Leaf Color Polymorphism as a Mechanism of Within-individual Resource Partitioning in the Purple Pitcher Plant, *Sarracenia purpurea*

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Abstract

Resource partitioning occurs when two or more species have evolved traits that allow them to use a shared limiting resource in different ways, thereby reducing competition and promoting coexistence. In principle, the same phenomenon could occur between tissues of a single individual, if those tissues shared a limiting resource. In nitrogen deficient environments, like bogs, carnivorous plants such as *Sarracenia purpurea*, the purple pitcher plant, have evolved leaves modified to capture insects and acquire nitrogen from insects. Unlike most plants, whose leaves are nearly identical in appearance, individual pitcher plants have leaves (pitchers) that differ substantially in relative amounts of red and green coloration, *i.e.* they display leaf color polymorphism. This study addresses the possibility that pitcher plants use leaf color polymorphism as a mechanism of within-individual resource partitioning.

I sampled the contents and photographed the hoods of 31 *S. purpurea* (five pitchers per plant) in Mud Lake Bog in Cheboygan, MI in order to determine the relationship between within-plant color variation and the biomass and types of prey captured. Plants with greater leaf color polymorphism captured significantly more overall prey biomass, hymenopteran biomass, and dipteran biomass, and more species of Hymenoptera, Coleoptera, Hemiptera, and Arachnida. In no case was biomass or number of species negatively correlated with within-plant color variation, suggesting that there is no substantial cost of leaf color polymorphism. These results suggest that purple pitcher plants have pitchers that vary in color, at least in part, because such variation is a mechanism of within-individual resource partitioning

Introduction

Resource partitioning is the division, usually by two or more species, of a shared, limiting resource. It is widely accepted that resource partitioning allows species with similar resource requirements to coexist by reducing competition (Schoener 1974). Many examples exist of sympatric species using a shared resource in different ways. For instance, MacArthur (1958) found that five North American warbler species forage at different heights in spruce trees, presumably to avoid competing for the same prey.

Resource partitioning may also occur between individuals of the same species. For example, brown trout individuals feed at different times based on social rank. Dominant individuals feed at more favorable times while inferior individuals feed at less desirable times. This temporal division of food resources allows brown trout to use the same limited resources with reduced competition (Alanärä et al. 2001). Certain bumblebee species also exhibit intraspecific resource partitioning (Johnson 1986). Individual bumblebees partition flowers based on size matching between the flower corolla and the bee’s tongue length. In rare cases, resource partitioning has been described between genotypes within a population. For instance, Hunt et al. (2008) examined the distribution patterns of genotypes of bacterioplankton from the family

Vibrionaceae and found that genotypic clusters had developed distinct microhabitat preferences that allowed them to partition dissolved nutrients and organic particles based on size, distribution season, and whether they were free-living or associated with suspended particles or zooplankton. In principle, resource partitioning could also occur between different tissues within a single individual, if those tissues share a limiting resource. We are aware of only a single example of within-individual resource partitioning. Some studies have suggested that in some tree species, sun and shade leaves differ in size, pigment, and lobe or leaf margin patterns (Givnish 1988; McMillen & McClendon 1979; Murphy et al. 2012; Talbert & Holch 1957).

Within-individual resource partitioning may be more common for species that 1) live in resource deficient environments and 2) have more than one body part involved in gathering the limiting resource. Bogs are particularly low in nitrogen, and contain a disproportionate number of carnivorous species that use their leaves to capture nitrogen by catching insects and other invertebrates (Juniper et al. 1989). The leaves of the carnivorous purple pitcher plant, *Sarracenia purpurea*, are unusual in two ways. First, unlike the leaves of most plants, which have evolved to capture carbon and are nearly uniformly green, the leaves of pitcher plants have evolved to capture both carbon and nitrogen; as a consequence, individual leaves have both green and red tissue (Figure 1). Second, unlike the leaves of most plants, which are nearly identical in appearance, the leaves of pitcher plants are often highly variable in appearance, ranging from mostly green to mostly red on the same plant. Green tissue is used for photosynthesis, or carbon capture, while red tissue promotes nitrogen by attracting insects (Joel & Gepstein 1985). It is possible that, by having leaves (hereafter referred to as pitchers) that vary in color and are therefore attractive to different types of prey, individual plants can minimize competition among pitchers for the same prey types and thereby increase overall nitrogen capture by the plant.

Figure 1. Individual *Sarracenia purpurea* pitchers contain both red and green tissue

A variety of pitcher characteristics, such as color (the primary focus of this study), nectar, UV reflectance patterns, hood size, venation, and aperture size, are thought to influence prey capture (Bennett & Ellison 2009; Cresswell 1991; Karowe & Lopez-Nieves, unpublished;

Newell & Nastase 1998; Schaefer & Ruxton 2008). Plants that have more red coloration and red venation capture more prey (Nastase & Newell 1998; Schaefer & Ruxton 2007). Although these studies suggest that red coloration increases for prey capture, they do not explain why pitchers on the same plant vary, often considerably, in relative amounts of red and green coloration. In principle, if one level of redness is optimal for capturing prey, all pitchers on a plant should be that single optimal color.

Several studies have suggested that pitcher morphology, in place of or in addition to coloration, influences prey capture. Creswell found that pitchers with larger openings capture more prey biomass (1993). Similarly, larger pitchers tended to capture more prey biomass (Creswell 1993; Green and Horner 2007; Heard 1998). In contrast, Newell and Nastase (1998) did not find a significant relationship between overall pitcher size, pitcher opening size, and prey capture. Instead they suggest that color, rather than size or shape, affects prey capture.

As mentioned, pitcher plants are unusual in that leaves (pitchers) on the same plant vary in red and green coloration; they exhibit within-plant leaf color polymorphism. One potential explanation is that color polymorphism allows pitchers on the same plant to avoid competing among themselves for similar types of prey. For instance, greener pitchers on a plan may capture certain types of prey while redder pitchers on the same plant may capture other types. If so, then color polymorphism would allow the plant to capture a wider variety of prey types, and thereby maximize plant-wide nitrogen acquisition.

