

Seed Size Variation in the Cleistogamous species, *Ruellia humilis*

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Abstract

Plant mating systems describe the relative production of self and outcrossed seeds in a population. Mating systems effect the genetic variation, individual fitness, population persistence and evolutionary change in a population. Cleistogamous species exhibit a mixed mating system in which both closed, cleistogamous (CL) flowers and open, chasmogamous (CH) flowers are produced on individual plants. Understanding the selective forces that favor the stable production of both flowers may provide insight into conditions that favor mixed mating systems in general. Using *R. humilis* as a study system the seed sizes between CL and CH flowers were examined to determine any potential differences between the flowers.

Keywords: cleistogamous, chasmogamous, seed size, mating systems

Introduction

Cleistogamous species provide strong evidence of stable mixed mating systems (Oakley and Winn 2008). However, the production of heteromorphic flowers prompts questions about the effects of cleistogamous species on evolution (Goodwillie Kalisz and Eckert 2005). Cleistogamous species are capable of producing cleistogamous flowers (CL) that are closed and self-fertilize and chasmogamous flowers (CH) that can be pollinated (Oakley Moriuchi and Winn 2007). The production of both flowers gives cleistogamous species a fitness advantage as they are able to mate with others by siring another individual's seeds and with themselves (Oakley et al. 2007). The genetic variation between plants depends on their degree of relatedness and affects how the species responds to natural selection (Oakley & Winn 2008).

CH and CL flowers differ in flowering times and vary in abundance at different times of the year (Oakley & Winn 2008). Since the flowers produced determine the parental fitness, it can be concluded that

the different flowers have more fitness at different times (Oakley et al. 2007). The production of both CL and CH flowers is then an adaptive phenotype that allows the plants to maintain a level of fitness for an extended period of time (Oakley & Winn 2008).

The reliability and the energetic cost of producing CL flowers are thought to offset the effects of inbreeding depression (Oakley & Winn 2008). However, CH flowers are capable of two functions that CL flowers are not. CH flowers increase the genetic variation in a population by mating with other individuals by utilizing pollinators (Oakley & Winn 2008). The increase in allele recombination could increase the plant's ability to adapt to changes in the environment (Oakley & Winn 2008). This gives the CH flowers an evolutionary advantage over CL flowers (Goodwillie et al. 2005). Outcrossing also increases the fitness of male plants since they are able to sire the offspring of other individuals as well as their own (Oakley & Winn 2008). CL flowers do not benefit from

the recombination of alleles that would increase their ability to adapt to changes when there is more competition (Goodwillie et al. 2005). CL offspring are genetically identical whereas CH offspring are more dissimilar (Oakley & Winn 2008). As a result when CL offspring are more likely to experience a reduction in fitness when there is competition (Oakley & Winn 2008). CH siblings would not experience a large reduction in competition because of the genetic variation between them (Oakley & Winn 2008). Therefore, when the environment undergoes changes and competition increases, chasmogamous reproduction would be favored. When there is a stable environment and low competition cleistogamous reproduction would be selected for because they are less risky to produce (Oakley et al. 2007).

The differences between the mating patterns results in offspring that vary in their genetic similarity (Goodwillie et al. 2005). CL flowers are less costly but more reliable for the plant to produce, yet they do not benefit from genetic variation (Oakley et al. 2007). On the other hand, CH flowers provide the genetic variation needed for evolution but rely on pollinators to breed (Oakley et al. 2007). The differences in the flowers could have an impact on the fitness of the seeds produced by the different types of mating. As a first step towards understating the cost of reproduction via CH and CL flowers we examined seed size differences between CH and CL flowers and hypothesized that CH seeds would be larger.

Methods

The system used to study size differences was *Ruellia humilis* Nutt. (Acanthaceae) or fringleaf wild ruellia. *Ruellia humilis* exhibits mixed mating system by producing both the closed CL flowers and the open CH flowers. Three CH and CL seeds were collected from each of

eight maternal *R. humilis* individuals at Lake Carl Blackwell. The seeds were photographed using a Leica stereomicroscope with an attached digital camera system. Each seed image was measured for length, width and surface area using ImageJ. We then performed a two-way analysis of variance (ANOVA) to determine whether seed types, maternal family, or their interaction influenced seed size traits.

