THE EFFECT OF MAMMALIAN HERBIVORY ON INFLORESCENCE ARCHITECTURE IN ORNITHOPHILOUS BABIANA (IRIDACEAE): IMPLICATIONS FOR THE EVOLUTION OF A BIRD PERCH

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Pollinators are usually invoked as the primary selective agents responsible for most variation in floral design (Johnson, 2006; Kay and Sargent, 2009) and inflorescence display (Jordan and Harder, 2006). Convergent evolution of floral traits (Fenster et al., 2004) and studies of morphological trait matching (Anderson and Johnson, 2008; Anderson et al., 2010b), pollinator ecotypes (Johnson, 1997; Anderson et al., 2010a), natural selection (Herrera et al., 2006; Harder and Johnson, 2009) and the mapping of pollinator shifts on to phylogenies (Johnson et al., 1998; Beardsley et al., 2003) each provide evidence supporting pollinator-driven variation in floral characters and its adaptive basis. However, non-pollinating agents may also influence selection on floral traits and in such cases they can be either antagonistic or reinforce pollinator-driven selection (Strauss and Whittall, 2006; Rausher, 2008). Unfortunately, relatively little is known about biotic selection imposed by non-pollinating agents, and the extent to which they may have shaped floral and inflorescence morphology.

An important source of antagonistic selection on reproductive traits is herbivory by mammals or invertebrates. Herbivores can directly reduce the fitness of plants when they consume reproductive tissue, or indirectly by feeding on vegetative organs, thereby decreasing the amount of resources available for reproduction (Zamora et al., 1999). Herbivores can affect various traits involved with pollination, including phenology (Gómez, 1993; Strauss et al., 1996), floral display (Karban and Strauss, 1993; Gómez, 2003), floral morphology (Strauss et al., 1996; Leege and Wolfe, 2002; Strauss et al., 2004), nectar characteristics (Krupnick et al., 1999), and pollen quantity and performance (Strauss et al., 1996; Delph et al., 1997). Herbivory can also affect the evolution of floral traits when they are tightly linked with defense traits (Irwin and Adler, 2006). Although herbivores can reinforce selective pressures by pollinators, more often their effects appear to be antagonistic (Strauss and Whittall, 2006). For example, pollinators often select for large or showy displays (Peakall and Handel, 1993; Chittka and Thomson, 2001), but due to their visibility these can experience greater levels of herbivory than less showy displays (Gómez, 2003; Cariveau et al., 2004).

Here, we investigated the potential consequences of herbivory on the evolution of inflorescence architecture in bird-pollinated Babiana, a genus of geophytes largely endemic to the Cape region of southern Africa. Our studies focused on two closely related species that have similar red flowers pollinated primarily by sunbirds (Goldblatt and Manning, 2007a), but...
which differ in inflorescence architecture and floral display size. A major goal of our study was to understand the costs and benefits of their contrasting inflorescence designs in the context of pollination and herbivory.

*Babiana ringens* (L.) Ker Gawl. (Iridaceae: Crocoideae) occurs sporadically on nutrient-poor, sandy soils in the Western Cape province of South Africa, and its self-compatible flowers are visited almost exclusively by malachite sunbirds (Fig. 1A) (Anderson et al., 2005; De Waal et al., 2012). A specialized bird perch in this species facilitates foraging by sunbirds. The perch is a modified inflorescence axis in which the growth of apical side branches have been suppressed and flowers occur mostly at ground level. A single (infrequently two) sterile bract is evident at the top of the perch and occasionally the bud subtended by the bract may give rise to a single flower (Fig. 1C). The base of the perch supports one, or less often two horizontal branches, with 1–20 flowers per branch, which are probed by birds when they are in an upside-down position. Anderson et al. (2005) demonstrated that perch removal reduced the fertility and outcrossing rate of plants in comparison with unmanipulated plants. They proposed that the naked inflorescence axis in *B. ringens* functions to attract sunbirds and to position them more effectively for cross-pollination. Their findings raise the question of what selective forces may have been responsible for the origin of ground-level flowering and the specialized bird perch of *B. ringens*.