Following the same logic, if resource partitioning occurs among pitchers within an individual plant, plants with greater leaf color polymorphism should attract more overall biomass and/or more types of prey. In fact, the extent of within-individual color variation varies among plants within a population. Some plants are less variable; all of their pitchers have a similar red to green color ratio. Other plants are more variable; their pitchers with quite different red to green color ratios. Accordingly, this study addresses the possibility that individual plants use color variation between pitchers as a mechanism of within-individual resource partitioning by asking:

1. Do plants with more color variation among their pitchers capture more types of prey overall?
2. Do plants with more color variation among their pitchers capture more prey biomass?
3. How do different elements of pitcher size impact prey capture?
4. Do plants with more size variation capture more types of prey or more prey biomass?

Materials and Methods

Study Species

Sarracenia purpurea, the purple pitcher plant, is one of over 600 species of carnivorous plants worldwide (Schnell 2002). It has pitcher shaped leaves that trap prey in collected rainwater (Juniper et al. 1989). Pitcher plants are typically found in nitrogen-limited environments, like bogs (Juniper et al. 1989). *Sarracenia purpurea* occurs throughout the northeastern United States, the Great Lakes region, and southern Canada. Although it is the most widely distributed pitcher plant species, *S. purpurea* is threatened or endangered in several states including Florida, Georgia, Illinois, and Maryland (USDA 2012).

Prey are attracted by the pitcher hood as well as nectar on the lip, or peristome, of the pitcher opening (Bennett & Ellison 2009; Newell & Nastase 1998). Small hairs on the pitcher hood create difficult walking conditions for prey, causing them to fall into the pitcher. These downward pointing hairs, together with slippery waxy pitcher walls make it difficult for insects to crawl out; consequently, they often drown in the water (Newell & Nastase 1998). In New England, pitcher plants derive approximately 50% of their nitrogen from prey capture (Ellison & Gotelli 2001); a similar value was observed in my study site in northern Michigan (Karowe & Foss-Grant unpublished data). Unlike those of most pitcher plant species, pitchers of *S. purpurea* do not secrete digestive enzymes; rather they rely on a community of decomposers, or inquilines, within the pitcher to break down prey and make nutrients available to the plant (Juniper et al. 1989).

Study System

For this study, 31 *S. purpurea* were sampled in Mud Lake Bog in Cheboygan County, MI (46° 61'N 84°59'W) during the summer of 2012. In Mud Lake Bog, mats of *Sphagnum* moss create acidic conditions with an average pH of 3.25 (Glassman & Karowe, in review; Small 1972). Plants were chosen to include the wide range of within-plant leaf color polymorphism present in Mud Lake Bog. Based on visual inspection, plants were initially determined to have a low, medium, or high level of within-plant leaf color polymorphism. Within plants with low color polymorphism, pitchers were similar in coloration, while plants with high color polymorphism contained pitchers that differed considerably in red vs. green coloration. Within each plant, five pitchers that as a group reflected that plant's level of color polymorphism were chosen for more detailed color analysis (described below). Sampled pitchers were restricted to those that appeared to be actively capturing prey. Pitchers with dried hoods, holes, and tears were excluded from sampling, as were those covered by spider webs, since spider webs may reduce prey capture by up to 10% (Hart et al. 2009).

Effect of within-plant leaf color polymorphism on prey capture

Color determination

At the start of the study, the hood of each of five pitchers was photographed to determine the degree of color variability within each individual plant. Adobe Photoshop was used to quantify the percent red on each pitcher hood. The Quick Selection tool was used to select the entire pitcher hood, extending from the top of the pitcher hood to the lower edge of the area covered by hairs (Figure 2, left).

Figure 2. (left) Area of pitcher hood used to find the total number of pixels (total amount of green and red) and (right) - area, within the dashed lines, selected by Photoshop and quantified as red.

The total number of pixels within the selected area, displayed in the Histogram window, was recorded. Using the Histogram window, the 25th percentile red value was also recorded. The Select → Color Range tool was used to quantify the percent red on each pitcher hood, using the 25th percentile red value as a guide. The fuzziness feature allows the operational definition of "red" to include a range of color values on either side of the 25th percentile red value. A fuzziness of 200 was used because it produced the closest correspondence between the area selected by Photoshop and the area seen as red by the researcher. The range feature was set to 100 to allow all areas of the selection to be analyzed by the Color Range tool. The eyedropper tool was used to select the 25th percentile red value on the pitcher hood. The color range feature automatically selects all sections of the pitcher hood within the range of the 25th percentile red value (Figure 2, right). Percent red was calculated by dividing the number of red pixels by the total number of pixels on the pitcher hood. Within-plant color variation was quantified as range (highest – lowest) and standard deviation of percent red among the five pitchers on each plant. A range from 10-20% red might have a different effect on prey capture than a range from 70-80% even though the ranges for such plants are equal. For this reason, relative range and relative standard deviation were calculated for each plant by dividing the range and standard deviation in percent red by the mean percent red. It is possible that other pitcher characteristics might influence prey capture (Creswell 1993; Green and Horner 2007; Heard 1998). For this reason total pitcher length from the bottom of the keel to the top of the hood, aperture width and length, hood width and height, and keel width were also measured (Figure 3).

Figure 3. Pitcher total length (left), pitcher opening length and width (middle), and pitcher hood width and height (right); measured in millimeters.

Prey collection and identification

The water and prey contents of each pitcher were removed using a turkey baster. To ensure that all prey were collected, a 10cc syringe was used to push de-ionized water into the base of each pitcher to facilitate prey collection with the turkey baster. The contents of each pitcher were filtered onto a Whatman #1 filter paper using a vacuum filter, and the prey were placed in 70% EtOH. The liquid filtrate from all pitchers was combined, divided equally, and replaced into the pitchers sampled. Insects with at least a head were identified using a dissecting microscope. If no heads were present in the sample, prey bodies were used. Prey heads and bodies were counted and identified to the order, family, or genus level where possible.

Prey composition was quantified by identifying the total number of prey types captured by a plant as well as the number of different orders and families represented among the prey captured by a plant. After identifying the prey contents of each pitcher, samples were dried and weighed using a Mettler Toledo XS205 balance. Weights for each order as well as overall prey biomass were calculated for each pitcher and plant.

Statistical Analysis

Both parametric Pearson's and nonparametric Spearman's correlation analyses were used to assess the relationships among measures of color variability on a plant and each measure of variation in prey types captured by pitchers on the same plant. Both sets of tests produced approximately the same results, but Spearman's rho generally detected stronger relationships between plant and prey variables. Since QQ plots revealed that the majority of plant and biomass variables departed from normality, nonparametric Spearman's correlations were used for most analyses.

