A Thesis Presented to

The Faculty of Alfred University

A Horse's World, How Colorful is it?

Julie Barr

In Partial Fulfillment of

the Requirements for

The Alfred University Honors Program

May 7th, 2021

Under the Supervision of:

Chair: Dr. Beth Johnson

Committee Members:

Dr. Danielle Gagne

Margaret Shank

Honors Foreword:

Introduction

For centuries, horses have been domesticated for multipurpose use for work on farms to extracurricular riding, and competing. Having such extensive use begs the question of how does a horse see their world? Traditionally, it was believed that horses were colorblind and incapable of thought (Hanggi, 2005). For humans, the primary experiences that we have been through the eyes of the individual, and it can be challenging to see the world through a different lens. The basis of human vision is unique mainly to primates, all of which possess what is known as trichromatic vision (Carrol, Murphy, Neitz M, Hoeve, & Neitz J, 2001). This trichromatic vision is representative of the three cones that are present in humans and other primates. These cones are specialized cells found in the eye that sense color. In addition to the cones, rods also help the world be interpreted. These rods are also specialized cells in the eye, however, the rods function to sense light, and are responsible for vision in environments with low light (Carrol et al. 2001).

When comparing these modalities of vision in humans to those in horses, there is one very large difference. This is that horses only possess two cones as opposed to human's three cones (Smith & Goldman, 1999). Although they knew that horses only had two cones, the question remained of what colors they were able to perceive. A study done by Carrol et al. (2001) exposed horse eyes to different wavelengths of light and measured their pupillary responses. It was found that the color we perceive as green, or 539 nm wavelength was a horse's maximum wavelength sensitivity (Carrol et al., 2001). This means that looking both above and below this wavelength, a horse's sensitivity to those wavelengths decreases. The results of the

study done by Carrol et al. (2001) found that horses have both a short cone and a medium/long cone, and their spectrum of visible colors is shown in Figure 1.

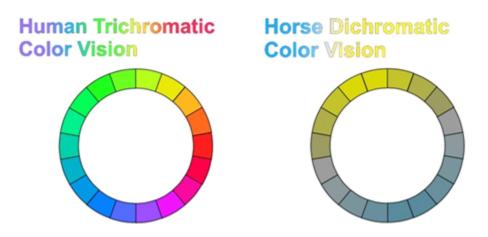


Figure 1. Compares the human trichromatic color vision to the hypothesized colors perceived by the dichromatic horse

These studies showed what range of colors that a horse was able to respond to, but additional behavioral studies are required to determine how a horse can perceive the varying colors. To do this, extensive training is required. One of the largest methods of training horses is using conditioning (Hanggi, 2005). There are two main classes of conditioning, operant and classical. The decision to choose between these two classes was made based on a study done by Lansade and Calandreau (2018) that showed that the use of a conditioned reinforcer alone was ineffective in maintaining a previously trained task. A conditioned reinforcer is a stimulus that initially elicited no response by the participant but is paired with something rewarding to make the initial stimulus rewarding. This makes the previously indifferent stimulus rewarding to the participant. Operant conditioning, on the other hand, makes use of a primary reinforcer, which is something that is reinforcing to behavior in of themselves and needs no additional associations to

be reinforcing. Additional studies done by Martinex de Andino and McDonnel (2017) showed that even from a young age that foals were able to effectively learn and retain a task learned through operant conditioning.

Based on the findings from previous studies, I will be using operant conditioning with carrots as primary reinforcers to teach my horse, Elsa, to distinguish green from blue, red, and purple. I hypothesize that Elsa will be unable to accurately discriminate between green and red but will be able to accurately discriminate between green and gray, blue, and purple.

Methods

The horse utilized for this experiment is the researcher's horse, Elsa. She is a 7-year-old thoroughbred mare who has been under consistent under-saddle work for a year and a half.

The colors chosen for this experiment were chosen by first using a hue, saturation, and luminance (HSL) picker to select a mid-tone green. From there, blue, gray, red, and purple were all designed using the following luminance equation to maintain the same perceived brightness across all colors (Shanley, 2020). The R represents the value of red, G represents the value of green, and B represents the value of blue in the color. The values for each color as well as their six-digit name, or HEX code are shown below in Table 1.

