

Patterns of pollen removal and deposition in tristyous *Pontederia cordata* L. (Pontederiaceae)

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In tristylous *Pontederia cordata* (Pontederiaceae), conspicuous differences in the size of pollen grains and discrete variation in the length of reproductive organs provide a suitable experimental system for the study of fine-scale pollination events. At a population of *P. cordata* at Pothole Lake, Ontario, the majority of flowers are visited by bumble bees which remove on average 45% of the pollen during single visits to previously unvisited flowers. The amount and proportion of pollen removed are significantly different among floral morphs and stamen levels. Deposition of the three pollen types on the bodies of *Bombus* spp., *Apis mellifera* and *Melissodes apicata* is non-random: large- and medium-size pollen tends to remain in greatest concentrations where it is initially deposited, whereas small-size pollen is displaced from the proboscis to more posterior body parts, probably as a result of grooming activities. Stigmatic pollen loads of individual flowers following single bumble bee visits indicate that the mid-styled morph captures the largest total pollen load, and the short-styled morph the smallest. The largest proportion of compatible pollen grains is deposited on stigmas of the long-styled morph. Pollen load data from "single visit" flowers is in general agreement with previously published population surveys involving multiply-visited flowers.

KEY WORDS: Pollination – tristily – *Pontederia cordata* – *Bombus* – flowering – pollen – deposition.

CONTENTS

Introduction	317
Natural history of <i>Pontederia cordata</i>	318
Materials and methods	319
Study site	319
Pollen removal	319
Pollen partitioning on pollinators	320
Pollen deposition on stigmas	320
Results	321
Pollen removal from flowers	321
Pollen partitioning on pollinators	321
Pollen deposition on stigmas	322
Discussion	324
Acknowledgements	328
References	328

INTRODUCTION

Tristyly is a genetic polymorphism in which populations are composed of three floral morphs that differ in a suite of floral characters including style length,

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anther height, pollen size and self-incompatibility. Using experimental pollinations, Darwin (1877) elucidated the breeding relationships of tristylous plants and proposed the first hypothesis to account for the function of the floral heteromorphism. He suggested that the reciprocal arrangement of sex organs promotes insect-mediated cross-pollination between anthers and stigmas of equivalent height and thus disassortative pollination among floral morphs. The hypothesis is contingent on deposition of pollen from the three anther levels on different locations of a pollinator's body. According to the hypothesis, the body parts will subsequently contact stigmas of different morphs and deposit compatible pollen. The hypothesis, therefore, has two components: (1) the three pollen types are partitioned on the bodies of pollinators, and (2) the pollens are then differentially deposited on the three stigma types. Studies of tristylous species in *Lythrum* (Ornduff, 1975; Mulcahy & Caporello, 1970) and *Pontederia* (Price & Barrett, 1984; Glover & Barrett, 1983, 1986; Barrett & Glover, 1985) have been designed to evaluate whether in natural populations the floral polymorphism influences the composition of stigmatic pollen loads in the manner predicted by Darwin. Because of the emphasis on pollen deposition patterns, pollen partitioning on pollinators and other aspects of the pollination process of tristylous species are largely unstudied.

Here we present detailed information on the dynamics of the pollination process in tristylous *Pontederia cordata* L. (Pontederiaceae). The species is particularly useful for this type of study since it possesses conspicuous size differences among pollen grains produced at different anther levels. This allows the investigator to distinguish compatible from incompatible pollen and to measure fine-scale pollination events (Barrett & Wolfe, 1986). The discrete variation in reproductive organ length that exists within and between flowers also enables the influence of variable floral morphology on pollination to be examined. The major objectives of our study are: (1) to quantify the schedule and amount of pollen removed from anthers by pollinators in the floral morphs of *P. cordata*; (2) to test Darwin's hypothesis that the reciprocal arrangement of sex organs within tristylous flowers results in differential placement of pollen from the three anther levels on the bodies of pollinators; and (3) to quantify the size and composition of pollen loads deposited on stigmas following bumble bee visits.

