SPATIAL VARIATION IN POLLINATOR VISITATION AND POLLEN LIMITATION ON FEMALE REPRODUCTIVE SUCCESS WITHIN A POPULATION OF AN ALPINE SPECIES

Masteroppgave i økologi
av
Sverre Lundemo

Institutt for naturforvaltning
Norges Landbrukshøgskole, Ås, 2004

SPATIAL VARIATION IN POLLINATOR VISITATION AND POLLEN LIMITATION ON FEMALE REPRODUCTIVE SUCCESS WITHIN A POPULATION OF AN ALPINE SPECIES

Summary

Pollen limitation influence together with resource limitation the reproductive output of plants. In this study I investigated how the degree of pollen limitation varied spatially between three sites within a population of *Dryas octopetala* L., a dwarf shrub, at an alpine location in southern Norway. To demonstrate if pollen limitation was evident, comparisons between supplementary pollinated flowers and control flowers were conducted. The sites were situated along an elevation gradient. Significant pollen limitation was only demonstrated for the mid-elevation site, while the other sites were resource limited. Pollinator visitation rates were measured to establish whether there was an association with seed set. Although there was no association between seed set and the number of pollinator visits, pollinator visitation rates influenced flower longevities of *Dryas*. Sites with low visitation rates had long-lived flowers and vice versa. Unexpectedly, flower longevities were shortest at the highest elevation site. Weather conditions likely influenced pollinator activity, and the plants responded to this by producing flowers of longevities more adjust to the visitation frequency.

Key words: Pollen limitation, pollinator visitation, *Dryas octopetala*, flower longevity, spatial variation.

Front page picture by Arne Anderberg

Introduction

Pollen availability and/or abiotic, environmental factors may affect the reproductive success of flowering plants (Bierzychudek 1981, Burd 1994). Infrequent visits by pollinators or insufficient transfer of pollen may cause a limitation on the number of seeds a plant can produce. Larson and Barrett (2000) mention that pollen limitation could also be an adaptive consequence to the posession of certain reproductive traits that are especially fit for an environment. Burd (1994) likewise suggests pollen limitation as a common outcome of resource strategies in a fluctuating pollination environment. This adaptation will result in a surplus production of ovules in many of the flowers, meaning that the pollinators will not be able to supply the flowers with enough pollen to ensure fertilization of them all (Burd 1995). The question then is whether the plant would be able to set more seeds if additional pollen was supplied.

Pollen limitation has previously been documented in many species through studies comparing the resulting reproductive success after natural and supplementary pollination with outcross pollen (Young and Young 1992, Burd 1994). Burd found that significant pollen limitation was demonstrated in some years or at some sites for 62 % of the species investigated. His results indicate that the pollination environment is rather dynamic and varies spatially, during a season or temporarily among seasons. When additional pollen supply (through supplementary pollination) leads to increased seed set in a plant, pollen limitation is demonstrated (Bierzychudek 1981, Zimmerman and Pyke 1988). A flower that receives supplementary pollination gets an opportunity to distribute more resources to seed production, either through the production of larger seeds (increased quality) or more seeds (increased quantity). This could possibly lead to a reallocation of resources within the plant, as surplus pollinated flowers require more than “their usual share”. The reallocated resources can be drawn from stored energy in the plant or the plant can redirect energy from other parts of the plant to such flowers. It is possible that other nearby flowers thereby will experience a reduction in resource availability and hence set a lesser amount of seed (Zimmerman and Pyke 1988), although Fox (1992) found this not to be the case in a study of Alaskan willows.

Often one would expect a higher production of seed set in supplementary pollinated flowers than in naturally pollinated flowers (Burd 1994), but that may not always be so. Haig and Westoby (1988) showed that when both pollen and abiotic resources constrain seed production simultaneously, seed production should not respond to an increase in pollen supply. This would be the case if the resource availability for some reason is limited, either by the aspect of sharing a common resource pool with other plants of different or the same species, by edaphic conditions (i.e. availability of nitrogen, phosphorus or water) or by other environmental constraints like wind, temperature, precipitation or predation. Available resources can also be viewed as part of trade-offs between survival and reproduction, and between male and female reproductive function (Olivieri et.al. 1994). If pollen limitation is demonstrated, there is still the question whether this will be the case in a longer perspective. Supplementary pollination could lead to an increase in seed set one year, but at the same time the plant may incur a cost by tapping its resource pool more than usual. This could entail a reduction in the plant’s reproduction during consecutive seasons. Thus a plant can be pollen limited during a single season, but lifetime reproductive success may be limited by resources (Ramsey 1995 and references therein, Alexandersson and Ågren 1996). Constant pollen limitation will favor traits that either increase pollen receipt or decrease the dependence on pollinators for pollen receipt (Haig and Westoby 1988). However, pollen limitation is seldom constant, resulting in a variation in the strenght and direction of selection on reproductive traits that reduce pollen limitation (Ramsey 1995).

Because alpine species likely live under more extreme weather conditions than e.g. temperate species, and pollen limitation is often related to the activity of pollinators, the reproductive success of plants at high altitudes/latitudes is often considered to be strongly pollen limited (Totland 1997, Totland and Eide 1999).

Successful fertilization may depend upon the microclimate in several ways. As the climate will vary on large and small scales, it is conceivable that there is a variation in pollinator visitation frequency to flowers, both on a large scale between populations and on a small scale within a population (Herrera 1995, Sullivan and Titus 1996). This could have secondary effects on pollen limitation, resulting in a spatial variation in reproductive success based on the spatial variation in pollen limitation (e.g. Santandreu and Lloret 1999). Pollinators regulate their visits to a species as a response to inter- and intrafloral density (see e.g Kunin 1997). Pollinator activity is also dependent upon the physical environment (e.g. wind, temperature, humidity, light). These factors will in turn cause changes in pollinator abundance or foraging behaviour, leading to temporal variation in pollen limitation (Ramsey 1995 and references therein). Wind and temperature may also influence exposure and receptivity of the style, and survival and germination of pollen grains (Corbet 1990). It is conceivable that edaphic conditions vary on a small and large scale, thereby causing a spatial inter- and intra-population variation in reproductive success.

Many alpine plants ensure fertilization of fruits with being self-compatible or wind-pollinated. Self-compatibility can act as insurance so that the plant at least will be able to set some seed but, due to a lack of genetic recombination, the following seed production might be of a lesser amount and quality compared to an outcrossing. Extending the flower longevity, the time a flower remains open and functional, is another strategy to mitigate low pollinator visitation. It is possible to view flower longevity as an optimum between three factors; maintenance cost of sustaining an already open flower relative to the cost of constructing a new one, rate of pollen receipt and rate of pollen dissemination (ovule fertilization) (Ashman and Schoen 1994). Such longevity variation will also be evident when comparing populations of a species, as these factors are likely to affect sites to different degrees depending on abiotic and biotic conditions.

The main purpose of this study was to determine whether pollen limitation on seed set occur in the alpine/arctic *Dryas octopetala*, and if variation among individuals of a population growing in contrasting environments was evident.
An important goal was to examine if seed production increased after experiencing supplementary pollination, or if that would cause a negative impact on nearby flowers through reallocation of resources.

