



Tritanopic color matches and the middle- and long-wavelength-sensitive cone spectral sensitivities

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Abstract

Tritanopic color matches (i.e. matches that depend on the middle- (M) and long- (L), but not short- (S) wavelength-sensitive cones) were made between two half-fields: one illuminated by either a 405 or a 436 nm Hg spectral line; the other by a light of variable wavelength and radiance. Our purpose was to test between rival M- and L-cone spectral sensitivities, which should predict the tritanopic matches. The observers were tritanopes, in whom functioning S-cones are lacking, or normal trichromats, in whom artificial tritanopia was induced by a strong, violet adapting field. The wavelengths found to match the 405 and 436 nm lights agreed poorly with those predicted by the cone spectral sensitivities of Smith and Pokorny (1975) [*Vision Research*, 15, 161], while the 405 nm matching wavelength agreed poorly with that predicted by Stockman, MacLeod and Johnson (1993) [*Journal of the Optical Society of America*, A10, 2491]. Both matching wavelengths agreed well, however, with the predictions of the Stockman and Sharpe (2000) [*Vision Research*] M- and L-cone spectral sensitivities, which lie within the range of measured matches. © 2000 Elsevier Science Ltd. All rights reserved.

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1. Introduction

The three types of cone photoreceptor, each with different spectral sensitivity, are referred to as long-, middle- and short-wavelength-sensitive (L, M and S), according to the relative spectral positions of their peak sensitivities. This paper is one in a series (Stockman & Sharpe, 1998, 1999, 2000; Stockman, Sharpe & Fach, 1999) aimed at providing a definitive estimate of the human cone spectral sensitivities or cone fundamentals.

In this paper, we test between rival estimates of the M- and L-cone fundamentals by measuring color matches made under ‘tritanopic’ conditions; that is, under conditions in which the S-cones do not contribute to the match. Such color matches depend on just the M- and L-cones, and consequently should be predicted by any feasible pair of M- and L-cone fundamentals.

This type of test is most critical at short-wavelengths, where small uncertainties about the spectral sensitivities of the M- and L-cone fundamentals persist. These uncertainties remain for several reasons. First, the derivation of the M- and L-cone fundamentals typically depends on spectral sensitivities measured, respectively, in small groups of protanopes and deuteranopes. Because macular and lens pigment densities can vary considerably between individuals, the mean M-cone and L-cone spectral sensitivity functions obtained separately from small groups of observers are unlikely to reflect the same underlying densities. Second, cone fundamentals are usually defined as linear combinations of color matching data, known as color matching functions or CMFs. Although the Stiles (1955) 2° and Stiles and Burch (1959) 10° CMFs are believed to be reliable at short-wavelengths, there are doubts about the validity of the CIE 2° CMFs in this spectral region (see Alpern, 1976; Estévez, 1979; Stockman & Sharpe, 1999). Third, M- and L-cone isolation experiments in dichromats require the elimination of the S-cone response at shorter wavelengths. In some cases, these have been only partially successful.

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Tritanopic matching data already exist in the literature; most notably those of Wright (1952). Stockman, MacLeod and Johnson (1993) used Wright's data to make substantial adjustments to their M- and L-cone fundamentals at shorter wavelengths (see Fig. 6, below). There are, however, concerns about such a manipulation. First, Wright's data were measured with a 1.33° diameter target that was 56% smaller in area than the 2° diameter traditionally used to measure CMFs and to define cone spectral sensitivities. Consequently, Wright's data almost certainly reflect higher photopigment optical densities than are appropriate for a 2° field (see Fig. 7, below). Second, Wright's data are for only seven subjects of unknown L- or M-cone photopigment genotype. Given that polymorphisms are common, such a small group is unlikely to represent accurately the mean M- and L-cone spectral sensitivities in the population.

The primary aim of the current work was to measure tritanopic color matches with the standard 2° target. Tritan matches will vary, however, because of L- and M-cone polymorphisms. Below, we model the effects of the most common polymorphism — the substitution of alanine for serine at position 180 of the L-cone opsin — on the tritan matches. The L-cone genotypes of four of our 11 subjects are known (see below).

Following the suggestion of Pokorny and Smith (1993), we used the short-wavelength 405 and 436 nm Hg lines as test lights. Spectral lines are nearly monochromatic, so that individual differences in macular and lens pigment density primarily affect the radiance of the lines, rather than their wavelength distributions. For lights of broader spectral bandwidth, however, individual differences in pre-receptor lens and macular pigment density can skew the wavelength distributions that reach the photoreceptor. Since the Hg lights that we used were produced by a high-pressure Hg arc lamp, they have a bandwidth of ~4 nm, which we must take into account in our calculations (see below).

Under tritanopic conditions, the Hg lines can be uniquely matched by longer-wavelength spectral lights that yield the same relative absorptions in the L- and M-cones. Such pairs of lights are known as tritanopic metamers. The simplest way of producing tritanopic conditions is to use subjects who suffer from tritanopia, a rare form of color vision deficiency in which functioning S-cones are lost or severely reduced in number (for a review, see Sharpe, Stockman, Jägle & Nathans, 1999b). In addition, it is possible to engender artificial tritanopia in color normals, simply by presenting near-threshold targets on an intense 420 nm background. One complication, however, is that such backgrounds bleach a fraction of the M- and L-cone photopigments,

for which a correction to the wavelength matches must be made (see below).

Our initial intention was, like Stockman et al. (1993), to use the tritanopic matches to 'refine' our M- and L-cone fundamentals (Stockman and Sharpe, 2000) at short-wavelengths by slightly adjusting the linear transformation from the CMFs to the fundamentals for consistency with the matches. However, unlike Stockman et al. (1993), we found that such adjustments were unnecessary, because the tritanopic predictions of the original fundamentals were already within the range of the measured matches.

2. Methods

2.1. Subjects

Eleven subjects made matches: two tritanopes and nine color normals. Seven of the subjects, labeled T1–T7, were measured in Tübingen, and four, labeled S1–S4, were measured in San Diego. S4 was female; the rest, male.

2.1.1. Tritanopes

The two tritanopes, S4 and T7, were identified and tested as part of another study (Weitz, Miyake, Shinzato, Montag, Zrenner, Went & Nathans, 1992). S4 was extensively psychophysically tested in San Diego by Ethan Montag; T7 in Munich by Eberhart Zrenner. Both were found to be tritanopes on standard and specialized tests for color vision and color discrimination (see Weitz et al., 1992). In addition, both are heterozygous for the replacement of serine by proline at codon 214 in the gene that encodes the S-cone opsin (S4 and T7 were, respectively, subjects E1 and F in the Weitz et al., 1992 study). The ser²¹⁴pro substitution confers tritanopia via an autosomal dominant inheritance with high penetrance (Weitz et al., 1992). Although the mechanism is not known, it is believed that the substitution, which is in the transmembrane domain of the pigment, produces a protein that actively interferes with the viability or fidelity of the S-cone photoreceptor (for further details, see Weitz et al., 1992; Sharpe, Stockman, Jägle, Knau & Nathans, 1999a; Sharpe et al., 1999b).

