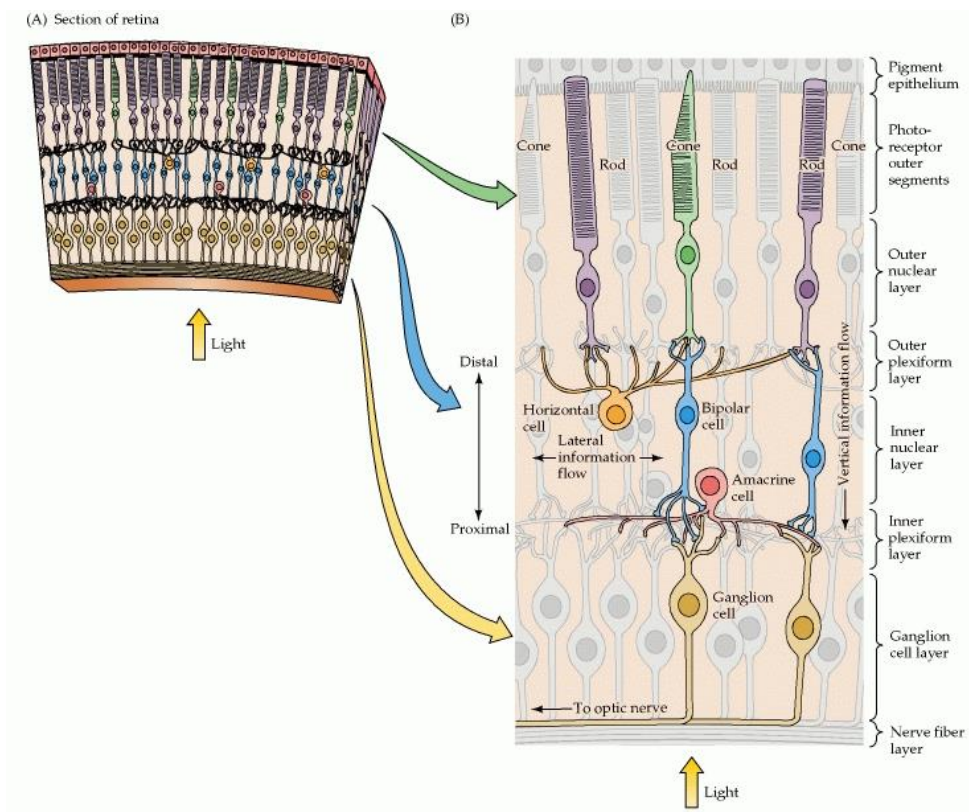


Figure 3 Structure of the retina. (A) Section of the retina showing overall arrangement of retinal layers. (B) Diagram of the basic circuitry of the retina(Purves, 2012).

The human retina contains two types of photoreceptors, rods and cones. Cones are responsible for day vision and rods are specialized for night vision. The distribution

of the photoreceptors across the surface of the retina has important consequences for the vision. The total number of rods in the human retina (about 120 million) far exceeds the number of cones (about 5-6 million). The density of rods is much greater than cones in throughout the most of the retina. However as mentioned before, this ratio between number of cones and rods changes dramatically in the macular lutea, and especially in fovea, where cone density is much greater than any other surface of the retina. In fact, the central 300 μm of the fovea (foveola) is rod free. The following figure (Fig. 4) depicts the distribution of photoreceptor cells in the human eye.

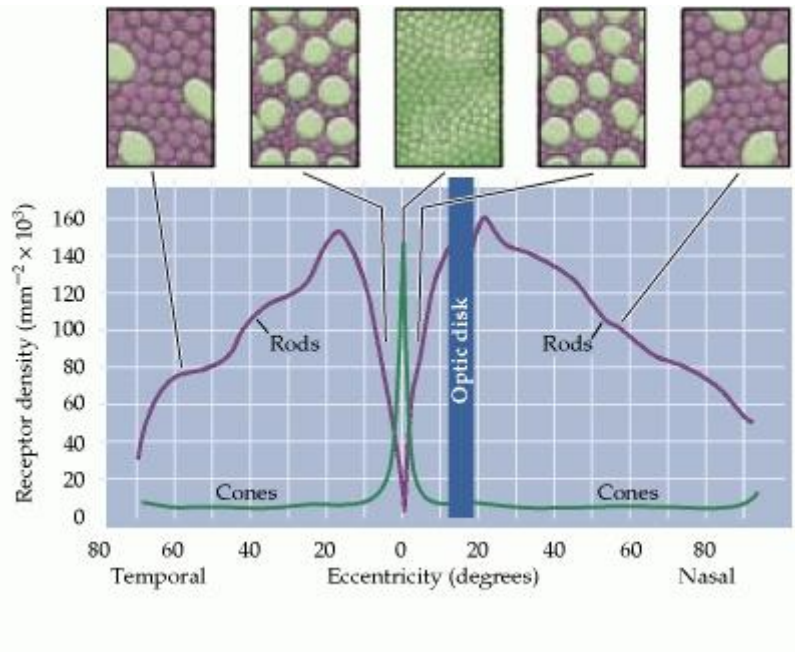


Figure 4 Distribution of Cones and Rods in the human retina(Purves, 2012)

There are three types of cone photoreceptors, which are responsible for color vision. The S-cones which are most sensitive at 420 nm, the M-cones sensitive at 534nm and the L-cones which are most sensitive at 564 nm. (Fig. 5)

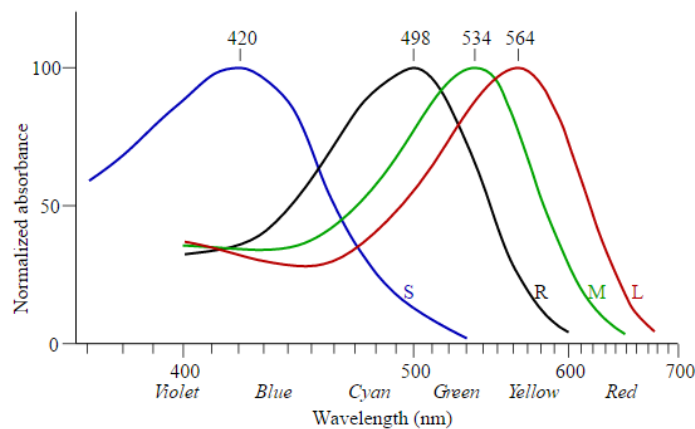


Figure 5 Absorbance of photoreceptors in different wavelengths.

1.3 Later stages of Visual Processing

As mentioned previously, the retina is responsible for the transformation of light to electrical signal, which is then transmitted by ganglion cells for further processing. The axons of the retinal ganglion cells stream toward the optic disc, where they become myelinated and together form the bilateral optic nerves. The optic nerves from each eye project to the optic chiasm, where fibers from each eye destined for one or the other side of the brain are sorted out and rebundled in the bilateral optic tracts, which project to three major subcortical targets: the pretectum, the superior colliculus (the structure which controls saccadic eye movements), and the lateral geniculate nucleus (LGN) (Kandel et al., 2000)(Fig. 6).

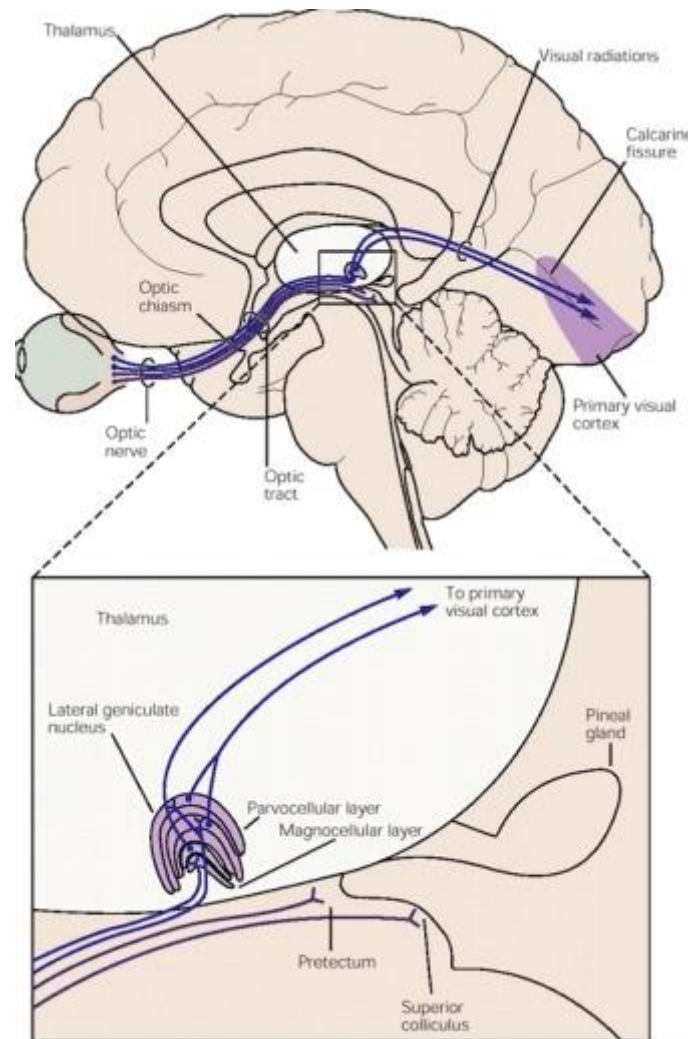


Figure 6 Simplified diagram of the projections from the retina to the visual areas of the thalamus (lateral geniculate nucleus) and midbrain (pretectum and superior colliculus)(Kandel et al., 2000).

The lateral geniculate nucleus is the main terminus for input to the visual cortex, as 90% of the retinal axons terminate there. Without this pathway visual perception is lost, although some very limited stimulus detection and movement toward objects in the visual field still is possible. This residual vision, possibly mediated by the visual

pathway passing through the superior colliculus, has been called blindsight(Kandel et al., 2000).

There are three types of retinal ganglion cells, P cells (parvocellular, or P pathway), M cells (magnocellular, or M pathway) and Konio cells (koniocellular, or K pathway). The three kinds of cells transfer different information and they transmit it to different layers of the lateral geniculate nucleus (LGN), which contains six layers of cell bodies separated by intralaminar layers of axons and dendrites. The layers are numbered from 1 to 6, ventral to dorsal(Fig.7)(Kandel et al., 2000).

The two most ventral layers of the nucleus contain relatively large cells and are known as the magnocellular layers; their main retinal input is from M ganglion cells. The four dorsal layers are known as parvocellular layers and receive input from P ganglion cells. Between the parvocellular layers there are some very thin layers, the konio layers. The three different layers project to three different layers of the primary visual cortex (V₁).M pathway projects to 4Ca layer of the V₁, the P pathway projects to 4Cb layer and koniocellular pathway projects to layer 3 of the V₁ . After V₁ the pathways continue to V₂ and other parts of the extrastriate cortex for higher processing.

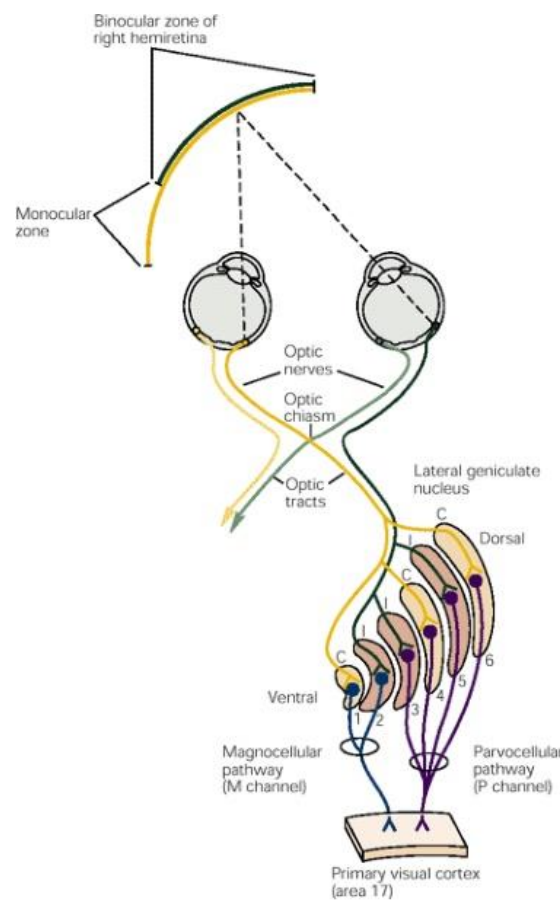


Figure 7 The lateral geniculate nucleus is the principal subcortical site for processing information. Inputs from the right hemiretina of each eye project to different layers of the right lateral geniculate nucleus to create a complete representation of the left visual hemifield. Similarly, fibers from the left hemiretina of each eye project to the left lateral geniculate nucleus(Kandel et al., 2000).

2. Extraocular Muscles

Movement of the eyes, is controlled by a set of six muscles, which form three complementary pairs. Also there is another extraocular muscle, which controls eyelid elevation (levator palpebrae). The eye's orientation is defined by three axes of rotation (horizontal, vertical and torsional), and eye movements can be described as rotations around these axes.

The six extraocular muscles, which control eye movement are the medial, lateral, superior and inferior recti, the superior and inferior oblique (Fig.8). The four recti originate from the tendinous ring around the optic canal, run forward in the orbit and insert into the eyeball at the medial, lateral, superior and inferior side respectively. The obliquus superior arises from the sphenoid bone just superior to the tendinous ring, runs forward to the trochlea, a fibrous pulley located at the superior and medial angle of the orbital anterior margin (Kjellgren, 2004).

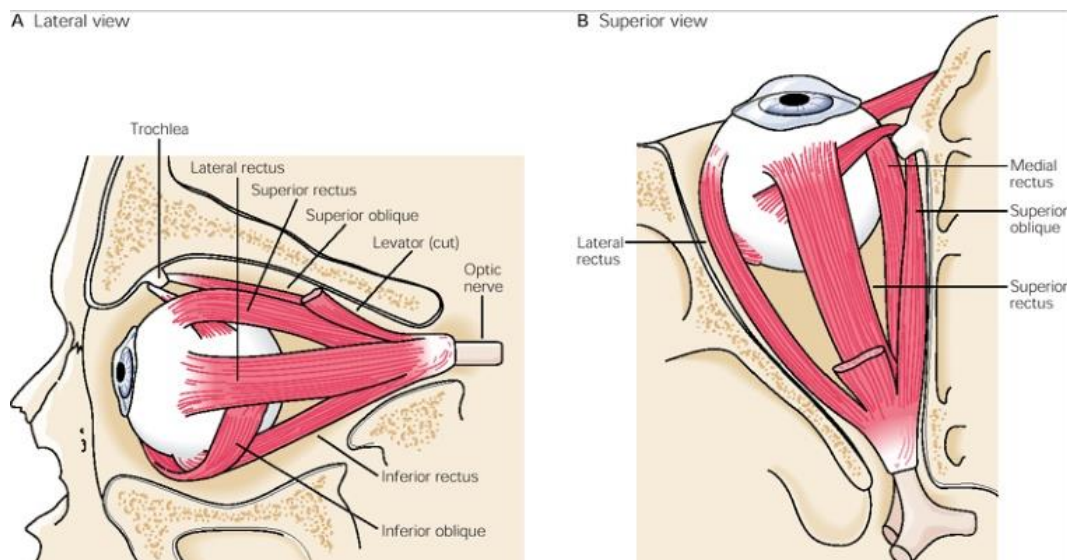


Figure 8 Lateral (A) and Superior (B) view of extraocular muscles in the human eye (Kandel et al., 2000).

Each one of the extraocular muscles, is responsible for one specific, or a combination of eye movements. The medial rectus adducts the eye and the lateral rectus abducts it. The actions of the four remaining muscles are complicated because they do not perform purely vertical or torsional rotations but a combination of the two. The following table (Table 1) presents actions of extraocular muscles.

Table 1 Primary and Secondary actions of extraocular muscles

Muscle	Primary Action	Secondary Action
Medial rectus	Abduction	None
Lateral rectus	Abduction	None
Superior rectus	Elevation	Intorsion
Inferior rectus	Depression	Extorsion
Superior oblique	Intorsion	Depression
Inferior oblique	Extorsion	Elevation

3. Gaze System

As mentioned before, the quality of vision is better in a certain area in the center of the retina (fovea), as the density of cone cells in this area is very high. However, the small size of this area (about 1.5 mm diameter) doesn't allow detection of a big part of the visual field. The anatomy of human eye, gives us the ability to observe objects in the world, but if we want to examine one of them we have to move fovea to it. This function is performed by the gaze system (Kandel et al., 2000).

The main function of the gaze system is to capture images of the surrounding environment towards the fovea area, and also stabilize the image even when the object moves or the head moves. Gaze system, consists of three subsystems: The Oculomotor system, the Head Movement system and the Fixation system.

3.1 The Oculomotor System

The oculomotor system, supports the observation of steady and not-steady images, as it is responsible for the movements of the eyes in the orbit. These movements are performed by the extraocular muscles mentioned in Chapter 2. The oculomotor system is responsible for three kind of ocular movements, the saccades, the smooth pursuit movements and the vergence movements.

3.1.1 Saccades

Saccades are fast ballistic, conjugate eye movements that people produce as they scan a visual image (Fig.9). They are typically followed by a fixation which is a period of time during which the eye is stationary(Liversedge, Gilchrist, & Everling). Saccades occur very frequently (up to 173.000 per day)(Abrams, Meyer, & Kornblum, 1989), and have a significant role in the construction of perceptual representation of the surrounding environment(Rayner, 1998).

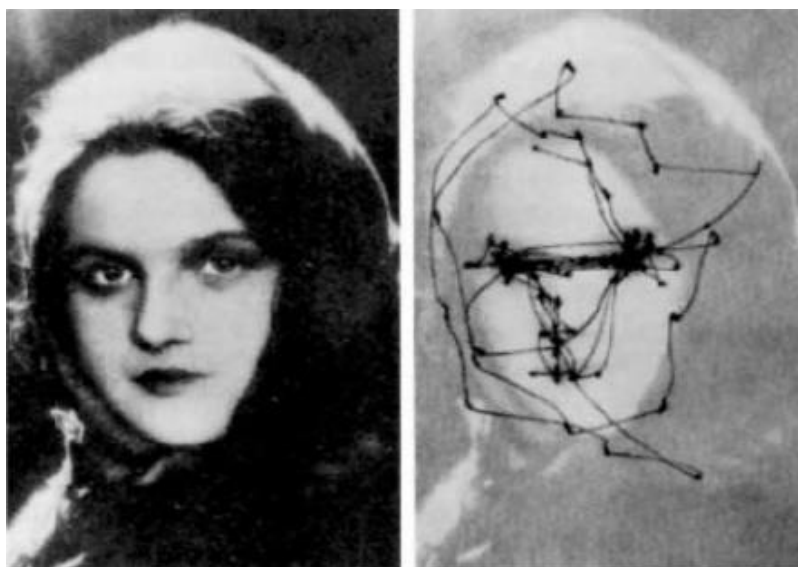


Figure 9 Photograph of a girl's face and record of the eye movements during free examination of the photograph with both eyes in one minute(Yarbus, 1967).

Saccades range in amplitude from the small movements made while reading, to the much larger movements made while gazing around a wider visual image. Saccades can be elicited voluntarily, but occur reflexively whenever the eyes are open, even when fixated on a target. The rapid eye movements that occur during an important phase of sleep are also saccades (Purves, 2012). The size of the target, determines the direction and amplitude of a saccade, but its velocity and duration cannot be controlled (Kandel et al., 2000).

Ordinarily there is no time for visual feedback to modify the course of the saccade and corrections to the direction of movement are made in successive saccades. Saccadic velocity can be slowed down only by fatigue, drugs and pathological states.

Saccades are one of the fastest movements produced by the human body ($30-700^\circ/s$) and also they have very small duration (about 30-100 msec).

3.1.2 Smooth pursuit eye movements

Smooth pursuit eye movements are responsible for the ability to observe moving objects in the world, as they center and stabilize image to the fovea (Spering, Kerzel, Braun, Hawken, & Gegenfurtner, 2005). The smooth pursuit eye movements keep the image of a moving target on the fovea, by moving the eyes as fast as the target.

The system requires a moving stimulus in order to calculate the proper eye velocity, which means that a verbal command or an imagined stimulus cannot produce smooth pursuit. Smooth pursuit movements have a maximum velocity of about $100^\circ/s$, much slower than saccades. Drugs, fatigue, alcohol, and even distraction degrade the quality of these movements (Kandel et al., 2000).

3.1.3 Vergence eye movements

Vergence movements are responsible for the ability to align the fovea of each eye with targets located at different distances from the observer. Unlike other eye movements, where the eyes move in the same direction (conjugate eye movements), vergence movements are disconjugate, they involve either a convergence or divergence of the lines of sight of each eye to see an object that is nearer or farther away. Convergence is one of the three reflexive visual responses elicited by interest in a near object. The other components of the so-called near reflex triad are accommodation of the lens, which brings the object into focus, and pupillary constriction, which increases the depth of field and sharpens the image on the retina (Purves, 2012). The dynamics of vergence are shown in Figure 10, the vergence movement to a sudden target appearance is slower than a saccadic movement, and after the target disappears the eyes gradually diverge again, maintaining the point of sight at a 'dark focus' about 1-2 m from the eyes (Bridgeman, 2012).

Vergence movements, show two types of responses according to their speed. Fast vergence is observed in response to a large and sudden change of disparity, for example when someone wants to change focus from near to far. Slow vergence is seen as a response to a small and slow change of disparity, such as tracking a target which moves slowly in depth. Convergence speed is usually greater than divergence

speed. The latency of the vergence movements is about 200 msec for retinal blur stimuli and 80-160 msec for retinal disparity stimuli(Wong, 2008).

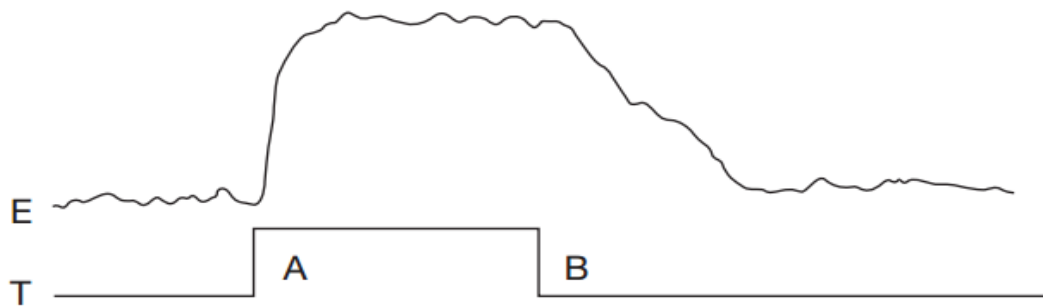


Figure 10 Vergence eye movements (E) in response to a near target (T). The target remains visible for 5s (from A to B)(Bridgeman, 2012).

3.2 The Fixation System

Human eyes are continually moving, even while our gaze system fixates on a target. Eye movements during fixations, are involuntary and very significant for our vision. We are able to detect stationary objects in the surrounding environment as neurons in the retina, mainly detect and compare changes in luminance and other aspects of objects, while in the other hand stationary images are ignored.

The three primary types of movements during visual fixations are: tremor, drifts and microsaccades (Fig. 11).



Figure 11 Cartoon representation of fixational eye movements in humans and primates. Microsaccades (straight and fast movements), drifts (curvy slow movements), and tremor (oscillations superimposed on drifts) transport the visual image across the retinal photoreceptor mosaic (Martinez-Conde & Macknik, 2007).

3.2.1 Tremor

Tremor (also called physiological nystagmus), is a high-frequency, involuntary movement of the eye. Tremor is a very fast (~90 Hz) eye movement with extremely small amplitude, which is approximately equal to the diameter of a cone in the fovea area (~ 0.5 arcmins)(Martinez-Conde & Macknik, 2007) . Usually tremor amplitudes and frequencies are usually near the level of the recording system's noise, which means that the recording of these eye movements is extremely difficult. Tremor's role on vision is not yet defined as its frequency is so high that the image's tremor is

not recognizable by the eye. Tremor is different in each eye with a possible effect on stereoscopic vision.

3.2.2 Drifts

Drifts are slow curvy movements, which occur in the same time period with tremor, between microsaccades. Drifts' velocity is less than 20 arcmin/s. They move the retinal image about 5-15 photoreceptors. They have been recorded both as conjugate and as disconjugate eye movements.

Drifts' existence was attributed to the instability of the extraocular muscles and to their antagonistic role with the microsaccades. However some recent studies have stated that they greatly contribute to the fixation's accuracy and that they prevent the image of a stable object from fading.

3.2.3 Microsaccades

Microsaccades are the fastest and largest of the three types of fixational eye movements. They are small saccades (mostly <0.5 degrees) occurring involuntary during fixation. They travel in a straight trajectory, carrying the retinal image across a range of several dozen to several hundred photoreceptor widths(Martinez-Conde & Macknik, 2007).

