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Using Community Science to Assess the Effect of Wing Pattern and Weather on Butterfly Behavior

Abbigail Merrill

University of Arkansas

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Abstract

Signaling in insects is used as communication and for attraction of mates. The physical appearance of the insect as well as conditions such as weather can play a role in visual signaling, by influencing the wavelengths of light available, and subsequent signal detection. We do not know, however, whether signals butterflies present broadly correlate with how they behave. In this study, we looked at the wing patterns and behavior of butterflies in Northwest Arkansas over a 3.5-year period to assess the relationship between wing pattern, weather, and behavior. We used observational data collected by hundreds of University of Arkansas students and Northwest Arkansas community members through surveys at both the Botanical Garden of the Ozarks and the general Northwest Arkansas region. We found that weather and wing color influenced general butterfly behavior. Butterflies were observed feeding more often on cloudy days than sunny days. Black and brown butterflies were observed feeding more often, while yellow and white butterflies were observed flying more often relative to other butterfly colors. We also found that there was an interaction between the effects of weather and wing color on butterfly behavior. White and yellow butterflies were observed feeding more and flying less on cloudy days than sunny days, relative to the other colors of butterflies. Furthermore, butterfly color influenced the choice of flower colors on which butterflies fed. More brown butterflies were observed on yellow flowers relative to other colors of butterflies. These results suggest that flower choice may be associated with butterfly wing pattern, and that different environmental conditions may influence butterfly behavior in wing-pattern-specific ways.

Key words: butterfly, pollinator, ambient light, wing pattern, visual signaling, community science

Introduction

Visual signaling is a form of communication that has a variety of functions for different animals. Signaling can be used for intraspecies communication while avoiding detection from predators of a different species. The male swordtail fish *Xiphophorus nigrensis* has UV ornamentation that increases their attractiveness to females but does not increase their risk of being detected by their predator, *Astyanax mexicanus*, because this species is not as sensitive to UV (Cummings et al., 2003). This allows for private communication among species. Visual features can be further highlighted during courtship and mating displays to attract attention from females. The male butterflies *Hypolimnas bolina* position themselves underneath females in a way that maximizes UV brightness, visible area, and flash-effect while they are fluttering as a part of their courtship ritual (White et al., 2015). Some insects signal mate quality through brightness rather than color. Females of the colorblind mantid *Psueomantis albofimbiata* signal higher mate quality to males through the brightness of their abdomen, with a brighter abdomen indicating better condition (Barry et al., 2015). Signaling is an important visual and behavioral tool used by a multitude of animals for communication.

Cloud coverage and ambient light play an important role in signaling. A higher presence of cloud coverage has higher levels of UV-light than clearer or sunnier conditions (Calbo et al., 2005). The ambient light environment an animal lives in also plays a role in signaling. Endler classified the different light environments as open, large gap, small gap, woodland shade, and forest shade (Endler, 1993). Forest shade, for example, is light coming from reflectance from leaves, not from

direct sunlight or open sky, while woodland shade is light coming from leaves as well as directly from the sky through canopy holes (Endler, 1993). Endler has classified colors to the light environments as well, with open and large gap being white, small gap being orange, woodland shade being blueish-green, and forest shade being yellowish-green (Endler, 1993; Endler, 1997). Many butterfly species exhibit polarized reflectance patterns, and these species are more likely to be found in forest habitats than open habitats (Douglas et al., 2007). This indicates that the ambient light conditions and cloud coverage in an environment influence which species will be present and capable of thriving in that environment. UV-light and ambient light are important environmental factors in influencing butterfly abundance. While it is well documented that ambient light environments can influence signal perception (Endler, 1993; Douglas et al., 2007), it remains unclear whether butterflies change their behavior in response to ambient light at the community level.

Community science, sometimes referred to as citizen science, is a research method in which the public is enlisted to obtain information for a study (Bonney et al., 2009). Community science can be used for both data collection and conservation efforts. Many pollinator species, including butterflies, face endangerment due to habitat loss, wildflower decline, and urbanization (Preston et al., 2012). For example, a well-known North American butterfly, the monarch (*Danaus plexippus*), faced an 81% population decline from 1999-2010 (Pleasants et al., 2013). Community science can be used to track these population declines and allow conservation groups to execute plans to protect and conserve pollinator populations. Community science is also useful for getting people involved in and educated about conservation. A survey of community science project leaders showed that 91% of community science projects in the United

States have a conservation focus (Lewandowski et al., 2016). Most community science project leaders also supply information to volunteers about threats to animal or plant populations, as well as potential conservation actions that can be taken (Lewandowski et al., 2016). Community science projects are being used to inform the public about conservation efforts and allow people to become engaged in conservation efforts.

One of the concerns of using community science data for conservation purposes is the accuracy of the data collected. A United Kingdom study comparing the data collected by The Big Butterfly Count, a community science group, and the UK Butterfly Monitoring Scheme, a research initiative with standardized recording protocol, found that the community science data produced comparable estimates of butterfly species abundance to the standardized protocol, although there is opportunity for possible misinformation when community science data is collected over short periods of time (Dennis et al., 2017). A community science project focused on data collection on pollinator communities found that community scientists did well with higher taxonomic level composition, bee abundance, bee richness, and bee community similarities, though community scientists did not accurately report information on specific species, indicating that community science may be limited to detection of community level changes (Kremen et al., 2011). The eButterfly project has also concluded that community science is useful for collection of information on species richness (Prudic et al., 2018). Another pollinator focused group called the Native Bee Watch, a group with high volunteer retention rates, found that researcher data correlates with community science data (Mason et al., 2019). Community science is not meant to replace research done by professionals; however, these

studies suggest that community science is helpful for obtaining widespread data and accurate at reporting population estimates.

