Longitudinal Assessment of Pupil Response to Red and Blue Light in Youth Hockey Players

Thesis

Presented in Partial Fulfillment of the Requirements for the Degree Master of Science in the Graduate School of The Ohio State University

By

Kristen Zulliger

Graduate Program in Vision Science

The Ohio State University

2022

Thesis Committee

Andrew Hartwick, OD, PhD, Advisor

Phillip Yuhas, OD, PhD

Catherine McDaniel, OD, MS

Abstract

Sports-related concussions in pediatrics and adolescents can be asymptomatic and often go undiagnosed. Pupil testing strategies may be used as a potential objective screening tool for concussion. The purpose of this work was to perform longitudinal assessments of pupil responses to pulses of red and blue light in youth hockey players to assess the function of the neural circuitry mediating the pupillary light reflex, while also determining the repeatability of these responses. In addition, assessments of cognitive function and surveys of symptoms were performed to determine the repeatability of these concussion screening tools across the hockey season.

There were 18 males, each 13 years old, enrolled in the cohort study at the start of the youth hockey season. Pupil responses to flashes of alternating red and blue light, applied at 0.1 Hz for 2 min, were recorded using the RAPDx pupillometer (Konan Medical). The data was collected in three sessions, each separated by approximately two months, conducted at the start, middle and end of the hockey season. Average pupil constriction during the first red versus first blue pulses, and the difference in constriction between the first and last pulses of red light were calculated. Symptoms were quantified with the Convergence Insufficiency Symptoms Survey (CISS) and Rivermead Post-Concussion Symptoms Questionnaire (RPCSQ). Cognitive function was measured using CogState Brief Battery Assessment.

The average difference between the pupil responses to the first red and the first blue flash, a marker of the contribution of melanopsin-driven responses to pupil constriction, was significantly correlated ($R = 0.61$; $P < 0.001$) with the change in pupil constriction that occurred across the pupil test, termed photopotentiation (measured as the difference in pupil responses to the first and last red flashes). Bland-Altman analyses demonstrated that there was not a significant difference in the pupil responses to the first red and blue flashes within individuals in the second visit ($P=0.16$) and third visit ($P=0.81$), relative to the initial visit, indicating good repeatability of these responses across the hockey season. There was significant improvement across the hockey season on the Learning Task of the CogState assessment in Bantam-aged players. Processing Speed, Attention, and Working Memory Speed Task scores were repeatable across the season. The CISS and RPCSQ scores were highly correlated ($R^2 = 0.70$, $P < 0.001$), meaning individuals with greater visual symptom severity, as assessed with CISS, will also report higher symptom severity on RPCSQ.

Individuals that show greater pupil responses to blue light, relative to red light of similar irradiance, could be interpreted as having a greater contribution of melanopsin-driven responses to the neural circuitry that mediates the pupillary light reflex. In this population of youth hockey players, this marker showed high repeatability across the playing season. Individuals exhibiting greater responses to the blue light showed a greater change in pupil constriction from the beginning to the end of the test, supporting a role for melanopsin in this adaptive response to repeated stimuli. Future work will assess the alterations in the responses to this pupil test in youth athletes post-concussion. Score

improvements in the Learning Task of CogState need to be accounted for by clinicians when comparing follow-up testing to baseline. The high correlation between the CISS and RPCSQ suggests that there is overlap between the two surveys.

Dedication

To my family and friends who have supported me throughout this process.

Acknowledgments

I would like to thank my advisor, Dr. Hartwick, for his support and guidance throughout my time in the Master of Vision Science program. I would also like to thank my defense committee members, Dr. McDaniel and Dr. Yuhas, for their thoughtful contributions to my thesis and defense. This work was funded by a pilot study grant from Ohio State University Chronic Brain Injury Program.

Vita

Bachelor of Science…….The Ohio State University May 2018

Doctor of Optometry (Expected).……The Ohio State University May 2022

Master of Science (Expected)……..The Ohio State University May 2022

Fields of Study

Major Field: Vision Science

Professional Field: Optometry

Table of Contents

List of Figures

Chapter 1. Introduction

Youth sports are a popular activity, with roughly 54% of high school students reporting participation (Centers for Disease Control and Prevention, 2017). There are numerous benefits to participation in youth sports. At least one hour of moderate-to-vigorous physical activity is recommended daily for children, and sports can help to achieve this goal. Physical activity in children has several benefits like cardiorespiratory and muscular fitness, healthy weight status, improved bone health, and increased cognitive function. Participation in youth sports also has psychosocial benefits like higher self-esteem, higher confidence, improved life skills, and a reduced risk of depression and anxiety (U.S. Department of Health and Human Services, 2018).

Sports-Related Concussion

A potential downside to youth sports is the possibility of experiencing a head injury. Sport-related concussion (SRC) is a traumatic brain injury (TBI) induced by mechanical forces. It can be caused from a direct impact to the head or from a force elsewhere on the body that is transmitted to the head. TBI is a leading cause of disability in the United States, and the pediatric and adolescent population accounts for approximately 65% of annual concussion diagnoses (Yue et al., 2016).

Although there has perhaps been less SRC-related research in ice hockey as compared to other contact sports such as football and boxing, ice hockey has one of the highest incidence rates of concussion in youth sports, being as high as 21.5 concussions per 1000 athlete exposures when using physician standardized direct observation, diagnosis, and treatment (Echlin et al., 2010). Starting in the 2011-2012 season, USA Hockey raised the body checking age from the PeeWee (ages 11 to 12 years) to Bantam (ages 13 to 14 years) division. There is evidence that increasing the age of body checking in youth hockey overall decreases the incidence of concussion (Ingram et al., 2019).

The signs and symptoms of a SRC can often be attributed to a functional disturbance as neuroimaging typically does not show structural injury (McCrory et al., 2017). The clinical diagnosis of a SRC can be difficult to reach because some youth and adolescent athletes with a SRC may experience no symptoms, and it has been reported that over half of SRC cases may go undiagnosed (Dhawan et al., 2017). Symptoms of a concussion can present in a variety of ways and are nonspecific. Currently, there is not a uniform, definitive way of diagnosing a concussion. A physician may use a combination of patient history, a concussion symptom survey, neurological signs, and brain imaging to reach a diagnosis (McCrory et al., 2017). In addition, SRCs can have chronic implications that affect the athletes well beyond the season of play, as emerging evidence links past history of SRC to changes in mood profiles that include ruminative thinking, hypervigilance, increased anxiety, panic, depression, apathy and sleep disruption (Sandel et al., 2017). Therefore, an improvement in the sensitivity and specificity of SRC

screening protocols could also aid in the development of prediction models that better identify kids that are at risk for clinically diagnosable SRC and chronic health issues.

Some commonly used symptom assessments are the Acute Concussion Evaluation, the Rivermead Post-Concussion Symptoms Questionnaire, and the Post-Concussion Symptom Score. They include a list of up to 25 symptoms, like headache, dizziness, balance problems, fatigue, confusion, difficulty concentrating, blurred vision, and light or noise sensitivity. A Likert scale is used to rank the degree to which a person agrees or disagrees with a statement on a five- or seven-point ordinal scale and is typically utilized in these assessments (Sullivan and Artino, 2013). They may ask if physical or mental activity exacerbate a given symptom. Some assessments may ask about the severity or frequency of each symptom. A total symptom score is typically computed by summating all the responses (Dziemianowicz et al., 2012).

The Rivermead Post-Concussion Symptoms Questionnaire does not have a diagnostic cut off value, but is used in tracking symptom severity over time. It has good reliability both at the one week and the six month period following a SRC (King et. al., 1995). Symptom surveys are dependent on self-report by the athlete, which is a major issue as many athletes will downplay their symptoms to get back onto the field. These surveys have also evolved over time, meaning they were not methodically developed nor scrutinized by the scientific community (Alla et. al., 2009).

A commonly used assessment of cognitive function is the CogState Brief Battery. It consists of a Detection Task, an Identification Task, a One Card Learning Task, and a One-Back Task to measure Processing Speed, Attention, Visual Learning, and Working

Memory. It can be performed in under 10 minutes and uses simple stimuli, along with simple tasks. When associated with neuropsychological tests it was determined to be valid in all four areas (Maruff et al., 2009).

CogState scores have been compared to those with and without vision symptoms who have suffered a concussion, using the Convergence Insufficiency Symptoms Survey (CISS). CISS is a questionnaire used in the detection and management of the convergence insufficiency binocular vision disorder (Bade et al., 2013). Common visual symptoms linked to concussion are photosensitivity, blurry vision, eyestrain, difficulty reading, and poor attention to near tasks, and many of these symptoms are assessed on the CISS. A score of 16 or higher is suggestive of convergence insufficiency in the pediatric population (Borsting et. al., 2003).

The presence of binocular vision-related symptoms, as measured with the CISS, with concussion is associated with significantly lower scores on the Attention, Learning, and Working Memory of CogState. A score of 13 or higher on the CISS is suggestive of a binocular vision disorder in the pediatric concussion population (Peiffer et al. 2020). Nearly 80% of individuals assessed within 30 days of suffering from a SRC exhibited a significant binocular vision dysfunction. This indicates there is a high prevalence of binocular vision impairment in those with a recent concussion (Peiffer et al., 2020).

Pupillary Light Reflex

The measurement of pupil size and reactivity to light, also known as pupillometry, may provide an objective, noninvasive, and rapid way to aid in the diagnosis of a

concussion and the determination of when an athlete should return to play. A previous study of pupillometry looked at rates and amounts of dilation in concussed and healthy adolescent athletes. It revealed that athletes suffering from an asymptomatic high acceleration head impact had a significant decreased percent change in pupil diameter, decreased pupil dilation velocity, and decreased maximum constriction velocity post-impact (Joseph et al., 2019). Carrick et al. (2021) revealed that in addition to having a history of concussion, concussion symptoms and gender significantly impact the PLR metrics when compared to healthy individuals. Another study suggested that pupillary light reflex (PLR) metrics are enhanced in acute adolescent concussion, but exercise diminished PLR metrics in healthy controls. Girls with concussion had a significantly longer time to 75% pupillary redilation than healthy controls (Master et al., 2020).