Through registration of pollinators I wanted to examine if pollinator visitation rates to *Dryas* flowers differed between the sites and if this was related to flower density of *Dryas*, temperature, light intensity and wind.

Another important question in this thesis concerns flower longevity; if it is subject to spatial variation, related to pollinator intensity or more dependent upon abiotic factors.

Methods

Study species

Dryas octopétala L. (Rosáceae), the mountain avens, is a perennial dwarf shrub occurring in contrasting habitats such as tundra, coastal cliffs, sand dunes, mountain meadows, dry slopes and birch forests, often on calcareous soil. It is a circumpolar species, and is found in a belt in the alpine and arctic zones from the British Isles through Scandinavia, Northern Russia and North America to Greenland. At least four subspecies and several varieties have been recognized (Hitchcock and Cronquist 2001). The hermaphroditic flower consists normally of eight (seldom 9 or 10) white petals (Lid and Lid 1998). The sex of each flower can vary from almost pure male with virtually nonexistent stigmas to dominant stigmas with very small anthers around. Such gender variation appears to reflect different degrees of environmental stress, with more stressful conditions causing more flowers to become pure male (Wada and Kanda 2000). This can possibly reflect a condition of andromonoecy or androdioecy in *Dryas* (Wada 1999). The bowl-shaped flowers are sun-tracking, a characteristic known as flower heliotropism (Kjellberg et.al. 1982). This trait enables the flowers to focus the incoming solar radiation and thus increasing the temperature in the centre of the flower (Corbet 1990). After pollination, the flowers develop many small nuts (achenes) with feather-shaped styles which aid their wind dispersal. The plant forms a mat that produce flowers throughout the growing season (Lid and Lid 1998). Plants increase their size through clonal growth, and individual clones can live more than 100 years (Crawford 1989). This growth form enables the plant to persist in extreme environments, where sexual reproduction is often restricted. Flower primordia in *Dryas* are formed during the previous summer, and so the weather might have an impact on seed production the consecutive year (Wookey et.al. 1995).

Study area

Fieldwork was conducted during July and August 2003. The study site was situated approximately 1.5 km northeast of Finse, Hardangervidda, Norway (60º36´56’’N-7º31´8’’E), on the southwest slope of the mountain Sanddalsnuten at 1554 m asl. Sandalsnuten is situated in the mid-alpine vegetation zone, which mostly consists of grass heaths dominated by *Juncus trifidus*, *Carex bigelowii* and *Festuca ovina* (Dahl 1984). The bedrock in the area consists mostly of phyllite and siliceous schist in some places, with some marble from cambro-silur (Dahl 1997). This gives rise to rather calcicolous communities, where *Dryas* is one of the dominating species, forming so-called *Dryas* heath. A large population of *Dryas* covers most of the southwest slope of Sandalsnuten.

Mean summer temperature (June, July and August) at Finse (1222 m asl) is 6.3 ºC (Aune 1993), average monthly precipitation for the same period is 89 mm (Førland 1993). Mean temperatures in June, July, August at the study site in 2003 were measured with temperature loggers (Tinytag 12 Plus G data loggers, Intab Interface-Teknik AB, Sweden) on the ground. The temperatures recorded for the respective months were 9.1ºC, 11.6 ºC and 8.9 ºC respectively, giving a grand mean of 9.9 ºC for the growth season.

Fieldwork

To study how pollen limitation varied spatially within the population, I chose three different sites along a gradient running from the bottom to the summit of Sandalsnuten. The lowest elevation site was almost at the bottom of the slope, where there was a transition to grassy heath (ca. 1450 m asl). The mid-elevation site was at ca. 1500 m asl, and the highest elevation site was just below the summit (ca. 1550 m asl). At this site *Dryas* has a more patchy distribution, and the total area covered by *Dryas* mats was distinctly smaller than in the other sites.

On each site, I selected 15 pairs (groups) of *Dryas*. Plants in a pair occurred 1-4 meters from each other. From each pair one plant was randomly assigned to function as a control, while the other should receive supplementary pollination. Experimental flowers were selected so they were in more or less the same state of development, and they were all selected as flower buds. On the control plants, three control flowers (CC) were selected at random. I selected three pairs of flowers (S+CS), which were situated as closely as possible to each other on the other plant, and marked them. One member of each pair was randomly assigned to receive supplementary pollination (S), while the other would serve as control (CS). In this way, the experiment involved 9 flowers from each pair of plants. As far as it was possible when searching for a pair (S+CS), I tried to select flowers growing on the same ramet of a plant.

For the supplementary pollination I selected several donor flowers growing 4 - 15 meters from the recipient. I carefully brushed the anthers across the stigmas with a fine-tipped forceps so as to not damage the flower. Every supplementary pollinated flower was given pollen from three different donor flowers, and all flowers were pollinated during two non-consecutive days. The second pollination was conducted with two donor flowers to each recipient flower. By using more than one donor I sought to make the pollination more effective. I pollinated the flowers at site A and B from 5-10 July, and those at site C from 11-14 July.

Depending on the outcome in terms of differences in seed production among experimental groups, the following interpretation of results can be made:

- S > CC and CC = CS: There is pollen limitation on the seed production on the whole plant level (i.e. increased seed production after supplementary hand pollination does not occur at the expense of seed production in the nearby control flower)
- S > CC and CC > CS: There is pollen limitation on the seed production, but only on the individual flower level (i.e. availability of both pollen and abiotic resources constrain seed production simultaneously)
- S = CC: There is no pollen limitation

All flowers in this part of the experiment were harvested after the achenes were starting to mature and become ready for dispersal. In every flower I counted the total number of achenes, and categorized every achene as mature (containing developed seeds), non-mature (no developing seeds within the nut) or intermediate/uncertain (showing conditions of both categories). I based the criteria for placing the achenes in one of the categories on size and shape of the individual achene relative to each specific flower’s variation in achene size/shape. Counting and categorization of the achenes was done under a stereo microscope.

The matured achenes/ovule ratio and the (matured+uncertain achenes)/ovule ratio showed the same trend, and all comparisons between the different treatments and areas were therefore done with the (matured+uncertain achene)/ovule ratio. This ratio is hereafter referred to as seed set.

Four flowers (1.03%) were lost due to grazing by domestic sheep (*Ovis aries*) and these were excluded from the experiment. All flowers contained styles at the time of pollination, and harvested flowers with no remains of these styles (aborted during the season) were included (6 flowers, 1.55%). This resulted in a total of 383 flowers for inclusion in the experiment. Minitab (Version 14.11, Minitab Inc. 2004) was used for calculating descriptive statistics and nested ANOVAs for these ratios. I used treatment type and site as fixed factors, group nested within site as a random factor and the interaction between tratment and site to examine the effects on seed set.