Although no evidence of S-cone function has been reported in the two tritanopes used in this study, many tritanopes retain some residual S-cone function under certain conditions (Pokorny, Smith & Went, 1981). To mitigate against such potential effects, as well as to maintain a constant level of pigment bleaching across all observers, we applied to the tritanopes the same conditions used to produce artificial tritanopia in color normals.

2.1.2. M- and L-cone genotype

All normal observers used in this study have ‘normal’ trichromatic color vision as defined by standard tests. Although it was not our intention to determine the L- and M-cone photopigment genotypes of our observers as part of this study, we were fortunate enough to know the genotypes of four of our observers from their participation in other studies.

The classification of photopigment genes is complicated by polymorphisms in the normal population, the most common of which is the frequent replacement of serine by alanine at codon 180 in exon 3. Approximately 56% of 304 Caucasian males with normal and deutan color vision have the serine variant (identified as L(S180)) and 44% the alanine variant (identified as L(A180)) for their L-cone gene (summarized in Table 1 of Stockman & Sharpe, 2000, from Winderickx, Battisti, Hibiya, Motulsky & Deeb, 1993; Neitz & Neitz, 1998; Sharpe, Stockman, Jägle, Knau, Klausen, Reitner & Nathans, 1998; and data from Schmidt, Sharpe, Knau & Wissinger, personal communication). In contrast, in the M-cone pigment, the alanine/serine polymorphism is much less frequent, 94% (Winderickx et al., 1993) or 93% (Neitz & Neitz, 1998) of males having the alanine variant.

Of our subjects, S1 is known to possess a single L(S180) and a single M(A180) photopigment gene (Nathans, personal communication), T1 similarly has a single L(S180) and a single M(A180) photopigment gene, T2 has an L(A180) gene and two M genes, and T4 has an L(S180) gene and four M genes (Schmidt et al., personal communication). The identities of the amino acids at position 180 of the M genes of T2 and T4 are not known, but are most likely alanine.

Below, we consider the effect of a change in spectral sensitivity from L(A180) to L(S180) on the tritan matches. According to the recent corneal spectral sensitivity measurements of Sharpe et al. (1998), the change in λ_{\max} from L(A180) to L(S180) is ~ 2.6 nm.

2.2. Apparatus

Measurements were made separately on two Maxwellian-view optical systems: one in San Diego and one in Tübingen (for a more detailed description of the latter, see Sharpe et al., 1998, 1999a). The short-wave-

length Hg line test lights of both systems originated from a 100 W Hg arc lamp, whereas the variable wavelength test lights originated from either a 50 W, 12 V tungsten halide lamp (San Diego) or a 75 W xenon-arc lamp (Tübingen). In San Diego, the 420 nm background was illuminated by the Hg lamp, and in Tübingen, by the Xe lamp. The images of the lights were less than 2 mm in diameter at the plane of the observer’s pupil.

Fine control over the radiance of the test lights was achieved by variable neutral density wedges positioned at arc or filament images, and by fixed neutral density filters inserted in collimated portions of the beams. The position of the observer’s head was maintained by a rigidly mounted dental wax impression.

2.3. Stimuli

Two vertically-bisected 2° half-fields were juxtaposed to make a circular bipartite field, which was presented in the center of a circular 16° diameter, 420 nm background field of varying radiance. The background wavelength was selected in San Diego by a 3-cavity, blocked interference filter (Ealing) with a full-width at half-maximum bandwidth (FWHM) of 11 nm, and in Tübingen by a grating monochromator (Jobin-Yvon H-10 Vis) with 2 mm entrance and exit slits, the output of which had an FWHM of 17 nm.

The wavelength of the standard half-field was rendered monochromatic by 3-cavity interference filters (Ealing in San Diego; Spindler and Hoyer in Tübingen) designed to transmit only either the 404.7 or the 435.8 nm lines of the high pressure Hg lamp. In practice, the Hg lines in a high-pressure lamp are broadened and shifted to longer wavelengths (see Elenbaas, 1951). Moreover, the ‘435.8 nm’ isolation filter used in Tübingen skewed the spectral distribution to longer wavelengths by an additional 1.4 nm. To avoid any errors caused by such wavelength shifts, we calibrated in situ the spectral ‘lines’ produced by our 100 W Hg lamps in combination with narrow-band interference filters. They were 405.2 and 438.2 nm in Tübingen and 405.2 and 436.3 nm in San Diego. The FWHMs of the lines were approximately 4 nm. A crucial spectroradiometric control was to ensure that the interference filters effectively blocked the unwanted Hg lines. One filter that

Table 1
Changes in the effective wavelengths of the Hg lights caused by changes in lens density of $\pm 25\%$ and changes in macular pigment density of $\pm 50\%$

Hg light (nm)	–25% Lens (nm)	+25% Lens (nm)	–50% Macular (nm)	+50% Macular (nm)
405.8	–0.16	+0.15	+0.03	–0.03
436.5	–0.04	+0.04	+0.02	–0.01
438.4	–0.03	+0.04	+0.02	–0.01

Table 2

The percentages of the M- and L-cone pigments estimated to be bleached by the 420 nm backgrounds used in San Diego and Tübingen

San Diego 420 nm background	Tübingen 420 nm background	% M-cone pigment bleached	% L-cone pigment bleached
10.8		7.1	6.1
	10.9	8.7	7.5
11.1		13.2	11.4
	11.2	16.0	14.0
11.4		23.2	20.5
	11.5	27.6	24.5
11.7		37.6	33.9
	11.8	43.1	49.2

was used in pilot measurements, but subsequently replaced, was nominally 404.7 nm, but leaked a small amount of the 435.2 nm line. This leak had the effect of shifting the matching wavelength to shorter wavelengths by approximately 8 nm!

The wavelength of the variable-wavelength half-field was selected at both sites by a Jobin Yvon H-10 monochromator with 0.5 mm slits, the spectral output of which could be adjusted by the subject. The spectral output of the monochromator was a triangular function of wavelength with a FWHM of 4 nm.

2.4. Corrections for Hg 'lines' of 4 nm bandwidth

Since the Hg lights had FWHMs of 4 nm, their visually-effective spectral distributions are slightly distorted by the L- and M-cone spectral sensitivities and altered by changes in prereceptor filtering. These effects can be easily calculated from the spectral waveforms of the Hg lights, and the cone fundamentals and macular and lens density spectra (Table 2, Stockman & Sharpe, 2000). We estimate, for an observer with normal macular pigment (0.35 at 460 nm) and lens (1.76 at 400 nm) densities, that the effective spectral 'lines' are shifted from 405.2 to 405.8 nm (+0.57 nm), from 436.3 to 436.5 (+0.17 nm), and from 438.2 to 438.4 nm (+0.16 nm). Individual differences in lens and macular pigment densities away from their mean values will slightly alter these wavelengths and their corresponding matches. We estimate that changes in macular density by $\pm 50\%$ from a peak density of 0.35, and changes in lens density by $\pm 25\%$ from a density of 1.76 at 400 nm, cause the changes in the effective wavelengths of the Hg 'lines' listed in Table 1.