Microsaccades' amplitude vary from 5-120 arcmins(Ditchburn, 1980), but they cannot be defined according to their amplitude alone, because small exploratory or voluntary saccades can be the same size as microsaccades. Microsaccades can be defined only operationally, as the involuntary saccades that are produced while the subject attempts to fixate(Martinez-Conde, 2006). They have a relatively constant duration of 25 msec and for this reason a linear correlation of their velocity with their amplitude is being observed (Fig. 12).

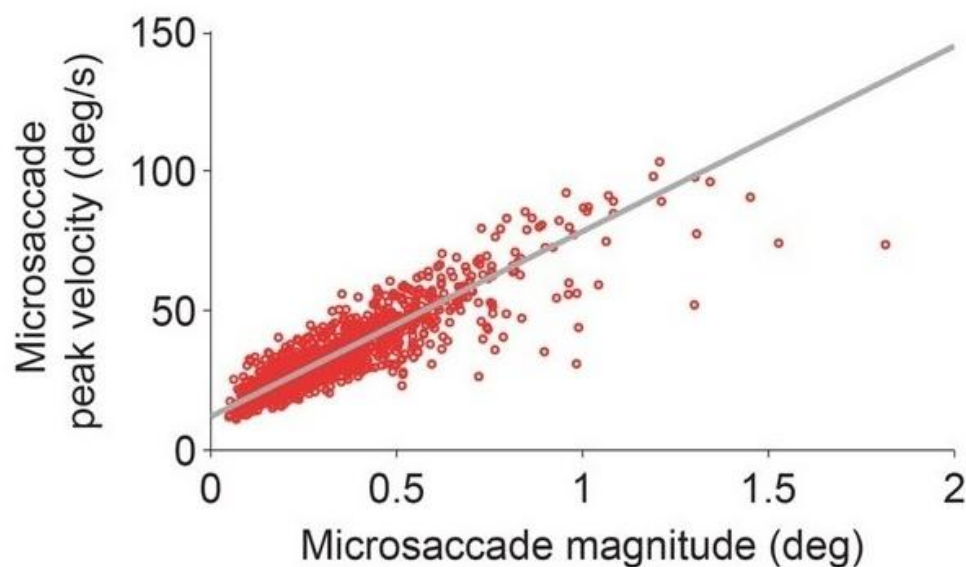


Figure 12 Microsaccadic peak velocity–magnitude relationship(McCamy et al., 2013)

A possible role for microsaccades is to correct displacements in eye position produced by drifts of the eye during fixation. For example, if drift carries the fixation target away from the fovea, microsaccades tend to bring the target back. Other studies suggest that microsaccades may counteract receptor adaptation on a short timescale and correct fixation errors on a longer timescale.

Microsaccades and voluntary saccades may be generated by the same mechanisms (Zuber, Stark, & Cook, 1965), however microsaccades cannot be voluntary movements. Microsaccades in the two eyes tend to be conjugate (Yarbus, 1967). The involuntary nature of microsaccades may indicate a subcortical control mechanism for their production.

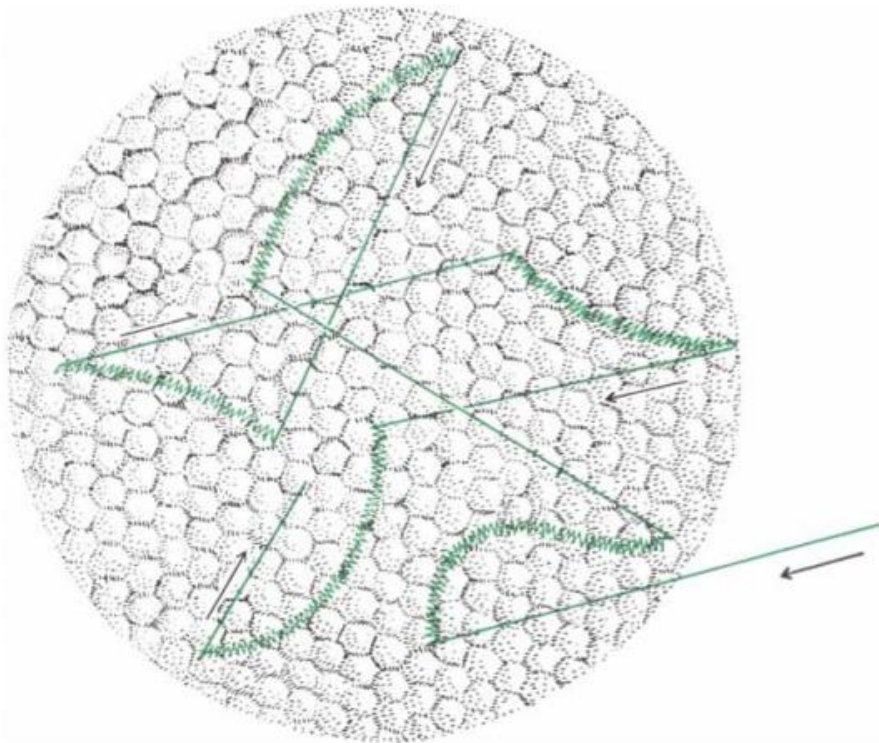


Figure 13 Eye movements that are halted in stabilized vision normally carry an image across the receptors of the retina as shown here. The three movements are a drift (curved lines) away from the center of vision, a faster flick (straight lines) back toward the center and a high frequency tremor superimposed on the drift. The magnitude of all these movements is very small; the diameter of the patch of the fovea shown above is only 0.05 mm (Pritchard, 1961).

3.2.4 Fixation Stability

Intrafixational eye movements (drift, microsaccades and physiological tremor) all lead to some degree of within-fixation instability. Vision researchers have used many definitions and measurements to evaluate the fixation stability. There are two common measures, which are used widely for the evaluation of fixation stability.

The first one is the Bivariate Contour Ellipse Area (BCEA), which is the area of the confidence ellipse in a bivariate distribution. This measure assumes normality of positions distributions (vertically and horizontally).

The second measure is the within-isolines area, which does not assume the normality of the underlying positions.

Each of these measures can be applied to data collected by eyetrackers, so we can determine the level of variability between fixations.

The calculation of BCEA will be further presented in chapter 5.5.

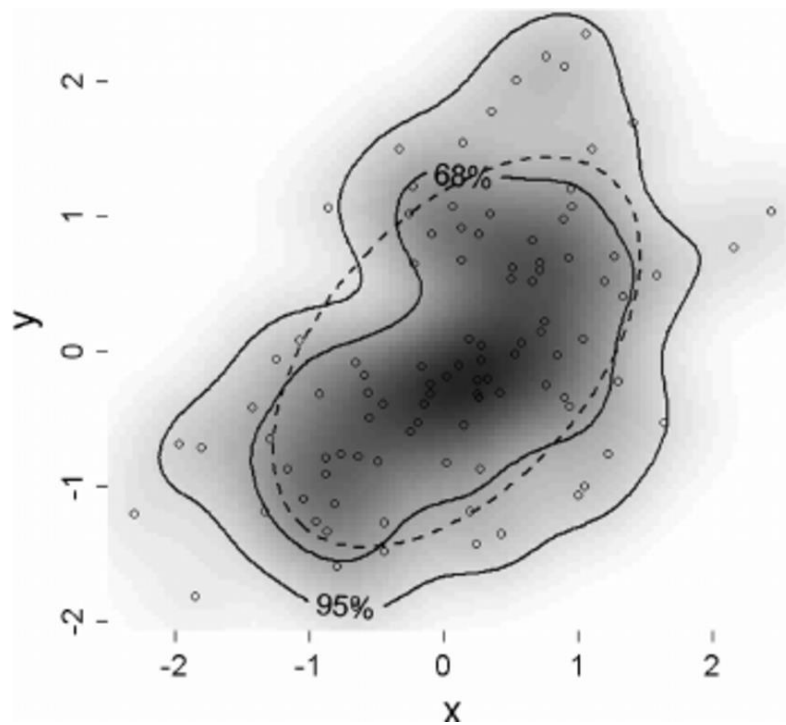


Figure 14 Scatterplot of 100 points drawn from a bivariate Gaussian random variable. 95 and 68% isolines are superimposed (solid lines). 68% BCEA is represented by a dashed line. Probability density is mapped to grey levels (Castet & Crossland, 2012).

3.3 The Head Movement System

We have presented previously the ocular movements, when the head is still, but usually when we look around our head is moving. The size of the voluntary and involuntary movements of head affects the amplitude and direction of eye movements.

The head movement system, is controlled by two sub-systems, the vestibular-ocular system and the optokinetic system.

3.3.1 Vestibular-Ocular System

The Vestibular-Ocular system also called Vestibular-Ocular Reflex (VOR) stabilizes retinal images during head movements by counter-rotating the eyes at the same speed as the head but in the opposite direction. Image stabilization by vestibule processing is much faster and more efficient than visual processing, performed by other kinds of eye movements. This is because visual information takes about 100 msec to travel from the visual cortices through a series of brain structures, to the ocular motoneurons that move the eyes. On the contrary, vestibular information takes only about 7-15 msec to travel from the vestibular sensors, through the brainstem, to the ocular motoneurons. This short latency allows the eyes to compensate for the rapid oscillation of the head(Kandel et al., 2000).

3.3.2 Optokinetic System

The optokinetic system, functions supplementary to the vestibular-ocular reflexes. The vestibular apparatus is not a perfect transducer of head movement. It has two serious problems. First, it habituates. In the dark nystagmus does not continue as long as the head moves, but gradually slows and stops. It stops because the semicircular canals habituate exponentially with a time constant of 5 s. Brain stem circuitry extends the time constant of vestibular nystagmus to 15 s, but during sustained rotation the vestibular signal ultimately fails and the eyes begin to move in space. Second, the canals do not respond well to very slow head movement. To compensate for such deficiencies in the vestibular apparatus the optokinetic system provides the central vestibular system with visual information that is used to stabilize the eyes(Kandel et al., 2000).

4. Eye movements during reading

During reading, people move their eyes to different locations, with saccade movements of different latency. The variation of latency between movements during reading is related to variation in the local and global processing requirements of the text (Yang & McConkie, 2001). These saccade movements have different amplitude but also different direction (to the front or to the back of the text). Between saccadic movements there are fixations with variable duration times.

Although the vast majority of eye movements (saccades) progress with the text, readers do not necessarily fixate at each word. Skipping short words is relatively common, and some long words receive more than one fixation. Approximately 10% to 15% of saccades (called regressions) move the eyes in the direction opposite to word order (Rayner, Juhasz, & Pollatsek, 2008). Regressions are a natural part of the reading process. In cases when they are eliminated, by forcing presentation of just one word after the other, comprehension of the text falls dramatically (Schotter, Tran, & Rayner, 2014).

Typically eyes move from one word to another, but they sometimes make an additional fixation on the same fixated word, a case which is called within-word refixation. The probability of refixating to a word, depends on the eyes' initial landing on the word. This happens because, as it is known, the location of the eyes in a word affects the amount of visual information which can be extracted from a word. The closer the eyes are to the middle of the word, the more letters of the word can be processed (Nazir, 1991). Nearly two-thirds of the words are fixated, some words being skipped in the first eye pass on the line of text. Only a few skipped words are subsequently fixated following an inter-word regression.

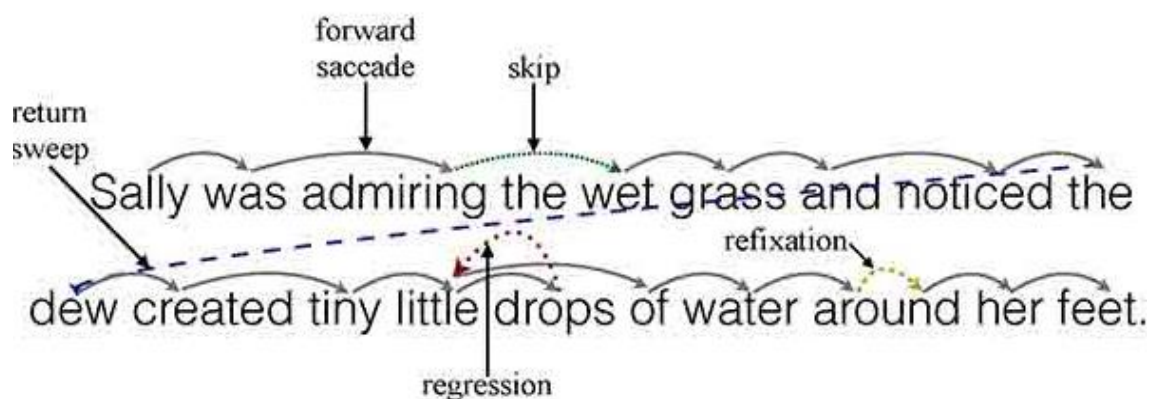


Figure 15 Example of eye movements during reading (Rayner et al., 2015).

4.1 The basic Characteristics of Eye Movements during Reading

The eyes during reading are moving (saccadic eye movements which last approximately 20-40 ms), or they are relatively stable (when they fixate). For skilled readers, the duration of fixations is about 200–250ms(Rayner et al., 2008). Visual information which is acquired from the text is obtained mainly during the fixations, as vision is suppressed during the saccades(Wolverton & Zola, 1983).

Most of the time during reading, the eyes move forward through the text from left to right across the line. At the end of each line, readers move their eyes to the beginning of the next line (via a return sweep). This return sweep is often inaccurate, as a backward corrective saccade is needed to fixate in the beginning of the next line. Even with this corrective saccade, the location of the left-most fixation on a line is typically 5–7 letter spaces from the first letter on the line. Likewise, the last fixation on a line is typically approximately 5–7 letters from the end of the line.(Rayner et al., 2008).

As mentioned before, skilled readers move their eyes backwards (regressions), to read previously processed words. It is assumed that regressions are associated with comprehension difficulties. However, most of the regressions that readers make are actually quite short saccades (often going back only a word or two in the text) and probably reflect oculomotor variability (i.e., overshooting the target word) or word recognition problems.

The average amplitude of saccades during reading is approximately 7-8 letter spaces, which is equal to 2-3 degrees of visual angle, for normal sized texts. In reading, the number of letter spaces is more important than the visual angle. If text size is held constant, but the viewing distance is altered (so more letters fall into a degree of visual angle when the text is held far from the eyes), the number of letters rather than the visual angle determines how far the eyes move (Morrison & Rayner, 1981).

The average fixation duration during reading, is considered to be about 200-250 ms and the average saccade size about 7-8 letter spaces, but there is considerable variability on those measurements.

Table 2 Developmental Characteristics of Eye Movements during Reading (K. Rayner, 1998)

	Grade level						
	1	2	3	4	5	6	Adult
Fixation Duration (ms)	355	306	286	266	255	249	233
Fixations per 100 words	191	151	131	121	117	106	94
Regression Frequency (%)	28	26	25	26	26	22	14

An important factor which affects the above described characteristics is the level of education of each reader. In the following table (Table 2) we can see the different characteristics between adults and students. It is obvious that adults who are also skilled readers make less fixations during reading, which also are fixations of shorter

duration than students. The percentage of regression movements is also less for skilled readers.

All the characteristics mentioned are related to eye movements which occur while reading texts in alphabetic systems like Greek, English etc. Eye movements and their characteristics are affected while reading non-alphabetic texts, like Japanese, Chinese etc. Fixation durations are longer and the saccades are shorter, because each character conveys more information than a letter or a word in an alphabetic system.

4.1.1 Fixation Duration during Reading

The average fixation duration is about 200-250 ms. Fixation durations during reading may vary from shorter durations about 50-75 ms to longer, about 500-700 ms, the following figure (Fig.16) shows the frequency distribution of fixation durations during reading.

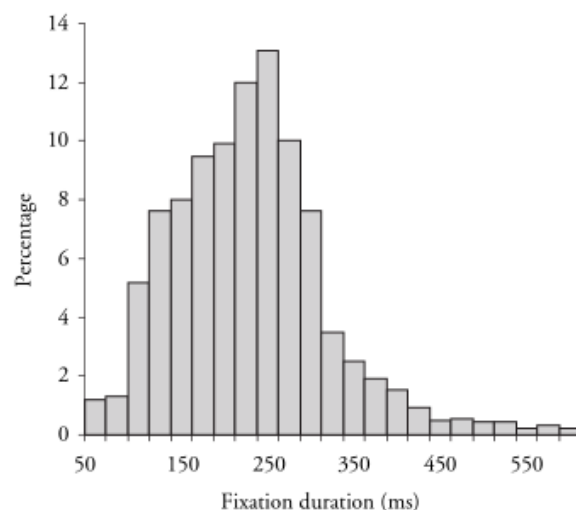


Figure 16 Fixation Duration Distribution (Rayner et al., 2008)

It has been shown that word frequency has a great effect on it. Readers spend more time on lower frequency words than on higher frequency words. Words that are highly predictable from previous context are also fixated for less time. For this reason, function words are fixated only about 35% of the reading time, while content words are fixated about 85% of the time (Starr & Rayner, 2001).

It has been shown that words with alternating case (e.g. AlTeRnAtInG cAsE) were fixated longer than those which were presented in normal case (Juhasz, Liversedge, White, & Rayner, 2006), giving more evidence on the processing models.

The initial fixation location, is another factor which affects fixation duration, as we can see in the following figure (Fig. 17) fixation duration is longer when the fixation is near the center of short words and to the left of the center of long words. This

phenomenon applies both to first fixations and second fixation(Liversedge, Gilchrist, & Everling, 2011).

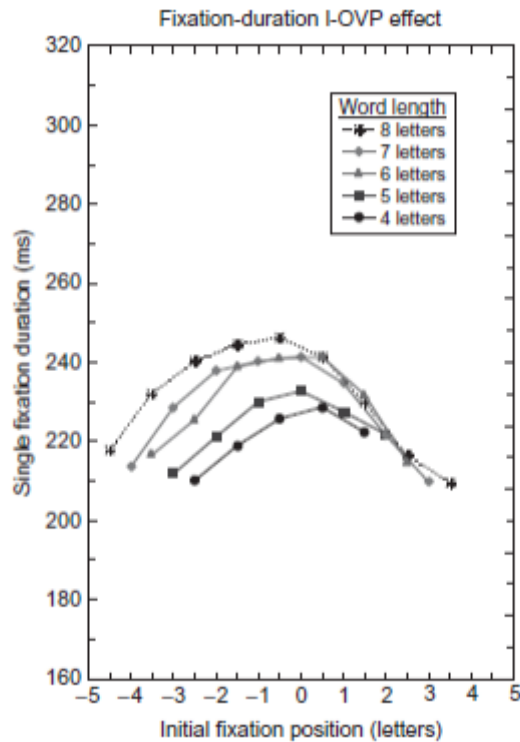


Figure 17 Fixation duration as function of initial fixation position.

Fixation duration is also affected by monocular vision. Johansson et al. showed that monocular reading results significant increase in fixation duration at 8.9% (Johansson, Pansell, Ygge, & Seimyr, 2014).

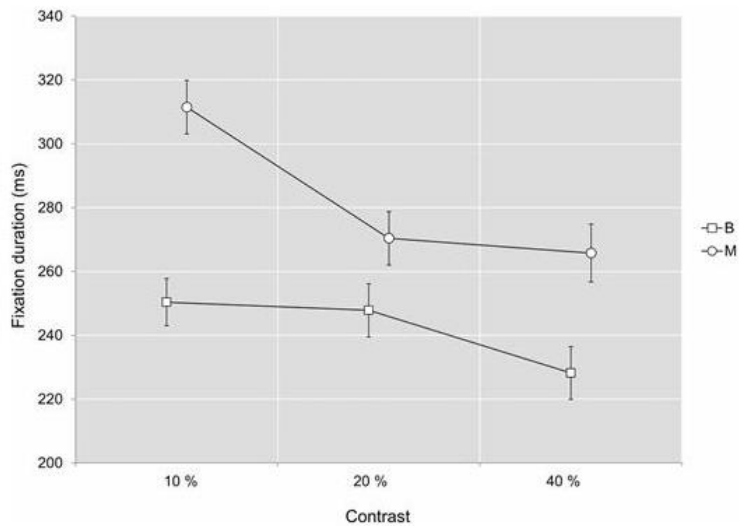


Figure 18 Binocular (squares) and monocular (circles) fixation duration at three different levels of contrast (Johansson et al., 2014)

Similarly to fixation duration, some saccades are only 1 letter space, while others are as long as 20–25 letter spaces (Fig. 19). We should mention that those bigger saccades, occur most of the times after regressions as the reader wants to continue reading from the part of the text where he or she was prior to launching the regression.

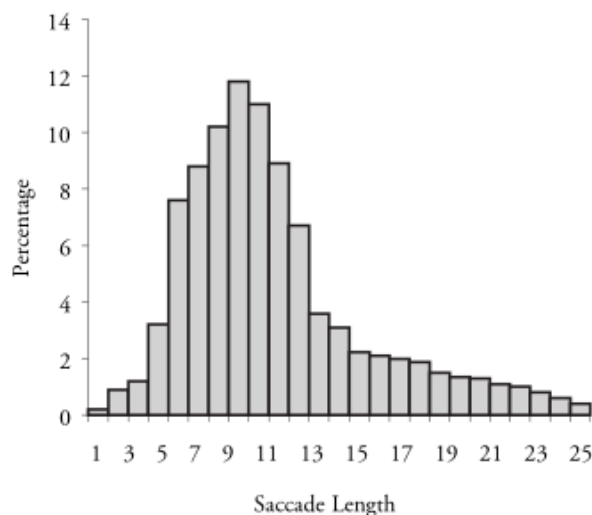


Figure 19 Forward Saccade Length Distribution (Rayner et al., 2008)

Recently researchers Staub and Benatar (Staub & Benatar, 2013) have analyzed distributions of fixation durations and concluded that individual readers vary in the time required for normal perceptual and linguistic processing during eye fixations. They analyzed correlations between the ex-Gaussian parameters fitted to individual readers' distributions, and suggested that the location of a given reader's distribution along the horizontal axis and the degree of skew may be independent. Their analysis also suggested that differences in these distributions reflect both the speed of word recognition and the frequency with which readers encounter disruption or difficulty during comprehension processes, though more work must be done to address this.

4.1.2 Word skipping and initial landing sites in words

Some words are skipped during the first eye pass. These are mainly the shortest words. The likelihood of word skipping dramatically decreases with word length, dropping from about 0.76 for 1- and 2-letter words to about 0.42 for 4-letter words and 0.05 for words of 9–10 letters. Word skipping also depends strongly on the location of the prior eye fixation on the line of text, that is, the eyes' launch-site distance to the beginning of a word; for launch sites of 1 letter, the probability of skipping a 5-letter word is near 0.65, but this decreases to about 0.30 for launch sites of 7–8 letters, and gets near zero for launch sites of 10 letters and more (Liversedge et al., 2011).

In a manner similar to word-skipping behavior, the eyes' landing sites in words greatly vary with word length and launch site. The effect of word length exemplifies

in the well-known preferred viewing (/landing) position effect (or PVP/PLP effect) (Rayner, 1979). This shows, in languages read from left to right that the eyes' landing position in words is most frequently towards the center of short words (5 letters) and slightly to the left of the center of long words.

As shown in Figure 20, there is still some variability in where the eyes land on words, the typical Gaussian-shape distribution of landing sites extending from the very-beginning to the end of words.

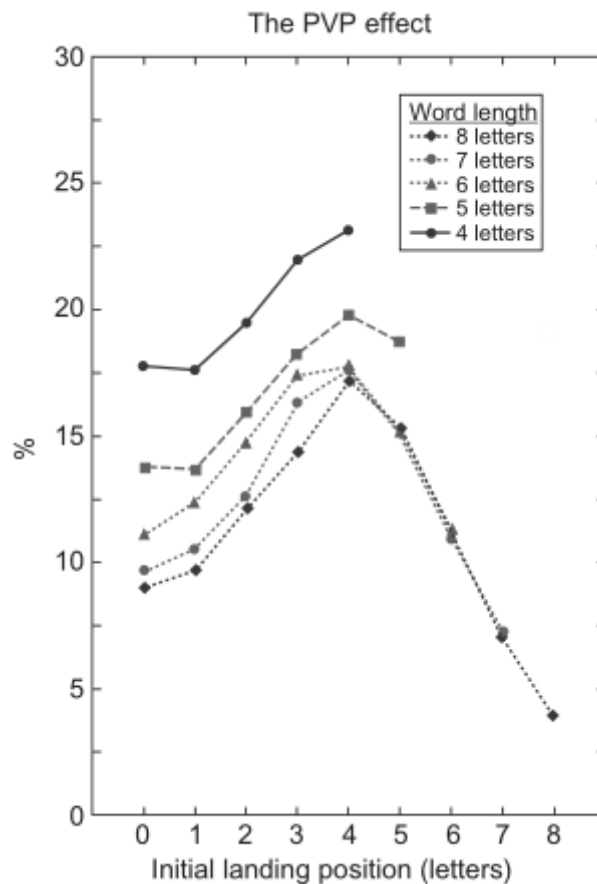


Figure 20 the preferred viewing position effect as shown by the distribution of the eyes' initial landing sites in words for the case of adult readers (Vitu et. al, 2001; Rayner, 1971)

4.1.3. Probability of Refixation

The probability of refixating a word is lower when the eyes initially land in the central region of the word than when they first fixate the beginning or end of the word. This first phenomenon has a typical U shape, with a minimum at the center of short words and to the left of the center of long words; it is referred to as the refixation-optimal viewing position effect (or refixation-OVP effect). It is related to another phenomenon, the OVP effect, showing that a word presented in isolation is more easily and more quickly identified when the eyes first fixate near the word's

center. The refixation-OVP effect was originally shown during the recognition of isolated words presented at variable locations relative to a previously displayed fixation point (Liversedge et al., 2011).