NATURAL HISTORY OF *PONTEDERIA CORDATA*

Pontederia cordata L. (Pontederiaceae) is a long-lived, emergent aquatic native to eastern North America and Central and South America (Lowden, 1973). Populations inhabit a wide range of wetland habitats and vary in size from isolated colonies to dense monospecific stands containing thousands of inflorescences. Populations usually contain all three floral morphs (hereafter referred to as the L, M and S morph), and in many populations the S morph is over-represented in comparison with isoplethic expectations (Barrett, Price & Shore, 1983; Morgan & Barrett, 1988). Inflorescences bear either long-, mid- or short-styled flowers that are purple-blue, zygomorphic, uniovulate and produce concentrated nectar (55% sucrose equivalents) during the 6–8 h anthesis period (Wolfe & Barrett, 1987). Many species of insects visit flowers for nectar and/or pollen; bumble bees are the most abundant pollinators in North America (Price & Barrett, 1984; Wolfe & Barrett, 1988). In Ontario, where the study was

conducted, flowering commences in early July and lasts approximately two months with inflorescences producing about 200 flowers, with an average of 20 open per day during peak blooming (Wolfe & Barrett, 1987). Further details of the floral biology and tristylous syndrome of *P. cordata* are available in Price & Barrett (1982, 1984).

MATERIALS AND METHODS

Study site

All work was conducted at Pothole Lake, Leeds and Grenville County, Ontario. The lake is approximately 800 m long and varies from 50 to 300 m in width. *Pontederia cordata* is the most abundant plant in the littoral zone and grows along the shoreline and in shallow areas in the middle of the lake. The Pothole Lake population contains the three floral morphs, and at peak flowering in 1982 the morph ratio was: L=0.31, M=0.27 and S=0.42 ($N=332$ inflorescences). Using this ratio and taking into account the number of pollen grains produced at each anther level, the frequency of pollen types (population pollen pool) was large-sized pollen (hereafter l)=0.12, mid-sized pollen (hereafter m)=0.18 and small-sized pollen (hereafter s)=0.70 (see Price & Barrett, 1984 for method of determining pollen production). Further information concerning the flowering phenology, pollinator fauna and foraging behaviour of pollinators at the population are presented in Wolfe & Barrett (1987, 1988).

Pollen removal

The number of pollen grains removed from previously unvisited flowers of *P. cordata* by a single bumble bee was examined between 10.00 and 12.00 hours on August 4 and 5, 1983. Naturally foraging bumble bees (*Bombus griseocollis*) were followed during short visits to previously bagged inflorescences. For each visitation run, the bag was removed and the observer stood by the inflorescence until a bee arrived. After a bee had probed five flowers, its visit was terminated by gently knocking it away from the inflorescence. All anthers from five flowers were then removed with forceps and stored in vials containing 70% ethanol. This was repeated for four inflorescences per morph, for a sample of 20 flowers per morph. Long- and mid-level anthers were removed with forceps. Short-level anthers were difficult to remove, so anthers and part of the perianth tube were excised together. To determine the number of pollen grains in unvisited flowers, undehisced anthers from 10 flowers (2 flowers \times 5 inflorescences) were collected and stored in 70% ethanol. Each sample of anthers was subjected to modified acetolysis. The procedure digests most plant material (anther sac, perianth tube) but leaves the pollen grain wall intact. To count pollen grains, samples of acetolyzate were placed on a haemocytometer and counted under a compound microscope (see Price & Barrett, 1984 for method). Data were analysed using goodness-of-fit tests to assess the hypothesis that pollen removal is independent of morph or anther level.

The schedule of pollen removal from flowers through the day was examined in August 2, 1983. Prior to flower opening, five inflorescences of each morph with at least 25 flowers were tagged. Each hour from 08.00 to 15.00 hours, two flowers

were removed from each inflorescence and placed in 70% ethanol. Prior to placing the flowers in vials, stigmas were removed so that only pollen from the anthers would be examined. Following acetolysis, pollen grains were counted under a compound microscope. Data were analysed using goodness-of-fit tests to assess the hypothesis that the schedule of removal does not differ among morphs or anther levels.