Recent phylogenetic and taxonomic evidence suggests that the sister species of *Babiana ringens* is *B. hirsuta* (Lam.) Goldblatt & J. C. Manning (Goldblatt and Manning, 2007b; Schnitzler et al., 2011; P. Goldblatt, Missouri Botanical Garden, personal communication). *Babiana hirsuta* is distributed along the West Coast of South Africa in sandy soils and is largely restricted to a narrow zone close to the high-water mark. It forms large populations consisting of many hundreds of plants and is self-incompatible and clonal with individual genets producing dozens of ramets (De Waal et al., 2012). *Babiana hirsuta* has branched inflorescences with large floral displays of 30–125 flowers per inflorescence (Fig. 1B). The plants are regularly visited by small, southern double-collared sunbirds that rob flowers of nectar but are ineffective pollinators, with legitimate pollination occurring through more infrequent visits by larger malachite sunbirds (Geerts and Pauw, 2009; De Waal et al., 2012). Plants are often strongly pollen-limited (De Waal, 2012). Sunbirds usually alight on the curved upper part of the inflorescence axis and proceed to move up and down the inflorescence using the numerous side branches as perches. We hypothesize that the specialized bird perch in *B. ringens* originated from a *B. hirsuta*-like ancestor through suppression of side branch development, leaving only one or two basal flowering side branches (Fig. 1C). This model of inflorescence evolution is supported by the presence of vestigial bracts on the apices of *B. ringens* perches, each of which subtends a sterile bud. The occasional failure to suppress the development of these buds leads to apical flowering in some *B. ringens* individuals (Fig. 1C), suggesting a homology to the apical flowering of *B. hirsuta*.

If this scenario is true, the transition from the *Babiana hirsuta*-like inflorescence architecture to that found in *B. ringens* would have required a large reduction in floral display size. Several potential selective forces could drive this transition including a decrease in display size to escape herbivores (Gómez, 2003), selection to reduce geitonogamous pollination and/or pollen discounting (Klinkhamer and de Jong, 1993; Harder and Barrett, 1995), or low soil nutrients favoring small plant size and hence reduced floral displays (Galen, 1999; Burkle and Irwin, 2009). However, other than herbivory, none of these factors can explain why a reduction in display size should be accompanied by the evolution of ground-level flowering, especially since tall displays have frequently been shown to be more attractive to pollinators (Cariveau et al., 2004; Anderson, 2010) leading to increased seed production (e.g., Ehrén et al., 2002). We hypothesize that ground-level flowering could evolve if mammalian herbivores preferentially forage on apical flowers and, as a result, basal flowers are less likely to be eaten by herbivores.

Based on these arguments, our study had the following three specific objectives: (1) to investigate whether the reduced floral display size of *B. ringens* is associated with reduced soil resources in terms of nitrogen and phosphorus; (2) to document the incidence and effects of mammalian herbivory on inflorescences in populations of *B. ringens* and *B. hirsuta*; and (3) to test the prediction that, in the presence of herbivory, *B. hirsuta* inflorescences manipulated to produce only basal flowers have higher seed production than those manipulated to produce only apical flowers. These objectives were accomplished through surveys of populations on the West Coast of the Western Cape Province of South Africa and through a field experiment in which we manipulated the presence and absence of herbivores and the architecture of inflorescences.