To measure pollinator visitation rates to *Dryas* and co-flowering species, I recorded the number of visits by insects during 10 minute periods. A 1 m^2 quadrate was used when conducting the measurements. The quadrate was moved to a new patch of flowers for each measurement. Before the measurements started I counted the number of open flowers of all species within the quadrate. I positioned the quadrate before the 10 minute periods so that it always contained from 5 – 100 *Dryas* flowers. At least 20 observation periods were conducted in each site (A, B and C). Date and time were recorded and temperature and light intensity (Li-Cor Model 250, Li-Cor Inc., Environmental Division, Lincoln, Nebraska) measured before each period started. Measurements were conducted between 10 a.m. and 7 p.m. During the visitation periods I tried to move as little as possible to avoid disturbing the insects. A flower visit was defined to have occurred when the insect touched the anther and/or the stigma. Revisits to the flower was counted as two visits. The visiting insects were categorized into four groups; large flies (Muscidae), small flies/mosquitos (e.g., Chironomidae, Sciaridae), hoverflies (Syrphidae) and butterflies (Lepidoptera sp.). A total of 65 visitation periods were conducted. The sampled material was processed in Minitab. I used one-way ANOVA and scatter plots with regression (stepwise regression) lines to demonstrate any relationship between visitation rates and flower densities (versus *Dryas* density or density of all flowers except *Dryas*).

I marked 5 quadrates ($1m^2$) to describe the flowering phenology within each of the three sites. Every second day, I counted the number of open *Dryas* flowers in these quadrates until the amount of flowers was less than one-tenth of the maximum flower number counted.

To examine if flower longevity differed among the sites I also marked three flower buds with split drinking straws in different colours. I measured the time from the opening of the flower bud (when the petals start to unfold) to withering. The resulting data was processed in Microsoft Excel (MS Office 2000, Microsoft Corporation), and one-way ANOVA was performed in Minitab to demonstrate if there was any difference in longevity between the sites.

Results

Pollen limitation

Seed set differed considerably between sites and treatments (Table 1, 2). Supplementary pollinated flowers (S) set on average 20.0 % more seed than control flowers on control plants (CC) for all sites combined. For site A, supplementary pollinated flowers set 13.2 % less seed than flowers on control plants, while B and C experienced increases (60.0 % and 36.0 % respectively). Control flowers on supplementary pollinated plants (CS) set 29.4 % and 12.0 % less seed than control flowers on control plants (CC) in site A and C, respectively, while seed set in site B increased with 20.0 %.

However, ANOVA analyses showed that the differences in seed set between treatments was only statistically significant for site B when comparing S flowers and CC flowers, and only for site A when comparing CS flowers and CC flowers (Table 3).

Table 1 Nested ANOVA on the effects of pollination treatments (supplementary pollinated, control on supplementary pollinated plants and control on control plants), site (A, B and C) and the treatment x area interaction. Group = pair of *Dryas* plants (consists of S, CS and CC flowers). S = supplementary pollinated, CS = control on supplementary pollinated plants, CC = control on control plants. Df = degree of freedom, Adj MS = adjusted mean squares, F = F-ratio, P = significance level.

		Df	Adj MS	F	P
S vs. CC	Treatment	1	0.17022	5.65	0.022
	Area	2	1.01687	36.33	< 0.001
	Treatment x Area	2	0.22001	7.30	0.002
	Group (Area)	40	0.02799	0.93	0.591
	Error	40	0.03012		
CS vs. CC	Treatment	1	0.05066	1.41	0.242
	Area	2	0.94474	32.23	< 0.001
	Treatment x Area	2	0.15106	4.20	0.022
	Group (Area)	40	0.02931	0.82	0.739
	Error		0.03595		

Table 2 Seed set of the different treatment groups (S, CS, CC) within the three sites (A, B, C). S= flower on experimental plant receiving hand pollination, CS= control flower on experimental plant, CC= control flower on control plant, n = number of groups within each treatment (consisting of both S, CS and CC flowers).

	Treatment group			
Area	S Mean±SE (n)	CS Mean±SE (n)	CC Mean±SE (n)	Area mean Mean±SE
A	0.59±0.16 (14)	0.48±0.16 (14)	0.68±0.15 (14)	0.58±0.16
B	0.72±0.14 (14)	0.54±0.19 (14)	0.45±0.17 (14)	0.57±0.17
C	0.34±0.18 (15)	0.22±0.19 (15)	0.25±0.21 (15)	0.27±0,19
Treatment mean	0.54±0.16 (43)	0.41±0.18 (43)	0.45±0.18 (43)	0.47±0.17

Table 3 P-values from one-way ANOVA on the effect of pollination treatments (supplementary pollinated, control on supplementary pollinated plants, and control on control plants) within each site (A, B and C). S = supplementary pollinated, CS = control on supplementary pollinated plants, CC = control on control plants.

Site	S vs CC	CS vs CC
A	0.138	0.002
B	< 0.001	0.230
C	0.210	0.749

Pollinator visitation

Flower visitation rates to *Dryas* varied greatly between the three sites. The highest visitation rate per flower ratio was at the highest elevation site (C), whereas the lowest visitation rate was at the mid-elevation site (B). At the lowest elevation site (A), visitation rate was intermediate to B and C (Fig 1.).

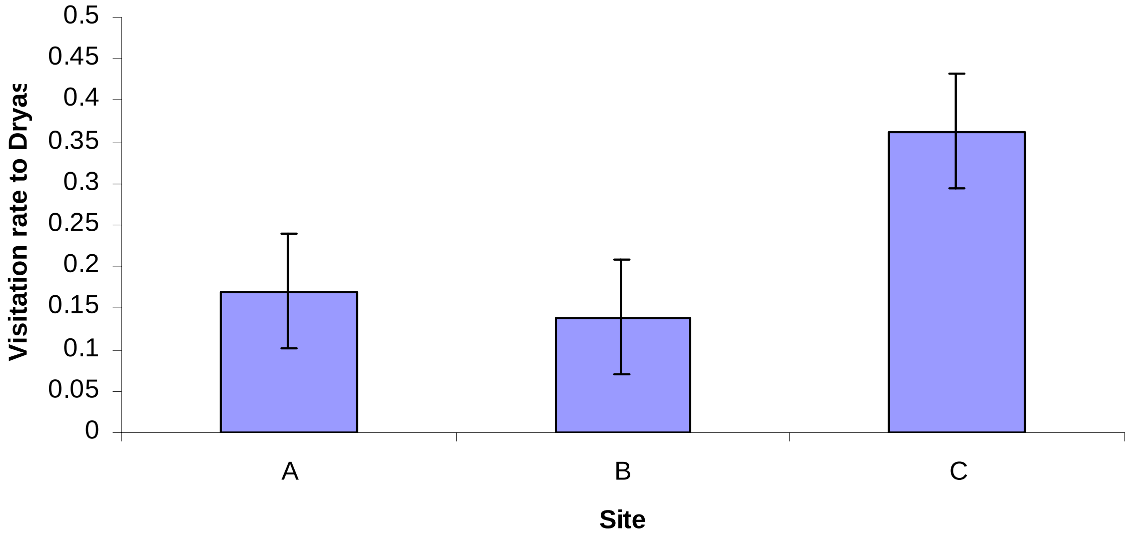

Fig. 1 Mean visitation rates ± SE to *Dryas* during 10 minutes at the three sites (n = 65)

The total number of *Dryas* flowers inside the quadrates was about the same at all sites (mean±SD: A= 43.3±32.9, B= 48.9±35.5, C= 44.3±35.4, entire population = 45.6±34.2). Simple linear regression showed that visitation rate per *Dryas* flower was not related to the number of *Dryas* flowers per quadrate in site A or B, while there was a significant correlation in site C (Fig. 2, Table 3). The same regression showed that visitation rate to all flowers combined (except *Dryas*) was only significant in site B (Fig. 3, Table 4).