Because the effect of within-plant colors variation was of particular interest, and because average percent red was significantly positively correlated with both measures of color variation (range and standard deviation of percent red), stepwise regression was also conducted using only pitcher color variables. To determine whether the apparent effect of color variation was simply due to its positive association with average color (i.e. plants that had redder pitchers on average also had more variable pitchers), stepwise regressions were first conducted with mean percent red forced in at the first step. This approach removed the effect of average redness before testing for an effect of variation in redness.

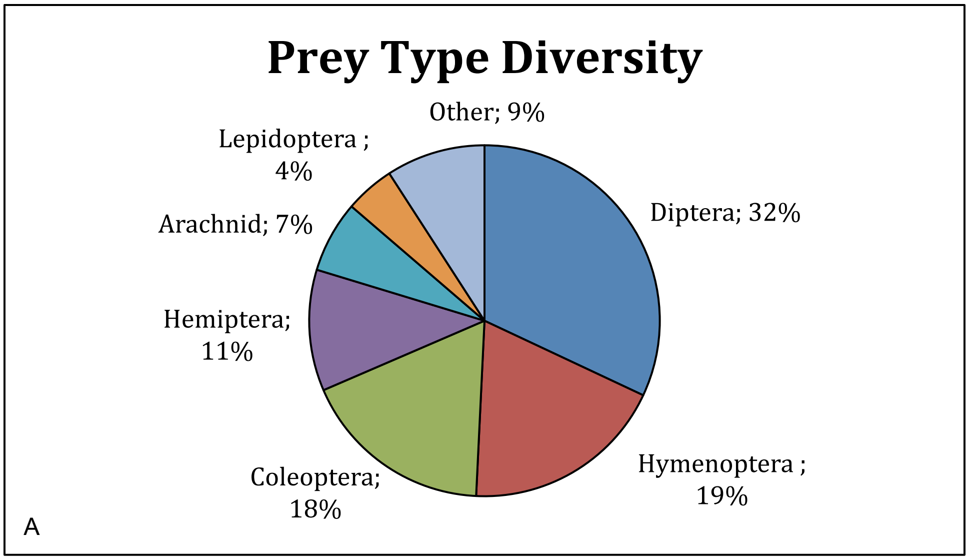
Principle components analysis was also used to determine the effect of all 17 color and shape variables independent of each other. Initially 10 principal components were created from these 17 variables. Only the six with values greater than one were used in multiple regression analyses.

Results

Prey distribution summary

In total, 1502 prey individuals were collected from the 155 sampled pitchers. There were 197 unique prey types (hereafter "species") and 16 orders identified in the samples. The 197 identified prey species included 63 fly species, 21 ant species, 16 species of other hymenopterans, 35 beetle species, 22 hemipteran species, 13 arachnid species, and 9 lepidopteran species (Figure 4A). The most common orders by number of individuals were Diptera (flies), Hymenoptera (ants and wasps), Coleoptera (beetles), Hemiptera (true bugs), and Araneae (spiders and mites). The most common prey types were mites, calyptrate muscoid flies, chironomid midges, and chrysomelid leaf beetles. Least common prey types included thrips, bark lice, millipedes, and grasshoppers. On average, 10 prey individuals were identified per pitcher, and 48 prey individuals from 19 prey species were identified per plant.

In total, 639.26 mg of prey was collected during sampling. Hymenopterans accounted for the largest amount of prey biomass with a total of 200.00 mg, followed by Dipterans (144.00 mg) and Coleopterans (123.00 mg; Figure 4B). On average, pitchers captured 4.12 mg of prey; plants on average captured 20.62 mg of prey.



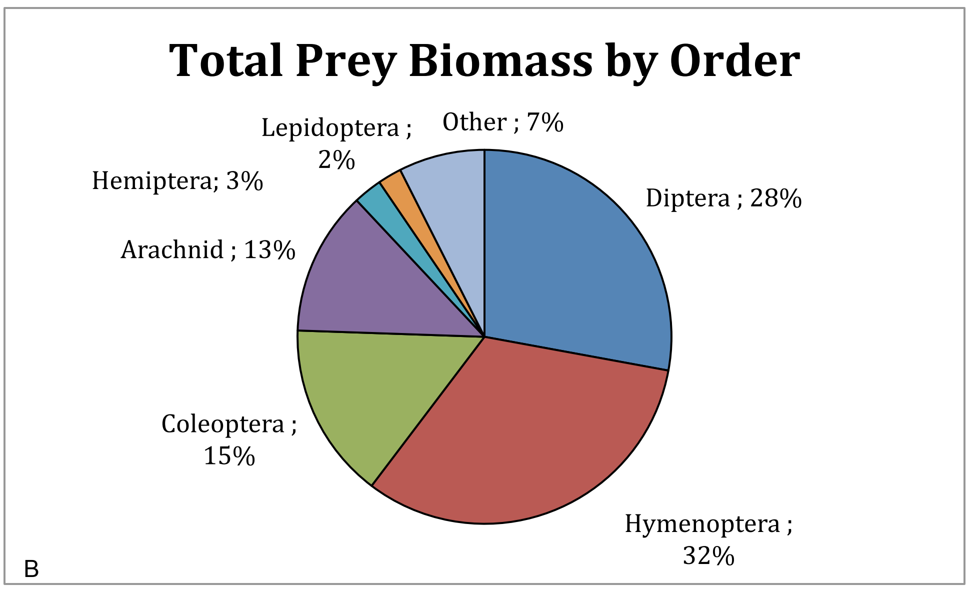


Figure 4. Percentages of total number of prey species captured (A) percentages of prey biomass captured by order (B).

Effect of color and color variation on prey capture

Prey Biomass

Plants with more color variation among their pitchers captured more prey overall (Table 1), indicated by highly significant positive correlations between total prey biomass and both measures of within-plan color variation: range in percent red (Rho = 0.59, p < 0.0005; Figure 5) and standard deviation of percent red (Rho = 0.54, p = 0.002). Range and standard deviation of percent red were also significantly positively correlated with biomass of two prey orders: Diptera (Rho = 0.46, p = 0.009 for both) and Hymenoptera (Rho = 0.55, p = 0.002, Figure 5, and Rho = 0.48, p = 0.007, respectively). Together, these two orders accounted for 60% of all prey biomass. Biomass of captured Coleoptera, Arachnida, and Hemiptera were not significantly correlated with either measure of within-plant color variation (Table 1).