In making these calculations, we used macular and lens optical density spectra and L- and M-cone spectral sensitivities tabulated in Stockman and Sharpe (2000) and assumed that the lights had a Gaussian spectral waveform (which is approximately correct). The changes in effective wavelength are relatively small. In terms of the change in the matching wavelength, we estimate, again using the Stockman and Sharpe fundamentals, that a change in the effective wavelength of the

405.8, 436.5 and 438.4 nm Hg lights of 0.10 nm causes a change of 0.07, 0.17 and 0.18 nm, respectively, in their longer-wavelength matching wavelengths. Thus, even at 405.8 nm, the combined effect of a 25% change in macular density and a 50% change in lens density results in an effective wavelength change of the Hg line of less than 0.20 nm (and a change in matching wavelength of less than 0.14 nm). As we will demonstrate below (see Figs. 5 and 6), compared with the differences between measured 405.8 nm matches and those predicted by the Smith and Pokorny (1975) fundamentals or by the Stockman et al. (1993) fundamentals, the changes caused by typical variations in macular and lens density are trivial.

2.5. Calibration

The radiant fluxes of test and background fields were measured in situ at the plane of the observers' pupil in Tübingen with a calibrated silicon photodiode (Model SS0-PD50-6-BNC, Gigahertz-Optics) and picoammeter (Model 486, Keithley), and in San Diego with a calibrated Graseby radiometer. We carried out our own additional calibration checks. Both instruments were cross-calibrated against each other and against another silicon photodetector (Gigahertz-Optics), which was calibrated against the German National Standard (Braunschweig). The devices agreed to within 0.01 log₁₀ unit from 400 to 700 nm.

The monochromators and interference filters were also calibrated in situ: in Tübingen with an Instrument Systems CAS-140 Spectroradiometer (Instrument Systems GmbH, Compact Array Spectrometer), and in San Diego with an EG&G Spectroradiometer. The wavelength scales of the two spectroradiometers and the Jobin-Yvon monochromators were calibrated against low pressure mercury sources.

2.6. Procedure

Matches were measured with central fixation. To avoid S-cone detection of the test fields, we carried out the matches as close as possible to contrast threshold.

We found that reliable matches could be made with the 405.8 nm test field 0.6 log unit above threshold, but that the 436.5 or 438.4 nm field had to be as high as 0.9 log unit above threshold. This difference arose because changes in the matching wavelength produced more distinct changes in color appearance near the 405.8 nm matching wavelength than near the 438.4 and 436.5 matching wavelengths.

At each background radiance, subjects were first presented with the half-field illuminated by the Hg line, and were asked to adjust its radiance until it appeared to be just at threshold. Each threshold setting was made three times and the results averaged. By the removal of calibrated 0.6 or 0.9 log unit neutral density filters, the half-field could be set to either 0.6 or 0.9 log unit above its contrast threshold for the main experiment.

Subjects were next presented with the second, variable-wavelength half-field, and were requested to adjust

its wavelength and radiance to match the half-field illuminated by the Hg line. After each match, the subject was asked to report on the quality of the match. The task was complicated, and required some practice, because changes in radiance or wavelength could both cause changes in apparent color or intensity. Thus, subjects had to readjust the radiance after any change in wavelength and vice versa. Generally, subjects found a satisfactory match to the 405.8 nm half-field at the three lowest 420 nm background radiances shown in Fig. 1, below. A satisfactory match was also found to the 436.5 or 438.4 nm half-field at the same radiances, but the match was less well defined. When there was a range of wavelengths that produced a match, subjects were asked to set the middle of the range.

Each data point shown is averaged from three settings made on each of three or four separate experimental runs.