Intrinsically photosensitive retinal ganglion cells (ipRGCs) play a major role in regulating the PLR and circadian rhythm entrainments, and more information on these neurons is provided in the next section. Traditionally, it was believed that the rods and cones were the photoreceptors that solely initiated this reflex, but rodless and coneless mice still exhibited a PLR, although with an increased constriction latency and higher threshold irradiance compared to the wild type mice (Lucas et al., 2001). ipRGCs were later shown to not only contribute to the constriction of the pupil at high irradiances, but also to the maintenance of the diameter of the pupil under steady photopic conditions (Lucas et al, 2003). Cones contribute minimally to the pupil diameter under photopic low and high irradiances after 10 seconds of steady light (McDougal & Gamlin, 2010).

After receiving light information, the retinal ganglion axons travel via the optic nerve, the nasal fibers decussate at the optic chiasm, and then the axons travel via the optic tract. The majority of the axons in the optic tract synapse in the ipsilateral lateral geniculate nucleus (LGN) of the thalamus, but some of them synapse in the ipsilateral olivary pretectal nucleus of the midbrain. The pretectal nucleus projects to and synapses bilaterally in the Edinger-Westphal nuclei. The Edinger-Westphal preganglionic parasympathetic axons efferently project along the oculomotor nerve and synapse in the ciliary ganglia. The postganglionic axons project along the short ciliary nerves until they innervate the iris sphincter muscle to cause pupillary constriction (Yoo & Mihaila, 2020).

Since ipRGCs play a large role in mediating the PLR, their contribution can be measured by assessing pupil responses to specific light stimuli. They are most sensitive to blue light and can fire for several seconds after the onset of light (Berson et al., 2002). By comparing pupil responses to red and blue light, the functional properties of ipRGC responses can be indirectly assessed.

Being able to measure the contribution of ipRGCs via pupil assessments has many potential clinical applications. Exacerbation of migraine symptoms in light conditions exhibited by migraine sufferers has been attributed to a population of ipRGCs that project to pain centers located in the posterior thalamus, which are innervated by the trigeminovascular pathway. Light stimuli can be linked to the sensation of pain (Noseda et al., 2010). Similarly, photophobia is a common symptom in traumatic brain injury (TBI), and a neural mechanism involving ipRGC altered function is suspected.

By using alternating blue and red light, certain functional properties of ipRGCs can be indirectly assessed. Their amount of contribution can be assessed by looking at the delay of pupil re-dilation after exposure to blue light, relative to the re-dilation after exposure to red light. Although a recent study by Yuhas et al. (2017) did not find an ipRGC hypersensitivity to light in TBI patients, there was greater pupil response variability, which may suggest a more heterogeneous ipRGC function in TBI patients.

A study explored the repeatability of pupil responses to red and blue light in healthy subjects. While it found no significant changes in the repeated measures, limitations include that it only assessed repeatability within the same day, and only used continuous monochromatic light, either red or blue, in each eye (Herbst et al., 2011). Repeatability can be better longitudinally assessed by collecting pupil responses separate times over the course of several months.

Anatomy and Physiology of ipRGCs

Photoreceptors within the mammalian retina capture light energy and transform it into electrical signals. Rods and cones were the only known photoreceptors up until the 21st century when significant evidence began to support the presence of a photoreceptor in the inner retina. Early in the 1990s, mice with degenerated outer retinal layers showed circadian entrainment properties that were no different than mice with intact retinas, even though no photoreceptor outer segments were present (Foster et al., 1991). This suggested that there may be a different photoreceptor collecting nonvisual light information. A pivotal study by Berson et. al. (2002) utilized retrograde fluorescent tags

from the suprachiasmatic nucleus (SCN) to label RGCs that projected to this brain location for the master circadian clock. After pharmacologically blocking rod and cone-driven activity, light-evoked light responses persisted in the SCN-projecting RGCs, leading to the identification of a new photoreceptor: the intrinsically photosensitive retinal ganglion cell (Berson et al., 2002).

Around the same time, a novel opsin of a photopigment, named melanopsin, was being identified in the mammalian retina (Provencio et al., 1998). Melanopsin-containing retinal ganglion cells were found to project via the retinohypothalamic tract (RHT) to the SCN (Hannibal et al., 2002). Hattar et al. (2002) conclusively demonstrated that melanopsin was the photopigment contained in ipRGCs.

Melanopsin is expressed on the dendrites, cell body, and axons of ipRGCs, up until the optic disc (Hattar et al., 2002). In primates, ipRGCs exhibit the largest dendritic tree of any retinal ganglion cell but account for only 0.2% of the ganglion cell population (Dacey et al., 2005). In general, ipRGCs show a transient peak in the spike rate that decreases to a plateau in response to light. The spike response is sluggish, with a delay in response to a dim light and it lasts for many seconds, even minutes, after an intense stimulus ends. Spiking responses in ipRGCs can be driven by direct light stimulation of the melanopsin photopigment (intrinsic responses) or by synaptic input from bipolar cells that receive rod and cone-driven signals (extrinsic responses). An example of the intrinsic response can be seen in Figure 1. The peak spectral sensitivity for the intrinsic response is around 484 nm, which corresponds to the wavelength of blue light (Berson et al., 2002).

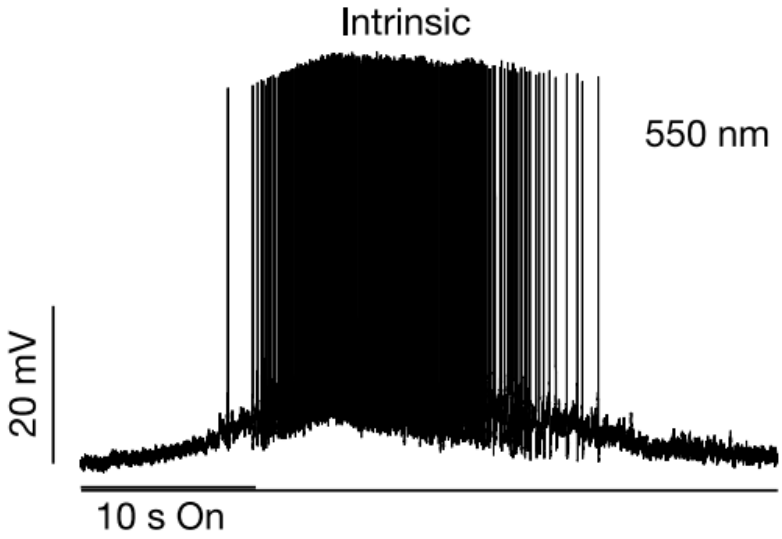


Figure 1: ipRGC Intrinsic Light Response

A delayed and prolonged intrinsic response to light is observed with a 10 second light stimulus (Dacey et. al., 2005).

There are several subtypes of ipRGCs, which are categorized based on their morphology, photosensitivity, and other properties. The M1 cells were the first discovered subclass of ipRGCs. The M1 subtype is classified based on its high expression of melanopsin, stratification of dendrites in the OFF sublamina of the inner plexiform layer, and innervation of brain areas involved in subconscious, nonvisual forming behaviors (Hattar et al., 2006). They also have the largest and fastest intrinsic photoresponses, but smallest soma (Hattar et al., 2009).

The M2 subtype is characterized by its stratification of dendrites in the ON sublamina of the inner plexiform layer, its medium-sized soma, and its smaller and slower response to intrinsic light relative to the M1 subtype (Hattar et al., 2009). Although they are less photosensitive than M1 cells, the M2 subtype can fire action potentials at higher frequencies. The M2 cells are as numerous as M1 cells, but the dendritic fields of M2 cells are more branched and larger than those of the M1 cells. There are significant overlaps in the receptive fields of both M1 and M2 subtypes, which indicates distinct functionality in how the cells signal light to the brain (Schmidt & Kofuji, 2009).

The M3 subtype of cells, also called the bistratified subtype, resemble ON alpha cells, have extremely large cell bodies, radiate thick dendritic branches in the OFF and ON sublamina of the inner plexiform layer, and have the weakest and slowest photoresponses relative to the M1 and M2 cells (Hattar et al., 2009). They account for less than 10% of all ipRGCs. These may not constitute a true cell type based on their low

spatial density, disabling them from fully tiling the retina (Berson et al., 2010). Their synaptic inputs and sensitivity to light have yet to be determined.

Another subtype of ipRGCs is the M4 cell. The large and radiating dendritic branches stratify in the ON sublamina of the inner plexiform layer and exhibit a large soma (Estevez et sl., 2012). Although they closely resemble M2 cells, M4 cells exhibit a weaker melanopsin immunoreactivity (Berson et al., 2010) and a specific transgenic reporter of melanopsin expression for M2 cells does not label M4 cells (Schmidt et al., 2008). The M4 cell's intrinsic response threshold is slightly higher than the M2 subtype. Although most ipRGCs are more traditionally believed to be involved in nonvisual roles, the M4 cells retinotopically project to the dorsal lateral geniculate nucleus (dLGN) and are suspected to play a role in enhancement of local contrast (Estevez et sl., 2012).

The last major subtype of ipRGCs is the M5 cell. The dendrites are small and bushy, which greatly contrast the broad, radiating dendrites of the M4 subtype (Estevez et al., 2012). Functionally in mice, it exhibits chromatic opponency with ultraviolet being excitatory and green light being inhibitory. Like the M4 subtype, the M5 cell is suspected to play a visual role. It projects axons to the dLGN that may play a role in color perception (Stabio et al., 2018).

A novel M6 subtype has recently been discovered. This bistratified M6 cell has the smallest, yet most abundantly branched dendritic field out of all the known ipRGCs. The melanopsin expression of M6 cells is low, along with having a weak light response dependent on melanopsin. Similar to the M4 and M5 subtypes, the M6 cells project to the dLGN and are suspected to play a role in pattern vision (Quattrochi et al., 2019).

The M1 subtype has been further categorized based on the expression of the transcription factor Brn3b. The Brn3b-negative M1 ipRGCs send projections to the SCN, which involves the entrainment of circadian rhythms. The Brn3b-positive M1 ipRGCs send projections to the olivary pretectal nucleus, which is involved in the pupillary light reflex. There are also sparse, Brn3b-positive ipRGCs that project to the thalamus, midbrain, and hypothalamus. The two M1 subtypes do not project to the same brain targets, indicating they are involved in specific light-driven activity (Li & Schmidt, 2018).

Purpose

The purpose of this study was to perform longitudinal assessments of pupil function, visual symptoms, and cognitive function in a cohort of 13 year-old hockey players during their first season in which body checking is legal in the sport. Compared to other youth sports, minor hockey has a relatively long season with game schedules often beginning in early September and ending in early March. The length of the season suggests that hockey might be a particularly suitable sport for longitudinal studies that assess changes in neural structure and function across an entire season.