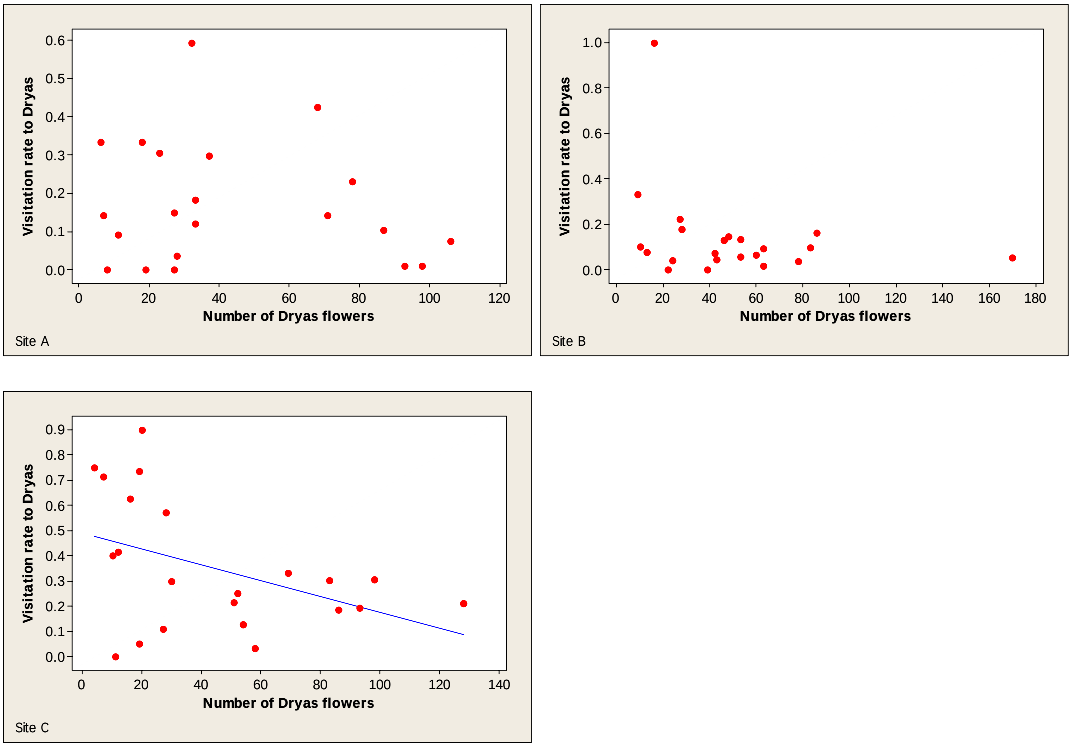

Fig. 2 Relationship between the number of *Dryas* flowers within 1m² and visitation rates to *Dryas* during 10 min. periods in site A-C. Significant relationship shown with regression line; Visitation rate to *Dryas* = 0.492 - 0.00316 number of *Dryas*.

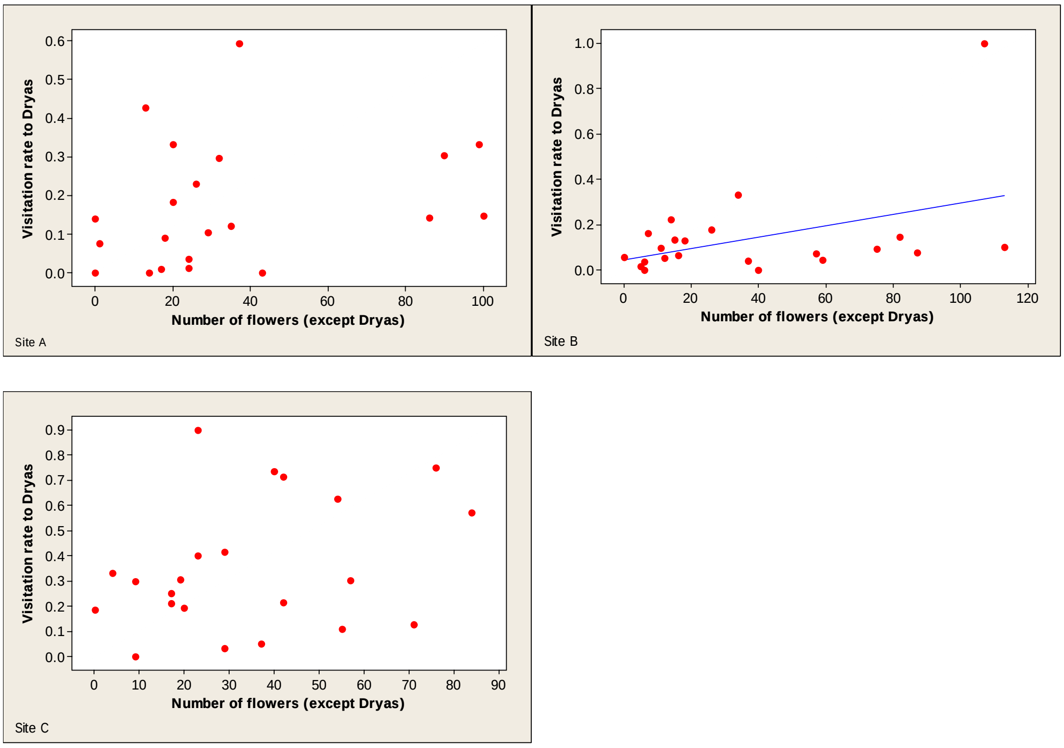

Fig. 3 Relationship between the total number of flowers (except *Dryas*) and visitation rates to *Dryas* in 1m² during 10 min. periods in site A-C. Significant relationship shown with regression line; Visitation rate to *Dryas* = 0.0454 + 0.00250 number of flowers (except *Dryas*).

Table 4 P-values of simple linear regression between the number of *Dryas* flowers/all flowers except *Dryas* and visitation rate to *Dryas* at the three sites.

	A	B	C
Number of *Dryas* vs. visitation to *Dryas*	0.482	0.201	0.044
Number of flowers (except *Dryas*) vs. visitation to *Dryas*	0.245	0.049	0.158

A total of 570 visits were observed during the periods. Large muscoid flies represented 61.4 % of all visits, while small muscoids represented 37.7 % of the visits. Syrphidae (hoverflies) and Lepidoptera (butterflies) combined only made up 0.09 % of the visits to *Dryas*. Large muscoids dominated among the pollinators at site A and C, while small muscoids was by far the most abundant group at site B (Fig. 4).