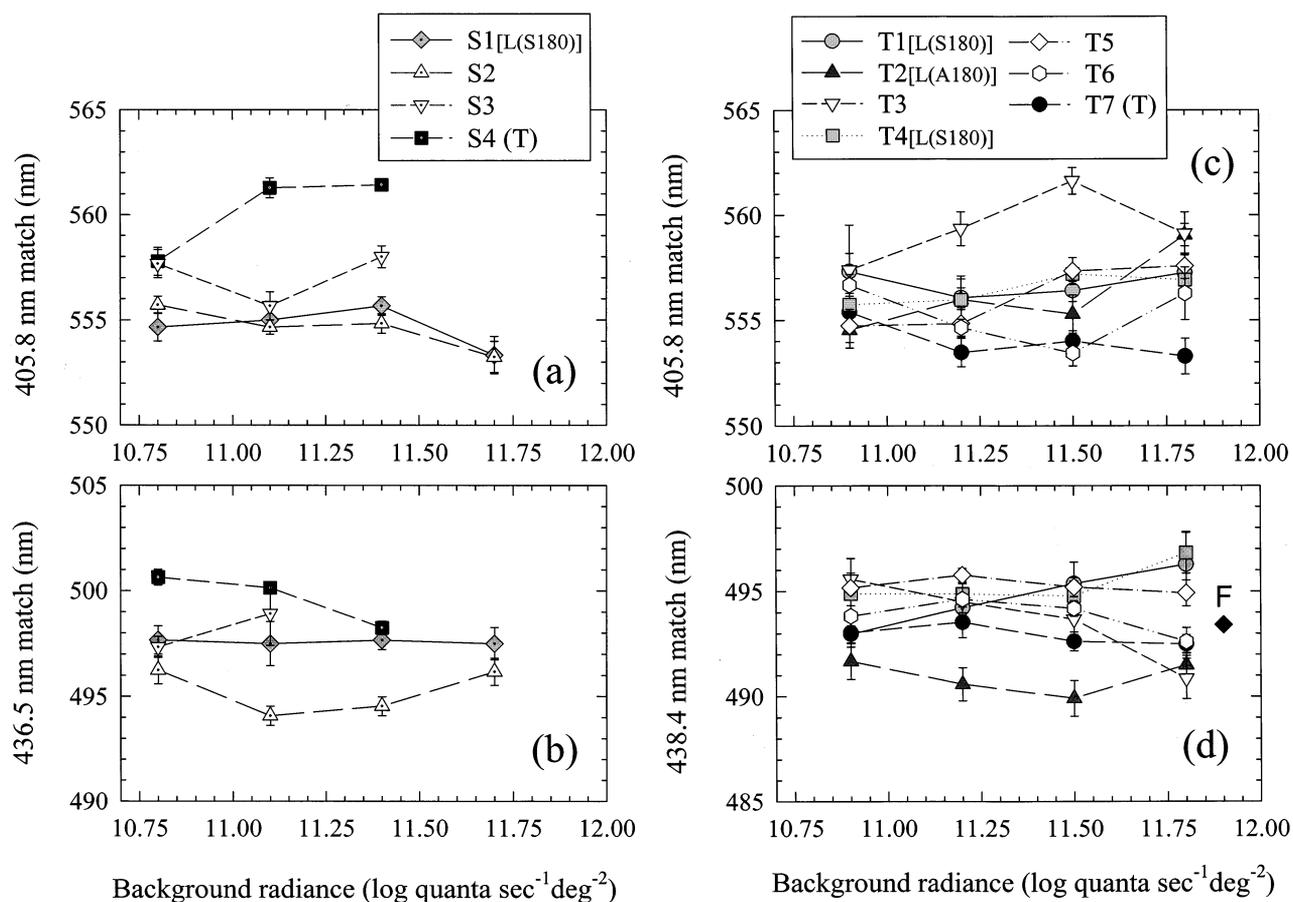


Fig. 1. Wavelengths that matched Hg lines as a function of the radiance of a 420 nm background for three color normal subjects (S1–S3, dotted symbols) and a tritanope (S4, black dotted squares) measured in San Diego (a,b) and six color normal subjects (T1–T6, undotted symbols) and a tritanope (T7, black circles) measured in Tübingen (c,d). (a) 405.8 nm matches, San Diego. (b) 436.5 nm matches, San Diego. (c) 405.8 nm matches, Tübingen. (d) 438.4 nm matches, Tübingen. The black diamond labeled 'F' shows the mean matches made at 11.9 log quanta s⁻¹ deg⁻² by seven normal subjects and a tritanope in a preliminary study conducted in Freiburg. The light gray symbols (circles, squares and dotted diamonds) indicate the subjects known to be L(S180), while the dark gray symbols (triangles) indicate the subject known to be L(A180). The error bars are ± 1 standard error.

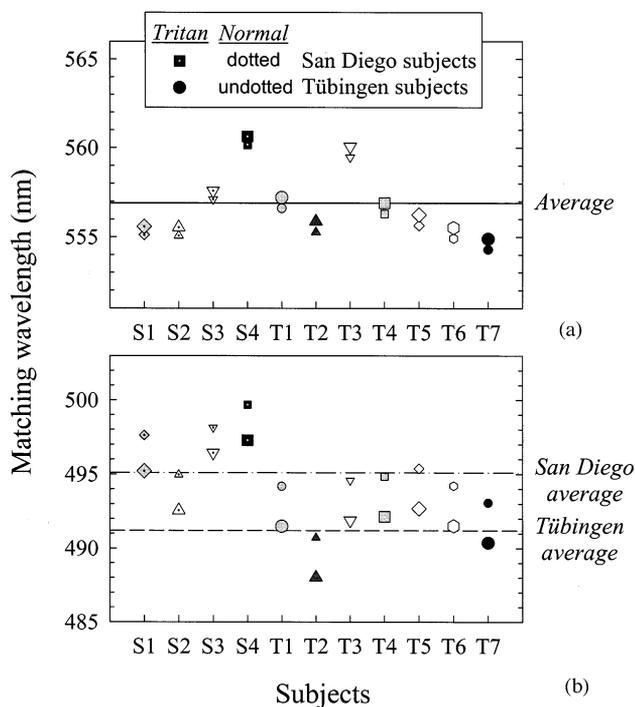


Fig. 2. Mean wavelengths (larger symbols) that matched (a) 405.8 nm (S1–S4 and T1–T7; overall average = 556.9 nm, horizontal line), and (b) 436.5 nm (S1–S4; average = 495.1 nm, horizontal dashed line) or 438.4 nm (T1–T7; average = 491.2 nm, horizontal dot–dashed line) after corrections for bleaching. Smaller symbols show mean matching wavelengths before corrections for bleaching. The symbols used for each subject are the same as in Fig. 1.

3. Results

The results of the matching experiment are shown for all 11 subjects in Fig. 1. Panels (a) and (b) show the 405.8 and 436.5 nm matches, respectively, made in San Diego, and panels (c) and (d), the 405.8 nm and 438.4 nm matches, respectively, made in Tübingen. The same symbols for the matches are used throughout the paper: dotted symbols denote San Diego subjects (S1–S4); undotted symbols, Tübingen subjects (T1–T7); filled symbols, tritanopes (S4, T7); light gray symbols, subjects known to be L(S180) (S1, T1, T4); and dark gray symbols, subjects known to be L(A180) (T2).

The lowest 420 nm background levels of 10.80 or 10.90 log quanta $s^{-1} \text{ deg}^{-2}$ were found during preliminary measurements to be the lowest at which all subjects were able to obtain good matches. In general, the matches at the three lowest levels shown in Fig. 1 were rated good by all subjects, but began to deteriorate at the fourth level for some subjects (11.70 log quanta $s^{-1} \text{ deg}^{-2}$ in San Diego, and 11.80 log quanta $s^{-1} \text{ deg}^{-2}$ in Tübingen). Also shown in Fig. 1d as the filled diamond labeled ‘F’ is the average of matches made by eight observers in a pilot study carried out in Freiburg some years earlier — on a similar apparatus to the one

later used in Tübingen and with the same filters. The older results are consistent with the newer ones.

One drawback of using intense 420 nm backgrounds is that they bleach some of the M- and L-cone photopigments. We therefore had to correct the matches made at the lowest three levels for the effects of photopigment bleaching (we did not use the matches made at the highest level in the subsequent analysis, since some subjects found them to be unsatisfactory). To make the corrections, we adjusted estimates of the M- and L-cone spectral sensitivities to lower underlying photopigment optical densities in order to mimic the effects of bleaching. We then calculated the changes in the wavelengths of the lights found to match the Hg lines that would be caused by restoring the bleached M- and L-cone photopigment optical densities to their unbleached densities. The details of the corrections, and a short discussion of the effects of bleaching are given in the Appendix (see Fig. 8, below).

Fig. 2 shows the average matches across the three lowest 420 nm radiances before (smaller symbols) and after (larger symbols) correction for photopigment bleaching for (a) the 405.8 nm test light and (b) the 436.5 and 438.4 nm test lights. On average 405.8 matches 556.9 nm (all subjects, horizontal solid line, standard error 0.57 nm); 436.5 matches 495.1 nm (San Diego subjects, horizontal dashed line; standard error 1.03 nm) and 438.4 matches 491.2 nm (Tübingen subjects, horizontal dot–dashed line; standard error 0.59 nm). The corrections for bleaching shift the mean 405.8 nm match by about 0.5 nm to longer wavelengths, and the mean 436.5 and 438.4 nm matches by about 2 nm to shorter wavelengths. Such corrections are necessary, given that bleaching almost certainly affects the matches (see Appendix A), but they inevitably add a level of uncertainty. However, the corrections are much smaller than the discrepancies between the matches and the predictions of the Smith and Pokorny (1975) cone fundamentals (see Fig. 5). Moreover, in the case of the Stockman et al. (1993) fundamentals, they actually improve the agreement between the matches and the predictions at all matching wavelengths.

