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Neutral point testing of color vision in the domestic cat

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ABSTRACT

Despite extensive study, the basic nature of feline spectral sensitivity is still unresolved. Most electrophysiological studies have demonstrated two photopic receptors within the cat's retina, one most sensitive to longer wavelengths near 560 nm and the other most sensitive to shorter wavelengths near 460 nm, providing the neuroretinal basis for dichromatic vision. A few studies, however, have detected a third photopic receptor most sensitive to medium wavelengths between 500 and 520 nm, overlapping in spectrally sensitivity with the feline scotopic receptor, that potentially could allow trichromatic vision. Indeed, one behavioral study has demonstrated trichromatic vision in cats, but a flaw within its experimental design raises the possibility that achromatic intensity cues might have allowed the accurate identification of medium wavelength targets. This study tested for a spectral neutral point in the domestic cat using a two-choice discrimination task. The positive targets were created using monochromatic light from various single wavelength light emitting diodes (LEDs) combined with a white light of variable intensity, while the negative targets were created using white light of variable intensity. Trials were performed with varying intensities of positive and negative targets, from brighter positive targets to brighter negative targets, to eliminate achromatic intensity cues. Two cats with prior experience with two-choice discrimination tasks, one male and one female, successfully discriminated monochromatic light from 456 nm to 497 nm and from 510 nm to 524 nm, but both failed to discriminate monochromatic light at 505 nm over multiple trials. These results provide strong evidence that cats are dichromatic with a neutral point near 505 nm. This neutral point is nearly identical to the neutral point of the human deuteuranope, making feline vision a more accurate a model for red-green colorblind individuals than normal trichromats.

The feline visual system has been fundamental to understanding the mammalian visual system (Hubel and Wiesel, 1962), but the electrophysiological characterization of the cat's retina has outpaced comparable behavioral analyses. Many earlier behavioral experiments underestimated the full capability of the feline visual system because of limitations in experimental technique: inadequate illumination (Clark and Clark, 2013, 2014), poor target size and contrast (Linberg et al., 2001; Loop et al., 1979), and physical and timing constraints (Clark and Clark, 2013, 2014).

Based on electrophysiological studies, the cat is likely dichromatic. From the initial identification of a single photopic receptor at 560 nm (Granit, 1942), there is compelling evidence of at least one additional photopic receptor at 460 nm (Rabin et al., 1976; Schuurmans and Zrenner, 1981). Currently, however, there is conflicting data about the possible presence of a third photopic receptor between 500 and 520 nm (Crocker et al., 1980; Guenther and Zrenner, 1993; Loop et al., 1987; Ringo et al., 1977; Wienrich and Zrenner, 1984) that overlaps in spectrum with the single scotopic receptor at 500 nm (Daw and Pearlman, 1969).

Because of this disagreement within the electrophysiological data, there are two potential outcomes for behavioral studies of cat color vision. If the cat has dichromatic vision, a neutral point should occur between 500 and 510 nm (Jacobs, 1993; Kelber et al., 2003; Neitz et al., 1989). At least four studies, however, predict that the cat has trichromatic vision, either from cone receptors with rod input at 500 nm (Nelson, 1977) or from a third, spectrally distinct photopic receptor (Crocker et al., 1980; Ringo et al., 1977; Wienrich and Zrenner, 1984). With trichromatic vision, no neutral point should be demonstrated for medium wavelength light.

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An earlier behavioral study attempted to demonstrate the lack of a neutral point in cats by comparing green targets (spectral peak near 500 nm) with purple targets that putatively created equivalent spectral peaks in the short and long wavelength spectrum (Kezeli et al., 1987). Based on the cat's ability to consistently distinguish purple from green stimuli despite many slight variations in spectral intensity, the authors concluded that cats must have trichromatic vision. The major problem with their conclusion, however, is that analysis of the spectral graphs characterizing their visual targets demonstrated that the purple targets had consistently much higher saturations of short and long wavelengths than the green targets, allowing simple intensity discrimination to generate the correct responses.

To more accurately perform neutral point testing in the domestic cat, it is important to minimize achromatic clues (Jacobs and Neitz, 1986; Neitz et al., 1989). Most modern sources of "white" light are not uniform across the spectrum, but instead balance intensity peaks in the blue spectrum against smaller intensity peaks in the red and yellow spectrum to create "white." For this study, the positive targets were created by adding monochromatic light from single wavelength light emitting diodes (LEDs) to a variably intense white LED light, allowing precise control of the overall desired illumination. The negative targets were created using an identical variably intense white LED without the additional monochromatic light. Thus, there was precise control of the overall illumination for both positive and negative targets and the discrimination task was limited to the detection of added monochromatic light. In addition, experimental conditions were adopted to optimize behavioral performance, including no dietary restrictions, unlimited time, bright photopic ambient lighting conditions, and limited trials per day guided by subject demand (Clark and Clark, 2013, 2014).