These colors were all printed onto circles and were placed on a whiteboard with nine clear dividers to hold the circle. These nine positions on the board were spaced evenly laterally and longitudinally and were hung on the wall of a 12' x 12' wooden stall. A training regimen was also implemented for both the initial training period and testing period and can be found fully on page 14. A barrier was also introduced during the later phases of initial training and the

testing period to prevent social cues from the researcher to influence Elsa's decisions on which circle to choose.

Table 1. The values of R (red), G (green), and	B (blue) as well as the six-digit name, or HEX
code, for each color.	

Color	Green	Blue	Red	Purple
R	32	56	255	212
G	196	140	70	61
В	30	255	85	255
Hex	20C41E	388CFF	FF4853	D43DFF

Results

The chi-square tests of independence showed that there was a significant difference between the observed and expected number of touches for both gray and blue. There was not, however, a significant difference between the observed and expected number of touches for both red and purple. This means that Elsa was not able to accurately distinguish red and purple from the green but was able to accurately distinguish gray and blue from green.

Discussion

Throughout this experiment, Elsa was able to accurately identify the task at hand at an astonishing speed. Elsa was able to distinguish the green and gray ratio that was much higher than seen in any of the other test colors. I believe that this was due in part to the use of gray in the initial training phases. Since there had already been documented abilities of horses to distinguish between gray and other colors (Macuda & Timney, 1999), the gray circle was utilized as a control to expose Elsa to the testing conditions before they were presented for the test colors.

Even during the calibration periods for each test color, it became fairly evident which colors she could distinguish between. There was uncertainty for the first few minutes with each test color as she was learning which one she was going to be reinforced for picking. For the blue, it became very clear that she was able to accurately pick out the green from the blue even as it was rotated into different positions. The experience that Elsa had with the purple was also similar to the blue, but there was a higher degree of uncertainty shown even during the testing period. This resulted in a chi-square value that was not significant, although it was fairly close to being significant. Further trials with purple would help to clarify if horses can accurately distinguish purple from green but based on my observations I believe that horses can discriminate green from purple, just not as accurately.

These observations from the blue and purple trials were unlike the occurrences during the trial with red. Even during the calibration period, Elsa would touch the red circle three or four times in a row without reinforcement before going to the green circle. In the other test colors, once Elsa had a few minutes of practice, she was indicating that she could clearly distinguish the colors. By the end of the calibration period with the red, however, Elsa was still not consistently

choosing the green circle over the red. This is further supported by the chi-square results, where the observed values were the same as the expected values, showing that she was unable to accurately discriminate between red and green.

Overall, the hypothesis that Elsa would be able to accurately distinguish green from gray, purple, and blue but not red was partially supported. It is statistically supported that Elsa could distinguish blue and gray from green, but the hypothesis that Elsa could distinguish purple from green was not supported. It was also supported that Elsa was unable to distinguish green and red, but further testing could be utilized to further validate these results. The following link and QR code leads to a video created throughout the training and testing process:

https://youtu.be/drXU2Su7Tw4.



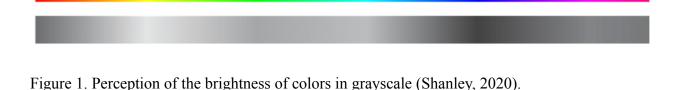
Introduction

Although horses have been used in a domestic setting for hundreds of years, their vision is still something that is being fully understood. There have been established traditional misconceptions that horses are colorblind, the only function as conditioned-response animals, and have poor acuity and depth perception (Hanggi, 2005). To counter the idea that horses are thoughtless and colorblind, vast research has been done into the ability of a horse to discriminate color as well as their cognitive capabilities.

While training, a question that frequently arises can be whether to utilize operant or classical conditioning for the task at hand. Lansade and Calandreau (2018) demonstrated that the use of a conditioned reinforcer alone was ineffective when maintaining an operant conditioned task. A conditioned reinforcer is when an association has been formed with a previously unconditioned stimulus and becomes associated with something that rewards the participant. Even from a young age, foals can efficiently learn and retain an operant task (Martinez de Andino & McDonnel, 2017). Through the extension of a foal's ability to learn through operant conditioning, an older horse is also more than capable to utilize operant learning of even complex tasks. Primary reinforcers themselves are reinforcing to begin with and do not need any additional association to become rewarding. Based on the higher efficacy of operant conditioning over classical conditioning for target training in foals as well as increased efficacy with a primary reinforcer, I chose to use operant conditioning with carrots acting as a primary reinforcer for my experiment.