**MATERIALS AND METHODS**

**Study sites and surveys of soil**—Our study involved four *Babiana hirsuta* populations on sand dunes within 100 m of the sea and six *B. ringens* populations each located on sandy soils in fynbos, several kilometers inland (Table 1). To determine whether low soil nutrients are associated with the small floral displays of *B. ringens*, we collected soil samples from all of the *B. hirsuta* sites and four of the six *B. ringens* sites. We removed ca. 1 kg of soil from the root depth of *Babiana* at each site. The soil was then subsampled and percentage N calculated using an elemental analyzer (Eurovector EA 3000, Eurovector, Milan, Italy). Available P was calculated according to the methods of Bray and Kurtz (1945), by extracting 2 g of soil in Bray II solution. The extract was filtered through Whatman No. 2 filter paper and analyzed colorimetrically using the Malachite Green method (Motomizu et al., 1983). To investigate reproductive success, we calculated percentage fruit set per inflorescence from randomly sampled individuals in two *B. hirsuta* and six *B. ringens* populations as the number of fruits divided by the total number of flowers per inflorescence. Sample sizes for fruit set and the numbers of flowers per plant are provided in Table 1. These species differ significantly in the mean number of flowers per inflorescence (Table 1), and using percentage fruit set allowed us to explore limitations to fruit set and make inferences regarding the pollinator environment.

**Incidence and effects of herbivory**—We estimated the overall incidence of herbivory in three *B. hirsuta* populations. At Lamberts Bay and Rocher Pan, we randomly positioned a 100-m transect in each population and recorded whether flowering stems that touched the transect had been eaten or not. A total of 54 and 48 inflorescences were scored for evidence of herbivory at the Lamberts Bay and Rocher Pan sites, respectively. At Veldrif, we randomly located 110 plants damaged by herbivores and recorded whether apical or basal flowering branches were removed. We compared the height and number of side branches of inflorescences that were eaten by mammalian herbivores with those that were intact. In addition, we investigated the incidence of herbivory in *B. ringens* at Mamre, Hopefield, and Rondeberg. At each of these sites, plants were much less dense, and so we recorded the incidence of herbivory for every mature plant encountered, until we reached 50 plants each from Mamre and Hopefield and 30 from Rondeberg. Although we never directly observed grazing by antelope, the damage observed on plants probably resulted from the most common large browser in the area, the Cape grysbok (*Raphicerus melanotis*), which is frequently implicated in nocturnal plant herbivory (Skinner and Chimimba, 2005).
Fig. 1. Inflorescences of sunbird-pollinated Babiana ringens and B. hirsuta from the Western Cape Province of South Africa. (A) Inflorescence axis of B. ringens with ground-level flowers; apical flowers are suppressed, and only a sterile bract is evident (arrow). The axis is used as a perch by foraging malachite sunbirds. (B) B. hirsuta, which has multiple flowering side branches distributed along the entire inflorescence axis. (C) B. hirsuta (left) and three B. ringens plants exhibiting different degrees of apical floral suppression. The middle two B. ringens plants are rare forms that produce apical flowers, and the arrow indicates the sterile bract. (D) B. hirsuta with apical flowers removed experimentally. (E) B. hirsuta with basal flowers removed experimentally. (F) Inflorescence stalk of B. hirsuta after antelope grazing removed most of the apical portion of the inflorescence.
Reproductive success of apical vs. basal flowering inflorescences—To explore whether herbivory in populations of a B. hirsuta-like ancestor may have played a role in the evolution of ground-level flowering in B. ringens, we conducted a manipulative experiment in a B. hirsuta population at Velddrif, spanning an area of 500 × 50 m, by examining herbivory and seed production of plants manipulated to produce either apical or basal flowers. The primary objective of this experiment was to determine to what extent plants exposed to mammalian herbivores would experience position-dependent herbivory of flowers. We used mesh wire (65 mm aperture) and metal stakes to create fenced cages to exclude mammalian herbivores, and these were paired with equivalent uncaged treatments potentially exposed to herbivores. The cages were “open-top” so as not to deter pollinating sunbirds from visiting plants inside the cages, and the cages were on average ca. 1.75 m in diameter and 1.2 m tall. Thus, caged and uncaged treatments should receive similar pollinator service but differ in their exposure to mammalian herbivory. Our observations indicated that sunbirds accessed plants in cages by either entering through the holes in the wire or by simply flying over the sides of the fences.