The experimental subjects, a 4-year-old female (F1) and a 9-year-old male (M1), were neutered or spayed brown tabby mixed breed domestic cats that had been previously trained on two-choice discrimination tasks. The cats were examined to ensure normal ocular anatomy and focus. The experiment was conducted according to the guidelines of the ISAE Ethical Treatment for Animals (Sherwin et al., 2003) and EU directive 2010/63/EU for animal experiments.

The experiments were performed in a windowless, 3.4 m by 3.1 m uniform white room with light beige carpeting. The room was lit by two 13-W compact fluorescent bulbs (60 W incandescent equivalent), producing a luminance of 140 lux at the target areas. Measurements were made with a Lighting Passport Colorimeter (Asensetek®, New Taipei City, Taiwan) with wavelength tolerance of ±0.5 nm and luminance tolerance of ±5%. The experimental area was 200 cm × 150 cm, enclosed by 150 cm tall barriers, and bisected by a 120 cm panel to place the choice point 80 cm from the entrance (Fig. 1A). The subjects demonstrated readiness for each experimental trial by waiting outside the testing room door, resulting in a typical yield of 4–8 trials per day.

The targets were comprised of two glass panels placed 25 cm posterior to the choice point (Fig. 1A). The panels were covered with 0.3 neutral density (ND) semi-translucent sheets to capture the projected light. Single wavelength and white LEDs were embedded in 10 cm square, 3 cm tall white boxes that were placed a variable distance behind the glass target panels. The distance was set to project the required target size and light intensity onto semi-translucent sheets, as determined by the angle of light dispersal from the LED. The white light sources were two white LED flashlights (Cree XM-L-TG White Flashlight, Cree, Durham, NC) with manual focus and manual control of beam diameter. Each flashlight was fitted with a ND 2–400 variable filter (Neewer 52 mm Fader ND 2–400 Variable Filter, Neewer®, Shenzhen, Guangdong, China) to control light intensity. The flashlights were mounted in