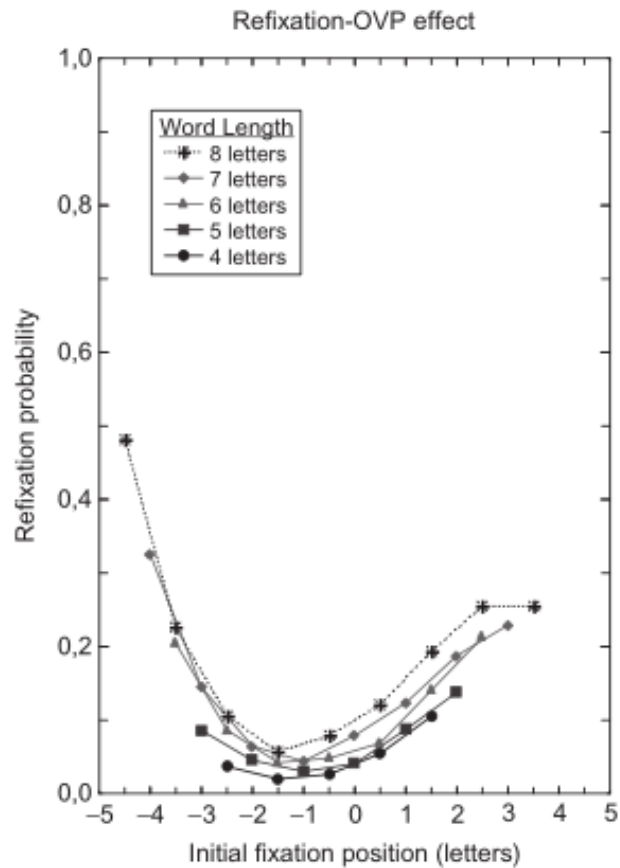


Figure 21 Refixation probability as a function of Initial Fixation Position(Liversedge et al., 2011).

4.1.4 Fixational Eye Movements during Reading

In Chapter 3 we have discussed the existence of eye movements, even when the eye is theoretically still (fixation). Likewise the three basic types of movements during fixations are present in fixations during reading

Older studies suggested that microsaccades (using the classical definition of a saccade smaller than 12 min arc), almost never occur during reading (Kowler & Anton, 1987; Kunitz & Steinman, 1968; Schnitzer & Kowler, 2006). Microsaccades might be expected in such tasks if only to clean up any errors in landing position left behind by large, primary saccades. However, microsaccades—and corrective saccades in general—are rare when saccadic targets are spatially extended shapes, rather than small target points(Collewijn & Kowler, 2008).

The original studies about this topic, reported that microsaccades occurred on 1.7% of fixations, and in other studies on 4.8% of fixations(Kunitz & Steinman, 1968), while the median fixation duration for these fixations (containing microsaccades) was 535 ms and 520 ms in each case, in contrast to 285 to 305 msec for fixations with no microsaccades.

Later studies have reported larger microsaccades (Martinez-Conde, Macknik, Troncoso, & Dyar, 2006) using a steady target, than the microsaccades found in the classical studies, which used the contact lens optical lever to determine saccadic movements. The results of these studies has created a confusion between researchers because the role of saccades, with amplitude greater than 20 arcmins, was undisputed (Collewijn & Kowler, 2008), as they are able to bring the image to a more central retinal location. Those studies have also revealed a direct link between suppression of microsaccades and visual fading, which means that there is a strong relationship between microsaccades production and visibility during fixation. Considering the conclusions from recent studies we can assume that the presence of microsaccades could be significant for a demanding task like reading.

In the present moment, there are not any studies that we know of regarding the existence and the role of microsaccades in fixations during reading, except the original studies which used the original definition of Microsaccadic movements, and this will be a major issue we want to discuss in this study, as we expect to find more microsaccades (including saccades bigger than 20 arcmins) using an algorithm for saccade detection.

4.1.5. Evaluation of Reading Performance

Clinical researchers have used several measures to evaluate reading performance as a whole. The evaluation of reading performance is very important for clinical purposes, as patients facing eye diseases (e.g. AMD) are facing also reading difficulties. Such measures are reading speed, threshold print size and reading acuity among others.

Reading speed is a strong predictor of visual ability and vision-related quality of life. From this, we would expect reading performance to be one of the more important outcome measures for judging the effectiveness of therapeutic interventions and vision rehabilitation (Rubin, 2013).

Several test charts have been used to quantify reading performance, like MNREAD test for low vision reading acuity which allows us to measure reading speed across a range of letter sizes but there are only two different texts, while the International Reading Speed Tests (IReST) has more advantages, as it is available in multiple languages and the number of texts (10) make it possible to do longitudinal studies without repeating passages, one of the main disadvantage of IReST charts is that it is available only in one font size so we can't evaluate the reading performance in smaller font size.

4.2 Perceptual Span in Reading

As we know vision is better in the center of the fovea (foveola) and worst in the periphery (parafovea). This anisotropy lead researchers to question the extraction of information from the parafoveal vision.

The perceptual span is the number of letters that can be extracted from a single fixation(Liversedge et al., 2011). Several studies have concluded that perceptual span is relatively small, as it ranges from 3-4 letters to the left of the fixation up to 14-15 letters to the right of fixation, however in order to identify words we need information that can be taken only up to about 7-8 letters to the right of the fixation (George W. McConkie & Rayner, 1975; Starr & Rayner, 2001).

There is also evidence that the characteristics of the writing system affect the asymmetry of the span, and the results are very different for languages with ideographic scripts, like Chinese and Japanese. Ikeda and Saida (Ikeda & Saida, 1978) found that the perceptual span for readers of Japanese was about 13 character spaces, which is considerably smaller than that of English readers.

Another important finding, we should mention is that the size of perceptual span is not constant but varies as a function of text difficulty (A. W. Inhoff, Pollatsek, Posner, & Rayner, 1989; Rayner, 1986). Rayner found that the size of the span was smaller when text was difficult to read. When fourth-grade children were given reading-level-appropriate text, the size of their perceptual span was similar to that of adult readers. However, when they were given college-level text, their span became much smaller.

The fact that there is a higher probability of skipping a word if it is a short function word or if it's highly predictable, is a simple indication that there is some kind of parafoveal process. There are many low-level factors that affect reading behavior through the parafoveal process. Some of these effects are the following. Saccade length is influenced by both the length of the fixated word and the next word. The further the launch site of a saccade, the closer to the beginning of the word the landing site is. Also, if the beginning of a word contains an orthographically irregular letter, the initial landing position of the eyes shifts to the beginning of the word(Liversedge et al., 2011).

There is little evidence on the effect of higher-level factors on the landing position of the eyes. There is however, high interest in the effect of parafoveal words on fixation durations. It has been indicated that if parafoveal information is denied, reading rates decrease rapidly(Liversedge et al., 2011).

4.3 The control of eye movements during reading

Readers make unconsciously two important decisions, while reading, when and where to move the eyes. We have a lot of evidence which suggest that these two decisions are made independently of each other (Rayner, 1998; Rayner, Kambe, & Duffy, 2000).

A very important factor which affects the decision about where to move the eyes during reading, is word length. There is evidence that saccade lengths are significantly decreased when information about word boundaries is withheld (Rayner, 1998). As mentioned before, there is some variability in where the eyes land in a word, the readers usually fixate between the beginning and the center of the word, this location was termed the preferred viewing location by Rayner (1979).

The location of fixation in a word is also related to where the saccade before the fixation was launched. The majority of saccades land towards the center of a word, but this is related to the location of the previous fixation (G. W. McConkie, Kerr, Reddix, & Zola, 1988; Rayner, Sereno, & Raney, 1996).

In several studies, there are evidences that the orthographic regularity of the initial letters of a word, can influence in a significant way where the eyes land in a word (Beauvillain, Dore, & Baudouin, 1996; Hyona & Olson, 1995; Radach, Inhoff, & Heller, 2004) . These effects are very small but also reliable.

The decision about when to move the eyes is strongly related to the properties of the text that is being processed. The amount of time spent reading a word is closely related to lexical, syntactic, and discourse variables. The frequency with which a word occurs in language reliably affects the fixation durations on words (Albrecht Werner Inhoff & Rayner, 1986; Juhasz & Rayner, 2003; Rayner & Duffy, 1986).

Other studies have shown that the familiarity of a word affects fixation times on words even when frequency is statistically or experimentally controlled (Juhasz & Rayner, 2003; Williams & Morris, 2004). Other lexical variables that have been found to affect fixation durations are the age at which words are acquired and the concreteness of a word's referent (Juhasz & Rayner, 2003). Another factor which determines when the eyes move during reading, is the word length, however, these effects of word length are usually not seen (or are rather small) in first fixation duration and single fixation duration, but are quite robust in gaze duration, where time spent refixating the word is taken into account (Rayner et al., 2008).

PART 2. EXPERIMENT

5. Participants & Methods

5.1 Participants

A group of 14 persons participated in the study. Five of them were male and nine female. Their mean age was 30.6 years, ranging from 25 to 36 years. All of them were native Greek speakers. We chose our participants in a way so that we assured similar education level for all of them. The participants did not have any ocular or systemic pathology. The experiments were performed with the participants' habitual refractive correction (if needed) as long as visual acuity was better than 0.0 logMAR in each eye. None of them had previously seen the text used for this experiment. Distance VA was measured (at 4 m) using the European-wide ETDRS visual acuity charts, that were developed in the University of Crete. Near visual acuity was measured (at 40 cm) again using the European-wide ETDRS visual acuity charts. Eye dominance was evaluated using the hole in card method.

The characteristics of the study's participants are presented in the following table.

Table 3 Baseline Characteristics of Study Participants

Subject	Sex	Age	Dominant Eye	VA far (logMAR)			VA near (logMAR)			Glasses
				OD	OS	Bin.	OD	OS	Bin.	
1	M	26	OD	0,00	-0,04	-0,04	0,02	0,00	-0,04	Yes
2	F	29	OD	-0,26	-0,16	-0,28	-0,10	-0,08	-0,14	No
3	F	32	OS	-0,22	0,00	-0,26	-0,04	-0,06	-0,02	Yes
4	F	30	OD	-0,20	-0,16	-0,18	-0,16	-0,10	-0,16	No
5	F	33	OS	-0,04	0,00	-0,06	-0,06	0,00	-0,04	Yes
6	M	26	OD	-0,16	-0,16	-0,26	-0,16	-0,16	-0,20	No
7	M	25	OD	-0,24	-0,20	-0,24	-0,18	-0,08	-0,20	No
8	M	32	OD	-0,22	-0,14	-0,28	-0,24	-0,20	-0,20	Yes
9	M	36	OD	-0,12	-0,12	-0,16	-0,06	-0,02	-0,12	No
10	F	34	OS	-0,20	-0,30	-0,30	-0,16	-0,16	-0,14	No
11	F	36	OD	-0,12	-0,12	-0,16	-0,06	-0,02	-0,12	No
12	F	32	OS	-0,16	-0,08	-0,22	-0,08	-0,06	-0,10	Yes
13	F	28	OS	-0,20	-0,06	-0,24	-0,20	-0,08	-0,16	No
14	F	30	OD	-0,28	-0,28	-0,28	-0,06	-0,12	-0,20	No

5.2 Eye Movements Recording

Eye movements were monitored using an Eye-Link II eye tracker (SR Research Ltd.)(Fig. 22). The EyeLink II has a high resolution (noise-limited at $<0.01^\circ$) and maximum data rate of 500 samples per second (Hz). The EyeLink II system consists of three miniature cameras mounted on a padded headband. One head-tracking camera was used to detect infrared markers in the world, while two eye cameras focus on the left and right eyes respectively.



Figure 22 The EyeLink II eye tracker

The EyeLink II system consists of a Host PC, a display PC, the headband and a PCI card. The Host PC connects to the headband and powers four infrared markers (for head tracking) that are mounted on the corners of Display PC's monitor. It also hosts the EyeLink II Host application where you can control the tracker and change the options of the recording. The Display PC runs experiment software for control of the Host PC and presents the stimuli to the monitor. The headband has the cameras which record the eye movements. Finally, the PCI card is connected with the headband and is hosted in the Host PC. It performs the powerful image processing required to achieve the high temporal and spatial resolution of the system. The EyeLink II system is presented in the following figure (Fig. 23).

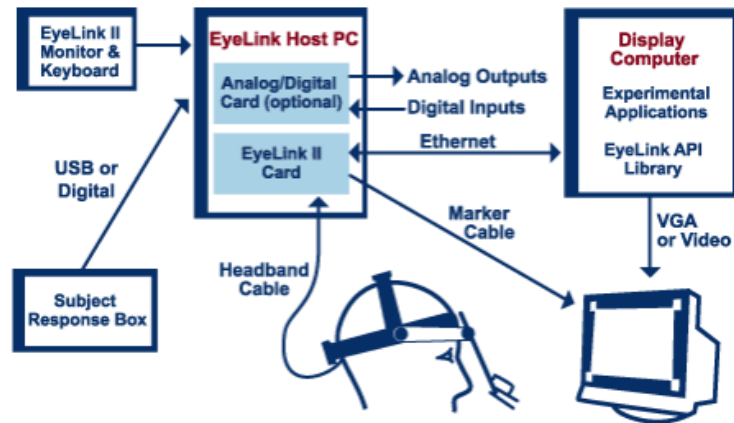


Figure 23 The EyeLink II System

Data used in this study were recorded with the pupil tracking mode at a rate of 500Hz. Viewing and recording were binocular. Recordings from the right eye were taken analysis of fixation stability, and binocular recordings were used for microsaccades detection. The calibration and validation points were presented in a high resolution monitor (SONY, GDM 520), which was supported by a high resolution graphic card (VSG 2/5, Cambridge Research System).

5.2.1 The pupil tracking mode

The iris of the eye is distinguishable from the pupil. The pupil can be separated by the surrounding iris optically. This can be especially sharpened with the use of infrared light which will be nearly entirely absorbed once entering the eye, consequently making the pupil much darker than the surrounding iris (Young & Sheena, 1975).

The recording apparatus must be held completely still in relation to the head. This tracking method has several advantages, comparing with other tracking methods like limbus tracking. One major advantage is that the pupil is less covered by the eyelids than the limbus which allows recordings with higher resolution. The disadvantage is that the difference in contrast is lower between the pupil and iris than between the iris and sclera-thus making the border detection more difficult(Young & Sheena, 1975).

5.3 Reading Text

A paragraph consisted of 141 words and 911 characters was used in this experiment. The print size was 0.4 logMAR at a distance of 40 cm. The text contained simple sentences about the island of Crete.

Η Κρήτη, το πέμπτο μεγαλύτερο νησί της Μεσογείου, βρίσκεται στο νοτιότερο άκρο της Ευρώπης. Ο περήφανος και φιλόξενος κρητικός λαός φημίζεται για τον πολιτισμό, το καινοτόμο πνεύμα του, τις διατροφικές του συνήθειες. Η Κρήτη συνδυάζει το βουνό με τη θάλασσα, το παλιό με το καινούργιο, την αρχαία ιστορία με τη σύγχρονη. Αποτελεί σταυροδρόμι πολιτισμών λόγω της γεωγραφικής της θέσης. Είναι από τους πιο δημοφιλείς ελληνικούς τουριστικούς προορισμούς. Στην Κρήτη το αφιλόξενο και άγονο ορεινό τοπίο συνδυάζεται αρμονικά με τους εύφορους πράσινους κάμπους, το ξανθό της αμμουδιάς, το βαθύ μπλε της θάλασσας και όλα μαζί με το γαλάζιο του ηλιόλουστου ουρανού. Το πράσινο των πεδιάδων, το γκρι των απόκρημνων βράχων και το μπλε βαθύ του πελάγους δημιουργούν έναν απίστευτο συνδυασμό που ταξιδεύει τον περιηγητή. Τα φυτά της περιοχής προσθέτουν τα δικά τους χρώματα και αρώματα μέσα στα οποία ζουν σπάνια είδη ζώων.

Figure 24 the text used in this experiment, which contains simple sentences about the island of Crete

5.4 Experimental Procedure

The observer was seated in front of the display screen at a distance of 40 cm with the head stabilized using a chin rest to limit any head movements. The illuminance of the observer's eye was measured with a luxmeter and the luminance of the reading text was measured with a photometer. Prior to each experiment, a calibration/validation session was conducted. The calibration was a five-point cross figure. After the validation, the display screen was switched off and the session commenced. The observer was instructed to read the text silently. The observer was asked to close his eyes until he was asked to open them. He was also asked to first look in a black spot at the bottom of the reading card and then start reading. He was informed that we needed him to comprehend the sentences and for this reason, he would have to answer five questions afterwards.

The experiment was conducted to evaluate the characteristics of eye movements in different luminance conditions. Participants read the text two times, the first while the lights of the room were switched off and the cornea illuminance was set to mesopic levels, of about 1.0 lux. The chart background's luminance was measured with a photometer at right below 1 cd/m². The second measurement was conducted in photopic conditions and the cornea illuminance was measured with a luxmeter at about 60 lux. Using the photometer we have measured the chart background luminance at 55 cd/m².

Calibration and validation was conducted after each session. After each recording, comprehension was evaluated using five questions on the sentences and the text.

5.5 Data Analysis

For further analysis, of the recorded data, we developed an algorithm using MATLAB and the statistical analysis of the results was implemented using Microsoft Excel, IBM SPSS 22 and GraphPad Prism 6. The significance value was set to $\alpha=0.05$ for all the statistic tests performed in this study.

The data files used as input to the MATLAB script consisted of 8 columns of data for each participant of the experiment, which represented the timestamp for each recording moment (recording in every 2 ms, as sampling frequency was 500Hz), the corresponding vertical and horizontally positions x and y for both eyes and the number of trial (we recorded two trials for the two different luminance conditions) and the number of fixation, which was separated by EyeLink II parser software.

For every individual participated in this study we have excluded manually the recorded data before and after reading the text. The algorithm we have developed, also excludes the recordings which correspond to blinks.

One main function of this algorithm, is to identify the fixations which occur after forward and backward (regressions) fixations, so we can analyze separately the two categories of fixations. The sorting between fixations is performed by identifying the first and last fixations in each line of the text, which allows us to detect the fixations which occur while the eyes perform a return sweep saccade (change of line). From the remaining fixations we were able to compare two consecutive fixations by the horizontal position x and separate forward and backward fixations. In this study fixations which occurred after forward fixations and regressions will be called simply as forward and regression fixations correspondingly. The participants' eyes were blinking in some fixations, which have been excluded from the analysis.

Using the algorithm, we can also calculate the reading speed for each trial, by dividing the number of words which consist the text, by the reading time in minutes.

$$reading\ speed = \frac{number\ of\ words}{reading\ time}$$

In order to describe the variability of the eye position in a fixation, we used a measure to quantify the stability for each fixation. We have used the Bivariate Contour Ellipse Area (BCEA). R. M. Steinman was the first to introduce this measure, by calculating the confidence ellipses of bivariate normal distributions for recordings in experiments with steady target (Steinman, 1965). This measure requires normality for both measured positions (vertical and horizontal), to assume bivariate normal distribution and the dispersion of those positions can be describe by an ellipse whose area is analogous to the standard deviation of a univariate distribution.

The area of such an ellipse which contains the central $(1 - \alpha) \times 100\%$ of a bivariate normal population is:

$area = \pi\chi^2(a)\sqrt{\lambda_1}\sqrt{\lambda_2}$, where χ^2 denotes the chi-square statistical value which corresponds to the selected confidence level and λ_1, λ_2 are the eigenvalues of the covariance matrix Σ .

This means that the area is also equal to $area = \pi\chi^2(a)|\Sigma|^{1/2}$.

We have also to note that:

$$\det(\Sigma) = \det \begin{bmatrix} \sigma_{11} & \sigma_{12} \\ \sigma_{21} & \sigma_{22} \end{bmatrix} = \det \begin{bmatrix} \sigma_1 & \rho\sigma_1\sigma_2 \\ \rho\sigma_1\sigma_2 & \sigma_2 \end{bmatrix} = \sigma_{11}\sigma_{22}(1 - \rho^2)$$

Also we replace the chi-square value with $2k$ where k is half the value of chi-square statistic, and we have the final formula for the contour ellipse area:

$$area = 2k\pi\sigma_1\sigma_2\sqrt{1 - \rho^2}$$

ρ is the product-moment correlation of these two position components, and k establishes the confidence limit for the ellipse, the probability of a given observation falling within it being given by:

$$P = 1 - e^{-k}$$

Where e is the base of the natural logarithm.

In our experiment we have used a value of $k=1.14$ which corresponds to a probability of 68%. The bivariate contour ellipse area as calculated in the present experiment encloses that portion of the retinal surface where the target image would be found 68% of the fixation time.

This measure is expressed in (arcmin)², and the appropriate conversions are included in the algorithm to convert the input data positions from pixels to minutes of arc, considering the distance of reading.

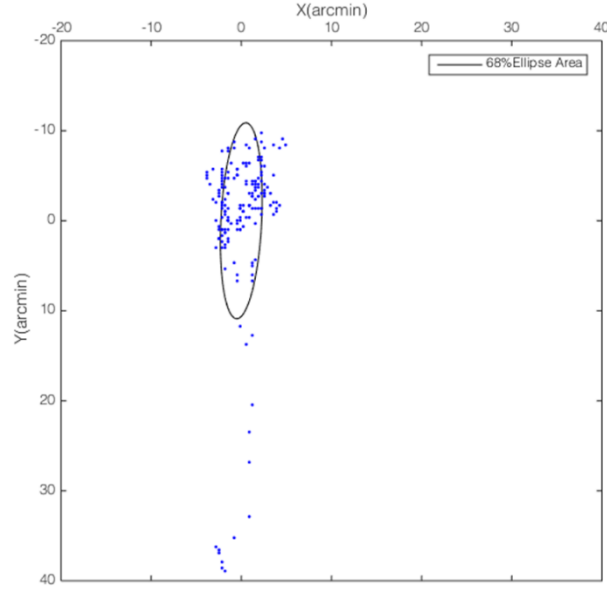


Figure 25 Scatter Plot of recorded eye positions and the 68% Bivariate Contour Ellipse

Another important function of the implemented algorithm is the use of appropriate functions to detect the microsaccades in fixations and calculate their characteristics. We have used a velocity threshold based algorithm, which was introduced by R. Engbert & R. Kliegl (Ralf Engbert & Kliegl, 2003; R. Engbert & Mergenthaler, 2006) and we have adapted functions from EYE-EEG extension (Dimigen, Sommer, Hohlfeld, Jacobs, & Kliegl, 2011).

Before applying the microsaccades detection algorithm, raw data were smoothed by using a running average on velocity samples to suppress high frequency noise by using a cumulative sum:

$$\vec{x}_n = \vec{x}_o + \Delta t \sum_{i=1}^n \vec{v}_i$$

The velocities were computed from raw data, for both eyes according to a moving average procedure:

$$\vec{v}_n = \frac{\vec{x}_{n+2} + \vec{x}_{n+1} - \vec{x}_{n-1} - \vec{x}_{n-2}}{6\Delta t}$$

Where Δt is 500 ms in our case as the sampling frequency was 500Hz.

The detection of microsaccades by using E&K algorithm is based on a velocity threshold which is applied in the 2D velocity space to determine the movements which are faster from the threshold we set. Thresholds are computed separately for horizontal η_x and vertical η_y components, in units of median based SDs:

$$\eta_{x,y} = \lambda \cdot \sigma_{x,y}$$

Where $\sigma_{x,y} = \langle v_{x,y}^2 \rangle - \langle v_{x,y} \rangle^2$ and $\langle \cdot \rangle$ is the median estimator.

In our case we have selected $\lambda=4$ and also we have requested a minimum 3 samples duration to consider it a candidate for microsaccade.