Both total prey biomass and dipteran biomass were also significantly positively correlated, albeit not as strongly, with the average percent red of pitchers within a plant (Rho = 0.43, p = 0.015, Figure 4, and Rho = 0.39, p = 0.042; Table 1). Biomass of captured Hymenoptera (Figure 6), Coleoptera, Arachnida, and Hemiptera were not significantly correlated with average percent red (Table 1).

Table 1. Relationships between measures of within-plant color (mean, range, and standard deviation of percent red) and measures of captured prey biomass. All Rho and p-values are from Spearman rank correlations.

Redness measure		Total prey biomass	Diptera biomass	Hymenoptera biomass	Coleoptera biomass	Hemiptera biomass	Arachnid biomass
Range	Rho	0.593	0.460	0.545	0.037	0.003	0.284
	p-value	**>0.0005**	**0.009**	**0.002**	0.845	0.988	0.122
Standard deviation	Rho	0.541	0.463	0.477	-0.024	0.062	0.233
	p-value	**0.002**	**0.009**	**0.007**	0.898	0.741	0.207
Average	Rho	0.434	0.368	0.330	-0.069	0.134	0.250
	p-value	**0.015**	**0.042**	0.070	0.714	0.472	0.176

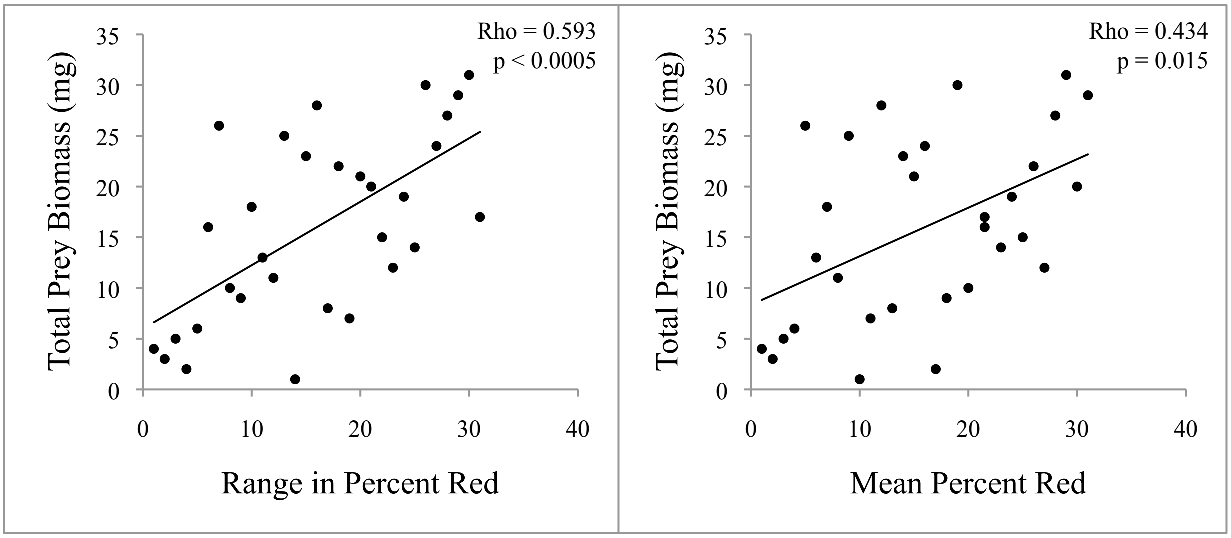


Figure 5. Relationship between total prey biomass and range in percent red (left) and total prey biomass and mean percent red (right). Rho and p-values are from Spearman's rank correlations.

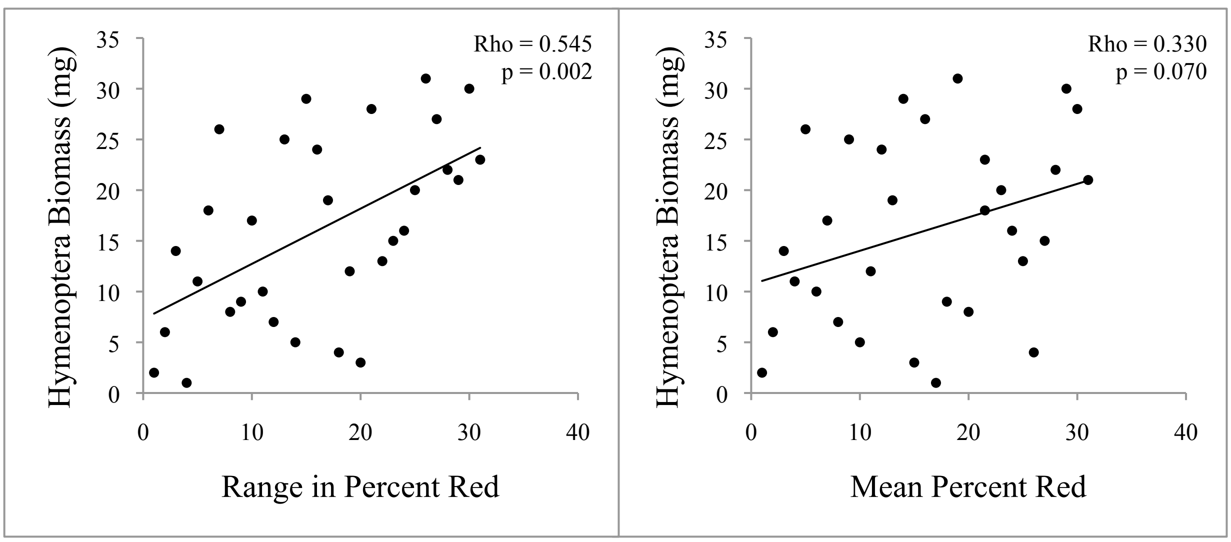


Figure 6. Relationship between Hymenoptera biomass and range in percent red (left) and Hymenoptera biomass and mean percent red (right). Rho and p-values are from Spearman's rank correlations.