In this study, we gather information on the behavior and abundance of different butterflies in Northwest Arkansas through surveys filled out by community scientists. One goal of this study is to provide information about the correlation between butterfly colors and their behavior and plant preferences. This study also aims to involve the community in collecting information about native species and hopefully inspire interest in the butterfly community in Northwest Arkansas.

Literature Review

Butterfly Vision

Butterfly vision is an intricate and complex process that has key differences compared to the eyes of humans. Butterflies have compound eyes composed of many ommatidia that are arranged in a hemisphere (Stavenga & Arikawa, 2006). The outer portions of the eye, the facet lenses, each associate with a crystalline cone (Stavenga & Arikawa, 2006) (Figure 1). These work together to form the imaging optics responsible for projecting light onto the photoreceptors and focus light into the rhabdom, thus enhancing light absorption by the visual pigments (Stavenga & Arikawa, 2006). An ommatidium in a butterfly contains nine photoreceptors, each of which can be sensitive to different wavelengths of light. The sensitivity to these wavelengths of light can be adjusted by filtering pigments, which are concentrated in clusters around the rhabdom and function to selectively absorb specific wavelengths of light (Stavenga, 2002). Butterfly visual pigments, called rhodopsins, are located in the rhabdomere. The process of butterfly vision functions through the absorption of light by these visual pigments (Stavenga & Arikawa, 2006).

In most butterfly species there is also a membrane at the back of the ommatidium, the tapetum. Incident light that enters through the rhabdom without being absorbed is reflected by the tapetum and travels back through the eye, enhancing the opportunities for rhodopsins to be excited by light (Stavenga, 2002). The combined elements of a butterfly's eyes function to allow for one of the most important senses required for butterflies.

Vision is not the same for all species of butterflies, which is reflected in the contrasting abilities of different species to see different colors (Briscoe & Bernard, 2005). One of the ways we measure butterfly color sensitivity is through eyeshine. Eyeshine is the light that is reflected off of the tapetum; it can be used to tell the observer the color of the filtering pigments present in the eye, and therefore the wavelengths of light available to rhodopsins. Junonia coenia has a homogenous blue eyeshine, contrasting with Vanessa cardui, which has a homogenous orange eyeshine, while Nymphalis antiopa and Siproeta stelenes have a more heterogenous eyeshine than the previous two species (Briscoe & Bernard, 2005). This means that J. coenia has better visual abilities for detecting the color blue, while V. cardui is more inclined to see the color orange. This suggests that vision varies between species of butterflies. Another study examining the difference in color vision between species found that *Bicyclus anynana* butterflies were heterogenous in the eyeshine pattern in the ventral eye area and had yellow-reflecting ommatidia in the dorsal eye area (Stavenga, 2002). In contrast the majority of the ommatidia in Heliconius *melpomene* eyes were red, while the dorsal ommatidia reflected a mixture of yellows (Stavenga, 2002). This study indicates that *H. melpomene* has better visual abilities for detecting the color red, while *B. anynana* can more easily detect the color yellow. Vision also may differ in the sexes of some species, as illustrated by *B. anynana*, where males had larger eyes than the

females under two different rearing conditions (Everett et al., 2012). This study also found that increasing the rearing temperature led to increases in eye size, mostly accounted for by an increase in facet lens number (Everett et al., 2012). Thus, butterfly vision is variable across species and between sexes, potentially contributing to the different behaviors and preferences (floral and wing pattern) of different species.

Vision is an important sensory modality for butterflies, and influences their behavior, particularly their mate choice behavior. Male *H. melpomene* butterflies search for females using predominantly visual cues (Jiggins et al., 2004). For example, male *H. melpomene* butterflies from four parapatric populations use color patterns for mate detection and choose females that exhibit their own color pattern opposed to females of different color patterns (Jiggins et al., 2004). Butterflies use both wavelengths of light visible to humans and UV light to detect and choose mates (Obara & Hidaka, 1968). For example, male *Pieris rapae crucivora* recognize females using a mixture of near-UV light and visible light reflected by the wings of the female (Obara & Hidaka, 1968). The hind wing of the female reflects between 30%-40% of near-UV light, while the male almost entirely absorbs it and only reflects 5% (Obara & Hidaka, 1968). These differences allowed the male to detect and differentiate females from males. Male butterflies of the species *P. r. crucivora* also are more active in UV-rich environments, and spend longer amounts of time searching for females and approach and copulate more often with females in the shade, which has relatively higher amounts of UV light (Obara et al., 2008).