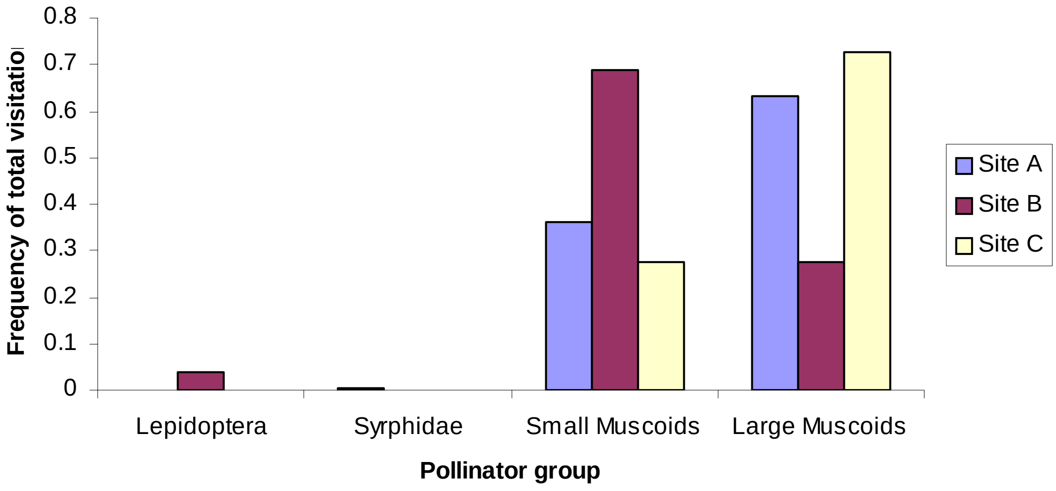

Fig. 4 Visitation rates by the different pollinator groups to flowers at the three sites.

Flowering phenology

Site A and B had almost identical flowering periods, while the highest elevation site had a later and shorter flowering period (Fig. 5). The three sites had almost equal numbers of flowers during their flower peak, and terminated flowering at about the same time.

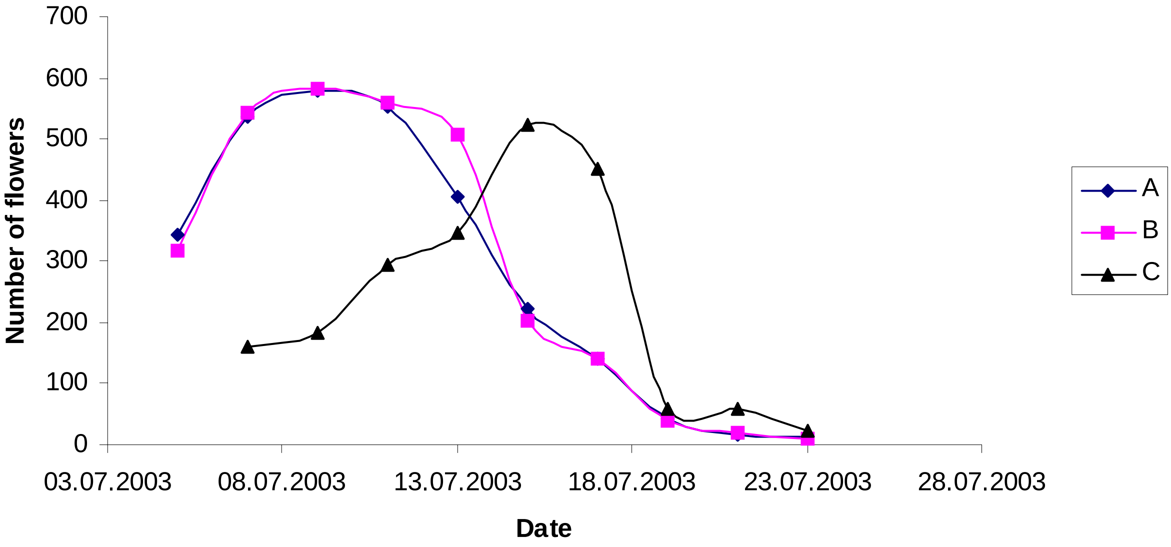

Fig. 5 The total number of open *Dryas* flowers within 1m² quadrates at the three sites.

Flower longevity

The flower longevity differed significantly among the sites (Table 5). Flowers in site C were open for a markedly shorter time compared with B and A (29.6 % and 25.2 % respectively) (Fig. 6).

Table 5 One-way ANOVA on the difference in flower longevity among sites. Df = degree of freedom, MS = mean squares, F = F-ratio, P = significance level.

	Df	MS	F	P
Site	2	29.867	73.5	< 0.001
Error	42	0.406		

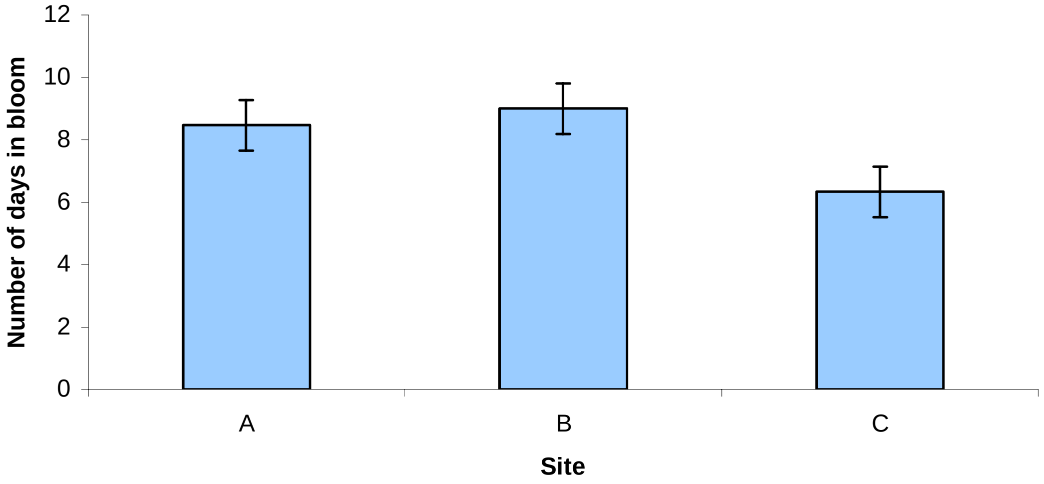

Fig. 6 Mean duration from opening to withering (flower longevity) ± SE of *Dryas* flowers at the three different sites.

Wind measurements were done to see if there was a variation in this abiotic factor between the sites (Fig. 7). Not surprisingly, the average wind velocity at the highest elevation site was always higher than at the other sites. Average wind speed at site A and B was in general rather similar. Maximum wind velocity was also almost always highest at site C.

