POPULATION SEX RATIOS AND GENDER SWITCHING IN JACK-IN-THE-PULPIT (*ARISAEMA TRIPHYLLUM*: ARACEAE)

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Project Summary

Diphasy is the ability for an individual plant to express a particular sexual mode in any given season based on circumstances rather than genetics (Schlessman 1988). This form of gender expression is common in the genus *Arisaema*, and has been widely documented in *Arisaema triphyllum*. (Shaffner 1922; Rust 1980; Bierzychudek 1982, 1984a, 1984b; Policansky 1987; Barriault 2010). Much of the research done on this species so far has focused on the relationship between size and gender expression in individual plants (Lovett-Doust and Cavers 1982; Bierzychudek 1984a; Policansky 1987; Vitt 2003). This study had two main objectives, the first of which was to investigate the role of population sex ratio in gender switching in an *A. triphyllum* population, while the second objective was to investigate the role of different environmental factors and management activities on plant size and population sex ratios in two different populations of *A. triphyllum*.

The first objective was studied by using pollen composition as a proxy for population sex ratio. 123 females were pollinated in the spring of 2009 using one of four pollination treatments: three hand pollination treatments (1. single male, 2. multiple male, and 3. natural pollination plus multiple male) and one natural pollination treatment. Each individual plant that was part of the study was marked with a number, and its gender and size were measured and recorded in the summer of 2009 and 2010. I hypothesized that females pollinated with pollen from a single male would be much more likely to be male in the following season than females pollinated with pollen from a mix of ten different males. There was no significant correlation found between pollination treatment and a plant's probability of changing gender in 2010. However, larger females were more likely to remain female, while smaller females were more likely to switch gender in 2010. To complete second objective I studied two different populations within

Florsheim Nature Preserve in Lincolnshire, IL, one that had a female biased sex ratio and one with a male biased sex ratio in 2009. Because light is often a limiting resource in understory plants, hemispherical photography was used to analyze canopy cover and light transmittance above 30 male, 30 female, and 30 vegetative plants in each population. Additionally, a vegetation survey was completed in order to categorize both the woody and herbaceous communities in each of the two populations. Management activities were qualitatively compared between the two populations based on information received from park staff. I hypothesized that both canopy structure and community composition would differ in populations with differing population sex ratios, and that light levels would be associated with plant size and gender, with increased light leading to larger, female plants. The canopy photos showed no significant difference in light availability between populations, but one population had a significant correlation between plant size and transmitted light. The vegetation survey showed very different herbaceous communities in each population, and also a difference in total canopy cover and tree basal area.

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Chapter 1 – Sex Ratio and Gender Switching in *Arisaema triphyllum* (Jack-in-the-Pulpit)

Introduction

Diphasy is defined as a sexual system in which individuals belong to a single genetic class but express a particular gender in any one season according to circumstances (Schlessman 1988). Gender switching, or the ability for an individual organism to reproduce as a male in one breeding season and as a female in the next, is a reproductive strategy most commonly seen in the genus *Arisaema* (Clay 1993). The most common hypothesis describing gender switching in plants states that if the efficiency of reproduction as a male or female varies with age or size, then an individual that could change to the more advantageous sex would be more fit than an individual that must remain the same sex throughout its life (Schlessman 1988). This theory is based on the size advantage model that was first presented by Ghiselin (1969) to explain sequential hermaphroditism in animals. Because there are increased costs of reproduction for females relative to males, there is a correlation between size and gender, with females exhibiting a larger size, with greater resources than males.

Arisaema triphyllum (Jack-in-the-Pulpit) is a woodland perennial common in the eastern half of the United States with a diphasic sexual system where gender is controllable and reversible in either direction (Schaffner 1922). Individual plants of this species are almost exclusively dioecious, with only a very small percentage of monecious individuals found in any one population (Schaffner 1922; Ewing and Klein 1982; Levine and Feller 2004; Barriault et al. 2009). Many studies of the species *A. triphyllum* have shown that there is a strong correlation between size and gender in a population, however size is not the sole determinant of a plant's gender (Ewing and Klein 1982; Bierzychudek 1982, 1984a; Policansky 1987; Vitt 2003). The exact effects of genetic and environmental conditions on gender, particularly the cues that might lead to a switch in gender expression, are areas that are still not widely understood.

Population sex ratio affects the relative success of being male versus being female. Bierzychudek (1984b) showed that the reproductive success of individuals in a population of *Arisaema triphyllum* is directly dependent on the sex ratios of that population. Because individuals of the rarer sex in a population have a higher reproductive value (Fisher 1930), a sex ratio bias in a gender switching population will confer a reproductive advantage onto whichever sex is rare in a given season (Vitt 2003). The overlapping size of male and female plants within *A. triphyllum* populations indicates that another factor, possibly sex ratio, influences gender determination. The amount of control that an individual has over when and in what direction it can change its gender can lead to greater fitness for an individual plant. The composition of pollen that an individual female receives can be taken as a proxy for the sex ratio of that female's population. In this regard, it would be greatly advantageous for a female plant to be able to discriminate between pollination by one male, and pollination by many males, in order to ensure the greatest amount of reproductive success in any given season. Because *A. triphyllum* is self-incompatible, it is possible that there is a mechanism for pollen source identification in these plants.

Objectives

Much of the work investigating the causes of sex change in the genus *Arisaema* has focused on applying the size advantage hypothesis to gender switching in these populations. My research will investigate sex ratios and gender switching in a population of *Arisaema triphyllum*. I tested the hypothesis that the sex ratio of a population influences gender switching in these species by manipulating the composition of the pollen used to pollinate each female. I hypothesize that females pollinated with pollen from multiple males will remain female at a higher rate than those pollinated with pollen from a single male.

Materials and Methods

Study species

Jack-in-the-Pulpit (*Arisaema triphyllum*) is an herbaceous perennial found throughout eastern North America. Individual plants overwinter as a corm and produce a single inflorescence consisting of a spadix and spathe (Bierzychudek 1982). The reproductive strategy of *A. triphyllum* is unique in that individuals express gender switching or diphasy. *A. triphyllum* plants, when reproductive, tend to be male when small and female when large (Bierzychudek 1982). Bisexual individuals are known to exist, but are not very common, and only three were observed in this study. The pollinators for *A. triphyllum* are believed to be a combination of fungus gnats (*Mycetophilidae* and *Sciaridae*) and a thrip (*Heterothrips arisaema*) (Barriault et. al 2010). It is thought that these pollinators do not receive any benefit for their pollination services (Bierzychudek 1982); rather they are lured to the flowers by smell and then trapped inside the spathe of females. *A. triphyllum* is able to reproduce asexually as well as sexually by budding off the corm (Bierzychudek 1982; Boles et. al 1999; Vitt 2003).

Pollination experiment

In the spring of 2009, a sample of 123 *Arisaema triphyllum* females and 39 males from a population of plants within Florsheim Nature Preserve in Lincolnshire, IL, were located and marked with flags and small numbered tags. The GPS location of each plant was also recorded with a Nomad GPS unit. The females were haphazardly sampled from the population in mid-April and randomly placed into one of four pollination treatments.

> 1. control, i.e. naturally pollinated (np); 2. naturally pollinated plus addition of pollen from multiple males (npp); 3. saturated with pollen from a single male (sm); 4. saturated with pollen from multiple males (mm).

All plants not yet fully emerged and with the spathe enclosing the spadix were covered with pollinator exclusion bags from the time of emergence to eliminate natural pollination. Individuals who had already emerged by the time I began my field work were randomly placed into treatments one and two because I could not ensure that they had not already been naturally pollinated. Plants in these treatments were allowed to undergo natural pollination, with additional pollen being added to plants in treatment two after a period of a few weeks, but before all the male flowers in the population were gone. This was done to investigate any confounding factors from the naturally pollinated plants also being the plants that were the first to emerge. Plants which had been enclosed in pollination bags before opening were randomly assigned to either treatment three or four. Every female flower on the inflorescence was given the same treatment. To hand pollinate plants in treatments two through four the entire spathe was carefully removed using a razor blade. Each stigma was then brushed with pollen that had been collected using a sable bristled paintbrush. Plants in treatments three and four were then re-covered with pollinator exclusion bags until all male flowers in the population were gone.

The pollen from each male individual was collected by removing a small portion of the spathe and dumping the pollen that had fallen to the bottom into a labeled glass vial. The pollen from each male was randomly chosen to either stand alone as pollen to be used in a single male treatment (treatment 3), or was used as a part of a 10 plant pollen mixture to be used in the multiple male treatments (treatments 2 and 4). Pollen was used the same day in which it was collected to minimize questions of pollen viability.

Measurements of leaflet number, length and width of the center and right hand leaflet, and base pseudostem diameter were taken for each of the tagged plants once the plants had reached their maximum size in June 2009, and again in June of 2010. The four linear leaf measurements were summed to provide a proxy size for total leaf area. The leaf area of a subset of plants was also determined in 2009 in order to test if the length and width measurements of the plants in the study correlate with the leaf area measure, as has been shown in previous work (Vitt 2001). This was done by tracing the leaf outlines onto plain white paper, cutting out the shapes, and then weighing each one on a balance. I then took the weight of five separate 1 cm^2 pieces of paper from the same ream and averaged them to get the weight of 1 cm^2 of white paper. The area of the cut out leaf shapes could then be calculated from this measure. A linear regression analysis done in Microsoft Excel showed a strong correlation between the sum of the four leaflet measures (length and width of the center and right-hand leaflets) and the leaf area ($R^2 = 0.916$), confirming my decision to use only the sum of leaflet measures in my analyses (all further mention of leaf size will refer to this sum).

In September of 2009 I collected all of the infructescences from the females in each of the treatment categories. However, I was able to find only 25% of the infructescences. Each recovered infructescence was photographed, and fruits and seeds were counted and weighed, and an average fruit and seed weight were calculated for each individual plant. In June of 2010, the gender and size of each permanently marked female plant that could be relocated were measured and recorded the same way as in 2009.

Data Analysis

All data were analyzed using the R Statistical Package Version 2.11.1 (The R Foundation for Statistical Computing. 2008) unless otherwise noted.

Size and Gender

Leaf and pseudostem size for every female were measured in 2009 and 2010, and the means were compared with a one-way ANOVA across pollination treatment (single male (sm), multiple males (mm), naturally pollinated (np), natural pollination plus multiple males (npp)). Student's t-tests were run to compare mean sizes across years within each pollination treatment.

Pollination experiment

Gender data were analyzed from 2009 and 2010, and number of individuals that changed gender (female to female, female to male, female to vegetative, male to male, etc.) was calculated. A Fisher's exact test was used to test for a relationship between gender switching of females and pollination treatment. A logistic regression was conducted to determine the probability of gender change given a particular treatment, with pseudostem diameter, and leaf size as covariates. The model was created by simplifying a saturated generalized linear model with binomial errors. A X^2 test was used to test for any significant differences between fruit maturation and pollination treatment, and a logistic regression was used to test for a relationship between plant size and fruit production.

Spatial Analysis

A map of the population was created with ArcGIS (Version 9.3.1, Esri Inc., 2009). A Ripley's K analysis was conducted to compare overall dispersal of female plants in 2009 to that of the females who switched gender versus those that remained female in 2010. The Ripley's K analysis summarizes spatial dependence over a range of ten distances in the population. The tool computes the average number of neighboring features within each distance associated with each feature. If the average number of neighboring features is larger than the average concentration of features at that distance, than the distribution is considered clumped at that distance. The analysis was first run in its unweighted form to understand the general dispersal pattern of all of the female plants. The analysis was then run again, giving female plants that changed gender a weight of one and those that remained female a weight of two. The weighted Ripley's K analysis can then be compared to the unweighted analysis to see how much clumping there is over and above the amount expected in the population as a whole. This analysis does not take pollination treatment into account, but rather investigates spatial relationships of gender switching in female plants regardless of treatment.

Results

Size and Gender

Mean sizes of females in 2009 did not differ significantly across pollination treatments according to a one-way Anova (Figure 1-1a; Leaf, n=110, df=3, p=0.876; Pseudostem, n=115, df=3, p=0.487). Plants across all treatments were significantly smaller in both leaf size and pseudostem diameter in 2010 compared to 2009, according to a Student's t-test (Table 1-1, Leaf: t = 7.2, df = 130, p-value <0.001, Pseudostem: t = 9.8, df = 135, p-value < 0.001).

Pseudostem sizes in 2010 did differ significantly (Figure 1-1b), with individuals that were naturally pollinated having significantly larger pseudostem diameters than individuals from the multiple male (mm) hand pollination treatment (Anova, n=76, p = 0.047).

Pollination Experiment

Plants in each gender class were more likely to remain the same gender in 2010 when only naturally pollinated females were used (Table 1-2). Using size as a covariate, a logistic regression shows that larger females tended to remain female, while smaller females were more likely to switch gender (Figure 1-2, Leaf size: $t = 2.7$, $df = 38.4$, p-value = 0.009 , Pseudostem size: $t = 2.8$, $df = 40.7$, p-value = 0.007). While both pseudostem diameter and leaf size were positively associated with remaining female, pollination treatment had no significant effect on the probability of gender switching according to the model (Analysis of Deviance, $df = 67$, $p = 0.804$).

Whether a female individual from 2009 expressed a different gender in 2010 was not associated with pollination treatment according to a Fisher's exact test (Table 1-3, $df = 3$, $p=0.67$). Pollination treatment had no effect on whether or not an individual produced fruit ($X^2 = 2.2375$, $df = 3$, p-value = 0.5246). There was also no relationship between plant size and fruit production (Table 1-5).

Spatial Analysis

Gender switching in female plants shows a clumped distribution that is greater than the amount of spatial clumping of the overall population at all ten distances analyzed (Figure 1-3). Figure 1-4 shows the spatial locations of all plants in the population based on whether or not they switched gender in 2010.

Discussion

To date, work done on gender switching and *Arisaema triphyllum* has focused primarily on the correlation between an individual's size and its gender. However, size alone does not completely predict which gender a plant will exhibit in any given year (Bierzychudek 1984a; Vitt 2003). The four pollination treatments used in this study were chosen to represent different pollen compositions as a proxy for male versus female biased sex ratios. The hypothesis that pollen composition could influence gender switching was based on the idea that a reproductive advantage is conferred upon whichever sex is rarer in a population (Fisher 1930, Vitt 2003). If males in a population are rare it will be more likely for their pollen to fertilize a female, because they will have less pollen to compete with. However, no evidence was found that female plants are switching genders in response to pollen composition. Although not significant, a greater than expected number of females in both multiple male treatment groups actually became male in the following year, rather than remain female. This result is contrary to the rare sex hypothesis, and indicates that further study should be done in order to determine if there is a relationship between pollen composition and gender switching that was not able to be detected in this study, perhaps due to small sample sizes or confounding effects of pollen load.

Because of a lack of enough female plants, I was unable to test pollen load as a variable in addition to pollen composition. All of the hand pollinated plants were saturated with pollen in order to minimize any combination effects from pollen load. With pollen saturation on the hand pollinated females, I hypothesized that those individuals would produce more fruit than the naturally pollinated plants. However, there was no significant correlation between pollination treatment and whether or not an individual produced fruit. There was also no significant correlation between leaf size and fruit production seen in this population. However, it has been shown previously that producing and maturing a full infructescence requires a large expenditure of energy that the smaller plants often cannot afford (Bierzychudek 1984b). This could cause fruits or seeds to be aborted in order for the plants to survive. It is likely that the pollen saturation of the inflorescences overwhelmed most of the females in the study, evidenced by the limited amount of fruit produced and matured. This could have masked any response to pollen composition that may have been seen, and rather than producing fruit and then changing gender, they aborted the fruit to conserve stored resources. Additionally, the low fruit numbers seen from the naturally pollinated plants, as well as the minimal amount of fruit found in the population in 2010 (pers. obs.) indicates a generally pollen-limited population. Fungus gnats are the main *Arisaema triphyllum* pollinators, and are known to be relatively ineffective (Mesler et al. 1980; Barriault et al 2010) with pollen limitation being a common occurrence in *A. triphyllum* populations (Rust 1980; Bierzychudek 1982; Barriault 2010).

When looking at plant size and the probability of gender switching, larger female plants were more likely to remain female than smaller plants. This result supports the theory that cost of reproduction plays a role in determining a plants gender for the next year. In 2010, plants that were part of the naturally pollinated group had larger pseudostem diameters than the hand pollinated plants. The results do not indicate that pollen composition impacts size because there was no difference between the single male and multiple male treatment groups. Additionally, the mean size of females pollinated in each of the four treatments did not differ in the year in which they were pollinated, 2009, so previous size cannot account for the difference in size in 2010. Pseudostem diameter has been shown to be significantly correlated to corm size (Vitt 2001), meaning those plants with larger pseudostems likely were able to store a greater amount of photosynthate the previous year. Because the group of plants that were naturally pollinated were the only plants to retain their spathes (the others had theirs removed in order to aid in hand pollination), this might indicate that the spathe does a relevant amount of photosynthesizing for the plant.

In addition to pollen composition and plant size, environmental factors are also known to play a role in gender expression in *Arisaema triphyllum* plants. According to the Ripley's K analyses, females showed a clumped distribution based on whether they changed gender or not above and beyond the clumping that is seen in the population as a whole. This indicates a possible relationship between gender switching and environmental factors which is investigated further in Chapter 2.

Conclusions

Based on this study, there appears to be no correlation between the pollen composition received by female plants and the propensity of females to remain female or become male the following season. Because there were not enough females available, I was unable to test the effects of pollen load, which may have a more prominent effect on gender switching, especially in a plant that is known to often be pollen limited. Additionally, it is possible that there is a lag effect in seeing any differences in gender switching as a result of this experiment. *Arisaema triphyllum* plants are known to "decide" their gender for the next season by mid-growing season (Vitt 2003). Therefore it may take longer than one season for the effects of increased pollination or a reaction to differing pollen composition to be determined. While it is clear that plant size is not the only determinant of a plant's gender, a more detailed and longer term study of different pollen compositions, as well as different pollen loads would need to be done in order to fully understand the effects of pollen composition versus pollen load on gender switching.

Tables

Table 1-1. Mean leaf and pseudostem sizes (± 1 SE) for females in each pollination treatment in 2009 and 2010. Bold and * indicates a significant difference among treatments (anova, n=76, df = 3, p=0.047) Bold and ** denotes a significant difference between years from t-test (Leaf; t=7.2, df=130, p<0.001; Pseudostem; t=9.8, df=135, p < 0.001

	Mean leaf sizes (mm)		Mean pseudostem diam. (mm)	
Pollination treatment	2009	2010	2009	2010
multiple male	570.8±20.5	412.0±27.1	11.9±0.34	7.1±0.50
single male	545.0±22.2	407.5±32.6	11.7±0.48	7.6±0.67
natural pollination	554.7±17.1	454.7±30.8	12.4±0.44	**9.1±0.68**
natural plus multiple male	548.4±25.2	417.0±20.5	12.6±0.58	7.8±0.45
All treatments	554.9±10.6	**423.0±14.4****	12.1±0.23	**7.9±0.31****

Table 1-2. Number of individual plants that fell into each gender category in 2009 and 2010. Only naturally pollinated females included in counts for female plants.

		Gender in 2010		
		Female	Male	Vegetative
Gender in 2009	Female	10	6	6
	Male	7	17	11
	Vegetative	0	5	12

Table 1-3. Number of female individuals in each pollination treatment that expressed each gender in 2010. Expected numbers from Fisher's exact test in parentheses.

	Gender in 2010		
Pollination Treatment	Female	Male	Vegetative
Single male	7 (7.3)	7 (8.3)	7 (5.4)
Multiple male	6 (6.6)	10 (7.6)	3 (4.9)
Natural pollination	10 (7.6)	6 (8.7)	6 (5.6)
Natural plus multiple male	4 (5.5)	8 (6.4)	4 (5.6)

Table 1-4. Analysis of deviance table for logistic regression with binomial errors run on gender switching and plant size. Bold indicates significance.

Parameter	Std. error	Pr(>|z|)	Model null deviance	Df	Model residual deviance	Df
Leaf size 2009	0.003	**0.0914**				
Pseudostem diameter 2009	0.122	**0.0937**	89.16	68	77.88	66

Table 1-5. Analysis of deviance table for logistic regression model run on fruit production and plant size in 2009.

Parameter	Std. error	Pr(>|z|)	Model null deviance	Df	Model residual deviance	Df
Leaf size 2009	0.004	0.580				
Pseudostem diameter 2009	0.141	0.188	74.73	68	72.73	66

Figures

Figure 1-1. Mean leaf sizes and pseudostem diameters of female plants (±1 SE) for each pollination treatment (n values above bars) in (a) 2009 and (b) 2010. Treatment codes are mm = multiple males, np = natural pollination, npp = natural plus multiple male, sm = single male. * denotes significant difference (anova, n=76, df = 3, p=0.047).

