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COLOR VISION POLYMORPHISM AND ITS PHOTOPIGMENT BASIS IN A CALLITRICHID MONKEY (*SAGUINUS FUSCICOLLIS*)

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Abstract—The color vision of five saddle-backed tamarins (*Saguinus fuscicollis*) was studied. Behavioral tests of color discrimination and spectral sensitivity indicate that this species has a color vision polymorphism. Individual monkeys have either dichromatic or trichromatic color vision. Measurements of the spectral sensitivity of cones on this species were made on nine animals with the technique of electroretinogram (ERG) flicker photometry. Both the electrophysiological and the behavioral results suggest that there are four classes of cone pigment in this species. In addition to a short wavelength sensitive cone, apparently common to all tamarins, there are three classes of middle to long wavelength cone (λ_{max} = 545, 557 and 562 nm). Individual animals have either one or two of the latter types. The color vision variation in this species differs for males and females.

Color vision New world monkeys Polymorphism Photopigments

INTRODUCTION

The species making up *Callitrichidae*, the marmosets and tamarins, are small, agile animals whose natural habitats extend widely across South and Central America. These monkeys feed opportunistically on fruits and plants, insects and plant exudates (Kleiman, 1977; Terborgh, 1983; Sussman and Kinzey, 1984). As for many other aspects of primate behavior, success in this endeavor would seem to depend heavily on an ability to make discerning visual discriminations. As a step toward understanding the limits of this ability, we report here results from experiments on one central feature of vision in the saddle-backed tamarin (*Saguinus fuscicollis*), color vision, and provide measurements of the spectral properties of the photopigments that set the limits for this behavior.

The older literature on color vision in Callitrichid monkeys appears to be limited to a single test run on the common marmoset (*Callithrix jacchus*) by Miles (1958). The results indicated the presence of some color vision in this species, but suggested that it might be restricted in the same way as for human color defectives whose color vision is shifted in the protan direction. This outcome is an agreement with an earlier view of platyrrhine color vision, according which it was believed that all or most such species had color vision qualitatively similar to human protanopes or protanomalous trichromats (for a review see Jacobs, 1981). In recent years, however, it has become clear that this is incorrect. In particular, a series of studies on the squirrel monkey (*Saimiri sciureus*) revealed a striking color vision polymorphism with six distinct phenotypic variants (Jacobs, 1984). That polymorphism is known to result from individual variation in the complement of middle to long wavelength cone pigments (Mollon *et al.*, 1984; Bowmaker *et al.*, 1985; Bowmaker *et al.*, 1987). A principal motivation for the present research was to determine if a color vision polymorphism might be found in a Callitrichid monkey and, if so, whether its characteristics were similar to those established for the squirrel monkey, a Cebid species. We have found that, as for the squirrel monkey, the tamarin has a clear color vision polymorphism. The set of cone photopigments on which these polymorphisms depend are different for Cebid and Callitrichid species.

METHOD

Subjects

Five adult, saddle-backed tamarins (*Saguinus fuscicollis*)—three male (here identified as BO, HE, and JA) and two female (BE and TH)—

served as subjects in behavioral experiments. These animals were individually caged under standard colony conditions and tested daily. All of these animals, along with an additional four monkeys (all male), served as subjects in electrophysiological recording experiments designed to provide estimates of cone pigment spectra.

The apparatus and procedures used for both behavioral and electrophysiological experiments have been described in detail in recent publications; these are described only briefly here.

Behavioral experiments

Vision was tested in a three-alternative, forced-choice apparatus (Jacobs, 1983). The animal viewed three circular panels (2.5 cm dia) mounted along one wall of a test chamber. The discrimination is an oddity task in which two panels are illuminated identically. The animal was reinforced for selecting (by touching) the third, uniquely illuminated, panel. A correct choice resulted in the delivery of 0.1 ml of grape juice into a receptacle mounted beneath the stimulus panel. The panels were illuminated by an optical system located exterior to the test chamber. The panel receiving unique illumination was changed randomly from trial to trial. All of the functions of this test apparatus were computer controlled. Several different tests of visual capacity were run (for a more complete description see Jacobs, 1983; 1984).

(1) *Neutral point.* A first color vision test involved a search for a spectral neutral point. The light illuminating the positive panel was monochromatic (half-energy passband = 16 nm) and variable in wavelength while the light illuminating the negative panels was achromatic (4800 K) and held at a constant luminance of 3.5 cd/m^2. Over trials monkeys were required to discriminate the monochromatic from the achromatic lights as the wavelength of the monochromatic light was varied from 481 to 514 nm in steps of 3 nm. The intensity of the monochromatic light was varied in steps of 0.1 log unit over a range of $\pm$0.5 log unit around the intensity values required for a normal human trichromat to set the monochromatic and achromatic lights to equal brightness. Test trials were 2 sec in duration with an intertrial interval of 4 sec.

The five monkeys were first trained to discriminate both the 481 and 514 nm lights from the achromatic light. The intensities of the monochromatic lights were changed every 5 trials so as to cover the ranges indicated above. The subjects completed between 200 and 500 trials in each daily test session. Once the animal successfully discriminated the 481 and 514 nm test lights at all of its intensity values the intermediate wavelength range was tested. This process continued over test sessions until performance stabilized and then an additional 25 trials were run at each wavelength/intensity combination.