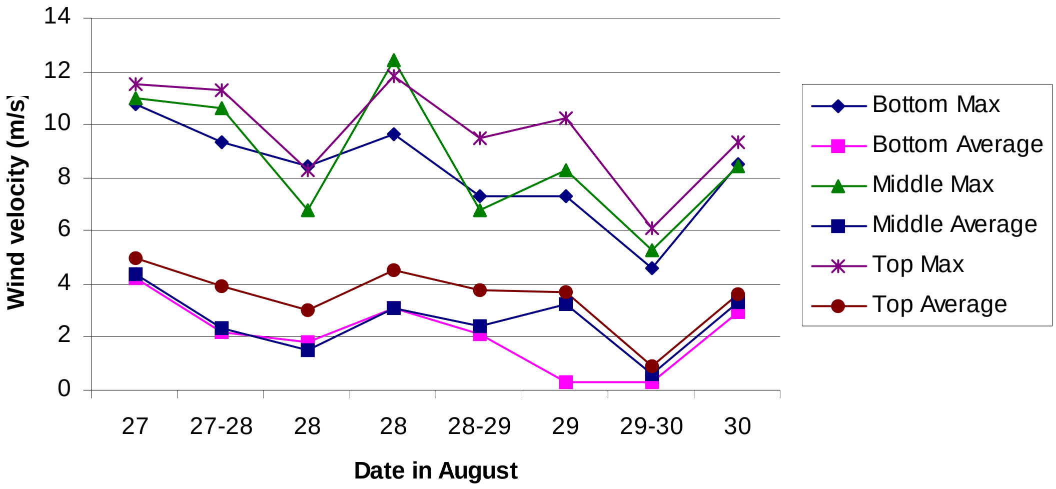

Fig. 7 Results from wind measurements at three different sites (A, B, C) at Mount Sandalsnuten in August 2003.

Temperature and light levels were both highest at site C. At this site light intensity was 20.3 % and 21.7 % higher and temperature was 26.7 % and 16.5 % higher than at site A and B, respectively. Whether temperature nor light was significantly correlated to visitation rates.

Discussion

Dryas octopetala flowers are large and conspicous in their habitat, and individual plants likely allocate a relatively large amount of their available resources to their floral display. According to Burd (1995) species with expensive flowers will experience more frequent pollen limitation because a larger amount of resources is allocated to pollinator attraction. This would suggest, together with the harsh environmental conditions the plants are exposed to, that female reproductive success of *Dryas* should be pollen limited.

However, my results show that the degree of pollen limitation in *Dryas* varies within the same population. At the lowest elevation site, probably the most benign site in terms of abiotic environmental conditions, it seems like pollen limitation is absent, and that supplementary pollination incurred a non-significant, negative effect on the plant’s reproductive success. The reason for that could be that the supplementary pollination has provided the stigmas with pollen of lower quality than naturally pollinated plants receives through pollinator activity. Another possible explanation is that pollen tube crowding or damage during the supplementary pollination could have negatively affected the flowers’ reproduction. A reduction in seed set could also occur because the high pollen loads of the supplementary pollinated flowers would attract pollen thieves, the pollen used may be inviable (e.g. through interspecific pollen transfer), or due to a statistical Type I error caused by a relatively low sample size (Young and Young 1992). The possibility that the plants in this site are limited by resources rather than pollen may contribute to explain the lack of pollen limitation, but not the actual reduction in seed set after pollen addition.

The plants in the mid-elevation site was pollen limited on the whole-plant level. Here the control flowers on both control plants and on supplementary pollinated plants exhibited similar seed set, while the supplementary pollinated flowers set significantly more seed. Thus, reproductive success of *Dryas* at site B do not appear to be resource limited. At site B the number of large muscoids acting as pollinators was very low compared to the other sites, and small muscoids was the most important pollinator group (Fig. 5). When the flower of a species such as *Dryas* do not exhibit a special structure that is adapted to a special type of pollinators, large pollinators are expected to be more effective than small and this may contribute to explain the significant pollen limitation at this site. Almost all visits to *Dryas* were conducted by dipteran species. These are in general unspecialized, but often abundant insects that normally are considered relatively ineffective as pollinators. However, Totland (1997) found in one study in the Finse area that dipterans were still able to provide *Ranunculus acris* flowers with adequate amounts of pollen as the pollinator assembly comprised many species and visitation activity was high. Although visitation rates were low at site B, and most of the visits were conducted by small dipterans, seed set was not especially different (when ignoring CC in site A) from A. Both temperature and light levels were about the same in site A and B and differences in edaphic conditions is therefore a possible explanation to the variation in pollen limitation between these sites.

At the highest elevation site *Dryas* showed a trend toward being pollen limited on a plant level, but this trend was not significant. Seed set at this site was considerably lower than at the other sites, possibly due to abiotic, environmental constraints on reproductive success. At this site the occurrence of pure-male flowers was higher than at the other sites (*personal observation*). This could indicate, as a consequence of resource limitation on reproductive success, that the plants at this site allocate more resources to the male function because producing fruit and seed demands more energy than producing pollen. This would be consistent with the results of Wada & Kanda (2000) who found that enhanced growth conditions through a warming manipulation resulted in a higher investment to the gynoecium of *Dryas* flowers. Since the plants at site C are resource limited, increased pollen availability both by the occurrence of a higher percentage of males and a high visitation rate would not benefit female reproductive success at this site.

Site C had higher wind velocities, but also higher temperatures and light intensities than the other sites. High visitation rates is often associated with high temperature and light levels, especially in high altitude habitats (McCall and Primack 1992). Wind speed may inhibit flight by pollinators mechanically and through increasing energy expenditure during flight (Brantjes 1981). The wind often made it difficult for insects to fly between flowers, and especially muscoids were often observed resting on the petals of *Dryas* flowers (*personal observation*). During periods with less wind this lack of feeding activity was compensated for through increased activity by the pollinators.

Visitation rates to *Dryas* were highest at site C, something that could be due to a lower competition for pollinators at this site. At site A and C the mean number of flowers were almost equal (mean±SD = 77.5±32.7, and 78.7±35.3 respectively), while B had a higher density of flowers (mean±SD = 86.5±36.9). The fact that site B had the lowest visitation rate could be influenced by the higher flower density at this site leading to intra- or interspecific competition for the attraction of the pollinators. The variation in visitation rates among sites corresponded to the differences in average flower longevity; the site with the highest visitation had the shortest longevity and vice versa. Thus, it appears that flowers regulate their longevity (actually timing of withering) according to the pollination intensity they experience.

Although flowers at site C had the highest visitation rates they also produced the smallest amounts of seed compared to the other sites. Thus, at this within-population scale, there was no relationship between the site-specific pollinator visitation rate and resulting seed set. Flowers at site C could have responded to supplementary pollination, but constraints from abiotic resource availability likely caused the reproduction to stay on a low level. At site B, the availability of abiotic resources was apparently sufficient, resulting in considerable higher seed set in flowers that received supplementary pollen compared to control flowers.

The lack of difference in seed set between CS flowers and CC flowers in site B and C showed that supplementary pollination did not lead to a reallocation of resources between flowers within *Dryas* plants in order to benefit from the additional pollen. In site A, CS flowers experienced a reduction in seed set compared to CC flowers, but since the S flowers also set less seed than CC flowers, this difference must probably have been due to other factors.

The flower longevity differences in my study is contrary to earlier studies, where flower longevity has been found to increase at higher altitudes (e.g. Fabbro and Körner 2003). Although the altitudinal difference between the sites in my study is small and all flowers belong to the same population, there is high inter-populational variation. Normally, rather similar flower longevities would have been expected, but differences in pollination intensity and spatial variation in climate and edaphic conditions cause a differentiation between the sites.

My results clearly demonstrate that the extent of pollen limitation varies spatially within a population. Weather and edaphic conditions appear to be major factors that regulate reproductive success and also influence the degree of pollen limitation in *Dryas* in this population. Future studies could benefit from taking this small-scale variation between patches into account when investigating pollinator visitation, flower longevity, pollen limitation, and female reproductive success.

