VISIBILITY THROUGH SIMULATED ATMOSPHERIC HAZE AND ITS RELATION TO MACULAR PIGMENT

By

LAURA FLETCHER

(Under the Direction of Billy R. Hammond)

ABSTRACT

The predominant factor limiting the detectability of distant targets is veiling due to atmospheric scattering, known commonly as haze. It has been suggested that yellow filters (in this case, the macular pigments, MP) that absorb this haze could extend visual range. This hypothesis was tested on 27 subjects with a wide range of MP optical densities. Visibility was measured by varying the amount of simulated blue haze needed to veil a sine-wave grating (7.5 cyc/deg). Visibility for this target under xenon light and shortwave deficient (SWD) light was also assessed. MP was significantly related to energy at threshold for both haze (r = 0.59, p < 0.01) and xenon (r = 0.60, p < 0.01) backgrounds, but not the SWD background. Thus, subjects with higher levels of MP could withstand more light before losing sight of the target, which is consistent with previous modeling by Wooten and Hammond (2002).

INDEX WORDS: Macular pigment, lutein, zeaxanthin, visibility, atmospheric haze

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CHAPTER 1

INTRODUCTION

Overview

The role of the carotenoids lutein (L) and zeaxanthin (Z) in the retina has been explored for decades (for an excellent history of the research, see Nussbaum, Pruett, & Delori, 1981). These xanthophyllic molecules, which are found primarily in green, leafy vegetables (e.g., spinach and kale), are selectively taken up by the retina to the exclusion of other dietary carotenoids. Once there, they imbed in the Henle fiber layer (Snodderly, Brown, Delori, & Auron, 1984) anterior to the photoreceptor outersegments. Because L and Z accumulate in greatest density in the central macular region of the retina, they are together termed the macular pigments (MP). The amount of MP in the retina is determined primarily by an individual's diet, thus MP optical density (MPOD) varies widely from person to person (ranging from so little as to be nearly immeasurable up to approximately 1.5 log units of optical density).

MP is known to absorb light between the wavelengths of about 400 to 520 nm, with peak absorption occurring at 460 nm (Snodderly, et al., 1984). The wide variety in density among individuals thus leads to a situation where the amount of shortwave (SW) light reaching an individual's retina can vary significantly (e.g., an optical density of 0.3 equates to approximately 50% of 460 nm light being prevented from reaching the retina; whereas an OD of 1.5 absorbs nearly 98%). The MP's location anterior to the photoreceptor outersegments, as well as the known absorption of SW light, has led researchers to propose that the pigments could be

effectively screening the foveal photoreceptors from shortwave light, which would have some important consequences for vision and for overall eye health.

Hypotheses of MP

It has been previously demonstrated that SW light is especially damaging to retinal tissue (Ham, Ruffolo, Mueller, Clarke, & Moon, 1978). Any reduction in actinic SW light reaching the retina would therefore have a protective effect by reducing accumulated damage over the lifetime. This could lead to preserved functioning, especially in the foveal region (Junghans, Sies, & Stahl, 2001). In addition, L and Z are effective antioxidants and their ability to quench free radicals in the retina (which is one of the most metabolically active tissues in the body) may also confer protection (Khachik, Bernstein, & Garland, 1997). Some positive evidence for these "protection hypotheses" of MP has come from observational research linking higher carotenoid consumption to a reduced risk of age-related macular degeneration (AMD; e.g., see Beatty, Boulton, Henson, Koh, & Murray, 1999 for a review).

While the protective function of MP is obviously quite important, it is unlikely that we evolved to accumulate these pigments for the protection they confer. The immediate optical benefits provided by filtration are the more likely cause for why we originally began to accumulate MP. In 1866 Schultz put forth what is now termed the acuity hypothesis of MP (as cited in Nussbaum, et al., 1981). Because shortwave light is most out of focus due to chromatic aberration by the lens, filtration of this out-of-focus light could lead to improvements in acuity. Walls and Judd (1933) later put forth several additional optical theories of the function of MP, including Shultz's "acuity hypothesis". They posited that filtration could also enhance chromatic borders by selectively absorbing one side of the border more than the other, thus making the edge more distinct. MP could reduce the discomfort caused by bright lights via a reduction of the