Environmental Effects on Pollinator Behavior

Anthropogenic factors affecting pollinator habitats can also influence pollinator behavior and abundance. The intensity of land use is one of the environmental components that affects butterflies. In Germany butterfly species diversity decreases with increasing land-use intensity and butterflies in intensively managed lands have longer flight periods and a larger number of generations per year than butterflies living in less intensively managed land (Borschig et al., 2013). As habitat patch size decreases, generalist species dominate species with lower dispersal power, narrower feeding niches, and lower reproductive rates in both Europe and North America (Ockinger et al., 2010). This study also showed that specialist butterflies, short-winged species of butterflies, and species with low reproduction are more likely to be harmed by habitat loss than generalist species of butterflies (Ockinger et al., 2010). Increasing land use intensity does not always directly harm pollinator populations. An increase in land use intensity can lead to a loss in flower diversity, which then can lead to a decrease in pollinator diversity (Weiner et al., 2014). A decrease in flower diversity is more harmful for specialist species, which rely on specific species of flowers for food, than for generalist species, which feed from a larger variety of flowers (Weiner et al., 2014). The space butterflies live in and the degree to which the area is urbanized affects what species will thrive and which species will decline.

Temperature is another key environmental factor that can affect butterfly behavior and abundance. A study examining the effect of temperature on pollinators across 40 grasslands found that 84% of the variation in pollinator activity is explained by ambient temperature and that lighter insects prefer habitats with lower temperatures (Kuhsel & Bluthgen, 2015). When looking at individual species instead of taxonomically broad behavior, *Junonia coenia* butterflies preferentially mate and court during the warmest times of the day (McDonald & Nijhout, 1996).

Mating activity is most frequently observed at intermediate temperatures and high light intensities for this species and lowering the light level lowers mating activity during both optimal and high temperatures. Altering temperature can also change the likelihood of survival for some species (Stuhldreher et al., 2014). The continental butterfly species *Erebia medusa* faces lower survival rates as winter temperatures increase (Stuhldreher et al., 2014). All of this indicates that butterfly behavior is not influenced by one singular component of the environment, there are multiple environmental factors that account for influencing their behavior.

The traits of plant species that compose a pollinator's environment, specifically flowering species, are another important factor in determining which pollinator species are present and how they interact with their surroundings. Butterflies have been shown to have preferences for certain flower colors (Pohl et al., 2011). For example, Speyeria mormonia and Phyciodes campestris prefer the orange flowers over yellow flowers of the plant *Dugaldia hoopesii*. Multiple butterfly species exhibit flower color preferences, in which they visit one flower species more than others based on color, though flower size and morphology plays a role as well for some species (Pohl et al., 2011). Some butterfly species also respond to manipulations of flower color (Pohl et al., 2011). Flower color can also be an indicator for nectar availability, as illustrated by the flower *Lantana camara*, which changes from a yellow color to a reddish-orange color after nectar has been removed (Barrows, 1976). Changes in flower color such as this could indicate to butterflies a plant that contains no nectar, which could possibly lead to less visitation to these flower colors. For a variety of butterfly species in southern England grasslands, there is a strong positive correlation between host plant abundance and butterfly abundance as well as a positive relationship between nectar abundance and butterfly abundance (Curtis et al., 2015).

Furthermore, butterflies that are sedentary rather than mobile have a steeper host plantabundance relationship (Curtis et al., 2015). Traits of the plants that compose a butterfly's environment can therefore influence which butterfly species are abundant and how these species behave.

Community Science

Butterfly vision and a butterfly's relationship with its environment can be examined in laboratory settings, but information on these topics can also be gathered by the public. Community science enlists community members for assistance in collecting scientific data (Bonney et al., 2009). Research using community science allows for information to be collected across a wide geographic range. It also engages the public and garners interest in science among the public. Students that engage in hands on scientific activities become more confident in their learning and have improved scientific reasoning skills (Beck & Blumer, 2012). Community science is an opportunity for active engagement for both students and non-students and is helpful for gathering data for scientific advancement as well as engaging the public in scientific methods.

Community science allows a widespread collection of information that can help scientists see what animals need in their natural habitat. A community science project in Japan focused on Little Tern conservation used community scientists to shed light on the preferred substrate of Little Terns, which then led to the Little Tern Project treating colony sites with the preferred substrate (Kobori et al., 2015). A project known as eButterfly is currently utilizing community scientists to better understand butterfly distribution and abundance. This information can be helpful for conservation strategies, as it allows scientists to track timing of migration and study impacts of global change on migration (Prudic et al., 2017). Learning the flight patterns these butterflies are following can allow scientists and conservationists to preserve the key migration areas and further protect these butterfly species.

Engaging the public in scientific research may allow people to see how their actions can impact the environment and can show people possible changes that can be made to protect certain species. Following a community science project on invasive plant species, a survey showed that 86% of the participants began considering which plants were invasive when purchasing plants, while 70% reported changing their behavior, and 43% reported discussing invasive plant species with others (Jordan et al., 2011). After the formation of Neighborhood Nestwatch, a project designed to improve knowledge about avian ecology and spread awareness for conservation initiatives, 56% of participants reported they changed some aspect of their behavior, such as planting shrubs that could act as food or shelter for birds (Evans et al., 2005). Many participants in this study also reported that they joined in order to educate their children on conservation efforts, demonstrating that community science can be used to spread conservation awareness to younger audiences (Evans et al., 2005).

For all these reasons, we are using a community science approach to explore the relationship between butterfly wing color and butterfly behavior. In this study, we use data collected by participants at the Botanical Garden of the Ozarks as well as students in Principles of Zoology and Animal Behavior to examine butterfly behavior and flower preferences.

