function of both loud and intermediate vocalization level flights is aggressive in nature.

We found no statistical difference between the loud and intermediate levels of flight vocalization in time until first feeding motion (Fig. 2). This indicates that we might have arbitrarily created the three levels of flight vocalization when Jacanas actually exhibit only two flight vocalization levels: e.g. flights with or without vocalization. If this were the case we would not have expected to see any differences in behavior between these two vocalization levels. However, wing-raising duration and occurrence of aggression were significantly different between loud flights and intermediate flights. We also observed a difference in the type of aggressive interaction occurring after the loud and intermediate vocalization level flights. After intermediate flights the interactions were usually comprised of landing, then chasing and attacking another Jacana on the ground, while those interactions occurring during the loud flights were most often diving air attacks. Because of these differences between intermediate flight vocalization level behavior and loud flight vocalization level behavior, we concluded that, within flights with vocalization, there are two separate levels of Jacana short-flight vocalizations.

Based on the observed differences in types of aggression occurring after loud and intermediate level flights, we believe that the intermediate level of flight vocalization may be used as a warning. We found aggression occurring after approximately half of the 30 intermediate flights sampled. This is the pattern we would expect to find if this level of call is in fact a warning. If the "warning" was heeded, no aggressive interaction would follow. As Jacanas' motives for heeding or not heeding a "warning" call are unknown, we would predict them to respond or not on a random basis. This would give us the approximately 50:50 split of aggression occurrences that we observed after intermediate level flights.

Ground vocalizations, in these same three levels of intensity, are also common. The functions of such loud and intermediate calls made from the ground should also be examined.

In our study we did not take separate data for males and females, because we were not able to sex adult Jacanas with confidence. We did observe both sexes of birds making all three levels of flight vocalization, however. An investigation into behavioral differences between males and females and between adults and juveniles could shed more light on the function of short-flight vocalizations.

In many species of birds, a primary function of vocalization is in anti-predatory behavior. However, because adult Jacanas do not exhibit anti-predator behaviors in the non-breeding season (Jenni and Betts, 1978), and because we observed no chicks or nests in the population during our study, we assumed that the observed levels of flight vocalization in our study were not related to such an anti-predator function.

Jacana short flight vocalization plays a significant part in aggressive interactions between conspecifics. Differentiated vocalization levels related to different types of behaviors should be useful and important for a species in which all adults attempt to hold overlapping territories for at least a portion of the year and where all adult females do not breed every breeding season, leading to intense intraspecific and intrasexual aggression.

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POLLINATOR ATTRACTANTS AND REWARDS IN THE PROTOGYNOUS FLOWERS OF *NYMPHAEA AMPLA*

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Abstract (SDK) Protogynous, hermaphroditic flowers of *Nymphaea ampla* offer differential pollinator rewards but similar visual cues in the pistillate and staminate stages. Both staminate and pistillate stages have a bright yellow coronal center, but only the staminate stage provides a pollen reward. By quantifying pollinator visits to flowers in the two stages, we found that the total duration of pollinator visits was greater in staminate stages, as predicted. Additionally, we found no significant difference between the number of pollinators visiting the staminate and pistillate stages, confirming that pollinators are unable to visually distinguish between the two. Our results indicate that *N. ampla*'s breeding system acts to maximize male and female reproductive success.

INTRODUCTION (NDP)

The water lily, *Nymphaea ampla*, is a protogynous, hermaphroditic aquatic herb. Flowers commonly persist for three to four days (Ratsirarson 1990). The pistillate stage, representing the female function, lasts for the first day of flowering and is characterized by a bright yellow receptacle, surrounded by three to four whorls of upright stamens. The receptacle contains a pool of exudate which has a sugar concentration of only 3-4% (Ratsirarson 1990). Because most pollinators require nectar with a sugar concentration of at least 15% (Faegri and van der Pijl 1966, cited in Ratsirarson 1990), Ratsirarson (1990) suggests that it is actually the pollen in *N. ampla* that provides the pollinator rewards on days two through four. Pollen is available only in the staminate stages when the male function of a *N. ampla* flower is mature. The staminate stage is characterized by a collapse of the inner whorls of stamens over the now exudate-free center. Pollen on coronal stamens gives the collapsed corona a bright yellow color similar to that of the pistillate receptacle.

Principal pollinators of *N. ampla* in the Río Tempisque marshland are *Apis mellifera*, the common honeybee, and *Brachygastra* spp. wasps (Alonso-Mejia, 1990). Both species are pollen gatherers that use visual cues. It appears that only the staminate stage offers pollinator rewards in the form of pollen. The pistillate stage of *N. ampla* relies on similarity in color to the staminate stage to attract pollinators, but offers no apparent rewards.

Because pollinators that rely on visual color cues will be unable to distinguish between the two *N. ampla* stages, we hypothesized that the rate of pollinator visitation to both pistillate and staminate stages will be the same. Further, if pollinators forage optimally, we proposed that they should spend more time gathering rewards from staminate-stage flowers than from pistillate-stage flowers. Thus, we predicted that the total duration of pollinator visits should be greater in the staminate-stage than in the pistillate-stage flowers.

METHODS (SDK)

We searched for flowers of *Nymphaea ampla* on an 82 x 20 m belt transect across the Río Tempisque marsh, Palo Verde National Wildlife Refuge, Costa Rica. The transect was located near the airstrip on the southwest side of the barbed wire fence.