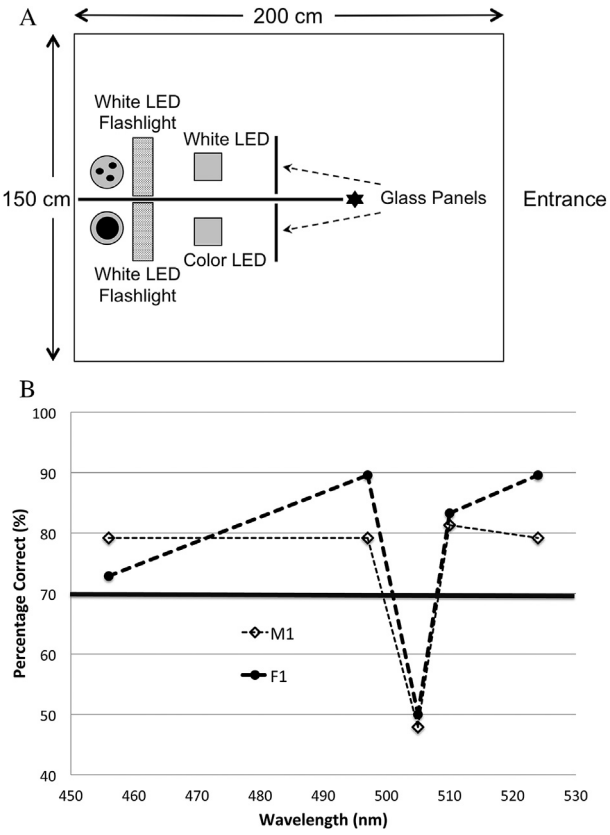

Fig. 1. A. Diagram of Experimental Area. The testing enclosure measured 200 cm × 150 cm, bisected by a 120 cm panel to place the choice point, marked with the star, 80 cm from the entrance. The targets were two glass panels covered with 0.3 neutral density semi-translucent sheets placed 25 cm posterior to the choice point. Single wavelength color (positive target) or white (negative target) LEDs were embedded in 3 cm tall white boxes (uniform grey in the figure) placed a variable distance posterior to the glass panels to project the desired circular target onto the glass panels. White LED flashlights of variable intensity and size were embedded into 25 cm tall white boxes (speckled pattern in the figure) placed 65 cm posterior to the glass panels. Those flashlights were adjusted so that the combined illumination of the white LED flashlights and embedded white or color LEDs measured at the desired intensities for both positive and negative targets. Identical treats were hidden behind the tall boxes at the end of the enclosure (grey circles) and were covered by a lid with a large central hole to provide access to the treat for the positive target and a perforated lid to allow scent but no access to the treat for the negative target. **B. Graph**. Percentage correct during two-choice discrimination testing of monochromatic light superimposed on white light versus equivalently bright white light. Both the nine-year-old male (M1) and four-year-old female (F1) scored greater than 70% correct (thick black line, binomial probability < 0.01) for all wavelengths longer and shorter than 505 nm, while at 505 nm their success rate dropped to chance levels.

20 × 25 × 8 cm rectangular boxes placed 65 cm behind the glass panels and oriented to project circles of light with a predetermined diameter onto each glass panel. The monochromatic LEDs were selected to include a short wavelength and a long wavelength target, then more thoroughly cover the 495 nm–515 nm range to encompass the expected neutral point from electrophysiological testing (Table 1).

Rewards, consisting of a few pieces of canned tuna fish (StarKist® Chunk Light Tuna, Pittsburgh, PA) mixed with solid, dry cat treats (Friskies® Party Mix Beachside Crunch, Nestle Purina, St. Louis, MO), were completely hidden in matching plastic containers behind the boxes that enclosed the flashlights. The container behind the positive target was covered with a lid with a large opening to allow access while the container behind the negative target was covered with a perforated lid to allow scent egress but no access (Fig. 1A).

Table 1
Parameters of light emitting diodes.

Light emitting diodes	Manufacturer listed spectrum	Measured spectrum	Measured 50% band	Manufacturer listed intensity	Measured intensity at target	Diameter at target
NTE30036	468 nm	456 nm	14 nm	3000 lux	160 lux	12 cm
Freedom Photon LED	–	497 nm	16 nm	5330 lux	290 lux	12 cm
Lumex SSL-LX5093UEGC	505 nm	505 nm	17 nm	2200 lux	230 lux	12 cm
AND412HG (Green)	505 nm	510 nm	15 nm	2200 lux	220 lux	9 cm
Kingbright WP7113ZGC	515 nm	524 nm	17 nm	1300 lux	400 lux	9 cm
Cree XM-L TG LED	White		–	32 000 lux	Variable	Variable

For each trial, pseudo-random Fellows sequences were used to minimize the effects of two-choice strategies (Fellows, 1967). Once the targets were adjusted to the proper luminance and size, one cat at a time was admitted into the testing room. During each trial, a human observer was positioned in the room beside the entrance, behind the cats as they faced the choice point and targets. The observer was careful to avoid influencing the cat's choice with any verbal or nonverbal cues. The trials were not timed but were terminated without recording a response if the cats did not demonstrate an interest in making a choice. If the cat proceeded towards the positive target, positive encouragement was administered, the cat was permitted to eat the treat, and the trial was recorded as a success. If the cat proceeded towards the negative target, the cat was removed from the room, both encouragement and the treat were withheld, and the trial was recorded as a failure.

Because both cats were experienced in two-choice discrimination tasks, they immediately began experimental trials without additional training. The first phase tested for short wavelength discrimination using as the positive target the 456 nm LED superimposed onto the white LED for a total combined illuminance of 350 lux and target size on the glass panel of 12 cm in diameter (Table 1). The initial negative target was a 0.7 log unit reduction (70 lux) white LED projection on the glass target of the same size. Consistent with our earlier studies, this phase was considered successfully passed if the positive target was chosen at a binomial probability of non-random occurrence at the 0.01 level: 7 consecutive correct choices, 9 correct out of 10 trials, 11 correct out of 13 trials, 13 correct out of 16 trials, or 16 correct out of 20 trials (Clark and Clark, 2013, 2014). The negative target illumination was then increased in 0.1 log units until its luminance was brighter than the positive target by 0.2 log units (480 lux). If the cat successfully discriminated the positive target at all combinations of light intensity, a verification step was performed using 48 trials at equal intensity (Jacobs, 1984), with the passing success rate set at 70% (34 correct choices).

The second phase tested for long wavelength discrimination using a consistent positive target of the 524 nm LED superimposed on the white LED for a total combined illuminance of 500 lux and target size on the glass panel of 9 cm in diameter (Table 1). The range of negative target luminance was from −0.3 log units to +0.2 log units (250 lux–680 lux). If the cat successfully discriminated all combinations of light intensities, a verification phase using 48 consecutive trials followed.