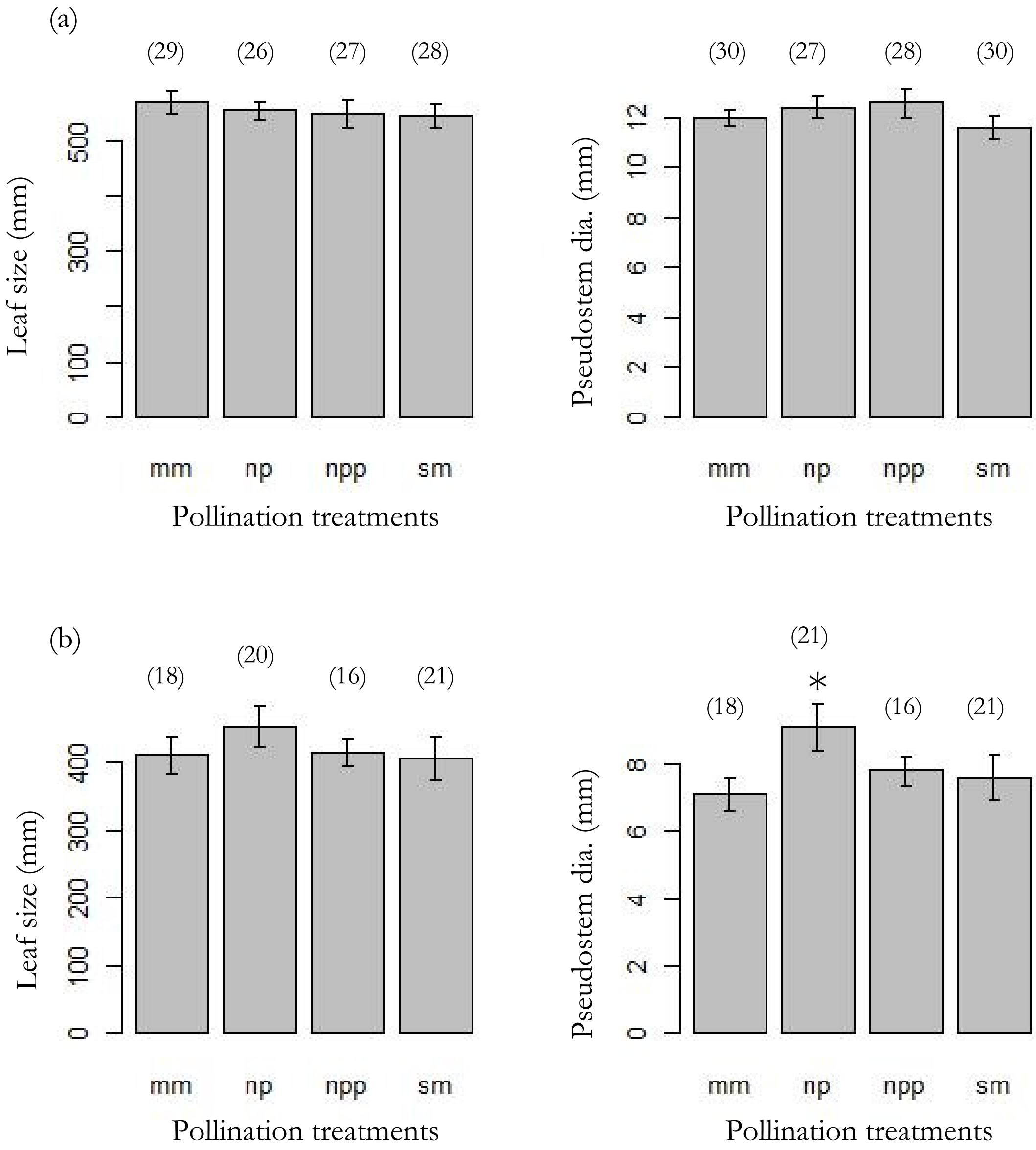


Figure 1-2. (a) Mean leaf size and (b) mean pseudostem diameter (±1 SE) of females in the pollination experiment that remained female in 2010 (yes, n=24; no, n=45). * denotes significant difference (a. t = 2.7, df = 38, p = 0.008; b. t = 2.8, df = 40, p = 0.006)

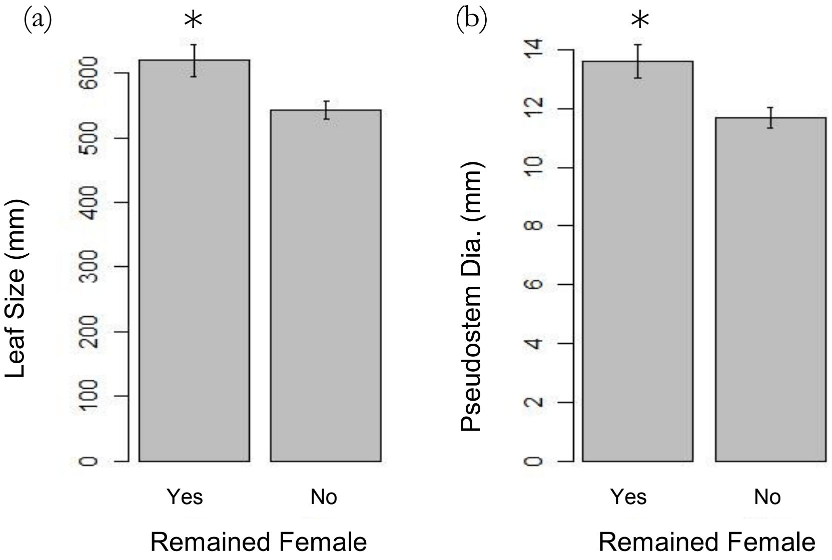


Figure 1-3. Graph showing results of Ripley's K analysis on spatial distribution for female plants. The distance along the x-axis represents the distance at which the distribution was analyzed, while the L(d) statistic on the y-axis is the K-function calculated by the analysis. Points above the expected line indicate a clumped distribution while points below indicate a dispersed distribution. The unweighted series includes all plants of the population equally, while the weighted series gives different weight to females that remained female (2) and those than changed gender (1).

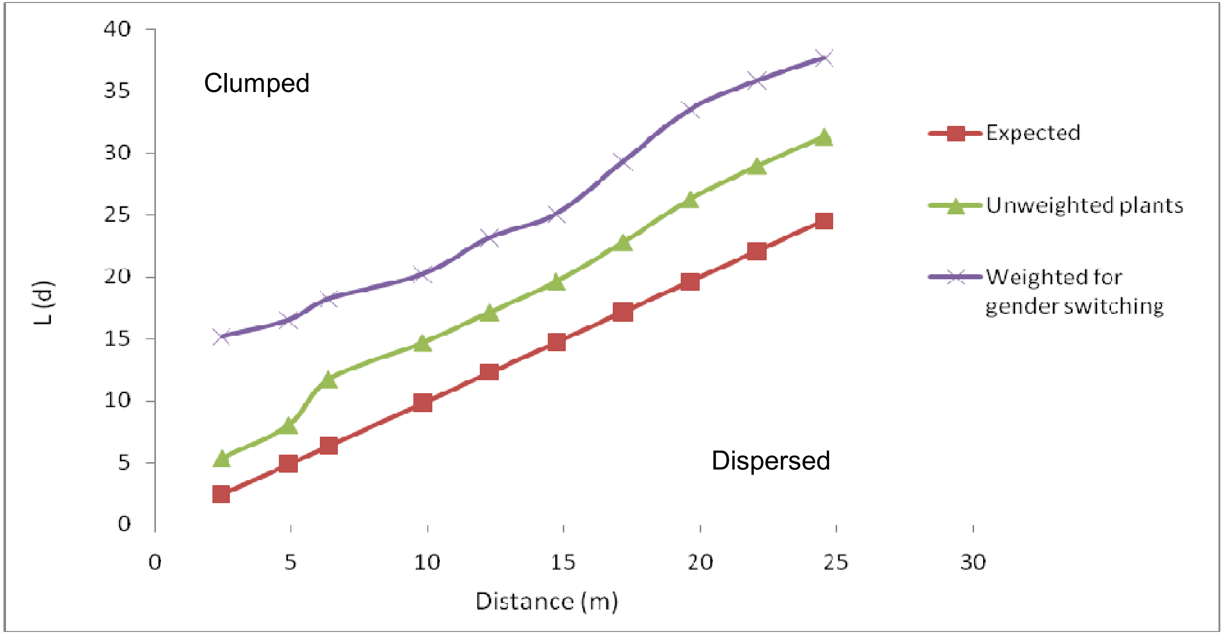
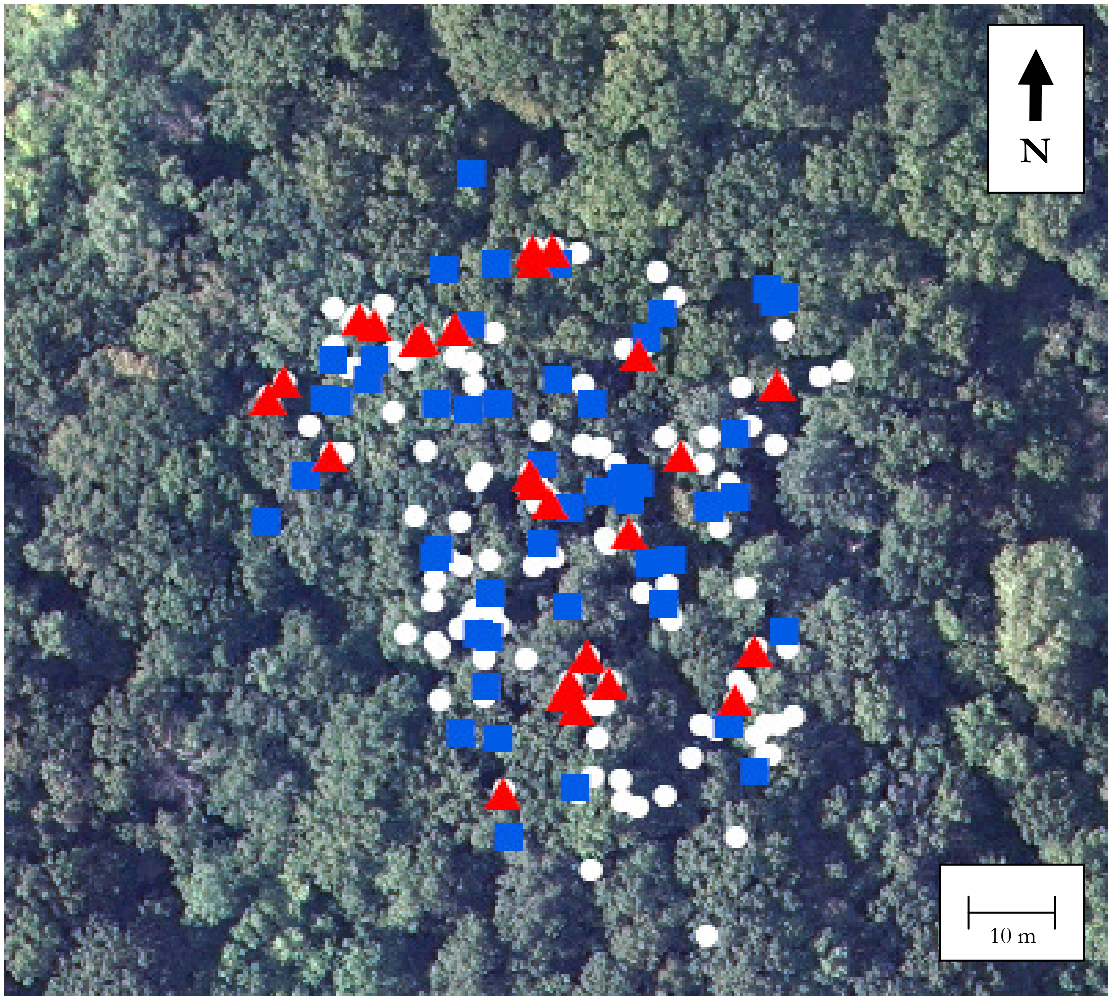


Figure 1-4. GIS map of female plants. Red triangles are plants that remained female from 2009 to 2010, while blue squares are plants that changed gender from 2009 to 2010. White dots represent males and vegetative plants in the population.



Chapter 2 – Environmental and Management effects on Population Sex Ratios in populations of *Arisaema triphyllum*

Introduction

The sex ratio of a population of dioecious plants affects the relative reproductive success of being male versus being female in that population. Bierzychudek (1984b) showed that the reproductive success of individuals in a population of *Arisaema triphyllum* is directly dependent on the sex ratios of the population. In most published studies of *A. triphyllum*, populations tend to be male-biased (Shaffner 1922; Lovett Doust and Cavers 1982; Bierzychudek 1982; Vitt 2003). In other plant species, environmental conditions have been shown to influence both the ratio of male to female flowers on an individual plant, as well as the ratio of male to female organs on perfect flowers (Freeman et al. 1980). It is also likely then, that environmental conditions will have an influence on the sex ratio of a dioecious population. Dioecious individuals with the ability to change sex will have a selective advantage over those that cannot in a patchy or changing environment (Freeman et al 1980). A 2009 study on the effect of nitrogen levels on understory plants showed that differences in past land usage can alter the growth and biomass of *A. triphyllum* plants (Fraterrigo et al. 2009). Because there is a correlation between size and gender, land alterations, and possibly certain management activities could then also lead to changes in population sex ratios.

One environmental factor of particular interest when investigating gender expression in *Arisaema triphyllum* is light level. Changes in forest canopy structure due to succession and or management activities can alter the understory light levels by either creating or minimizing gaps in the tree canopy. Light is often considered the most limiting resource for understory herbs, and increased light levels have been shown increase female:male sex ratios in *A. triphyllum* populations (Levine and Feller 2004). Additionally, photosynthetic rates have been shown to differ both between genders and also among different sized female individuals (Vitt 2001). Because of this association between light availability, photosynthetic rates, and gender, it is possible that canopy structure will be associated with population sex ratios and gender switching.

Objectives

Previous studies have shown that stressful environmental conditions tend to increase male-bias in populations of dioecious plants (Heslop-Harrison 1957; Freeman et all 1980; Schlessman 1988). Stressful environmental conditions are marked by decreases in available resources, which can make it less possible or profitable to reproduce as a female. Differences in competition levels, light levels, and management histories in an area all affect the environmental conditions and may influence the population sex ratio of *Arisaema triphyllum* plants in that area (Lovett-Doust and Cavers 1982). This study investigated relationships between community composition, canopy structure, and management history and their effect on the sex ratios of two populations of *A. triphyllum* in Northeastern Illinois.

Materials and Methods

Study site

Two populations of *Arisaema triphyllum* were investigated in the Florsheim Nature Preserve in Lincolnshire, Illinois. This site began as 40 acres of donated land in 1985, but now encompasses 110 acres of managed land. Burning, invasive control, mowing, and deer culling are all part of the management strategy of Florsheim Nature Preserve that began in 1995 (Pers. comm. Lydia Scott). Anywhere from 5-20 deer are culled each year at the site. The main invasive threats in the woodland area are buckthorn (*Rhamnus cathartica*), garlic mustard

(*Alliaria petiolata*), and oriental bittersweet (*Celastrus orbiculatus*) (Pers. comm. with Chris Fisher).

Figure 2-1 shows an aerial map of the park with the location of each population labeled. One population of Jack-in-the-Pulpits is located off the trail to the right immediately as you enter the woods (Figure 2-2a). This population consists of around 400 individuals and has a male-biased sex ratio (~3:1 in 2009). The area is characterized by a fairly open canopy and several ephemeral ponds that remain wet until early summer. There are three distinct levels of vegetation, an herbaceous layer, a shrub layer, and an upper canopy. The most recent burning of this area occurred in the fall of 2009.

The second population used in the study is located deeper into the woods between the fork of two trails, in an area that is somewhat drier with no ephemeral ponds (Figure 2-2b). This population consists of >1,600 individuals and has an even to slightly female-biased sex ratio (~1:1 in 2009). This area is characterized by a more closed canopy, with only two distinct vegetation layers, an herbaceous layer and an upper canopy layer. A shrub layer is present in certain areas, but is very sparse. This population also has a number of large fallen trees, as well as a very substantial litter layer consisting mainly of oak leaves. This area is very rarely burned, but had a small patch accidentally burned in 2008, before this project was begun.

Canopy Photos

In the summer of 2009, canopy photos were taken of ~90 plants (30 male, 30 female, and 30 vegetative) from each population. A Nikon E4500 camera was fitted with a Nikon FC-E8 fisheye lens, and each photo was taken at a height of 0.5m, with the camera oriented so the bottom of the photograph produced was always due north. All 90 plants from the male-biased population and the 30 vegetative plants from the female-biased population were randomly chosen and marked with an ID tag, while male and female plants from the female-biased population were randomly selected from plants previously marked as part of the pollination study (Chapter 1).

Vegetation Survey

In the summer of 2010, a vegetation survey was conducted at both populations in order to characterize the community composition of the two populations. In each population, three parallel transects were randomly placed perpendicular to the long edge of the population area. Each tree whose canopy hung over a transect was identified, and the diameter at breast height (dbh) and length of the transect it covered were measured and recorded. Additionally, five meter-square plots were randomly placed along each transect (total of 15 plots per site) to categorize the herbaceous layer. In each plot, plants were identified to species (genus where that was not possible) and assigned a percent cover.

Data Analysis

All data were analyzed using the R Statistical Package Version 2.11.1 (The R Foundation for Statistical Computing 2008) unless otherwise noted.

Size and Gender

Mean leaf and pseudostem size for each gender category (male, female, vegetative) for each population in each year (2009, 2010) were compared using a Student's t-test.

Canopy Photos

Each canopy photo was first edited using Microsoft Office Photo Manager, adjusting the brightness and contrast to ensure relative similarity and to even out the contrast between canopy and sky among photos (Figure 2-3a). This initial editing process was done in order to present a more uniform suite of photos into the analyzing software. Canopy/sky thresholds were ultimately determined using an automatic threshold algorithm based on edge detection and implemented in SideLook 1.1 (Noblis 2005) which is available as shareware at http://www.appleco.ch. The edge detection was calculated using the blue color channel, which is believed to provide the best contrast between canopy and sky (Frazer et al. 1999, 2001; Noblis and Hunziker 2005; Jarcuska et al. 2010). Calculating thresholds automatically with this edge detection algorithm has been shown to have advantages over manual thresholding, including increased repeatability, objectivity and accuracy (Noblis and Hunziker 2005). The canopy photos were ultimately analyzed for canopy openness and transmitted gap light using Gap Light Analyzer (GLA) Version 2.0 (Frazer et al 1999), which is available for free download at http://www.ecostudies.org/gla. After the thresholds were determined, the black and white images created in SideLook (Figure 2-3b) were run through GLA to calculate a suite of canopy cover variables; percent canopy openness (Cnpy Open), leaf area index (LAI 4Ring, LAI 5Ring), percent direct solar radiation (Trans Dir), percent diffuse radiation (Trans Dif), and percent total radiation (Trans Tot). A one-way Anova compared the six canopy cover variables across the two populations. Linear regressions were performed on the same variables to determine their effect on change in plant size within each population. The six canopy variables were then used as predictive variables in a logistic regression to estimate their effects on the probability of gender switching.

Vegetation Survey

Data collected on trees during the vegetation survey were analyzed by calculating an Importance Value (IV) for each species in each area. The importance values were calculated using relative cover (RC), relative density (RD), and relative basal area (RA) in the following equation: IV = RC + RD + RA. Relative cover measurements for each area were calculated by adding together the cover measurements for each species across all three transects and then dividing by the total cover of all species in the area. Relative density and basal area were calculated in similar fashions. Sample equations to calculate each of these variables for the species *Ulmus rubra* are as follows:

$RC_{\text{Ulmus rubra}}$ = Total transect cover length *U. rubra* (m)/Total transect cover length all species (m)
$RD_{\text{Ulmus rubra}}$ = Total # of *U. rubra* individuals / Total # of individuals of all species
$RA_{\text{Ulmus rubra}}$ = Total basal area of *U. rubra* (m^2)/ Total basal area of all species (m^2)

Once each relative measure was calculated, the three were added together to get the IV for each species. The IVs for each area were then graphed alongside each other to visualize compositional differences in the tree species in each area.

The herbaceous data was analyzed with Nonmetric Multidimensional Scaling using the Vegan package in R (http://vegan.r-forge.r-project.org/). Species occurring in less than 5% of the plots were dropped from the analysis. Percent covers were transformed into cover classes using Arcsine square root transformations. Dead wood, bare ground and leaf litter in each plot were used as environmental vectors.

Spatial Analysis

Maps of each population were created using ArcGIS (Version 9.3.1) to visualize the distributional patterns of size and gender in each population. A hotspot analysis (Getis-Ord G*) was used to visualize "hotspots," or clusters of large plants, and "cold spots," or clusters of small plants. The Getis-Ord G* analysis looks at a value assigned to each point within the context of the values of neighboring points. In this analysis, values for both plant size and canopy cover were used. The tool then takes the local sum for a feature and its neighbors and compares it proportionally to the sum of all of the features. If the value for the local sum is too large or small to be expected by chance alone, a hotspot or cold spot is identified respectively. The Getis-Ord G* tool returns a z-score for each point where a large positive z-score indicates a hotspot, or cluster of high values, and a low negative z-score indicates a cold spot or cluster of low values. Getis-Ord G* analysis was also used to show clusters of high and low values of the six canopy measures calculated from GLA. The hotspots and cold spots for plant size and canopy variables for each population were then compared to visualize any co-occurring patterns between plant size and canopy structure. A linear regression was performed on plant size and the canopy variables hotspots in the female-biased population by averaging the size and canopy measures of each plant within each of the size hotspots identified into eight different clusters. Two clusters were removed from the analysis because they only contained a single plant.

Results

Size and Gender

In both populations and across both years, female *Arisaema triphyllum* plants had the largest mean leaf size and pseudostem diameter, followed by males, and then vegetative plants (Table 2-1). Mean leaf size of female plants in both populations did not differ between years (male-biased, $t = 0.05$, $df = 19$, $p = 0.96$; female-biased, $t = -1.5$, $df = 57$, $p = 0.135$). Pseudostem diameters differed significantly across the two populations in both years, with mean pseudostem diameter of female plants in the male-biased population increasing from 2009 to

2010 ($t = 2.4$, $df = 10$, $p = 0.0364$), the mean pseudostem diameter of female plants in the female-biased population decreased over the same time period ($t = -4.2$, $df = 61$, $p<0.001$). Male plants did not differ significantly in pseudostem diameter (male-biased, $t = 0.41$, $df = 24$, $p = 0.688$; female-biased, $t = -1.2$, $df = 81$, $p= 0.235$), however male leaf size was significantly smaller in the female-biased population between 2009 and 2010 ($t = -8.8$, $df = 44$, $p < 0.001$).