The sex ratio of *N. ampla* is biased towards the male flowers, as the staminate

stage persists for two to three days compared to one day for the pistillate stage (Ratsirarson 1990). Because pistillate *N. ampla* flowers were relatively uncommon, we first located and tagged 15 individual pistillate flowers and then paired each of these with the nearest staminate flower. Neighboring staminate flowers were within 1 m of the focal pistillate flower in all sample pairs but one which was 3 m apart. We identified the pistillate stage by the presence of a pool of exudate in the center of the open (but immature) staminate corona. We identified the staminate stage by the presence of a closed (but mature) staminate corona. Flower pairs were tagged at 0900 when all *N. ampla* flowers in the study site had opened.

We first observed the pistillate-stage flower for 10 minutes and then the paired staminate-stage flower for the next 10 minutes. We recorded the number of pollinator visits, species of pollinator, and the duration of each pollinator visit. Observations began at 0930 and continued through 1145 on 12 January 1995.

RESULTS (SDK, NDP)

We found no significant difference in the rate of visitation that pollinators, (*A. mellifera* and *Brachygastra* spp. wasps) made to pistillate and staminate stage *Nymphaea* flowers (all pollinators: t= -0.782, df= 14, P= 0.45, bees: t= -1.468, df= 14, P= 0.16 wasps: t= 1.046, df= 14, P= 0.31; Fig. 1).

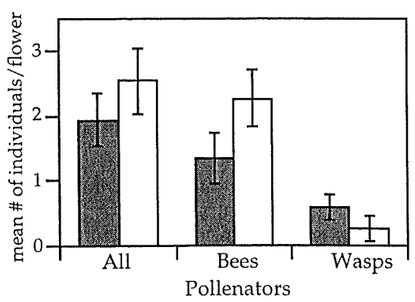

FIG. 1. Mean number of pollinator (± SE) visits per flower to pistillate- (grey bars, n=15) and staminate- (white bars, n=15) stage flowers of *Nymphaea ampla* within 10 minutes. Means were not significantly different for any type of pollinator visit.

The total duration of visits by all pollinators (bees and wasps combined) was not statistically different for the pistillate and staminate stages (t= -1.528, df= 14, P= 0.15; Fig. 2) nor did the total duration of visits by *Brachygastra* spp. wasps differ significantly between the two stages (t= 0.871, df= 14, P= 0.40; Fig. 2). However, the total duration of *A. mellifera* visits was significantly higher in staminate- versus pistillate-stage *Nymphaea* flowers (t= -2.320, df= 14, P= 0.036, Fig. 2).

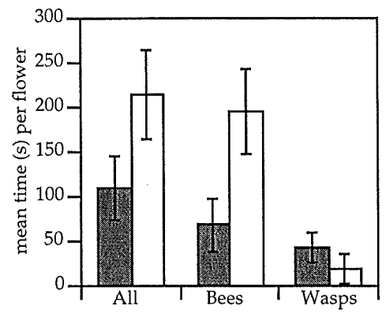

FIG. 2. Mean total duration of pollinator visits (± SE) per flower to pistillate- (grey bars, n=15) and staminate- (white bars, n=15) stage flowers of *Nymphaea ampla* during 10 minute observation periods. Total duration of bee pollinator visits was significantly different for pistillate and staminate stages.

DISCUSSION (NEP)

The data support our first hypothesis: total number of pollinator visits did not differ between pistillate and staminate stages of *N. ampla*. Foragers are evidently unable to distinguish between flowers which offer a reward and flowers which do not. We suggest that pollinators are equally attracted to the similar, bright coloration of flowers of both stages.

The total number of wasp visits was low (pistillate flowers: 9; staminate flowers: 4) and we found no statistical difference in the duration of wasp visits between the two stages of flowers. The low number of wasp visits may have contributed to the lack of

significant difference in duration of total pollinator visits. Total duration of bee pollinator visits, however, was significantly greater in the staminate stage than in pistillate stage. Although both flower stages attract pollinators, only the staminate stage provides a reward. Bees foraging in the pistillate-stage flowers find no such reward and leave in search of another flower (Fig. 2).

The staminate stage relies on its pollen reward to sustain bee foraging and maximize the success of the *N. ampla* reproductive function. More specifically, a pollen reward stimulates lengthy bee foraging and increases the potential for *N. ampla* pollen dispersal. In its pistillate stage, *N. ampla* invests little sugar in its nectar exudate and relies instead on its similarity in color to the staminate-stage flower to satisfy its female reproductive function. We speculate that lengthy pollinator visits are not essential to female reproductive success. Rather, brief pollinator visits are probably sufficient to introduce pollen to the receptacle. The reproductive strategy of *N. ampla* appears to maximize the success of both the male and female function while minimizing investment in pollinator rewards.

Presumably *N. ampla* is strictly protogynous, that is, it exhibits distinct female and male functions. It is possible, however, that the closed staminate corona dehisces pollen onto the stigmatic receptacle of the same flower. Thus, future study should examine the potential for self- fertilization in staminate-stage flowers. This study should also include an examination of the function of *N. ampla*'s nectar exudate. Though the exact function of the nectar secretion is not known, the exudate may facilitate collection and germination of the pollen grains (Ratsirarson 1990). The absence of exudate in the staminate stage of the flower might preclude pollen grain germination and act to limit self-fertilization. If self-fertilization does occur in *N. ampla*, it would be interesting to compare the fertilization success of selfed and outcrossed lilies as well as the germination success of selfed and outcrossed seeds.

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