There has been a vast expansion on the knowledge of color perception in horses that has shifted the initial belief that they are colorblind (Hanggi, 2005). Since these initial beliefs, Smith

and Goldman (1999) used a discrimination task between a test color and gray which suggested that horses' vision is at least dichromatic. During this test, varying intensities of gray were used, which showed that the brightness was irrelevant (Smith & Goldman, 1999). Although the brightness of the color may have been the same, the overall perceived brightness, or luminance, still varies between colors. As shown in Figure 1., each color will have different luminance even at the same brightness (Shanley, 2020).



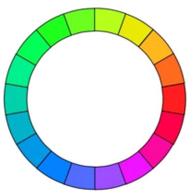
The luminance of different colors can be matched using mathematics so that if the hue was taken away from the color, all of the colors would be equivalent on the grayscale. By maintaining the same luminance across all colors, the discrimination of colors cannot be made based on the different brightnesses of each color. Additional behavioral studies done by Macuda and Timney (1999) showed that horses were able to discriminate between differing luminances, but had difficulty discriminating green and yellow from gray. This makes holding luminance as a constant across all colors presented crucial to the integrity of the study.

The eye contains two main types of specialized cells, rods and cones. The cones are responsible for collecting information from the environment regarding colors, whereas the rods are responsible for collecting information on light in the environment (Carrol, Murphy, Neitz M, Hoeve, & Neitz J, 2001). These rods and cones are collectively responsible for how the world is perceived through the eyes of the organism that they are in. The vision which humans have is trichromatic, meaning there are three cones present, but it has been indicated that horses are

dichromatic, or only have two cones (Carrol et al., 2001). These different cones are responsible for detecting light at varying wavelengths, and the color that is perceived is based on the information from these cones. A cone's length corresponds with the size of the wavelength it responds to. Shorter wavelengths correspond to blue, medium wavelengths corresponding with green, and long cones corresponding to red.

Using spectral sensitivity measurements taken through pupillary responses, it was determined that horses' maximum wavelength sensitivity was at 539 nm (Carrol et al., 2001). This maximum corresponds with the point at which the horse had the highest sensitivity to the color presented, and past this wavelength, there was decreased spectral sensitivity. The 539 nm wavelength of light corresponds with the color that humans would perceive as green. The results from the spectral sensitivity demonstrated that horses have both a short and a medium/long cone (Carroll et al., 2001). Since horses have a combined medium/long cone and a maximum wavelength falling in the green range, the higher wavelengths like red are not going to be perceived as what a person would see as red. To help visualize how horses would perceive the colors, Figure 2. Shows a rendition of a horse's color vision as depicted by Carrol et al. (2001).

Human Trichromatic Color Vision



Horse Dichromatic Color Vision

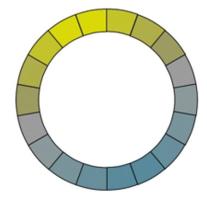


Figure 2. Compares the human trichromatic color vision to the hypothesized colors perceived by the dichromatic horse.

Although behavioral experiments on horse color vision demonstrated their ability to discriminate between test colors and gray, the discrimination amongst varying colors has not been studied as in-depth. With this in mind, the goal of my experiment was to use operant conditioning to train my horse, Elsa, to discriminate between the optimal spectral sensitivity color, green, and three test colors, blue, red, and purple. I hypothesize that Elsa will be able to accurately discriminate between gray, blue, and purple but not be able to accurately discriminate between green and red.

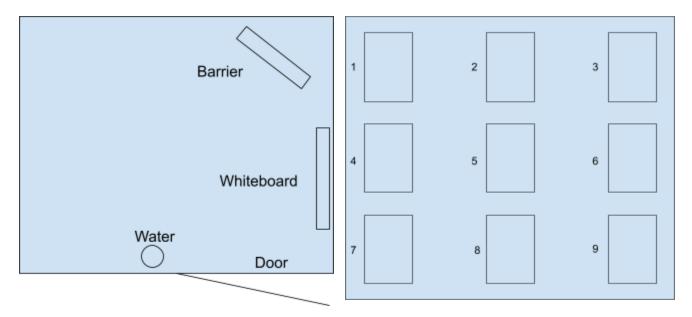
Method

Participant

The participant is a seven-year-old thoroughbred mare, who has been in training for a year and a half under saddle. She has been selected as she is owned by the researcher, and has the consistency of external factors like riding time in addition to the experimental training.