Canopy Photos

There were no significant differences between the two populations in any of the canopy variables measured (Table 2-2). In the female-biased population, linear regressions showed a positive relationship between an increase in pseudostem and leaf size of individual plants from 2009 to 2010 and all three measures of solar radiation (Table 2-3a, Figure 2-4). However, this same relationship was not seen in the male-biased population (Table 2-4b). No significant relationship between gender switching in females and any of the canopy variables were seen in the female-biased population (Table 2-4).

Vegetation Survey

A comparison of the Importance Values for each population shows a fairly similar species composition between the two areas. However, *Ulmus rubra* has a higher Importance Value in the female-biased population than in the male-biased population, and *Fraxinus alba* has a higher IV in the male-biased population than in the female-biased population (Table 2-5, Figure 2-5).

The herbaceous communities in each population are significantly different according to the NMDS model (p-value=0.003, stress=16.1, dimensions=3). According to the model, some of the species most indicative of a male-biased population plot include *A. bracteata*, *G. maculatum*, and *H. patula*, while species most indicative of a female-biased population plot include *V. riparia*, *A. dracontium*, *Q. rubra*, *Q. alba* and *T. radicans*. The environmental vectors of leaf litter and bare ground are correlated with the community differences with sites in the male-biased population having a higher occurrence of bare ground and sites in the female-biased population having higher amounts of leaf litter (Figure 2-6).

Spatial Analysis

Nearest neighbor analyses showed a significantly clumped distribution for both male and female plants in 2010 (Figure 2-7). GIS maps show the spatial distribution of the leaf and pseudostem sizes of individual plants in 2009 and 2010 in the male-biased (Figure 2-8) and the female-biased population (Figure 2-9). The areas of clusters of large and small plants determined in the hotspot analysis of the female-biased population can be seen circled in Figure 2-10. These circles overlay points representing the six different canopy variables studied. A regression performed on these hotspots illustrated an association between plant size and several canopy variables (Table 2-7). No significant clusters of large or small plants were found in the male-biased population however, Figure 2-11 shows a series of maps illustrating hotspots for each of the six canopy variables measured.

Discussion

Many studies have documented the relationship between size and gender in *Arisaema triphyllum* (Lovett Doust and Cavers 1982; Bierzychudek 1984a; Policansky 1987; Vitt 2003). The plants I studied in Florsheim Nature Preserve followed the same general pattern already established with female individuals generally being the largest plants, followed by male individuals and vegetative individuals being the smallest. While this general pattern is seen over and over again in *A. triphyllum* populations, the size cutoffs to transition from one gender to another between years fluctuate both within and among populations (Bierzychudek 1982). In this study, plants in each sex class differed significantly in size measures across populations and years.

Because natural selection will favor a pattern of resource allocation that maximizes the fitness of the individual, different patterns of resource allocation are expected in different ages and sexes of organisms, as well as in the same organisms growing in different places (Willson 1983). Because this study measured changes in both leaf size and pseudostem diameter reflecting differences in resource allocation between sexes and populations of *Arisaema triphyllum* could be investigated. Changes in leaf sizes from year to year indicate changes in current resources, while changes in pseudostem diameters indicate changes in stored resources (Vitt 2003). Based on pseudostem diameters, females in the male-biased population appear to increase the amount of resources they stored from 2009 to 2010, while female plants in the female-biased population on average had fewer resources stored in 2010 than they did in 2009. The existence of a tradeoff between growth and reproduction in females has been suggested before, with seedless female plants being both larger and more likely to remain female the next year than fruiting females (Bierzychudek 1984a). In this study, while the numbers of fruits or seeds were not compared across populations, many females in the female-biased population received hand pollinations (Chapter 1). Hand pollinations have been shown to be more efficient than natural pollination, especially in the case of *A. triphyllum* and result in a demographic cost of reproduction, seen as smaller size, lowered survivorship and decreased probability of reproduction (Bierzychudek 1982). Plants receiving hand pollination had smaller pseudostem diameters the following year, despite a constant leaf size across populations and years. This suggests an association between the resources required for fruit and seed production, leading to a decrease in the amount of resources that can be stored for the next year.

In the male plants, a completely different pattern of resource allocation was seen. While average pseudostem sizes remained constant across populations and years, average male leaf sizes increased in the female-biased population from 2009 to 2010. Vitt (2001) showed no correlation between pseudostem diameter and photosynthetic rate in males, as well as very little variation among male pseudostem sizes. With regard to reproductive costs, male plants produce pollen regardless of pollinator activity each season, while female plants rely on pollinators to determine the number of fruits that can be matured each season. Additionally, male plants' leaves senesce earlier in the summer than do female's, leaving them less time to collect and store resources. This more consistent reproductive demand on an individual plant's resources, may have led to plants evolving increased photosynthetic rates during the male-phase compared to the female-phase to compensate (Vitt 2001). Because pseudostem sizes remained constant across years, it is possible that stored resources in plants that decide to remain male are translated into larger leaf sizes in the following year, rather than larger pseudostem diameters. An overall increase in male plant average leaf size from one year to the next could then be ascribed to high-quality environmental conditions in the initial year.

One of the most influential environmental factors for most understory plants is light availability (Levine and Feller 2004). Both populations of plants had similar canopy structures and did not differ significantly in any of the canopy measures that were taken. However, the male-biased population is located on the edge of the wooded area very close to where the woods change over into prairie, while the female biased population is located farther into the woods, which has a significant affect on population light levels regardless of canopy structure. All plants in the female-biased population showed a significant relationship between the amounts of light available in 2009 and change in size from 2009 to 2010. Plants with the greatest light available in 2009 tended to get larger in 2010, while those that had the least light available tended to get smaller in 2010, based upon the hotspot analysis which showed that the canopy variable and plant size hotspots and cold spots overlapped. Increased plant size leads to an increased likelihood of being female, so increased light in an area could lead to a greater probability of a female-biased population sex ratio over time. The canopy in the area of the female-biased population was thinned several years prior to this study, while the same kind canopy opening is just now beginning in the area of the male-biased population. This difference is evidence for the association between increases in light gaps and an increase in the proportion of female plants over time.

Because the same patterns of light availability and change in plant size are not seen in both populations, it is likely that light availability is not the only factor influencing changes in plant size and gender. In addition to competition for light resources, *Arisaema triphyllum* plants also have to contend with competition from the herbaceous community for other resources. While the light levels from the canopy were similar between populations, the herbaceous communities in each area were significantly different. Although both populations are located within the same nature preserve, the local environment and management of each area is very different. The male-biased population is located in a much wetter area, with ephemeral ponds that last into early summer, while the female-biased population is located in a drier area with no ponds running through it. The differences in herbaceous communities between the two populations may also be due to the different management activities in each area of the preserve.

The area where the male-biased population is located has a more mature buckthorn stand that requires a greater amount of cutting and herbiciding than the area in which the female-biased population is located. In addition, the area of the male-biased population is burned more often and was even burned most recently in the fall of 2009. This occurred between the two seasons of size data collection, but before the data was collected in the vegetation survey. This burning provides an explanation for the large differences seen in the amount of leaf litter in each population, and can help to explain the differences in herbaceous communities in each area.

Because the size and gender of *Arisaema triphyllum* plants is influenced by factors in the previous season, there is likely to be a lag effect in seeing any changes in these variables due to environmental factors. Possible evidence for this can be seen in the associations between canopy and size in the two different populations. While a clear association between size and the canopy structure can be seen in the female-biased population, the same is not true for the plants in the male-biased population, despite the canopy "hotspots" and "cold spots" that were detected in the spatial hotspot analysis. Management of the canopy layer has been going on longer in the area of the female-biased population than it has in the area of the male-biased population, which could explain why the strong size associations are not yet seen in that population. This variable should be investigated over the next few seasons before ruling out that such a relationship exists in the male-biased population.

Conclusions

Resource allocation and tradeoffs are important when it comes to gender differences in *Arisaema triphyllum* plants. Differences in patterns of resource allocation were seen in both genders and both populations of *A. triphyllum* plants in this study. In order to better describe these differences, a more detailed investigation into environmental conditions would be necessary, including things like soil composition and moisture levels. Additionally due to the possible lag effects on seeing any changes, data should be collected for multiple seasons in order to fully understand the relationships between size, gender, and different environmental variables.

Differences in the herbaceous community around an individual plant can alter the level of competition it faces for resources, and increase the stress of the environment. This increased stress has been shown to increase the percentage of males in a population (Charnov and Bull 1977; Freeman et al. 1980; Day and Aarssen 1997), which could explain the sex ratio differences between the two populations in the study. While none of the management activities undertaken at Florsheim Nature Preserve are directed at *Arisaema triphyllum* specifically, the results can impact their population structures. This finding is evidence that management activities can often have impacts beyond their focal species, and monitoring of an entire community can reveal impacts on associated species that may not have been initially predicted.

Tables

Table 2-1. Mean leaf size ±1 SE in mm (a) and pseudostem diameter ±1 SE in mm (b) for each gender in the male-biased and female-biased populations in both 2009 and 2010. Values of n in parentheses. Bold denotes significant differences between years according to t-test. (Male leaf; t = -8.8, df = 44, p < 0.001; Vegetative Leaf; t = -2.8, df = 27, p= 0.009; Female Stem in male-biased; t = 2.4, df = 10, p = 0.0364 and in female-biased; t = -4.2, df = 61, p<0.001).

(a) Leaf size (mm)

	Male-biased		Female-biased	
	2009	2010	2009	2010
Female	558.1±19.9 (30)	559.5±22.8 (8)	554.9±10.6 (111)	522.6±17.7 (33)
Male	461.2±10.9 (30)	467.1±15.3 (16)	446.1±13.5 (35)	**413.1±11.9 (52)**
Vegetative	406.0±13.1 (30)	**379.7±28.5 (7)**	361.7±12.4 (29)	339.9±13.5 (42)

(b) Pseudostem diameter (mm)

	Male-biased		Female-biased	
	2009	2010	2009	2010
Female	10.4±0.38 (30)	**12.7±0.86 (8)**	12.1±0.23 (116)	**10.3±0.36 (33)**
Male	7.3±0.20 (30)	7.5±0.36 (16)	7.5±0.25 (34)	7.1±0.22 (50)
Vegetative	5.8±0.19 (30)	5.3±0.40 (7)	4.9±0.18 (29)	5.2±0.22 (42)

Table 2-2(a-f). Anova tables for canopy variables compared between the male-biased and female-biased populations.

(a) Canopy Openness

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	0.12	0.119	0.05	0.823
Residuals	176	420.21	2.387		

(b) Leaf Area Index 4

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	0.049	0.049	1.43	0.232
Residuals	176	5.99	0.034		

(c.) Leaf Area Index 5

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	0.0085	0.0085	0.43	0.511
Residuals	176	3.43	0.02		

(d) Diffuse Solar Radiation

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	6.05	6.05	1.02	0.313
Residuals	176	1040.87	5.914		

(e) Direct Solar Radiation

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	42.1	42.1	1.12	0.291
Residuals	176	6613.7	37.58		

(f) Total Solar Radiation

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Population	1	20.02	20.02	1.34	0.249
Residuals	176	2640.01	15		

Table 2-3a. Anova tables for linear regressions run on canopy variables and change in plant size in the female-biased population. **Bold** indicates significance.

(a)

Change in leaf size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	9714	9714.3	0.573	0.452
Residuals	54	916149	16965.7		
LAI 4 Ring	1	23	23.1	0.0013	0.971
Residuals	54	925840	17145.2		
LAI 5 Ring	1	15786	15786	0.9367	0.337
Residuals	54	910077	16853		
Direct solar radiation	1	99346	99346	6.4907	**0.0137**
Residuals	54	826517	15306		
Diffuse solar radiation	1	56722	56722	3.5242	**0.0659**
Residuals	54	869141	16905		
Total solar radiation	1	107304	107304	7.0788	**0.0102**
Residuals	54	818559	15159		

Change in pseudostem size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	6.51	6.51	0.9353	0.338
Residuals	56	389.83	6.96		
LAI 4 Ring	1	0.29	0.2895	0.0409	0.840
Residuals	56	396.05	7.0723		
LAI 5 Ring	1	18.96	18.96	2.8136	0.099
Residuals	56	377.38	6.7389		
Direct solar radiation	1	51.06	51.062	8.2818	**0.00566**
Residuals	56	345.28	6.166		
Diffuse solar radiation	1	37.59	37.587	5.8671	**0.0187**
Residuals	56	358.75	6.406		
Total solar radiation	1	58.64	58.642	9.7246	**0.00287**
Residuals	56	337.7	6.03		

Table 2-3b. Anova tables for linear regressions run on canopy variables and change in plant size in the male-biased populations

Change in leaf size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	0	0	0	0.998
Residuals	29	150643	5194.6		
LAI 4 Ring	1	2404	2404.4	0.470	0.498
Residuals	29	148238	5111.7		
LAI 5 Ring	1	5182	5181.7	1.033	0.318
Residuals	29	145461	5015.9		
Direct solar radiation	1	4768	4768.1	0.950	0.338
Residuals	29	145875	5030.2		
Diffuse solar radiation	1	3178	3178.3	0.625	0.436
Residuals	29	147464	5085		
Total solar radiation	1	5211	5211.2	1.039	0.316
Residuals	29	145432	5014.9		

Change in pseudostem size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	0.369	0.369	0.141	0.709
Residuals	29	75.70	2.610		
LAI 4 Ring	1	3.537	3.536	1.414	0.244
Residuals	29	72.53	2.501		
LAI 5 Ring	1	1.013	1.013	0.392	0.536
Residuals	29	75.05	2.588		
Direct solar radiation	1	0.712	0.712	0.274	0.605
Residuals	29	75.35	2.598		
Diffuse solar radiation	1	1.079	1.077	0.417	0.524
Residuals	29	74.99	2.586		
Total solar radiation	1	0.159	0.159	0.061	0.807
Residuals	29	75.91	2.617		

Table 2-4. Analysis of deviance tables for each canopy variable and gender switching in female plants in the female-biased population.

Parameter	Std. error	Pr(>|z|)	Model null deviance	Df	Model residual deviance	Df
Canopy Openness	2.833	0.962				
LAI 4 Ring	3.651	0.865				
LAI 5 Ring	2.430	0.380				
Direct Solar Radiation	0.046	0.446				
Diffuse Solar Radiation	0.123	0.844				
Total Solar Radiation	0.075	0.498	85.6	61	85.1	60

Table 2-5. A list of the calculated Importance Values for each species of tree in each of the two populations studied.

	Importance Values (IV)	
Species	FNP 1	FNP 2
Quercus alba	117.31	116.62
Ulmus rubra	59.06	104.41
Quercus rubra	37.13	38.81
Prunus serotina	3.92	15.53
Fraxinus americana	0.00	11.32
Carya ovata	4.76	8.27
Carpinus caroliniana	4.33	1.34
Prunus americana	11.10	1.32
Ulmus americana	0.00	1.23
Rhamnus cathartica	0.00	1.16
Fraxinus alba	59.58	0.00
Fraxinus pennsylvanica	2.81	0.00

Table 2.6. Anova tables for linear regressions run on clustered data based on size hotspots in the female-biased population for leaf (a) and pseudostem (b) size in 2010 and each of the six canopy variables studied. **Bold** indicates significance.

(a) Change in leaf size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	18652	18652	6.242	0.067
Residuals	4	8563	2140		
LAI 4 Ring	1	20529	20529	12.28	**0.025**
Residuals	4	6686	1671		
LAI 5 Ring	1	15712	15712	5.464	0.079
Residuals	4	11503	2876		
Direct solar radiation	1	18016	18016	7.834	**0.048**
Residuals	4	9199	2299		
Diffuse solar radiation	1	20924	20924	13.30	**0.022**
Residuals	4	6291	1572		
Total solar radiation	1	25047	25047	46.19	**0.0024**
Residuals	4	2168	542		

(b) Change in pseudostem size

Canopy variable	Df	Sum Sq.	Mean Sq	F value	Pr(>F)
Canopy Openness	1	11.84	11.84	6.242	0.067
Residuals	4	7.590	1.898		
LAI 4 Ring	1	13.14	13.14	8.345	**0.045**
Residuals	4	6.297	1.574		
LAI 5 Ring	1	8.506	8.506	3.113	0.152
Residuals	4	10.93	2.732		
Direct solar radiation	1	11.88	11.88	6.288	0.066
Residuals	4	7.556	1.890		
Diffuse solar radiation	1	13.67	13.67	9.490	**0.037**
Residuals	4	5.763	1.441		
Total solar radiation	1	16.45	16.45	22.05	**0.0093**
Residuals	4	2.984	0.746		

Figures

Figure 2-1. Aerial map of Lincolnshire, IL with the boundary of Florsheim nature preserve outlined, as well as the location of each population of *A. triphyllum* labeled.

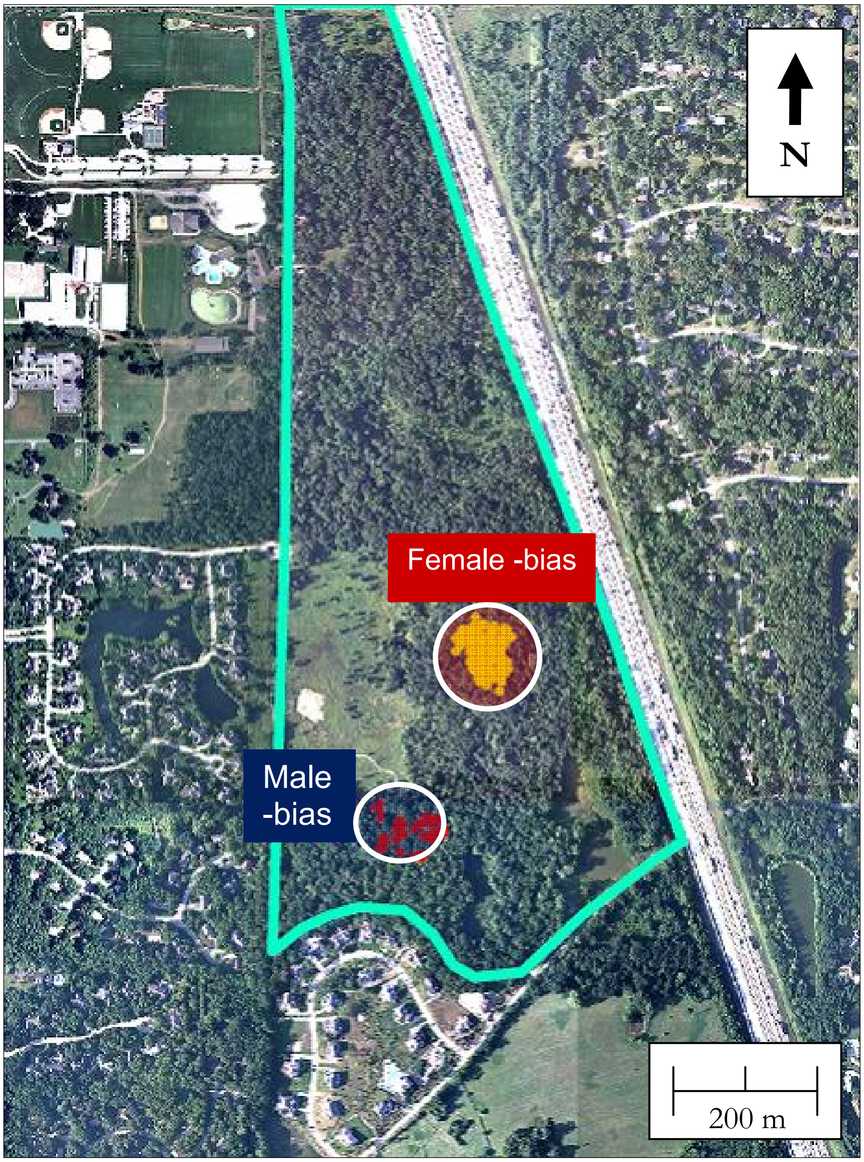


Figure 2-2. Images of Florsheim Nature Preserve *Arisaema triphyllum* (a) male-biased population and (b) female-biased population.

(a)

(b)

Figure 2-3. Examples of hemispherical canopy photos taken over plant number 301 both (a) after adjusting for contrast in Microsoft Office Photo Manager and (b) as a black and white image after the canopy/sky threshold was determined using SideLook 1.1.