As discussed above, tritanopic matches provide a useful way of testing between the rival fundamentals. Since tritanopes lack S-cones, their color matches should be predicted by any plausible M- and L-cone spectral sensitivity estimates. In Figs. 3, 5 and 6, we have replotted three different estimates of the M- and L-cone spectral sensitivities in the form of W. D. Wright (WDW) tritanopic $g(\lambda)$ coordinates by transforming them to Wright’s primaries of 480 and 650 nm and then equating them at 582.5 nm (the WDW $r(\lambda)$ coordinates are simply $1 - g(\lambda)$). Two advantages of plotting the tritanopic predictions in this way are, first, that WDW coordinates are independent of individual differences in macular and lens pigment densities, and, second, that Wright’s (1952) mean $g(\lambda)$ data for seven

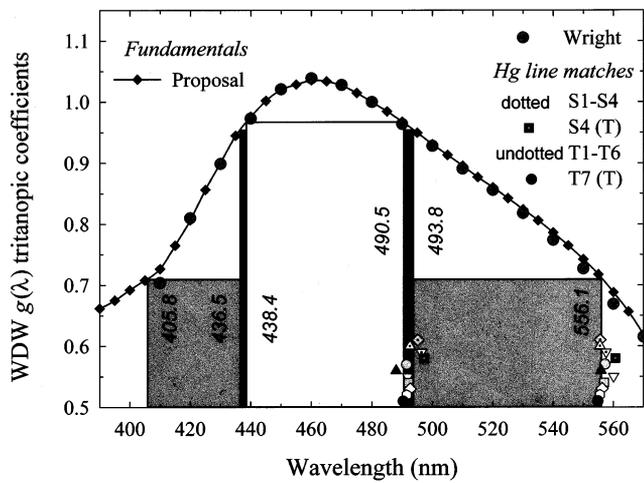


Fig. 3. Tritanopic predictions of Stockman and Sharpe (2000) M- and L-cone fundamentals (filled diamonds, continuous line), and the wavelengths found by 11 subjects to match either a 405.8 nm target light or a 436.5 nm target light or a 405.8 or a 438.4 nm target light under conditions that produce tritanopia in color normals (symbols as Figs. 1 and 2). The predicted matches are: 405.8 matches 556.1 nm; 436.5 matches 493.8 nm; and 438.4 matches 490.5 nm, as indicated by the outlines of the three large gray or white rectangles. Also shown are Wright's (1952) tritanopic $g(\lambda)$ coefficients (gray dotted circles).

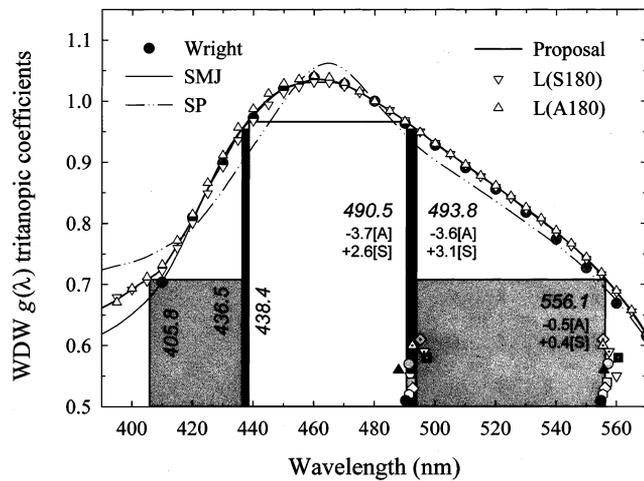


Fig. 4. Tritanopic predictions of Stockman and Sharpe (2000) M-cone fundamental and (i) the normal L-cone fundamental (continuous line, as Fig. 3), (ii) the L-cone fundamental with the underlying pigment shifted by 1.46 nm to shorter wavelengths to simulate the L(A180) spectral sensitivity (open triangles), and (iii) the L-cone fundamental shifted by 1.14 nm to longer wavelengths to simulate the L(S180) spectral sensitivity (open inverted triangles). For details, see text. The predicted matches, and the shift in the matches for L(A180) and L(S180) are: 405.8 matches 556.1 nm; -0.5 nm for L(A180) and $+0.4$ nm for L(S180); 436.5 matches 493.8 nm; -3.6 nm for L(A180) and $+3.1$ nm for L(S180); and 438.4 matches 490.5 nm; -3.7 for L(A180) and $+2.6$ nm for L(S180). Also shown are Wright's (1952) tritanopic $g(\lambda)$ coefficients (gray dotted circles); and the predictions of the Stockman and Sharpe (2000) fundamentals (thin continuous line) and Smith and Pokorny (1975) fundamentals (dot-dashed line).

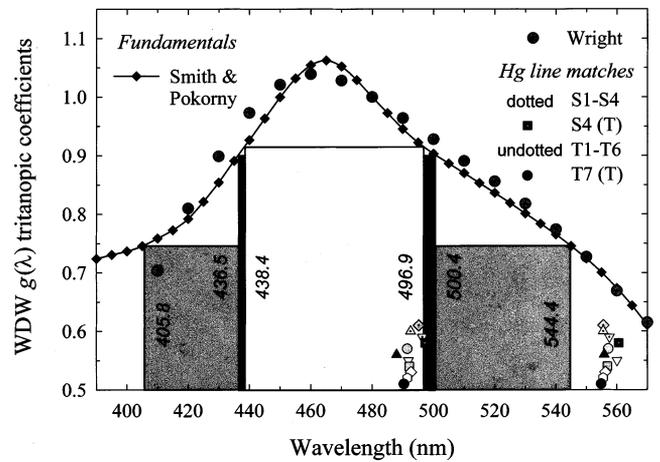


Fig. 5. Tritanopic predictions of the Smith and Pokorny (1975) M- and L-cone fundamentals (filled diamonds, continuous line); and the wavelengths found by 11 subjects to match either a 405.8 or a 436.5 nm target light or a 405.8 or a 438.4 nm target light. The predicted matches are: 405.8 matches 544.4 nm; 436.5 matches 500.4 nm; and 438.4 matches 496.9 nm, as indicated by the outlines of the three large gray or white rectangles. Other details as Fig. 3.