The evolving and continued understanding of the role of ipRGCs in both visual and nonvisual modalities is crucial for clinical applications and improvements in debilitating areas such as photophobia and TBI. Better understanding of a concussion's impact on the PLR may lead to the development of an objective screening test for a more accurate and timelier SRC diagnosis than current screening procedures. Testing paradigms that assess subtle changes in neural impairment and recovery in cases of

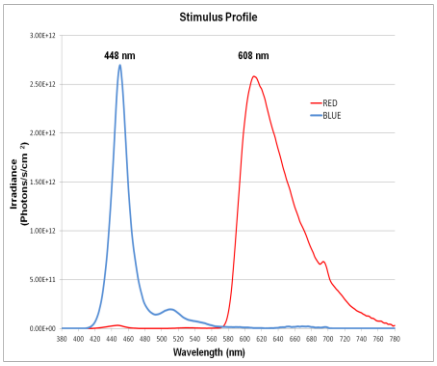
subclinical concussion could have considerable impact on youth athletics in terms of policy development, equipment design and rule change implementation that aim to make sport participation safer.

Chapter 2. Methods

Participants included 18 male hockey players, each 13 years of age, who were enrolled in a repeated measures cohort study at the beginning of the 2018-2019 youth hockey season in central Ohio. All participants had played hockey for at least 6 years, provided informed consent in agreeing to take part in study, and reported no recent (previous 2 months) concussions or hospitalizations due to sickness or injury. All testing, including pupil testing, were performed at a local hockey arena either before or after the player's evening practice. The symptom surveys and cognitive function testing were performed in a secluded room to ensure participant privacy and the pupil testing was performed in a dark, windowless room.

The RAPDx pupillometer (Konan Medical) was used to record pupil responses to various light stimuli. A 26.5° circular field of light stimuli was presented to both eyes, beginning with five seconds of red light, followed by five seconds of dark, then five seconds of blue light, followed by five seconds of dark. This totaled six pulses of alternating red and blue light applied at 0.1Hz for two minutes. The peak sensitivity for the red (long wavelength) light was at 608nm and the peak sensitivity for the blue (short wavelength) light was 448nm. Irradiance for each light was near 2.50×10^{12} photons/s/cm^2, close to the ipRGC threshold. Refer to Figure 2 for additional light stimulus information.

(a)



(b)

(c)

Figure 2: Light Stimulus

(a)RAPDx Pupillometer Device. (b) Profile of blue (448nm) and red (608nm) light stimulus. Irradiance near 2.50 x 10^{12} photons/s/cm^2. (c): Illustration of one minute of light stimulus, half of the total time of each session.

Artifacts due to blinking were removed by the RAPDx software or removed manually during data analysis when an abrupt change in pupil size was detected. To more easily handle the data, the data was binned into 0.25 second intervals and the diameter of the pupil was normalized for each eye, relative to the smallest pupil diameter (1.0 = 100% constriction) and largest pupil diameter (0 = 0% constriction). The pupil diameters were averaged between the two eyes at each ten second time interval (beginning with the onset of colored light). The average pupil constriction during the first red versus first blue pulses, and the change in constriction between the first and last pulses of red light, an adaptive response termed photopotentiation, were calculated.

Data collection occurred during three sessions, each separated by approximately two months, conducted at the beginning, middle and end of the hockey season. To assess the repeatability of photopotentiation across the three visits, Bland-Altman analyses were performed. This graphical analysis plots the difference between the two measurements vs. the mean of the two measurements to reveal agreement or bias between the two sets of data.

At each visit, participant's symptoms were quantified using the Convergence Insufficiency Symptoms Survey (CISS) and the Rivermead Post-Concussion Symptoms Questionnaire (RPCSQ). Both the CISS and RPCSQ were verbally administered. The CISS utilizes a Likert scale to report the frequency of fifteen different symptoms that are commonly associated with the binocular vision disorder convergence insufficiency (Bade et al., 2013). The RPCSQ utilizes a Likert scale to report the frequency of the most common symptoms experienced after a concussion, and to determine overall well-being

(King et al., 1995). The RPCSQ was modified for participants to report symptoms over the last 24 hours, not compared to before a head injury as indicated in the original questionnaire. Reference Figures 3 and 4 for examples of each survey.

Clinician instructions: Read the following subject instructions and then each item exactly as written. If subject responds with "yes" - please qualify with frequency choices. **Do not give examples.**
Subject instructions: Please answer the following questions about how your eyes feel when reading or doing close work. First think about whether or not you have the symptom. If you do, please tell me whether the problem occurs: Infrequently (not very often), Sometimes, Fairly Often, or Always.

		Never	Infrequently	Sometimes	Fairly often	Always
1.	Do your eyes feel tired when reading or doing close work?					
2.	Do your eyes feel uncomfortable when reading or doing close work?					
3.	Do you have headaches when reading or doing close work?					
4.	Do you feel sleepy when reading or doing close work?					
5.	Do you lose concentration when reading or doing close work?					
6.	Do you have trouble remembering what you have read?					
7.	Do you have double vision when reading or doing close work?					
8.	Do you see the words move, jump, swim or appear to float on the page when reading or doing close work?					
9.	Do you feel like you read slowly?					
10.	Do your eyes ever hurt when reading or doing close work?					
11.	Do your eyes ever feel sore when reading or doing close work?					
12.	Do you feel a "pulling" feeling around your eyes when reading or doing close work?					
13.	Do you notice the words blurring or coming in and out of focus when reading or doing close work?					
14.	Do you lose your place while reading or doing close work?					
15.	Do you have to re-read the same line of words when reading?					
To obtain score, total the number of "X"s in each column						
Multiply by the column value		x0	x1	x2	x3	x4
Sum 5 values						

SCORE: _____

Figure 3: The Convergence Insufficiency Symptoms Survey

The Rivermead Post-Concussion Symptoms Questionnaire*

After a head injury or accident some people experience symptoms which can cause worry or nuisance. We would like to know if you now suffer from any of the symptoms given below. As many of these symptoms occur normally, we would like you to compare yourself now with before the accident. For each one, please circle the number closest to your answer.

0 = Not experienced at all
1 = No more of a problem
2 = A mild problem
3 = A moderate problem
4 = A severe problem

Compared with before the accident, do you now (i.e., over the last 24 hours) suffer from:

Headaches	0	1	2	3	4
Feelings of Dizziness	0	1	2	3	4
Nausea and/or Vomiting	0	1	2	3	4
Noise Sensitivity, easily upset by loud noise	0	1	2	3	4
Sleep Disturbance	0	1	2	3	4
Fatigue, tiring more easily	0	1	2	3	4
Being Irritable, easily angered	0	1	2	3	4
Feeling Depressed or Tearful	0	1	2	3	4
Feeling Frustrated or Impatient	0	1	2	3	4
Forgetfulness, poor memory	0	1	2	3	4
Poor Concentration	0	1	2	3	4
Taking Longer to Think	0	1	2	3	4
Blurred Vision	0	1	2	3	4
Light Sensitivity, Easily upset by bright light	0	1	2	3	4
Double Vision	0	1	2	3	4
Restlessness	0	1	2	3	4

Are you experiencing any other difficulties?

1. _____	0	1	2	3	4
2. _____	0	1	2	3	4

*King, N., Crawford, S., Wenden, F., Moss, N., and Wade, D. (1995) J. Neurology 242: 587-592

06/23/08

Figure 4: The Rivermead Post-Concussion Symptoms Questionnaire

This original survey was modified for participants to report symptoms over the past 24 hours, not compared to before a head injury.

The cognitive function was tested using the CogState Brief Battery Computerized Concussion Assessment. It includes four tasks that measure Attention, Processing Speed, Memory, and Working Memory speed. The assessment was performed on a laptop computer. Participants were given instructions on how to complete each task, then completed practice trails to demonstrate understanding of the rules. Each task utilized playing cards as stimuli. Participants pressed the d and k keys of the laptop keyboard to signal yes or no responses for each task. Yes responses were selected using the dominant hand. Participants were instructed to answer as quickly and accurately as possible.

The Detection Task measures Processing Speed using a simple reaction time paradigm. The program asks questions such as "Has the card turned face up?" It continues for two minutes, or until 25 correct responses are recorded. The speed of correct responses is the primary outcome measured. The Identification Task measures Attention using a choice reaction time paradigm. The program asks questions such as "Is the face-up card red?" for two minutes, or until 35 correct responses are recorded. The speed of correct responses is the primary outcome measured. The One Card Learning Task measures Visual Learning using a pattern separation paradigm. The program asks questions such as "Have you seen this card before in this task?" while individuals are presented with six cards to learn, repeated randomly throughout the task, with eight distractor cards. It continues for three minutes, or until 42 responses are recorded. The percent of correct responses is the primary outcome measured. The One-Back Task measures Working Memory using an n-back paradigm. The program asks questions such as "Is this card the same as that on the immediately previous trial?" for three minutes, or

until 42 trials have been completed. The percent of correct responses is the primary outcome measured (Maruff, et al., 2009).

Chapter 3. Results

With the alternating red/blue flashing light sequence utilized in this protocol, there was a greater pupil constriction at the end of the test compared to the start, an adaptive property of the pupil response that has been termed photopotentiation. Individuals with high photopotentiation tended to show a more robust initial pupil constriction to blue light compared to red light. With successive pulses, the pupil exhibits greater constriction to red light, becoming more similar in response to the blue light. An example of high photopotentiation can be found in Figure 5. Individuals exhibiting low or no photopotentiation tended to show the same or similar pupil constriction to red light and blue light pulses, both initially and with successive pulses. The lack of difference between the responses to the initial red and blue pulses indicate a smaller contribution of melanopsin to the pupil response in these individuals. An example of low photopotentiation can be found in Figure 6.

The difference in initial response to the red and blue light pulse (a marker of melanopsin-mediated contribution to the PLR) compared to the difference in response to the first and last red pulse (the adaptive photopotentiation response) was further explored. There is a significant correlation ($R = 0.61$; $P < 0.001$) between these two values, which is illustrated in Figure 7. The difference in initial response to the red and blue light pulse is weakly correlated with the change from the first to the last blue pulse of light, which is illustrated in Figure 8 ($R = 0.33$).