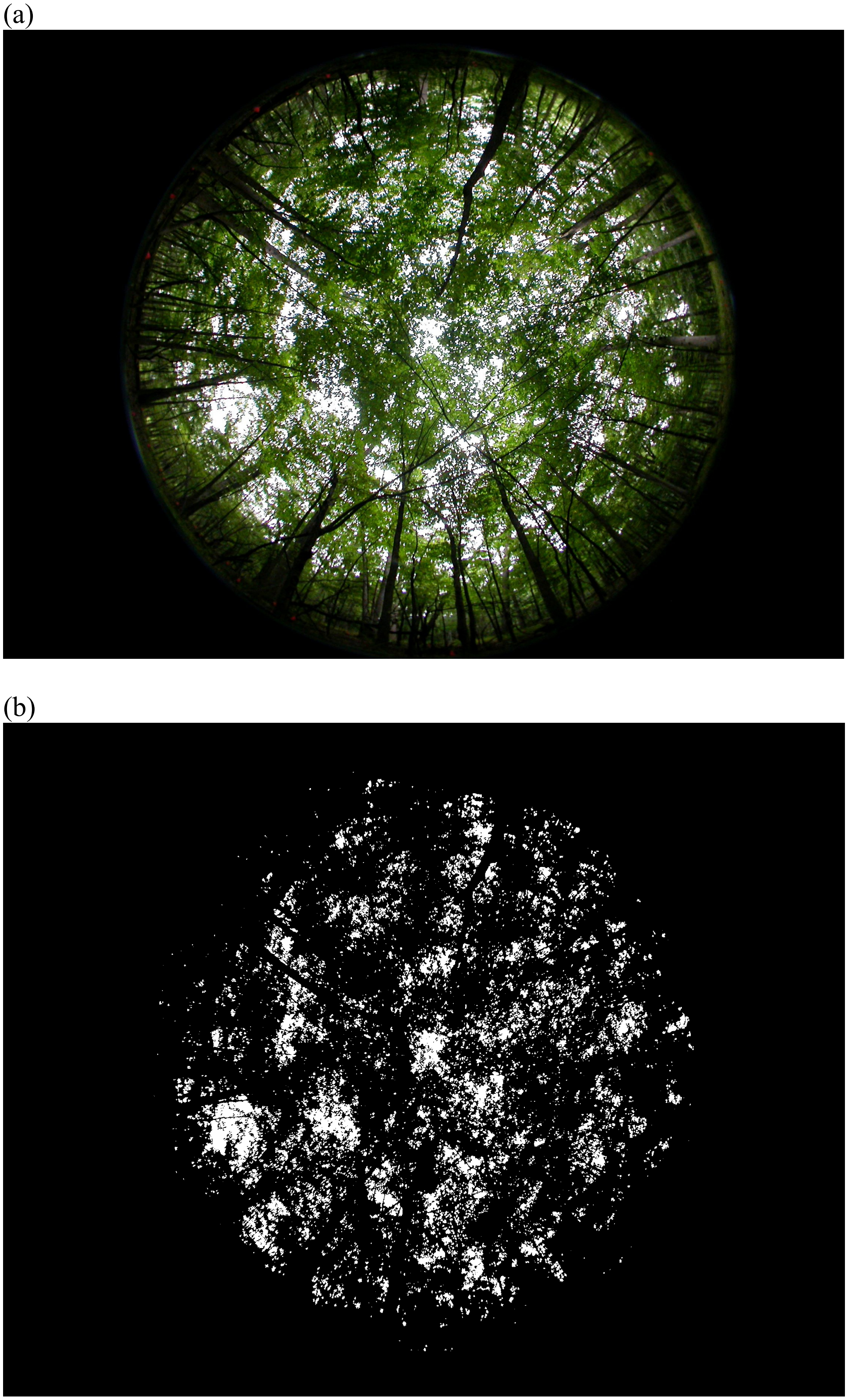
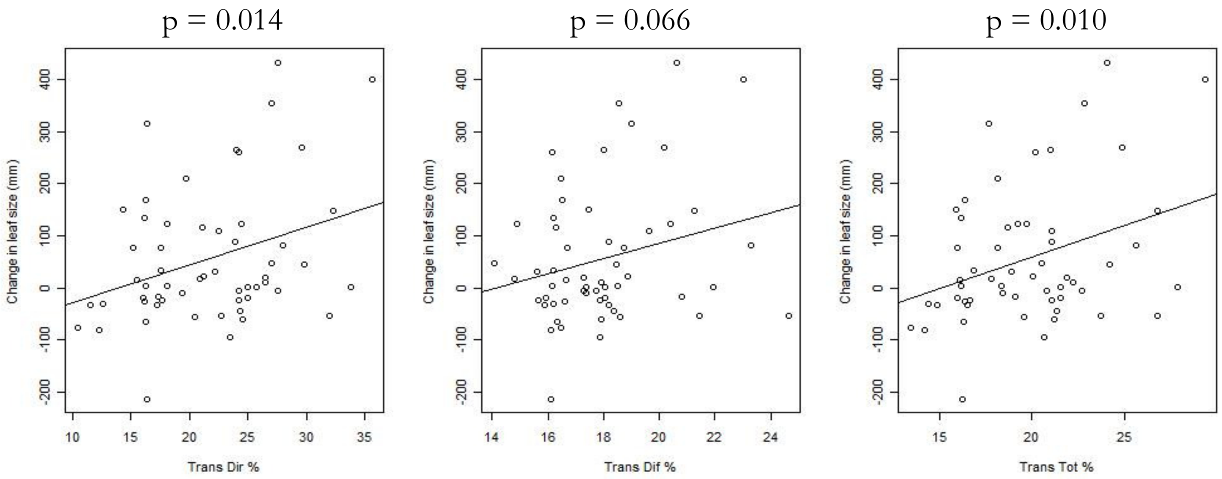


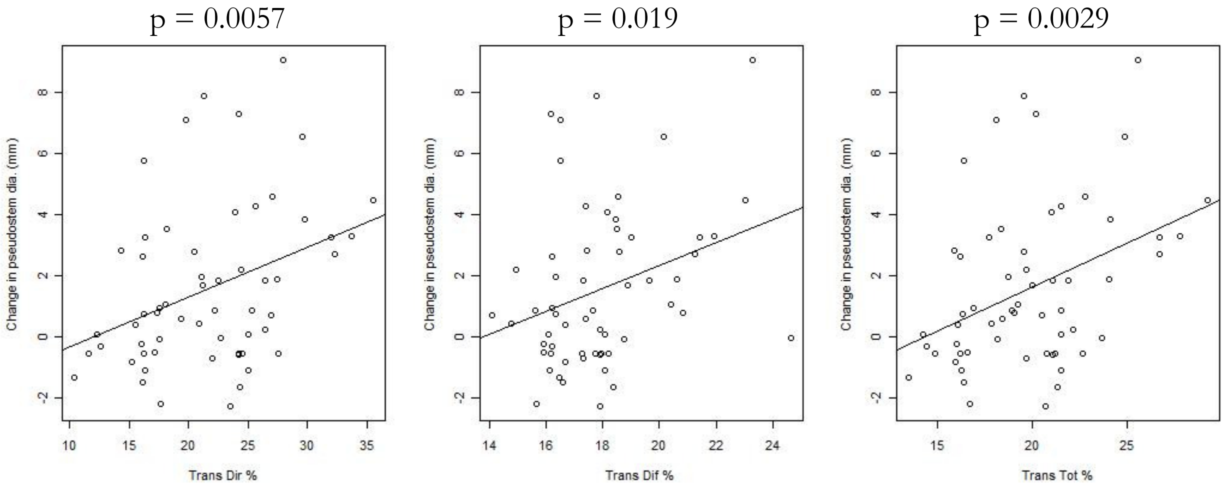
Figure 2-4. Linear regression on percent solar radiation measures (direct = dir, diffuse = dif, and total = tot) and change in plant size in the female-biased population.

(a) Leaf size



p = 0.014 p = 0.066 p = 0.010

(b) Pseudostem diameter



p = 0.0057 p = 0.019 p = 0.0029

Figure 2-5. Tree species importance values for each population plotted in order of importance in the female-biased population.

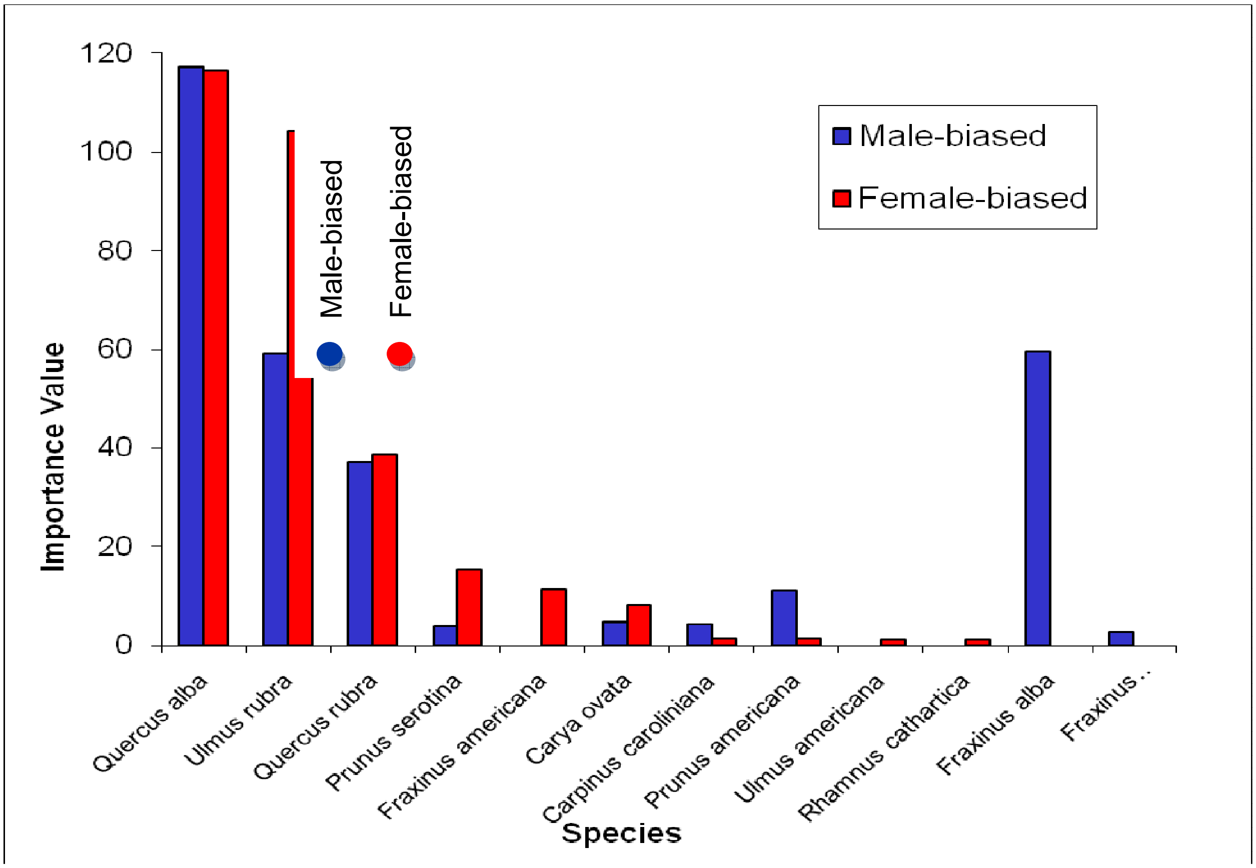


Figure 2-6. Ordination plot for herbaceous cover data from vegetation survey for each population, fitted with environmental vectors for the amount of leaf litter, bare ground, and dead wood in each plot. Species names represent data from the herbaceous vegetation survey.

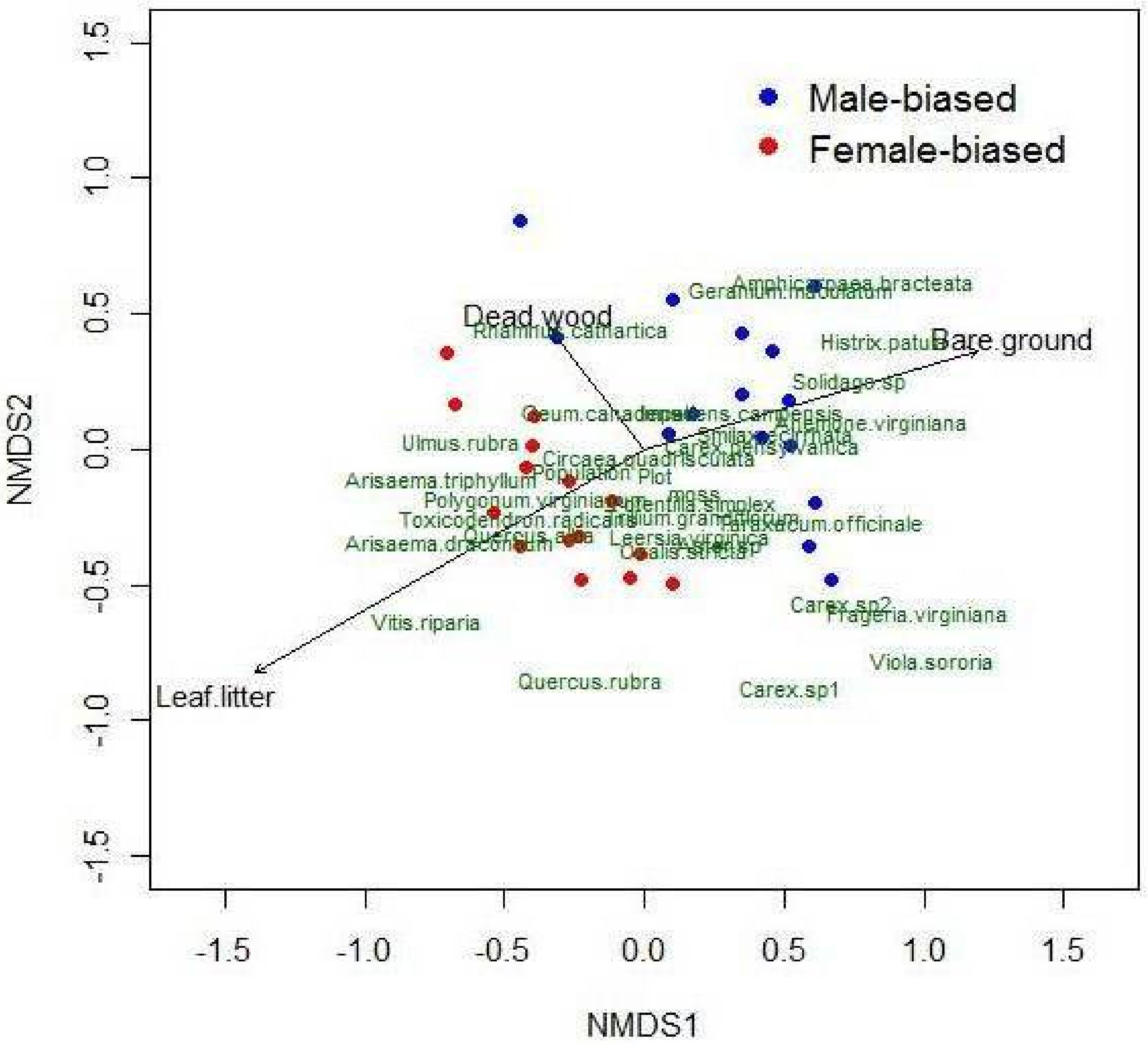
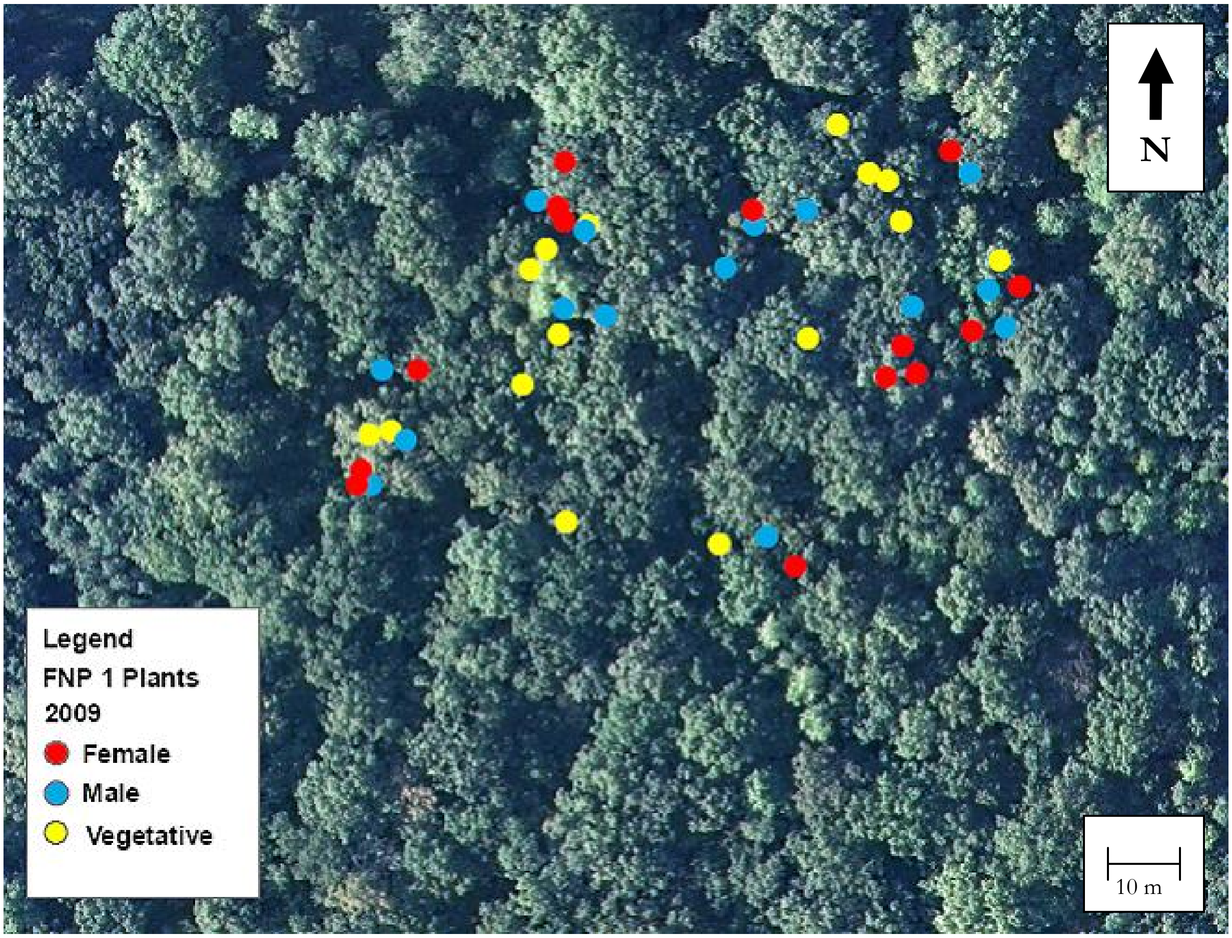
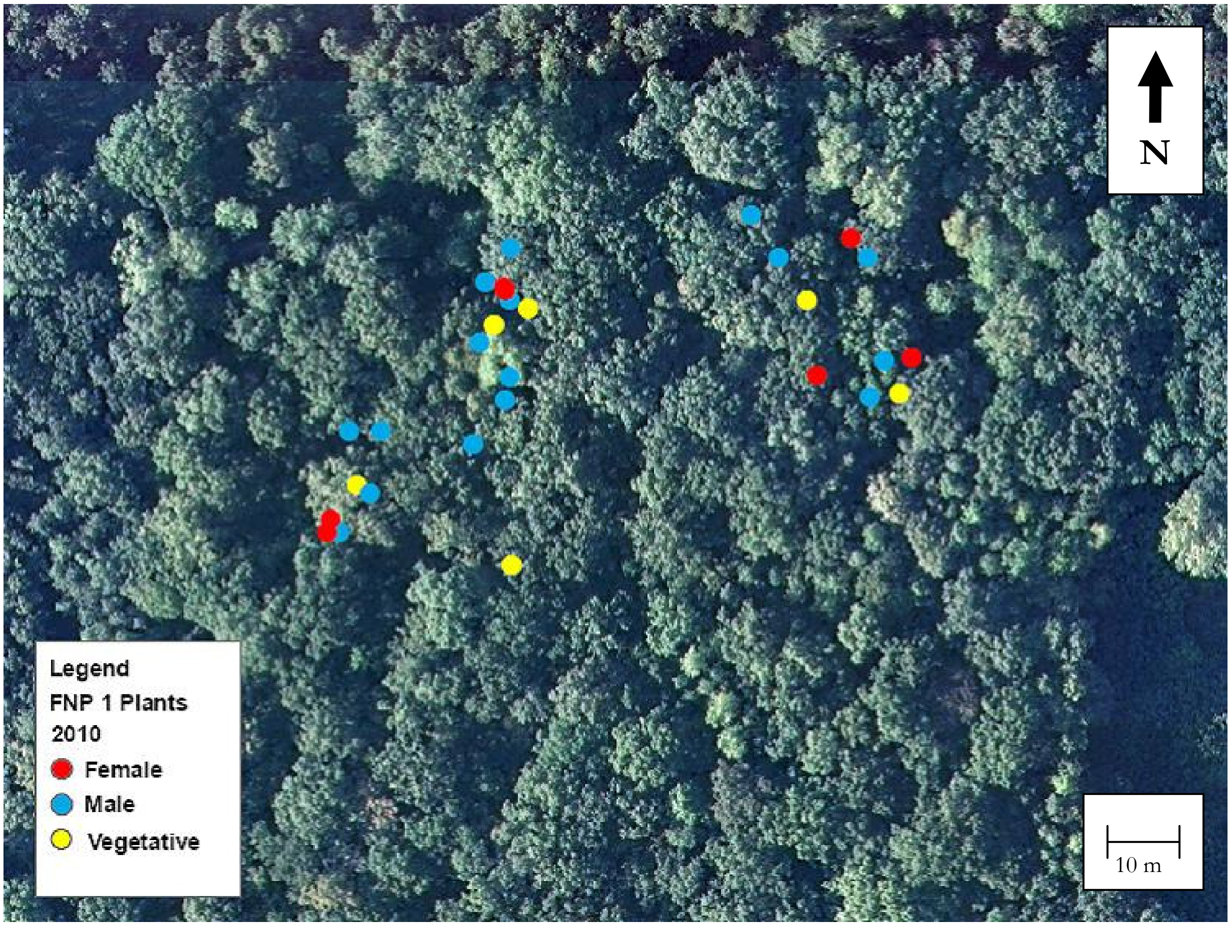


Figure 2-7. GIS maps showing the location and distribution of each plant along with its gender. Red circles indicate female plants, blue circles indicate male plants, and yellow circles indicate vegetative plants.