At this point we have to mention that microsaccades are traditionally defined as binocular movements (Ciuffreda & Tannen, 1995), which lead us to detect only the microsaccades which occur in both eyes with a temporal overlap, which means that if a microsaccade is detected in the one eye and another is detected in the second eye within the duration of the first microsaccade, we consider that we have detected a binocular microsaccade.

In the following figures (Fig. 26 and Fig. 27) we can see how the velocity threshold is applied to detect the microsaccades in 2D velocity space, and the graphical representation of smoothed eye movements in a single fixation, with the detected microsaccades.

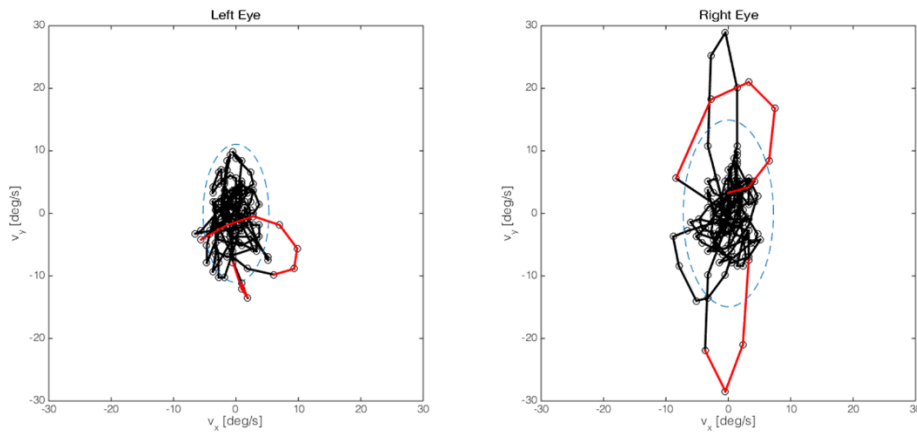


Figure 26 Velocities in 2D velocity space for both eyes, with velocity elliptic threshold and the velocities which correspond to microsaccade movements (with red).

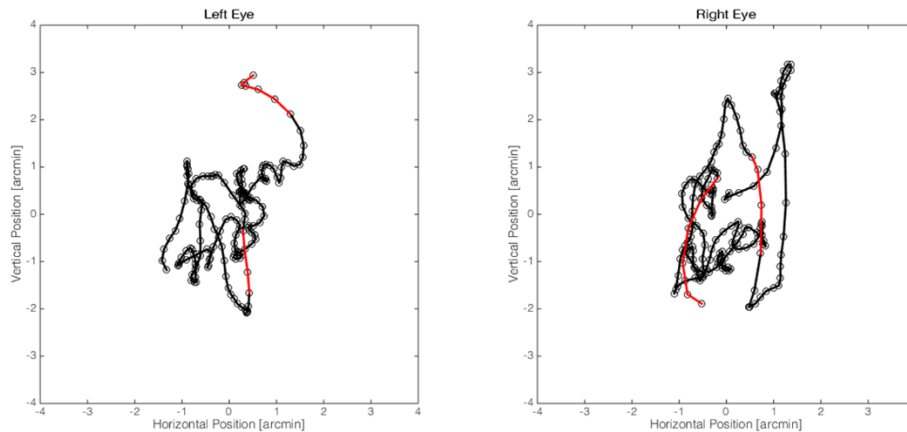


Figure 27 Eye movements of both eyes in a single fixations and the microsaccades (with red) detected by E&K algorithm

By using the appropriate functions the algorithm can also calculate several characteristics of the detected microsaccades, such as the angle, the peak velocity, the amplitude and the duration of each movement.

5.6 Ex-Gaussian Analysis

The ex-Gaussian distribution is often used to model response time (RT), since 1979 when Ratcliff introduced this analysis (Ratcliff, 1979). It is defined by adding two random variables, one from a normal distribution and the other from an exponential. In 2010 Staub et al., established that fixation duration in reading distributions can be well fitted by the ex-Gaussian distribution (Staub, 2010).

The ex-Gaussian function is a density function, which is actually derived via a convolution of the normal (Gaussian) and exponential density functions. The basic parameters of the ex-Gaussian function are μ , σ and τ . The first two describe the mean and the standard deviation of the Gaussian component of the density function, while τ describes the rate of the exponential component.

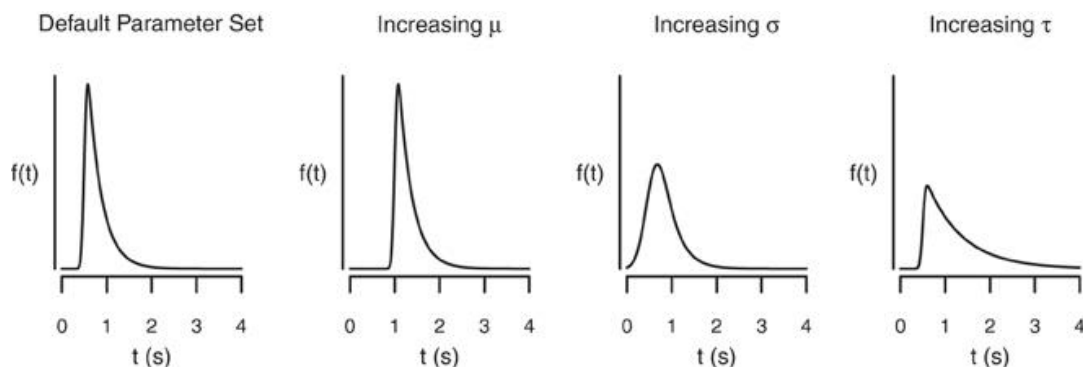


Figure 28 The shape of the ex-Gaussian density function according to the coefficients μ, σ and τ (Matzke, 2013)

The ex-Gaussian density function has some very interesting properties, as the mean of the ex-Gaussian distribution is the sum of μ and σ parameters, while the variance of the distribution is equal to the sum of σ^2 and τ^2 .

Ratcliff has suggested that there be at least 100 data points in the sample before the parameter estimates should be regarded as reliable (Ratcliff, 1979).

Ex-Gaussian density function can also be used as a good fit for other positive skewed distributions.

PART 3. RESULTS

6. Results

6.1 Number of Fixations

First of all we wanted to test, if there is a statistical significant difference to the total number of valid fixations (fixations after forward saccades, regressions and return sweep saccades).

All the variables, compared in this section, are normally distributed, as Shapiro Wilk test showed, so we can perform t-test to compare them. The following table presents the results.

Table 4 Shapiro-Wilk test results, which shows that there is not a statistical significant difference between the distributions and the normal distribution

	df	p
Total Number of Fixations (Low Luminance)	14	0,560
Total Number of Fixations (High Luminance)	14	0,904
Number of Forward Fixations (Low Luminance)	14	0,781
Number of Forward Fixations (High Luminance)	14	0,908
Number of Regressions (Low Luminance)	14	0,981
Number of Regressions (High Luminance)	14	0,296
Number of Return Sweep Fixations (Low Luminance)	14	0,685
Number of Return Sweep Fixations (High Luminance)	14	0,158

The mean of the total fixations number in low luminance was 149,71 (SD=38,18) and in high luminance 156,5 (SD=34,78). A paired t-test was performed, which showed that there is not statistical difference to the total number of fixations between the two luminance conditions (p=0,565).

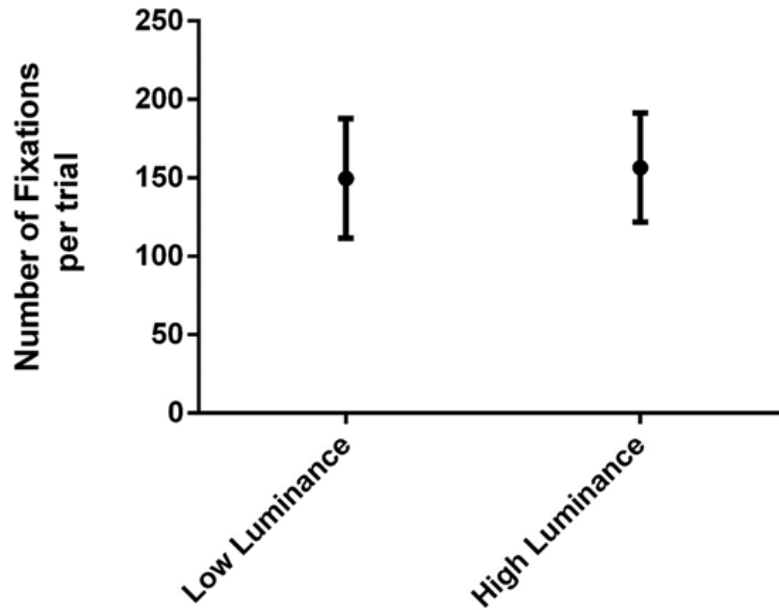


Figure 29 Total Number of Fixations per trial, in two luminance conditions, with mean±SD

The mean number of the forward fixations in low luminance was 109,21 (SD=27,45) and in high luminance 108,42(SD=24,54). Again the paired t-test showed that there is not any statistical significant difference between the two conditions (p=0,923).

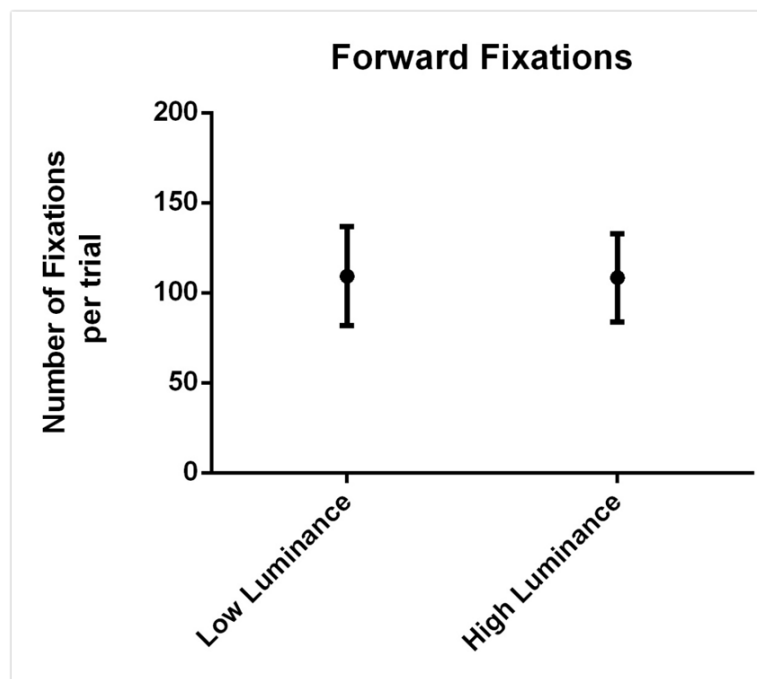


Figure 30 Number of Forward Fixations per trial in two different luminance conditions, with mean±SD

We have performed the same comparison for the number of fixations after backward saccades (regressions). The mean number of fixations in low luminance was 26,78(SD=11,92) and in high luminance 33,71 (SD=13,74). Again there was not statistical significant difference (p=0,07), and as the p value is close to the

significance level 0.05 we report also the 95% significance level which is (-0,921 , 14,78). This means that possibly with different number of subjects a significant difference could exist.

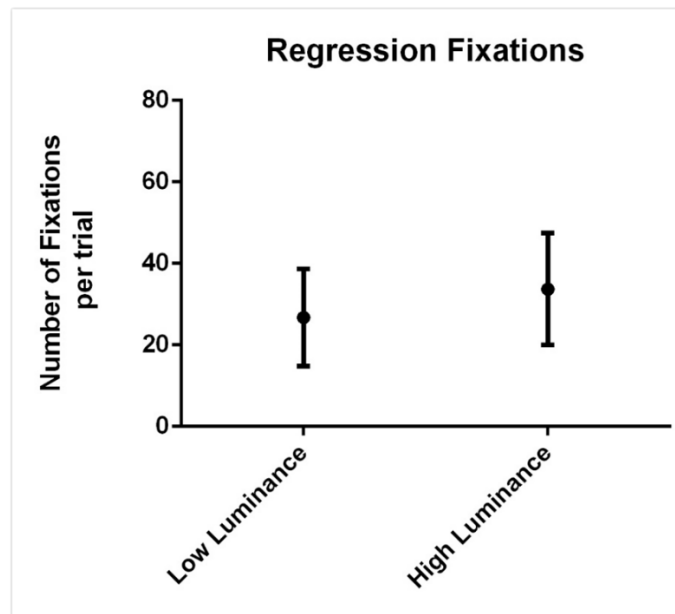


Figure 31 Number of Regression Fixations per trial in two different luminance conditions, with mean±SD

Finally we have compared the number of return sweep fixations. In low luminance the mean number of return sweep fixations was 13,714(SD=5,09) and in high luminance 14,357 (SD=5,41). The paired t-test showed that there is not statistical significant difference between the number of return sweep fixations in the different luminance conditions (p=0,745).

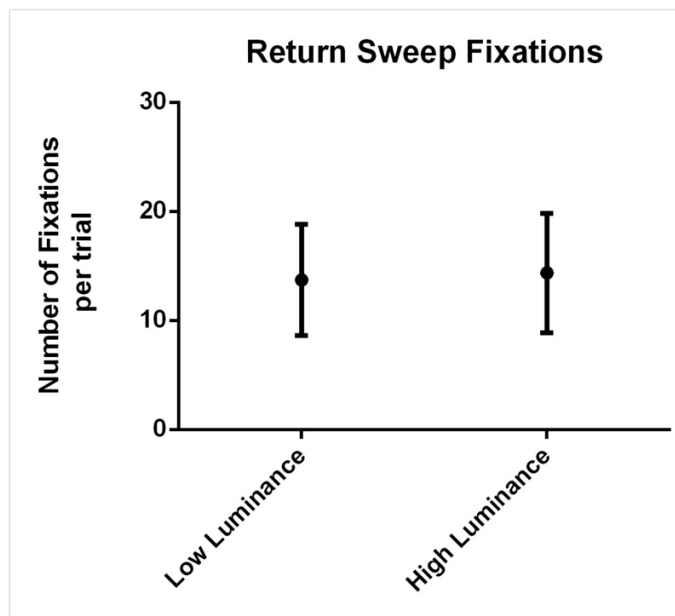


Figure 32 Number of Return Sweep Fixations per trial in the two different luminance conditions, with mean±SD

6.1.1 Percentage Distribution of Fixations

As we have seen in the previous chapter, the number of fixations is not affected by the change of luminance. But it is really important to examine if there is any difference in the percentage distribution.

As we can see in the following pie charts, in high luminance 69,42% were Forward Fixations, 21,15% were Regression Fixations and only the 9,42% of the total fixations were Return Sweep Fixations.

In low luminance 73,13% were Forward Fixations, 17,37% were Regression Fixations and 9,51% were Return Sweep Fixations.

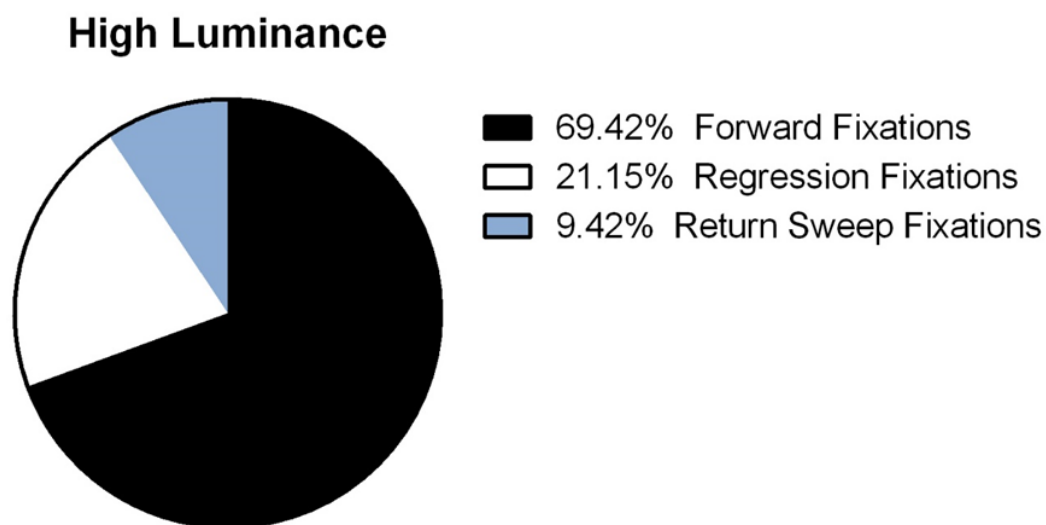


Figure 33 Average percentage of detected fixations, in high luminance conditions.

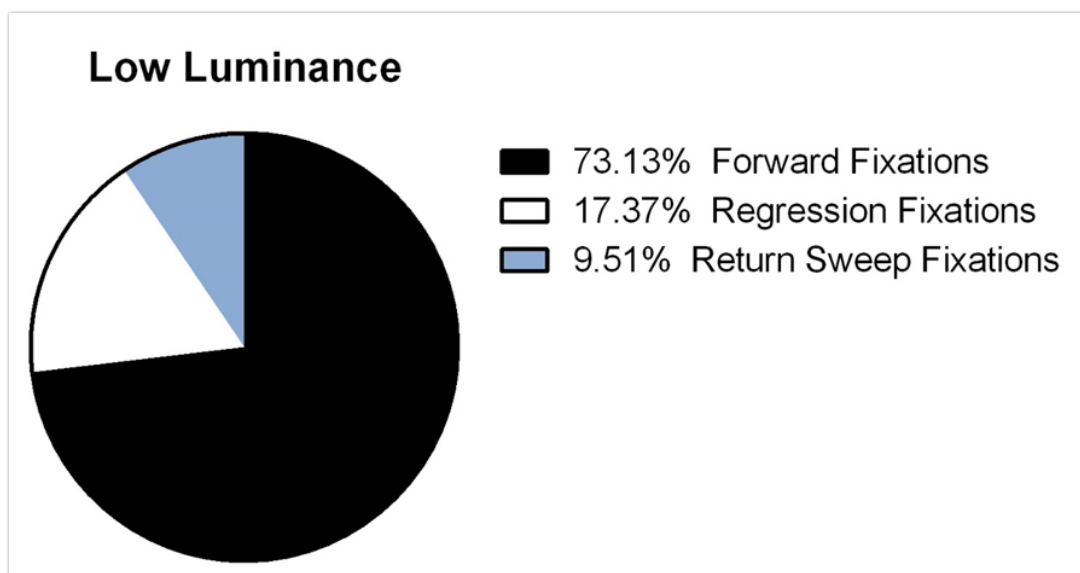


Figure 34 Average percentage of detected fixations, in low luminance conditions.

Multiple paired t-test have been performed to examine if there is any statistical significant difference between the percentages of each fixation category in low and high luminance conditions.

There is a statistical significant difference between the two conditions according to the percentage of forward fixations ($p=0.005$), and this means that the percent of forward fixations is higher in low luminance conditions (95% CI of the difference (1,28%-6,12%)).

Also there is statistical significant difference between the percentages of backward fixations in the two luminance conditions ($p=0,010$), and the percentage of backward fixations is higher in high luminance conditions (95% CI of the difference (-0,065,-0,010)).

Finally the comparison between the percentages of the return sweep fixations indicated that there is not a statistical significant difference between the two luminance conditions ($p=0.943$).

6.2 Fixation Duration & Fixation Stability

6.2.1 Fixation Duration

We had evidence from previous studies, that fixation duration is not distributed normally. In this study, as we have sorted the valid fixations in three different categories (forward fixations, regression fixations and return sweep fixations), we performed the same analysis for the forward and regression fixations, the two categories of fixations which are playing the most important role in obtaining information from the reading text.

We have performed Shapiro-Wilk test, to examine if fixation duration was normally distributed, which showed that for both fixation categories and for both luminance conditions the distributions were not normal ($p < 0.001$ for all distributions).

As the fixation duration is not normally distributed, we are using the median of each data set as a central tendency measure for the fixation duration.

In high luminance the median fixation duration was 220 ms for both forward and regression fixations. In low luminance the median fixation duration was 308 ms for forward fixations and 348 for regressions.

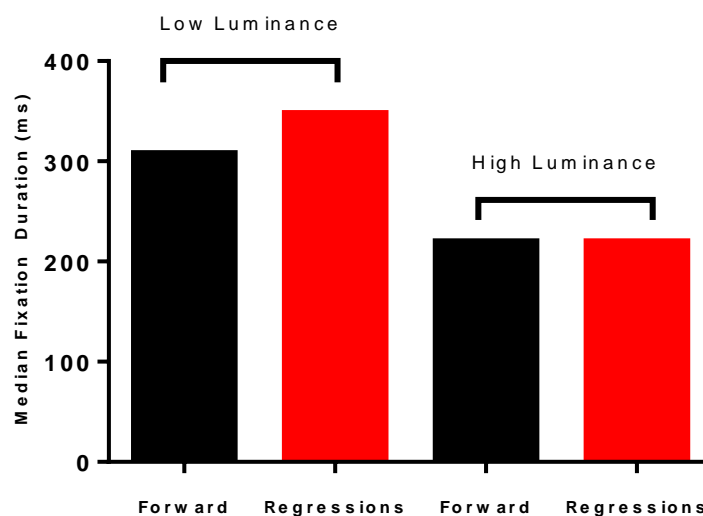


Figure 35 Median Fixation Duration in high and low luminance conditions. Highlighted with black the forward fixations' median duration and with red the backward fixations' median duration.

As we can see the median fixation duration is higher in lower luminance conditions, but there is not a big difference between the forward and backward fixations median duration.

6.2.2 Ex-Gaussian Analysis for Fixation Duration

We have mentioned in section 5.6, fixation duration distributions are well fitted by ex-Gaussian density function.

The fit an ex-Gaussian density function, we have used all the fixation durations we have detected in this study, and separated fixation durations in bins of 50 ms. The ex-Gaussian analysis has been performed four times, for the two different luminance conditions, and separately for the two categories of fixations (forward and regressions).

The following figures depict the ex-Gaussian density function in each condition for both forward and regression fixations.

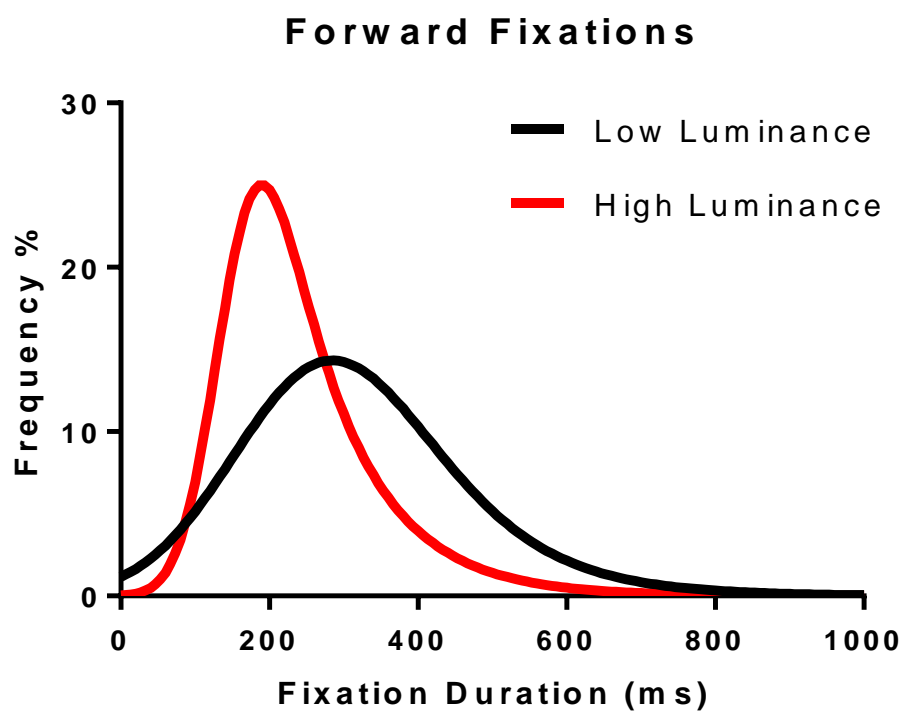


Figure 36 Ex-Gaussian density functions of Forward Fixation Duration in high (red) and low (black) luminance conditions

Regressions

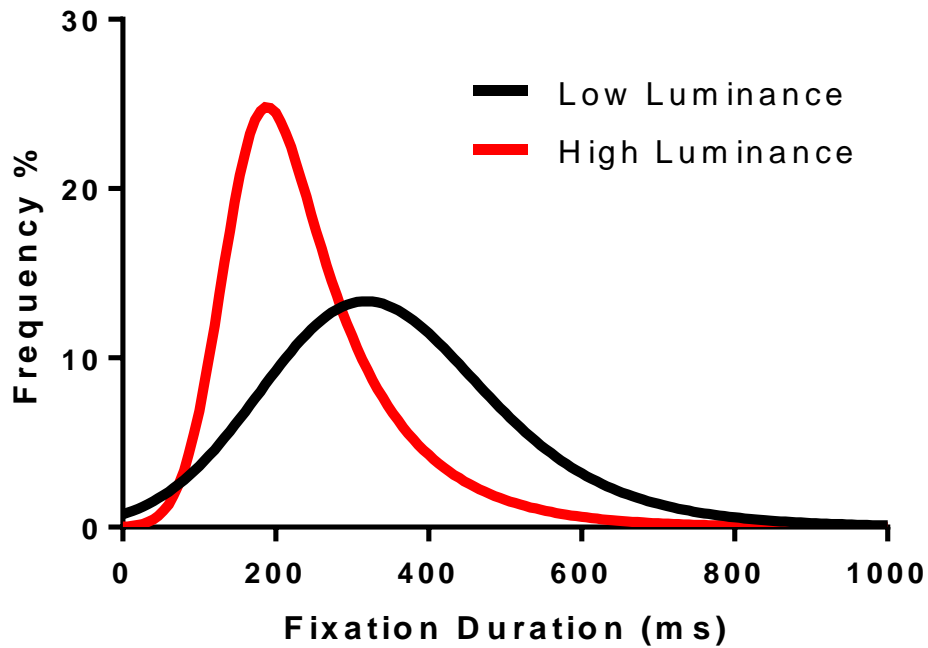


Figure 37 Ex-Gaussian density functions of regression fixation duration, in high (red) and low (black) luminance conditions

As we can observe from the figures, the density functions are likely similar for the two categories of fixations, but there is an obvious difference between low and high luminance as the density function is shifted to the left in high luminance. To understand better the difference we will report the values of the ex-Gaussian function's coefficients.