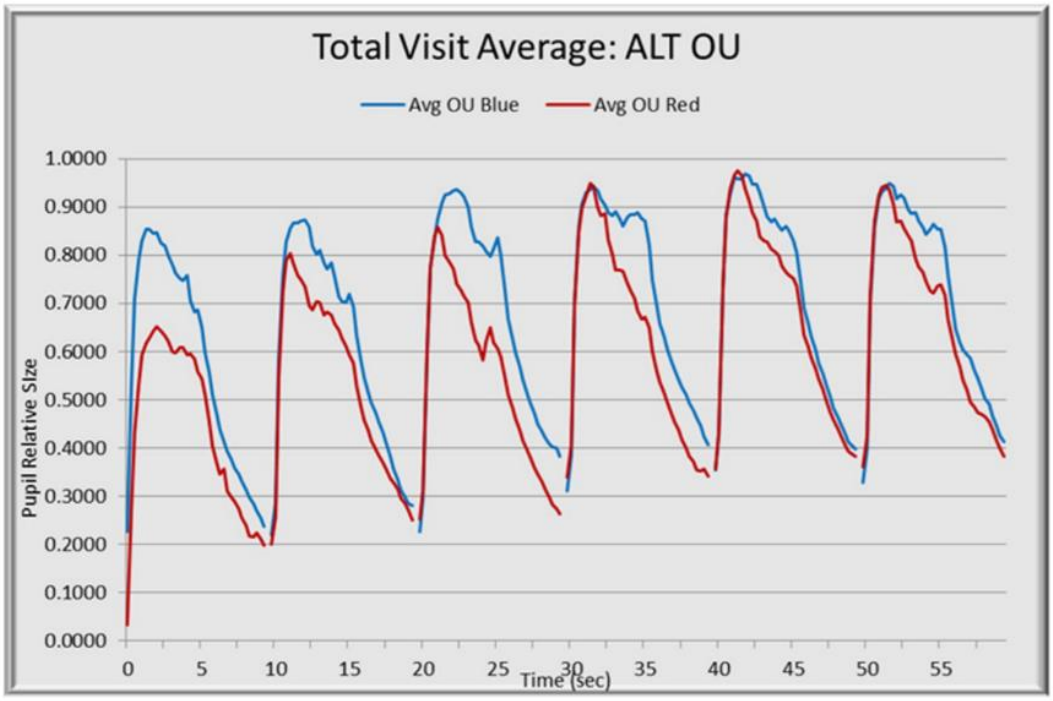


Figure 5: High Photopotentiation

This individual exhibits a more robust initial pupil constriction to blue light compared to red light. Over time, the pupil constriction in response to the red light becomes more similar to the constriction in response to the blue light.

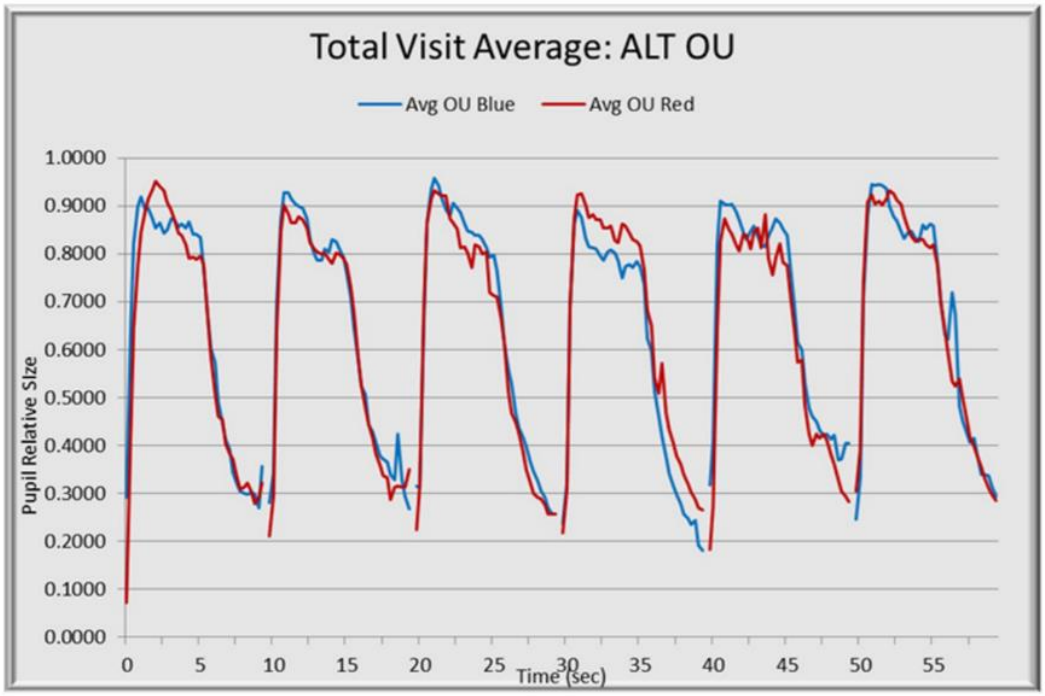


Figure 6: Low Photopotentiation

This individual exhibits little or no difference in initial pupil constriction to red light compared to blue light. This response remains similar over successive pulses.

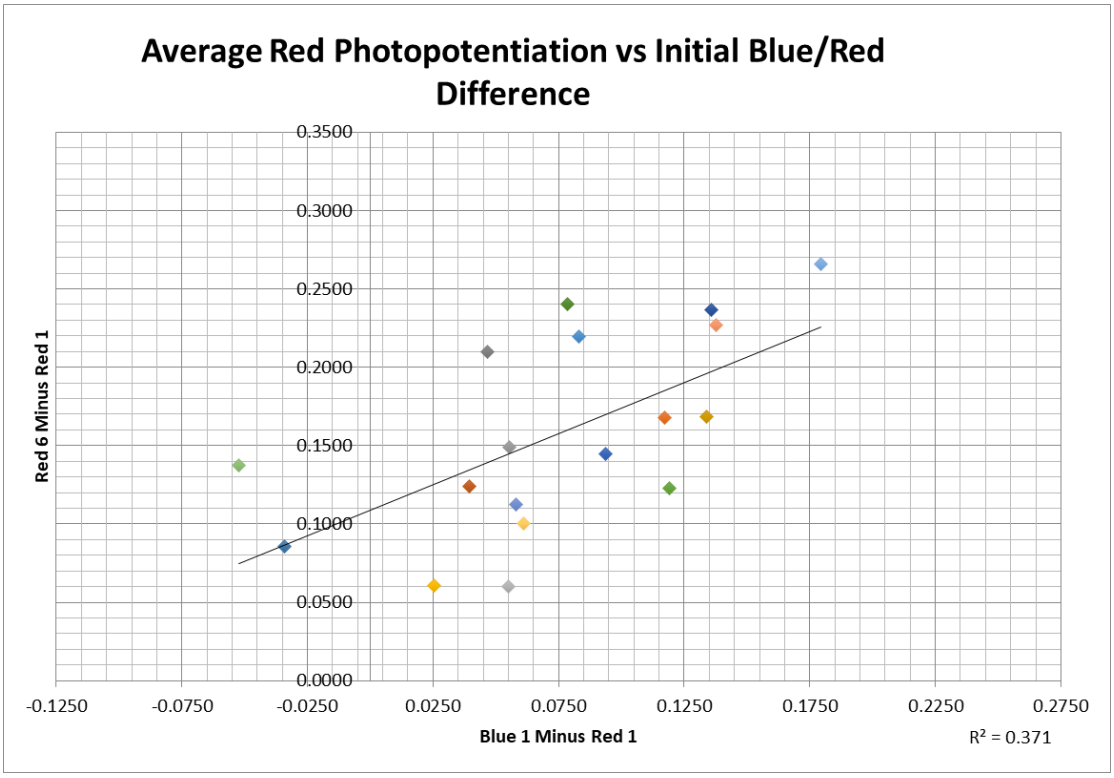


Figure 7: Average Photopotentiation vs. Initial Blue/Red Difference

The change from the first red to the last red pulse, an adaptive response termed photopotentiation, is highly correlated with the change in initial red and blue light pulses, the contribution of melanopsin ($R = 0.61$; $P < 0.001$).

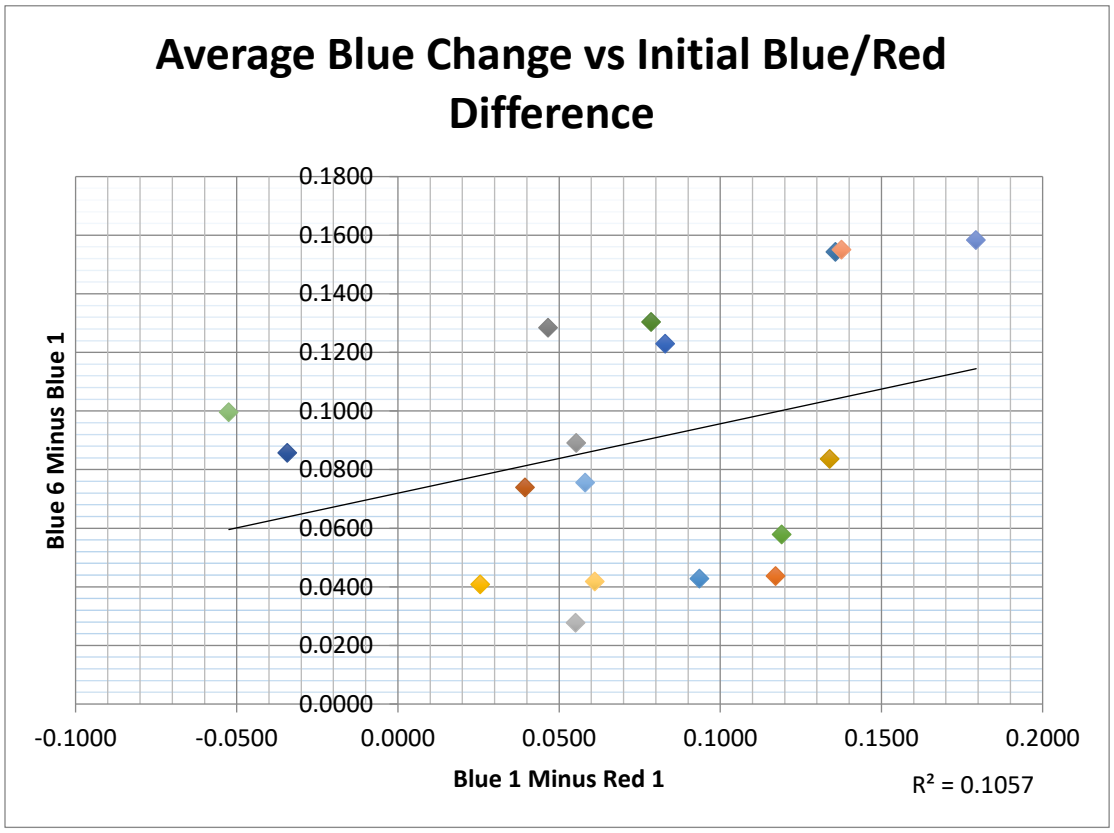


Figure 8: Average Blue Change vs. Initial Blue/Red Difference

The change from the first to the last blue pulse of light is weakly correlated with the change in initial red and blue light pulses, the contribution of melanopsin (R = 0.33).