(2) *Increment-threshold sensitivity.* Spectral sensitivity was measured using an increment-threshold procedure. For this test the three panels were continuously and equally illuminated. On each test trial light from a monochromator was added to one of the panels. The panel to which light was added varied randomly from trial to trial. The intensity of each test wavelength was varied in steps of 0.3 log unit over a range sufficient to titrate performance from 90 to 100% correct down to chance. This usually required from 4 to 6 intensity steps. As before, the ordering of test wavelengths was randomized and individual wavelength/intensity combinations were presented in blocks of 5 trials each. In each case the subjects were trained until their performance stabilized; then an additional 25 trials were run at each intensity/wavelength combination. Threshold was defined as the test light intensity required to produce performance at an average level of 57% correct (95% confidence level).

Three different measurements of spectral sensitivity were undertaken: (a) Complete spectral sensitivity functions were determined for all five subjects on achromatic (4800 K) backgrounds having a luminance of 25 cd/m^2. The test wavelengths ranged from 440 to 650 with step intervals of 10 nm. (b) Sensitivity to 540 and 640 nm test lights was measured for three subjects on achromatic backgrounds at several different luminance levels: 5.0, 8.6, 13.7, 25.4 and 50.0 cd/m^2. (c) Spectral sensitivity functions were measured for two subjects on orange backgrounds (32 cd/m^2); these were produced with a high pass filter (50% transmission at 583 nm). The test wavelengths ranged from 440 to 660 nm in steps of 10 nm.

(3) *Rayleigh match.* This classical color matching test was adapted for use with animal subjects. The light illuminating the negative panels was yellowish (dominant wavelength = 585 nm) set to a luminance of 6.1 cd/m^2. The light illuminating the positive panel came from a color mixer from which it was possible to

obtain a continuously variable mixture of 536 and 625 nm. The color mixture was initially set by a normal trichromatic human so as to produce an exact brightness and color match to the yellow light. In the discrimination tests the luminance of the mixed light was varied over a range of 0.8 log unit in steps of 0.1 log unit around the match values recorded for the human subject.

The monkeys were first trained to discriminate pure 536 and pure 625 nm light from the yellow. Once these discriminations were mastered, the animals were tested on 16 different mixture combinations (536 + 625 nm) spanning the complete range of possible proportions. A minimum of 25 test trials were run at each mixture/luminance combination; the match locations were defined as the midpoint of the mixture range over which performance did not differ significantly from chance ($P = 0.05$).

(4) *Wavelength discrimination.* Wavelength discrimination was measured for one monkey at 11 spectral locations that spanned the spectrum from 450 to 650 nm in steps of 20 nm. At each point the negative panels were illuminated with lights which had been passed through two identical interference filters (Ditric, half-energy passband = 20 nm). The positive light came from a grating monochromator. Over trials the wavelength of the latter was varied around that of the former in steps of 5 nm so as to determine the wavelength difference between the two required for successful discrimination. At each tested wavelength the luminances of the positive panel were set to 3.4 cd/m^2.

The method used to eliminate brightness cues in this test is illustrated elsewhere (Fig. 1, Jacobs, 1984). It involved first having the animal make a brightness match between the positive light and the negative lights when the two were of identical wavelength. Then, in the wavelength discrimination test proper, the intensity of the positive light was varied around these brightness equation values over a total range of 0.8–1.1 log units in steps of 0.1 log unit. Wavelength discrimination was measured in both spectral directions at each tested wavelength; the exceptions to this were the 450 and 650 nm lights where discrimination could only be measured toward longer and shorter wavelengths respectively. As in the other behavioral tests each wavelength/intensity combination was presented in a block of 5 trials. After the animal showed no further improvement in performance at each test wavelength, an additional 25 test trials were run at each of these combinations. Wavelength discrimination was specified as the difference in wavelength between the positive and negative lights required to maintain performance at a level of 60% correct.

Electrophysiological measurements of spectral mechanisms

The spectral properties of the cone pigments of tamarins were investigated by using the technique of electroretinogram (ERG) flicker photometry (Neitz and Jacobs, 1984; Jacobs and Neitz, 1987a). In this procedure the ERG is recorded from a corneal contact ring electrode. The stimulus, presented in Maxwellian view covering 53° of the central retina, was an interleaved train of light pulses from a test and a reference light. There was an equal period of no stimulation separating successive light pulses. In this experiment the pulse rate was set to 62 Hz. The ERGs elicited by the two lights were passed through active, narrow bandpass filters. When the effectiveness of the test and reference lights were greatly different, the averaged output from the filters appeared on an oscilloscope as a roughly sinusoidal waveform. A reversal of the relative intensities of the two produced a phase reversal in the output. The experimenter made a photometric equation by adjusting the intensity of the test light until the amplitude of the ERG was minimized and its phase was intermediate.

Tamarins were anesthetized with 12 mg of Ketamine hydrochloride mixed with 0.12 mg of acepromazine maleate (IM) and by subsequent IP injection of 10 mg/kg of sodium pentobarbital. The test eye was dilated by topical application of atropine sulfate (0.04%) and Phenylephredine HCl. The animals were placed in a padded head holder. Normal body temperature was maintained with a circulating hot water heater. The recording was done in a briefly illuminated room.