Prey species

Plants with more color variation among their pitchers did not capture more prey types overall (Table 2), indicated by lack of significant positive correlations between plant-wide number of prey species and both measures of within-plan color variation: range of percent red (Rho = 0.25, p = 0.18) and standard deviation of percent red (Rho = 0.21, p = 0.26). However, color variation appeared to influence the number of species captured within four prey orders: range and standard deviation of percent red were significantly positively correlated with number of species of Hymenoptera (Rho = 0.39, p = 0.032 and Rho = 0.37, p = 0.043, respectively), Coleoptera (Rho = 0.41, p = 0.020 and Rho = 0.38, p = 0.034, respectively), Hemiptera (Rho = 0.48, p = 0.006 and Rho = 0.44, p = 0.014, respectively), and Arachnida (Rho = 0.47, p = 0.007 and Rho = 0.41, p = 0.024, respectively) (Figure 7). In contrast, range and standard deviation of percent red were not significantly positively correlated with the number of Diptera species (Rho = 0.30, p = 0.10 and Rho = 0.27, p = 0.14, respectively).

The number of species within these four orders was also significantly positively correlated with the average percent red of pitchers within a plant (Hymenoptera, Rho = 0.37, p = 0.043; Coleoptera, Rho = 0.43, p = 0.016; Hemiptera, Rho = 0.47, p = 0.008; Arachnida, Rho = 0.44, p = 0.014; Table 2). In contrast, the total number of prey species and number of Diptera species were not significantly correlated with average percent red (Table 2).

Table 2. Relationships between measures of within-plant color (mean, range, and standard deviation of percent red) and measures of captured prey species. All Rho and p-values are from Spearman rank correlations.

Redness measure		Total prey species	Diptera species	Hymenoptera species	Coleoptera species	Hemiptera species	Arachnid species
Range	Rho	0.245	0.299	0.385	0.414	0.482	0.472
	p-value	0.184	**0.102**	**0.032**	**0.020**	**0.006**	**0.007**
Standard deviation	Rho	0.210	0.268	0.366	0.383	0.436	0.405
	p-value	0.258	0.144	**0.043**	**0.034**	**0.014**	**0.024**
Average	Rho	0.125	0.245	0.366	0.428	0.466	0.438
	p-value	0.503	0.183	**0.043**	**0.016**	**0.008**	**0.014**

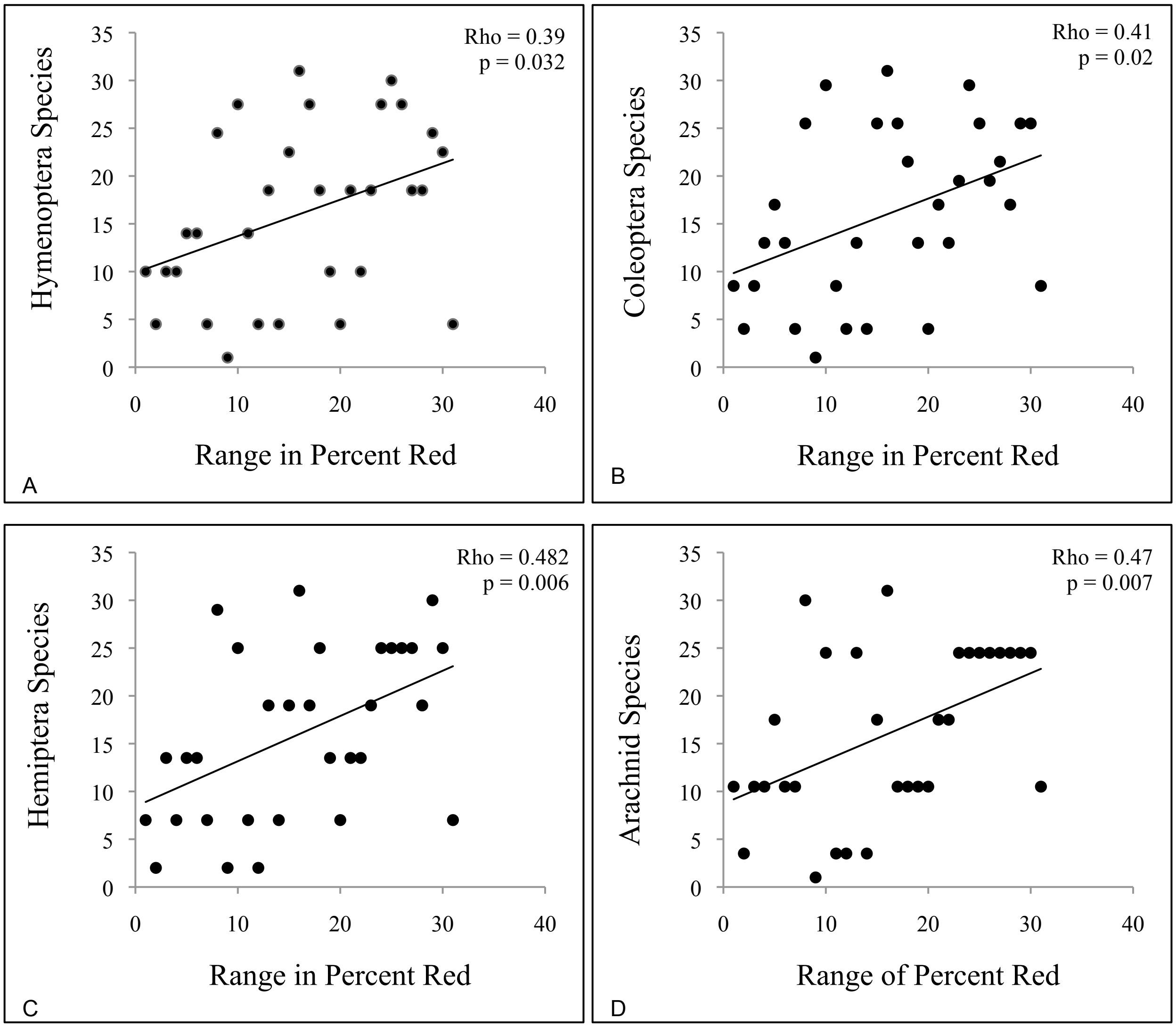


Figure 7. Relationship between range in percent red and Hymenoptera species (A), Coleoptera species (B), Hemiptera species (C), Arachnid species (D). Rho and p-values are from Spearman's rank correlations.

Effect of average pitcher size and size variation on prey capture

Prey biomass

Plants with more size variation among their pitchers did not capture more prey biomass (Table 3), indicated by lack of significant correlations between total prey biomass and all measures of plant size and shape variability. Total prey biomass and hymenopteran biomass were nearly significantly negatively correlated with average total pitcher length (Rho = -0.34, p = 0.064 for both), suggesting that shorter pitchers tend to capture more prey. However, biomass of captured Diptera, Hymenoptera, Coleoptera, Arachnida, and Hemiptera were not significantly correlated with any measure of plant size or within-plant size variation (Table 3).