Pollen partitioning on pollinators

To determine if pollen partitioning is evident on naturally foraging pollinators, three groups of bees were studied: bumble bees (*Bombus* spp.), honeybees (*Apis mellifera*) and the solitary bee *Melissodes apicata*. In early July 1983, 15 individuals of each group were captured while foraging on *P. cordata* in small jars containing ethyl acetate. Pollen was sampled from the ventral surface of three regions of each bee: mid-abdomen (hereafter called abdomen), base of proboscis (hereafter called head) and tip of proboscis. Pollen was removed by placing a cube of fuchsin-glycerine jelly (2 × 2 × 1 mm; Beattie, 1971) onto a pin and pressing this onto the desired body part. The cube was then melted on a microscope slide and the number of pollen grains counted under a compound microscope. Since each body part was subsampled, pollen counts do not represent the total borne on that part of the body. Data were analysed using goodness-of-fit tests to determine if deposition of pollen type was independent of body part.

Pollen deposition on stigmas

Pollen deposition was studied by determining the number of compatible and incompatible pollen grains deposited on stigmas of each morph from a single pollinator visit. Data were collected in a manner that also allowed the relationship between a flower's position in a visitation sequence and the number of compatible pollen grains received to be evaluated (pollen carryover). Naturally foraging bumble bees were used as they were the most common pollinator. Data were collected between 09.00 and 13.00 hours for approximately 25 days during July and August 1982.

TABLE 1. The number of pollen grains removed from flowers of *Pontederia cordata* following single visits by bumble bees (*Bombus griseocollis*). Values are based on 20 flowers pooled and represent the mean of four haemocytometer counts

Anther level/ floral morph	Number of pollen grains in unvisited flowers	Number of pollen grains removed from one visit	Percent of pollen grains removed from one visit
l/S	4000	2934	73
l/M	2977	1911	64
m/S	6044	2311	39
m/L	3822	2344	61
s/M	27600	12000	43
s/L	23555	7622	32

G-tests assess the hypothesis that the pattern of pollen removal is independent of anther level. The expected values are the number of pollen grains in unvisited flowers. 1. $G = 44.43$ (df = 2; $P < 0.001$) for between anther level heterogeneity (l/S + l/M vs. m/S + m/L vs. s/M + s/L). 2. *G* values for heterogeneity within anther levels (i.e., m/S vs. m/L): l = 0.726 (d.f. = 1; NS), m = 10.55 (d.f. = 1; $P < 0.0001$), s = 18.08 (d.f. = 1; $P < 0.0001$).

Inflorescences were bagged prior to flower opening to ensure that all pollen on stigmas was from the experimental visitation run. Before each visitation run inflorescences were trimmed to 10 flowers to facilitate recording the sequence of flowers visited by bees. While a bee foraged the visitation sequence was recorded with a tape recorder. Bees tended to visit less than 10 flowers per inflorescence, resulting in a preponderance of short runs for pollen carryover analysis.

Pollen grains were removed from the stigma by excising the pistil from the flower and pressing the stigma into small pieces of fuchsin-glycerin jelly which had been melted into place on microscope slides. Virtually all pollen grains are removed from the stigma if this technique is applied less than 30 min after pollen deposition. Later, the number and type of pollen (l, m, s) was recorded for each flower, using a compound microscope.

RESULTS

Pollen removal from flowers

A single bumble bee visit removes an average of 45% of the pollen grains from a previously unvisited flower of *P. cordata* (Table 1). The largest number of pollen grains are removed from short-level anthers while similar numbers are removed from long- and mid-level anthers. Percent removal varied with stamen height and was greatest in long-level anthers (68.5%), followed by mid-level anthers (50%) and short-level anthers (37.5%) (Table 1). Significant variation in removal also exists between each of the two corresponding anther levels in different morphs. Differences were most pronounced in the two mid-level anther sets (Table 1). Although a similar number of pollen grains were removed from mid-level anthers of the L and S morphs, this represents a smaller fraction of the pollen in the S morph.