Spectral sensitivity measurements were made at 10 nm steps from 440 to 640 or 650 nm. At each spectral location the intensity of the flickering monochromatic light was adjusted until the ERG it produced best nulled the response from the reference light. The reference light was achromatic (corneal radiance = 0.05 mW). The intensity settings required for equation at each test wavelength were brought to an equal quantum base and then compared to wavelength-dependent visual pigment nomograms (Dawis, 1981) using a computer routine.

The computer determined the spectral positioning (to the nearest nm) of the nomogram that gave the best fit to the array of sensitivity values.

A second experiment was run to determine if more than one middle to long wavelength cone mechanism was present in each retina. To accomplish this we searched for the presence of a chromatic adaptation effect by first establishing a flicker photometric equation between a 540 nm test light and a 630 nm reference light. These equations were made in the absence of accessory adaptation and then, alternately, in the presence of intense 540 and 630 nm adaptation. The latter were initially adjusted in intensity so that they elevated the threshold for obtaining a criterion response to the 540 nm test light by an additional 0.5 log unit.

RESULTS

Spectral neutral point

All five subjects learned to successfully discriminate the 481 and 514 nm test lights over their full range of intensities, but there was individual variation in the performances of the animals at the intermediate wavelengths. Monkeys BE and TH performed near perfectly on all of the intermediate test wavelengths at all intensity values. The other three subjects, however, were unable to successfully make some of these discriminations even after hundreds of test trials. Figure 1 summarizes this outcome. Plotted there for each of the five subjects is average performance recorded at each test wavelength over the final 25 trials. The values represent the lowest level of discrimination recorded at each wavelength. The results from the three subjects who failed the discrimination for some test wavelengths are in the top three panels. The discrimination failures of each of these monkeys were for a restricted range of intensity values, usually only 0.1 or 0.2 log units in size, and centered within the full intensity range; they were successful for those test lights both brighter and dimmer than these values. We assume the intensity values at which discrimination fails define the locations of brightness matches.

The results of this experiment indicate that there is individual variation in color vision among saddle-backed tamarins. Subjects BE and TH show no spectral neutral point in the 481–514 nm portion of the spectrum whereas each of the other three subjects does. This would suggest the former to have trichromatic color vision while the latter would conventionally be expected to be dichromatic. There is the hint of some further variation in color vision among the putative dichromats. If one takes the midpoint of the spectral range over which these animals failed the discrimination as the best estimate of the neutral point location, JA and HE have neutral point locations of 490 and 492 nm respectively, while that for BO is somewhat longer—497 nm. Differences in neutral point locations of this sort are known to be associated with variation in the type of the dichromacy in both humans (Bailey and Massof, 1974) and squirrel monkeys (Jacobs, 1984).

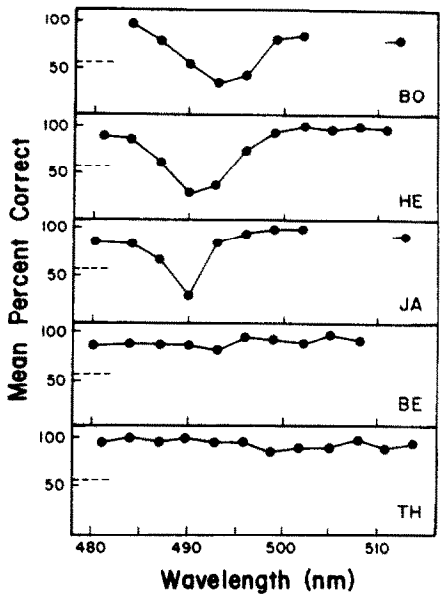

Fig. 1. Neutral point tests results for five tamarins. Each plotted point is the mean asymptotic performance level for a discrimination of monochromatic versus achromatic light. The dashed lines to the left show the performance levels required for significant discrimination ($P < 0.05$).

Increment-threshold spectral sensitivity

The full spectral sensitivity functions determined on achromatic adapting backgrounds are shown in Figs 2 and 3. Each solid point these represents a threshold determined from performance on the final 25 test trials. The lines were drawn through these points by eye. Figure 2 contains the functions for the three subjects believed on the basis of the neutral point test to be dichromats. These three are similar in form each having two locations of maximal sensitivity —in the short wavelengths at 440–450 nm and in the middle wavelengths from about 510 to 550 or 560 nm—with an intermediate region of

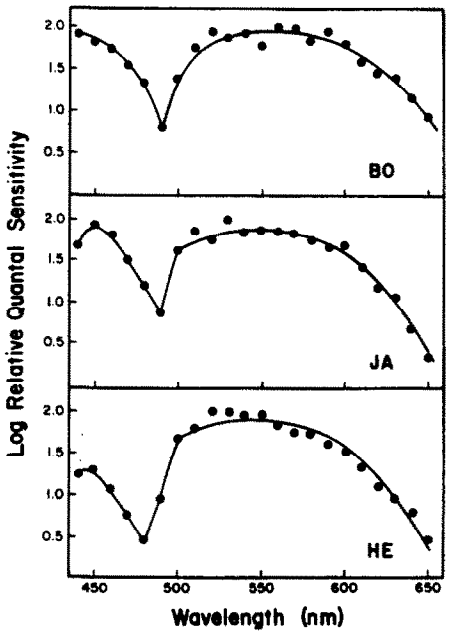

Fig. 2. Increment-threshold spectral sensitivity functions for three dichromatic tamarins. The continuous lines were drawn by eye through the data points. Background luminance = 25 cd/m^2.

greatly depressed sensitivity at 480–490 nm. Beyond that, there are some less prominent individual variations. In particular, HE is relatively less sensitive in the short wavelengths while BO shows substantially slower falloff in sensitivity toward the long wavelengths than the other two subjects. These functions are generally similar to increment-threshold spectral sensitivity functions reported for human dichromatic subjects (Zrenner, 1983).