Materials

The stall used for this demonstration was a 12' x 12' wooden box stall with metal bars along the front. Within this stall, there is a water bucket, a whiteboard, and a barrier. The orientation of the stall is shown in Figure 3. and has a fluorescent light directly above the



whiteboard to minimize glare. The whiteboard was set up and labeled as shown with nine clear

Figure 3. Shows the orientation of the stall setup as well as the setup of the whiteboard.

dividers to act as holders for the position of each circle (Figure 1). Position five on the whiteboard acts as the initial calibration position for each training session.

The colors that were used in this experiment are gray, green, red, blue, and purple. Luminance is defined as the perceived brightness of a color. If the hue, or value of color, is taken away each one of the colors would be the same shade of gray. To ensure that the luminance is constant across each color, the following equation is used to calculate the perceived luminance. Luminance = $\frac{(299*R) + (587*G) + (114*B)}{1000}$ with the R being the value of red, G being the value of green, and B being the value of blue (Shanley, 2020). Each color was chosen using a hue, saturation, and luminance (HSL) color picker and changed until all colors had a luminance value of 128. The initial green color was picked by the researcher and had a mid-range luminance value to be able to ease the matching of the other colors. Chi-square tests of independence were conducted to compare the observed and expected values between green and each test color.

Color	Green	Blue	Red	Purple
R	32	56	255	212
G	196	140	70	61
В	30	255	85	255
Hex	20C41E	388CFF	FF4853	D43DFF

Table 1. The values of R (red), G (green), and B (blue) as well as the six-digit name, or HEX code, for each color.

Procedure

Each training session consisted of a total of 20 minute timed periods. In the initial training phases, before the test colors were introduced, each occurrence of the desired behavior was reinforced with a carrot as a primary reinforcer as well as a bridging stimulus from a clicker when the desired behavior was performed. During the initial training, reinforcement occurred on a continuous reinforcement schedule with the desired behavior being reinforced each time that it was performed. During the testing phase, reinforcement occurred on a fixed interval, with reinforcement being given during the first ten minutes of the training but no reinforcement for the last ten minutes of training. The progression to the next step of the training regiment was based upon the overall frequency of the desired behavior, or natural progression to the next step. The training regiment for the initial training phases as well as the test phases are listed below.

Initial Training Regimen:

- 1.) Touching the whiteboard
- 2.) Touching the green circle on the whiteboard
- 3.) Touching the green circle
- 4.) Touch the green circle with the gray circle present
- 5.) Introduce the barrier

Test Color Procedure:

- 1.) The green circle rotated for three minutes
- 2.) Introduction and rotation of test color three times over seven minutes
- 3.) Reinforcement stops and the frequency of each colored circle touched is recorded for ten minutes. During these ten minutes, the circle positions are rotated randomly three times approximately three minutes apart.

Results

During the initial targeting training sessions, the first training day started with a frequency of 1.1 reinforcements per minute, and on the last day of training had a frequency of 2.7 reinforcements per minute. The peak number of reinforcements per minute occurred on day seven with 4.85 reinforcements per minute as shown below in Figure 4.



Figure 4. Frequence of reinforcements per minute across initial training days.

During the testing of color discrimination, the green versus gray had 37 reinforcements during the calibration period, and 21 green touches with 2 gray touches during the testing period (Figure 5). In the blue test, there were 49 reinforcements during the calibration period with 26 green touches and 6 blue touches during the testing period (Figure 5). For the red testing, there were 38 reinforcements during the calibration period with 19 green touches and 19 red touches during the testing period (Figure 5). The final testing color was purple, and there were 32 reinforcements during the calibration period with 12 green touches and 5 purple touches during the test period (Figure 5).