effects of glare or dazzle. Finally, MP could absorb atmospheric haze, which is predominantly shortwave, thereby improving vision outdoors. The first three of these hypotheses have all been tested empirically in multiple studies (e.g., Reading & Weale, 1974; Engles, Wooten, & Hammond, 2007; Renzi & Hammond, 2010; Stringham & Hammond, 2007, 2008); however, the visibility hypothesis (i.e., the idea that outdoor vision can be improved by selective absorption of blue haze) has remained largely untested. The sole previous empirical study of the visibility hypothesis was a small within-subjects study of contrast sensitivity in the presence of simulated haze. (Hammond, Wooten, Engles, & Wong, 2012). Visibility was assessed by measuring contrast sensitivity at 8 cycles per degree (cpd) with varying levels of MP (simulated with a variable path length filter filled with a solution that matched the absorbance spectrum of MP). They found the largest improvements in contrast sensitivity occurred for the initial addition of 0.25 units of optical density, and a plateau effect for MPOD above approximately 0.50 at 30' retinal eccentricity.

Visibility Hypothesis of MP

Our atmosphere is filled with particles of various sizes, which come from both natural and human-made sources. They range in size from very small (e.g., air molecules, radius of 10^{-4} µm) to quite large (e.g., rain droplets, radius of about 10^3 µm; Wooten & Hammond, 2002). Light from the sun travels through our atmosphere, and interacts with these particles in several ways; for example rays may be absorbed, or, more importantly for vision, scattered by these atmospheric particles. The way in which elastic scatter (i.e., scatter in which the frequency of the incident light is preserved) occurs depends on several factors: the size of the scattering particle, the distance between particles and their spatial arrangement, and the refractive index of the particles as compared to their surrounding medium (as reviewed by Engles, 2011). Particle size,

as compared to the wavelength of light with which it is interacting, is likely the most important of these. In 1871, Lord Rayleigh demonstrated that when a particle in the atmosphere is much smaller than the incident wavelength of light, the light is scattered according to the inverse fourth power of wavelength (as cited in Bohren & Fraser, 1985). Thus, shorter wavelengths (i.e., "blue" light) are scattered with greater efficiency than longer wavelengths; this is termed Rayleigh scatter.

Due to the abundance of larger particles in the atmosphere, pure Rayleigh scatter is almost never observed. In fact, it has been calculated that the maximum color purity the sky can have is 42% (at a peak of 475 nm; Bohren & Fraser, 1985). In 1908, Gustav Mie expanded Lord Rayleigh's model to encompass particles of all sizes. The model is quite complex, but essentially, larger particles scatter light more effectively, but this scatter is independent of wavelength (e.g., clouds are made up of large water droplets, thus when they scatter light they appear as mixture of all wavelengths – white).

Nevertheless, Rayleigh scatter is the predominant reason for the apparent blueness of the sky (other factors are the solar spectrum of light and the spectral sensitivity of the human eye). The resultant "blue haze" through which we view distant objects, is the most significant limiting factor for how far one can see outdoors (Wooten & Hammond, 2002). When viewing a distant target, light reflected from the target is scattered out of the optical path to the observer. As SW light is scattered out of the observational path more efficiently, the longer wavelengths from the target are more likely to reach the eye of the observer. This can result in a viewing situation where the target is rendered somewhat SW deficient, and is seen on a predominantly SW background (see Figure 1). Prior research has demonstrated that absolute thresholds for a yellow target (i.e., SW deficient) on a blue background are reduced when viewed through a yellow filter

(Luria, 1972). Additional research on the effect of yellow lenses on contrast sensitivity has given mixed results (e.g., Kelly, Goldberg, & Banton, 1984 found no improvement in contrast sensitivity; Wolffsohn, Cochrane, Khoo, & Wu, 2000 did find improvements). This lack of consistency is likely due to the fact that few filter studies have measured MP optical density (MPOD) in subjects. The MP is an effective filter of SW light, and adding an additional filter may either have no effect on performance, or may reduce the overall luminance to such a degree that performance is actually impaired.