Table 3. Relationships between measures of average plant size and within-plant size variation measures of captured prey biomass. All R and p-values are from Spearman rank correlations.

Size measure		Total prey biomass	Diptera biomass	Hymenoptera biomass	Coleoptera biomass	Hemiptera biomass	Arachnid biomass
Average total	Rho	-0.337	-0.253	-0.337	0.055	0.018	-0.038
length	p-value	**0.064**	0.170	**0.064**	0.768	0.923	0.838
Total length	Rho	-0.135	-0.049	-0.067	0.120	-0.065	0.071
variance	p-value	0.470	0.794	0.722	0.520	0.726	0.703
Average hood	Rho	-0.286	-0.207	-0.148	0.052	-0.263	-0.057
width	p-value	0.119	0.263	0.428	0.780	0.152	0.763
Hood width	Rho	-0.072	0.168	-0.107	-0.154	-0.110	-0.133
variance	p-value	0.701	0.367	0.566	0.409	0.555	0.475
Average hood	Rho	-0.095	0.011	0.026	-0.013	-0.119	0.064
length	p-value	0.610	0.952	0.889	0.946	0.523	0.731
Hood length	Rho	0.062	0.190	0.116	-0.029	0.009	-0.009
variance	p-value	0.742	0.306	0.533	0.875	0.960	0.963
Average opening	Rho	-0.134	-0.120	-0.023	-0.056	-0.163	0.007
width	p-value	0.471	0.522	0.901	0.763	0.381	0.970
Opening width	Rho	-0.225	0.070	-0.253	-0.333	-0.086	-0.191
variance	p-value	0.224	0.708	0.170	0.067	0.644	0.305
Average opening	Rho	-0.016	0.095	0.296	0.197	-0.077	0.018
length	p-value	0.933	0.609	0.106	0.288	0.682	0.923
Opening length	Rho	-0.004	0.047	0.182	0.098	0.204	0.103
variance	p-value	0.984	0.800	0.326	0.600	0.272	0.580
Average keel	Rho	0.052	0.153	0.059	0.088	0.022	-0.021
width	p-value	0.782	0.413	0.752	0.638	0.904	0.912
Keel width	Rho	-0.051	-0.084	0.028	0.115	-0.010	0.001
variance	p-value	0.787	0.653	0.880	0.539	0.958	0.994

Prey Species

Unlike mean and variation in pitcher color, mean and variation in pitcher size did not appear to influence the number of prey species captured, indicated by a lack of significant correlations between all plant-wide measures of prey species captured and all measures of within-plant average size and size variation (Table 4). The only correlations that approached significance were between average hood width and the number of hemipteran and arachnid species; in both cases, the correlations were negative.

Table 4. Relationships between measures of average plant size and within-plant size variation measures of captured prey species. All R and p-values are from Spearman rank correlations.

Size measure		Total prey species	Diptera species	Hymenoptera species	Coleoptera species	Hemiptera species	Arachnid species
Average total	Rho	-0.025	-0.197	-0.177	-0.145	-0.223	-0.286
length	p-value	0.893	0.288	0.341	0.435	0.229	0.118
Total length	Rho	-0.087	0.060	0.041	0.124	0.039	-0.016
variance	p-value	0.641	0.748	0.828	0.506	0.835	0.931
Average hood	Rho	-0.028	-0.179	-0.201	-0.139	-0.315	-0.325
width	p-value	0.881	0.335	0.279	0.456	**0.084**	**0.075**
Hood width	Rho	-0.280	-0.287	-0.235	-0.293	-0.274	-0.104
variance	p-value	0.127	0.117	0.203	0.110	0.135	0.579
Average hood	Rho	0.073	-0.079	0.013	0.089	-0.072	-0.089
length	p-value	0.696	0.672	0.946	0.633	0.700	0.633
Hood length	Rho	0.037	-0.004	0.108	0.004	0.033	0.114
variance	p-value	0.842	0.982	0.561	0.982	0.859	0.541
Average opening	Rho	-0.053	-0.024	-0.096	-0.001	-0.167	-0.143
width	p-value	0.778	0.897	0.607	0.996	0.370	0.443
Opening width	Rho	-0.330	-0.282	-0.236	-0.147	-0.227	-0.219
variance	p-value	0.070	0.124	0.200	0.429	0.219	0.238
Average opening	Rho	0.204	0.224	0.100	0.088	-0.033	-0.049
length	p-value	0.271	0.227	0.594	0.639	0.858	0.795
Opening length	Rho	0.046	-0.125	-0.127	-0.118	-0.112	-0.056
variance	p-value	0.806	0.504	0.494	0.529	0.550	0.765
Average keel	Rho	-0.007	-0.082	-0.063	-0.005	-0.001	-0.268
width	p-value	0.970	0.660	0.737	0.978	0.995	0.145
Keel width	Rho	-0.073	0.075	-0.069	0.041	0.015	-0.079
variance	p-value	0.696	0.688	0.713	0.825	0.936	0.672

Correlations among independent variables

Plant average percent red was highly correlated with both measures of within-plant color variation: range in percent red (Rho = 0.74, $p < 0.0005$; Table 5) and standard deviation of percent red (Rho = 0.73, $p < 0.0005$; Table 5). Plant range and plant standard deviation were also highly correlated (Rho = 0.98, $p < 0.0005$; Table 5). Range and standard deviation of percent red were significantly negatively correlated with two measures of plant size and shape: average total pitcher length (Rho = -0.63, $p < 0.0005$ and Rho = -0.61, $p < 0.0005$, respectively) and average pitcher hood width (Rho = -0.44, p = 0.012 and Rho = -0.45, p = 0.010, respectively). Averages of hood length, opening width, opening length, and keel width were not significantly correlated with either measure of within-plant color variation (Table 5). Variances of total length, hood width, hood length, opening width, opening length, and keel width were also not significantly correlated with either measure of within-plant color variation (Table 5). Overall, redder pitchers tended to be longer with larger hoods.