We erected 30 cages in which each of three different manipulations (described below) were performed on 2–5 plants per manipulation (i.e., up to 15 experimental plants in each cage). Each plant in the 30 cages was paired with a nearby (<3 m) uncaged counterpart. Pairs of caged and uncaged treatments (blocks) were distributed across the site to control for spatial environmental variation. In total, there were ca. 100 inflorescences for each manipulation in each of the caged and uncaged treatments. The three experimental manipulations, set up in the uncaged and caged treatments before flowering commenced, were: (1) all basal (lower) side branches of inflorescences removed to leave single apical side branches (Fig. 1E); (2) all apical (upper) side branches of inflorescences removed to leave a single basal branch so that plants resembled the architecture of B. ringsens (Fig. 1D); and (3) unmanipulated inflorescences. A total of 600 inflorescences were included in the experiment (100 × 3 manipulations, in each of the caged and uncaged treatments). We tagged all plants by treatment and replicate code and, after 5–7 wk when fruits were ripe, the fruits were harvested, and total seed set per inflorescence was counted and averaged for each manipulation in each treatment–block combination. In the uncaged treatment, we scored whether the inflorescence stalks had been damaged by herbivores, and this was recorded, herbivores consistently consumed only the upper axis and side branches, but seldom the entire inflorescence (C. de Waal, personal observation, Fig. 1F). Where this preference was recorded, herbivores consistently consumed only the upper parts of the inflorescence, leaving the basal parts to continue flowering (Lamberts Bay: N = 4; Rocher Pan: N = 19). At Velddrif, 2.7% of plants damaged by herbivores had all flowering branches removed, whereas 97.3% had at least one basal flowering branch left intact (N = 110). In contrast, B. ringens plants exhibited much lower levels of herbivory with 14% at Mamre, 0% at Hopefield, and 17% at Rondeberg. When herbivores damaged B. ringens plants, they consistently removed the apical parts of the perch but not the ground-level flowers (Mamre: N = 7, Rondeberg: N = 5).

Surveys in two populations of B. hirsuta (Velddrif and Lamberts Bay) revealed variable effects of herbivory on the height of inflorescences (Fig. 2A). A general linear mixed model was

### Table 1. Number of flowers per inflorescence, percentage fruit set, and soil nutrient levels (N and P) in four Babiana hirsuta populations and six B. ringens populations from the Western Cape Province of South Africa. GPS locations of all sites are given.