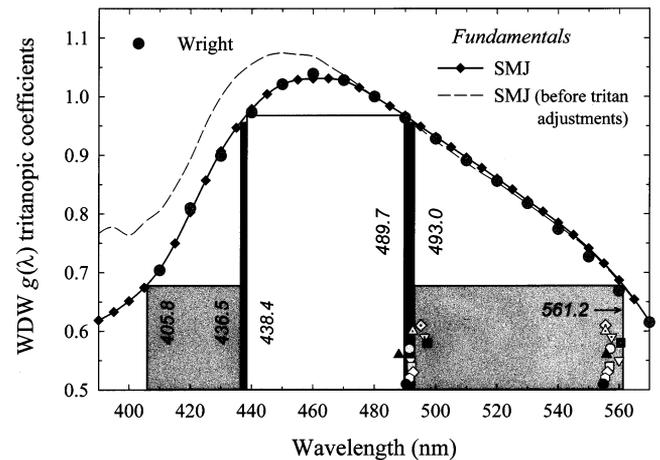


Fig. 6. Tritanopic predictions of the Stockman et al. (1993) M- and L-cone fundamentals based on the CIE 1964 10° CMFs adjusted to 2° (filled diamonds, continuous line); and the wavelength matches made by 11 subjects. The predicted matches are: 405.8 matches 561.2 nm; 436.5 matches 493.0 nm; and 438.4 matches 489.7 nm, as indicated by the outlines of the three large gray or white rectangles. The predictions of the Stockman et al. (1993) M- and L-cone fundamentals based on the Stiles (1955) 2° CMFs before their adjustment for consistency with Wright's data are indicated by the dashed line. Other details as Fig. 3.

tritanopes (gray dotted circles) are tabulated in the same form, so allowing straightforward comparisons. The tritanopic matches predicted by a $g(\lambda)$ function are any two wavelengths that have the same $g(\lambda)$ value, which, of course, also have the same relative M- and L-cone spectral sensitivities. Also shown in Figs. 3–6, using the same symbols as in Fig. 2, are the mean matches for each subject made in Tübingen and San Diego.

Fig. 3 shows the $g(\lambda)$ function (filled diamonds and continuous line) calculated from the proposed fundamentals of Stockman and Sharpe (2000). These fundamentals, which are described in the companion paper (Stockman & Sharpe, 2000), were based on spectral sensitivity measurements made throughout the visible spectrum in nine protanopes and 20 deuteranopes, the majority of which possessed just a single longer wavelength photopigment gene (Sharpe et al., 1998). The match predictions can be seen by following the outlines of the three large rectangles from 405.8, 436.5 and 438.4 nm. They are 556.1 nm for the 405.8 nm match (0.8 nm shorter than the mean of 556.9 nm); 493.8 nm for the 436.5 nm match (1.3 nm shorter than the mean of 495.1); and 490.5 nm for the 438.4 nm match (0.7 nm shorter than the mean of 491.2 nm). Importantly, each prediction lies within the range of the measured matches.

We could make small adjustments to the Stockman and Sharpe fundamentals so that they would predict the mean matches more precisely (by changing slightly the weights on the blue CMF), but first we must ensure that such adjustments are justified. An important consideration is the range of tritan matches that should be expected from the common L(A180) and L(S180) polymorphism, which causes a shift in the corneally-measured L-cone λ_{\max} of about 2.6 nm (Sharpe et al., 1998). We estimated the tritan matches predicted by the L(A180) and L(S180) polymorphisms by shifting the photopigment underlying the Stockman and Sharpe L-cone fundamental by 1.46 nm to shorter and 1.14 nm to longer wavelengths, respectively (the 1.46:1.14 ratio corresponds to the 56:44 ratio of L(S180):L(A180) found in 304 male Caucasians (see Table 1 of Stockman & Sharpe, 2000)).

To make the calculations, we adjusted the L-cone spectral sensitivity of Stockman and Sharpe (2000) to the photoreceptor level by removing the effects of the macular pigment and lens pigment, and then adjusted it to an optical density (absorbance) spectrum. A peak macular pigment density of 0.35, a lens density of 1.76 at 400 nm, and a peak photopigment optical density of 0.5 were assumed (see, for the appropriate density spectra, Stockman and Sharpe (2000)). We then shifted the spectrum by either 1.14 or 1.46 nm (at its λ_{\max}) along a log wavelength scale (Mansfield, 1985; MacNichol, 1986), and adjusted it back to a corneal spectral sensitivity, by reversing the previous calculations.

Fig. 4 shows the L(S180) (open inverted triangles) and L(A180) (open triangles) $g(\lambda)$ predictions, and the predictions for the normal L-cone function (continuous line). The change from L to L(A180) shifts the predicted 405.8, 436.5 and 438.4 nm matches by -0.5 , -3.6 and -3.7 nm, respectively, while the change from L to L(S180) shifts them by $+0.4$, $+3.1$ and $+2.6$ nm, respectively. The predicted changes are con-

sistent in direction with, but larger in size than, the differences between the measured matches for subjects known to be L(S180) (light gray symbols) and the subject known to be L(A180) (dark gray symbol), which are -3.3 nm (predicted -6.3 nm) from L(S180) to L(A180) for the 438.4 nm (Tübingen) match, and -0.7 nm (predicted -0.9 nm) from L(S180) to L(A180) for the 405.8 nm match.

Though the L(S180) versus L(A180) polymorphism has a sizable effect on the predicted 436.5 and 438.4 nm matches, it has comparatively little effect on the 405.8 nm match. A smaller effect is found at 405.8 nm because the underlying L-cone photopigment spectra have similar slopes at 405.8 nm and its matching wavelength, but dissimilar slopes at 436.5 and 438.4 nm and their matching wavelengths.

By comparing the matches for subjects of known and unknown genotype, we speculate that the majority of our subjects (S1, S3, T1, T4–T7) are L(S180), two (S2, T2) are L(A180), and two (S4, T3) may fall into another category. Calculations similar to those carried out above for the L-cone polymorphism suggest that the matches for the M(S180) genotype, relative to the mean M, should shift by $+4.0$, $+0.7$ and $+0.4$ nm for the 405.8, 436.5 and 438.4 nm lights, respectively. Since S4 and T3 both make longer-wavelength matches to 405.8 nm than the average, they may therefore have an M(S180) photopigment, the probability for which is about 7% (see above).

The L(S180) and L(A180) predictions shown in Fig. 4 (open triangles) delimit the range of matches that should be expected for subjects with a 'normal' L-cone photopigment and with the typical M(A180), M-cone photopigment. Wright's data (gray dotted circles) fall within the L(S180) to L(A180) range except at 410 nm, and between 510 and 560 nm. As we show below, these small discrepancies probably arise because Wright's data were collected using a smaller field of 1.33° diameter, so that the photopigment optical densities were higher in his experiments. For comparison, the predictions of the Smith and Pokorny (1975) fundamentals (dot-dashed line) and Stockman et al. (1993) (thin continuous line) are also shown, both of which are discussed later.

The analysis of the L-cone polymorphism and the effects of macular and lens pigment density variation (see above) suggest that the Stockman and Sharpe (2000) fundamentals are consistent with the tritan measurements. The predicted matches lie within the range of measured matches, and therefore well within the range of matches expected on the basis of individual variability. An adjustment of the Stockman and Sharpe (2000) fundamentals, so that they more precisely predict the mean tritan matches, is not warranted.