The data collection occurred approximately two months apart during the hockey season. Bland-Altman analysis was performed to better assess the repeatability of the difference in pupil responses between the initial blue and red pulse between each of the three visits. As illustrated in Figure 9, there was no significant change in the difference between visit one and two ($P = 0.16$), visit one and three ($P = 0.81$), and visit two and three ($P = 0.18$).

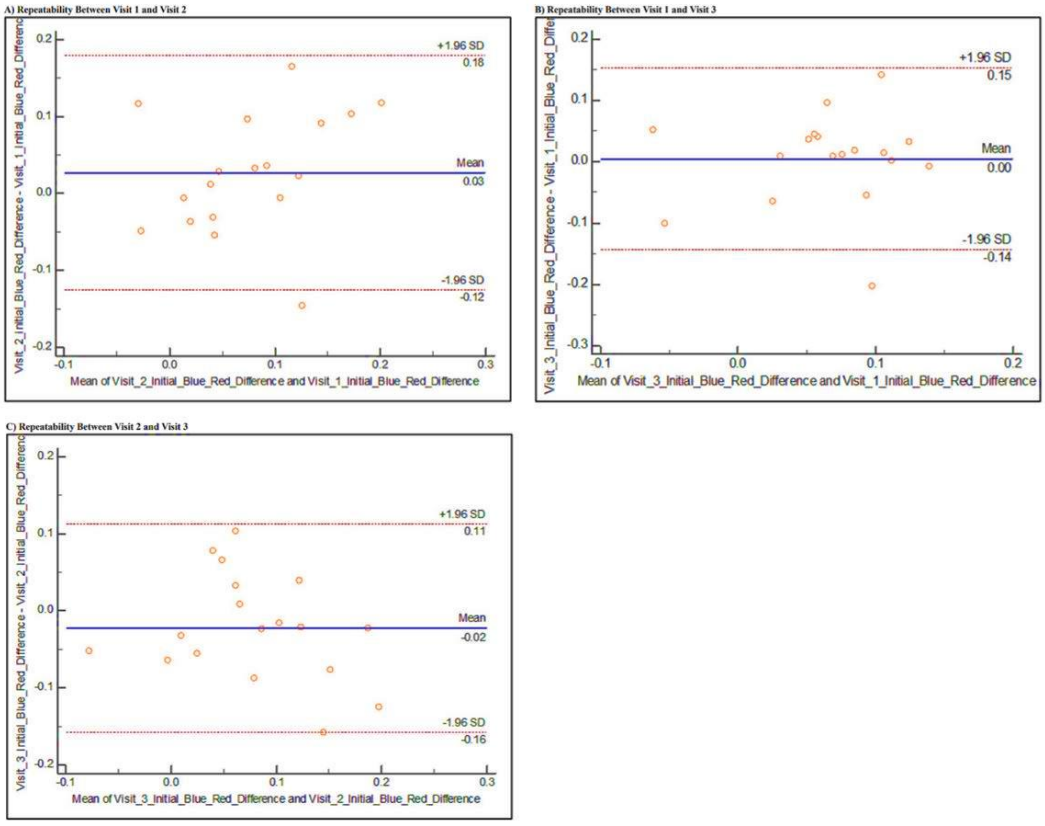


Figure 9: Bland-Altman Analysis of Repeatability Between Visits

There was no significant change in the difference between visit one and two (P = 0.16), visit one and three (P = 0.81), and visit two and three (P = 0.18), indicating repeatability of the responses across visits.

The CogState scores for the Learning, Attention, Processing Speed, and Working Memory Speed Tasks were compared across the season. The CogState scores are standardized to a score that is near 100. There was a significant increase in scores for the Learning Task from visit one to visit two and from visit one to visit three ($P < 0.01$, 1-way RM ANOVA). There was a significant, but small decrease in scores for the Processing Speed Task from visit one to visit two ($P = 0.03$). There were no significant score differences at subsequent visits from baseline for the Attention and Working Memory Speed Tasks ($P > 0.05$). The CogState scores across the season are illustrated in Figure 10.

Bland-Altman analysis was performed to better assess the repeatability of the CogState scores across the visits. Between visit one and two, the bias score for the Learning Task was 7.91 and the Processing Speed Task bias score was -5.47. There were insignificant bias scores of 0.41 and 0.68 for Attention and Working Memory Speed, respectively. Between visits one and three, the bias scores were -3.20, 0.24, 13.14 and 1.54 for Processing Speed, Attention, Learning and Working Memory. The repeatability measures are seen in Figures 11 and 12.

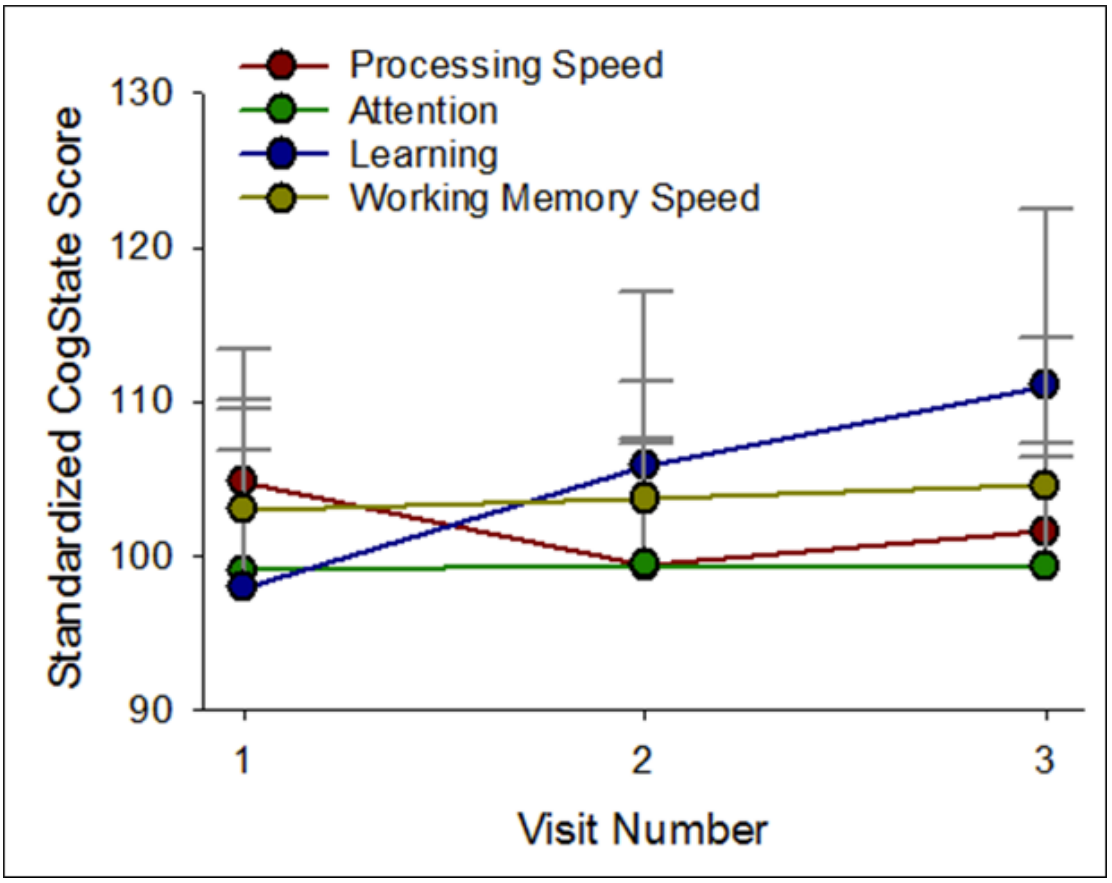


Figure 10: Cogstate Scores Across the Season

The Learning Task exhibited a significant increase in scores from visit one to visit two and from visit one to visit three ($P < 0.01$, 1-way RM ANOVA). The Processing Speed Task exhibited a significant, but small decrease in scores from visit one to visit two ($P = 0.03$). There were no significant differences in scores across subsequent visits for the Attention and Working Memory Speed Tasks ($P > 0.05$).