As a result of the large number of pollen grains removed by a single bee visit and high pollinator densities at the site (see Wolfe & Barrett, 1988), pollen is rapidly depleted from flowers (Fig. 1). On August 2, each anther level contained less than 40% of its original pollen complement 90 min after dehiscence. The amount of pollen remaining in flowers continued to decrease throughout the day, but at a reduced rate. There was a significant difference in the pattern of pollen removal both among morphs ($G=169.65$, d.f. = 14,35, $P<0.001$) and anther levels ($G=90.06$, d.f. = 14,35, $P<0.001$). Despite the appearance of being "empty", anthers still contained some pollen when flowers began to wither at 15.00 hours. The proportion of pollen left in flowers was greatest in the M morph at all seven sampling intervals. Similarly, short-level anthers always had a greater fraction of their pollen complement remaining in flowers than did long- and mid-level anthers.

Pollen partitioning on pollinators

Pollen from the three anther levels is initially placed on different regions of the ventral surface of a pollinator's body during entry into a flower. The distribution of pollen on naturally foraging pollinators indicates how well this stratification is maintained. In general, the pollen types were distributed in a similar manner on the three bee groups that were examined (Table 2). In each group, χ^2 analysis

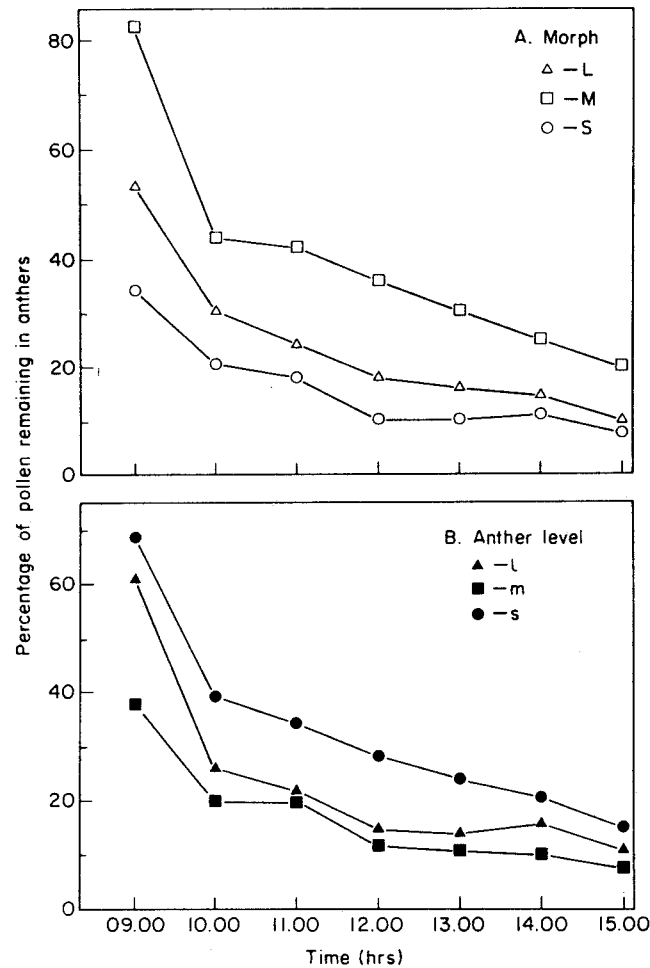


Figure 1. Removal of pollen from flowers of *Pontederia cordata*. Values represent the percent of the original pollen complement remaining in anthers at intervals through the day. Pollen removal from the floral morphs (A) and anther levels (B) with time is significantly heterogeneous $G=201.25$, $P<0.001$ and $G=322.75$, $P<0.001$, respectively.

indicated that pollen types were not located at random on the body. Pollen from long-level anthers (l) was most abundant on the abdomen, m pollen was most common on the head, and the location of s pollen varied among species. In honey bees and bumble bees s pollen was most common on the abdomen, while in *Melissodes* it was more common on the head. Thus l and m pollen tends to remain on the body parts on which they are initially deposited (abdomen and head, respectively). In contrast, s pollen is displaced from the proboscis to posterior body parts. The number of pollen grains, irrespective of type, was considerably smaller on the proboscis than on the abdomen or head (Table 2).