Methods

<u>Study species:</u> This research surveys lepidoptera from across northwestern Arkansas. Because butterflies were identified by color rather than species, we do not know the exact species included. However, based on colors reported by participants, some likely species include *Danaus plexippus* (Monarch), *Papilio glaucus* (Tiger Swallowtail), *Junonia coenia* (Common Buckeye), *Strymon melinus* (Gray Hairstreak), *Vanessa virginiensis* (American Lady), *Vanessa cardui* (Painted Lady), *Chlosyne nycteis* (Silvery Checkerspot), *Physciodes tharos* (Pearl Crescent), *Colias philodice* (Clouded Sulphur), and *Colias Eurytheme* (Orange Sulphur).

<u>Study site:</u> One of the primary sites of observation was the Botanical Garden of the Ozarks (BGO), Fayetteville, AR, located at 36°08'12"N and 94°07'06"W (Figure 2A). The BGO is 44 acres in size, has twelve themed gardens, and contains a native butterfly house. There are an estimated 80,000 visitors every year, and an average of 18,000 people are educated about butterflies and pollinator gardens through the Botanical Garden's various programs. Animal Behavior students completed their observations at a second site, Wilson Park, Fayetteville, AR, located at 36.072994 N and 94.163239 W in 2017,2018, and 2019. Wilson Park is a 22.75 acre park located in the center of Fayetteville. Wilson Park has a spring, pond, playground, and walking trail. Additional study areas included various locations throughout Northwest Arkansas where Principles of Zoology students conducted their observations, and conducted their surveys in residential neighborhoods, city and state parks, farms, and wilderness areas throughout the

region. Most students recorded the latitude and longitude of their starting point. Animal Behavior students in 2020 also conducted their butterfly surveys throughout the region due to COVID shutdowns at the University of Arkansas. Their survey locations were also recorded (Figure 2). Northwest Arkansas is composed of wet and dry prairies and the Boston Mountains.

Experimental design: Observations were collected by Northwest Arkansas citizens and University of Arkansas students enrolled in Principles of Zoology and Animal Behavior over a duration of 4 years, from April 2017 to November 2020. Animal Behavior students, Principles of Zoology students, and Botanical Garden visitors were asked to collect similar observational data, but were given different instructions concerning the duration of their survey. Participants were instructed to note date, time, color of the butterfly, activity of the butterfly (flying, feeding, sitting), size of the butterfly (small, medium, large), and color of the flower the butterfly was on if it was on a flower. Participants were instructed to pick one main color for the butterflies and the flowers. Principles of Zoology and Animal Behavior students were also asked to record weather conditions (sunny, cloudy, partly cloudy, rain). In Principles of Zoology, students were asked to note latitude and longitude at the start of their walk, and to collect butterfly observation data over a 30-minute walk during a 7-10 day period in the last week of September and first week of October. Observations were collected on paper and submitted in class (Supplemental Figure 1). For Animal Behavior, students went on a 30-minute walk at Wilson Park on the Friday closest to April 16. Observations were completed in groups and collected on paper. In 2020, due to COVID shutdowns, students went for a 30-minute walk on their own, wherever they were located, instead of as a class. Botanical Garden Participants were not given a time limit and

collected data throughout the year. Data from all participants were compiled into an excel spreadsheet for analysis.

Data processing: After all the data were entered, we then separated butterfly colors into the most likely primary color or colors, because sometimes participants picked more than one color. For consistency, one researcher reclassified all butterfly colors. We then filtered out rare responses in the following ways: For analyses involving butterfly color, we removed all colors with less than 1% responses, leaving us a subset including the butterfly colors yellow, black, blue, brown, orange, and white. For analyses involving size, we removed the few butterfly observations where participants selected multiple sizes, creating a subset in which only one size was selected: small, medium, or large. For analyses involving activity, we categorized feeding as the dominant behavior when feeding was selected along with an additional behavior, and excluded records where both fly and sit were selected. For analyses involving weather, we created a subset containing sunny, cloudy, and partly cloudy weather, as those were the predominant selected weather options (responses of rainy, cold, warm, and specific temperatures were rare). For analyses involving flower color, we created a new category, "multi" for the records where multiple colors were selected, giving us the final options of blue, multi, green, orange, pink, purple, red, yellow, and white.

<u>Statistical analysis:</u> To determine if butterfly size or color affected observed activity or the flower and plant colors butterflies landed on, we conducted chi-square tests. We also assessed

the effect of weather, time of day, and survey year on observed butterfly color, size, and behavior using chi-square tests.

To account for correlative effects of butterfly color and size, and to test for any interactive effects of butterfly color and weather, we conducted a series of nominal logistic regression models. We first conducted a model with butterfly color, weather, and an interaction term of butterfly color * weather as factors and butterfly activity (feed, fly, sit) as the dependent variable. To determine if there was an interactive effect of butterfly size and weather on observed activity, we conducted a nominal logistic model with butterfly size, weather, and an interaction term of butterfly size*weather as factors. To determine if there was an effect of weather, butterfly color, butterfly size, and time of day on observed activity, we conducted a nominal logistic model with weather, butterfly color, butterfly size, time of day, and an interaction term of weather and butterfly color as factors. To determine if there was an effect of butterfly color and size on activity, we conducted a nominal logistic model using butterfly color, butterfly size, and an interaction term of size on activity, we conducted a nominal logistic model using butterfly color, butterfly size, and an interaction term of butterfly color *butterfly size as factors.