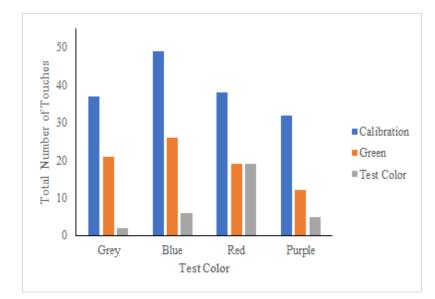


Figure 5. Number of reinforcements during the calibration period and the total number of touches during the testing period for each test color.

The ratio of touches for green: test color was also calculated and plotted as shown in Figure 6. The highest ratio of green: test color touches was 10.5:1 green to gray touches, with the lowest ratio being 1:1 for green: red (Figure 6). The green: the blue ratio was 4.3:1 and the green: purple ratio was 2.4:1 (Figure 6).

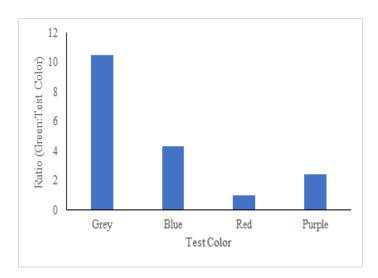


Figure 6. Ratio of green:test color across all colors.

A two-way chi-square of independence was conducted to analyze if there was a significant difference between the expected and observed frequency of touches between the green and the test color circles. There was a significant difference in the observed versus expected frequency of both the gray and blue test colors. For the grey test color, $X^2(1,1) = 15.696$, p < 0.001. The blue test color had a $X^2(1,1) = 12.5$, p < 0.001. Elsa was able to pick out the green over both the gray and blue. There was, however, no significant difference between the observed and expected frequencies of touches for the red or purple. The red test color had a $X^2(1,1) = 0$, p = 1 and for purple $X^2(1,1) = 2.882$, p = 0.09. Red and purple circles were both not significantly distinguishable between the green circle.

Discussion

During the testing color phases, the results were very similar to what I hypothesized. Due to the significant difference shown for both the red and the blue, it is indicated that Elsa was able to distinguish both blue and grey from green. This, however, is not the case for red or purple. When looking at the frequencies for distinguishing red and green, the observed frequencies were the same as the expected frequencies, meaning that Elsa was unable to accurately distinguish red and green. This is similar to the results with purple, however, there is a larger chi-value, which indicates that she can distinguish purple and green, but not accurately enough to show a significant difference between the observed and expected number of touches. This supports my hypothesis that Elsa could distinguish between green and blue, but not red. It also supports that Elsa could distinguish purple to a lesser extent than green and blue. Possible confounds during the study could have been provided from physical cues directing Elsa to choose one color over the other during the testing period. To prevent this, the task could be taught independently of a researcher in the stall so that there would be no external cues provided.

Throughout this experiment, Elsa surprised me at nearly every step. The rate at which she learned to associate the whiteboard and the green circle was astonishing and she only continued to get better at it. When looking at Figure 4, there is a decrease in the frequency of reinforcements the day that a new training step was introduced with a steady increase after the first day of a new training step. The only step that took Elsa back a fair amount was the introduction of the barrier on day eight. She seemed rather distressed with being unable to see me initially. It took her a few minutes of looking around the barrier at me and pacing before she settled down and went back to the task at hand. The session on day eight ended early due to Elsa biting the board off of the wall, but the following days were back to normal reinforcement frequencies with the barrier present. Due to previous studies indicating that horses can distinguish green from gray, gray was utilized as a training tool to reinforce choosing green over gray are a large factor of why the ratio of green: gray touches were twice as high as any other testing color. The use of the green: gray ratio and testing acted more as

exposure to the next phase of testing that lacked reinforcement and provided a control ratio of two colors that she could distinguish between. The following link provides a video from the training and testing throughout the experiment: <u>https://youtu.be/drXU2Su7Tw4</u>.

During the testing with the blue, it was very clear that Elsa was able to distinguish the blue from the green. Throughout the initial calibration period, it was very rare that she chose the blue over the green more than once in a row before repeatedly and continually touching the green circle even as it was rotated across different positions. This, however, was not the case while testing with red. Even during the calibration period, Elsa would touch the red circle three or four times and check-in for reinforcement before going to the green circle. As the calibration period continued and the circles were rotated, there was not much improvement for her picking green over red. It seemed as though she would realize which position she would be reinforced for touching and continually choose that position. Once the circles were rotated, however, she would go back and forth between them until she understood which circle was in each position. This is reflected in the testing period where the ratio of green: red touches was 1:1, showing that the color she chose was at random and she was not associating one in particular with reinforcement (Figure 4).