Pollen deposition on stigmas

Pollen deposition on stigmas of *P. cordata* was investigated by examining the size and composition of pollen loads resulting from one bumble bee visit. Total pollen loads were greatest in the M morph, least in the S morph, and intermediate in the

TABLE 2. The partitioning of different size pollen grains of *Pontederia cordata* on the bodies of three pollinator groups. Values are the mean number of pollen grains originating from different anther levels on each of three body parts. $N = 15$ Bees per species. χ^2 Analysis performed on pollen counts totalled over 15 bees tests the hypothesis that deposition of pollen type is independent of body part

Pollen type	Body part		
	Abdomen	Head	Proboscis
A. <i>Apis mellifera</i>			
l	35.1	12.1	1.9
m	9.1	19.5	1.5
s	8.2	3.5	2.3
	$\chi^2 = 247.12$; d.f. = 4; $P < 0.001$		
B. <i>Bombus</i> spp.			
l	49.4	15.6	1.0
m	10.1	16.3	2.1
s	65.1	7.9	1.5
	$\chi^2 = 411.8$; d.f. = 4; $P < 0.001$		
C. <i>Melissodes apicata</i>			
l	28.0	2.2	0.2
m	19.8	21.7	1.3
s	16.5	27.0	8.5
	$\chi^2 = 220.48$; d.f. = 4; $P < 0.001$		

L morph, (Table 3). Stigmas in each of the three morphs received a substantial number of incompatible pollen grains with over 75% of the stigmatic pollen loads of flowers of the M and S morphs composed of incompatible pollen. The capture of compatible pollen varied among the morphs with flowers of the S morph receiving the smallest pollen load and the L morph the largest. Many pollinator visits failed to deposit compatible pollen. This effect was most pronounced in flowers of the S morph. Approximately 40% of flower visits to the S morph resulted in no deposition compared to 26% and 21% in the L and M morphs, respectively (Fig. 2).

Examination of pollen carryover indicated that the proportion of compatible pollen deposited during an inflorescence visit is generally highest in the first flowers visited (Fig. 3). However, a characteristic feature of the data is the high degree of variation in pollen grain deposition within and between visitation runs. The variation prevents any generalizations from being made concerning

TABLE 3. Deposition of pollen grains on stigmas of *Pontederia cordata* following a single bumble bee visit. Flowers were sampled from 15 July-5 August 1982 at Pothole Lake. Values are mean \pm s.d. N is number of flowers. Compatible pollen loads are in italics

Floral morph	N	Pollen type			Total
		l	m	s	
L	60	22.8 \pm 33.8	10.3 \pm 22.8	5.4 \pm 8.0	37.9 \pm 44.6
M	85	8.1 \pm 24.7	<i>15.1 \pm 24.9</i>	45.1 \pm 85.8	68.3 \pm 96.6
S	58	4.5 \pm 15.3	9.2 \pm 20.1	<i>4.6 \pm 10.8</i>	18.5 \pm 27.0