Once accurate wavelength discrimination was confirmed for short and long wavelengths, the third phase tested wavelengths to cover the predicted neutral point using similar criteria. The positive targets for this phase were LEDS of 510 nm at 350 lux and 9 cm target size, 505 nm at 350 lux and 12 cm target size, and 497 nm at 500 lux and 12 cm target size (Table 1). Finally, to test for brightness discrimination ability alone without added monochromatic light, trials were conducted using a consistent 350 lux white LED as the positive target compared with an identical white LED with intensity set to −0.5 to −0.1 log units as the negative target.

M1 took 44 trials to successfully discriminate 456 nm at log −0.7, then gradually improved his performance to resolve all combinations of light intensity utilizing far fewer trials. In the verification stage, he successfully chose the positive target 38 out of 48 trials (79.2%) at equal intensity (Fig. 1B). F1 took only 21 trials to pass 456 nm at log −0.7, then successfully resolved all combinations of light intensity and passed the verification phase with 35 out of 48 trials (72.9%) at equal intensity (Fig. 1B).

Both animals resolved the longer wavelength test with far fewer trials, passing the first comparison (log −0.3) utilizing the minimum of 7 trials. They both resolved all combinations of light intensity and the verification stage, with M1 passing the verification phase with 38 out of 48 correct (79.2%) and F1 passing the verification phase with 43 out of 48 correct (89.6%) (Fig. 1B).

Similarly, for the 510 nm LED target, both cats passed all combinations of light intensity and completed the verification phase with more than 80% accuracy. For the 505 nm LED, however, both cats passed the log −0.2 comparison, but neither could successfully discriminate the log −0.1 and equal intensity negative targets. M1 failed the log −0.1 (24 out of 48 successful responses, 50%) and the log 0.0 (equal intensity, 23 out of 48 successful responses, 47.9%), then also failed the verification phase with an identical 47.9% success rate. F1 failed the log −0.1 (24 out of 48 successful responses, 50%) and the log 0.0 (equal intensity, 25 out of 48 successful responses, 52.1%), then failed the verification phase with a 50% success rate. Finally, for the 497 nm LED, both cats were able to pass all combinations of light intensities and also completed the verification phase, M1 with a 79.2% success rate and F1 with a 93.8% success rate (Fig. 1B).

In the final phase, using brightness only as the discriminating factor, both cats were able to resolve the positive target successfully to a minimum 0.2 log unit difference (280 lux compared with 350 lux). They were unable, however, to use brightness alone to discriminate less than the 0.2 log unit differences between the positive and negative targets, identical to their performance for 505 nm wavelength target.

These results provide strong evidence that cats have dichromatic spectral sensitivity with a neutral point around 505 nm. This neutral point matches those found in the electrophysiological studies that detected only long (550–560 nm) wavelength and short (450–460 nm) wavelength photopic receptors (Daw and Pearlman, 1969; Granit, 1942; Rabin et al., 1976; Schuurmans and Zrenner, 1981). Both subjects were consistently able to discriminate longer and shorter wavelengths but were unable to resolve the 505 nm LED target over numerous trials with relatively equivalent brightness. This neutral point is almost identical to that of the human deuteuranope, also centered at 505 nm (Neitz et al., 1989). When considering the cat as a model for human vision, feline vision should be considered more similar to a red-green colorblind individual than a normal trichromat.

This study had too few trials away from the expected neutral point to provide useful information on relative spectral sensitivities of short and long wavelength cones. A greater density of long

wavelength cones in the feline retina might imply greater sensitivity to long wavelength monochromatic light, as shown by the cats' more rapid progression when detecting longer versus shorter wavelength targets. Further investigation of sensitivity at longer and shorter wavelengths is required, however, to more accurately describe the spectral sensitivity of the feline visual system, especially since “white” LED lights have relatively more intense blue saturation, possibly making discrimination between the white and blue LEDs more difficult. This difference, rather than a difference in cone saturation, might have accounted for the decreased accuracy of short wavelength discrimination.

This study has other important limitations, including testing only a few subjects using a limited number of trials. The use of two trained cats, while a small sample, is consistent with the number of subjects used in previous feline behavioral studies. Additionally, while conducting fewer trials increased the weight of each trial within the results, the rapid progression through each phase was crucial to maintaining motivation through the important verification phases. By the end of all phases, each cat discriminated the positive target at an equivalent illumination to the negative target at least twice with a binomial probability less than 0.01, yielding an overall level of statistical significance less than 10^{-4} for all wavelengths other than 505 nm.