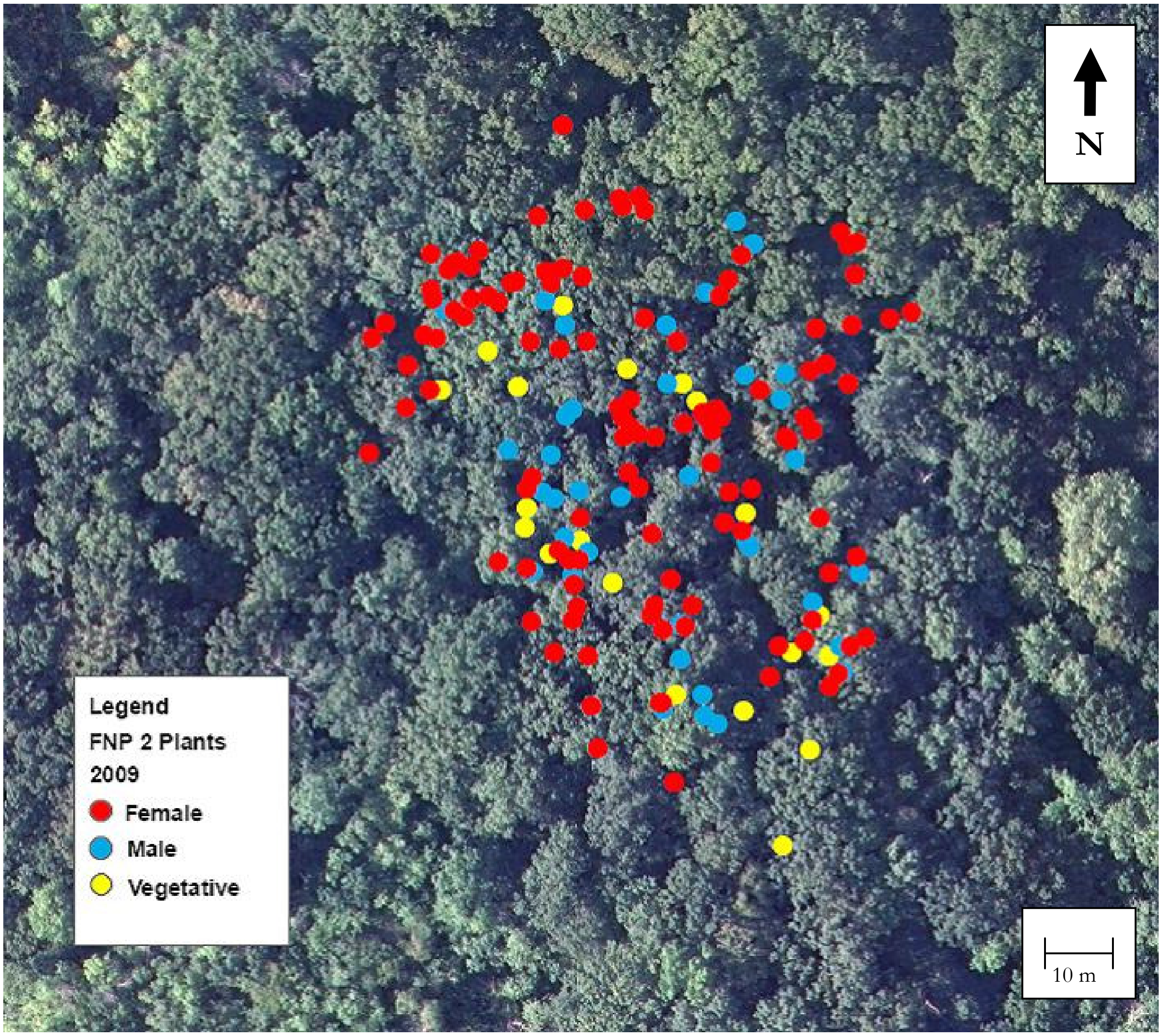
(a) Male-biased 2009



(b) Male-biased 2010



(c) Female-biased 2009



(d) Female-biased 2010

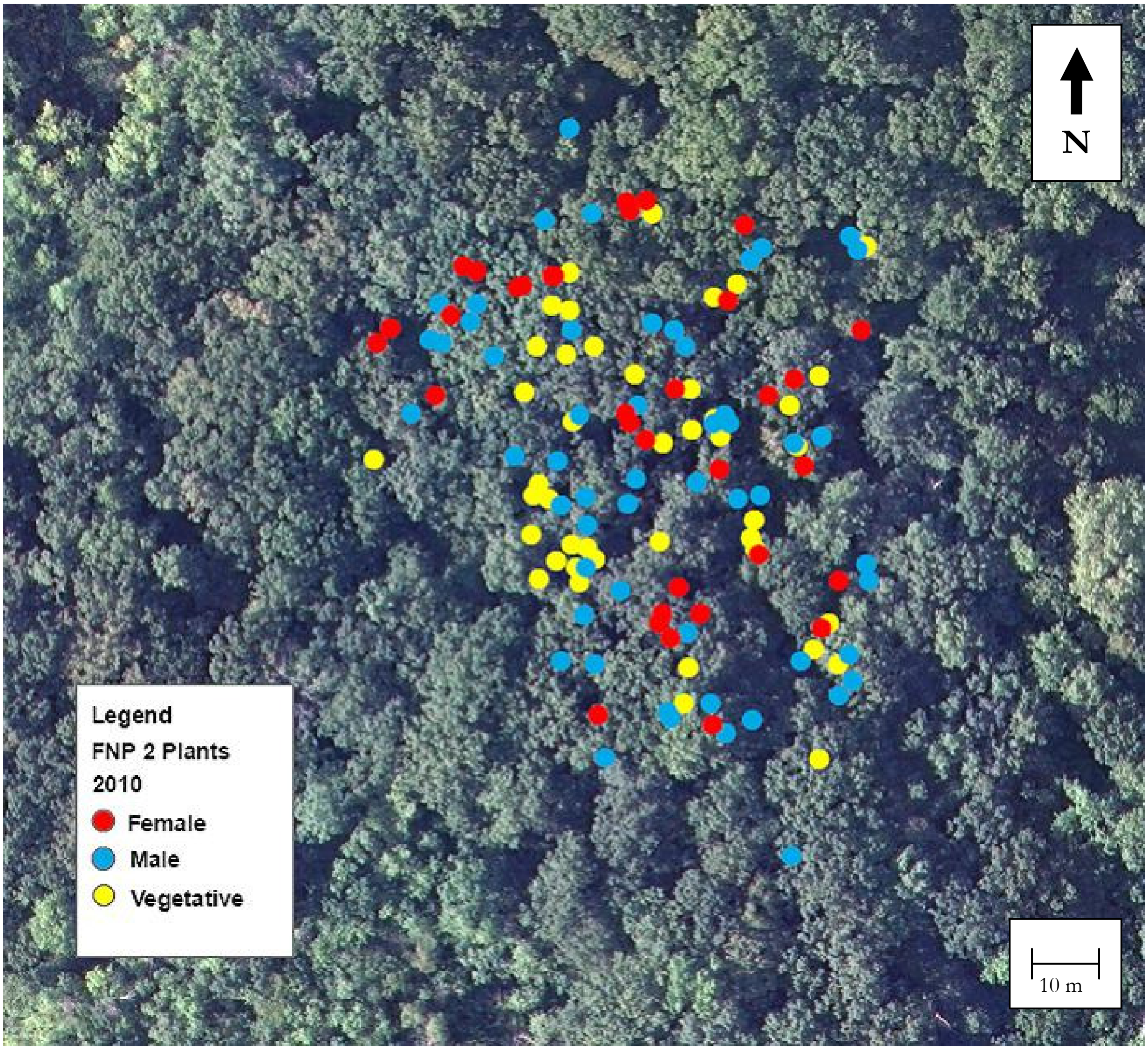
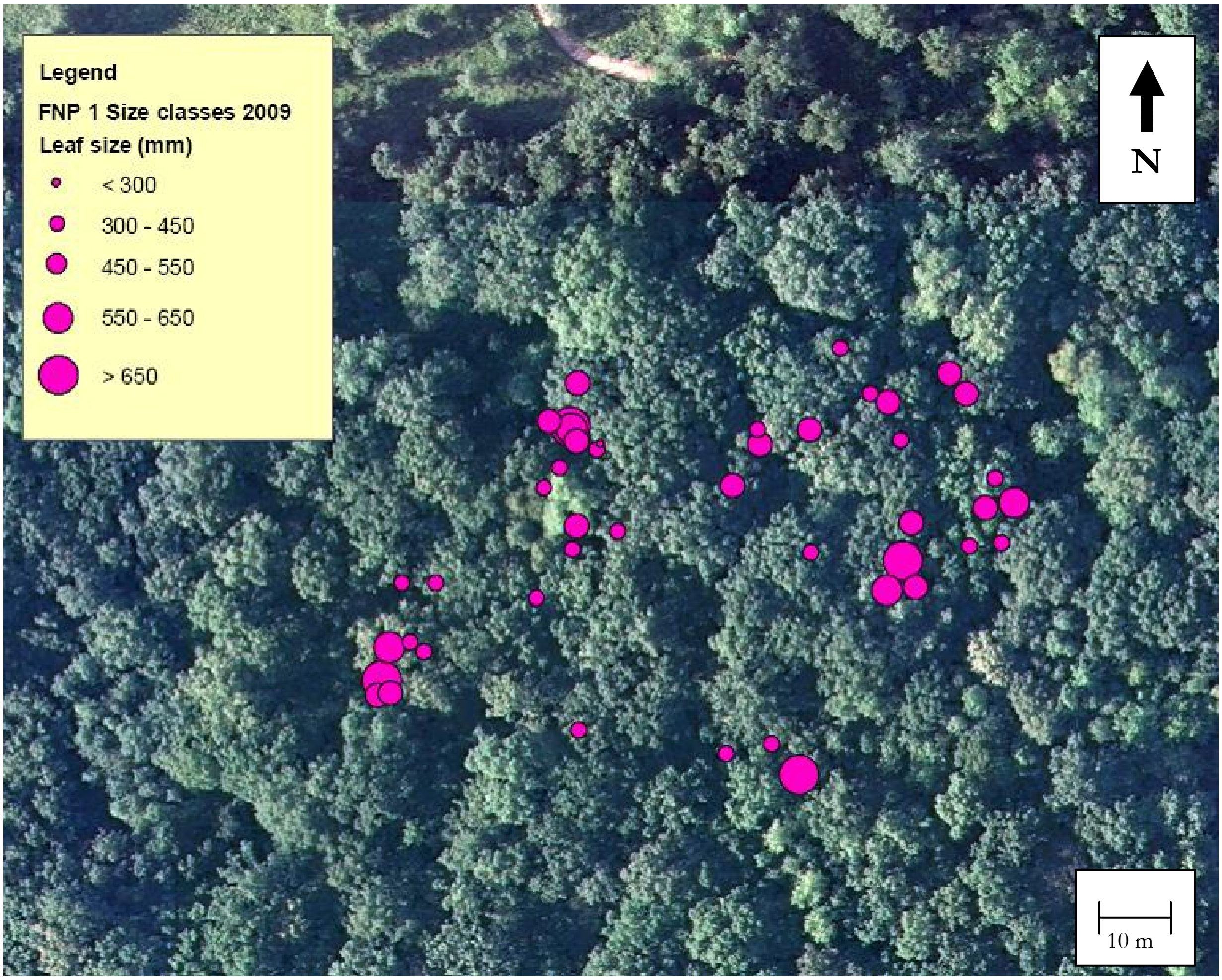
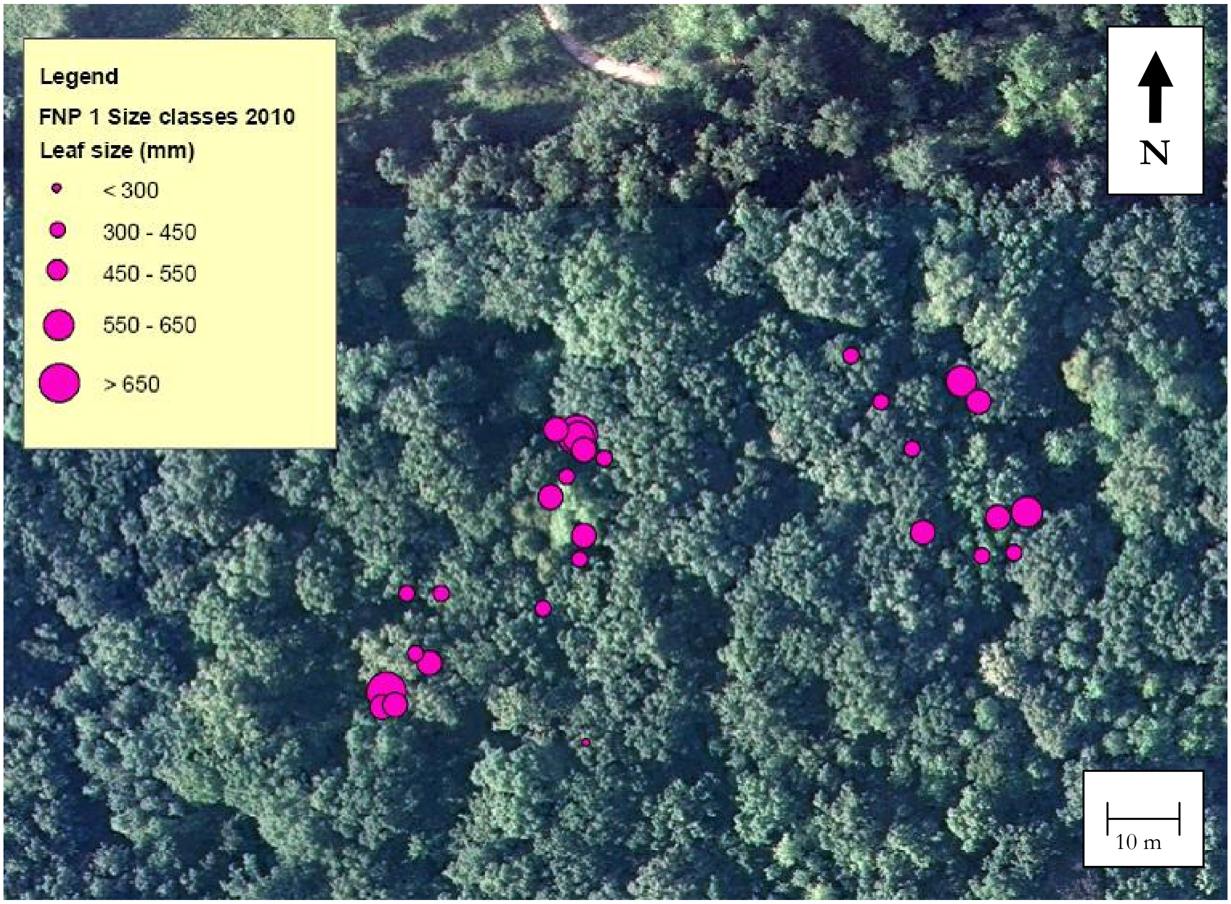


Figure 2-8. GIS maps showing distributions of plant size in the male-biased population with larger circles indicating larger plants and smaller circles indicating smaller plants.