A long-term study is neccessary to reveal whether these results vary among seasons and to see if increased seed set after supplementary pollination leads to reduced lifetime reproductive success.

Acknowledgements

I would like to thank The High Mountain Ecology Research Station at Finse for providing me with acommodation, Jorun Nyléhn for her assistance in the field work and my supervisor Ørjan Totland for all help and comments on the manuscript during this study.

Literature cited

Alexandersson, R., Ågren, J. 1996. Population size, pollinator visitation and fruit production in the deceptive orchid *Calypso bulbosa*. Oecologia 107:533-540.

Ashman, T., Schoen, D.J. 1994. How long should flowers live? Nature 371:788-791.

Aune, B. 1993. Temperaturnormaler: normalperiode 1961-1990. Det Norske meteorologiske institutt, Oslo.

Bierzychudek, P. 1981. Pollinator limitation of plant reproductive effort. American Naturalist 117:838-840.

Brantjes, N.B.M. 1981. Wind as a factor influencing flower-visiting by *Hadena bicruris* (Noctuidae) and *Deilephila elpenor* (Sphingidae). Ecological Entomology 6:361-363.

Burd, M. 1994. Bateman's principle and plant reproduction: The role of pollen limitation in fruit and seed set. Botanical Review 60:83-139.

Burd, M. 1995. Ovule packaging in stochastic pollination and fertilization environments. Evolution 49:100-109.

Corbet, S.A. 1990. Pollination and the weather. Israel Journal of Botany 39:13-30.

Crawford, R.M.M. 1989. Studies in plant survival. Blackwell Scientific, Oxford.

Dahl, E. 1984. En oversikt over plantesamfunn på Finse. Rapporter fra Høyfjellsøkologisk forskningsstasjon, Finse, Norge. Universitetene i Bergen og Oslo.(in norwegian). 1: 30 pp.

Dahl, S.O. 1997. Berggrunnsgeologi og geomorfologi på Finse. Rapporter fra Høyfjellsøkologisk forskningsstasjon, Finse, Norge = Reports from The High Mountain Ecology Research Station, Finse, Norway. Bergen ; Oslo : Universitetene i Bergen og Oslo (in norwegian). 16-18.

Fabbro,T., Körner, C. 2003. Altitudinal differences in flower traits and reproductive allocation. Flora 199:70-81.

Fox, J.F. 1992. Pollen limitation of reproductive effort in willows. Oecologia 90:283-287.

Førland, E.J. 1993. Precipitation normals, normal period 1961-1990. The Norwegian Meteorological Institute, Oslo.

Haig, D., Westoby, M. 1988. On limits to seed production. American Naturalist

131:757-759.

Herrera, C.M. 1995. Microclimate and individual variation in pollinators: flowering plants are more than their flowers. Ecology 76:1516-1524.

Hitchcock, C.L., Cronquist, A. 2001**.** Flora of the Pacific Northwest. University of Washington Press, Seattle, WA. 730 p.

Kjellberg, B., Karlsson, S., Kerstensson, I. 1982. Effects of heliotropic movements of flowers of *Dryas octopetala* on gynoecium temperature and seed development. Oecologia 54:10-13.

Kunin, W.E. 1997. Population size and density effects in pollination: pollinator foraging and plant reproductive success in experimental arrays of *Brassica kaber*. Journal of Ecology 85:225-234.

Larson, B.M.H., Barrett, S.C.H. 2000. A comparative analysis of pollen limitation in flowering plants. Biological Journal of the Linnean Society 69:503-520.

Lid , J., Lid, D.T. 1998. Norsk flora. 6th ed. Oslo: Det Norske Samlaget (in norwegian).

McCall, C., Primack, R. B. 1992. Influence of flower characteristics, weather, time of day, and season on insect visitation rates in three plant communities. American Journal of Botany 79:434-442.

Olivieri, I., Couvet, D., Slatkin, M. 1994. Allocation of reproductive effort in perennial plants under pollen limitation. The American Naturalist 144:373-394.

Ramsey, M. 1995. Causes and consequenses of seasonal variation in pollen limitation of seed production in *Blandfordia grandiflora* (Liliaceae). Oikos 73:49-58.

Santandreu, M., Lloret, F. 1999. Effect of flowering phenology and habitat on pollen limitation in *Erica multiflora*. Canadian Journal of Botany 77:734-743.

Sullivan, G., Titus, J.E. 1996. Physical site characteristics limit pollination and fruit Set in the dioecious hydrophilous species, *Vallisneria americana*. Oecologia 108:285-292.

Totland, Ø. 1997. Limitations on reproduction in alpine *Ranunculus acris*. Canadian Journal of Botany 75:137-144.

Totland, Ø., Eide, W. 1999. Environmentally-dependent pollen limitation on seed production in alpine *Ranunculus acris*. Ecoscience 6:173-179.

Wada, N. 1999. Factors affecting the seed-setting success of *Dryas octopetala* in front of Brøggerbreen (Brøgger Glacier) in the high Arctic, Ny-Ålesund, Svalbard. Polar Research 18:261-268.

Wada, N., Kanda, H. 2000. Notes on floral traits and gender expression of *Dryas octopetala* under a simulated environmental change. Polar Bioscience, 13:147-151.

Wookey, P.A., Robinson, C.H., Parsons, A.N., Welker, J.M., Press, M.C., Callaghan, T.V., Lee, J.A. 1995. Environmental constraints on the growth, Photosynthesis and reproductive development of *Dryas octopetala* at a high Arctic polar semi-desert, Svalbard. Oecologia 102:478-489.

Young, H.J., Young, T.P. 1992. Alternative outcomes of natural and experimental high pollen loads. Ecology 73:639-647.

Zimmerman, M., Pyke, G.H. 1988. Reproduction in *Polemonium*: assessing the factors limiting seed set. The American Naturalist 131:723-738.

Appendix 1

Compilation of data on seed set in *Dryas octopetala* from the three sites

Site	Group	Treatment	(Seeds+unsecure)/ovules
A	1	S	0.70175439
A	1	S	0.84615385
A	1	S	0.54098361
A	1	CS	0.28301887
A	1	CS	0.15714286
A	1	CS	0.66666667
A	1	CC	0
A	1	CC	0.54545455
A	1	CC	0.69014085
A	3	S	0.8877551
A	3	S	0.78873239
A	3	CS	0.77647059
A	3	CS	0.5625
A	3	CC	0.86538462
A	3	CC	0.66071429
A	3	CC	0.72131148
A	4	S	0.36111111
A	4	S	0.44285714
A	4	S	0.31818182
A	4	CS	0.49152542
A	4	CS	0.76712329
A	4	CS	0.38095238
A	4	CC	0.64473684
A	4	CC	0.5625
A	4	CC	0.42105263
A	5	S	0.28846154
A	5	S	0.46551724
A	5	S	0.64705882
A	5	CS	0.18367347
A	5	CS	0.54385965
A	5	CS	0.65
A	5	CC	0.52212389
A	5	CC	0.64893617
A	5	CC	0.55714286
A	6	S	0.23255814
A	6	S	0.38
A	6	S	0.76271186
A	6	CS	0.42857143
A	6	CS	0.22916667
A	6	CS	0.19117647
A	6	CC	0.09375
A	6	CC	0.86666667
A	6	CC	0.8
A	7	S	0.85148515
A	7	S	0.66666667
A	7	S	0.81632653
A	7	CS	0.79661017