<table>
<thead>
<tr>
<th>Site</th>
<th>Flowers per inflorescence ± SD (N)</th>
<th>% fruit set ± SD (N)</th>
<th>% N</th>
<th>P (mg/kg)</th>
<th>GPS South</th>
<th>GPS East</th>
</tr>
</thead>
<tbody>
<tr>
<td>B. hirsuta:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Lamberts Bay</td>
<td>87.57 ± 26.74 (34)</td>
<td>4.16 ± 4.74 (15)</td>
<td>0.03</td>
<td>116.0</td>
<td>32°07′50.3&quot;</td>
<td>18°18′20.3&quot;</td>
</tr>
<tr>
<td>Elands Bay</td>
<td>67.58 ± 19.63 (50)</td>
<td></td>
<td>0.04</td>
<td>87.0</td>
<td>32°19′06.5&quot;</td>
<td>19°18′27.0&quot;</td>
</tr>
<tr>
<td>Rocher Pan</td>
<td>—</td>
<td></td>
<td>0.03</td>
<td>74.0</td>
<td>32°36′64.7&quot;</td>
<td>18°15′57&quot;</td>
</tr>
<tr>
<td>Velddrif</td>
<td>54.11 ± 15.41 (45)</td>
<td>5.28 ± 9.48 (98)</td>
<td>0.02</td>
<td>44.4</td>
<td>32°45′03.5&quot;</td>
<td>18°10′14.8&quot;</td>
</tr>
<tr>
<td>B. ringens:</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Scarborough</td>
<td>5.86 ± 1.29 (35)</td>
<td>82.49 ± 16.83 (33)</td>
<td>0.03</td>
<td>3.0</td>
<td>34°11′35.7&quot;</td>
<td>18°23′00.6&quot;</td>
</tr>
<tr>
<td>Victoriasdal</td>
<td>5.84 ± 1.72 (50)</td>
<td>87.75 ± 15.37 (24)</td>
<td>0.03</td>
<td>9.7</td>
<td>34°20′18.4&quot;</td>
<td>21°14′13.0&quot;</td>
</tr>
<tr>
<td>Dekrietz</td>
<td>3.66 ± 0.63 (15)</td>
<td>94.26 ± 11.59 (19)</td>
<td>0.02</td>
<td>2.7</td>
<td>34°11′27.5&quot;</td>
<td>21°27′34.5&quot;</td>
</tr>
<tr>
<td>Hopefield</td>
<td>9.16 ± 3.32 (50)</td>
<td>77.25 ± 19.02 (14)</td>
<td>—</td>
<td>—</td>
<td>33°06′51.5&quot;</td>
<td>18°24′01.5&quot;</td>
</tr>
<tr>
<td>Rondeberg</td>
<td>9.25 ± 2.90 (30)</td>
<td>67.49 ± 26.12 (12)</td>
<td>0.02</td>
<td>2.3</td>
<td>33°24′56.5&quot;</td>
<td>18°18′14.0&quot;</td>
</tr>
<tr>
<td>Mamre</td>
<td>12.94 ± 3.41 (50)</td>
<td>77.95 ± 19.76 (29)</td>
<td>0.03</td>
<td>3.1</td>
<td>33°31′08.4&quot;</td>
<td>18°28′23.3&quot;</td>
</tr>
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RESULTS

Soil characteristics and fruit set comparisons—There were no significant differences in the nitrogen content of soils occupied by B. ringens and B. hirsuta (Mann–Whitney U test, Z = 0.612, P = 0.540). However, the coastal soils occupied by B. hirsuta had phosphorus levels that were an order of magnitude higher than at the inland B. ringens sites (MWU, Z = 2.327, P = 0.020, Table 1). There were striking differences between the two species in the reproductive success of populations. Babiana ringens populations were characterized by high percentage fruit set (77–94%), whereas fruit set in B. hirsuta populations was considerably lower (4–5%).

Evidence and effects of herbivory—Inflorescences of both Babiana species experienced herbivory, although levels of herbivory were much higher in B. hirsuta populations than in B. ringens populations. Fifty-three percent and 47% of B. hirsuta inflorescences from Lamberts Bay and Rocher Pan, respectively, were damaged by herbivores. Animals appeared to prefer the upper parts of the inflorescence, chewing off the upper axis and side branches, but seldom the entire inflorescence (C. de Waal, personal observation, Fig. 1F). Where this preference was recorded, herbivores consistently consumed only the upper parts of the inflorescence, leaving the basal parts to continue flowering (Lamberts Bay: N = 4; Rocher Pan: N = 19). At Velddrif, 2.7% of plants damaged by herbivores had all flowering branches removed, whereas 97.3% had at least one basal flowering branch left intact (N = 110). In contrast, B. ringens plants exhibited much lower levels of herbivory with 14% at Mamre, 0% at Hopefield, and 17% at Rondeberg. When herbivores damaged B. ringens plants, they consistently removed the apical parts of the perch but not the ground-level flowers (Mamre: N = 7, Rondeberg: N = 5).
In 20 of the 30 blocks, at least one manipulated inflorescence with only apical branches remaining (manipulation 1) had all of its flowers consumed. Thirty-four percent of inflorescences with this manipulation were affected in this manner. In 10 of 30 blocks, at least one unmanipulated inflorescence had its apical portions damaged by herbivory, and 10% of all unmanipulated plants in the uncaged treatment were affected in this way. In striking contrast, manipulated inflorescences in the uncaged treatment with only basal flow- ers (manipulation 2) experienced no herbivory (Fisher-exact test, \( P < 0.01 \)), with all basal flowering branches intact at the end of the experiment.