In addition to atmospheric factors, visibility is also affected by the color and luminance of the target object, as well as individual differences in observers. If these are taken into account, the visible range for an object can typically be calculated by also factoring in the reduction in contrast by the atmosphere between an object and its background (which is frequently, but not always, the horizon; Duntley, 1948). The individual factor of greatest interest here is the previously mentioned highly variable intraocular filter, the MP. Prior modeling by Wooten and Hammond (2002) has suggested that for individuals with equal Snellen acuity, a person with high MP would have a 30 percent increase in visual range over a person with no MP. For individuals who are required to perform difficult tasks outdoors as part of their daily job requirements (e.g., pilots), this could translate to a meaningful difference in job performance. *Summary and Hypotheses*

The present study aimed to test the visibility hypothesis of MP in a controlled, but ecologically valid, way. Xenon light, when paired with a specialized glass filter, can almost exactly approximate atmospheric haze (see Appendix 1 for spectral information). Thresholds for a SW deficient target superposed on this haze background should be related to an individual's MP levels. MP should also be related to thresholds for the same target superposed on a

broadband xenon background (because of the large quantity of SW light in the xenon spectrum). Thresholds should be unrelated to MP for the SW deficient target superposed on a SW deficient background because neither target nor background will be absorbed by MP. This SW deficient background condition, which spectrally obviates the effect of the MP, can also be used as a control for individual differences in task performance, as well as individual differences in contrast sensitivity. When controlling for these individual differences, the relationships between MP and thresholds in the haze and xenon conditions should be strengthened. The effect of the MP can be spatially obviated by measuring thresholds for the grating target on the same three backgrounds viewed parafoveally. The thresholds obtained in these conditions should be unrelated to MPOD, providing additional support that the filtration of the MP is the driving factor for target visibility.

CHATPER 2

METHODS

Subjects:

A total of 27 (16 female; 11 male) subjects were recruited for this study. All subjects were recruited from the University of Georgia population. Subjects ranged in age from 18 - 29 (Mean = 21.3; SD = 2.9). Informed consent was obtained from all subjects prior to any experimental procedures, and the experiment followed the guidelines of the Declaration of Helsinki as well as the University of Georgia Institutional Review Board. Inclusion criteria included: ability to perform study tasks, no history of relevant ocular disease, Snellen acuity better than 20/40 (corrected), and age between 18 and 30 years. Two subjects were excluded from analysis for corrected Snellen acuity worse than 20/40.

Assessment of Macular Pigment Optical Density:

Macular pigment optical density (MPOD) was measured in the right eye only of all subjects. Measurement of only one eye is possible because of the good interocular agreement for MP (Hammond & Fuld, 1992). Measurement was done using a Macular Densitometer (Macular Metrics, Rehoboth, MA), which allows for the detailed measurement of retinal levels of lutein and zeaxanthin in a noninvasive manner and in free-view. The psychophysical procedure used, heterochromatic flicker photometry (HFP), has been extensively validated for measuring MP (Wooten, Hammond, Land, & Snodderly, 1999). Briefly, a 458 nm measuring light (which is strongly absorbed by MP) is alternated in counter phase with a 570 nm reference light (which falls outside the absorption spectrum of MP). The differential between the "blue" and "green" light caused by absorption of the "blue" by MP creates the appearance of a flickering stimulus. Subjects can then adjust the radiance of the "blue" measuring light to achieve a point of null flicker (i.e., when the "blue" and the "green" are perceptually equally bright).

Because of large individual differences in temporal vision, the flicker rates of the stimuli were customized for each subject (known as customized HFP, or cHFP). This customization procedure has been previously described in detail by Stringham, Hammond, Nolan, Wooten, Mammen, Smollon, & Snodderly (2008). Essentially, a single measure of temporal vision known as the critical flicker fusion was obtained for each subject, and this value was used to calculate the ideal setting for each stimulus. Critical flicker fusion (CFF) was measured with the 570 nm reference light to avoid the influence of MP. The light is presented in square wave, with the frequency increased until the point at which subjects can no longer perceive flicker, and the stimulus perceptually fuses (i.e., the CFF). One ascending and descending method of limits threshold were averaged to obtain CFF.