To determine if there was an effect of weather and time of day on butterfly color, butterfly size, or observed activity, we conducted a nominal logistic model using weather, time of day, and an interaction term weather*time of day as factors. To determine if there was an interactive effect of butterfly color and size on flower color selected, we conducted a nominal logistic model with butterfly color, butterfly size, and an interaction term of butterfly color*butterfly size as factors. To determine if there was an effect of butterfly color, size, and weather on flower color chosen

or butterfly activity, we conducted nominal logistic models with butterfly color, butterfly size, weather, and an interaction term between weather and butterfly color as factors, and flower color and butterfly activity as dependent variables. We also ran tests to see if there were differences in the sizes and colors of observed butterflies in different survey years.

Since we conducted 23 chi-squared tests, we used a Bonferonni corrected p-value of 0.002 for our chi-squared tests. We conducted 7 nominal logistic models, and used a corrected p-value of 0.007 for these models. All chi-squared analyses and all nominal logistic models were conducted in JMP Pro 15.

Ethical statement: No butterflies were harmed during this study; all observations were nocontact. No humans were harmed in the conducting of this experiment; students participated in this as part of their class requirements and community scientists were volunteers.

Results

Effect of butterfly color and size on butterfly behavior

Butterfly color was correlated with butterfly behavior in data collected by BGO participants (P<0.0001, χ 2=265.040, n=1,971) and University of Arkansas students (P<0.0001, χ 2=64.172, n=1,758) (Figures 3, 4). At BGO, white butterflies were seen flying more than feeding or sitting, while brown butterflies were seen feeding more than sitting or flying. University of Arkansas students saw brown butterflies sitting more than feeding or flying. Butterfly size was correlated

with butterfly behavior at the BGO (P<0.0001, χ 2=31.192, n=1,970), but not in University of Arkansas data (P=0.0058, χ 2=14.526, n=1,779). However, butterfly size was correlated with butterfly color in both the BGO (P<0.0001, χ 2=556.917, n=2,006) and University of Arkansas (P<0.0001, χ 2=277.126, n=1,753) data. Black butterflies were more likely to be large than small or medium, and brown and white butterflies were more likely to be small (Figures 5, 6). However at the BGO, when butterfly color, butterfly size, and an interaction term of butterfly color and size were included in a nominal logistic model, we found that only butterfly color and the interaction term significantly influenced activity, suggesting butterfly color may be more important than butterfly size in predicting butterfly behavior (butterfly color:

P<0.0001, $\chi 2=93.971$; butterfly size: P=0.0131, $\chi 2=12.651$; butterfly color*butterfly size: P=0.0019, $\chi 2=43.242$; n=1,933). From data collected by University of Arkansas students, we found that only butterfly color significantly influenced activity (nominal logistic model, factor effects: butterfly color: P=0.0011, $\chi 2=29.436$; butterfly size: P=0.1261, $\chi 2=7.191$; butterfly color*butterfly size: P=0.0539, $\chi 2=31.100$; n=1,751).

Effect of butterfly color, butterfly size, and weather on flower choice

Main butterfly color was predictive of the color of the flower butterflies were seen on in both BGO (P<0.0001, χ 2=179.103, n=1,276) and University of Arkansas (P=0.0001, χ 2=80.936, n=879) data (Figures 7, 8). White butterflies were seen on green flowers more than the other colors of butterflies at the BGO. Weather also influenced the color of flower a butterfly was seen on (P<0.0001, χ 2=58.396, n=614) (Figure 11). Butterflies were seen on orange and red flowers more often on cloudy days than in other weather conditions, and on multicolor flowers more often on partly cloudy days than in other weather conditions.

Butterfly size had an effect on flower color chosen at the BGO (P<0.0001, $\chi 2=93.829$, n=1,271), but not in the University of Arkansas data (P=0.0067, $\chi 2=33.343$, n=891). Large butterflies were seen on orange flowers more than the other sizes of butterflies at the BGO. A nominal logistic model with the factors butterfly color, butterfly size, and an interaction term of butterfly color and size showed that there was an interactive effect of butterfly color and size on flower color choice at the BGO (butterfly color: P=0.0003, $\chi 2=78.013$; butterfly size: P=0.0080, $\chi 2=32.734$; butterfly color*butterfly size: P<0.0001, $\chi 2=135.793$; n=1,252), but not in University of Arkansas data (butterfly color: P=0.0022, $\chi 2=70.164$; butterfly size: P<0.0001, $\chi 2=148.124$; butterfly color*butterfly size: P=0.8401, $\chi 2=67.464$; n=876). When weather was taken into account, we lost the effect of butterfly size on flower color choice (Nominal logistic model, factor effects: butterfly color: P<0.0001, $\chi 2=291.738$; butterfly size: P=0.2849, $\chi 2=18.693$; weather: P<0.0001, $\chi 2=89.518$; weather*butterfly color: P=0.2187, $\chi 2=89.518$; butterfly color*butterfly size: P=0.8081, $\chi 2=61.439$; n=605).