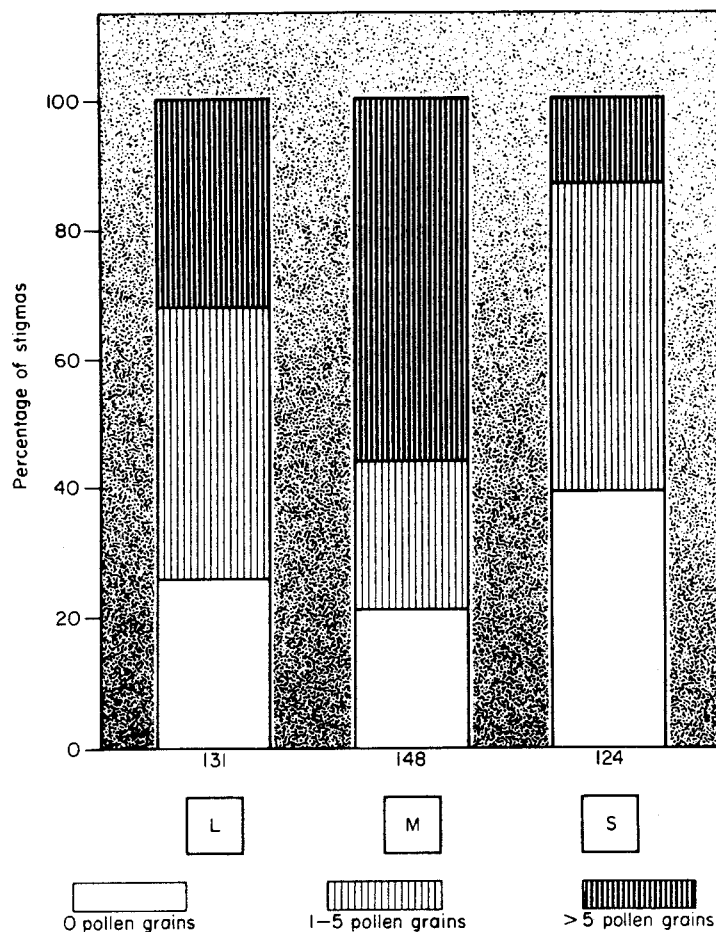


Figure 2. The fraction of *Pontederia cordata* stigmas that received different amounts of compatible pollen from a single bumble bee visit. Sample size (no. of flowers) is given below bars. Empty space = no pollen deposited; bottom diagonal hatch = 1-5 pollen grains; top diagonal hatch = > 5 pollen grains.

differences in the patterns of pollen carryover among the floral morphs or pollen types. An example of the high degree of variation within a single run can be seen by considering the number of compatible pollen grains deposited by a bumble bee on nine successively visited flowers of a single mid-styled inflorescence. These were: 20, 10, 2, 12, 0, 42, 0, 5 and 30. This variation in the number of compatible pollen grains deposited on consecutively visited flowers was typical of most of the 66 runs examined. Figure 3 illustrates the average percent of the pollen load composed of compatible pollen that was deposited on successively visited flowers in each of the floral morphs. The flatness of the curves, particularly after the first flower had been visited, implies that the residency time of compatible pollen on a pollinator's body is sufficient to ensure transport to additional inflorescences.

DISCUSSION

Darwin (1877) hypothesized that the function of the stamen-style polymorphism in heterostylous plants is to promote intermorph pollen transfer

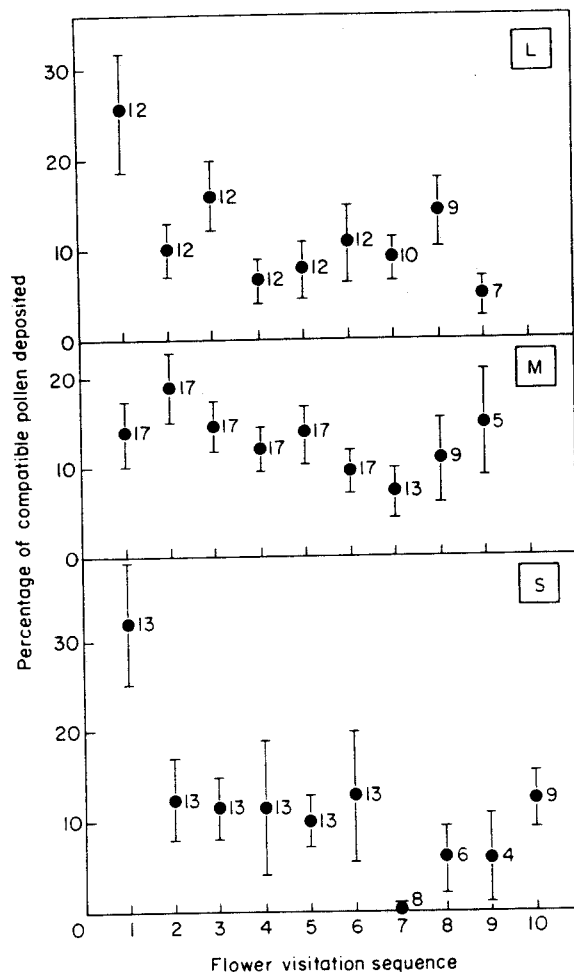


Figure 3. Deposition of compatible pollen in relation to a flower's position in a visitation sequence within a single inflorescence. Values are mean (\pm S.E.) percent of compatible pollen grains deposited on flowers and were obtained by averaging values for all visitation runs conducted for each morph. Sample sizes (number of flowers) are given.