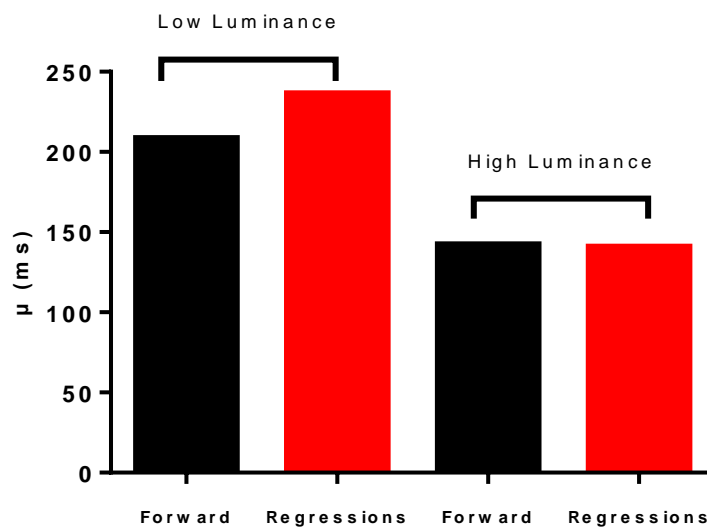


Figure 38 μ parameter values in different luminance conditions

The value of μ parameter is higher in low luminance, which explains why the density function is shifted to the right. Another important information is that μ value is relatively the same between forward and regression fixations in both conditions of luminance.

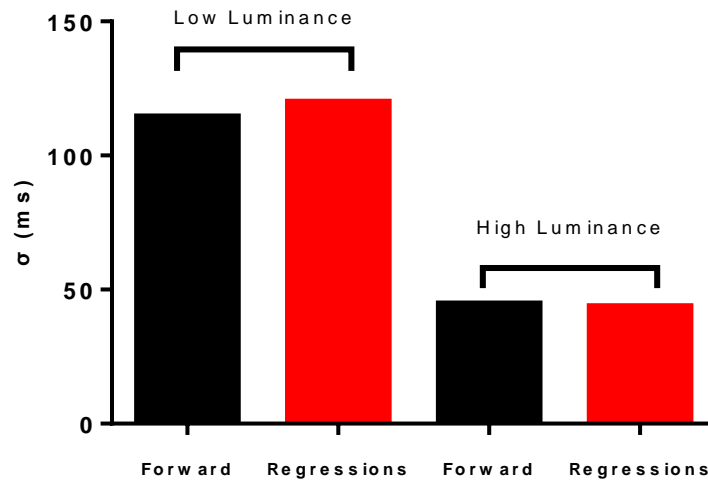


Figure 39 Values of σ parameter for the different conditions of luminance

We know that as the parameter σ increases, the density function becomes wider. In our case parameter σ is affected by low luminance, as its value is greater than in high luminance. Similar to μ parameter the value of σ parameter is relatively similar between forward and regression fixations.

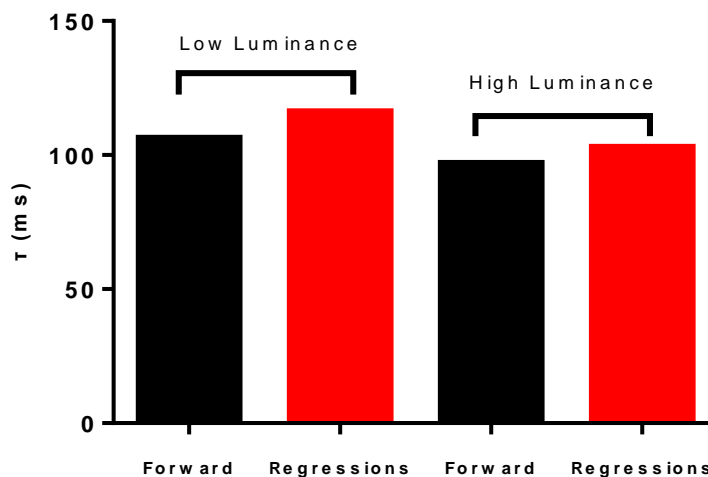


Figure 40 Values of τ parameter for the different conditions of luminance

Parameter τ remains relatively stable in both luminance conditions, but we should mention that the parameter's value is slightly greater for regression fixations.

6.2.3 Fixation Stability

In previous chapters we have discussed about the fixation stability and the measures used to be evaluated. In our case we have calculated the 68% Bivariate Contour Ellipse Area for each valid fixation (forward & regression).

Likewise fixation duration, we have sorted the calculated data in four different categories based on luminance conditions and fixation category (forward or regression).

We have performed Shapiro-Wilk test to examine if 68% BCEA is distributed normally. The test showed that the distributions are statistically significantly different from a normal distribution in each case ($p < 0.001$).

The median 68% BCEA is illustrated in the following figure. The median 68 % BCEA in high luminance was 50,16 arcmin² for forward fixations and 46,90 arcmin² for backward fixations and in low luminance 72,18 arcmin² for forward fixations and 76,45 arcmin² for backward fixations.

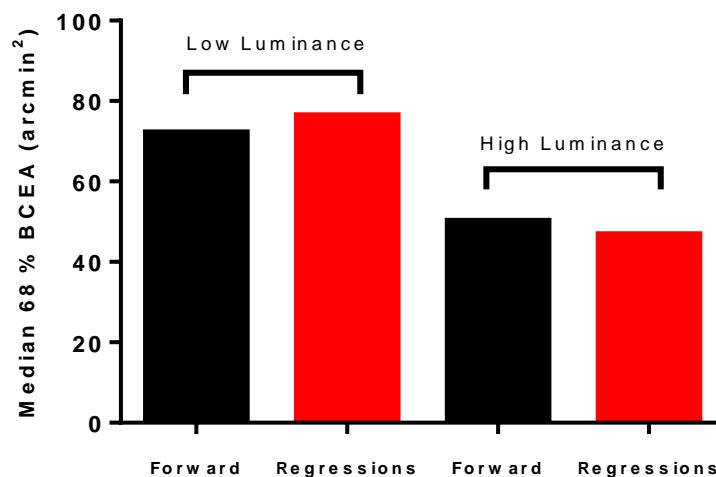


Figure 41 Median 68% BCEA in high and low luminance

As we can see the median 68% Bivariate Contour Ellipse Area is greater in low luminance for both forward and regressions.

6.2.4 Ex-Gaussian Analysis for 68% BCEA distributions

The distributions of 68% BCEA have positive skewness therefore we tried to apply ex-Gaussian analysis to examine if it is a good fit.

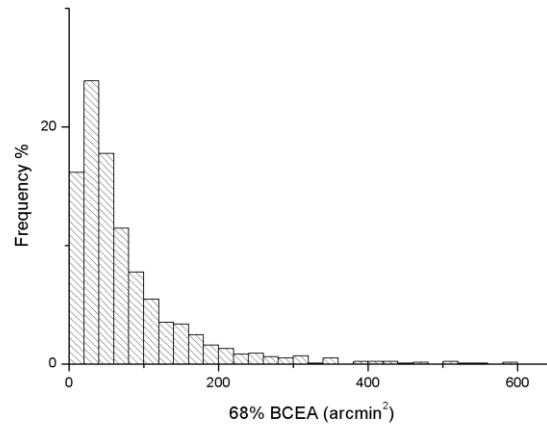


Figure 42 An example distribution of 68% Bivariate Contour Ellipse Area

The fit an ex-Gaussian density function, we have used the 68 % BCEAs from all the fixations we have detected in this study, and separated them in bins of 20 arcmin². The ex-Gaussian analysis has been performed four times, for the two different luminance conditions, and separately for the two categories of fixations (forward and regressions).

The following figures depict the ex-Gaussian density function in each condition for both forward and regression fixations.

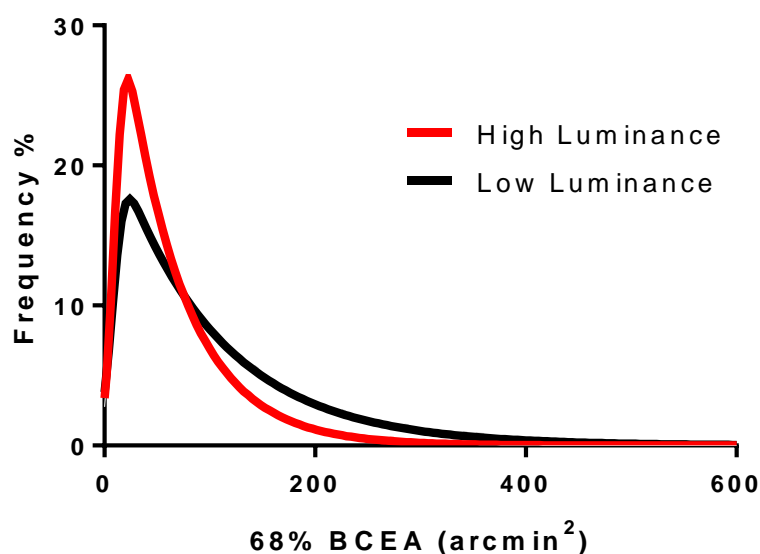


Figure 43 Ex-Gaussian density functions of 68% BCEA in high (red) and low (black) luminance conditions (forward fixations)

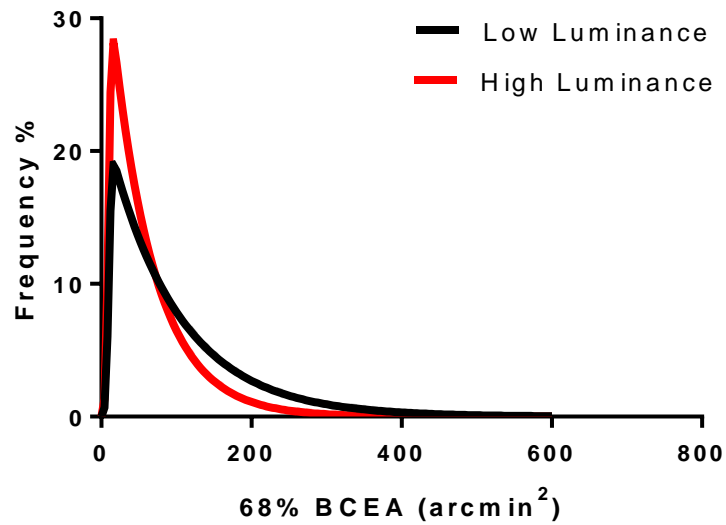


Figure 44 Ex-Gaussian density functions of 68% BCEA, in high (red) and low (black) luminance conditions (backward fixations)

As we can see density functions are less wide comparing to fixation duration ex-Gaussian functions. Also the two graphs appear relatively the same, as there are not big differences between forward and regression fixations. The main difference is observed between the two luminance conditions. To understand better the differences we will highlight the differences between the ex-Gaussian functions' coefficients μ , σ and τ .

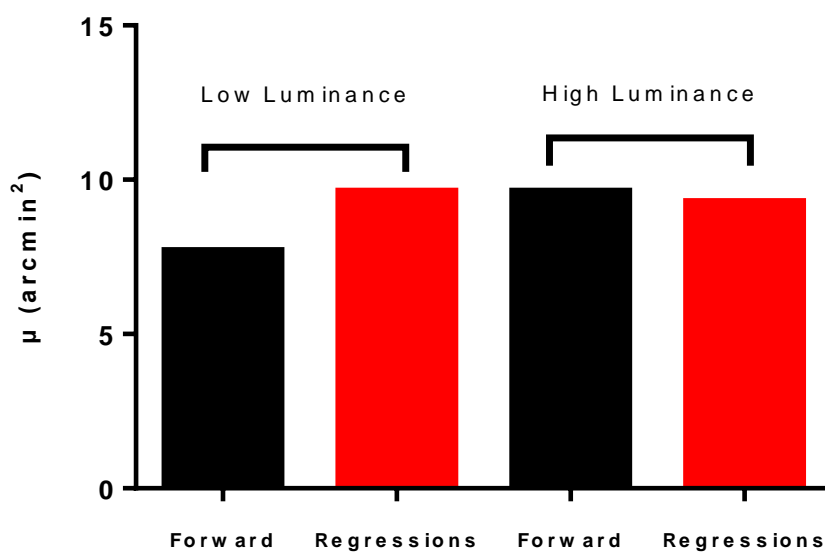


Figure 45 μ parameter in different luminance conditions

Value of μ parameter remains relatively stable, in high and low luminance. We can spot a difference for the forward fixations in low luminance which is responsible for the slight shift to the left of the ex-Gaussian function.

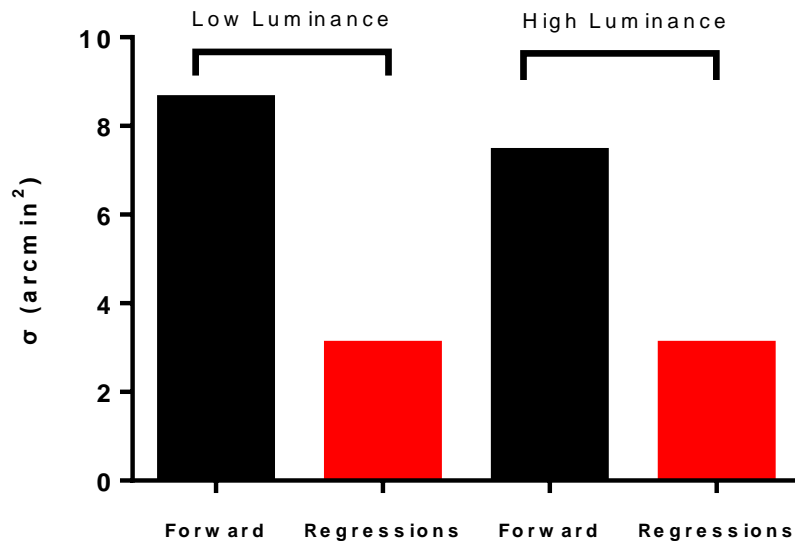


Figure 46 σ parameter in different luminance conditions

The value of parameter σ remains relatively stable for both forward and regression fixations in low and high luminance, but as we can see σ is lower in regressions than forward fixations. This means that the Gaussian component is wider in forward fixations, as the variance is greater than in regression fixations.

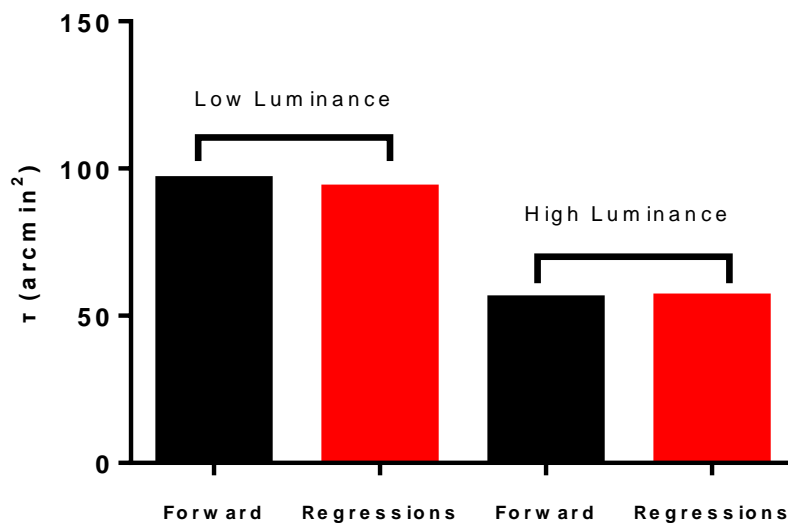


Figure 47 parameter τ in different luminance conditions

Parameter τ is higher in low luminance conditions for both forward and backward fixations. As $1/\tau$ expresses the rate of the exponential component of the ex-Gaussian function, we can conclude that in lower luminance the function decreases slower, therefore we have more fixation with bigger fixation areas in low luminance.

6.2.5 Relationship between Fixation Duration & Fixation Stability

An important question we are trying to answer, is whether fixation duration is associated with fixation stability.

We performed correlation analysis for both categories of fixations in high and low luminance conditions.

For forward fixations, the Pearson correlation coefficient was $r=0.1835$ ($p<0,0001$) in high luminance conditions and $r=0.0945$ ($p=0,0005$) in low luminance.

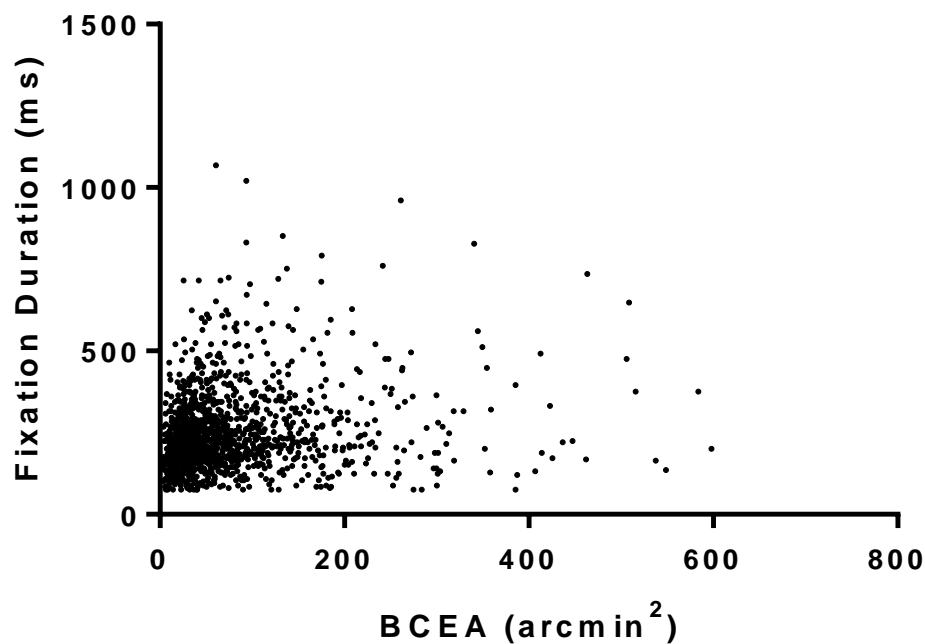


Figure 48 Scatterplot of Fixation Duration and BCEA of forward fixations in high luminance conditions

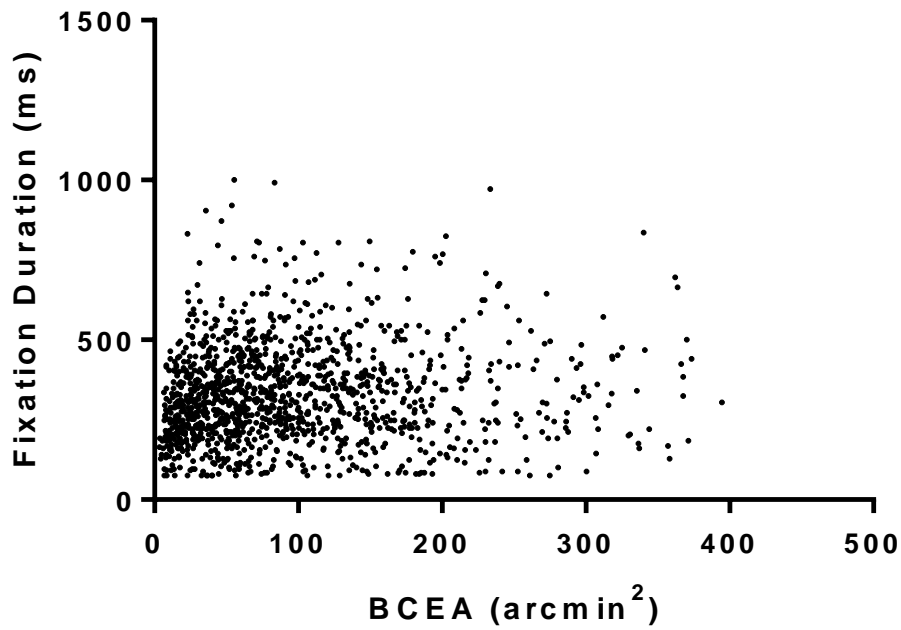


Figure 49 Scatterplot of Fixation Duration and BCEA of forward fixations in low luminance conditions

For backward fixations, the Pearson correlation coefficient was $r=0,161$ ($p=0,0006$) in high luminance and $r=0,176$ ($p=0,0012$) in low luminance.

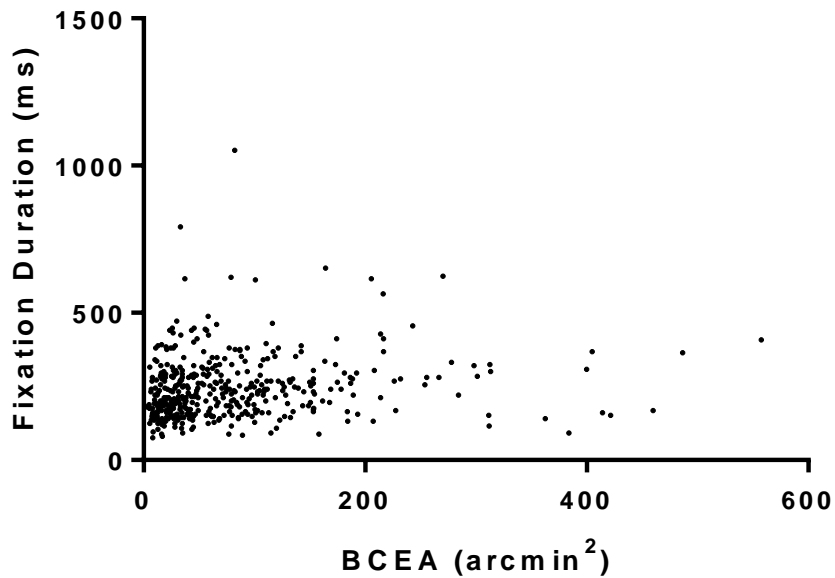


Figure 50 Scatterplot of fixation duration and BCEA of backward fixations in high luminance conditions

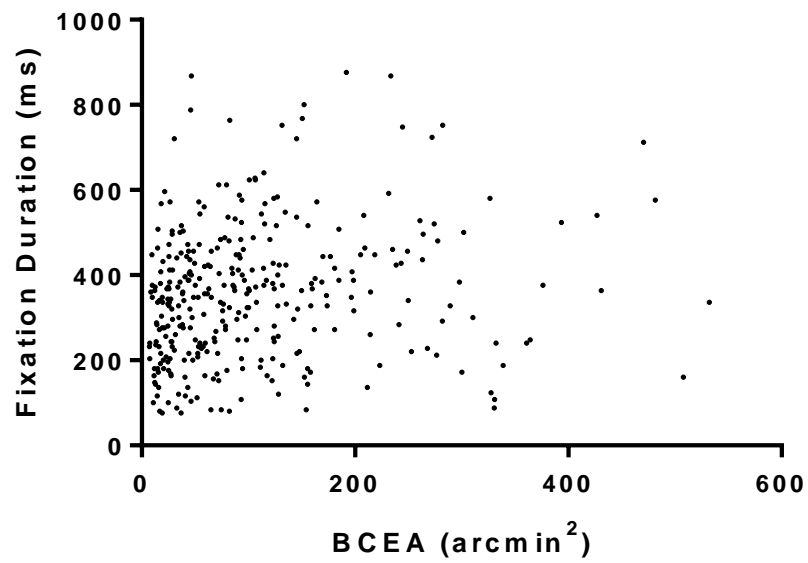


Figure 51 Scatterplot of fixation duration and BCEA of backward fixations in low luminance conditions

Correlation analysis showed that we have a weak correlation between fixation duration and 68% Bivariate Contour Ellipse Area, which means that there is not a linear association between the duration of fixation and the stability of each fixation.