A detailed spatial profile of MPOD was obtained by using stimuli of various sizes. Because MP is sampled at the edge of a stimulus when using HFP (Hammond, Wooten & Snodderly, 1997), using increasingly larger stimuli allows for measurement at increasing retinal eccentricities (e.g., a target 1[°] in size will measure MPOD at 30' retinal eccentricity). MPOD was measured at retinal eccentricities of 7.5', 30', 60', and 120'. A parafoveal measure at 7° was obtained by having subjects fixate a red light to the left of the target while making their judgments. The parafoveal measure falls outside of where MP is optically measurable, and thus serves as a reference point for calculating the optical density at the other retinal eccentricities.

Assessment of Visibility Thresholds

A three-channel Newtonian-view optical system with a 1000 W xenon-arc lamp (Thermo Oriel Instruments, Stratford, CT) light source was used to obtain thresholds for the visibility of a sine-wave grating with varying background conditions. The target was viewed monocularly (right eye only), and head position was made stable with a combination chin-and-forehead rest assembly. Channel one was used to create the various backgrounds; channel two was used to create a 0.5 degree grating target; and channel three was used to create a fixation light for the parafoveal measures. The target channel contained a sine-wave grating which was 7.5 cycles per degree, and rendered shortwave deficient (SWD; cutoff = 570 nm) by a chromatic filter (Corning, 51300; Oriel, Stanford, CT). A SWD target most closely resembles what is likely to happen when viewing targets at a distance outdoors (i.e., SW light is most effectively scattered out of the optical path creating a situation in which SWD targets are viewed on a background that is SW heavy; see Wooten & Hammond, 2002).

The grating was kept at a constant brightness (110 nW) and its visibility was tested under three different background conditions, the presentation order of which was randomized for each subject. A blue haze condition was created with a chromatic filter (Schott glass Filter #BG34, UQG Optics Ltd., Barrington, NJ). The spectrum of this filter almost exactly replicates that of atmospheric haze (see Appendix 1). A broadband condition intended to mimic sunlight with the absence of haze was created with the xenon-arc light source made less intense with neutral density filters. The third background condition was a SWD (558 nm; half-power bandwidth = 8 nm, Edmund Optics, Barrington, NJ), background which falls outside the absorption spectrum of MP. This condition was included to essentially obviate the effect of MP, and thus serve as a control for individual differences in the task.

Alternating ascending and descending thresholds were obtained using the method of limits. An average of three ascending and three descending trials were obtained for each subject, but this could be as few as three in each direction or as many as five in each direction (for a total of 6 - 10 trials) depending on subject consistency. Threshold was taken to be the average background luminance at each transition point. For each trial, the experimenter-controlled wedge was set somewhere between 150 - 300 units above or below (depending on direction) the previously determined threshold value, to prevent subjects from using time elapsed since the beginning of the trial as a cue when making a threshold judgment. This threshold determination was followed by an abbreviated three-alternative forced choice procedure in which the subject made determinations about whether the grating was tilted to the left, to the right, or was straight up and down. Background luminance was set to roughly the threshold value obtained in the method of limits trials, and was increased or decreased until the subject could correctly identify the orientation of the grating approximately 66 percent of the time.

For the parafoveal assessment, the three background conditions were repeated while the subject fixated a red point of light placed 5° nasally. The same ascending and descending method of limits procedure was used, followed by an abbreviated three-alternative forced choice procedure. For all subjects, the order of the background conditions was randomized but the foveal measurements always took place prior to the parafoveal measures.

The final analysis was conducted using threshold values obtained using the method of limits procedure. The three-alternative forced choice procedure was found to be less reliable, presumably due to the well-known "oblique effect". Previous studies (e.g., Campbell, Kulikowski & Levinson, 1966) have demonstrated reduced visibility for oblique gratings compared to gratings oriented vertically or horizontally.

CHAPTER 3

RESULTS

See Table 1 for descriptive statistics (means and standard deviations) for MPOD. Average MPOD was relatively high for this sample (an OD of 0.46 at a retinal eccentricity of 30°), but the range was quite large (0.15 - 0.91 at 30° eccentricity) and follows a normal distribution. While most individuals have a MP spatial profile that closely resembles an exponential decay function, some individuals are known to have secondary peaks or troughs in their profiles (Hammond, et al, 1997). For these individuals, the convention of choosing MPOD at 30° retinal eccentricity as the dependent variable may not accurately reflect their true MP status. Thus, a composite MP measure for each subject may also be desirable. An exponential curve was fit to each subject's MP profile, and a MP area under the curve (AUC) value was obtained for each subject. All analyses were conducted using the conventional 30° retinal eccentricity measure of MPOD as well as the AUC measure.