between anthers and stigmas of similar height. This will occur if pollen from the three anther levels is differentially placed on the bodies of pollinators and subsequently deposited on the appropriate height stigmas. The idea has received only limited support from work on distylous species (Darwin, 1877; Ford, 1964; Rosov & Serebtsova, 1958; Oleson, 1979) but had not been previously studied in tristylous species. Data from this study indicates that pollen adhering to bees foraging on *P. cordata* is not distributed at random. In the three bee groups examined, l and m pollen was most common on the body region on which it was initially deposited, whereas s pollen was not. Since all body parts have pollen in frequencies that deviate from random, it is clear that some degree of partitioning is maintained.

Several factors influence the distribution and composition of pollen on a pollinator's body. Darwin (1877) noted that *P. cordata* is one of the few heterostylous species with zygomorphic (bilaterally symmetrical) flowers. In contrast to flowers with radial symmetry, zygomorphic flowers offer a limited

number of orientations that a pollinator may adopt during entry and feeding (Faegri & van Pijl, 1979). Anther level positions within tristylous flowers result in pollen deposition on specific locations of the pollinator. For example, short-level anthers can only contact the proboscis. However, the proboscis may also touch long- and mid-level anthers while entering the perianth tube. This may account for the occurrence of all pollen types on the proboscis whereas the abdomen appears to pick up primarily l pollen.

Once pollen adheres to a pollinator's body, other factors disturb the original placement pattern. Bees regularly groom from their bodies into pollen baskets on their hind legs (Heinrich 1979). Grooming will displace pollen and may account for the different amounts of pollen found among body parts. Cleaning behaviour usually involves movement of pollen from the anterior to the posterior end of a bee's body. Since bees regularly clean their proboscis and head during nectar feeding, this may explain why the proboscis bore few pollen grains in the three bee species examined. In contrast, l pollen, which is initially deposited near the hind legs, may not experience such a high level of displacement.

The amount of pollen that adheres to a body part is also affected by its surface area (Cruden & Miller-Ward, 1981). The proboscis, which has a relatively small area, may thus be expected to bear a smaller number of pollen grains than the larger head and abdomen. The distribution of pollen on a pollinator's body is likely to be dynamic and will undergo modification as a result of foraging behaviour. However, placement of pollen on body parts that are difficult for the bee to groom will aid in maintaining pollen partitioning. In *P. cordata* this appears to involve parts of the head and body region where l and m pollen are preferentially deposited. Despite differences in the size, morphology and foraging behaviour (Wolfe & Barrett, 1987) in all of the three main bee groups that visit *P. cordata* at Pothole Lake some degree of partitioning is maintained during feeding. This indicates that close coadaptation between *Pontederia* and its pollinators is not a prerequisite for pollen stratification and compatible pollen transfer to occur.

The spatial distribution of pollen types on pollinators is helpful in explaining the observed patterns of stigmatic deposition. First, as mentioned earlier, the body part that deposits compatible pollen is different for the three morphs (abdomen—L morph; head—M morph; proboscis—S morph). The largest number of compatible pollen grains is borne on the abdomen and the least on the proboscis. This pattern may cause the level of compatible pollination to be highest in the L morph, lowest in the S morph and intermediate in the M morph as observed in our study. Second, some flower visits by bees do not deposit compatible pollen. The probability of an unsuccessful visit was most pronounced in the S morph (39%). This is most likely due to the high incidence of bees with few, if any, compatible pollen grains on their proboscis.

In a study that documented the foraging behaviour of pollinators of *P. cordata*, Wolfe & Barrett (1987) found that honey bees preferentially visited flowers of the S and M morphs to collect pollen from long-level anthers. As a result of preferential collection, the corbicular pollen loads of honey bees were found to be mainly composed of l pollen. Not surprisingly, the most common pollen type on bodies of honey bees was l pollen despite the relatively low frequency of this pollen in the population. A second association was found in *Melissodes*. In this bee, s pollen was most common on the body and in pollen baskets. Although the

species does not appear to forage in a non-random fashion with respect to morph, the preponderance of s pollen appears to be associated with the presence of specialized hairs on the proboscis. Laberge (1956) has suggested that the function of these hairs in *Melissodes*, a specialist of *Pontederia* spp, is to remove pollen from the concealed short-level anthers of tristylous flowers in the genus.