The treatment (caged or uncaged) for the whole model did not significantly affect seed set (\( F_{1,29} = 0.01, P = 0.910 \)), but both the inflorescence manipulation (\( F_{2,58} = 5.08, P = 0.009 \)) and the treatment by manipulation interaction (\( F_{2,58} = 3.57, P = 0.034 \)) accounted for significant variation in total seed set. In the uncaged treatment, manipulated inflorescences with only apical flowering side branches remaining (manipulation 1) experienced a significant reduction in total seed set per inflorescence (65% reduction, Fig. 3) compared to inflorescences of the same manipulation that were fenced to exclude herbivores (LSD, \( P < 0.05 \)). In contrast, inflorescences with basal flowers (manipulation 2) in the caged treatment set similar numbers of seeds to basal inflorescences in the uncaged treatment (LSD, \( P > 0.05 \)). In the uncaged treatment, inflorescences with only basal flowering branches intact set significantly more seed than inflorescences with only apical flowering branches remaining (LSD, \( P < 0.05 \)). However, total seed set did not differ significantly between unmanipulated inflorescences.

Effects of herbivory on the reproductive success of apical vs. basal flowers—Caged plants in our field experiment showed no signs of mammalian herbivory, indicating that the cages were effective in keeping herbivores out. Herbivores variably affected plants of the three different manipulations in the uncaged treatment (Pearson \( \chi^2 = 38.19, df = 2, P < 0.0001 \)). In 20 of the 30 blocks, at least one manipulated inflorescence with only apical branches remaining (manipulation 1) had all of its flowers consumed. Thirty-four percent of inflorescences with this manipulation were affected in this manner. In 10 of 30 blocks, at least one unmanipulated inflorescence had its apical portions damaged by herbivory, and 10% of all unmanipulated plants in the uncaged treatment were affected in this way. In striking contrast, manipulated inflorescences in the uncaged treatment with only basal flow- ers (manipulation 2) experienced no herbivory (Fisher-exact test, \( P < 0.01 \)), with all basal flowering branches intact at the end of the experiment.

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in uncaged and caged treatments (LSD, \( P > 0.05 \), Fig. 3). Data on fruit and seed set per flower from the field experiment are provided in Appendix 1.

**DISCUSSION**

The main findings of our study are: (1) Populations of *B. hirsuta* occur on phosphorus-rich soils, and natural fruit set is very low, whereas those of *B. ringens* occur on phosphorus-poor soils and have high levels of fruit set. (2) Populations of *B. hirsuta* experience considerably higher levels of mammalian grazing of inflorescences than *B. ringens*, resulting in significant losses of reproductive potential. (3) There are significant position-dependent effects of herbivory in populations of *B. hirsuta*, with plants manipulated to display basal flowers exhibiting higher seed set in the presence of herbivores than those manipulated to display apical flowers. Next, we discuss the ecological and evolutionary implications of our results and propose a model for the evolution of ground-level flowering and the specialized bird perch of *B. ringens*.

Our approach, involving a contrast between *B. hirsuta* and *B. ringens*, is based on the assumption that these two species are closely related and that the naked bird perch that characterizes *B. ringens* originated from a common ancestor with a branched inflorescence similar to *B. hirsuta*. Two lines of evidence support this hypothesis. First, a recent molecular phylogeny of *Babiana* (Schnitzler et al., 2011) indicates that the two species are closely related to one another, and the most recent taxonomic revision places the two as sister species (Goldblatt and Manning, 2007b; P. Goldblatt, Missouri Botanical Garden, personal communication). Second, in some *B. ringens* plants, vestigial flowers are produced at the apical region of the perch and produce fruit (Fig. 1C), indicating that the developmental regulation of functional apical flowers has not been completely lost. The occurrence of these apical flowers is consistent with the hypothesis that the ancestral inflorescence condition also involved apical flowering. On the basis of this hypothesis, we propose the following model for the evolution of the naked inflorescence axis of *B. ringens*.