Results

The analysis did not reveal any significant differences in seed length ($F_{1,27} = 0.25$, $P = 0.62$), width ($F_{1,27} = 1.55$, $P = 0.22$), nor surface area ($F_{1,27} = 0.39$, $P = 0.54$) for seed measurements as a whole (Figure 1). However, we found a significant interaction between seed type and maternal family for seed width ($F_{7,27} = 6.52$, $P = 0.0001$) and surface area ($F_{7,27} = 4.83$, $P = 0.0012$), but not seed length ($F_{7,27} = 1.39$, $P = 0.25$) (Figure 2).

Discussion

Seed size can be an important indicator of plant fitness with larger seeds producing more fit plants. As we did not find any differences between CH and CL seed sizes there may not be a significant difference in adult plant fitness levels. Other studies have found that in cleistogamous species, CH and CL individuals have similar seed sizes as well as fitness related traits. The similar fitness levels between CH and CL plants provide an explanation of the stability of mixed mating systems. We found that variation in *R. humilis* could be partially explained by maternal families. The significant differences suggest a genetic component to seed size, however differences could also be due to environmental differences. To determine the role of genetics in seed size, hand pollinations would need to be performed. To further this

study, a growth chamber experiment would be done in which the CH and CL seeds are planted and followed throughout their lifecycle to determine any differences in fitness related traits such as germination rates, leaf and flower production, and size of adult plants.

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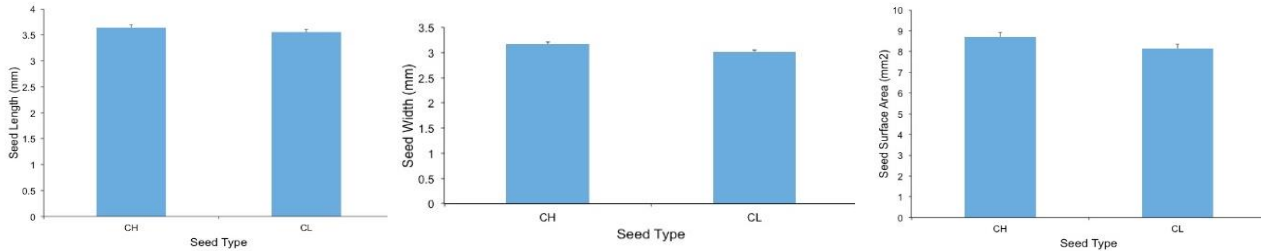


Figure 1 - Seed size differences between CH and CL seeds

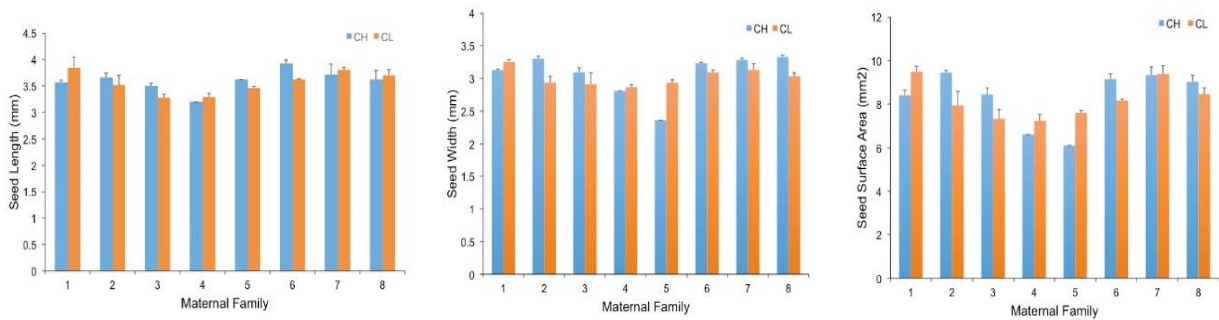


Figure 2 - Seed size differences in maternal families

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