6.3 Microsaccades

Using the algorithm described in section 5.5 of this study, we were able to detect the microsaccades which occurred in each fixation the eye tracker's software has detected. The basic questions we want to present in this section is how often microsaccades occur during reading fixations, and their characteristics in both luminance conditions, separately for forward and backward fixations.

6.3.1. Number of Microsaccades

In total 1729 microsaccades have been detected in high luminance conditions, from all participants in this study, 1317 of them in 1460 forward fixations and 412 in 456 backward fixations. In low luminance 1730 microsaccades have been detected, 1379 of them in 1353 forward fixations and 351 in 335 backward fixations.

The average number of microsaccades per forward fixation was 0,891 (SD=0,707) microsaccades/fixation in high luminance conditions and 0,948 (SD=0,771) microsaccades/fixation in low luminance conditions. The average number of microsaccades per backward fixation was 0,903 (SD=0,707) microsaccades/fixation in high luminance conditions and 1,023 (SD=0,749) microsaccades/fixation in low luminance conditions.

As we can see in the following comparison bar chart, the average number of microsaccades per fixation is slightly higher in backward fixations. Also the frequency is higher in low luminance conditions.

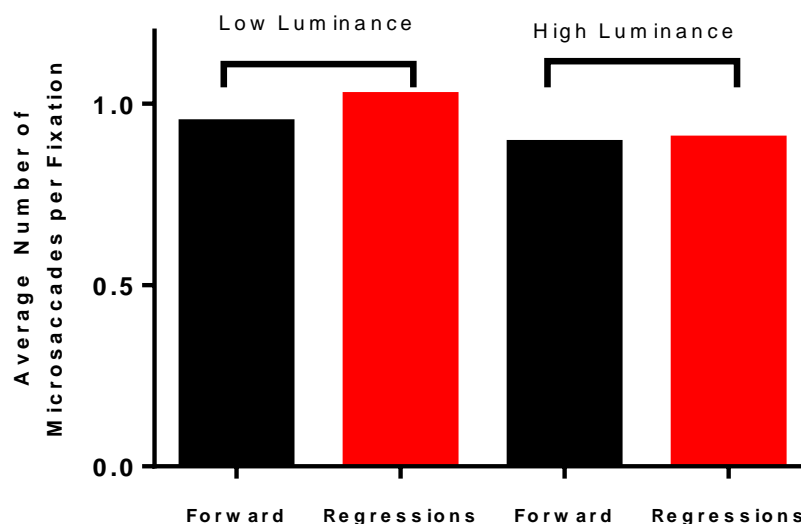


Figure 52 Average Number of Microsaccades per Fixation

In the following charts we present how the number of microsaccades per fixation is affected by the increase of fixation duration. We had to group the fixations in groups

based on fixation duration. The fixations are grouped in 75 ms groups (75-125, 125-175 etc.). We should mention that as we have discussed in previous sections of this study, fixation duration is not distributed normally, therefore for higher fixation durations the samples were low which means that for fixations with a duration higher than 700 ms the results might not be accurate.

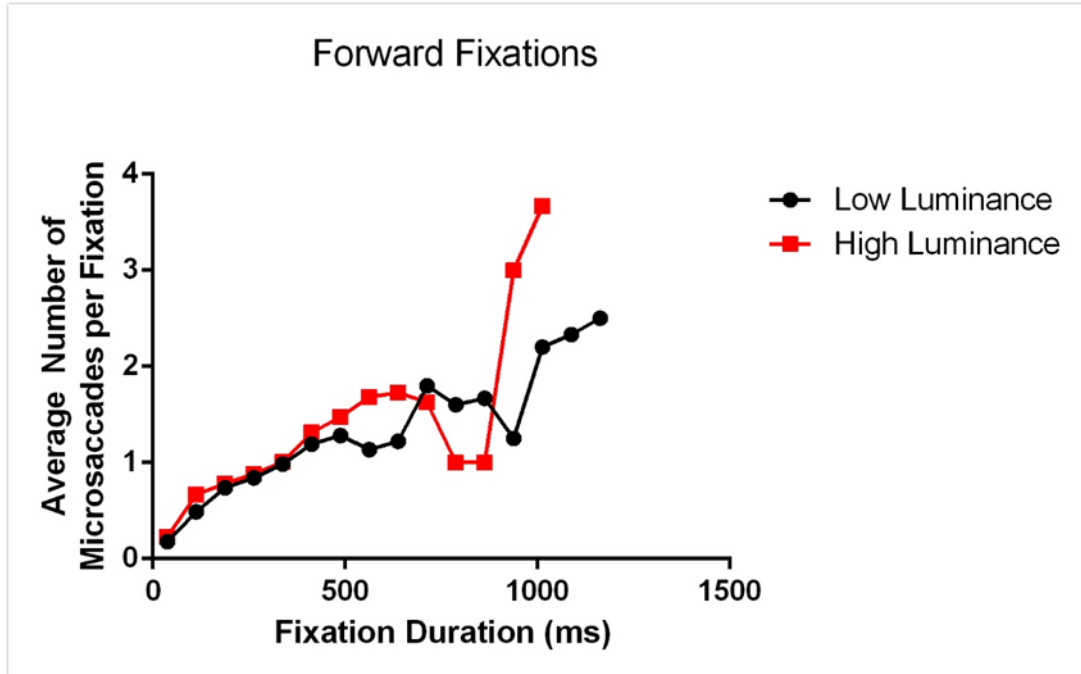


Figure 53 Average Number of Microsaccades per forward fixation as a function of fixation duration

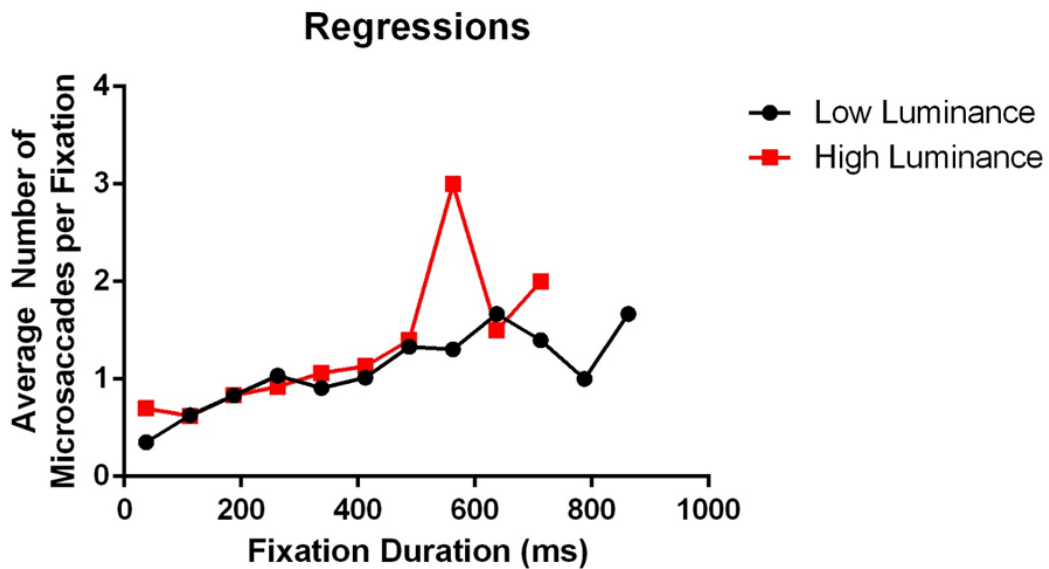


Figure 54 Average Number of Microsaccades per backward fixation as a function of fixation duration

As we can see the number of microsaccades per fixation increases as fixation duration increases, so we can assume that dependency between the two variables exists.

The average rate of microsaccades per second, for forward fixations was 3,98 microsaccades/s (SD=3,42) in high luminance and 3,18 microsaccades/s (SD=2,86) in low luminance. For fixations which occurred after a backward saccade the average rate of microsaccades was 4,05 microsaccades/s (SD=3,39) in high luminance conditions and 3,33 microsaccades/s (SD=2,97) in low luminance conditions.

We noticed that despite the average number of microsaccades per fixation is higher in low luminance conditions, the average rate of microsaccades per second is higher in high luminance conditions.

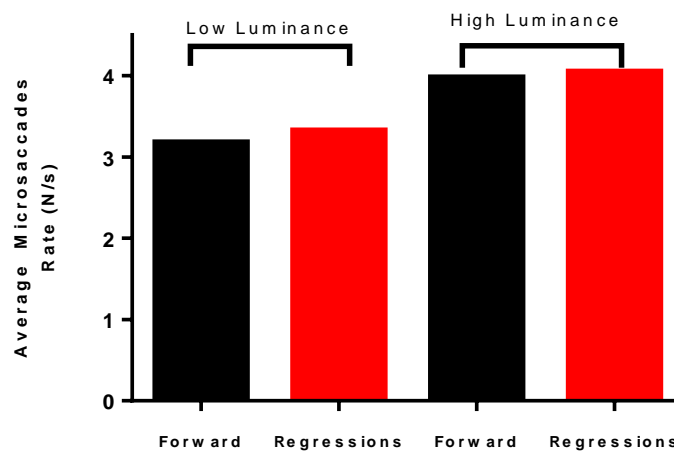


Figure 55 Average Microsaccades Rate in high and low luminance

The average microsaccades rate is presented in the following figures as a function of fixation duration.

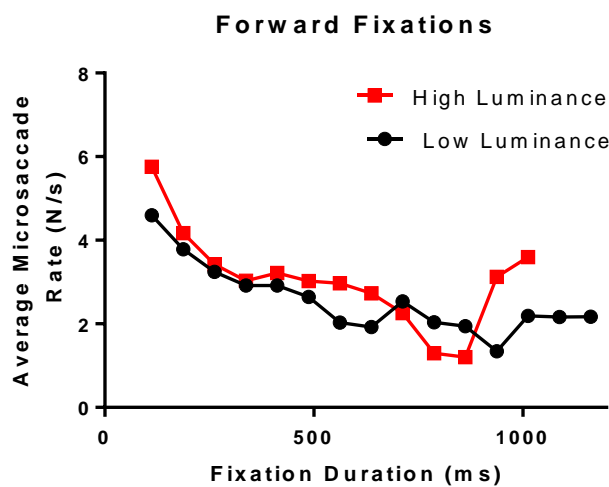


Figure 56 Average Microsaccade Rate as a function of Fixation duration for forward fixations

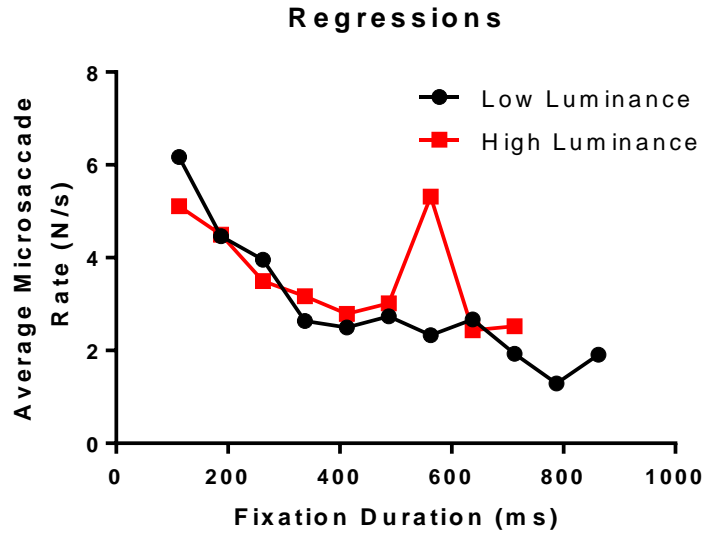


Figure 57 Average Microsaccade Rate as a function of fixation duration for fixations occurred after a regression

As we can see in both categories of fixations, the average rate of microsaccades is decreasing as fixation duration increases.

In the following figures we can see how the average number of microsaccades per fixation is affected as a function of fixation stability (68% Bivariate Contour Ellipse).

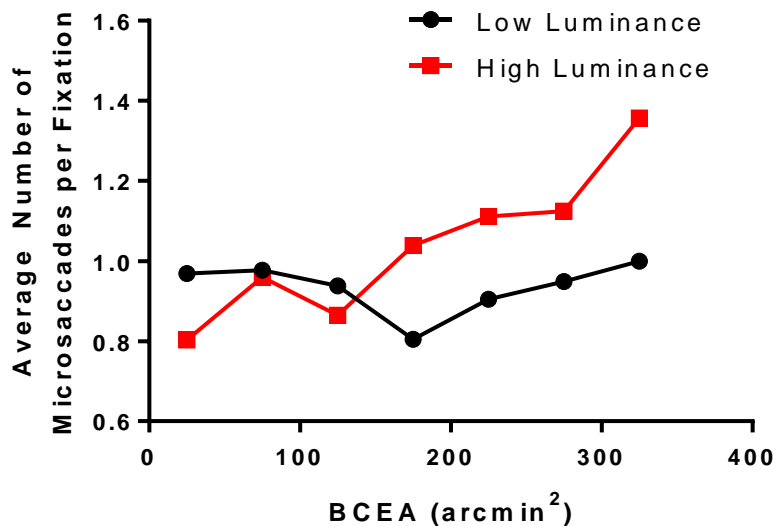


Figure 58 Average Number of Microsaccades per Fixation as a function of 68% BCEA for forward fixations

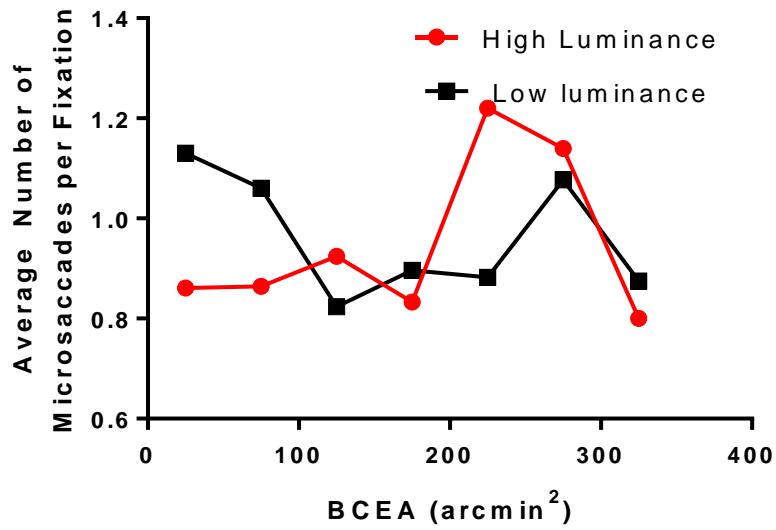


Figure 59 Average Number of Microsaccades per Fixation as a function of BCEA for backward fixations

Previously, we have depicted the Average Microsaccade Rate (N/sec) as a function of fixation duration. In the following graphs we present the average microsaccade rate as a function of 68% BCEA.

The distributions of 68% Bivariate Contour Ellipse Areas, are not normal and well fitted by ex-Gaussian density function as we have shown before. This means that for BCEAs larger than 200 arcmin² we only have a few fixations, so the results may not be accurate, especially for the microsaccades detected in regressions.

For fixations which occurred after forward fixations, the average microsaccade rate remains relatively stable for both luminance conditions.

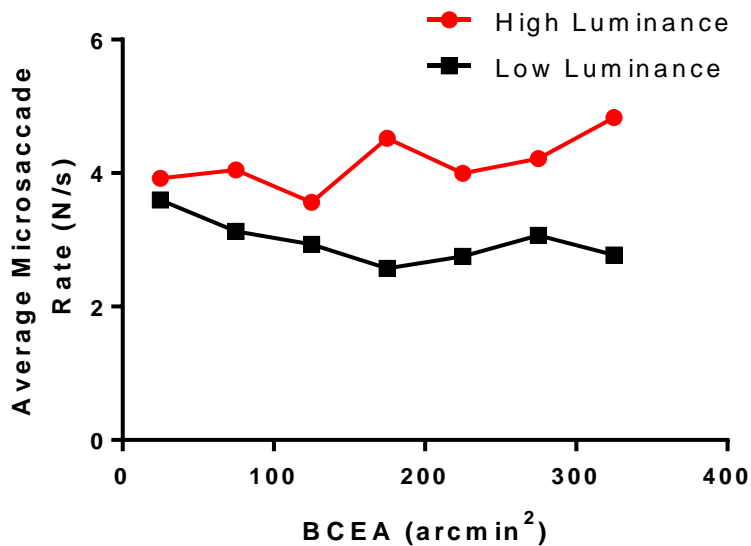


Figure 60 Average Microsaccade Rate as a function of 68% BCEA for forward fixations

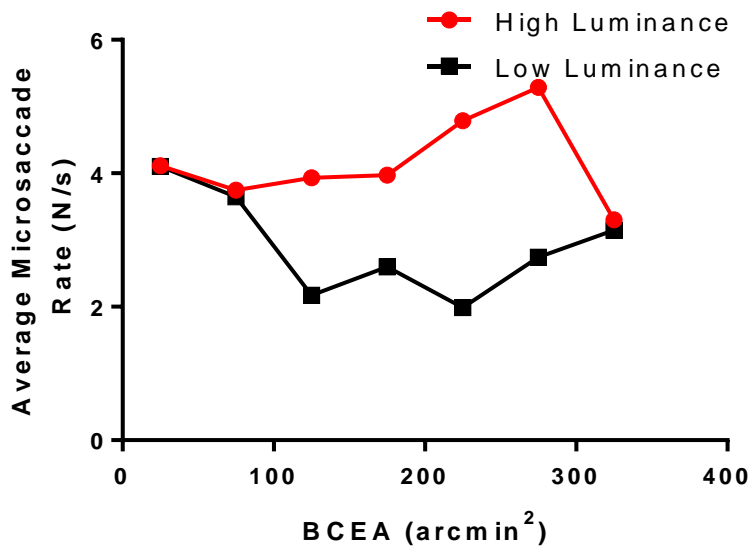


Figure 61 Average Microsaccade rate as a function of 68% BCEA for backward fixations

6.3.2 Characteristics of Microsaccades during reading

6.3.2.1 Amplitude and Peak Velocity

In this section of this study we will analyze the characteristics of the microsaccades the algorithm have detected, separately for fixations after forward and backward saccades.

It is known from previous studies that peak velocity and amplitude of microsaccades are linearly correlated, a property which is also valid for saccades.

For both categories of fixations, forward and regressions we found a statistical significant correlation between those two variables. In high luminance conditions, the Pearson correlation coefficient was $r=0,64$ ($p<0,001$) for fixations occurred after forward saccades and $r=0,68$ ($p<0,001$) for fixations occurred after regressions. In low luminance conditions, the Pearson correlation coefficient was $r=0,64$ ($p<0,001$) for fixations which occurred after forward saccades and $r=0,60$ ($p<0,001$) for fixations occurred after backward saccades.

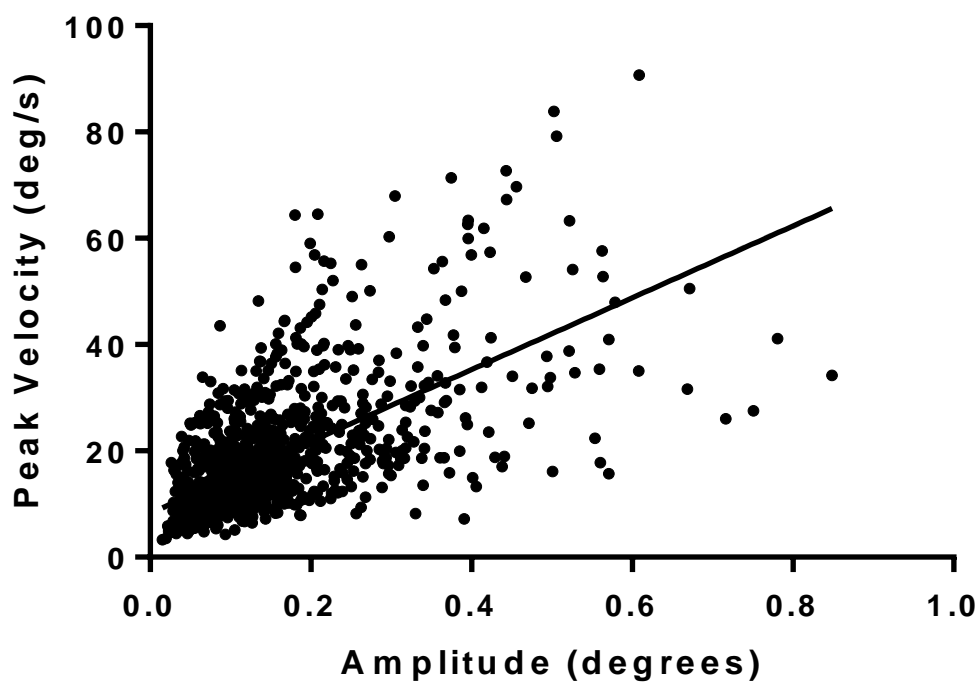


Figure 62 Scatterplot of Amplitude and Peak Velocity of microsaccades detected in forward fixations in high luminance conditions

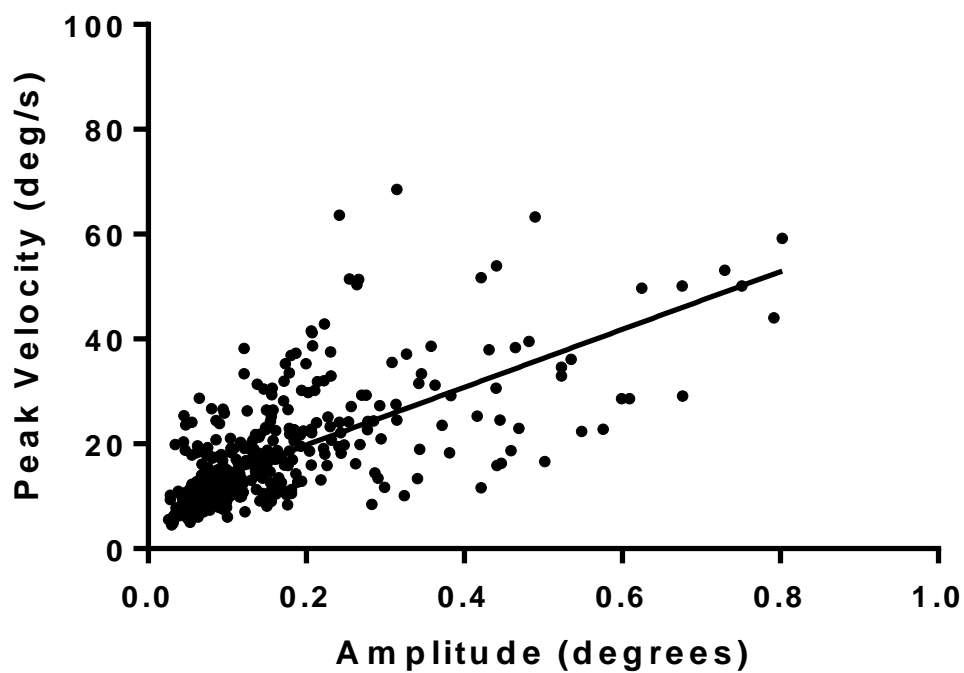


Figure 63 Scatterplot of Amplitude and Peak Velocity of microsaccades detected in fixations occurred after backward saccades in high luminance conditions

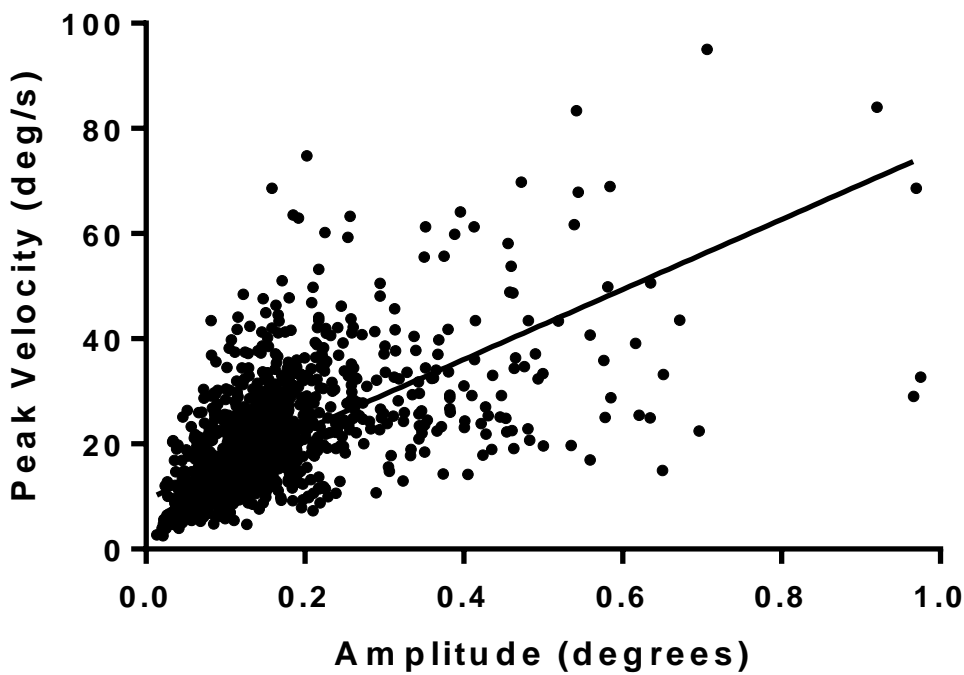


Figure 64 Scatterplot of Amplitude and Peak Velocity of microsaccades occurred after forward fixations in low luminance conditions

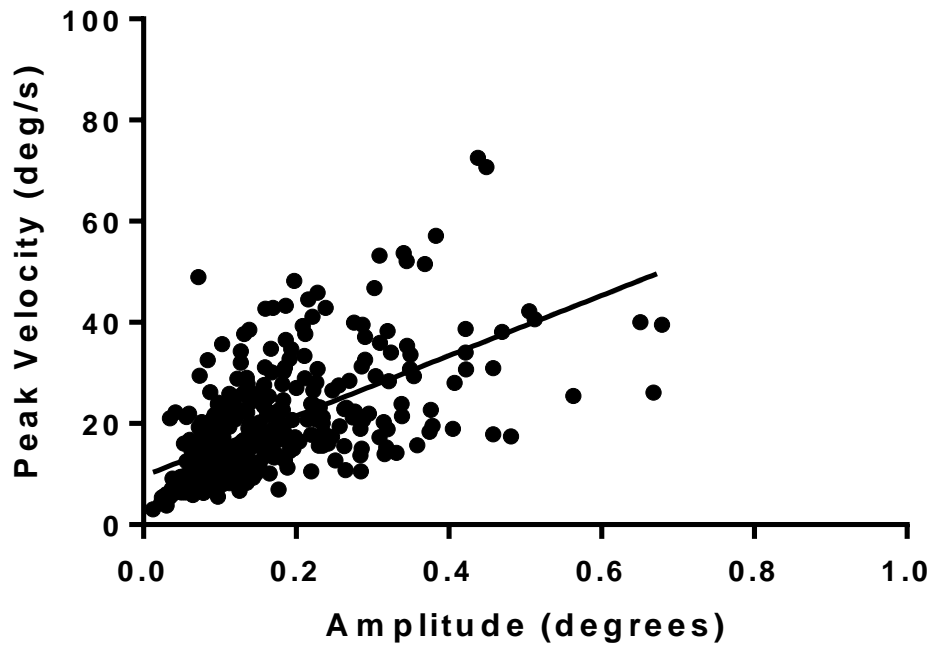


Figure 65 Scatterplot of Amplitude and Peak Velocity of microsaccades detected in fixations which occurred after backward saccades in low luminance conditions

Using Shapiro-Wilk test we have confirmed that both peak velocity and amplitude for microsaccades detected in both categories of fixations, in high and low luminance conditions are not normally distributed (p value was less than 0,001 for all distributions).