Both average total pitcher length and average pitcher hood width were also significantly negatively correlated with the average percent red of pitchers within a plant (Rho = -0.69, $p < 0.0005$, and Rho = -0.46, p = 0.009; Table 5). All other size and shape metrics were not significantly correlated with average percent red (Table 5). There were several significant correlations between measures of pitcher size (Table 6). Average pitcher length was significantly positively correlated with average hood width (Rho = 0.59, p = 0.001; Table 6), average hood length (Rho = 0.46, p = 0.010; Table 6), average opening width (Rho = 0.38, p = 0.033; Table 6), and average opening length (Rho = 0.44, p = 0.014; Table 6). Overall, longer pitchers tended to have larger hoods and pitcher openings.

Table 5. Spearman's correlations among measures of pitcher color variability and measures of size and shape.

		Average	Standard deviation	Range
Plant	Rho		0.726	0.741
	p-value	.	**<0.0005**	**<0.0005**
Standard deviation	Rho	0.726		0.981
	p-value	**<0.0005**	.	**<0.0005**
Range	Rho	0.741	0.981	
	p-value	**<0.0005**	**<0.0005**	.
Average total length	Rho	-0.693	-0.606	-0.630
	p-value	**<0.0005**	**<0.0005**	**<0.0005**
Total length variance	Rho	-0.210	-0.281	-0.283
	p-value	0.256	0.126	0.123
Average hood width	Rho	-0.462	-0.454	-0.444
	p-value	**0.009**	**0.010**	**0.012**
Hood width variance	Rho	0.092	-0.082	-0.059
	p-value	0.624	0.659	0.751
Average hood length	Rho	-0.155	-0.049	-0.057
	p-value	0.406	0.794	0.760
Hood length variance	Rho	0.241	-0.032	-0.020
	p-value	0.192	0.863	0.917
Average opening width	Rho	-0.212	-0.285	-0.253
	p-value	0.253	0.121	0.170
Opening width variance	Rho	0.350	-0.030	-0.053
	p-value	0.054	0.873	0.775
Average opening length	Rho	-0.285	-0.150	-0.135
	p-value	0.120	0.421	0.469
Opening length variance	Rho	-0.170	-0.256	-0.236
	p-value	0.360	0.164	0.201
Average keel width	Rho	-0.012	0.076	0.096
	p-value	0.949	0.683	0.608
Keel width variance	Rho	-0.058	0.118	0.109
	p-value	0.758	0.526	0.559

Table 6. Spearman's correlations of pitcher size and shape metrics.

Size measures		Average total length	Total length variance	Average hood width	Hood width variance	Average hood length	Hood length variance	Average opening width	Opening width variance	Average opening length	Opening length variance	Average keel width	Keel width variance
Average total length	Rho		0.379	0.585	-0.282	0.458	-0.158	0.383	-0.254	0.439	0.308	0.202	0.005
	p-value	.	**0.035**	**0.001**	0.124	**0.010**	0.396	**0.033**	0.169	**0.014**	0.092	0.275	0.977
Total length variance	Rho	0.379		0.200	-0.061	0.191	0.180	0.281	0.235	0.005	0.215	0.011	0.243
	p-value	**0.035**	.	0.281	0.745	0.303	0.331	0.126	0.203	0.977	0.246	0.954	0.188
Average hood width	Rho	0.585	0.200		-0.094	0.775	0.020	0.799	0.081	0.552	-0.051	0.298	-0.124
	p-value	**0.001**	0.281	.	0.615	**<0.0005**	0.915	**<0.0005**	0.665	**0.001**	0.785	0.104	0.507
Hood width variance	Rho	-0.282	-0.061	-0.094		-0.122	0.549	0.021	0.386	-0.311	-0.054	-0.115	-0.319
	p-value	0.124	0.745	0.615	.	0.513	**0.001**	0.912	**0.032**	0.088	0.771	0.536	0.081
Average hood length	Rho	0.458	0.191	0.775	-0.122		0.124	0.708	0.017	0.574	-0.026	0.272	-0.184
	p-value	**0.010**	0.303	**<0.0005**	0.513	.	0.507	**<0.0005**	0.926	**0.001**	0.888	0.139	0.323
Hood length variance	Rho	-0.158	0.180	0.020	0.549	0.124		0.101	0.437	-0.133	0.170	-0.065	-0.339
	p-value	0.396	0.331	0.915	0.001	0.507	.	0.589	**0.014**	0.477	0.359	0.727	0.062
Average opening width	Rho	0.383	0.281	0.799	0.021	0.708	0.101		0.100	0.408	-0.143	0.235	-0.168
	p-value	**0.033**	0.126	**<0.0005**	0.912	**<0.0005**	0.589	.	0.591	**0.023**	0.443	0.203	0.365
Opening width variance	Rho	-0.254	0.235	0.081	0.386	0.017	0.437	0.100		-0.368	-0.014	0.051	-0.024
	p-value	0.169	0.203	0.665	**0.032**	0.926	**0.014**	0.591	.	**0.042**	0.939	0.787	0.897

Average opening length	Rho	0.439	0.005	0.552	-0.311	0.574	-0.133	0.408	-0.368		0.269	0.184	-0.109
	p-value	**0.014**	0.977	**0.001**	0.088	**0.001**	0.477	**0.023**	**0.042**	.	0.143	0.323	0.560
Opening length variance	Rho	0.308	0.215	-0.051	-0.054	-0.026	0.170	-0.143	-0.014	0.269		-0.096	-0.033
	p-value	0.092	0.246	0.785	0.771	0.888	0.359	0.443	0.939	0.143	.	0.606	0.858
Average keel width	Rho	0.202	0.011	0.298	-0.115	0.272	-0.065	0.235	0.051	0.184	-0.096		0.282
	p-value	0.275	0.954	0.104	0.536	0.139	0.727	0.203	0.787	0.323	0.606	.	0.125
Keel width variance	Rho	0.005	0.243	-0.124	-0.319	-0.184	-0.339	-0.168	-0.024	-0.109	-0.033	0.282	
	p-value	0.977	0.188	0.507	0.081	0.323	0.062	0.365	0.897	0.560	0.858	0.125	.

Because several color and shape measures were highly correlated with each other, two multivariate techniques were used to assess the relative importance of pitcher characteristics on prey capture. Stepwise regression with all 17 color, size, and shape variables identified only range in percent red as a significant predictor of total prey biomass ($R = 0.44$, $p <0.012$). However, other variables were identified as significant predictors of biomass of specific prey orders. Dipteran biomass was significantly correlated with plant average percent red ($R = 0.42$, $p = 0.018$). Hymenopteran biomass was significantly positively correlated with range of percent red ($p = 0.020$) and significantly negatively correlated with standard deviation of percent red ($p = 0.050$); together, these two variables explained 32.8% of the variation in hymenopteran biomass. In addition, biomass of "other" prey was significantly positively correlated with standard deviation of hood length ($R = 0.072$, $p < 0.0005$).