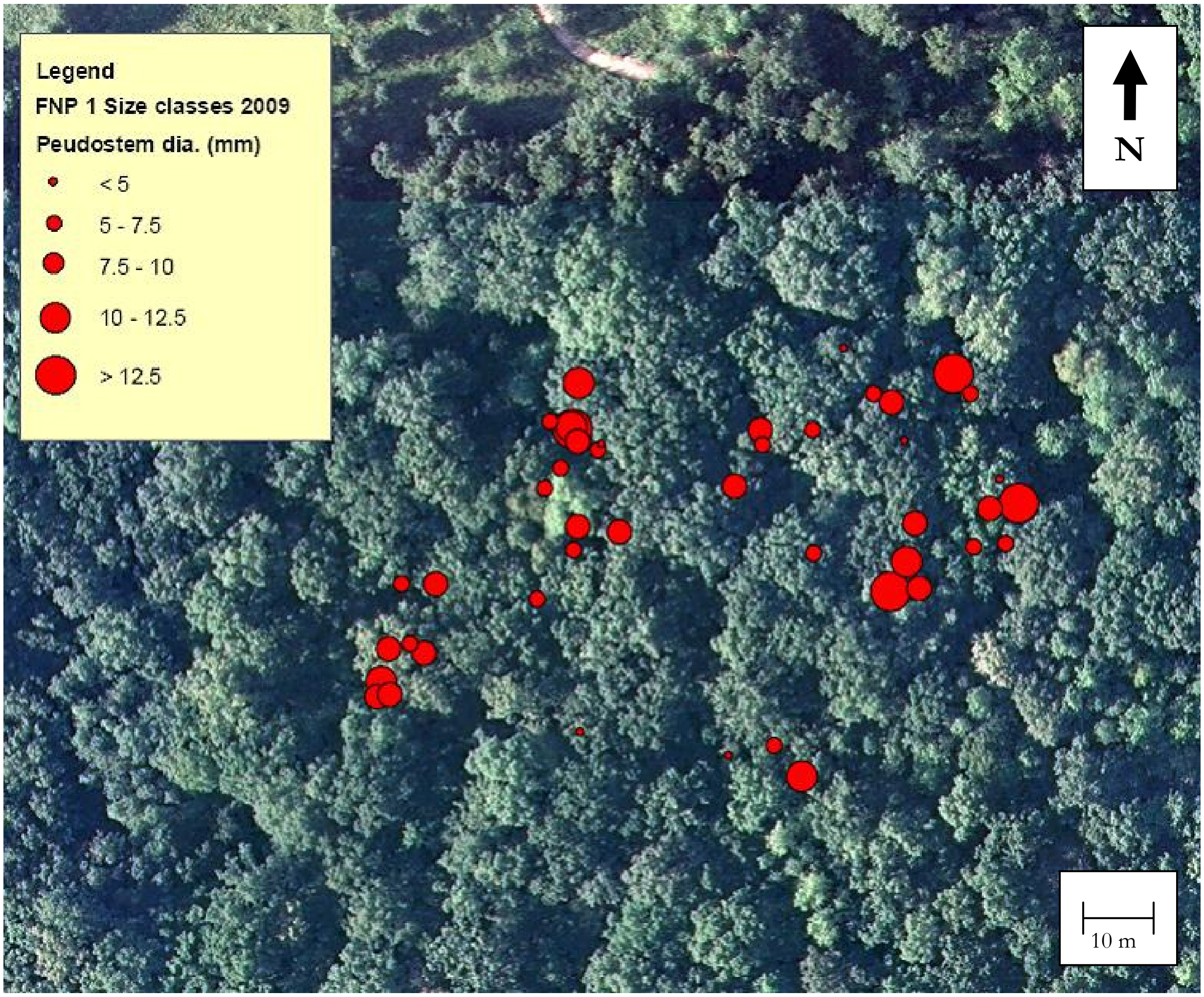
(a) Leaf size 2009



(b) Leaf size 2010



(c) Stem size 2009



(d) Stem size 2010

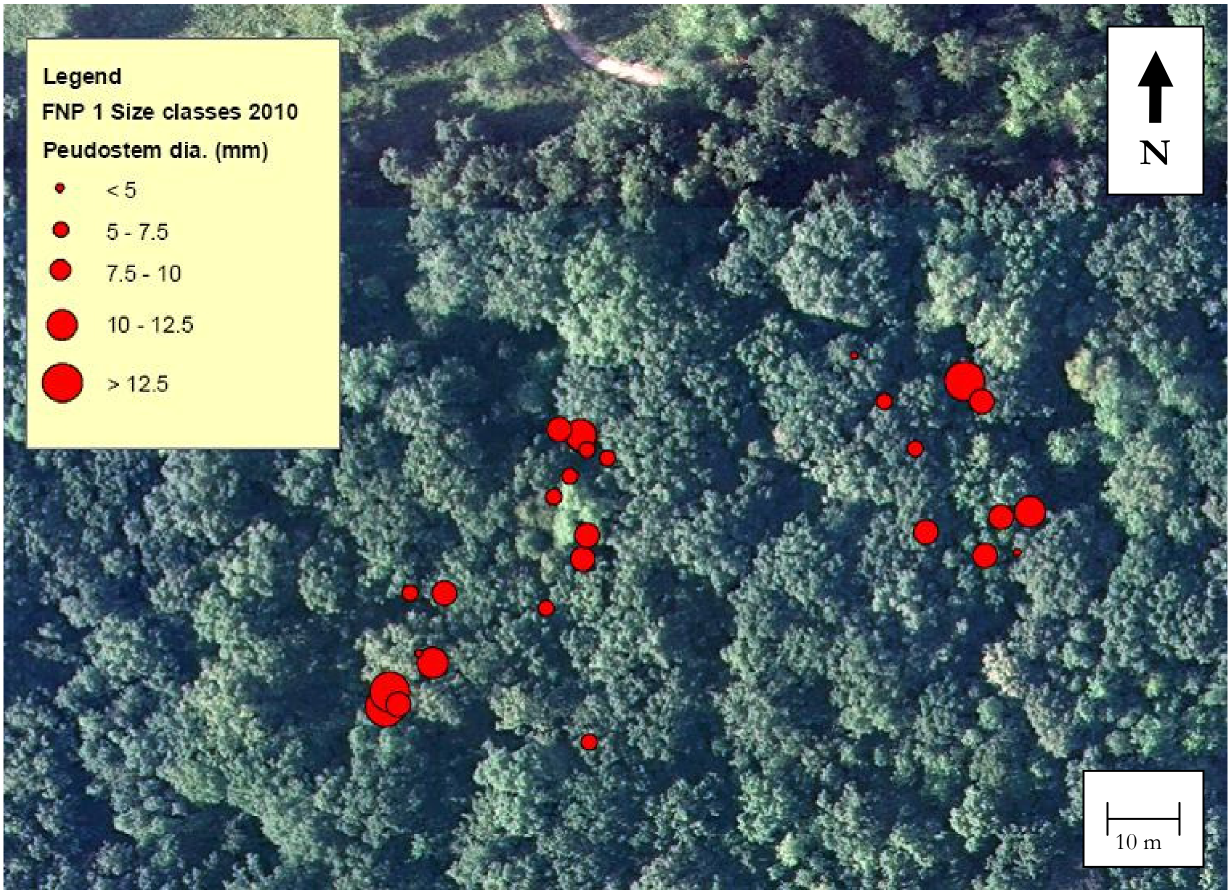
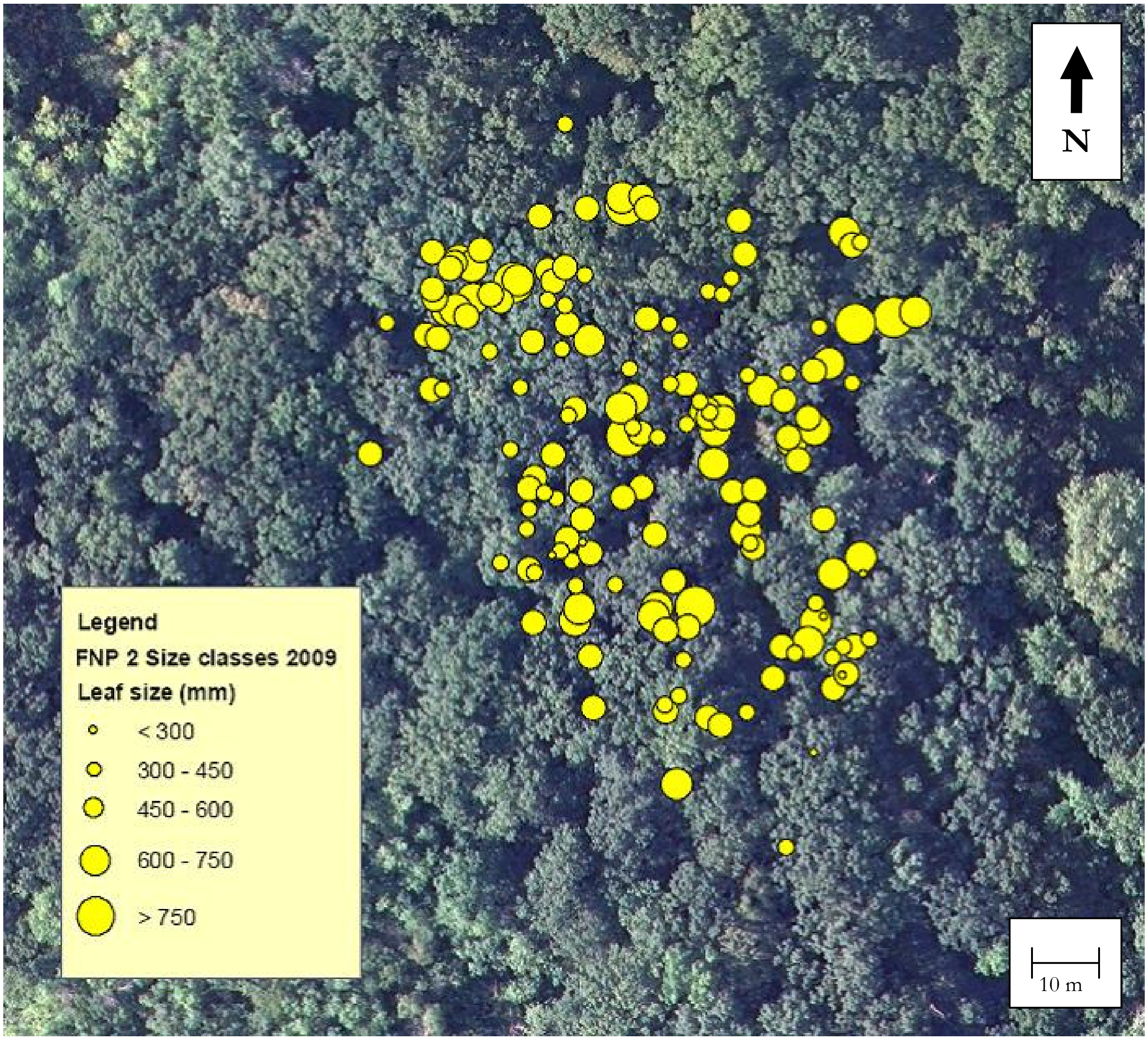
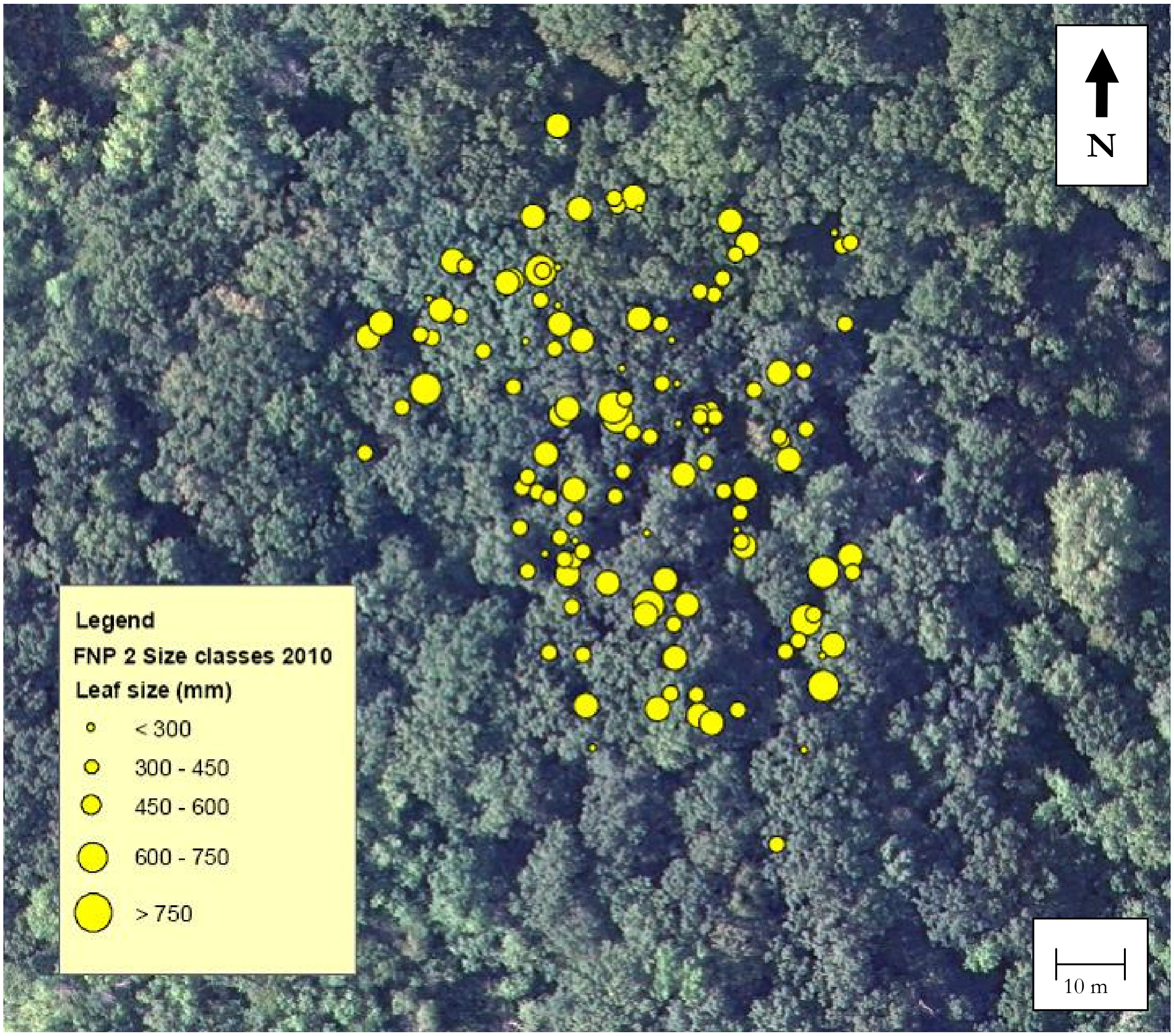


Figure 2-9. GIS maps showing distributions of plant size in the female-biased population with larger circles indicating larger plants and smaller circles indicating smaller plants.

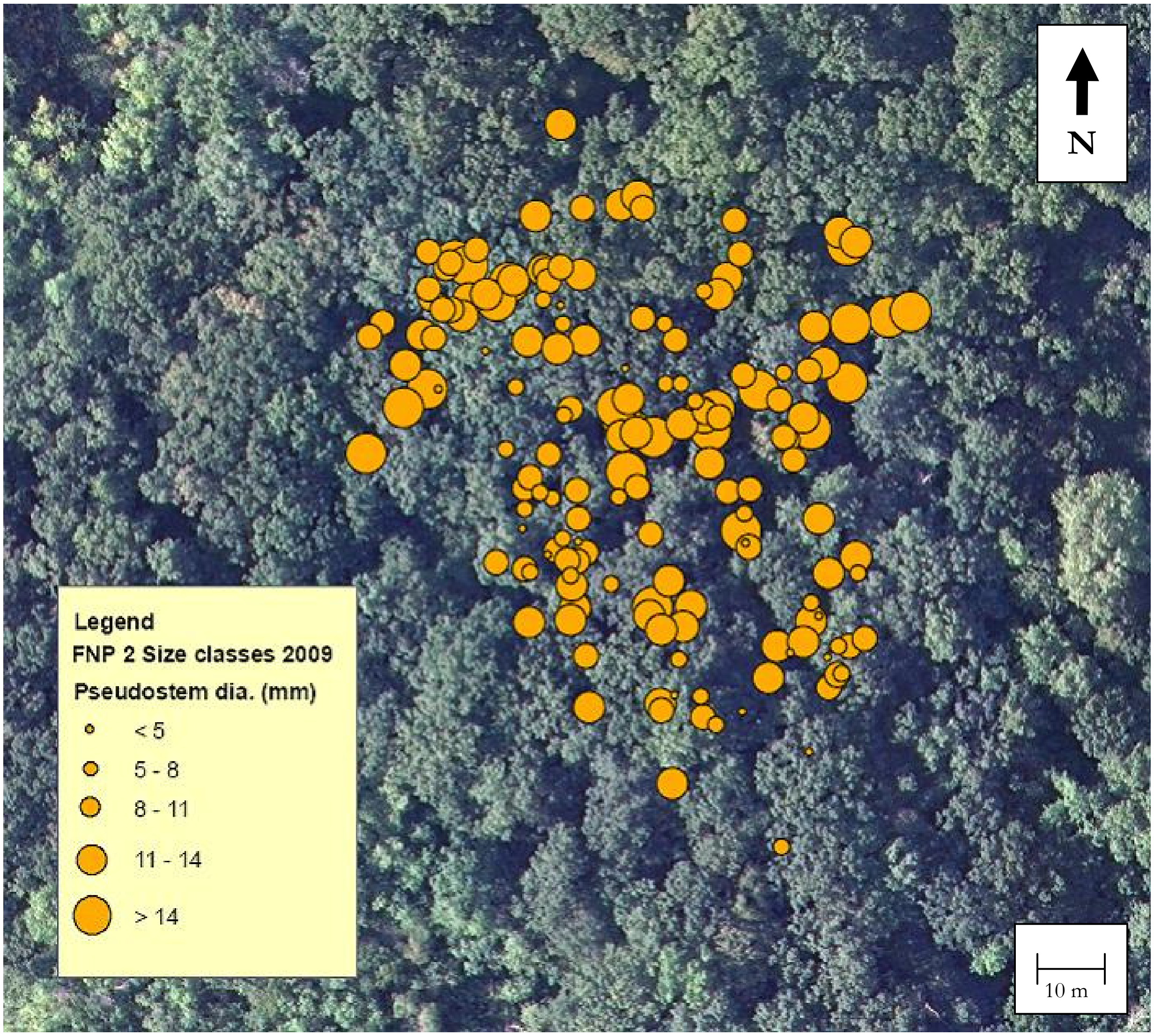
(a) Leaf size 2009



(b) Leaf size 2010



(c) Stem size 2009



(d) Stem size 2010

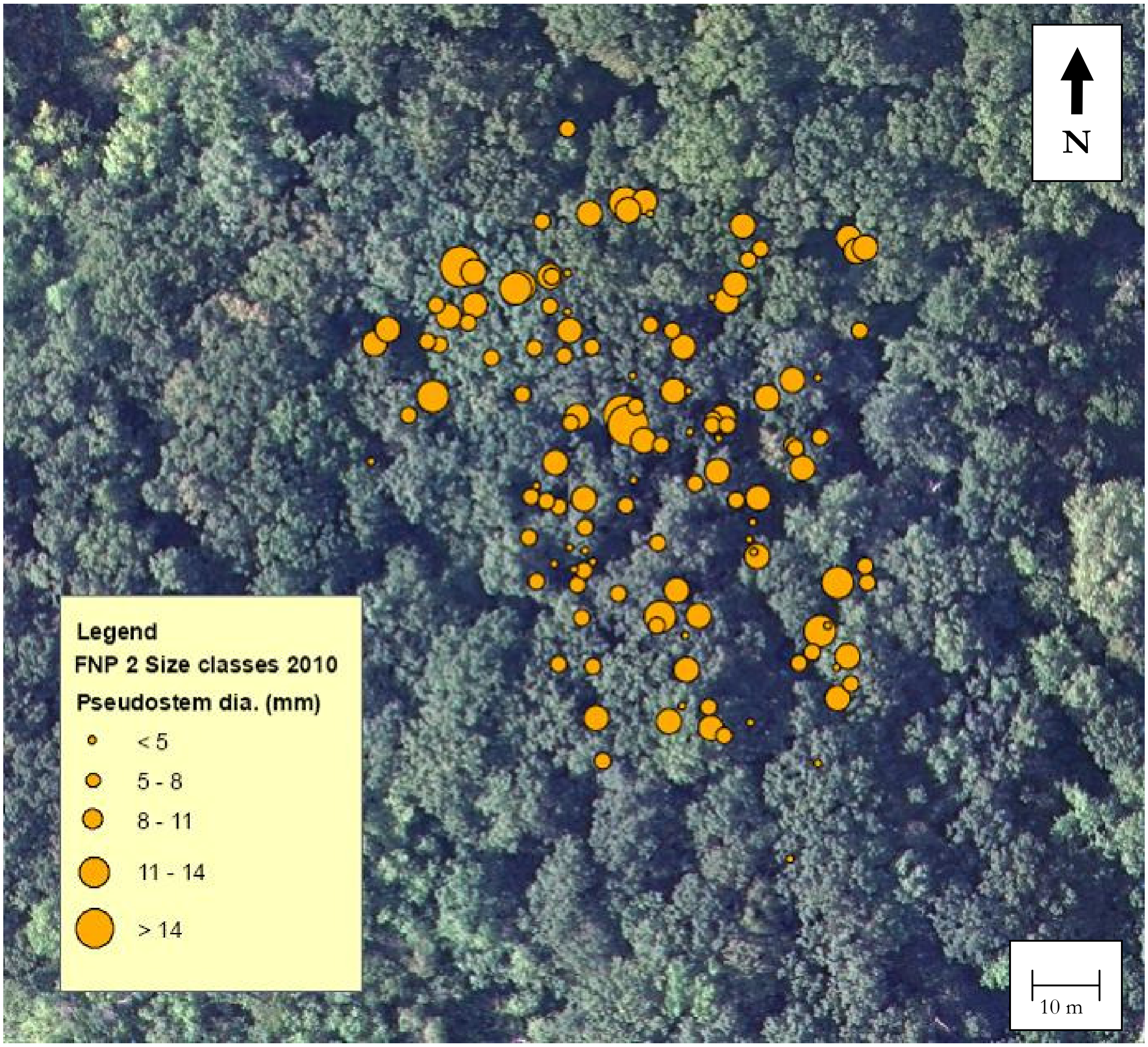
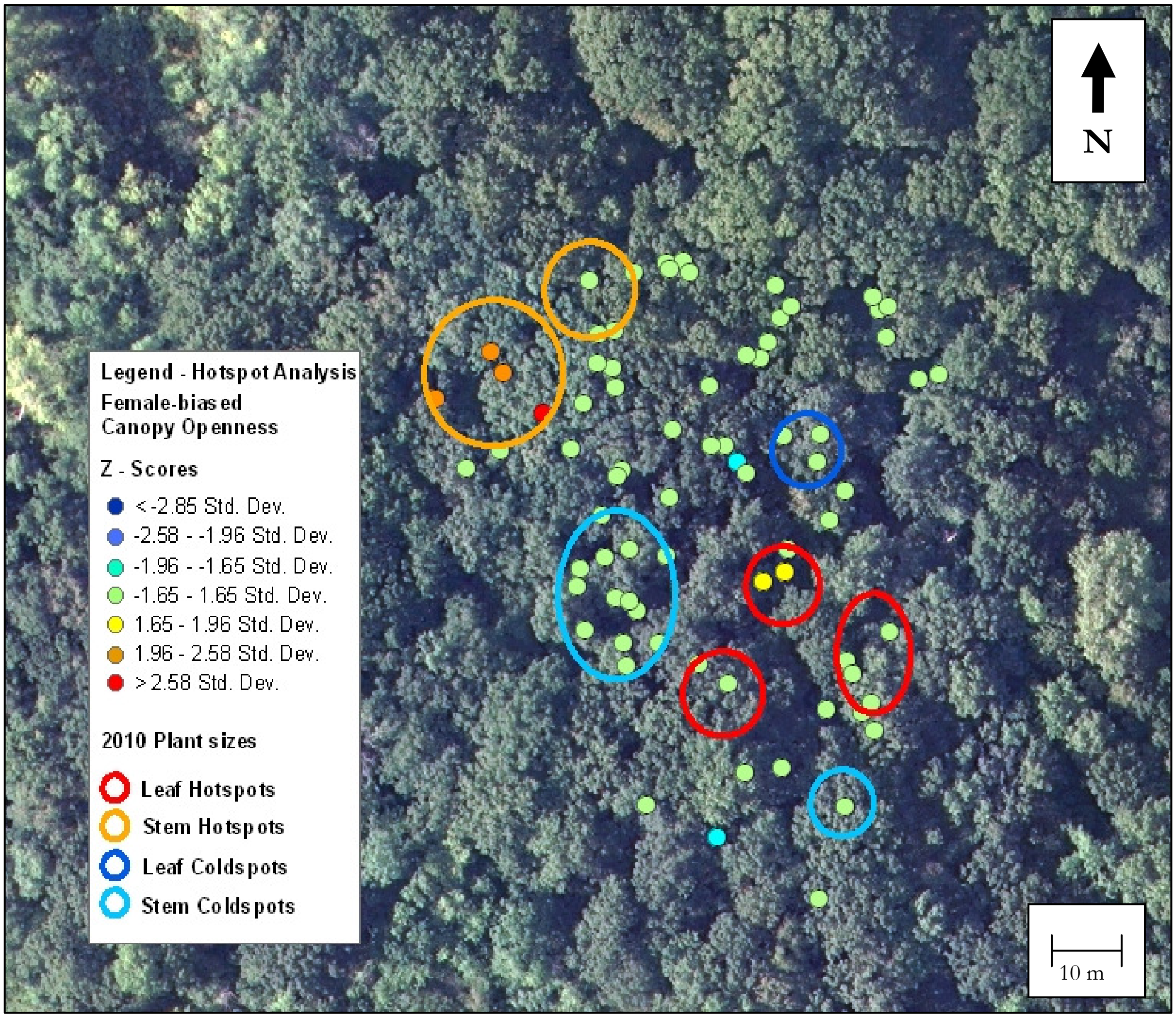
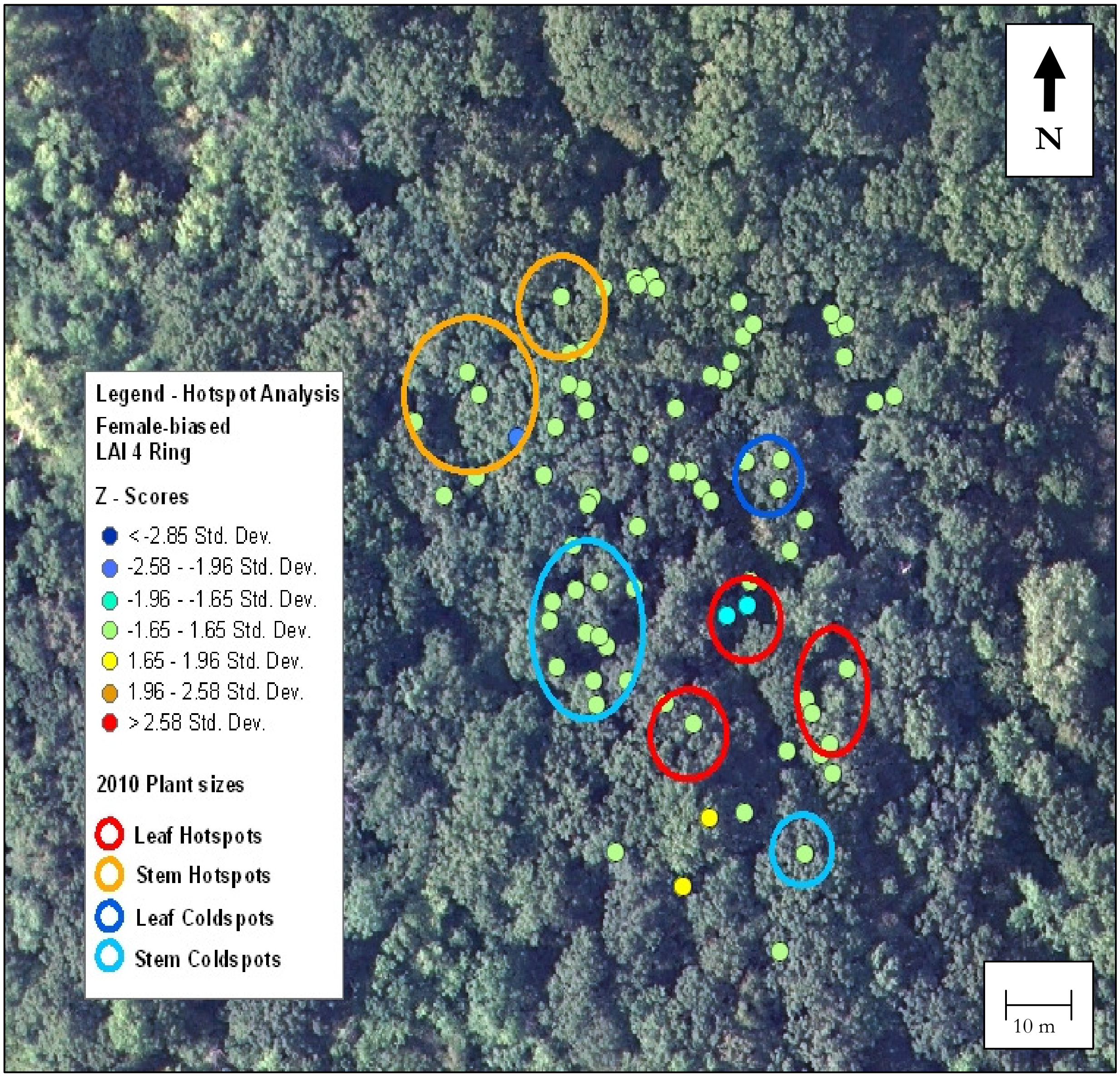


Figure 2-10. GIS maps showing the overlap of size and canopy variable hotspots in the female-biased population. The large orange and red circles are areas of large stem and leaf size respectively, while the large light and dark blue circles are areas of small stem and leaf size respectively. The dots represent each plant where a canopy photo was taken, with more reddish colored dots indicating hotspots of higher values for each variable and bluish colored dots indicating cold spots of lower values. The canopy variables investigated were (a) Canopy Openness, (b) LAI 4 Ring, (c)LAI 5 Ring, (d) % diffuse solar radiation, (e) % direct solar radiation, and (f) % total radiation.