Pearson-product moment correlations were performed to determine the association between MPOD and log energy at threshold for each of the three background conditions (see Table 2). Statistical significance was set at p < 0.05. MPOD at 30' as well as the composite value was highly correlated with log energy required to lose sight of the target for both the haze (r =0.59, p < 0.01 at 30' eccentricity; see Figure 2) and the xenon (r = 0.60, p < 0.01 at 30' eccentricity; see Figure 3) background conditions. As expected, MPOD was unrelated to log energy at threshold for the 558 nm background condition (r = 0.21, p = 0.31; see Figure 4).

Thresholds obtained in the 558 nm background condition can be used to control for individual differences in task performance. Semi-partial correlations between MPOD and log energy in the haze and background conditions while controlling for thresholds in the 558 nm background condition were also performed (see Table 3). Partialing out the shared variance between the background conditions did not significantly strengthen the relationship between MPOD and both haze (r = 0.61, p < 0.01 at 30' eccentricity) and xenon (r = 0.62, p > 0.01 at 30' eccentricity) thresholds.

Unexpectedly, MPOD was also significantly related to log energy at threshold for all three parafoveal conditions (see Table 2). However, when a semi-partial correlation between MPOD and the parafoveal thresholds is computed while controlling for the foveal thresholds in that condition, the relationship disappears (see Table 4). This relationship is thus possibly driven by the very strong correlation between foveal and parafoveal thresholds for each condition (e.g., r = 0.70, p < 0.01 for the haze background – with similar values for the other two conditions); though potential additional reasons for the significant correlations are addressed in the discussion section.

CHAPTER 4

DICUSSION

The accumulation of the MP in primates likely evolved due to its immediate optical benefits, which arise from its ability to screen the foveal cones from SW light. The purpose of the present study was to empirically test the visibility hypothesis of MP (i.e., that selective filtration of SW-dominant atmospheric haze can extend visual range outdoors) in an ecologically valid way.

When viewing objects close-up or at intermediate distances, a viewer can rely on both chromatic and luminance edges for object detection; however, at greater distances, edges tend to become isoluminant, and chromatic differences may be the only reliable indicator of an edge (Horvath, Gorraiz, & Raimann, 1981). Hansen and Gegenfurtner (2009) analyzed approximately 700 calibrated color images of natural scenes and found that isoluminant edges were not more or less common than pure luminance edges, and that in fact luminance and chromatic edges occur independently of one another. This lends credence to the idea that the MP could confer an advantage when viewing objects at a distance. By selectively filtering the background relative to the target, this chromatic border is enhanced leading to greater visibility.

Indeed, in the present study we found that individuals with higher levels of MP required more simulated haze to lose visibility of a SWD grating target. Log energy at threshold varied by a factor of two between individuals with the highest and lowest levels of MP implying that an individual with high MP would be able to detect a target at a much greater distance (i.e., more atmospheric haze between them and the target) than an individual with low MP. Filtration by the

MP is enhancing the presence of a chromatic edge, leading to greater visibility. Unlike the previous study by Hammond et al (2012), the effect of MP did not seem to plateau for subjects Perhaps there is some optical difference between simulating high levels of MP with an artificial filter (as in the Hammond study) and truly high tissue levels (as in the present study). There is, however other evidence for a leveling-off of the optical effects of MP (e.g., Reading & Weale, 1974) so this possibility should be examined more closely in future studies.

For the broadband xenon background, which closely approximates the spectrum of sunlight, this same relationship is found. Xenon light has a significant SW component making filtration by the MP a possible factor when detecting the target on a xenon background. This filtration of the background relative to the target enhances its visibility by creating a luminance edge between the two. As previously mentioned, luminance edges are important when viewing natural scenes close-up or at intermediate distances (per Horvath, et al, 1981). In our data, there is approximately a two-fold difference in amount of energy required to lose sight of the target between the individuals with the highest and lowest MP levels, and MP is positively related to the amount of energy in the background at threshold. This relationship between MP and log energy at threshold is not observed when the SWD target is superposed on a 558 nm background because neither the target nor the background are absorbed by the MP.