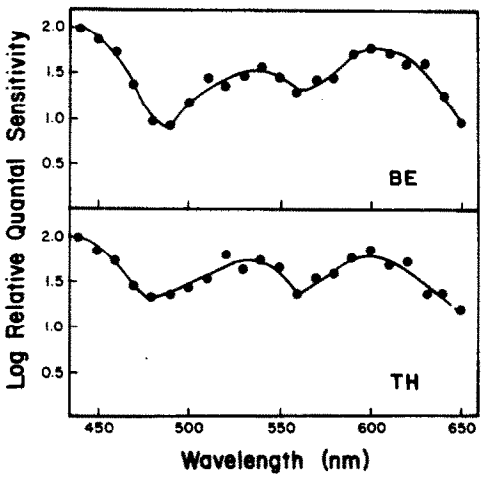

Fig. 3. Increment-threshold spectral sensitivity functions for two trichromatic tamarins. The continuous lines were drawn by eye through the data points. Background luminance = 25 cd/m^2.

The increment-threshold spectral sensitivity functions measured for subjects BE and TH are in Fig. 3. Unlike those for the three subjects of Fig. 2, these show three spectral peaks: at 440 nm, at about 540 nm, and at 580–600 nm. The two intermediate regions of lowered sensitivity are at about 480–490 nm and at about 560 nm. The major difference between the two functions is that sensitivity falls off more rapidly at the long test wavelengths for BE than for TH. In general form these functions are similar to those obtained from known trichromatic subjects (Sperling and Harwerth, 1971).

If monkeys BO, JA and HE are dichromats, as is strongly indicated by the results shown in Figs 1 and 2, then thresholds measured in the middle to long wavelengths should, to a great extent, be determined by the action of a single photopigment type. If so, spectral sensitivity measured in this region provides a prediction of the absorption spectrum of the underlying photopigment. To obtain such a prediction increment thresholds were determined for 540 and 640 nm test lights on several achromatic backgrounds. Table 1 summarizes the results of such determinations. The values entered there are log (540/640) sensitivities. It can be seen that: (a) there is only small variation in relative sensitivity for the thresholds measured for each subject across the full span of backgrounds examined implying that, indeed, only a single spectral mechanism controls this behavior, and (b) that these ratios vary on average by less than 0.1 log unit for subjects JA and HE, but that BO is relatively much more sensitive to the long test wavelength. That latter outcome is in accord with the indications from the full spectral sensitivity functions (Fig. 2).

The λ_{max} values of the photopigments best accounting for these long wavelength sensi-

Table 1. Increment thresholds determined for three tamarins

Luminance (cd/m^2)	Monkey BO	JA	HE
5.0	—	1.34	1.21
8.6	0.77	1.19	1.20
13.7	0.65	1.09	1.06
25.4	0.79	1.16	1.06
50.0	0.77	1.29	1.25
Mean	0.75	1.21	1.25
SD	0.06	0.10	0.10
λ_{max}	557	545	546

The values tabled are the log ratios of sensitivity measured for 540 and 640 nm test lights at several different adapting levels. The methods used to derive the λ_{max} estimates are detailed in the text.

tivities are also given in Table 1. These were computed to the nearest nm using a computer routine. They are based on the assumptions that the underlying photopigments have a shape given by the wavelength-dependent visual pigment nomogram appropriate for this part of the spectrum (Ebrey and Honig, 1977), that photopigment density is negligible under these test conditions, and that in this species prereceptoral absorption does not vary between 540 and 640 nm. The estimated spectral peaks of the middle-wavelength cones for JA and HE are virtually identical, 545 and 546 nm respectively, while the middle wavelength cone of BO appears to be shifted significantly further toward the long wavelengths having a peak at 557 nm.

To try to gain insight into the nature of the short wavelength mechanism in this species we measured complete spectral sensitivity functions for the two dichromats having identical middle wavelength cones (JA and HE) when they were concurrently adapted to long wavelength light. The idea was to differentially depress the sensitivity of the longer cone as much as possible while still permitting measurement of the spectral sensitivity function. Two complete functions were determined for each monkey. The actual thresholds measured were very similar for the two animals. The spectral sensitivity function plotted in Fig. 4 is thus the average for these two animals.