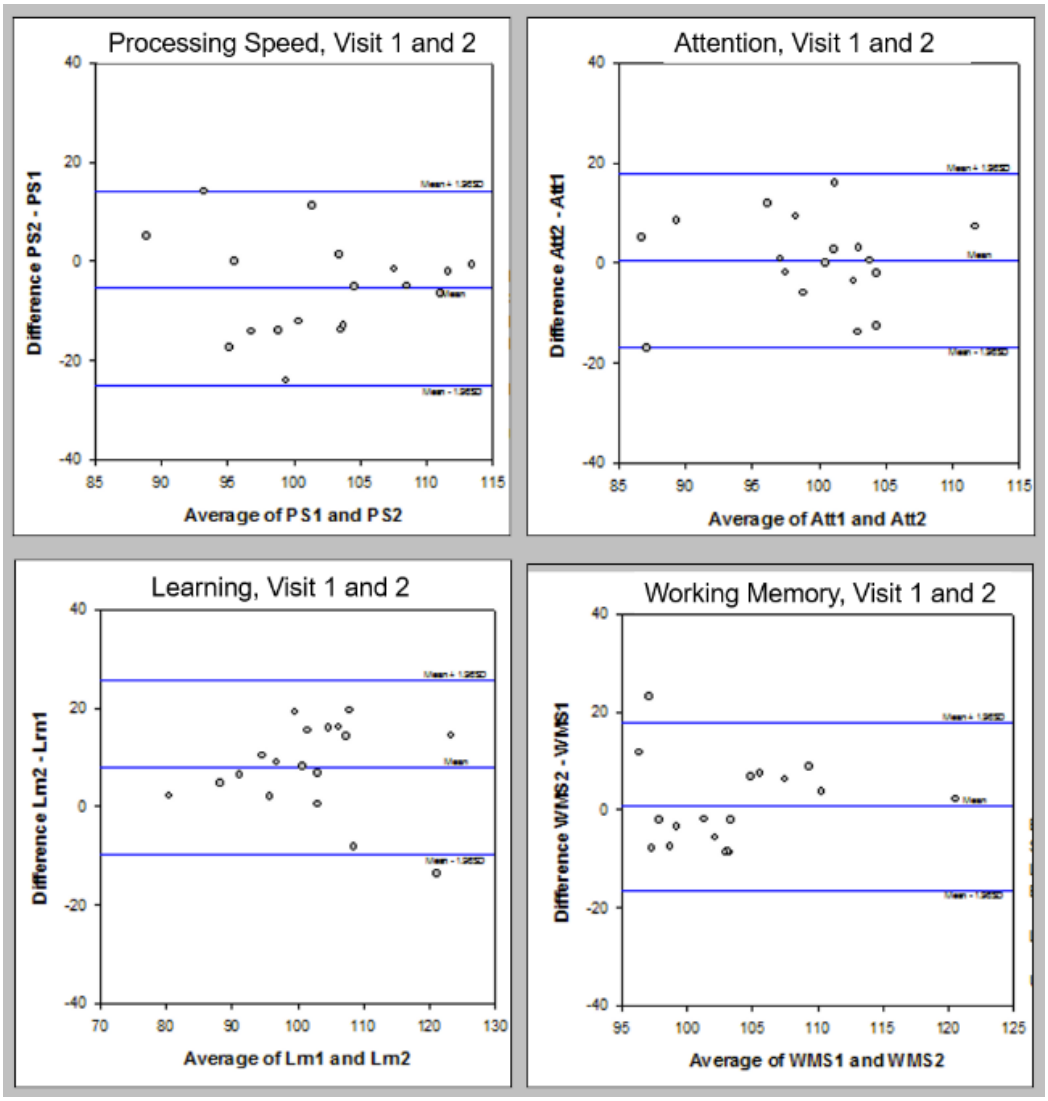


Figure 11: CogState Bland-Altman Analysis between Visit One and Two

The bias score for the Learning Task was 7.91 and the Processing Speed Task bias score was -5.47 between visit one and visit two. The Attention and Working Memory Speed Tasks exhibited insignificant bias scores of 0.41 and 0.68, respectively.

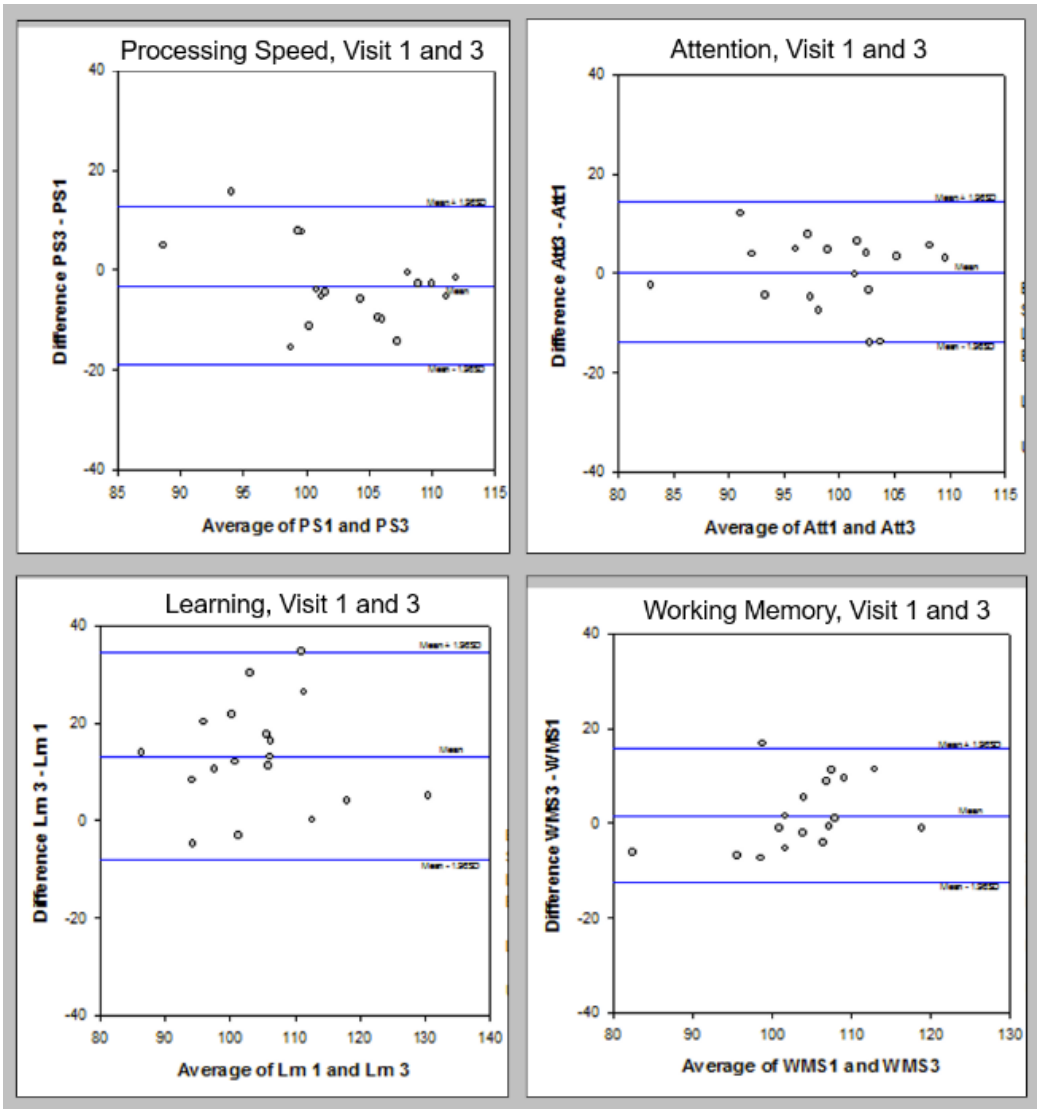


Figure 12: CogState Bland-Altman Analysis between Visit One and Three

The bias scores were -3.20, 0.24, 13.14 and 1.54 between visit one and visit three for Processing Speed, Attention, Learning and Working Memory Tasks, respectively.

Scores from the CISS and the Rivermead (RPCSQ) were further assessed. The CISS has vision-specific questions, which the RPCSQ has questions tailored to the overall well-being of a person. There was a strong correlation between the CISS and RPCSQ scores ($R^2 = 0.70$, $P < 0.001$). This is illustrated in Figure 13.

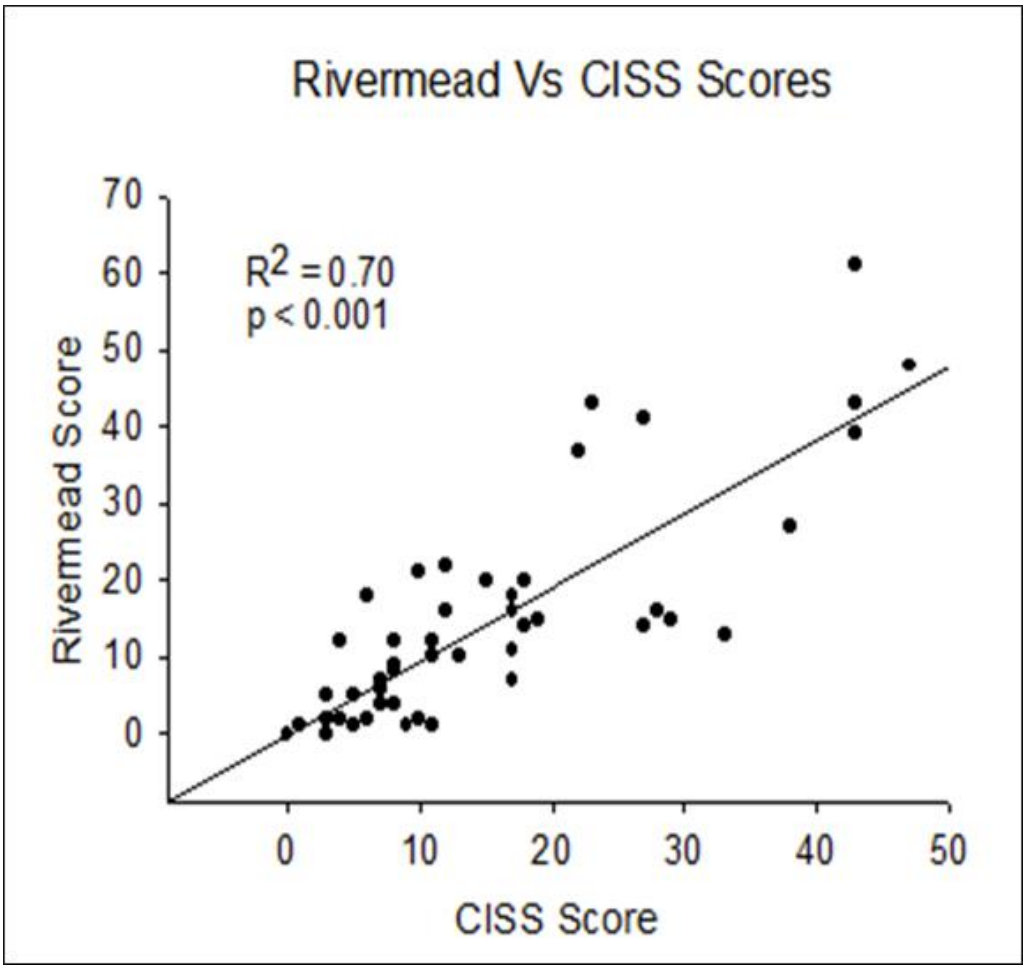


Figure 13: Rivermead and CISS Score Correlation

The CISS and RPCSQ scores were highly correlated ($R^2 = 0.70$, $P < 0.001$).