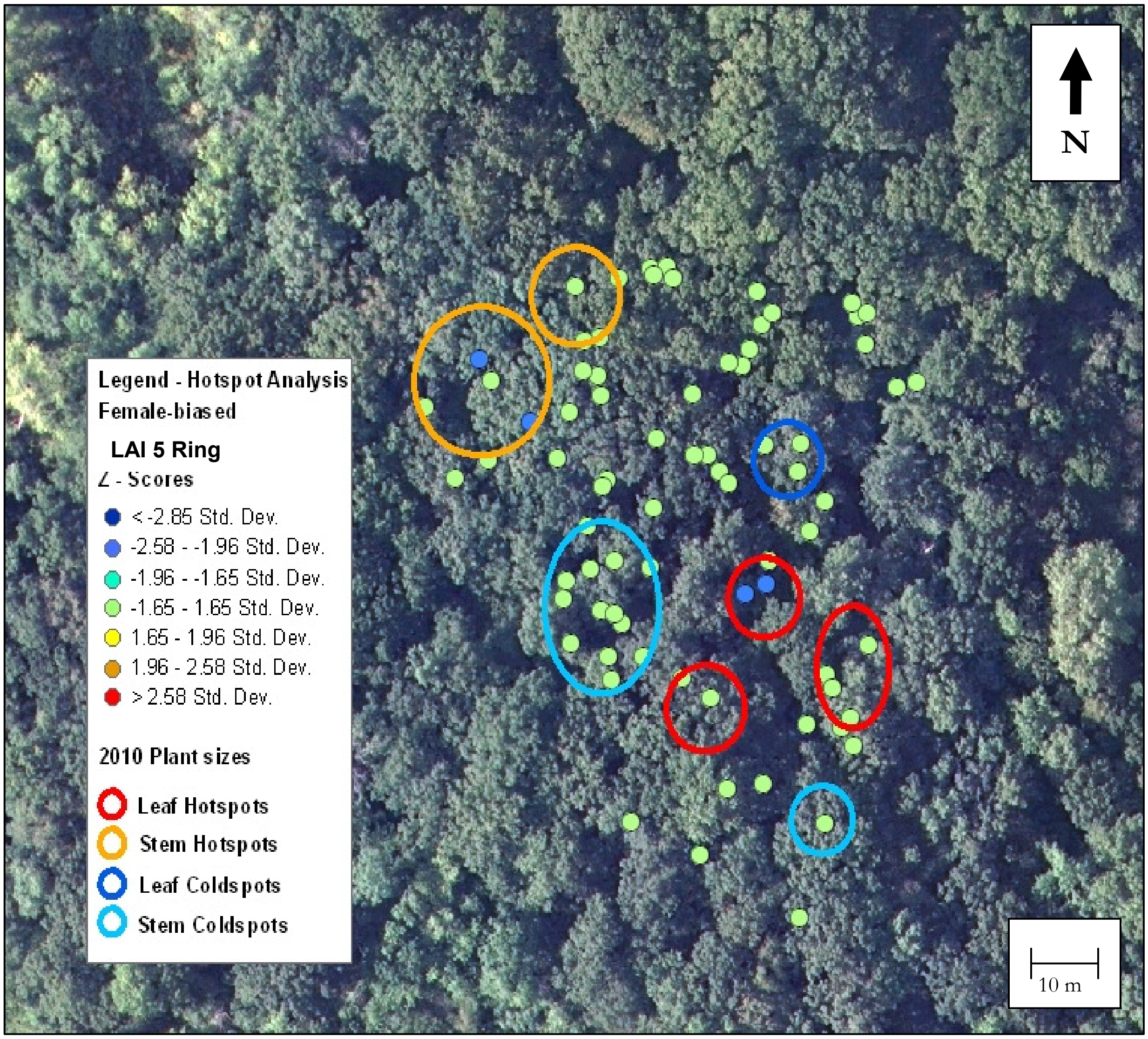
(a)



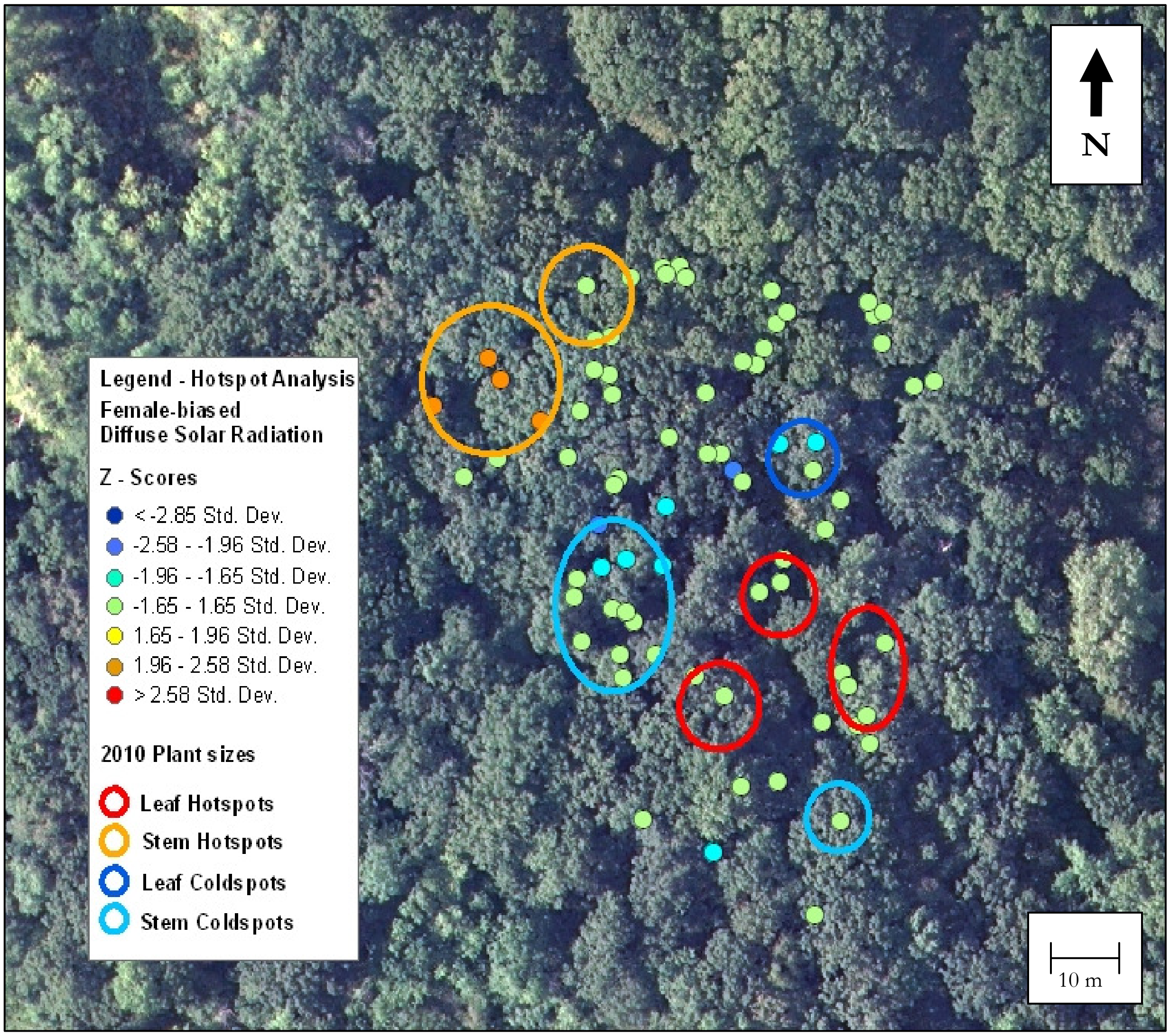
(b)


Legend - Hotspot Analysis
Female-biased
LAI 4 Ring
Z - Scores
● < -2.85 Std. Dev.
● -2.58 - -1.96 Std. Dev.
● -1.96 - -1.65 Std. Dev.
● -1.65 - 1.65 Std. Dev.
● 1.65 - 1.96 Std. Dev.
● 1.96 - 2.58 Std. Dev.
● > 2.58 Std. Dev.
2010 Plant sizes
○ Leaf Hotspots
○ Stem Hotspots
○ Leaf Coldspots
○ Stem Coldspots
N
10 m

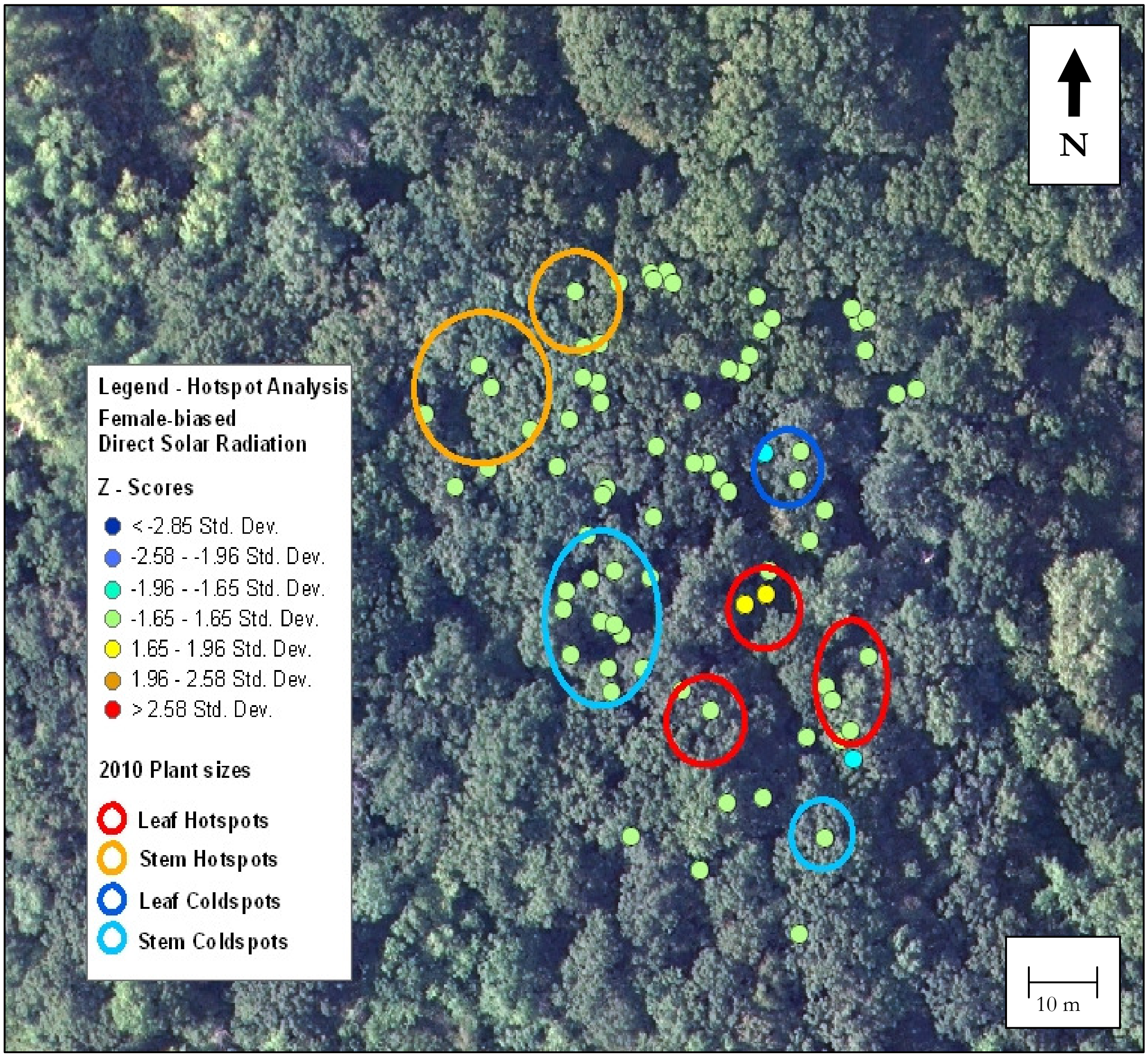
(c)


N
Legend - Hotspot Analysis
Female-biased
LAI 5 Ring
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
2010 Plant sizes
Leaf Hotspots
Stem Hotspots
Leaf Coldspots
Stem Coldspots
10 m

(d)


N
Legend - Hotspot Analysis
Female-biased
Diffuse Solar Radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
2010 Plant sizes
Leaf Hotspots
Stem Hotspots
Leaf Coldspots
Stem Coldspots
10 m

(e)


N
Legend - Hotspot Analysis
Female-biased
Direct Solar Radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
2010 Plant sizes
Leaf Hotspots
Stem Hotspots
Leaf Coldspots
Stem Coldspots
10 m

(f)

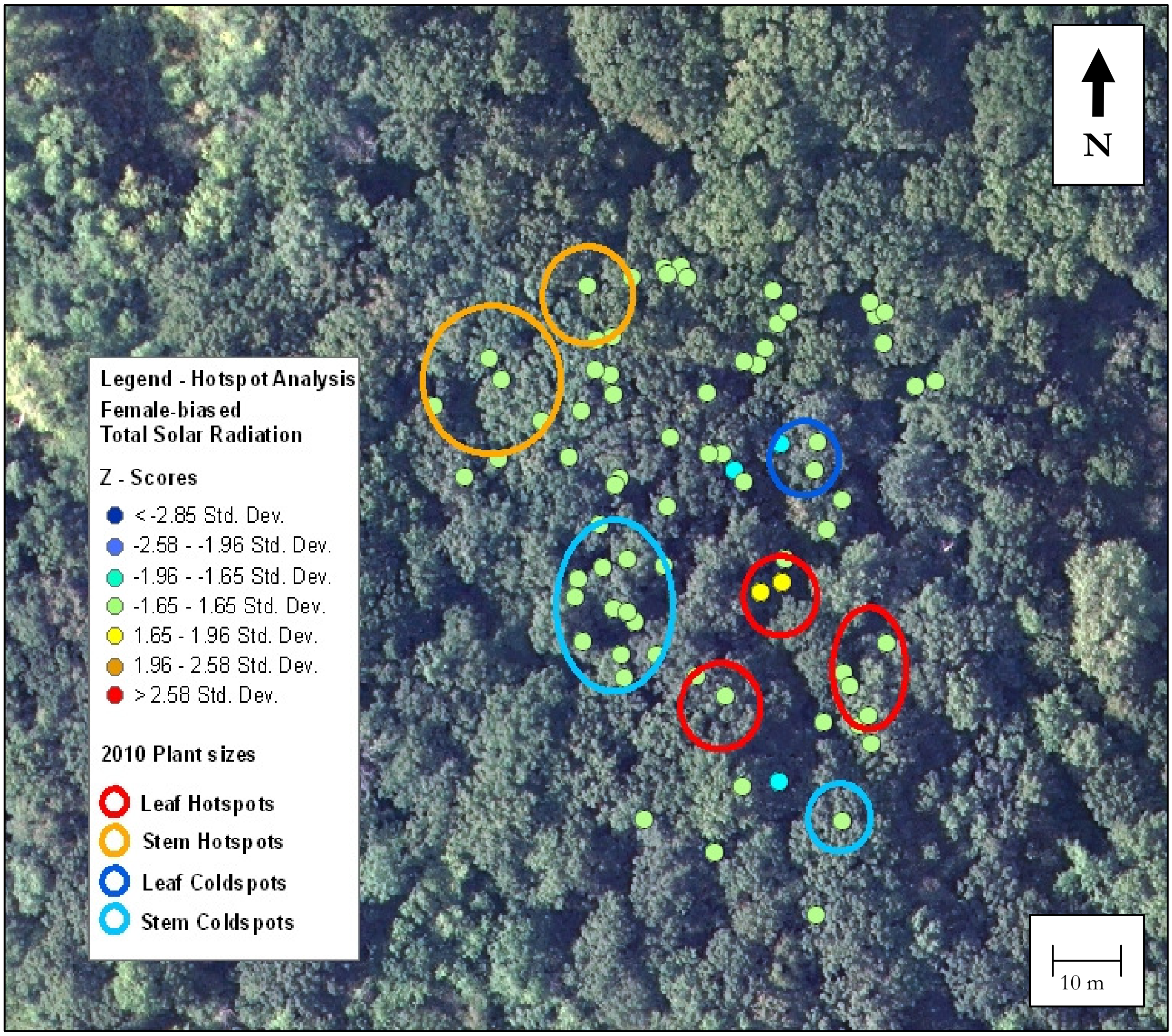
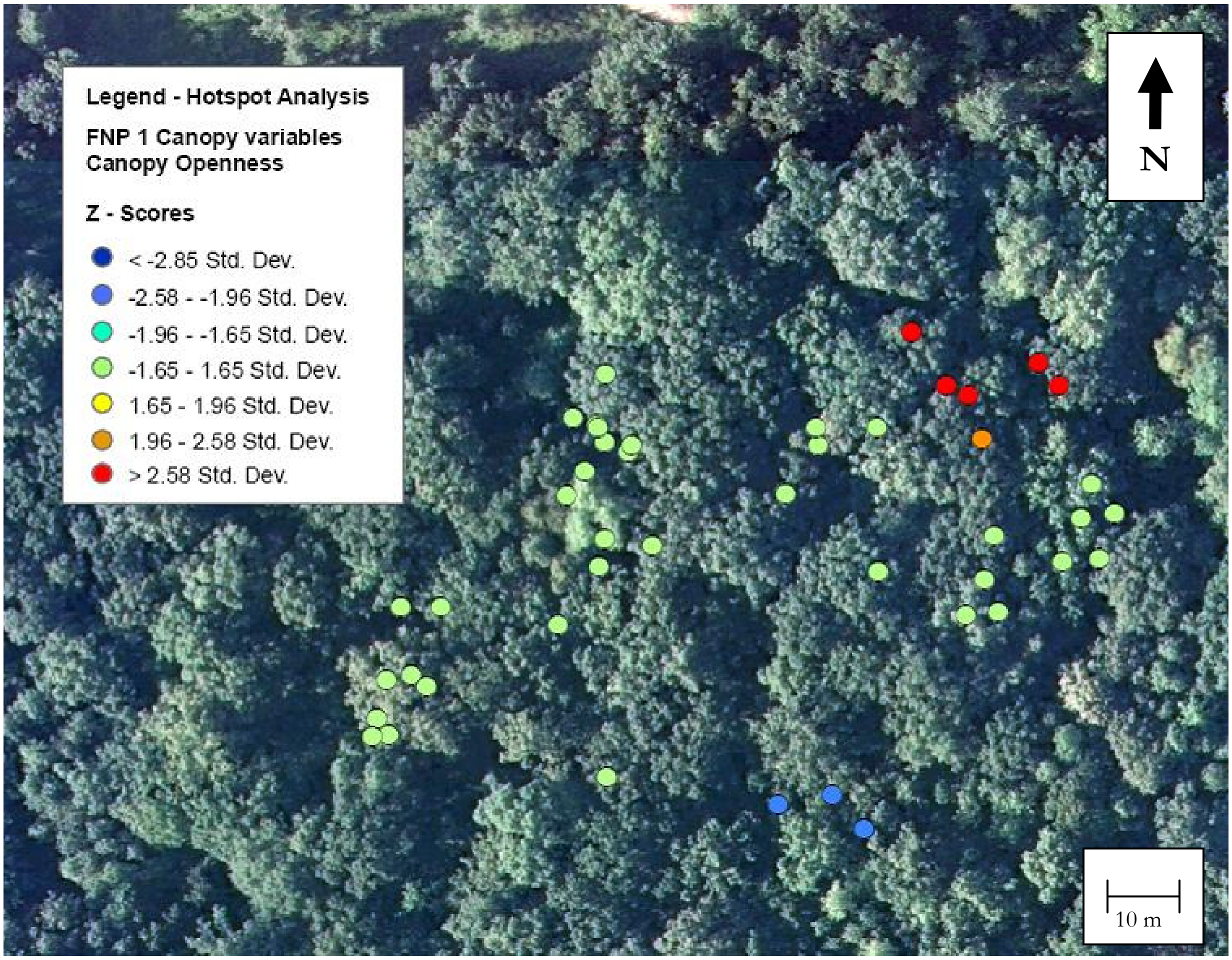
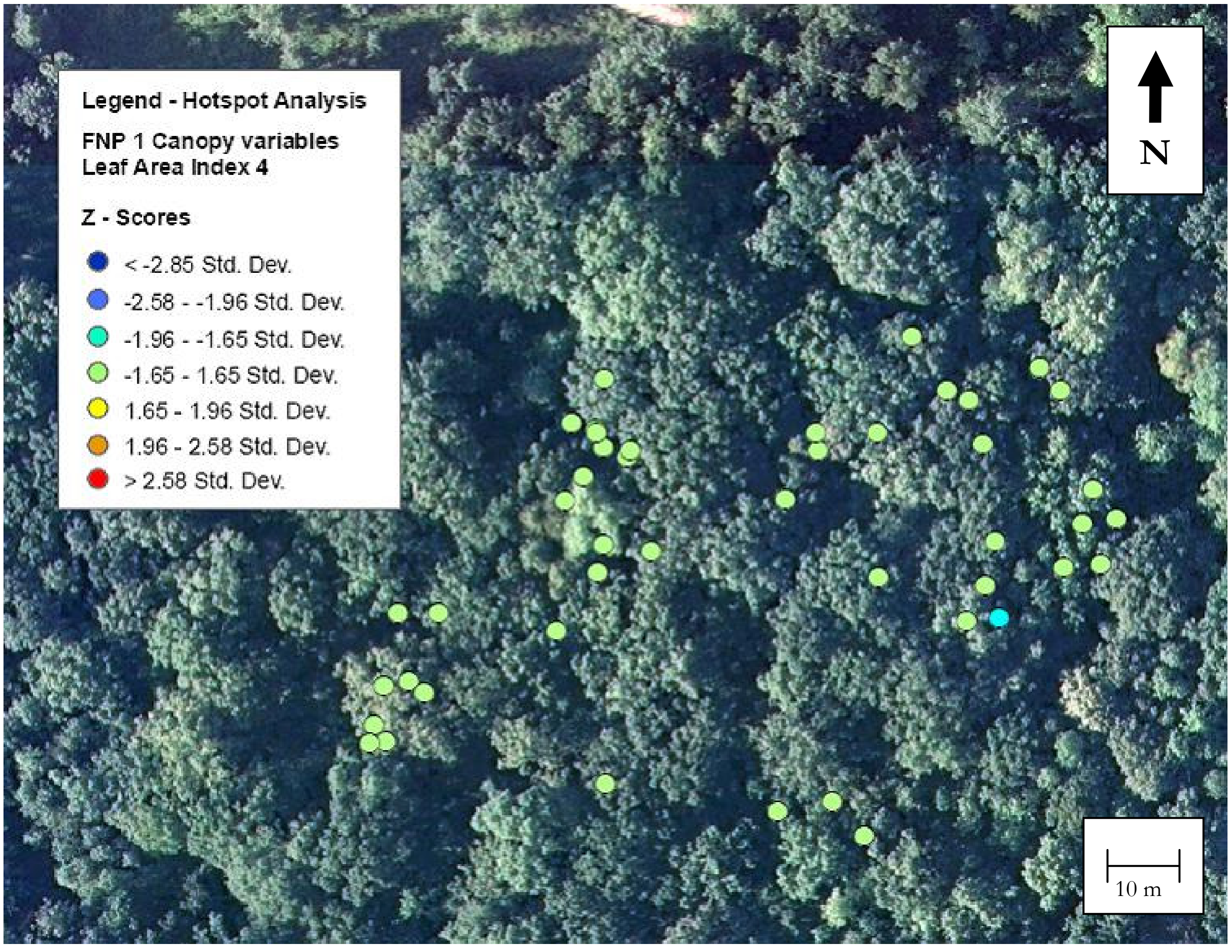