Figure 4 shows that with long wavelength adaptation these two dichromatic monkeys have maximum sensitivity at the shortest test wavelength, 440 nm. From that point, sensitivity falls off rapidly to about 510 nm, stays at a constant low level from 510 to 570 nm and then drops again, becoming unmeasurable beyond 600 nm. Note that by comparison to the spectral sensitivity functions measured for these two animals under achromatic adaptation (Fig. 2) there is no minimum in the sensitivity function between two regions of higher sensitivity. Since one of the two photopigments that contribute to this behavior had been established to have a peak at about 545 nm, it was possible to use that information to try to estimate what the second pigment type might be in these dichromatic animals. To do this a computer routine was employed with which an attempt was made to fit the threshold values of Fig. 4 by providing one known photopigment ($\lambda_{max} = 545$ nm) and having the computer search for a second photopigment which in combination with the 545 nm pigment provided the best fit to the total data

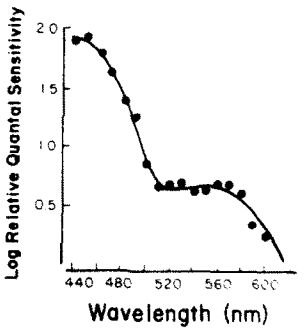

Fig. 4. Average increment-threshold spectral sensitivity functions for tamarins JA and HE. The background was an orange light. The curve drawn through the data points is that for the best fitting, summative pair of wavelength-dependent visual pigment nomograms having λ_{max} values of 545 nm (5%) and 433 nm (95%).

array. There are two uncertainties associated with this approach. One is that prereceptoral absorption is not known for this species. We made the assumption that lens absorption in this species might not be greatly different from that of the squirrel monkey and used those values (Neitz and Jacobs, 1984). A second assumption involves the nature of the interaction between the outputs from the two pigment types in this test situation. We examined both linear addition of two pigment types and linear subtraction of the two. The former provided the best fit to the data. The solid line in Fig. 4 is that for the best fitting summative pair of pigments having λ_{max} at 545 nm (5%) and 433 nm (95%). Although not providing quite such a good account of the data the linear subtraction procedure yielded nearly the same estimate for the peak of the short wavelength pigment (436 nm). Thus, although the approach is somewhat indirect, and based on the assumptions noted above, this experiment suggests that the short wavelength photopigment in the saddle-backed tamarin has peak sensitivity in the region of 433–436 nm.

Rayleigh matches

The results of Figs 1 and 3 indicate that BE and TH probably have trichromatic color vision. If so, they should be able to set dichromatic color equations in the middle and long wavelength part of the spectrum. These equations also provide information about the nature of the trichromacy; accordingly, we attempted to determine Rayleigh mayches for both of these subjects.

Both TH and BE learned to successfully discriminate the light from the color mixer at its

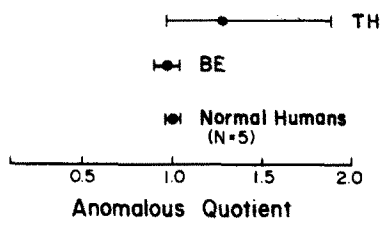

Fig. 5. Rayleigh match results for tamarins BE and TH and for five normal human trichromats. The matching ranges and midpoints are given for the two monkeys subjects. The data point for the humans is the average of their individual matches; the horizontal line encloses the total range of those matches.

pure red and pure green settings from the standard yellow light over the full range of the intensity settings of the former. Following this, they were tested over a large range of intermediate red/green mixtures with variation in intensity of the mixture as detailed above. Both animals failed to successfully discriminate some red/green mixtures from yellow. This behavior persisted even after very extensive experience in this test situation (>150 test trials at each mixture/intensity combination). These failures define the dichromatic matches, i.e. the Rayleigh equations.

Rayleigh matches for the two animals were specified as the midpoint of the range of red/green mixtures where performance dropped below chance (95% confidence) over the final 25 test trials. These are shown in Fig. 5 where the matches are expressed as anomalous quotients, i.e. the ratios of green light to red light in the mixture at the match relative to the analogous ratio obtained from normal human trichromats. For comparison, that figure also illustrates the Rayleigh matches obtained from five normal humans tested in the same situation. Two features of these results bear note: the two monkeys made different Rayleigh matches, and both on average behaved somewhat differently from the normal human trichromats. Relative to that latter standard, TH required relatively more green light to make a Rayleigh equation while the equation for BE was very slightly to the red side of that for the humans. These results imply that TH and BE do not have the same two photopigments in the middle to long wavelength portion of the spectrum, and that neither has the exact complement of the normal human trichromat. Subject TH was poorer at this task than BE. We have no explanation for this difference.

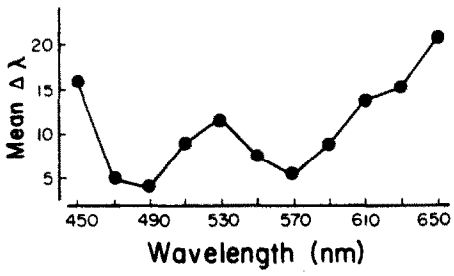

Fig. 6. Wavelength discrimination function for tamarin BE. Each plotted point is the $\Delta\lambda$ value at the indicated wavelength.

Wavelength discrimination

In an attempt to gain some indication of the acuteness of color vision in the tamarin, wavelength discrimination was measured for one of the trichromatic subjects, BE, at 11 spectral locations spaced at 20 nm steps from 450 to 650 nm. These wavelengths were tested in a randomized order. It required about 5 months of daily testing to obtain these data so at the end of that period the very first wavelength examined was re-tested to make sure that there had been no overall shift in ability over this period. None was found. The results are summarized in the form of a standard wavelength discrimination function in Fig. 6. Each point there represents a measurement of the change in wavelength ($\Delta\lambda$) required from each test wavelength in order to raise discrimination performance from chance to a level of 60% correct.