Chapter 4. Discussion

Traditionally, it was believed that rods and cones were primarily involved in the pupillary light reflex, but this reflex is still present in rodless and coneless mice (Lucas et al., 2001). Another type of cell in the retina, the intrinsically photosensitive retinal ganglion cell, is primarily responsible for maintaining the PLR. The ipRGCs express melanopsin (Hattar et al., 2002) and are most sensitive to blue light (Berson et al., 2002). The contribution of this melanopsin-driven response can be further explored by comparing the pupil responses to alternating red and blue flashes of light. There is sustained pupil constriction following light offset that is mediated by ipRGCs at irradiances above the threshold of melanopsin (Gamlin et al., 2007). Rods significantly contribute to the pupil response to steady light at irradiances below the threshold of melanopsin (McDougal and Gamlin, 2009). The blue flashes of light are above this threshold, therefore this pupil response is believed to be mediated by ipRGCs. The red flashes of light are below this threshold, therefore this pupil response is believed to be mediated by rods and cones. The gradual increase in pupil constriction in response to red pulses compared to blue pulses over time is termed the adaptive process of photopotentiation. This effect may be driven by the signaling of melanopsin in ipRGCs, as evidenced in rodents (Zhu et al., 2007).

Individuals who show a greater pupil response to blue light relative to red light of similar irradiance may have a greater contribution of melanopsin-driven response to the neural circuitry that mediates the pupillary light reflex. Those exhibiting greater blue light initial responses showed greater photopotentiation. Further exploration of the relationship between the contribution of melanopsin and the amount of photopotentiation indicates there is a significant correlation between these two values.

It is important that both the melanopsin initial response and the adaptive photopotentiation were tested. They correlate in the healthy hockey player population, but this could be different in athletes with concussion. An athlete with a SRC may exhibit deficient photopotentiation with a normal initial melanopsin contribution. It is also important that the light was near the threshold of melanopsin, as subjects exhibited responses above or below the threshold. This may have contributed to the strong correlation between the initial melanopsin response and photopotentiation. If the light was brighter, a ceiling effect is expected to create a cluster of the data due to all subjects exhibiting photopotentiation. This ceiling effect is exhibited when the blue light is above the threshold of melanopsin, which is supported by the weak correlation between the contribution of melanopsin and the difference in the first and last blue pulse, shown in Figure 8. The signaling of melanopsin in ipRGCs has been studied in rodents (Zhu et al., 2007), but these findings support a role for melanopsin in this adaptive response to repeated stimuli in healthy humans.

When exploring the repeatability of pupil responses between the three visits, there was no significant change in the initial difference between red and blue responses for any

of the visits. This indicates good long-term repeatability of this suspected melanopsin-dependent parameter. In contrast, a previous study suggests that individuals with a history of mild TBI exhibit greater variability in pupil responses over the span of two to four weeks (Yuhas et al. 2017). The suspected melanopsin-dependent parameter seems to be a characteristic stable across many months and not a random occurrence in the healthy adolescents.

The CogState scores for the Learning, Attention, Processing Speed, and Working Memory Speed Tasks were compared between the visits. There was a significant improvement in scores for the Learning Task across the hockey season. The Processing Speed, Attention, and Working Memory Speed Task scores were more repeatable throughout the season. A practice effect that improves the score the more times it is preformed may be present for the Learning Task, but not for the other tasks. This suggests that clinicians may need to factor in a score improvement for the CogState Learning Task when comparing scores to baseline over time. An individual who scores the same on the Learning Task a few months after baseline may be suspected of a brain injury, although no significant score change is present.

Computerized neurocognitive testing is commonly used by sports medicine physicians to assess the presence of concussion. However, to our knowledge, this is the first study to perform these assessments at multiple time-points across the season of play in youth hockey players. Although test-retest reliability of computerized neurocognitive testing is only moderate (MacDonald and Duerson, 2015), the ability to compare the results within-individual across multiple (three) time-points throughout the hockey

season help to lessen the effects of inter-individual variation that can affect the results from cross-sectional studies that compare case groups (those with SRC) to control groups (those without SRC).

There was a strong correlation between the CISS and RPCSQ scores, suggesting that individuals with higher severity of visual symptoms may also report higher severity of concussion symptoms. This suggests that there is survey overlap, meaning each of these questionnaires may not be as specific to the intended area of concussion or vision symptom screening. There is no need to utilize both surveys as either is acceptable to screen for a concussion. However, caution should be used when screening for a concussion with only a survey due to the self-report nature of it.

One limitation to the study is the small sample size of only 18 participants, which were recruited from the same hockey team in Columbus, Ohio. The data collection occurred over three sessions, and required both pupil readings and survey questions, which is time-consuming for the subjects. Pupil responses were recorded during the evening at each session, and this consistency was important as there is evidence that melanopsin activity is under circadian modulation (Münch et. al., 2012). It would be useful to explore the pupil responses in a different geographical location, a different season of the year, or time of day to see if these variables would have a significant effect on the pupil responses, whether that be on the repeatability measures or the contribution of melanopsin and photopotentiation.

Future studies may benefit from exploring the variability in pupil responses and contribution of melanopsin and photopotentiation in both a control group of subjects and

those who have been diagnosed with a mild traumatic brain injury. By investigating the differences between these two groups, pupil response testing may provide a more objective and efficient way to aid in the diagnosis of a sports-related concussion. The on-site pupil testing and administration of surveys may provide a convenient and accessible way to screen for a concussion. Pupil variability was exhibited in individuals with mild TBI (Yuhas et al., 2017). Although the healthy athletes in our study are young and learning, three of the four CogState parameters and the pupil responses remained stable across five months. If any of these values change significantly over time, it may indicate a SRC.

Bibliography

Alla, S., Sullivan, S. J., Hale, L., & McCrory, P. (2009). Self-report scales/checklists for the measurement of concussion symptoms: a systematic review. *British journal of sports medicine*, *43 Suppl 1*, i3–i12. https://doi.org/10.1136/bjsm.2009.058339

Bade, A., Boas, M., Gallaway, M., Mitchell, G. L., Scheiman, M., Kulp, M. T., Cotter, S. A., Rouse, M., & CITT Study Group (2013). Relationship between clinical signs and symptoms of convergence insufficiency. *Optometry and vision science: official publication of the American Academy of Optometry*, *90*(9), 988–995. https://doi.org/10.1097/OPX.0000000000000012

Berson, D. M., Castrucci, A. M., & Provencio, I. (2010). Morphology and mosaics of melanopsin-expressing retinal ganglion cell types in mice. *The Journal of comparative neurology*, *518*(13), 2405–2422. https://doi.org/10.1002/cne.22381

Berson, D. M., Dunn, F. A., & Takao, M. (2002). Phototransduction by retinal ganglion cells that set the circadian clock. *Science (New York, N.Y.)*, *295*(5557), 1070–1073. https://doi.org/10.1126/science.1067262

Borsting, E. J., Rouse, M. W., Mitchell, G. L., Scheiman, M., Cotter, S. A., Cooper, J., Kulp, M. T., London, R., & Convergence Insufficiency Treatment Trial Group (2003). Validity and reliability of the revised convergence insufficiency symptom survey in children aged 9 to 18 years. *Optometry and vision science : official publication of the American Academy of Optometry*, *80*(12), 832–838. https://doi.org/10.1097/00006324-200312000-00014

Carrick, F. R., Azzolino, S. F., Hunfalvay, M., Pagnacco, G., Oggero, E., D'Arcy, R., Abdulrahman, M., & Sugaya, K. (2021). The Pupillary Light Reflex as a Biomarker of Concussion. *Life (Basel, Switzerland)*, *11*(10), 1104. https://doi.org/10.3390/life11101104

Centers for Disease Control and Prevention. 2017 State and Local Youth Risk Behavior Survey Data. https://www.cdc.gov/healthyyouth/data/yrbs/data.htm. Atlanta, GA: Centers for Disease Control and Prevention.

Dacey, D. M., Liao, H. W., Peterson, B. B., Robinson, F. R., Smith, V. C., Pokorny, J., Yau, K. W., & Gamlin, P. D. (2005). Melanopsin-expressing ganglion cells in primate retina signal colour and irradiance and project to the LGN. *Nature*, *433*(7027), 749–754. https://doi.org/10.1038/nature03387

Dhawan, P. S., Leong, D., Tapsell, L., Starling, A. J., Galetta, S. L., Balcer, L. J., Overall, T. L., Adler, J. S., Halker-Singh, R. B., Vargas, B. B., & Dodick, D. (2017). King-Devick Test identifies real-time concussion and asymptomatic concussion in youth athletes. *Neurology. Clinical practice*, *7*(6), 464–473. https://doi.org/10.1212/CPJ.0000000000000381

Dziemianowicz, M. S., Kirschen, M. P., Pukenas, B. A., Laudano, E., Balcer, L. J., & Galetta, S. L. (2012). Sports-related concussion testing. *Current neurology and neuroscience reports*, *12*(5), 547–559. https://doi.org/10.1007/s11910-012-0299-y

Echlin, P. S., Tator, C. H., Cusimano, M. D., Cantu, R. C., Taunton, J. E., Upshur, R. E.,

Hall, C. R., Johnson, A. M., Forwell, L. A., & Skopelja, E. N. (2010). A prospective study of physician-observed concussions during junior ice hockey: implications for incidence rates. *Neurosurgical focus*, *29*(5), E4. https://doi.org/10.3171/2010.9.FOCUS10186

Estevez, M. E., Fogerson, P. M., Ilardi, M. C., Borghuis, B. G., Chan, E., Weng, S., Auferkorte, O. N., Demb, J. B., & Berson, D. M. (2012). Form and function of the M4 cell, an intrinsically photosensitive retinal ganglion cell type contributing to geniculocortical vision. *The Journal of neuroscience: the official journal of the Society for Neuroscience*, *32*(39), 13608–13620. https://doi.org/10.1523/JNEUROSCI.1422-12.2012

Foster, R. G., Provencio, I., Hudson, D., Fiske, S., De Grip, W., & Menaker, M. (1991). Circadian photoreception in the retinally degenerate mouse (rd/rd). *Journal of comparative physiology. A, Sensory, neural, and behavioral physiology*, *169*(1), 39–50. https://doi.org/10.1007/BF00198171

Gamlin, P. D., McDougal, D. H., Pokorny, J., Smith, V. C., Yau, K. W., & Dacey, D. M. (2007). Human and macaque pupil responses driven by melanopsin-containing retinal ganglion cells. *Vision research*, *47*(7), 946–954. https://doi.org/10.1016/j.visres.2006.12.015