N
Legend - Hotspot Analysis
Female-biased
Total Solar Radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
2010 Plant sizes
Leaf Hotspots
Stem Hotspots
Leaf Coldspots
Stem Coldspots
10 m

Figure 2-11. GIS map showing hotspots calculated for the six different canopy variables in the male-biased population. The dots represent each plant where a canopy photo was taken, with more reddish colored dots indicating hotspots of higher values for each variable and bluish colored dots indicating cold spots of lower values. The canopy variables investigated were (a) Canopy Openness, (b) LAI 4 Ring, (c)LAI 5 Ring, (d) % diffuse solar radiation, (e) % direct solar radiation, and (f) % total radiation.

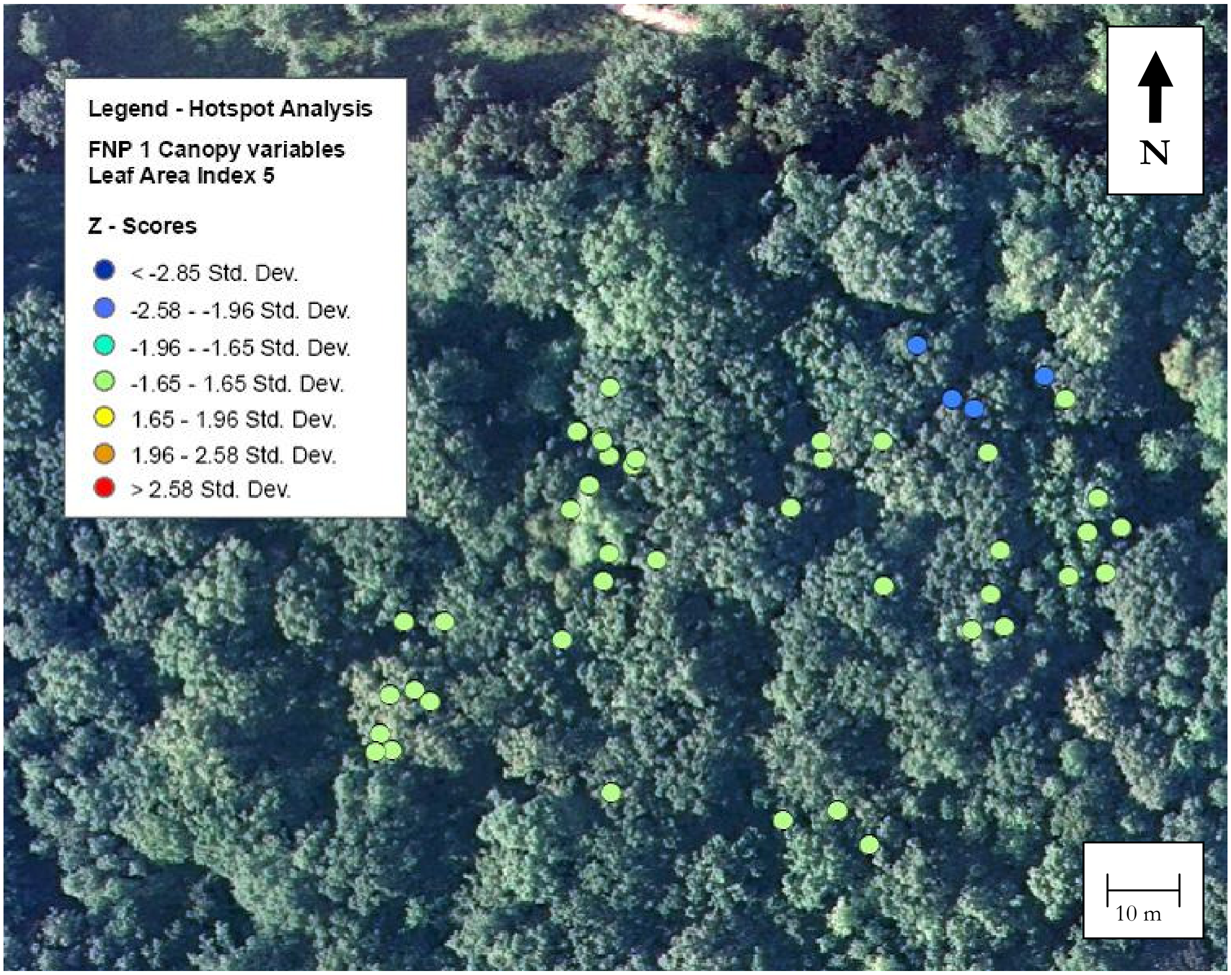
(a)



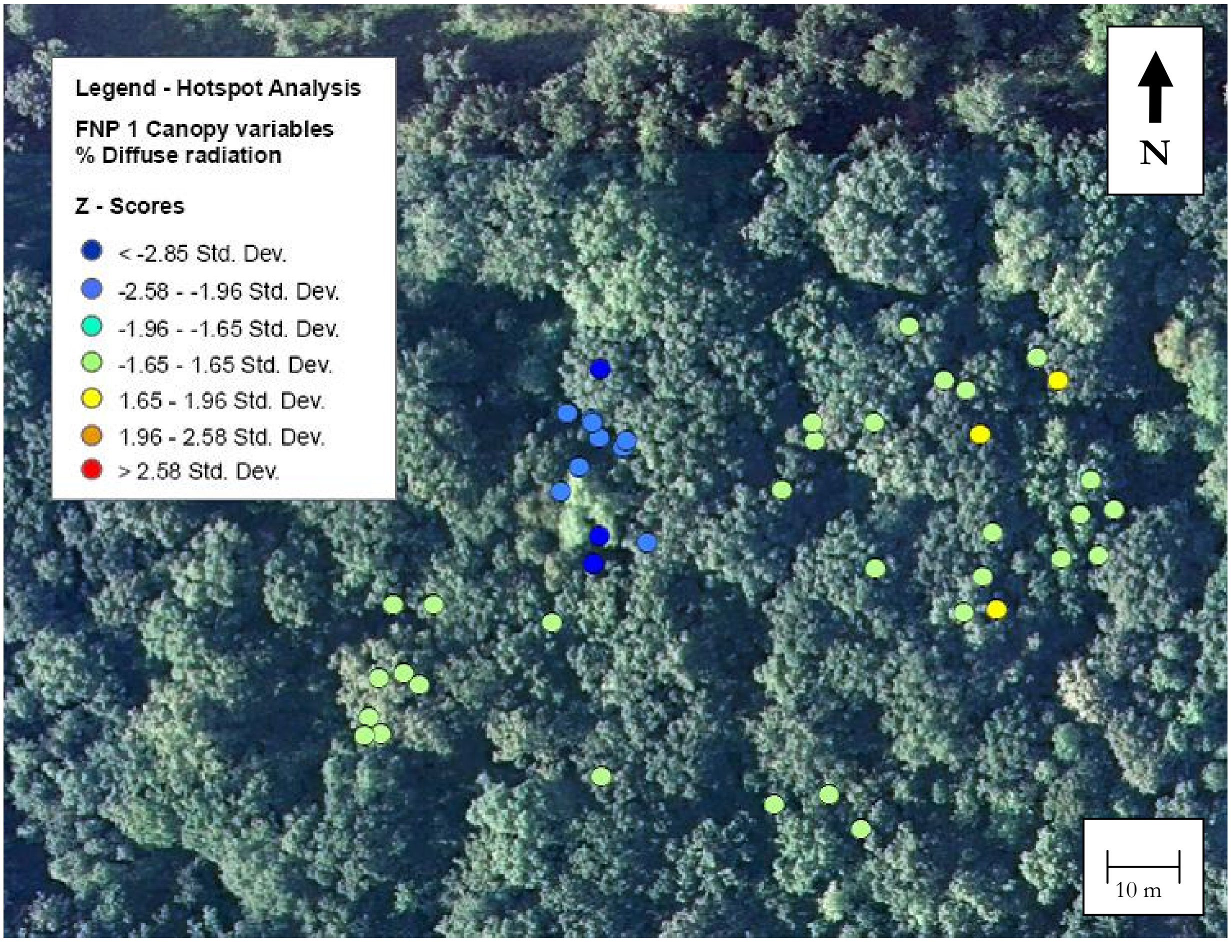
(b)


Legend - Hotspot Analysis
FNP 1 Canopy variables
Leaf Area Index 4
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
N
10 m

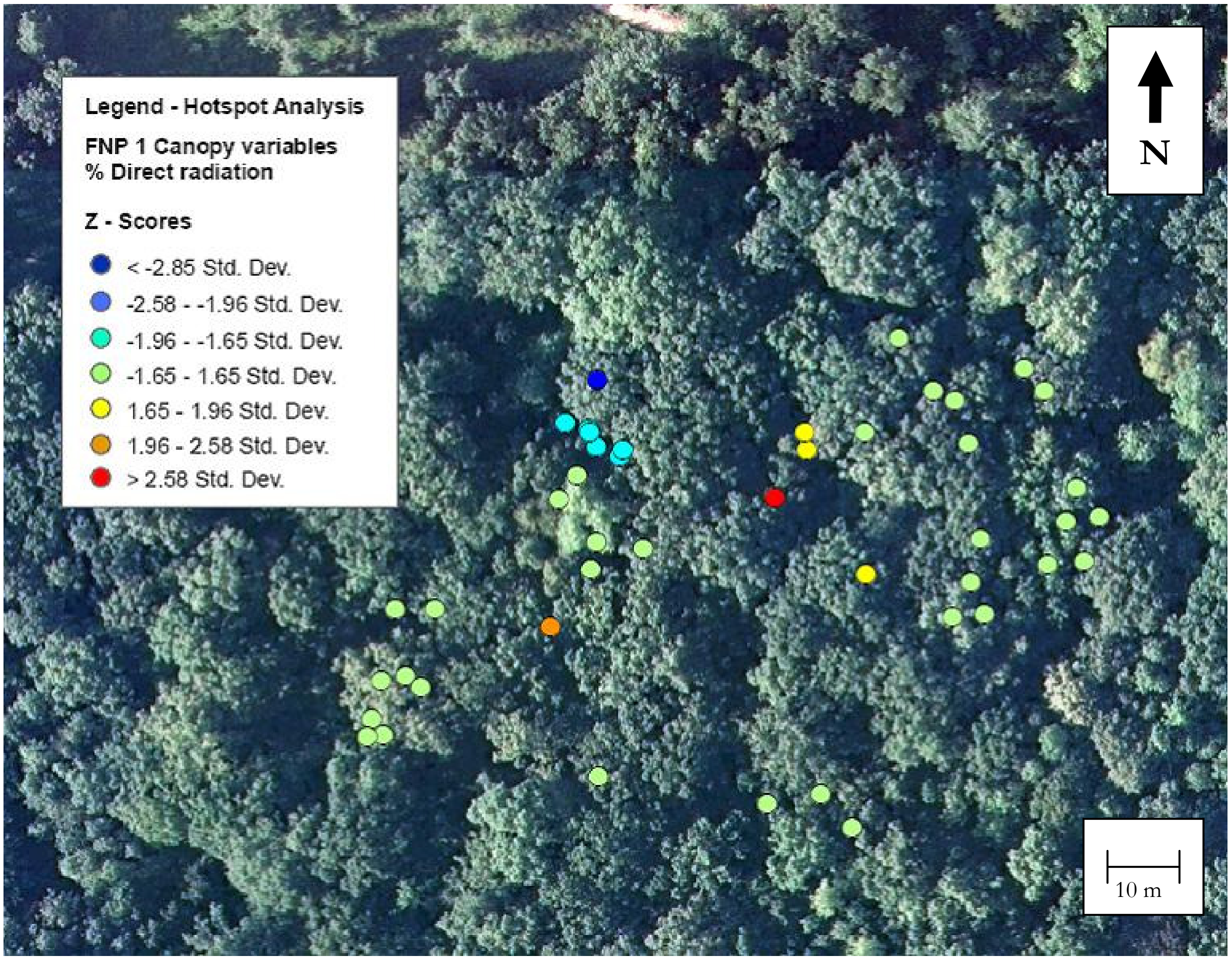
(c)


Legend - Hotspot Analysis
FNP 1 Canopy variables
Leaf Area Index 5
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
N
10 m

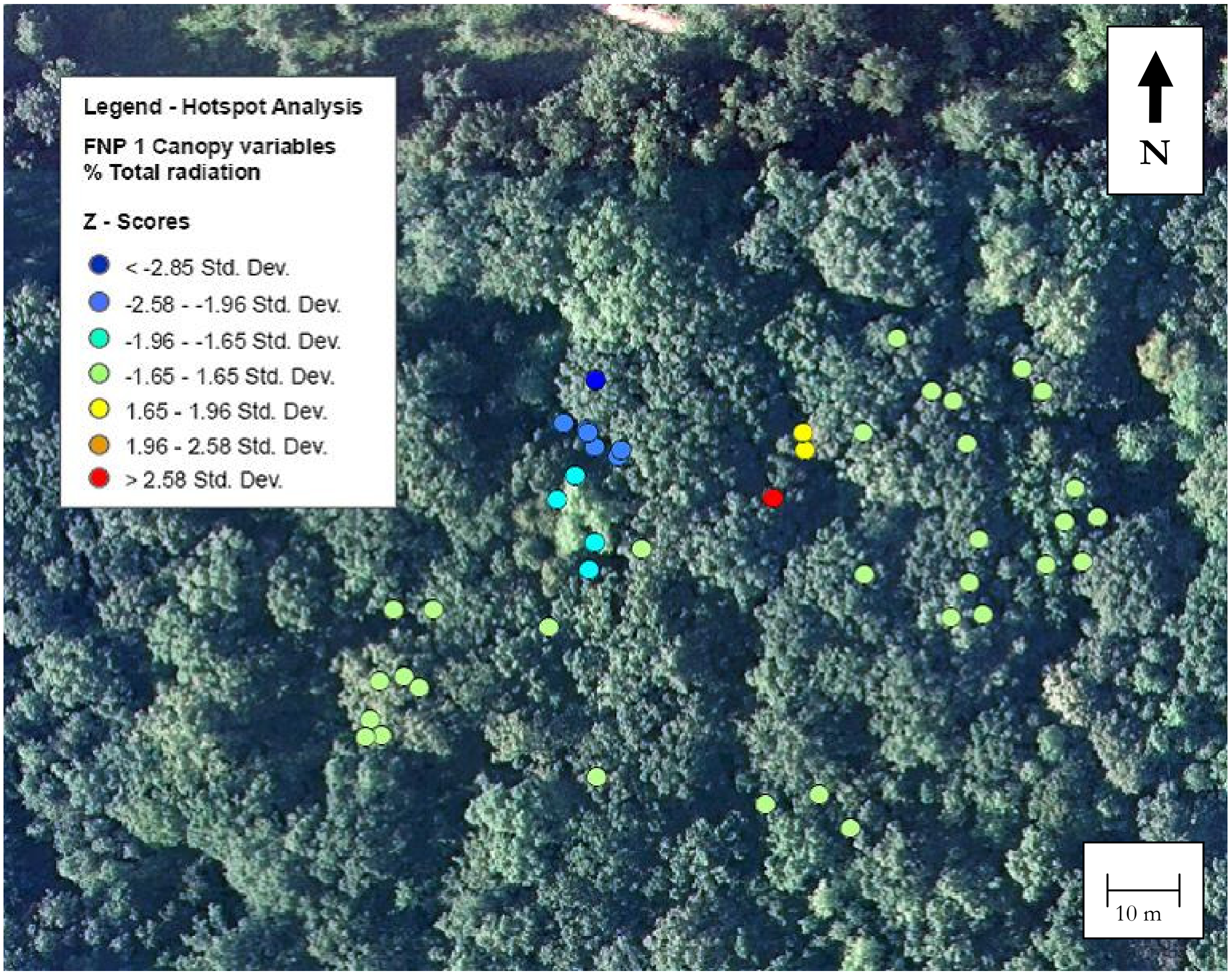
(d)


Legend - Hotspot Analysis
FNP 1 Canopy variables
% Diffuse radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
N
10 m

(e)


Legend - Hotspot Analysis
FNP 1 Canopy variables
% Direct radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
N
10 m

(f)


Legend - Hotspot Analysis
FNP 1 Canopy variables
% Total radiation
Z - Scores
< -2.85 Std. Dev.
-2.58 - -1.96 Std. Dev.
-1.96 - -1.65 Std. Dev.
-1.65 - 1.65 Std. Dev.
1.65 - 1.96 Std. Dev.
1.96 - 2.58 Std. Dev.
> 2.58 Std. Dev.
N
10 m

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