The wavelength discrimination function for BE shows two regions of most acute discrimination, at 490 and 570 nm, which are separated by a spectral region of worsened discrimination. At the location of most acute discrimination a wavelength difference of 4 nm could be discriminated. This is considerably poorer than the performance of a normal human tested in this situation, but similar to the capacity established for squirrel monkeys (Jacobs, 1984). The ability of this subject to make successful wavelength discriminations tails off at both the shortest and the longest test wavelengths. The form of this function with its two minima is characteristic of that measured for other trichromatic primate subjects, both human (Wright, 1947) and monkey (DeValois *et al.*, 1974; Jacobs, 1984). In general BE was somewhat worse at discriminating among the long wavelengths than among the shorter wavelengths. This would be consistent with the indication from the previous experiment that her color vision is slightly shifted in the protan direction relative to that of the normal human (see Wright, 1947).

Table 2. Effects of chromatic adaptation on ERG flicker photometric equations of 540 and 630 nm lights

Subject	Adaptation effect
TH	+0.08
BE	+0.04
JA	0.00
HE	0.00
BO	−0.01
805	0.00
807	0.00
812	−0.01
813	−0.01

The values, indicating the size of the effect, are in log units. Those subjects also tested in behavioral experiments are given letter designations.

Number of photopigment types contributing to the photopic ERG

The stimulus conditions used for recording flicker photometric ERGs, which included high frequency flicker and bright light adaptation, would be reasonably expected to obviate any contributions from rods and short wavelength cones. The equations so recorded should, consequently, reflect only the operation of those cones with peak absorption in the middle to long wavelengths. As a first step we sought to determine whether or not any evidence for a differential chromatic adaptation effect could be seen in these recordings. Presence of such an effect would indicate more than one cone type in middle to long wavelengths.

Table 2 summarizes the results of this test for the nine subjects. The values entered there are a measure of the average additional 540 nm light (given in log units) required to complete the 540/630 nm equation when the eye was adapted to a bright 540 nm light, relative to the amount required when the eye was similarly adapted to a 630 nm light. In the typical experiment differences in intensity of the test light of 0.03 log unit or greater led to easily detectable changes in response; the equations also had a very high reset reliability. Three of the behavioral subjects (BO, HE and JA) and all four of the additional subjects for this experiment showed no chromatic adaptation effect, i.e. their 540/630 nm equation values were independent of the nature of the chromatic adaptation. TH and BE both showed clear chromatic adaptation effects, the magnitude of the effect being consistently larger for TH than for BE. This experiment indicates TH and BE have more than one type of photopigment in the middle to long wavelengths while the other seven subjects have only one. For the five original subjects these diagnoses are consistent with results from the behavioral experiments.

Spectral sensitivity of the middle-wavelength cones in the tamarin

The complete spectral sensitivity functions determined by ERG flicker photometry for each of the seven dichromatic monkeys are given in Fig. 7. The solid circles are sensitivity values at each test wavelength while the line drawn through each function is the curve for the best fitting nomogram the peak location of which is given on each curve. These photopigment nomograms provide close fits to the sensitivity data. Three different middle wavelength pigments are identified by this procedure: four

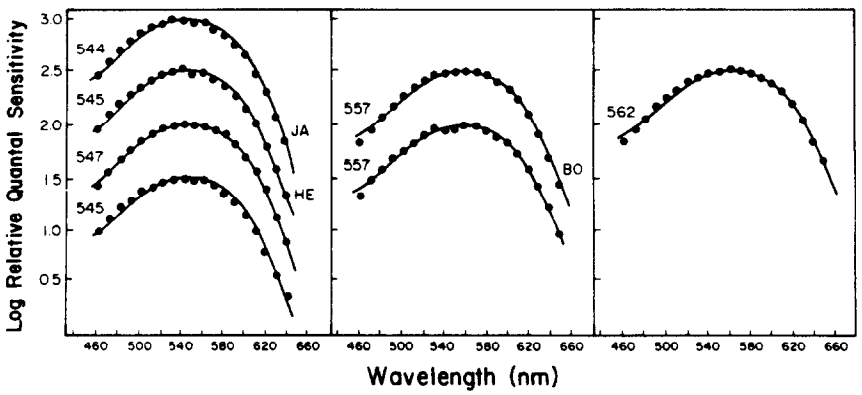

Fig. 7. Flicker photometric spectral sensitivity functions obtained from seven dichromatic tamarins. The solid circles are the sensitivity values obtained by equating the effectiveness of monochromatic test lights and an achromatic reference light. The lines are the best fitting visual pigment nomograms, the λ_{max} value of which is given for each function. The individual functions are arbitrarily positioned on the sensitivity axis. The monkeys that also served as subjects in the behavioral experiments are individually identified.