Based upon these results with the red, I was curious to see how well she would be able to distinguish green and purple. It appears that she can distinguish them, as she was indicating much heavier to the green circle during both the calibration and testing period, and had a ratio more than twice that of the red (Figure 4), but the degree to which she distinguished them was not enough to be significant. This means that the distinctions between the colors seen could be due to random chance. The discrepancy in Elsa's ability to distinguish red and purple is understandable when looking at the two colors from a wavelength standpoint. These two colors

are on opposite extremes on the wavelength spectrum, with the color that we see as purple having a low wavelength, which is similar to that of what we perceive as blue, whereas red has a very high wavelength (Carrol et al., 2001). Although expected, the creation of purple using a combination of both red and blue was an interesting combination to choose from.

Overall, Elsa was able to significantly distinguish gray and blue from the green but was unable to significantly distinguish red and purple from green. These results align with physiological responses from spectral sensitivity studies that were conducted by Carrol et al. (2001), as the higher nm wavelength of red was had the lowest discrimination. Although spectral sensitivity studies indicate what wavelength of light that a horses' eye is responding to, behavioral tests like the one that I conducted are crucial to determine how this input is perceived through a horses' perspective.

Future studies on horse vision could explore what other types of electromagnetic waves that horses could see, be it ultraviolet, radio, or any other type of wavelength in addition to optical. Another direction to explore is how horses' color discrimination could improve through the use of color vision correction lenses to see if this would improve a horses' discrimination across colors with a higher wavelength, like red.

References

- Blackmore T.L., Foster T.M., Sumpter C.E., Temple W. (2008). An investigation of colour discrimination with horses (*Equus caballus*). *Behav Processes*, 78(3), 387-396. https://doi.org/10.1016/j.beproc.2008.02.003.
- Byosiere S.E., Chouinard P.A., Howell T.J., Bennett P.C. (2019) The effects of physical luminance on colour discrimination in dogs: A cautionary tale. *Applied Animal Behaviour Science*, 212, 58-65. https://doi.org/10.1016/j.applanim.2019.01.004.
- Carroll J., Murphy C.J., Neitz M., Hoeve J.N., Neitz J. 2001. Photopigment basis for dichromatic color vision in the horse. *Journal of Vision*, 1(2). <u>https://doi.org/10.1167/1.2.2</u>
- Hanggi E.B. (2005). The Thinking Horse: Cognition and Perception Reviewed. *AAEP Proceedings*, 246-255.
- Jacobs G.H. (2018). Photopigments and the dimensionality of animal color vision. *Neuroscience* & *Biobehavioral Reviews*, 86, 108-130. https://doi.org/10.1016/j.neubiorev.2017.12.006.
- Lansade, L. and Calandreau, L. (2018). A conditioned reinforcer did not help to maintain an operant conditioning in the absence of a primary reinforcer in horses. *Behavioral Processes*, 146, 61-63. https://doi.org/10.1016/j.beproc.2017.11.012.
- Macuda T. and Timney B. (1999) Luminance and chromatic discrimination in the horse (Equus caballus) *Behav Processes*, 44(3), 301-307. DOI: 10.1016/s0376-6357(98)00039-4.
- Martinez de Andino, E., McDonnel, S. (2017). Evaluation of operant learning in young foals using target training. *Applied Animal Behaviour Science*, 193, 67-72. https://doi.org/10.1016/j.applanim.2017.02.021.

- Preacher, K. J. (2001, April). Calculation for the chi-square test: An interactive calculation tool for chi-square tests of goodness of fit and independence [Computer software]. Available from http://quantpsy.org.
- Shanley, C. (2020, September). *Mixing Colors of Equal Luminance Part 2*. Design and Sketch. <u>https://medium.com/sketch-app-sources/mixing-colours-of-equal-luminance-part-2-3e10c</u> <u>07c947c</u>.
- Smith S.A., Goldman L.B. (1999). Color discrimination in horses. Applied Animal Behaviour Science, 62(1), 13-25. https://doi.org/10.1016/S0168-1591(98)00206-8.