Despite the increased awareness of the importance of male function to plant fitness, few studies have examined the process of pollen removal in species with free pollen. Webb & Bawa (1983) studied pollen removal in *Malva viscus arborius*, a tropical shrub, and found that most occurred in the first four hours of a flower's "one-day" anthesis period. While comparing the efficiency of pollen foraging in different bees, Strickler (1979) estimated that more than 70% of the pollen produced in a flower of *Echium vulgare* is removed during a single visit. In contrast, only 7–15% of the pollen in flowers of *Phlox pilosa* and *P. glaberrima* is removed by a single butterfly visit (Levin & Berube, 1972). In *P. cordata*, on average 45% of the pollen is removed during the first visit to a flower by a bumble bee. This level of pollen removal coupled with high pollinator densities at Pothole Lake (Wolfe & Barrett, 1988) results in rapid deletion of pollen from anthers.

The position of anthers within a flower is also likely to affect pollen removal in *P. cordata*. A single visit by a bumble bee to an unvisited flower removes different amounts of pollen from the three anther levels. Approximately four times as many pollen grains are removed from short-level anthers compared to long- and mid-level anthers because of the large number of pollen grains produced by these anthers (see Price & Barrett, 1982). However, if expressed as a proportion of the number of pollen grains per anther level, a single visit removes 38% of the pollen from short-level anthers, 50% from mid-level anthers and 69% from long-level anthers. Although the amount of pollen removed may be variable, our observations indicate that virtually all visits to previously unvisited flowers by bumble bees remove pollen. In contrast, not all visits deposit pollen (see below). Although most pollinator visits remove and deposit pollen, the probability is greater that early visits to previously unvisited flowers will remove rather than deposit pollen.

Studies of pollen deposition on naturally pollinated stigmas of heterostylous species indicate asymmetries in pollen transfer among the floral morphs (for a review see Ganders, 1979). In *P. cordata* (Price & Barrett, 1984, Barrett & Glover, 1985; Glover & Barrett, 1983) the M morph usually receives the largest total pollen load and the S morph the smallest. In addition, the L morph is most likely to experience legitimate pollination while the M and S morphs frequently display random pollination. Data from the present study involving single visits to previously unvisited flowers are consistent with those from bulk collections of stigmas from multiply-visited flowers. The largest total pollen loads were found in the M morph (\bar{x} 68 pollen grains per stigma) and the smallest loads were deposited on stigmas of the S morph (\bar{x} 18 pollen grains per stigma). With respect to the composition of pollen loads, the percent of the total pollen load composed of compatible (legitimate) pollen was considerably higher in the L morph (59%) than in the M (22%) or S (25%) morphs. The factors influencing these asymmetries in pollen deposition are complex and poorly understood. Pollen production differences among anther levels, pollinator positioning and interactions among pollen grains and stigmatic papillae are all likely to play a role.

A characteristic feature of data from studies of pollen deposition and carryover is that the number of pollen grains deposited on successively visited flowers differs to a large degree (Thomson & Plowright, 1980; Price & Waser, 1982). This was also found in *P. cordata* with approximately 30% of pollinator visits ineffective in depositing compatible pollen. Often these "blanks" occurred in the middle of a sequence of flowers on a single inflorescence visit. Aside from increasing the residency time of pollen on insects, and hence carryover distance, this phenomenon is unlikely to greatly influence seed set patterns. Flowers of *P. cordata* at Pothole Lake receive at least 10–20 pollinator visits per day and as a consequence multiply-visited flowers receive large numbers of compatible pollen grains. Since flowers of *P. cordata* possess a single ovule pollen loads are sufficient to maintain high levels of seed set. The relationships between pollen loads and seed set patterns are considered elsewhere.

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