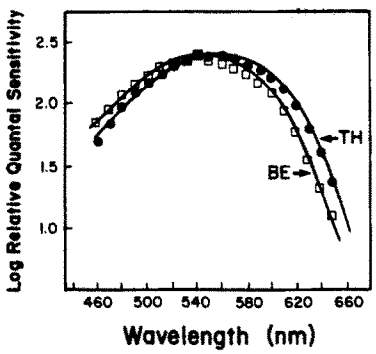

Fig. 8. Flicker photometric spectral sensitivity functions for two trichromatic tamarins. These were obtained under the same conditions as those shown in Fig. 7. The lines drawn through the data points were fit by eye. Absolute differences in sensitivity between the two subjects are preserved in this graph.

animals (left panel of Fig. 7) had λ_{max} values in the range from 543–545 nm, two (middle panel) had λ_{max} values at 557 nm, while a final animal had a photopigment having a λ_{max} at 562 nm (right panel). The three monkeys who served as behavioral subjects are separately identified in Fig. 7. As was predicted by the behavioral results, two of these (JA and HE) have the same middle wavelength photopigment; the photopigment of the third monkey (BO) is, as in the behavioral experiments, peak shifted some 12 nm toward the longer wavelengths.

The spectral sensitivity functions determined for the two trichromatic subjects, BE and TH, are shown in Fig. 8. The curves were drawn through the data points by eye. It is obvious that the spectral sensitivity curves of the middle wavelength cones in the two animals are different with the sensitivity function for TH shifted somewhat toward the long wavelengths relative to the function for BE. Each of these curves must reflect contributions from two classes of cone. The question of the identity of the cone classes in each of these trichromats is taken up below.

DISCUSSION

The results of the several behavioral tests make clear that individual saddle-backed tamarins may have any of several distinct forms of color vision including both dichromatic and trichromatic variants. This species, like ours, has a color vision polymorphism. The polymorphism differs from that of the human, however, in that the variant forms of color vision must be present at much higher frequency in the tamarin than in the human. The discovery of four different color vision phenotypes in five animals would seem even more surprising if not for the recent work on color vision and its mechanisms in the squirrel monkey. Viewed in that context, the color vision polymorphism of the saddled-backed tamarin is certainly striking, but not wholly unexpected.

As for the squirrel monkey, the color vision polymorphism of the tamarin arises from a corresponding variation in the cone pigment complement. The dichromatic animals examined in both the behavioral and electrophysiological experiments provide a clear indication of what two of the possible middle wavelength cone pigments must be. The estimates of the λ_{max} values for monkeys JA and HE were 545 and 546 nm, respectively, from the behavioral experiment; the ERG estimates were 544 and 545 nm for these same two animals. In conjunction with the further two dichromats having the same cone pigment (Fig. 7, left panel), it appears that one of the middle wavelength pigments of the tamarin has a λ_{max} value of about 545 nm. The middle wavelength cone pigment of the dichromat BO as estimated from both behavioral and electrophysiological experiments has a λ_{max} value of 557. In conjunction with results from the second dichromat of this type (Fig. 7, center panel), the second middle wavelength cone pigment of the tamarin may be taken to have a λ_{max} value of about 557 nm. The dichromatic subject whose results are illustrated in the right panel of Fig. 7 had a third type of cone pigment with a λ_{max} value of 562 nm. In sum, the measurements made on dichromatic tamarins indicate the presence of at least four different classes of cone pigment in this species: a short wavelength pigment whose peak sensitivity has not thus far been very precisely specified, but probably is in the region of 433–436 nm, and three classes of middle wavelength cone having peaks at about 545, 557 and 562 nm. The total number of tamarins available for examination was somewhat limited; consequently, we cannot be certain that all of the photopigment variation in this species has been seen. That conclusion is provisionally assumed, however, in the remainder of the discussion.

Two different types of trichromatic color vision were discovered in the tamarin. Both the behavioral and ERG experiments indicate that these two arise from different cone complements. Which pigments are present in these trichromatic monkeys? TH was consistently

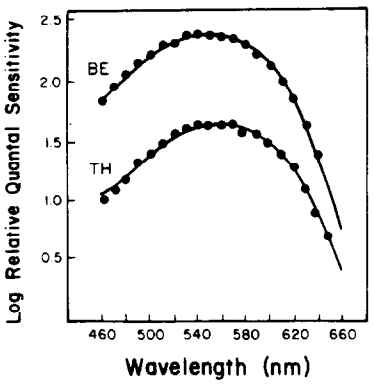

Fig. 9. Flicker photometric spectral sensitivity functions for two trichromatic tamarins. The data points are from Fig. 8. The curves are those for the best fitting linear summation of two nomogram photopigment curves. The peaks and proportions used to obtain these best fits were: BE—545 nm (65%) + 557 nm (35%); TH—545 nm (27%) + 562 nm (73%). The two functions are arbitrarily positioned on the sensitivity axis.

more sensitive to the long wavelengths than BE in all of the measurements. If, as suggested above, only three middle wavelength pigments are present in this species, this implies that BE must have the 545 and 557 nm cones while TH has the 545 and the 562 nm cones. This assignment of cone complement is consistent with the sensitivity measurements of behavioral and ERG experiments, as well as with Rayleigh matches. Since we know from earlier measurements on the squirrel monkey that the magnitude of the chromatic adaptation effect measured in the ERG experiment is related to the size of the spectral separation of the underlying cone pigments (Jacobs and Neitz, 1987a), this assignment of pigments would also be in accord with the fact that TH gave a significantly larger chromatic adaptation effect than did BE.