The median microsaccades' amplitude is illustrated in the following figure. The median amplitude in high luminance conditions was 0,105 degrees (SD=0,105) for forward fixations and 0,105 degrees (SD=0,134) for backward fixations. In low luminance conditions the median amplitude was 0,12 degrees (SD=0,11) for forward fixations and 0,132 degrees (SD=0,11) for backward fixations.

The microsaccades we detected in low luminance conditions had larger amplitude than in high luminance conditions. Also in both conditions the amplitude was slightly larger for microsaccades, detected in fixations that occurred after a backward saccade.

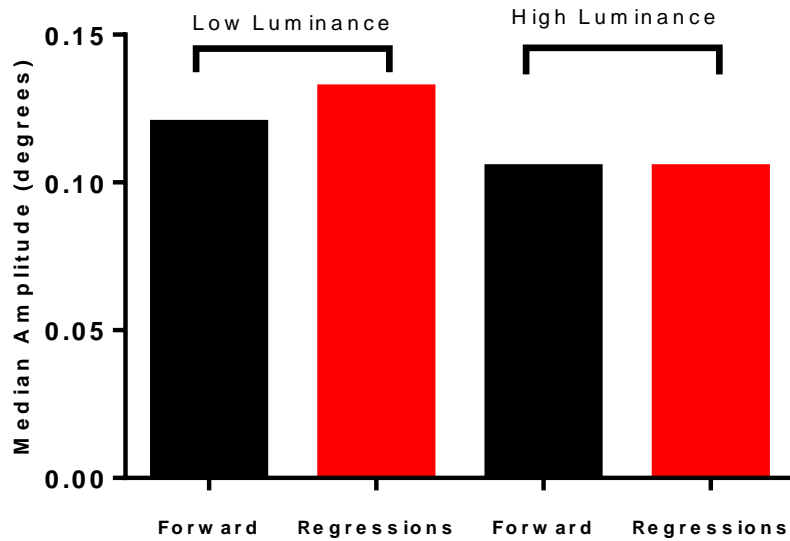


Figure 66 Median Microsaccades' Amplitude in high and luminance conditions

The median microsaccades' peak velocity is illustrated in the following figure. The median peak velocity in high luminance conditions was 14,32 deg/s (SD=11,11) for forward fixations and 13,78 deg/s (SD=10,82) for backward fixations. In low luminance conditions the median amplitude was 16,75 deg/s (SD=11,98) for forward fixations and 16,81 deg/s (SD=11,33) for backward fixations.

The median peak velocity was higher in low luminance conditions for fixations which occurred after forward and backward saccades. Which was expected as the amplitude and peak velocity of a microsaccade are highly correlated, as we have seen previously.

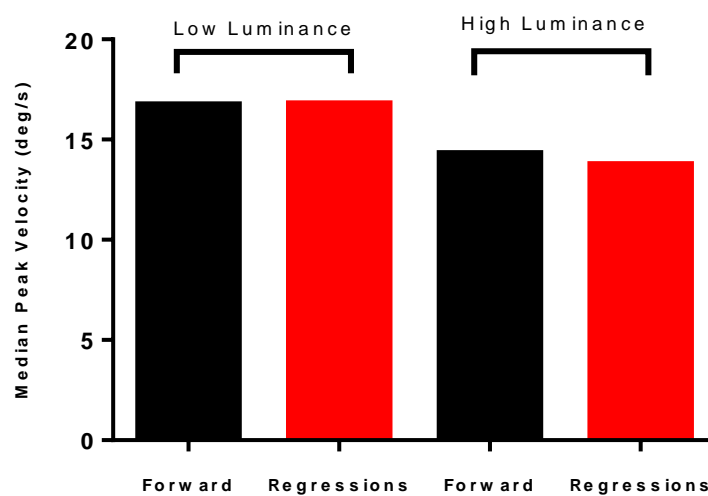


Figure 67 Median Microsaccades' Peak Velocity in high and low luminance conditions

As discussed before, ex-Gaussian density functions fit well to positive skewed distributions. The microsaccades' amplitude and peak velocity distributions are also positive skewed so we can apply ex-Gaussian analysis.

To apply ex-Gaussian analysis in amplitude distributions we have grouped data in bins with a range of 0,025 degrees. In the following figures we present the ex-Gaussian functions which fitted well the amplitude distributions.

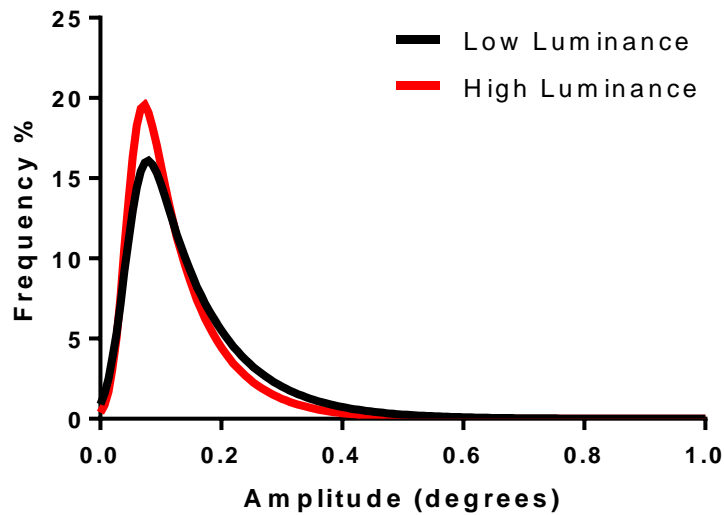


Figure 68 ex-Gaussian density functions of Amplitude for forward fixations in high and low luminance conditions

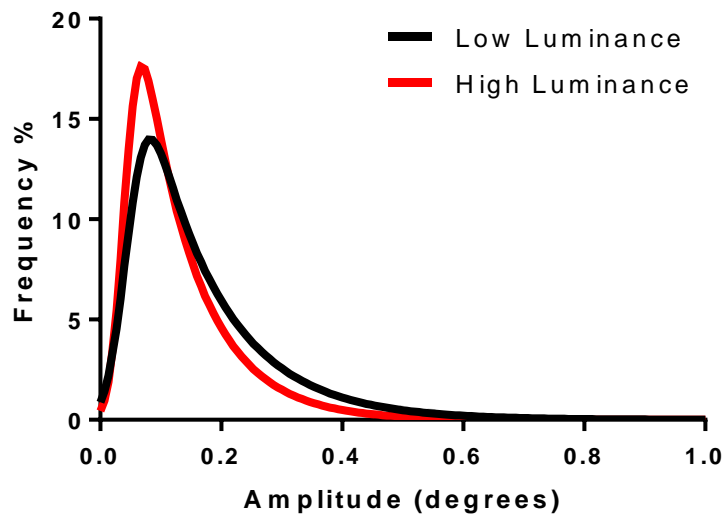


Figure 69 ex-Gaussian density functions for fixations which occurred after backward saccades in high and low luminance

The μ parameter remained constant for both high and low luminance conditions for fixations which occurred after forward saccades. We can see that only in high luminance μ was lower for fixations which occurred after regressions, and the ex-Gaussian density function was shifted slightly to the left.

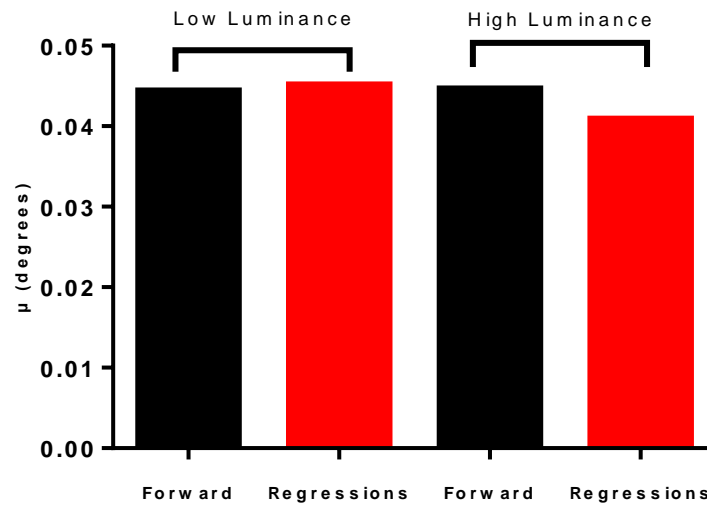


Figure 70 μ parameter of ex-Gaussian density function of microsaccades' amplitude in high and low luminance conditions

The σ parameter is higher in low luminance condition which is the reason why the ex-Gaussian density function is wider in low luminance conditions.

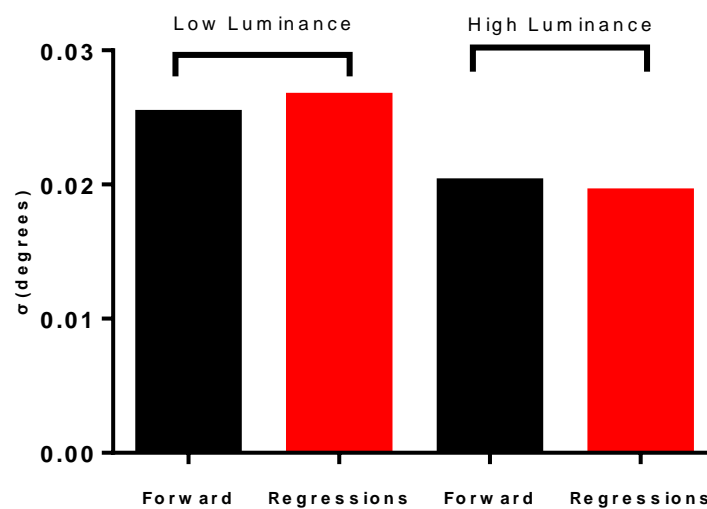


Figure 71 σ parameter of ex-Gaussian density function of microsaccades' amplitude in high and low luminance conditions

Parameter τ increases in low luminance conditions for both categories of fixations, this means that in high luminance the rate of the exponential component of the ex-Gaussian function decreases faster, so we can conclude that in low luminance conditions there are more microsaccades with high amplitude than in high luminance conditions.

Also the parameter τ is lower for microsaccades detected in fixations which occurred after backward saccades. In that case we have less fixations with high amplitude.

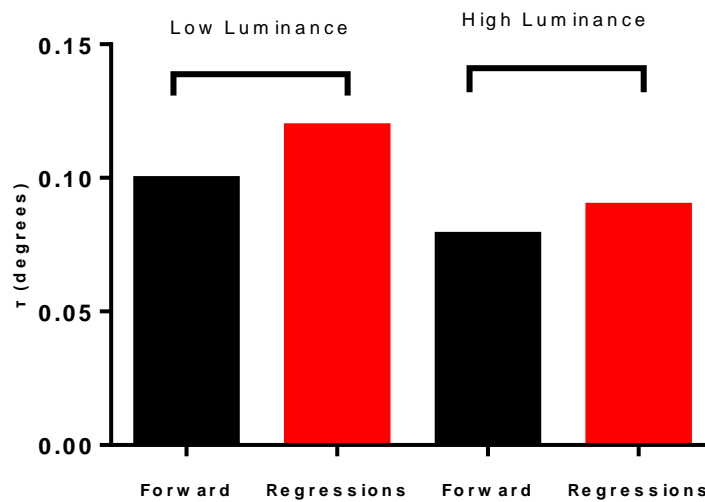


Figure 72 τ parameter of ex-Gaussian function of microsaccades' amplitude in high and low luminance conditions

To apply ex-Gaussian analysis in peak velocity distributions we have grouped data in bins with a range of 4 degs/sec. In the following figures we present the ex-Gaussian functions which fitted well the peak velocity distributions distributions.

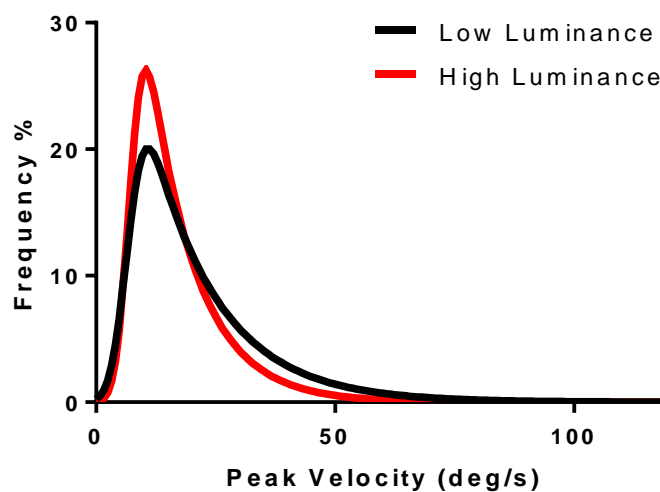


Figure 73 ex-Gaussian functions of microsaccades peak velocity in high and low luminance conditions for fixations which occurred after forward fixations

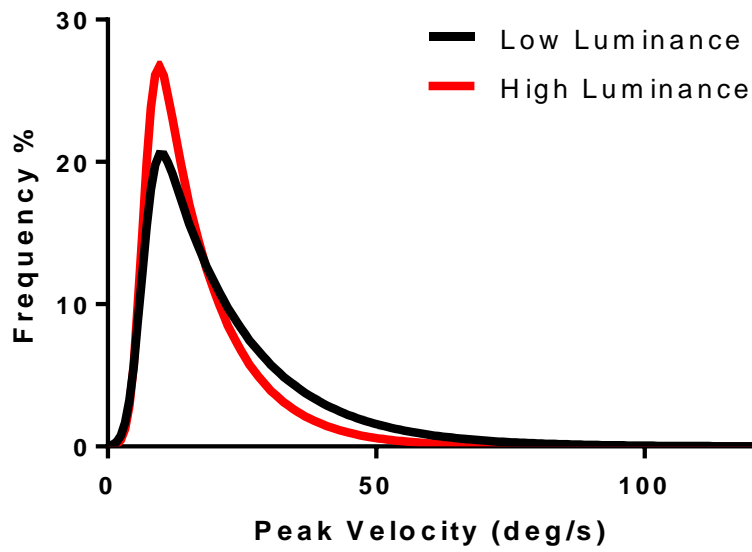


Figure 74 ex-Gaussian functions of microsaccades peak velocity in high and low luminance conditions for fixations which occurred after backward saccades

The μ parameter is higher in high luminance conditions, and as we can see the ex-Gaussian function for low luminance is shifted slightly to the left. ()

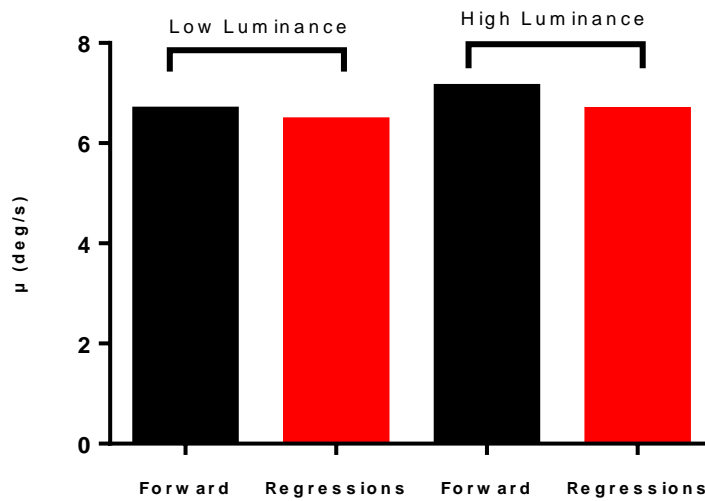


Figure 75 μ parameter of ex-Gaussian function of peak velocity

Parameter σ is higher in low luminance conditions for both categories of fixations, and the density function is wider. Also for microsaccades detected in regressions the parameter is lower than in forward fixations.

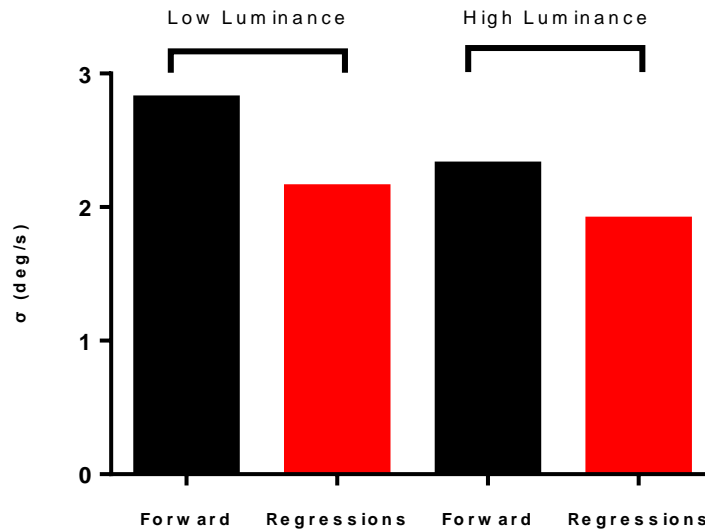


Figure 76 σ parameter of ex-Gaussian function of peak velocity

Parameter τ is higher in low luminance conditions, which means that the exponential component of the ex-Gaussian density function decreases slower, therefore the frequency of higher peak velocities is higher in low luminance.

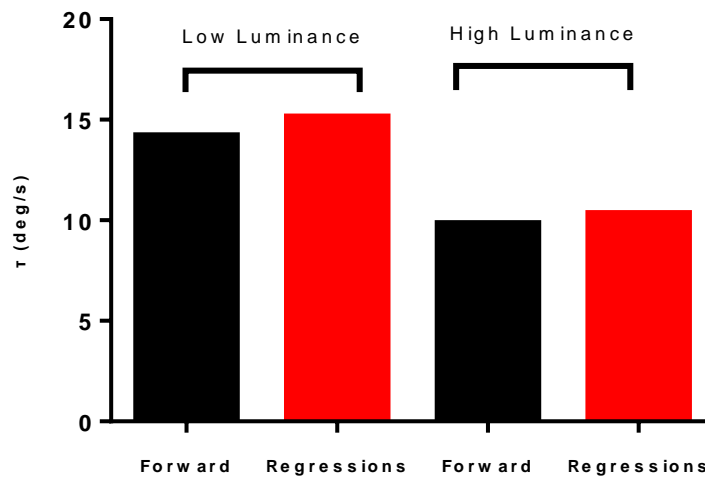


Figure 77 τ parameter of ex-Gaussian function of peak velocity

6.3.2.2 Orientation of microsaccades during reading

Another important characteristic of microsaccades is their orientation. Using the algorithm, as described in methods, we are able to calculate the angle of microsaccades.

In the following figures, we present the angular distribution of the detected microsaccades in forward fixations, in high and low luminance conditions.

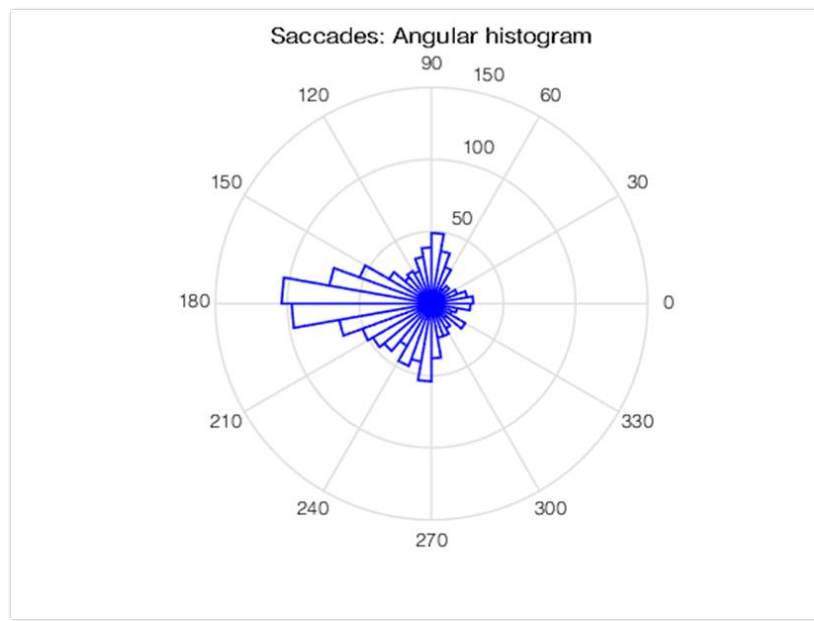


Figure 78 Angular distribution of microsaccades' orientation in high luminance for forward fixations

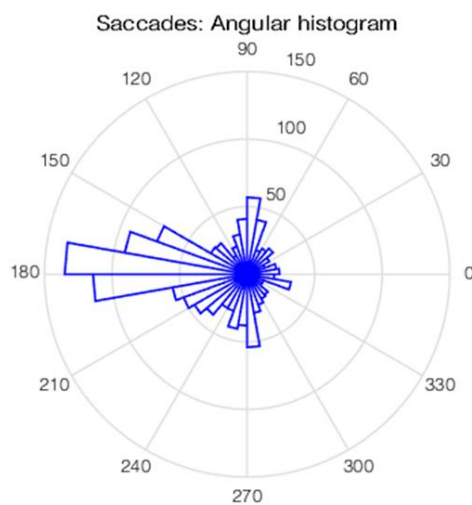


Figure 79 Angular distribution of microsaccades' orientation in low luminance for forward fixations

As we can see the majority of microsaccades, detected in forward fixations were left-orientated. In high luminance conditions 66,7% and in low luminance conditions 65,4% of the total number of microsaccades detected in forward fixations were left-orientated.