Effect of weather on butterfly color, butterfly size, and butterfly behavior

Because BGO participants were not asked to note weather conditions, effects of weather were only analyzed using data collected by University of Arkansas students. Weather had an effect on observed butterfly behavior (P=0.0001, χ 2=23.429, n=1,281) (Figure 10). Butterflies were seen feeding more on cloudy days than other weather conditions. However, weather did not have a significant effect on either observed butterfly color (P=0.0035, χ 2=26.221, n=1,240) (Figure 9) or observed butterfly size (P=0.3469, χ 2=4.464, n=1,249). A nominal logistic model with the variables weather, butterfly color, and an interaction term of weather and butterfly color showed

that only butterfly color and the interaction term had an effect on butterfly behavior (weather: P=0.0156, χ 2=12.252; butterfly color: P=0.0001, χ 2=35.247; weather*butterfly color: P=0.0006, χ 2=46.842; n=1,239). A nominal logistic model with the variables weather, butterfly size, and an interaction term of weather and butterfly size showed that none of these variables had an effect on butterfly behavior using our Bonferroni correction (weather: P=0.0439, χ 2=9.801; butterfly size: P=0.0261, χ 2=11.042; weather*butterfly size: P=0.0124, χ 2=19.494; n=1,248).

Effect of time of day on butterfly color, butterfly size, and butterfly behavior

Time of day had an effect on observed butterfly color in data collected by both BGO (P<0.0001, $\chi 2=77.760$, n=1,764) and University of Arkansas (P<0.0001, $\chi 2=37.396$, n=1,569) participants. Orange butterflies were seen more in the evening than the other colors of butterflies. Time also had an effect on observed butterfly behavior in BGO data (P<0.0001, $\chi 2=52.577$, n=1,719), but not in University of Arkansas data (P=0.242, $\chi 2=5.470$, n=1,617). At the BGO, butterflies were seen feeding more in the morning than the other times of day. Time of day did not have an effect on observed butterfly size (BGO: P=0.0121, $\chi 2=12.841$, n=1,749; University of Arkansas: P=0.3061, $\chi 2=4.821$, n=1,585). A nominal logistic model with the variables time, weather, and an interaction term between time and weather showed that only the interaction term had an effect on butterfly color (time: P=0.0299, $\chi 2=19.928$; weather: P=0.0189, $\chi 2=21.330$; time*weather: P=0.0063, $\chi 2=39.197$; n=1,113). A nominal logistic model with the variables time, weather, and an interaction term between time and weather showed that none of these variables had an effect on butterfly behavior or butterfly size.

Effect of butterfly color, butterfly size, time, and weather on butterfly behavior

A nominal logistic model with the variables butterfly color, butterfly size, time, weather, and an interaction term of weather and butterfly color showed that only butterfly color and the interaction term had an effect on butterfly behavior under our Bonferroni correction (butterfly color: P=0.0005, χ 2=31.252; butterfly size: P=0.3706, χ 2=4.270; time: P=0.0443, χ 2=9.779; weather: P=0.0075, χ 2=13.941; weather*butterfly color: P<0.0001, χ 2=58.270; n=1,106).

Discussion

Our study, which integrates data collected by many community scientists, indicates that butterfly behavior is influenced by multiple factors. Primary butterfly color and size both influence butterfly behavior. Weather and time of day influence butterfly behavior as well. Furthermore, a butterfly's choice of flower color is influenced by multiple factors as well. Primary butterfly color, size, and weather all influenced the choice of flower color a butterfly was observed on. Our results did not conclude that weather conditions correlated with which primary butterfly colors were observed. However, time of day did have an influence on which primary butterfly

Our results suggest that butterfly color is broadly correlated with butterfly behavior. BGO participants recorded brown butterflies feeding more than the other colors of butterflies, while Principles of Zoology and Animal Behavior students recorded brown butterflies sitting more than the other colors of butterflies. Though only the color of the butterfly was recorded, these data could suggest that some species or families of butterflies are more active than others. Furthermore, these data could be used to predict where certain species of butterflies are more likely to be found. Butterflies that are recorded sitting and feeding more than the other colors of butterflies may be less likely to be found in open areas with no flowering plants or substrate on which they could land. Future studies should explore the relationship between flowering plant availability and the abundance of different butterfly species as well as broad scale behavioral differences between butterfly families.

Our results show that environmental conditions also influenced butterfly behavior, with weather having an influence on butterfly activity. These data suggest that weather conditions can be used to predict how a butterfly will behave. These results support previous research that shows that temperature influences butterfly behavior (Kuhsel & Bluthgen, 2015), as days with more sunlit conditions are generally warmer than cloudy conditions keeping all other variables the same. Our findings also support studies that show butterfly activity is influenced by UV-light, such as work with *P. rapae crucivora* that indicates that shady conditions, that is conditions with higher amounts of UV-light, are more favorable for copulation (Obara et al., 2008). Our data show that butterflies prefer to feed in cloudy conditions than partly cloudy or sunny conditions. This information could indicate that high levels of UV-light are important for a butterfly's detection of optimal foraging sites. The bird species *Rupicola rupicola, Corapipo gutturalis,* and *Lepidothrix serena* behave differently based on the ambient light conditions they're in (Endler & Thery, 1996). Our research indicates that butterflies may alter their behavior based on ambient light conditions as well.

Amount of cloud cover had an influence on the color of flower on which a butterfly was recorded. This could suggest that the amount of UV light in the environment affects a butterfly's

visual abilities. Previous research has shown that butterflies have preferences for flower colors (Pohl et al., 2011). If flower color preferences are changing based on cloud coverage, it is possible that the light in the environment is affecting how the butterfly sees the flowers. In forests, the presence of clouds changes the color of ambient light from greenish to white (Endler, 1993). The changing ambient light in our study could have affected how the butterflies perceived the flower colors. Multiple bird species alter their display methods for courtship based on the ambient light conditions (Endler & Thery, 1996). This is another indicator that the amount of cloud coverage affects how color is seen by animals. Previous studies have shown that altering environmental rearing conditions such as temperature can alter facet lens number (Everett et al., 2012). If the weather conditions in this study were consistent over the span of several days, this could provide further evidence that temperature and environmental conditions influence butterfly vision. Future studies should examine specific butterfly species' preferences in flower color given multiple options in both sunny and cloudy conditions, as well as the effect of rearing light environment on adult butterfly flower preference.