Stepwise regressions with average percent red forced in at the first step indicated that, after the effect of average percent red was removed, range in percent red was a nearly significant predictor of total prey biomass ($p = 0.067$). Together the two variables together explained substantially more variation in prey biomass than did mean percent red alone (20% vs. 9%). However, when the effect of range in percent red was removed by forcing it in at step one, average percent red was not a significant predictor of total prey biomass after the effect of range in percent red was removed at step one ($p = 0.99$), and the two variables together explained no more variation in prey biomass than did range in percent red alone (20% vs. 20%).

Principal components analysis using all 17 color and shape variables indicated that only one principal component (PC2) was nearly significantly correlated with total prey biomass. PC2 had large positive loadings for all four measures of color variation (range and standard deviation in percent red, and range/mean and standard deviation/mean), and a large negative loading for standard deviation of opening width. Mean percent red had a small negative loading on PC2. Therefore, both stepwise regression and principal components analysis suggest that pitcher plants with more variable coloration, independent of average pitcher redness, capture more total prey biomass.

Discussion

The results of this study provide the first evidence that pitcher plants appear to use leaf color polymorphism as a mechanism of within-plan resource partitioning. Plants with more variable pitchers captured more prey in the orders Hymenoptera, Coleoptera, Hemiptera, and Arachnida, but not Diptera. As a consequence, plants with pitchers that vary more in color capture more biomass overall. This is one of the few examples of within-individual resource partitioning and, to our knowledge, the first demonstration of this phenomenon for any resource other than light (Givnish 1988; McMillen & McClendon 1979; Murphy et al. 2012; Talbert & Holch 1957).

While this study did not address prey behaviors that could result in resource partitioning, several possibilities exist. Many Hymenoptera, Coleoptera, and Hemiptera visit flowers for nectar and pollen (Bennett & Ellison 2009; Bernhardt 2000; Larson et al. 2001). It is possible that individual flower-visiting species differ in the amount of red they find most attractive. For instance, one hymenopteran species might be most attracted to pitchers that are 30% red while another hymenopteran species might be most attracted to pitchers that are 50% red. This could reflect similarity to the flower species each hymenopteran visits most frequently and/or finds most rewarding. Color variation could also enhance capture of individuals within a single prey species. For instance, given the tendency of individual pollinators to display constancy toward one flower type during a foraging bout, two wasps of the same species could be attracted to pitchers of different color, because the two wasps are currently exhibiting constancy to flowers that differ in redness (Chittka & Raine 2006; Chittka et al. 1999). If so, each individual wasp might be attracted to a pitcher of different redness. A similar scenario could explain the positive effect of color polymorphism on capture of Coleoptera and Hemiptera, some of which also forage on flowers.

It appears that flies respond to both variation in redness and average redness; plants with both redder pitchers and pitchers that display color variation capture more fly biomass and more fly species. Although red detection is likely the same between flies and other insects, higher average redness might attract more flies because, above a low threshold of redness, pitchers are essentially equally attractive. Carrion feeding flies, for instance, might be equally attracted to any level of redness because even slightly red pitchers resemble carrion. Alternatively, flies may be attracted by the odor of decaying prey within a pitcher, which would be correlated with prey biomass within a pitcher. This and other studies have show that average redness is correlated with total biomass. Our results also show that, while variation in redness is a better predictor or total prey biomass, average redness is very strongly correlated with variation in redness, and therefore also strongly correlated with total prey biomass. If this were the case, color variation would not affect behavior of these flies. It is plausible that plants use leaf color polymorphism to partition only fly species that are not carrion feeders. Our observation that fly biomass was positively correlated with both average redness and variation in redness may indicate that our fly sample consisted of both carrion feeding species that responded to average redness, and other fly species that responded to color variation.

It is also possible that plants actually do use leaf color polymorphism to partition fly species, but uncertainty in identification obscured this relationship. Although flies made up the largest portion of prey types found, they were the most difficult to identify. Often flies were fragmented and small. Different levels of digestion cause changes in prey color, which might have caused us to place the same type of fly into different categories. We were often unable to confidently place a species or family label on a fly specimen and generally sorted them into morpho-species. Perhaps this uncertainty obscured a real relationship between color variability and flies captured.

In addition to the total amount of red on a hood, the arrangement of red color could also that influence the types of prey captured (Schaefer & Ruxton 2008). Pitchers with greater red venation receive more visits from potential prey, and may have more successful captures (Newell & Nastase 1998). Striped veins along the pitcher hood guide prey into the dark and fatal pitcher (Biesmeijer et al. 2005). Other studies suggest, while venation is important, pitchers with contrasting venation color patterns capture more prey (Juniper et al. 1989; Moran et al. 1999). While pitcher venation generally appears red to the human eye, it may appear dark to prey that do not have photoreceptors to pick up on red pigmentation (Chittka & Waser 1997). Another possibility is that venation patters reflect UV light to attract insects. It has been suggested that insects are attracted to UV light (Craig and Bernard 1990). Perhaps pitchers on an individual plant exhibit different UV patterns on their hoods that attract different types of insects. Future studies should consider the influence of amount and pattern of red venation as well as other venation manipulations on prey capture.

All significant correlations between biomass or number of species and within-plant color variation were positive, suggesting that in terms of prey captured, there is no substantial cost of leaf color polymorphism. It is better for a pitcher plant to contain pitchers that vary in color.
All pitcher plants Mud Lake Bog display leaf color polymorphism, albeit at different levels. One possible explanation is that level of sunlight impacts color variation on a plant. I observed plants in both sun and shaded areas and found that plants with lower leaf color polymorphism typically occur in shaded areas of the bog. These plants also tended to be greener (Schnell 1979). Perhaps greener shaded plants reflect a tradeoff between carbon capture and carnivory; these plants need to devote more tissue to photosynthesis rather than prey capture. Perhaps light availability impacts both pitcher coloration and prey capture. These results suggest that, despite light availability, purple pitcher plants contain pitchers that vary in color, at least in part, because such variation is a mechanism of within-individual resource partitioning.

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