The evolutionary transition in inflorescence architecture from a *B. hirsuta*-like ancestor to *B. ringens* can be divided into three “phases”, which may have taken place simultaneously or have occurred sequentially. (1) A reduction in floral display size. (2) The loss of apical flowers over basal flowers. (3) The maintenance of a sterile inflorescence axis. We begin by discussing potential factors that may have led to a reduction in display size. The number of flowers in inflorescences of *B. ringens* is much lower than in *B. hirsuta*, which has flowering side branches along most of the inflorescence axis (Fig. 1, Table 1). It seems unlikely that herbivory alone would have driven the reduction in display size that is now characteristic of *B. ringens*, because contemporary populations of *B. hirsuta* experience high levels of herbivory yet still produce large floral displays. Larger displays frequently attract more pollinators (Emms et al., 1997; Miyake and Sakai, 2005; Fenster et al., 2006) and may be maintained in *B. hirsuta* because of competition for pollinators in the coastal environments in which the species grows and in which reproductive success is chronically pollen limited (De Waal et al., 2012).

Resource constraints associated with growing on nutrient-poor inland soils (Witkowski and Mitchell, 1987) could have been a factor selecting for the relatively small floral displays characteristic of *B. ringens*, although of course other factors (e.g., reductions in geitonogamy, degree of pollinator service) may also have been involved. Available phosphorus is one of the most important growth-limiting elements in Cape ecosystems, especially for plants such as *Babiana*, which have no cluster root systems to enhance phosphorus acquisition (Lambers et al., 2006). Moreover, phosphorus and nitrogen are both essential elements in the development of flowers (Loehwing, 1940). The coastal habitats occupied by *B. hirsuta* experience a strong marine influence and elevated available phosphorus levels (Table 1; also see Witkowski and Mitchell, 1987), which may enable *B. hirsuta* to produce and maintain large displays. However, it is important to emphasize that this explanation for the evolution of reduced display size in *B. ringens* owing to resource constraints does not explain why a reduction in flower number is coupled with basal flowering, rather than a uniform change in flowers per branch across the entire inflorescence. Thus the coupling of reduced display size and basal flowering implicates additional selective pressures such as herbivory.

Whereas variation in display size can be explained by numerous factors, our study demonstrates that in *B. hirsuta* the position in which flowers are produced on an inflorescence (i.e., basal vs. apical) determines their likelihood of being eaten by grazing antelope. Furthermore, differences in inflorescence architecture affect fitness in terms of total seed set. For example, inflorescences with only apical flowers (Fig. 1E) experienced a 65% reduction in total seed set when exposed to herbivores. In contrast, manipulated *B. hirsuta* inflorescences with only basal flowers that most resembled *B. ringens* (Fig. 1D), were not consumed by herbivores, and there was no significant difference in seed set between the fenced and unfenced treatments. In the presence of herbivores, plants with basal flowering branches experienced higher reproductive success than plants with only apical flowering branches. These results clearly demonstrate that in *B. hirsuta*, basal flowers and fruits are less likely to be eaten by herbivores and that basal flowering can reduce the effects of herbivory. This finding may account for why herbivory levels in *B. ringens* were substantially lower than in *B. hirsuta*, although in the absence of observational data, we cannot rule out the possibility that this difference is associated with reduced herbivore pressure in *B. ringens* habitats.