If these pigment assignments are correct than it ought to be possible to use their combination to account for the spectral sensitivity curves. Figure 9 shows the results of such an attempt. It is based on our previous demonstration that the ERG flicker spectral sensitivity functions of trichromatic squirrel monkeys could be well fit by linearly summing pairs of photopigment curves in various combinations (Jacobs and Neitz, 1987a). The solid circles in Fig. 9 are the ERG spectral sensitivity results of BE and TH. The curves drawn through those data points are the best fitting linear sums of two photopigment nomograms having peak sensitivities of 545 and 557 nm (for BE) and 545 and 562 nm (for TE). It can be seen that these combinations provide excellent fits to the spectral sensitivity functions.

The conclusion from this analysis of ERG and behavioral experiments is that *Saguinus fuscicollis* has three different middle wavelength cones: individual animals have either any one of these and dichromatic color vision, or a pair of these pigments and trichromatic color vision. We have found five of these six possible outcomes. The sixth photopigment/color vision combination would be a trichromacy based on the presence of 557 and 562 nm cones. Whether that combination in fact occurs is not known. If so, it will be interesting to determine the nature of color vision, if any, that can be supported by the presence of two cone types whose spectral peaks are separated by only 5 nm.

During the course of this investigation, Travis *et al.* (1985) made microspectrophotometric (MSP) measurements of the cone pigments in another Callitrichid monkey, the common marmoset (*Callithrix jacchus*). Their results appear similar to those we have detailed for the tamarin. They found individual differences in the location of the wavelength of peak sensitivity of the middle wavelength pigments. Some marmosets had only a single middle wavelength pigment (λ_{max} = either 545, 558 or 565 nm), while others had two of these pigments types (545 + 565 nm). It would appear that much the same photopigment and color vision polymorphism characterizes both marmosets and tamarins.

A unique feature of the polymorphism of color vision in the squirrel monkey is that although individual female monkeys may have either trichromatic or dichromatic color vision, all males are dichromatic (Jacobs, 1984; Jacobs and Neitz, 1985, 1987a). That conclusion is based on the examination of more than 70 animals. The sample of tamarins we have examined is much smaller, but a similar picture emerges: of seven males tested all were dichromats while both of the females were trichromatic. Examination of squirrel monkey color vision pedigrees supports a simple explanation for this gender-related difference in color vision (Jacobs and Neitz, 1987a). The explanation assumes that the three middle wavelength pigments are produced by the activity of genes at a single locus on the X-chromosome. There are three alleles at this locus; their individual actions result in the production of any one of the three cone types. With only one X-chromosone, males receive only one of these pigment types and are dichromatic. Females have the possibility of receiving different alleles on their two

X-chromosomes. In conjunction with random X-chromosome inactivation this would result in two classes of middle wavelength cone pigment and trichromatic color vision. On the present evidence it would seem likely that this genetic scheme, first suggested to characterize squirrel monkey color vision, is likely also to account for the transmission of color vision in the tamarin.

This investigation suggests that there are several ways in which color vision and its retinal mechanisms in the tamarin, a Callitrichid, are similar to those of the squirrel monkey, a Cebid. These are: (a) both species have a polymorphism of color vision, (b) these polymorphisms arise from individual variation in the presence of three types of middle wavelength cone pigment, and (c) there is a sex-related difference in colour vision in the two species that appears to have a common genetic explanation. What differs for the two species is the identity of the three middle wavelength cones. For the squirrel monkey these have average λ_{max} values of 538, 551 and 561 nm (Jacobs and Neitz, 1987a) whereas for the tamarin the values obtained from similar types of measurements are about 545, 557 and 562 nm. There is evidence that some other Cebid species have the same pigment complement as the squirrel monkey (Jacobs and Neitz, 1987b) and that, as noted above, at least one other Callitrichid species has the same pigments as the tamarin (Travis *et al.*, 1985). There are more than a dozen species of tamarins and marmosets and thus further work on this problem will be required before definitive conclusions are possible. It now appears, however, that Cebid and Callitrichid monkeys have one class of middle wavelength cone pigment in common (λ_{max} = c.562 nm), but differ in the spectral positioning of their other two pigment types.

Finally, the results of this investigation reinforce the view that there are significant differences in the color vision of platyrrhine and catarrhine monkeys (Jacobs, 1983). The former show striking color vision polymorphism reflecting variation in middle and long wavelength cone pigments; these variations are traceable to the action of multiple alleles at a single locus on the X-chromosome. On present evidence, on the other hand, catarrhine monkeys seem uniformly trichromatic reflecting the fact that the two types of middle and long wavelength cone pigment are common to all members of a species. This presumably indicates that catarrhine monkeys have two pigment loci on their X-chromosomes with only infrequent alleles (Jacobs, 1986). An intriguing commonality of platyrrhine and catarrhine color vision is that all species from both classes so far examined have a photopigment with a λ_{max} in the region of 561–565 nm (Bowmaker *et al.*, 1983; MacNichol *et al.*, 1983; Travis *et al.*, 1985; Jacobs and Neitz, 1987b; present study). This photopigment is presumably identical to that originally extracted from the chicken retina (Wald *et al.*, 1955), iodopsin, and later discovered to be present in a number of other avian species (Lythgoe, 1972). It is tempting to imagine that this ubiquitous photopigment might be the primordial cone pigment of the primates.

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