Our results show that butterfly color has an influence on the color of flower on which a butterfly will be seen. White butterflies were seen more often on green flowers or plants and brown butterflies were seen more often on yellow flowers. A butterfly's preferences for flower color has scientific value by providing information on butterfly vision. Previous studies have shown that butterflies see color differently (Briscoe & Bernard, 2005; Stavenga, 2002). Our data showing that different colors of butterflies have different preferences for flower colors could suggest that the visual appearance of a flower is an important factor in a butterfly's choice on where to land or feed. Another possible reason some butterfly colors are more attracted to some

flower colors could be for camouflage. There are multiple documentations of camouflage tactics in butterfly species, such as the butterfly *Polygonia c-album* mimicking a dead dried out leaf pattern (Brakefield et al., 1992) and *Memphis philumena* mimicking the vein pattern of leaves (Salazar and Julian, 2008). Our research could provide insight on butterflies choosing flower colors they can camouflage against. Furthermore, linking flower color with butterfly color can indicate which flower species to protect and keep an abundance of to attract the butterflies that are frequently seen on them. Previous community science research has led conservationists to identify preferred substrate for a bird species, and then make more of that substrate available to the species (Kobori et al., 2015). Similar measures could be taken using the results of this and similar studies. More flowers could be planted that correlate with the color of butterfly they attract. Using the information gathered by community members, we can infer where conservation efforts are needed and formulate plans to enact conservation efforts.

Community science has been shown to be effective at gathering information on species richness (Prudic et al., 2018), which is reflected through this study. Although we don't have information on specific species recorded, we do have records of the color of butterflies seen by participants. From the data gathered by community scientists, we can see that some butterfly colors were seen more in certain years and less in others, which could possibly indicate a decline in the abundance of some species. White butterflies were recorded most in 2017, with a decline in recordings in the years after. Blue butterflies and brown butterflies were recorded most in 2018, and less in the following years as well. Though we don't have information on which specific species were seen, the primary colors recorded could still assist with understanding some butterfly's abundance and distribution, similar to the work done by eButterfly (Prudic et al., 2017). These community

science data allow for insight into the abundance and potential need for conservation efforts for butterflies in Northwest Arkansas.

Throughout the course of this study, hundreds of individuals were introduced to pollinator behavior. This exposure can allow for participants to become involved in conservation efforts designed to protect butterflies. Sparking an interest in conservation efforts is efficient when people can get hands on experience and understand the need for such efforts (Evans et al., 2005). For butterflies, these conservation interests can be helpful for the protection of *D. plexippus*, a species that has faced a large population decline (Pleasants et al., 2012). Rather than just telling people about butterfly numbers and citing statistics, this community science research has been showing people butterfly diversity and abundance. The participants of this study helped to provide insight on the behavior and preferences of the butterflies of Northwest Arkansas. Getting members of the community of all ages involved with butterfly data collection can be useful for garnering interest in conservation, as well as identifying conservation measures that need to be taken.

Conclusions

This study used community science to examine multiple factors that could affect butterfly behavior and preferences. Because it was decided it would be more reliable to ask community scientists to list the primary color rather than attempt to identify a butterfly and potentially provide incorrect information, we cannot provide data for any specific species of butterflies. However, this study does provide broad information on the behavior and abundance of butterflies in Northwest Arkansas, as well as how these behaviors correlate with primary color, size, weather, and time of day. Future research should be done to determine which butterfly species prefer which flower species, as well as how specific butterfly species act in different weather conditions. We hope the high involvement and large amount of data collected in this study can serve as evidence and act as inspiration for other scientists considering using community science for their research efforts.

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Tables

Table 1. Chi-squared tests using the independent variables of butterfly color, butterfly size, weather, time of day, and year.

		BC	60		UARK			
Test	χ^2	p-value	DF	Ν	χ^2	p-value	DF	Ν
Activity by	265.040	< 0.0001	10	1971	64.172	< 0.0001	10	1758
butterfly color								
Flower color	179.103	< 0.0001	40	1276	80.936	0.0001	40	879
by butterfly								
color								
Butterfly	556.917	< 0.0001	10	2006	277.126	< 0.0001	10	1753
color by size					-			
Activity by	31.192	< 0.0001	4	1970	14.526	0.0058	4	1779
size								
Flower color	93.829	< 0.0001	16	1271	33.343	0.0067	4	891
by size								
Butterfly	NA	NA	NA	NA	26.221	0.0035	10	1240
color by								
weather								
Activity by	NA	NA	NA	NA	23.429	0.0001	4	1281
weather								
Size by	NA	NA	NA	NA	4.464	0.347	4	1249
weather								
Flower color	NA	NA	NA	NA	58.396	< 0.0001	16	614
by weather								
Butterfly	77.760	< 0.0001	10	1764	37.524	< 0.0001	10	1569
color by time								
of day								
Activity by	52.577	< 0.0001	4	1719	5.470	0.242	4	1617
time of day								
Size by time	12.841	0.0121	4	1749	4.821	0.306	4	1585
of day								

Table 2. Nominal logistic regression model with factors of weather, butterflycolor, and an interaction term of weather and butterfly color on butterfly activity.