MP was positively related to background energy at threshold for all three conditions when viewed parafoveally. There are a few potential non-mutually exclusive reasons for why this unexpected result may have occurred. First, the fixation point was placed only 5 degrees out. While MPOD has decreased considerably by 5 degrees, some individuals may still have enough MP at this spatial location to be optically significant. Five degrees was chosen because putting the fixation point any farther out would have made the task more difficult and less reliable.

Second, all but two subjects were naïve to psychophysical experiments. Making parafoveal judgments about a grating is a difficult task, and there may have been some amount of "cheating" (i.e., glancing over at the target) when subjects were making their decisions about whether the grating was visible or not. Subjects appear not to have been making only foveal judgments during the parafoveal assessment, since all subjects had foveal thresholds that were markedly lower than their parafoveal thresholds. Nevertheless, the parafoveal thresholds obtained in this study may be some combination of foveal thresholds and subjects' true parafoveal thresholds. Finally, there may be some effect of MP that is not necessarily optical. As previously noted, L and Z imbed in the Henle fiber layer anterior to the foveal cones. There are considerable individual differences in the number of foveal cones (Curcio, Sloan, Packer, Hendrickson & Kalina, 1987), and these differences extend out to several degrees of eccentricity. If more photoreceptors, which would presumably lead to improved spatial vision, also led to more "space" for accumulating MP, then a relationship between MP and measures of spatial vision may be seen even in tasks for which the optical effects of MP should not play a role.

In sum, the present study provides support for the visibility hypothesis of the MP. The next obvious step is to conduct a similar study with subjects with a wide range of MPOD in an outdoor setting for maximum ecological validity. Finding that the same relationship exists outside the carefully controlled laboratory setting would perhaps provide the impetus for making measurement of MP (and subsequent supplementation for individuals found to have low levels) in individuals whose job performance could be enhanced by better vision outdoors.

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7.5'	30'	60'	120'	AUC
0.55 ± 0.15	0.46 ± 0.13	0.32 ± 0.14	0.15 ± 0.10	42.1 ± 17.2

Table 1. Descriptive statistics (mean \pm s.d.) for MPOD*

*Specified by retinal eccentricity

Table 2. Zero-order Pearson product-moment correlation coefficients for associations between

	30' MPOD	MP AUC
Foveal	0.59^{*}	0.61^{*}
Haze log E		
Foveal	0.60^{\dagger}	0.60^{\dagger}
Xenon log E		
Foveal	0.21	0.31
558 nm log E		
Parafoveal Haze	0.59^{\dagger}	0.50*
log E		
Parafoveal Xenon	0.62^{\dagger}	0.55^{\dagger}
log E		
Parafoveal 558 log	0.45*	0.39
Е		

MPOD and log energy at threshold

* p < 0.05

p < 0.01

Table 3. Semi-partial correlation coefficients for associations between MPOD and foveal log energy at threshold

	30' MPOD	MP AUC
Haze log E	0.61^{+}	0.54^{*}
Residuals		
Xenon log E	0.62^{+}	0.52^{+}
Residuals		

 $\ddagger p < 0.01$

Table 4. Semi-partial correlation coefficients for associations between MPOD and parafoveal log energy at threshold controlling for foveal thresholds

	30' MPOD	MP AUC
Haze log E	0.31	0.39
Xenon log E	0.21	0.26
558 nm log E	0.08	0.04

* *p* < 0.05



Figure 1. Rayleigh scatter creates a situation in which we frequently view shortwave-deficient targets on a shortwave background





Figure 2. The relationship between MPOD at 30' retinal eccentricity and haze background energy at threshold



r = 0.60, p = 0.001

Figure 3. The relationship between MPOD at 30' retinal eccentricity and xenon background energy at threshold





Figure 4. The relationship between MPOD at 30' retinal eccentricity and 558 nm background energy at threshold