We obtained results from our field experiment that were unexpected and therefore requires explanation. Unmanipulated plants produced similar amounts of seed to both the manipulated inflorescences in the caged treatment, and inflorescences with only basal flowers intact in the uncaged treatment (Fig. 3). In addition, there were no seed production differences between unmanipulated plants in the caged and uncaged treatments. Because no flowers were removed from the unmanipulated inflorescences, we would have expected greater total seed production than in these other two treatments. It is possible that sunbirds preferred manipulated inflorescences as they offered a more obvious perch on which to alight on arrival. This may have resulted in higher visitation rates, or the more accessible perch may have enabled more effective positioning for feeding thus increasing cross pollen deposition. Another possibility is that because of larger floral displays, unmanipulated inflorescences experienced more geitongamous pollen transfer possibly leading to less compatible fertilizations and/or higher abortion of fruits. Also, flower removal in manipulated inflorescences may have affected maternal investment to fruits and seeds causing the observed higher fruit and seed set per flower than in unmanipulated inflorescences (Appendix 1), thus counterbalancing their initially lower flower.
number. Finally, *B. hirsuta* individuals may produce “excess” flowers as a bet-hedging strategy either for years in which resources are more abundant or to compensate for losses due to herbivory. The latter may help to explain why unmanipulated plants and those with basal flowering branches had similar seed production. Compensation could also explain why seed production was similar between caged and uncaged unmanipulated plants. At this stage, we have no data to distinguish between these alternative hypotheses that might help to explain the lower than expected seed production of unmanipulated inflorences in our experiment. However, low fertilization and herbivory rates (10%) of unmanipulated plants probably contributed to the fact that no seed production differences were found between caged and uncaged inflorences of this treatment.

Where seed production is assured by reliable pollinator service but limited by resources, the best strategy for bird-pollinated *Babiana* may be to produce fewer flowers that are protected from herbivory by their basal position. During the course of evolution, basal flowering necessarily results in the suppression of apical branches and would have given rise to a naked inflorescence axis that served as a bird perch. This specialized structure is maintained in contemporary populations of *B. ringens* because it is an important adaptation for bird pollination, functioning to increase bird visitation and the quantity and quality of seed produced (Anderson et al., 2005). Moreover, as demonstrated by our manipulative experiment, the presence of a perch structure without apical flowers is also associated with the avoidance of herbivory. Although it seems unlikely that mammalian herbivory alone would have driven the evolution of the specialized bird perch that characterizes *B. ringens*, the results of this study suggest that it may have played a role.

LITERATURE CITED


**APPENDIX 1.** Proportion fruit set (±SE) and seed set per flower (±SE) of Babiana hirsuta inflorescences in a field experiment at Velddrif, Western Cape, South Africa. The inflorescence manipulations were (1) all lower side branches removed leaving one apical flowering branch; (2) all upper side branches removed leaving one basal flowering branch; (3) unmanipulated inflorescences. The inflorescence manipulations were compared between caged and uncaged treatments (see Methods for further details).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Proportion fruit set ± SE</th>
<th>Seed set per flower ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caged</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Manipulation 1</td>
<td>0.06 ± 0.01</td>
<td>2.28 ± 0.44</td>
</tr>
<tr>
<td>Manipulation 2</td>
<td>0.07 ± 0.01</td>
<td>2.29 ± 0.35</td>
</tr>
<tr>
<td>Unmanipulated</td>
<td>0.06 ± 0.01</td>
<td>2.03 ± 0.19</td>
</tr>
<tr>
<td>Uncaged</td>
<td></td>
<td></td>
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<tr>
<td>Manipulation 1</td>
<td>0.03 ± 0.01</td>
<td>1.01 ± 0.24</td>
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<tr>
<td>Manipulation 2</td>
<td>0.13 ± 0.02</td>
<td>2.58 ± 0.35</td>
</tr>
<tr>
<td>Unmanipulated</td>
<td>0.05 ± 0.01</td>
<td>2.26 ± 0.22</td>
</tr>
</tbody>
</table>