Weather and Butterfly Color on Acti				
Variable	p-value	χ^2	DF	Ν
Weather	0.0156	12.2515259	4	1239
Butterfly Color	0.0001	35.2474908	10	1239
Weather*Butterfly Color	0.0006	46.8420005	20	1239

Weather and Size on Activity (subset)				
Variable	p-value	χ^2	DF	Ν
Weather	0.0439	9.80076787	4	1248
Size	0.0261	11.0420957	4	1248
Weather*Size	0.0124	19.4938925	8	1248

Table 3. Nominal logistic regression model with factors weather, butterfly size, and an interaction term of weather and butterfly size on butterfly activity.

Table 4. Nominal logistic model with the factors weather, butterfly color, butterfly size, time of day, and an interaction term of butterfly color and weather on butterfly activity.

Butterfly Color, Size, Time, Weather on	Activity			
(subset)				
Variable	p-value	χ^2	DF	Ν
Butterfly Color	0.0005	31.2522173	10	1106
Size	0.3706	4.27044773	4	1106
Time	0.0443	9.77908727	4	1106
Weather	0.0075	13.94143	4	1106
Weather*Butterfly Color	< 0.0001	58.2704613	20	1106

Table 5. Nominal logistic regression model with the factors butterfly color, butterfly size, and an interaction term of butterfly color on butterfly size on butterfly activity.

Size and Butterfly Color on Ac				
Variable	p-value	χ^2	DF	Ν
Size	0.0130	12.6687533	4	3684
Butterfly Color	< 0.0001	94.9765543	10	3684
Size*Butterfly Color	< 0.0001	60.0048807	20	3684

Weather and Time on Activity (subset)				
Variable	p-value	χ^2	DF	Ν
Weather	0.0479	9.58988252	4	1154
Time	0.4364	3.78121107	4	1154
Weather*Time	0.1059	13.1777678	8	1154

Table 6. Nominal logistic regression model with the factors weather, time of day, and an interaction term of weather and time of day on butterfly activity.

Weather and Time on Butterfly Color (subset)				
Variable	p-value	χ^2	DF	Ν
Weather	0.0189	21.3303795	10	1113
Time	0.0299	19.9277236	10	1113
Weather*Time	0.0063	39.1968405	20	1113

Table 7. Nominal logistic model with the factors weather, time of day, and an interaction term of weather and time of day on butterfly color.

Weather and Time on Size (subset)				
Variable	p-value	χ^2	DF	Ν
Weather	0.4891	3.42682786	4	1122
Time	0.4968	3.37704516	4	1122
Weather*Time	0.3378	9.05337795	8	1122

Table 8. Nominal logistic model with the factors weather, time of day, and an interaction term of weather and time of day on butterfly size.

Figures



Figure 1. Diagram of the eye of a butterfly. Diagram from Stavenga & Arikawa, 2006.



Figure 2. Map of the survey sites. The red star indicates the Botanical Garden of the Ozarks. The yellow star indicates Wilson Park. Image provided by MJ Murphy. Figure from Merrill & Hirzel et. al., 2021.



Figure 3. Effect of butterfly color on butterfly activity using data collected by BGO participants. Figure from Merrill & Hirzel et. al., 2021.



Figure 4. Effect of butterfly color on activity using data collected by Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.



Figure 5. Effect of butterfly color on butterfly size using data collected by BGO participants. Figure from Merrill & Hirzel et. al., 2021.



Figure 6. Effect of butterfly color on butterfly size using data collected by Principles of Zoology and Animal Behavior Students. Figure from Merrill & Hirzel et. al., 2021.



Figure 7. Effect of butterfly color on flower color using data collected by BGO participants. Figure from Merrill & Hirzel et. al., 2021.



Figure 8. Effect of butterfly color on flower color using data collected by Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.



Figure 9. Effect of weather on butterfly color using data collected by Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.



Figure 10. Effect of weather on butterfly activity using data collected by Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.



Figure 11. Effect of weather on flower color using data collected by Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.



Figure 12. Effect of year on butterfly color using data collected by BGO participants, Principles of Zoology students, and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.

Color Key= the color of most of the butterfly or flower... WH=white, YW=yellow, O=Orange, RD=red, BL=blue, BK=black, BR=brown, PL=purple, GR=green, PK=pink

SM=small (pencil eraser to watch face, or slightly bigger) M= medium (about the size of a key) LG= large (bigger than a key)

Latitude:	Longitue	de:	nny /partly cloudy /cloudy /ra	
Date:	Time:	Your	age:	Gender: F M O
Butterfly	Color	Size	Activity	Flower/plant color
1	WH YW O RD BL BK BR	SM M LG	Feed Fly Sit	WH YW O RD BL GR PL PK
2	WH YW O RD BL BK BR	SM M LG	Feed Fly Sit	WH YW O RD BL GR PL PK
3	WH YW O RD BL BK BR	SM M LG	Feed Fly Sit	WH YW O RD BL GR PL PK
4	WH YW O RD BL BK BR	SM M LG	Feed Fly Sit	WH YW O RD BL GR PL PK

Supplemental Figure 1. Survey sheet for Principles of Zoology and Animal Behavior students. Figure from Merrill & Hirzel et. al., 2021.