Hannibal, J., Hindersson, P., Knudsen, S. M., Georg, B., & Fahrenkrug, J. (2002). The photopigment melanopsin is exclusively present in pituitary adenylate cyclase-activating polypeptide-containing retinal ganglion cells of the retinohypothalamic tract. *The Journal of neuroscience : the official journal of the Society for Neuroscience*, *22*(1), RC191. https://doi.org/10.1523/JNEUROSCI.22-01-j0002.2002

Hattar, S., Liao, H. W., Takao, M., Berson, D. M., & Yau, K. W. (2002). Melanopsin-containing retinal ganglion cells: architecture, projections, and intrinsic photosensitivity. *Science (New York, N.Y.)*, *295*(5557), 1065–1070. https://doi.org/10.1126/science.1069609

Hattar, S., Kumar, M., Park, A., Tong, P., Tung, J., Yau, K. W., & Berson, D. M. (2006). Central projections of melanopsin-expressing retinal ganglion cells in the mouse. *The Journal of comparative neurology*, *497*(3), 326–349. https://doi.org/10.1002/cne.20970

Hattar, S., Ecker, J. L., Dumitrescu, O. N., Chen, S. K., Wong, K. Y., Alam, N. M., Prusky, G. T., Berson, D. M. (2009). Functions and Target Innervations of Distinct Subtypes of Melanopsin Cells. *Invest. Ophthalmol. Vis. Sci., 50(13), 5027.*

Herbst K, Sander B, Milea D, Lund-Andersen H, Kawasaki A (2011) Test–retest repeatability of the pupil light response to blue and red light stimuli in normal human eyes using a novel pupillometer. Front. Neur. 2:10. doi: 10.3389/fneur.2011.00010

Ingram, B. M., Kay, M. C., Vander Vegt, C. B., & Register-Mihalik, J. K. (2019). The Effect of Body Checking Policy Changes on Concussion Incidence in Canadian Male Youth Ice Hockey Players: A Critically Appraised Topic. *Journal of sport rehabilitation*, *28*(7), 774–777. https://doi.org/10.1123/jsr.2018-0102

Joseph, J. R., Swallow, J. S., Willsey, K., Almeida, A. A., Lorincz, M. T., Fraumann, R. K., Oppenlander, M. E., Szerlip, N. J., & Broglio, S. P. (2019). Pupillary changes after clinically asymptomatic high-acceleration head impacts in high school football athletes. *Journal of neurosurgery*, 1–6. Advance online publication. https://doi.org/10.3171/2019.7.JNS191272

King, N. S., Crawford, S., Wenden, F. J., Moss, N. E., & Wade, D. T. (1995). The Rivermead Post Concussion Symptoms Questionnaire: a measure of symptoms commonly experienced after head injury and its reliability. *Journal of neurology*, *242*(9), 587–592. https://doi.org/10.1007/BF00868811

Li, J. Y., & Schmidt, T. M. (2018). Divergent projection patterns of M1 ipRGC subtypes. *The Journal of comparative neurology*, *526*(13), 2010–2018. https://doi.org/10.1002/cne.24469

Lucas, R. J., Hattar, S., Takao, M., Berson, D. M., Foster, R. G., & Yau, K. W. (2003). Diminished pupillary light reflex at high irradiances in melanopsin-knockout mice. *Science (New York, N.Y.)*, *299*(5604), 245–247. https://doi.org/10.1126/science.1077293

Lucas, R. J., Douglas, R. H., & Foster, R. G. (2001). Characterization of an ocular photopigment capable of driving pupillary constriction in mice. *Nature neuroscience*, *4*(6), 621–626. https://doi.org/10.1038/88443

MacDonald, J., & Duerson, D. (2015). Reliability of a Computerized Neurocognitive Test in Baseline Concussion Testing of High School Athletes. *Clinical journal of sport medicine : official journal of the Canadian Academy of Sport Medicine*, *25*(4), 367–372. https://doi.org/10.1097/JSM.0000000000000139

Maruff P, Thomas E, Cysique L, et al. Validity of the CogState brief battery: relationship to standardized tests and sensitivity to cognitive impairment in mild traumatic brain injury, schizophrenia, and AIDS dementia complex. *Arch Clin Neuropsychol*. 2009;24(2):165-178. doi:10.1093/arclin/acp010

Master, C. L., Podolak, O. E., Ciuffreda, K. J., Metzger, K. B., Joshi, N. R., McDonald, C. C., Margulies, S. S., Grady, M. F., & Arbogast, K. B. (2020). Utility of Pupillary Light Reflex Metrics as a Physiologic Biomarker for Adolescent Sport-Related Concussion. *JAMA ophthalmology*, e203466. Advance online publication. https://doi.org/10.1001/jamaophthalmol.2020.3466

McDougal, D. H., & Gamlin, P. D. (2010). The influence of intrinsically-photosensitive retinal ganglion cells on the spectral sensitivity and response dynamics of the human pupillary light reflex. *Vision research*, *50*(1), 72–87. https://doi.org/10.1016/j.visres.2009.10.012

McCrory, P., Meeuwisse, W., Dvořák, J., Aubry, M., Bailes, J., Broglio, S., Cantu, R. C., Cassidy, D., Echemendia, R. J., Castellani, R. J., Davis, G. A., Ellenbogen, R., Emery, C., Engebretsen, L., Feddermann-Demont, N., Giza, C. C., Guskiewicz, K. M., Herring, S., Iverson, G. L., Johnston, K. M., … Vos, P. E. (2017). Consensus statement on concussion in sport-the 5th international conference on concussion in sport held in Berlin, October 2016. *British journal of sports medicine*, *51*(11), 838–847. https://doi.org/10.1136/bjsports-2017-097699

Münch, M., Léon, L., Crippa, S. V., & Kawasaki, A. (2012). Circadian and wake-

dependent effects on the pupil light reflex in response to narrow-bandwidth light pulses. *Investigative ophthalmology & visual science*, *53*(8), 4546–4555. https://doi.org/10.1167/iovs.12-9494

Noseda, R., Kainz, V., Jakubowski, M., Gooley, J. J., Saper, C. B., Digre, K., & Burstein, R. (2010). A neural mechanism for exacerbation of headache by light. *Nature neuroscience*, *13*(2), 239–245. https://doi.org/10.1038/nn.2475

Peiffer AJ, MacDonald J, Duerson D, Mitchell G, Hartwick ATE, McDaniel CE. The Influence of Binocular Vision Symptoms on Computerized Neurocognitive Testing of Adolescents With Concussion. *Clin Pediatr (Phila)*. 2020;59(11):961-969. doi:10.1177/0009922820927477

Provencio, I., Jiang, G., De Grip, W. J., Hayes, W. P., & Rollag, M. D. (1998). Melanopsin: An opsin in melanophores, brain, and eye. *Proceedings of the National Academy of Sciences of the United States of America*, *95*(1), 340–345. https://doi.org/10.1073/pnas.95.1.340

Quattrochi, L. E., Stabio, M. E., Kim, I., Ilardi, M. C., Michelle Fogerson, P., Leyrer, M. L., & Berson, D. M. (2019). The M6 cell: A small-field bistratified photosensitive retinal ganglion cell. *The Journal of comparative neurology*, *527*(1), 297–311. https://doi.org/10.1002/cne.24556

Sandel, N., Reynolds, E., Cohen, P. E., Gillie, B. L., & Kontos, A. P. (2017). Anxiety and Mood Clinical Profile following Sport-related Concussion: From Risk Factors to Treatment. *Sport, exercise, and performance psychology*, *6*(3), 304–323. https://doi.org/10.1037/spy0000098

Schmidt, T. M., & Kofuji, P. (2009). Functional and morphological differences among intrinsically photosensitive retinal ganglion cells. *The Journal of neuroscience: the official journal of the Society for Neuroscience*, *29*(2), 476–482. https://doi.org/10.1523/JNEUROSCI.4117-08.2009

Schmidt, T. M., Taniguchi, K., & Kofuji, P. (2008). Intrinsic and extrinsic light responses in melanopsin-expressing ganglion cells during mouse development. *Journal of neurophysiology*, *100*(1), 371–384. https://doi.org/10.1152/jn.00062.2008

Stabio, M. E., Sabbah, S., Quattrochi, L. E., Ilardi, M. C., Fogerson, P. M., Leyrer, M. L., Kim, M. T., Kim, I., Schiel, M., Renna, J. M., Briggman, K. L., & Berson, D. M. (2018). The M5 Cell: A Color-Opponent Intrinsically Photosensitive Retinal Ganglion Cell. *Neuron*, *97*(1), 150–163.e4. https://doi.org/10.1016/j.neuron.2017.11.030

Sullivan, G. M., & Artino, A. R., Jr (2013). Analyzing and interpreting data from likert-type scales. *Journal of graduate medical education*, *5*(4), 541–542. https://doi.org/10.4300/JGME-5-4-18

U.S. Department of Health and Human Services. Physical Activity Guidelines for Americans, 2nd Edition. Washington, DC: U.S. Department of Health and Human Services; 2018. https://health.gov/paguidelines/second-edition/pdf/Physical_Activity_Guidelines_2nd_edition.pdf

Yoo H., Mihaila D.M. Neuroanatomy, Visual System, Pupillary Light Reflexes and

Pathway. [Updated 2020 Aug 10]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2020 Jan. https://www.ncbi.nlm.nih.gov/books/NBK553169/

Yue, J. K., Winkler, E. A., Burke, J. F., Chan, A. K., Dhall, S. S., Berger, M. S., Manley, G. T., & Tarapore, P. E. (2016). Pediatric sports-related traumatic brain injury in United States trauma centers. *Neurosurgical focus*, *40*(4), E3. https://doi.org/10.3171/2016.1.FOCUS15612

Yuhas, P. T., Shorter, P. D., McDaniel, C. E., Earley, M. J., & Hartwick, A. T. (2017). Blue and Red Light-Evoked Pupil Responses in Photophobic Subjects with TBI. *Optometry and vision science: official publication of the American Academy of Optometry*, *94*(1), 108–117. https://doi.org/10.1097/OPX.0000000000000934

Zhu, Y., Tu, D. C., Denner, D., Shane, T., Fitzgerald, C. M., & Van Gelder, R. N. (2007). Melanopsin-dependent persistence and photopotentiation of murine pupillary light responses. *Investigative ophthalmology & visual science*, *48*(3), 1268–1275. https://doi.org/10.1167/iovs.060