Fig. 5 shows the $g(\lambda)$ function predictions of the 2° M- and L-cone fundamentals of Smith and Pokorny

(1975), which are based on the Judd, Vos modified 2° CIE CMFs (filled diamonds and continuous line). Overall, these fundamentals predict both Wright's data and the Hg matches poorly. For the Hg lights, they predict that 405.8 nm should match 544.8 nm (12.1 nm too short); 436.5 nm should match 500.8 nm (5.7 nm too short); and 438.4 nm should match 497.2 nm (6 nm too long). Moreover, the predictions lie outside the likely range of matches suggested by the L-cone polymorphism at position 180 (see Fig. 4), and suggested by variability in the density of pre-receptor filters (see Table 1). The failure of the Smith and Pokorny fundamentals to predict tritanopic matches results primarily from their dependence on the CIE 2° CMFs, which, as others have pointed out, are inconsistent with tritanopic color matching data (Alpern, 1976; Estévez, 1979).

Fig. 6 shows the $g(\lambda)$ function predictions of the 2° M- and L-cone fundamentals of Stockman et al. (1993), which are based on the 10° CIE 1964 CMFs adjusted to 2° (filled diamonds and continuous line). Clearly, the Stockman et al. (1993) fundamentals predict Wright's data well, but that is hardly surprising since they were adjusted to be consistent with them. For the Hg lights, the Stockman et al. fundamentals predict that 405.8 nm should match 561.2 nm (4.3 nm too long); 436.5 nm should match 493.0 nm (2.1 nm too short); and 438.4 nm should match 489.7 nm (1.5 nm too short). The predictions at 436.5 and 438.4 nm are comparable to those of the Stockman and Sharpe fundamentals. The prediction at 405.8 nm, however, is much worse, lying

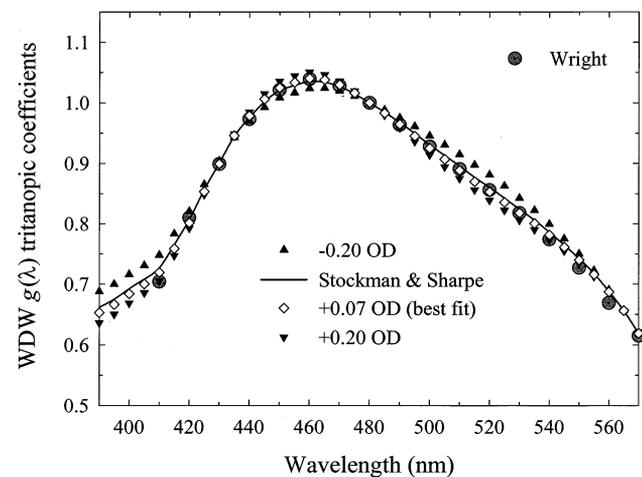


Fig. 7. Tritanopic predictions of the Stockman and Sharpe (2000) M- and L-cone fundamentals before (continuous line) and after (open diamonds) adjustments in photopigment optical density for best agreement with Wright's (1952) tritanopic $g(\lambda)$ coefficients (gray dotted circles). The best fitting change was an increase in optical density of 0.07. Changes in photopigment density of +0.20 (filled triangles) and -0.20 (filled inverted triangles) are also shown. The Stockman and Sharpe fundamentals were assumed to correspond to a photopigment optical density of 0.50.

entirely outside the range of the measured matches. An appeal to the L-cone polymorphism and to normal variation in pre-receptor filtering (see above) cannot save the Stockman et al. fundamentals at this wavelength.

The poor prediction of the tritan match at 405.8 nm light arises mainly because Stockman et al. (1993) based their fundamentals on Wright's data at 410 nm, which — for reasons we discuss below — are inconsistent with 2° data (see Fig. 7). Moreover, the small number of subjects (7) measured by Wright makes it statistically unlikely that his mean data accurately represent the normal population, so casting doubt on the generality of the adjustment made by Stockman et al.

Fig. 6 also shows the tritan predictions of the Stockman et al. (1993) fundamentals before they were 'adjusted' for consistency with Wright's (1952) tritanopic data (dashed line). In contrast to the unadjusted Stockman and Sharpe (2000) fundamentals shown in Fig. 3, the unadjusted Stockman et al. (1993) fundamentals are wrong at short-wavelengths.

4. Discussion

In conclusion, tritanopic matches provide an invaluable definition of the relative M- and L-cone spectral sensitivities at short-wavelengths. The matches to the 405.8 nm Hg spectral line are especially useful because little other information is available at such very short-wavelengths to guide derivations, and because the match is minimally affected by the L-cone polymorphism (see above). A high precision is obtained because changes as small as 0.02 log unit in the relative M- and L-cone sensitivities can result in changes of several nanometers in the predicted tritanopic matches. The measured 405.8-nm matches agree poorly with the matches predicted by the cone fundamentals of Smith and Pokorny (1975) and of Stockman et al. (1993), but well with the proposed Stockman and Sharpe (2000) fundamentals. The measured 436.5 and 438.4 nm matches agree poorly with the matches predicted by the cone fundamentals of Smith and Pokorny (1975), but well with the cone fundamentals of both Stockman et al. (1993) and Stockman and Sharpe (2000).

The mean L(A180) spectral sensitivity data from the Stockman and Sharpe (2000) study could not be directly used to predict the tritan matches to the Hg targets: although they were precise enough to estimate spectral sensitivity, they were too imprecise to predict the tritan matches, which can change by several nanometers in response to a very small change (e.g. as small as 0.02 log unit) in spectral sensitivity. Thus, we modeled the L(A180) and L(S180) matches by shifting a common underlying photopigment curve, as described above. Had we used the mean L(A180) data directly,

the differences between the predicted L(A180) and L(S180) matches would have been much larger than those found experimentally.

As noted above, Wright (1952) used a target of 1.33° diameter, rather than the standard 2° diameter target generally used for small-field color matches. Since cone outer segment length decreases with eccentricity, his data probably reflect slightly higher M- and L-cone photopigment optical densities than those measured with the larger target. We tested this by adjusting the photopigment optical densities of the Stockman and Sharpe (2000) fundamentals to best fit Wright's data (in a similar way to the adjustments for bleaching, described in detail in the Appendix, but in the opposite density direction).

Fig. 7 shows the tritanopic $g(\lambda)$ predictions of the Stockman and Sharpe (2000) fundamentals before (continuous line) and after (open diamonds) best-fitting photopigment optical density adjustments. Best agreement with Wright's (1952) tritanopic $g(\lambda)$ coefficients (gray dotted circles) was obtained with an increase in optical density of 0.07. For comparison, an increase (filled triangles) and decrease (filled inverted triangles) in optical density of 0.20 are also shown. The analysis illustrated in Fig. 7 suggests, as expected, that Wright's data are consistent with a slightly higher photopigment optical density than the density consistent with the 2° cone fundamentals.