In conclusion, these results strongly suggest that the feline visual system is indeed dichromatic with a clearly defined neutral point. The domestic cat is capable of making chromatic discriminations between light stimuli with predominantly longer or shorter wavelengths than 505 nm, but is incapable of discriminating wavelengths near 505 nm from equally intense white light.

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References

Clark, D.L., Clark, R.A., 2013. The effects of time, luminance, and high contrast targets: revisiting grating acuity in the domestic cat. Exp. Eye Res. 116, 75–78.
Clark, D.L., Clark, R.A., 2014. Optotype recognition visual acuity in the domestic cat. Res. Rev. Biosci. 8, 407–413.
Crocker, R.A., Ringo, J., Wolbarsht, M.L., Wagner, H.G., 1980. Cone contributions to cat retinal ganglion cell receptive fields. J. Gen. Physiol. 76, 763–785.
Daw, N.W., Pearlman, A.L., 1969. Cat colour vision: one cone proces or several. J. Physiol. 201.
Fellows, B., 1967. Chance stimulus sequences for discrimination tasks. Psychol. Bull. 67, 87–92.
Granit, R., 1942. The spectral properties of the visual receptor of the cat. Acta Physiol. Scand. 5, 219–229.
Guenther, E., Zrenner, E., 1993. The spectral sensitivity of dark- and light-adapted cat retinal ganglion cells. J. Neurosci. 13, 1543–1550.
Hubel, D., Wiesel, T., 1962. Receptive fields, binocular interaction and functional architecture in the cat's visual cortex. J. Physiol. 160, 106–154.
Jacobs, G.H., 1984. Within-species variations in visual capacity among squirrel monkeys (Saimiri sciureus): color vision. Vis. Res. 24, 1267–1277.
Jacobs, G.H., 1993. The distribution and nature of colour vision among mammals. Biol. Rev. 68, 412–471.
Jacobs, G.H., Neitz, J., 1986. Spectral mechanisms and color vision in the trew shrew (Tupaia belangeri). Vis. Res. 26, 291–298.
Kelber, A., Vorobyev, M., Osorio, D., 2003. Animal colour vision - behavioral tests and physiological concepts. Biol. Rev. 78, 81–118.
Kezeli, A.R., Maximov, V.V., Lomashvilli, N.J., Khomeriki, M.S., Tshkvediani, N.G., 1987. Participation of the green-sensitive cone mechanism of the cat retina in colour discrimination. Sechenov Physiol. J. USSR 73, 883–889 (in Russian).
Linberg, K.A., Lewis, G.P., Shaaw, C., Rex, T.S., Fisher, S.K., 2001. Distribution of S- and M-cones in normal and experimentally detached cat retina. J. Comp. Neur 430, 343–356.
Loop, M.S., Bruce, L.L., Petuchowski, S., 1979. Cat color vision: the effect of stimulus size, shape, and viewing distance. Vis. Res. 19, 507–513.
Loop, M.S., Millican, C.L., Thomas, S.R., 1987. Photopic spectral sensitivity of the cat. J. Physiol. 382, 537–553.
Neitz, J., Geist, T., Jacobs, G.H., 1989. Color vision in the dog. Vis. Neurosci. 3, 119–125.
Nelson, R., 1977. Cat cones have rod input: a comparison of the response properties of cones and horizontal cells bodies in the retina of the cat. J. Comp. Neur 172, 109–136.
Rabin, A.R., Mehaffey, L., Berson, E.L., 1976. Blue cone function in the retina of the cat. Vis. Res. 16, 799–801.
Ringo, J., Wolbarsht, M.L., Wagner, H.G., Crocker, R., Amthor, F., 1977. Trichromatic vision in the cat. Science 198, 753–755.
Schuurmans, R.P., Zrenner, E., 1981. Responses of the blue sensitive cone system from the visual cortex and the arterially perfused eye in cat and monkey. Vis. Res. 21, 1611–1615.
Sherwin, C., Christiansen, S., Duncan, I., Erhard, H., Lay, D., Mench, J., O'Connor, C., Petherick, J., 2003. Guidelines for the ethical use of animals in applied ethology studies. Appl. Anim. Behav. Sci. 81, 291–305.
Wienrich, M., Zrenner, E., 1984. Cone mechanisms and their colour-opponent interaction in monkey and cat. Ophthalmic Res. 16, 40–47.