The following figures present the angular distribution of microsaccades which have been detected in regressive fixations.

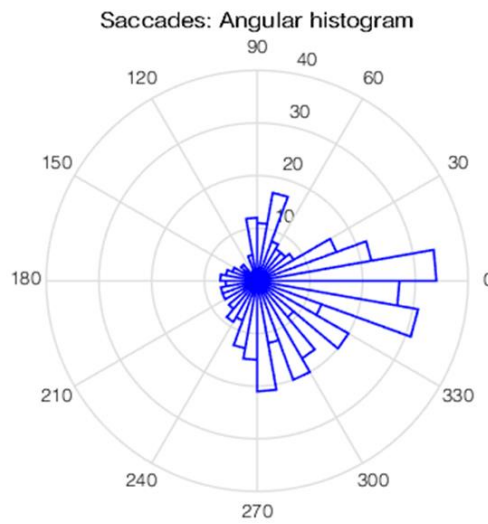


Figure 80 Angular distribution of microsaccades' orientation in high luminance for regressive fixations

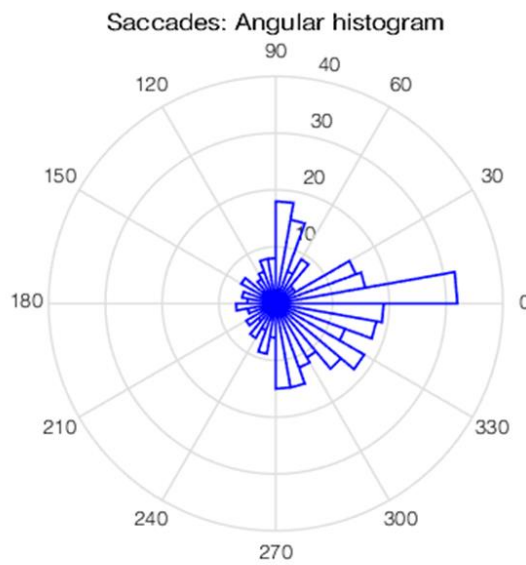


Figure 81 Angular distribution of microsaccades' orientation in low luminance for regressive fixations

The majority of microsaccades, detected in fixations which occurred after regressions were right-oriented. In high luminance conditions, 70% and in low luminance 71% of the detected microsaccades were right-oriented.

The orientation for the majority of the detected microsaccades was opposite to the previous saccade for both forward and regressive fixations.

PART 4. DISCUSSION

7. Conclusions & Future Work

In this study, we tried to evaluate fixations during reading, based mainly on fixation characteristics such as fixation duration, fixation stability and the microsaccades we have detected in each fixation and examine the effect of luminance in these factors. We have also separated the detected fixations, in three categories, based on the saccade which occurred before each fixation (forward, backward and return sweep saccades). We have analyzed all the factors we have described above for fixations which occurred right after forward and backward saccades as those fixations are responsible for obtaining visual information during reading. We have also used the ex-Gaussian analysis, as it is well known that ex-Gaussian functions fit well fixation duration distributions. As ex-Gaussian functions fit well positive skewed distributions, we have used ex-Gaussian analysis for 68% BCEA and microsaccades peak velocity and amplitude distributions.

We have shown that luminance didn't have any effect on the number of detected fixations (includes forward, backward and return sweep fixations), but also the number of each category separately. The percentage of fixations detected after forward saccades was higher in low luminance conditions, while the percentage of fixations detected after regressions was lower. We didn't also find any statistical significant difference in the percentage of return sweep fixations.

Using Shapiro-Wilk test for normality we have shown that both fixation duration and 68% BCEA (measure of fixation's stability) were not normally distributed. Median fixation duration and median 68% BCEA were higher in low luminance conditions for both forward and regressive fixations. The ex-Gaussian analysis showed also that fixation duration is affected from luminance. Parameters μ and σ increased in low luminance conditions while parameter τ remained stable. The ex-Gaussian analysis for 68% BCEA distributions showed that BCEA is affected also by luminance. The μ parameter remained relatively stable, also σ parameter was not affected by luminance, as it remained stable, but it was higher for forward fixations. Finally parameter τ was higher in low luminance.

We examined also if there was any relationship between fixation duration and fixation stability. For both forward and backward fixations, there was a statistically significant weak positive correlation in both high and low luminance conditions.

The average number of microsaccades per fixation was higher in low luminance conditions, while the average microsaccade rate (N/sec) was higher in high luminance conditions. This is an unexpected result but probably as the fixation duration is lower in high luminance, the mechanism of microsaccades generates more microsaccades in shorter time period to move the image towards the fovea. We have also shown that the average number of microsaccades increases as a function of fixation durations, while the average microsaccades rate decreases. The average number of microsaccades per fixation and the average microsaccades rate (N/sec) as a function of 68% BCEA remained relatively stable.

In a previous study (Otero-Millan, 2008), researchers have shown that the average microsaccade rate (N/sec) for microsaccades detected in image viewing, were lower than the rate we have found. But they have also shown that the average microsaccades rate (N/sec) is higher for more complicated images, and have suggested a strong relationship between microsaccade generation and target detection during visual search. This leads us to believe that for a demanding task like reading the average microsaccade rate will be higher, which is supported by other studies (Ko, 2010).

The basic characteristics of microsaccades have been detected, such as amplitude and peak velocity. We have found a statistically significant correlation between amplitude and peak velocity, which is supported by bibliography for microsaccades and saccades. Both amplitude and peak velocity were not normally distributed. The median amplitude of microsaccades was higher in low luminance conditions, and the median peak velocity was also slightly higher in low luminance, a result we expected as those measures are highly correlated.

We have applied ex-Gaussian analysis to amplitude and peak velocity, and in both cases showed a very good fit. The μ parameter of the amplitude's ex-Gaussian function remained stable for both luminance conditions, while σ parameter was higher in low luminance conditions. Parameter τ was higher in low luminance but also higher for regressive fixations comparing to forward fixations. For the peak velocity ex-Gaussian function, μ parameter was higher in high luminance, while parameters σ and τ were higher in low luminance conditions.

Finally the orientation of the microsaccades is very important to understand their role in reading. We have found that the majority of the detected microsaccades are orientated to the opposite direction of the previous saccade. This shows that the mechanism which generates microsaccades functions is possible the same as the saccades mechanism, in order to move the image towards the fovea at anytime, with microsaccades function being a corrective mechanism.

Further studies can apply the analysis we have presented in this study to unveil the effect of contrast, print size, age and other personality characteristics in fixations characteristics during reading. Future studies can also focus on analyzing the different role of the two eyes during reading, with an emphasis on the dominant eye. The same analysis can be performed to analyze reading for patients with ocular diseases or neurological disorders.

APPENDIX

ALGORITHM

```
clc
clear all
tic
%read excel file
filename = 'file.xlsx';
sheet = 1;
xlsrange = 'A2:H300000';
%Valid Trials
Trials=[5];
%number of words in text
num_words=141;
%read matrix with data
data=xlsread(filename,sheet,xlsrange);
%Input 8 columns
%1|Timestamp
%2|Trial Index
%3|Right Fixation Number
%4|Right Gaze x
%5|Right Gaze y
%6|Left Fixation Number
%7|Left Gaze x
%8|Left Gaze y
%remove fixations with blinks/lost track

%conditions to delete data rows when trial is 2 or 3 and when
x,y and fix are 0
%TF1=data(:,2)==2; %deletes trial 2 rows
%TF2=data(:,2)==3; %deletes trial 3 rows
TF3=isnan(data(:,3));
TF4=isnan(data(:,4));
TF5=isnan(data(:,5));
TF6=isnan(data(:,6));
TF7=isnan(data(:,7));
TF8=isnan(data(:,8));
% % %combine conditions
TFall = TF3|TF4|TF5|TF6|TF7|TF8;
%detect fixations with blinks or lost track etc
TFblinks=TF4|TF5|TF7|TF8;
z=data(TFblinks,2:3);
TF9=isnan(z(:,2));
z(TF9,:)=[];
z=unique(z,'rows');
%remove nans
```

```

data(TFall, :) = [];

% default values for microsaccade detection
SAMPLING=500; %Sampling Frequency
time=1000/SAMPLING; %time between two consecutive recordings
MINDUR=3; %Minimum Duration in Points
VFAC=4;
clusterdist=25;
clustermode=4;
smoothtype=2; %Default Value
degperpixel=0.0559529;
arcminperpix=3.35717431;
sacfin=[];
sacfin_reg=[];
matr=[];
%Dominant EYE RE for right eye or LE for left eye (default
value is RE)
OD='RE';
for i=Trials
    switch i
        case 1
            tit='Low Luminance';
        case 4
            tit='High Luminance';
    end
    data2=[];
    ind = data(:,2) == i;
    data2=data(ind, :);
    %Reading Speed
    RDTIME=max(data2(:,1))-min(data2(:,1)); %ms
    RDTIME=RDTIME/60000; %time in minutes
    RDSPEED(i)=num_words/RDTIME; %words per minute

    %fixation with blinks in current trial
    blinkfix=[];
    indblink=z(:,1)==i;
    blinkfix=z(indblink,2);
    blinkfix=blinkfix';

    %swap data for DE if necessary
    if OD == 'LE'
        tempfix=data2(:,3);
        tempx=data2(:,4);
        tempy=data2(:,5);
        data2(:,4)=data2(:,7);
        data2(:,5)=data2(:,8);
        data2(:,6)=tempfix;
        data2(:,7)=tempx;
        data2(:,8)=tempy;
    end

    fix_index=unique(data2(:,3));
    %find forward fixations
    for j=fix_index'

```

```

    %clear matrices
    data3=[];
    ind2=data2(:,3) == j;
    data3=data2(ind2,:);
    xmean(j,i)=mean(data3(:,4));
    ymean(j,i)=mean(data3(:,5));
end
%first-last fixations
first=min(fix_index);
last=max(fix_index);
num_for_fix=0;
%create a matrix for discarded fixations
discarded=[];
for j=(first+1):last
    %check if forward fixation
    if xmean(j,i) > xmean(j-1,i)
        num_for_fix=num_for_fix+1;
        for_fix(num_for_fix,i)=j;
    else
        discarded=[discarded;j];
    end
end
%eliminate fixations with blinks/track loss from for_fix
tempfor=for_fix(:,i);
tempfor=tempfor(~ismember(tempfor,blinkfix'));
for_fix(:,i)=[];
for_fix(1:length(tempfor),i)=tempfor;
fix_stats(1,i)=length(tempfor);
lastinline=[];
firstinline=[];
prevfirst=0;
prevlast=0;
%check for first/last fix in each line
for j=(first+5):(last-5)
    if xmean(j,i)>xmean(j-1,i) && xmean(j,i)>xmean(j-2,i)
    && xmean(j,i)>xmean(j-3,i) && xmean(j,i)>xmean(j-4,i) &&
    xmean(j,i)>xmean(j-5,i) && xmean(j,i)>xmean(j+1,i) &&
    xmean(j,i)>xmean(j+2,i) && xmean(j,i)>xmean(j+3,i) &&
    xmean(j,i)>xmean(j+4,i) && xmean(j,i)>xmean(j+5,i)
        if prevlast==0
            lastinline=[lastinline;j];
            prevlast=j;
        elseif (prevfirst>prevlast) && (j>prevfirst)
            lastinline=[lastinline;j];
            prevlast=j;
        end
    elseif xmean(j,i)<xmean(j-1,i) && xmean(j,i)<xmean(j-
    2,i) && xmean(j,i)<xmean(j-3,i) && xmean(j,i)<xmean(j-4,i) &&
    xmean(j,i)<xmean(j-5,i) && xmean(j,i)<xmean(j+1,i) &&
    xmean(j,i)<xmean(j+2,i) && xmean(j,i)<xmean(j+3,i) &&
    xmean(j,i)<xmean(j+4,i) && xmean(j,i)<xmean(j+5,i)
        if prevfirst==0
            firstinline=[firstinline;j];
            prevfirst=j;
        elseif (prevlast>prevfirst) && (j>prevlast)
            firstinline=[firstinline;j];

```

```

        prevfirst=j;
        end
    end
end
firstline(:,i)=firstinline;
lastline(:,i)=lastinline;
% %add first fixations to forward fixations & delete them
from discarded
% %fixations
% for_fix=[for_fix;firstline];
% for_fix(:,i)=sort(for_fix(:,i));
% for w=1:length(firstline(:,i))
% discarded(discarded==firstline(w,i))=[];
% end
%find fixations between first-last and delete them from
discarded
%fixations (change of line fixations)
choffline=[];
%merge last-first matrices
%TF11=lastline(:,1)==144;
%lastline(lastline==103)=[];
%lastline(TF11,:)=[];
%lastline=[10;lastline];
firstlast=[firstline(:,i),lastline(:,i)];
%find values between them
for w=1:length(firstline(:,i))
    change_line=(lastline(w,i)+1):1:(firstline(w,i)-1);
    choffline=[choffline;change_line'];
    fix_stats(3,i)=length(choffline);
end
%the remaining discarded are regressions
for w=1:length(choffline)
discarded(discarded==choffline(w))=[];
end
regressions(1:length(discarded),i)=discarded;
tempreg=regressions(:,i);
%eliminate fixations with blinks/track loss from regressions
tempreg=tempreg(~ismember(tempreg,blinkfix));
regressions(:,i)=[];
regressions(1:length(tempreg),i)=tempreg;
fix_stats(2,i)=length(tempreg);
count=0;
count1=0;
for j=fix_index'
    if ismember(j,for_fix(:,i))
        ind3=data2(:,3) == j;
        data3=data2(ind3,:);
        %clear x,y
        xr=[];
        xl=[];
        xrarc=[];

        %clear matrices
        sac=[];
        sacr=[];
    end
end

```

```

sac1=[];

%assign x,y values
xr(:,1)=data3(:,4);
xr(:,2)=data3(:,5);
xl(:,1)=data3(:,7);
xl(:,2)=data3(:,8);

count=count+1;
%bcea calculation
%x,y in arcmins
xrarc(:,1)=xr(:,1)*arcminperpix;
xrarc(:,2)=xr(:,2)*arcminperpix;

[bcearea(count,i),angle(count,i)]=bcea(xrarc(:,1),xrarc(:,2));
%area2(count,i)=bcea(xl(:,1),xl(:,2));

%fixation duration
fix_dur(count,i)=time*length(xr(:,1));
%microsaccades detection
%Velocity calculation
%clear velocity matrices
vr=[];
vl=[];
vr=vecvel(xr,SAMPLING,smoothtype);
vl=vecvel(xl,SAMPLING,smoothtype);
%Compute Velocity threshold for each eye
[msdxr,msdyr]=velthresh(vr);
[msdxl,msdyl]=velthresh(vl);
%Detection of microsaccades
sacr=microsacc(xr,vr,VFAC,MINDUR,msdxr,msdyr);
sac1=microsacc(xl,vl,VFAC,MINDUR,msdxl,msdyl);
%Detection of binocular microsaccades
[sac,monol,monor]=binsacc(sac1,sacr);
sac = saccpar(sac); % average saccade characteristics of
both eyes
sac = mergesacc(sac,(xl+xr)./2,clusterdist,clustermode);
% merge nearby saccades (e.g. glissades)

if ~isempty(sac)
% define saccade duration as difference between
saccade offset and
% saccade onset sample. In saccpar(), monocular
saccade durations
% of both eyes are averaged, leading to uneven values
(e.g.: 10.5
% samples) different from the difference between onset
and offset
% values (which are the monocular extremes).
% Instead: use difference between offset and onset
sac(:,3) = sac(:,2)-sac(:,1)+1;

% report saccade velocity/distance/amplitude as visual
angles

```

```

sac(:, [5 6 8]) = sac(:, [5 6 8]).* degperpixel;

% report saccade angles in degree rather than radians
sac(:, [7 9]) = sac(:, [7 9]) * 180/pi;
%add index of trial & fixation
sac(:, 10)=i;
sac(:, 11)=count;
sacfin=[sacfin;sac];
sacnum(count, i)=length(sac(:, 1));

%plot trajectory
if count==4 && i==4
    xls=xl*arcminperpix;
    xrs=xr*arcminperpix;
    xls=smoothdata(xl);
    xrs=smoothdata(xr);
    xrs(:, 1)=xrs(:, 1)-mean(xrs(:, 1));
    xrs(:, 2)=xrs(:, 2)-mean(xrs(:, 2));
    xls(:, 1)=xls(:, 1)-mean(xls(:, 1));
    xls(:, 2)=xls(:, 2)-mean(xls(:, 2));

    N=length(sac(:, 1));
    N2=length(xls(:, 1));
    N4=length(xrs(:, 1));

    figure(195)
    subplot(1, 2, 1)

    plot(xls(:, 1), xls(:, 2), 'ko')
    title('Left Eye')
    xlabel('Horizontal Position [arcmin]')
    ylabel('Vertical Position [arcmin]')

    for s=1:N2-1

line(xls(s:s+1, 1), xls(s:s+1, 2), 'Color', 'k', 'LineWidth', 2)
    end
    for s=1:N
        onset=sac(s, 1);
        offset=sac(s, 2);

line(xls(onset:offset, 1), xls(onset:offset, 2), 'Color', 'r', 'Line
Width', 2)
    end
    subplot(1, 2, 2)
    plot(xrs(:, 1), xrs(:, 2), 'ko')
    title('Right Eye')
    xlabel('Horizontal Position [arcmin]')
    ylabel('Vertical Position [arcmin]')
    for s=1:N4-1

line(xrs(s:s+1, 1), xrs(s:s+1, 2), 'Color', 'k', 'LineWidth', 2)
    end

```



```

        for s=1:N
            onset=sac(s,1);
            offset=sac(s,2);

line(xrs(onset:offset,1),xrs(onset:offset,2),'Color','r','Line
Width',2)
        end
        sh(1)=subplot(1,2,1);
        sh(2)=subplot(1,2,2);
        axis(sh,'square');

        N4=length(vl(:,1));
        N5=length(vr(:,1));

        vl=vl*degperpixel;
        vr=vr*degperpixel;
        [msdxr,msdyr]=velthresh(vr);
        [msdxl,msdyl]=velthresh(vl);
        figure(148)
        subplot(1,2,1)
        plot(vl(:,1),vl(:,2),'ko')
        title('Left Eye')
        xlabel('v_x [deg/s]')
        ylabel('v_y [deg/s]')
        for s=1:N4-1

line(vl(s:s+1,1),vl(s:s+1,2),'Color','k','LineWidth',2)
        end
        for s=1:N
            onset=sac(s,1);
            offset=sac(s,2);

line(vl(onset:offset,1),vl(onset:offset,2),'Color','r','LineWi
dth',2)

            hold on
            %text(s)
        end

        %plot elliptic threshold
        radiusxl = VFAC*msdxl;
        radiusyl = VFAC*msdyl;
        radiusl = [radiusxl radiusyl];
        phi=0:pi/150:2*pi;
        cx=radiusxl*cos(phi);
        cy=radiusyl*sin(phi);
        hold on
        plot(cx,cy,'--')

        subplot(1,2,2)
        plot(vr(:,1),vr(:,2),'ko')
        title('Right Eye')
        xlabel('v_x [deg/s]')
        ylabel('v_y [deg/s]')
        for s=1:N5-1

```

```

line(vr(s:s+1,1),vr(s:s+1,2),'Color','k','LineWidth',2)
    end
    for s=1:N
        onset=sac(s,1);
        offset=sac(s,2);

line(vr(onset:offset,1),vr(onset:offset,2),'Color','r','LineWi
dth',2)
    end
    %plot elliptic threshold
    radiusxr = VFAC*msdxr;
    radiusyr = VFAC*msdyr;
    radiusr = [radiusxr radiusyr];
    phi=0:pi/150:2*pi;
    cx=radiusxr*cos(phi);
    cy=radiusyr*sin(phi);
    hold on
    plot(cx,cy,'--')
    sh(1)=subplot(1,2,1);
    sh(2)=subplot(1,2,2);
    axis(sh,'square');
    end
else
    sacnum(count,i)=0;
end

```

```

    %Matrix with results for forward fixations
    %BCEA-Number of Microsaccades-Fixation Duration

```

```

results_for{i}=[bcearea(:,i),sacnum(:,i),fix_dur(:,i)];

```

```

%         testmat=1:10;
%         if ismember(count,testmat);
%         figure(count)
%         t=[];
%         t=time:time:fix_dur(count,i);
%         mesh(t,xr(:,1),xr(:,2))
%         hold on
%         plot(t,xl(:,1),'-')

%         figure(count+100)
%         plot(t,xr(:,2),'-')
%         hold on
%         plot(t,xl(:,2),'-')
%         end

```

```

% title(['time-BCEA, ' tit])
% xlabel('time (ms)')
% ylabel('BCEA (arcmin^2)')

% columns of [sac]:
% 1: saccade onset (sample)
% 2: saccade offset (sample)
% 3: duration (samples)
% 4: NaN (originally: delay between eyes in
samples)
% 5: vpeak (peak velocity)
% 6: saccade distance
% 7: saccade angle (based on distance)
% 8: saccade amplitude
% 9: saccade angle (based on amplitude)

%regression analysis
elseif ismember(j,regressions(:,i))
ind3=data2(:,3) == j;
data3=data2(ind3,:);
%clear x,y
xr=[];
xl=[];
xrarc=[];

%clear matrices
sac=[];
sacr=[];
sacl=[];

%assign x,y values
xr(:,1)=data3(:,4);
xr(:,2)=data3(:,5);
xl(:,1)=data3(:,7);
xl(:,2)=data3(:,8);

count1=count1+1;
%bcea calculation
%x,y in arcmins
xrarc(:,1)=xr(:,1)*arcminperpix;
xrarc(:,2)=xr(:,2)*arcminperpix;
bcearea_reg(count1,i)=bcea(xrarc(:,1),xrarc(:,2));

%fixation duration
reg_fix_dur(count1,i)=time*length(xr(:,1));
%microsaccades detection
%Velocity calculation
%clear velocity matrices
vr=[];
vl=[];
vr=vecvel(xr,SAMPLING,smoothtype);
vl=vecvel(xl,SAMPLING,smoothtype);
%Compute Velocity threshold for each eye

```

```

[msdxr,msdyr]=velthresh(vr);
[msdxl,msdyl]=velthresh(vl);
%Detection of microsaccades
sacr=microsacc(xr,vr,VFAC,MINDUR,msdxr,msdyr);
sac1=microsacc(xl,vl,VFAC,MINDUR,msdxl,msdyl);
%Detection of binocular microsaccades
[sac,monol,monor]=binsacc(sac1,sacr);
sac = saccpar(sac); % average saccade characteristics of
both eyes
sac = mergesacc(sac, (xl+xr)./2,clusterdist,clustermode);
% merge nearby saccades (e.g. glissades)

if ~isempty(sac)
    % define saccade duration as difference between
saccade offset and
    % saccade onset sample. In saccpar(), monocular
saccade durations
    % of both eyes are averaged, leading to uneven values
(e.g.: 10.5
    % samples) different from the difference between onset
and offset
    % values (which are the monocular extremes).
    % Instead: use difference between offset and onset
sac(:,3) = sac(:,2)-sac(:,1)+1;

    % report saccade velocity/distance/amplitude as visual
angles
sac(:, [5 6 8]) = sac(:, [5 6 8]).* degperpixel;

    % report saccade angles in degree rather than radians
sac(:, [7 9]) = sac(:, [7 9]) * 180/pi;
%add index of trial & fixation
sac(:,10)=i;
sac(:,11)=count;
sacfin_reg=[sacfin_reg;sac];
sacnum_reg(count1,i)=length(sac(:,1));
else
sacnum_reg(count1,i)=0;
end
end
end
polar_flipy=0;
sac_angle=[];
sac_angle=sacfin(:,7);

figure(34+i)
[t,r] = rose(sac_angle*pi/180,36); % angle in radians,
plot 10° bins
h = polar(t,r,'b-');
hline = findobj(gca,'Type','line');
set(hline,'LineWidth',1.2); % make line thicker
title('Saccades: Angular histogram','fontweight','bold')

```

```

        %Matrix with results for backward fixations
        %BCEA-Number of Microsaccades-Fixation Duration

results_reg{i}=[bcearea_reg(:,i),sacnum_reg(:,i),reg_fix_dur(:,i)];

end
for i=Trials
figure(170+i)
[t,r] = rose(angle(:,i)*pi/180,36); % angle in radians, plot
10° bins
h = polar(t,r,'b-');
hline = findobj(gca,'Type','line');
set(hline,'LineWidth',1.2); % make line thicker
title('BCEA: Angular histogram','fontweight','bold')
end
toc

```

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