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Appendix A. Corrections for bleaching

The 420 nm backgrounds used in these experiments bleach some of the M- and L-cone photopigments, for which a correction to the tritanopic matches must be made in order to make them applicable to non-bleaching light levels. The percentage of the M- and L-cone pigment bleached by each background can be calculated with the standard formula:

$$1 - p = \frac{I}{I + I_0}, \quad (1)$$

where p is the fraction of unbleached pigment, I is the radiance (or luminance) of the background, and I_0 is

the radiance (or luminance) of the background at which 50% of the photopigment is bleached. For the photopic (or cone) visual system and a white bleaching light, the half-bleaching constant, I_0 , is typically assumed to be 4.30 log td (Rushton & Henry, 1968). The 420 nm backgrounds that we applied, which were between 2.43 and 3.43 log td, would therefore be expected to bleach no more than 12% of the combined cone photopigments, even at the highest luminance. However, trolands underestimate luminance at short-wavelengths (e.g. Judd, 1951), and do not take into account the underlying cone spectral sensitivities. To estimate the percentage of the M- and L-cone photopigments bleached by the 420 nm background, we assumed that the half-bleaching constant (I_0) of 4.3 log td for white light is equivalent to one of 10.40 log quanta $s^{-1} \text{ deg}^{-2}$ at 555 nm, the peak of the luminosity function (i.e. we converted from trolands to quanta $s^{-1} \text{ deg}^{-2}$ at 555 nm). Since the quantal M-cone and L-cone spectral sensitivity differences between 555 and 420 nm are -1.52 and -1.59 log unit, respectively, the half-bleaching constant, I_0 , for the M-cones and for the L-cones for a 420 nm background should be 11.92 and 11.99 log quanta $s^{-1} \text{ deg}^{-2}$, respectively. The percentage bleaches, which were calculated with the use of Eq. (1) with I equal to the radiance of the 420 nm background, are tabulated in Table 2. The assumed cone spectral sensitivities are those of Stockman and Sharpe (2000).

Next, we estimated the effect of the bleaches on the tritanopic matches. For each background, we reduced the underlying photopigment optical densities of the unbleached M- and L-cone spectral sensitivities of Stockman and Sharpe (2000) according to the percentage of bleached pigment (from Table 2).

First, we corrected the M- and L-cone spectral sensitivities back to the photoreceptor level (Eq. (2)), and then adjusted them to optical density spectra (Eq. (3)). For instance, starting with the quantal spectral sensitivity of the L-cones [$L(\lambda)$], we calculated the spectral sensitivity of the L-cones at the photoreceptor level [$L_p(\lambda)$] by removing the effects of the lens pigment [$d_{\text{lens}}(\lambda)$] and the macular pigment [$d_{\text{mac}}(\lambda)$]:

$$\log[L_p(\lambda)] = \log[L(\lambda)] + d_{\text{lens}}(\lambda) + d_{\text{mac}}(\lambda), \quad (2)$$

where $d_{\text{lens}}(\lambda)$ and $d_{\text{mac}}(\lambda)$ are the (logarithmic) optical density spectra of the lens and macular tabulated in Table 1 of Stockman and Sharpe (2000). Then, we calculated the photopigment optical density of the L-cones [$L_{\text{OD}}(\lambda)$]:

$$L_{\text{OD}}(\lambda) = \frac{-\log_{10}[1 - L_p(\lambda)]}{D_{\text{unbleached}}} \quad (3)$$

We assumed, like Stockman and Sharpe (2000), a peak unbleached optical density ($D_{\text{unbleached}}$) of 0.5. $L_{\text{OD}}(\lambda)$ and $M_{\text{OD}}(\lambda)$, calculated according to Eqs. (2) and (3),

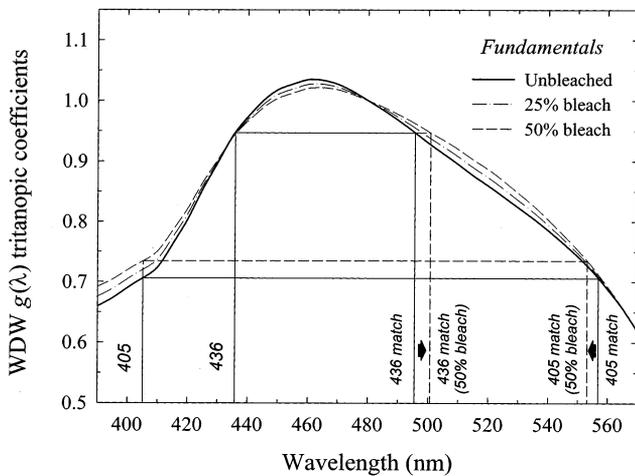


Fig. 8. Tritanopic predictions of the Stockman and Sharpe (2000) M- and L-cone fundamentals with no bleach (continuous line) and after bleaches of 25% (dot-dashed line) and 50% (dashed line) of both the M- and the L-cone photopigments. The vertical and horizontal continuous lines indicate the 405 and 436 nm tritanopic match predictions in the unbleached state; whereas the dashed lines indicate the predictions after a 50% bleach.

are tabulated in Table 1 of Stockman and Sharpe (2000).

Next, we recalculated the cone spectral sensitivities $[L'(\lambda)]$ for the lower, bleached peak photopigment optical densities (D_{bleached}), first at the retinal level, by inverting Eq. (3):

$$L'_p(\lambda) = 1 - 10^{-D_{\text{bleached}}L_{\text{OD}}(\lambda)}, \quad (4)$$

and then at the corneal level by adding back the effects of the two pre-receptoral filters:

$$\log[L'(\lambda)] = \log[L'_p(\lambda)] - d_{\text{lens}}(\lambda) - d_{\text{mac}}(\lambda) \quad (5)$$

The wavelengths that match the 405.8, 436.5 and 438.4 nm lights can then be calculated directly from each pair of bleached $[L'(\lambda)]$ and $M'(\lambda)$ cone spectral sensitivities. They are the wavelengths, in each case, at which the relative M- and L-cone spectral sensitivities are the same as at 405.8, 436.5 and 438.4 nm. In practice, we first transformed each pair of cone spectral sensitivities to Wright's primaries and then to Wright's $g(\lambda)$ chromaticity coordinates, as described above.

For each background, we compared the wavelengths that should match 405.8, 436.5 and 438.4 nm in the bleached state with the wavelengths that should match them in the unbleached state. The differences in nanometers between the two predictions are our estimate of the corrections that should be applied to the matches to take into account the effect of the bleaches. The corrections range from -1 to -4 nm for the 436.5 and 438.4 nm lights and from $+0.2$ to $+1$ nm for the 405.8 nm lights.

Two examples of the effects of bleaching are shown in Fig. 8 for 25% (dot-dashed line) and 50% (dashed

line) bleaches of both the M- and the L-cone photopigments. The vertical and horizontal lines trace out the 405 and 436 nm tritanopic match predictions [the wavelengths, λ_1 and λ_2 , at which $g(\lambda_1) = g(405)$, and $g(\lambda_2) = g(436)$] in the unbleached state (continuous lines) and following a 50% bleach (dashed lines). Bleaching shifts the 405 nm match to shorter wavelengths and the 436 nm match to longer wavelengths.

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