A	7	CS	0.30645161
A	7	CS	0.08163265
A	7	CC	0.70212766
A	7	CC	0.93617021
A	7	CC	0.90196078
A	8	S	0.55555556
A	8	S	0.63888889
A	8	S	0.51785714
A	8	CS	0.47058824
A	8	CS	0.62162162
A	8	CS	0.39393939
A	8	CC	0.47368421
A	8	CC	0.34545455
A	8	CC	0.66129032
A	9	S	0.92156863
A	9	S	0.69230769
A	9	S	0.61904762
A	9	CS	0.125
A	9	CS	0.69565217
A	9	CS	0.75
A	9	CC	0.06122449
A	9	CC	0.75
A	9	CC	0.90384615
A	10	S	0.80851064
A	10	S	0.89130435
A	10	S	0.62
A	10	CS	0.35555556
A	10	CS	0.54545455
A	10	CS	0.2745098
A	10	CC	0.88636364
A	10	CC	0.80952381
A	10	CC	0.77777778
A	11	S	0.60869565
A	11	S	0.64285714
A	11	S	0.67924528
A	11	CS	0.68518519
A	11	CS	0.19148936
A	11	CS	0.66
A	11	CC	0.92156863
A	11	CC	0.75510204
A	11	CC	0.65079365
A	12	S	0.57777778
A	12	S	0.35
A	12	S	0.10869565
A	12	CS	0
A	12	CS	0.45454545
A	12	CS	0.26666667
A	12	CC	0.81632653
A	12	CC	0.88333333
A	12	CC	0.66153846
A	13	S	0.88421053

A	13	S	0.12307692
A	13	S	0.82417582
A	13	CS	0.90789474
A	13	CS	0.87356322
A	13	CC	0.86792453
A	13	CC	0.75925926
A	13	CC	0.92307692
A	14	S	0.63636364
A	14	S	0.90697674
A	14	S	0.02857143
A	14	CS	0.54054054
A	14	CS	0.74626866
A	14	CS	0.2
A	14	CC	0.6
A	14	CC	0.75342466
A	14	CC	0.65
A	15	S	0.76470588
A	15	S	0.44827586
A	15	S	0.19354839
A	15	CS	0
A	15	CS	0.48
A	15	CS	0.6744186
A	15	CC	0.90322581
A	15	CC	0.87804878
A	15	CC	0.8
B	1	S	0.71428571
B	1	S	0.88888889
B	1	S	0.74242424
B	1	CS	0.28888889
B	1	CS	0.82539683
B	1	CS	0.81632653
B	1	CC	0.06818182
B	1	CC	0.41860465
B	1	CC	0.36538462
B	3	S	0.85294118
B	3	S	0.17460317
B	3	S	0.90909091
B	3	CS	0.72881356
B	3	CS	0
B	3	CS	0.54347826
B	3	CC	0.36206897
B	3	CC	0.53448276
B	3	CC	0.82692308
B	4	S	0.64814815
B	4	S	0.88571429
B	4	S	0.56756757
B	4	CS	0.76086957
B	4	CS	0.35294118
B	4	CS	0.65853659
B	4	CC	0.18518519
B	4	CC	0.75

B	4	CC	0.44
B	5	S	0.78181818
B	5	S	0.65454545
B	5	S	0.80769231
B	5	CS	0.8
B	5	CS	0
B	5	CS	0.62264151
B	5	CC	0.91304348
B	5	CC	0.72727273
B	5	CC	0.73684211
B	6	S	0.83098592
B	6	S	0.875
B	6	S	0.73333333
B	6	CS	0.93055556
B	6	CS	0.85106383
B	6	CS	0.85106383
B	6	CC	0.75757576
B	6	CC	0.76666667
B	6	CC	0.52941176
B	7	S	0.44680851
B	7	S	0.77083333
B	7	S	0.93220339
B	7	CS	0.71428571
B	7	CS	0.32786885
B	7	CS	0.45
B	7	CC	0.375
B	7	CC	0.34146341
B	7	CC	0
B	8	S	0.94642857
B	8	S	0.84057971
B	8	S	0.62903226
B	8	CS	0.74137931
B	8	CS	0.62857143
B	8	CS	0.9122807
B	8	CC	0.47916667
B	8	CC	0.34693878
B	8	CC	0.13636364
B	9	S	0.70454545
B	9	S	0.9047619
B	9	S	0.9
B	9	CS	0.72916667
B	9	CS	0.66666667
B	9	CS	0.85714286
B	9	CC	0.71186441
B	9	CC	0.20408163
B	9	CC	0.17333333
B	10	S	0.89830508
B	10	S	0.70833333
B	10	S	0.8
B	10	CS	0.51020408
B	10	CS	0

B	10	CS	0.7
B	10	CC	0.54901961
B	10	CC	0.90384615
B	10	CC	0.05769231
B	11	S	0.83333333
B	11	S	0.80952381
B	11	S	0.82142857
B	11	CS	0.22413793
B	11	CS	0.44827586
B	11	CS	0.04166667
B	11	CC	0.84210526
B	11	CC	0
B	11	CC	0.16666667
B	12	S	0.91525424
B	12	S	0.85714286
B	12	S	0.86666667
B	12	CS	0.53448276
B	12	CS	0.05882353
B	12	CS	0.77192982
B	12	CC	0.45614035
B	12	CC	0.0483871
B	12	CC	
B	13	S	0.26530612
B	13	S	0.54545455
B	13	S	0.34
B	13	CS	0.25581395
B	13	CS	0.76470588
B	13	CC	0
B	13	CC	0.73770492
B	13	CC	0
B	14	S	0.89285714
B	14	S	0.65671642
B	14	S	0.29347826
B	14	CS	0.14893617
B	14	CS	0.32941176
B	14	CS	0.15873016
B	14	CC	0.44827586
B	14	CC	0.80769231
B	14	CC	0.52054795
B	15	S	0.25925926
B	15	S	0.5
B	15	S	0.67567568
B	15	CS	0.35416667
B	15	CS	0.70588235
B	15	CS	0.9047619
B	15	CC	0.69230769
B	15	CC	0.4047619
B	15	CC	0.38297872
C	1	S	0
C	1	S	0.03529412
C	1	S	0.2625

C	1	CS	0.03333333
C	1	CS	0
C	1	CS	0.20253165
C	1	CC	0.12048193
C	1	CC	0.15068493
C	1	CC	0
C	2	S	0
C	2	S	0.35869565
C	2	S	0.04819277
C	2	CS	0
C	2	CS	0.14285714
C	2	CS	0.14606742
C	2	CC	0.92307692
C	2	CC	0.85555556
C	2	CC	0.75903614
C	3	S	0.42857143
C	3	S	0.38888889
C	3	S	0.61111111
C	3	CS	0.16666667
C	3	CS	0
C	3	CS	0.14
C	3	CC	0.44680851
C	3	CC	0.57142857
C	3	CC	0.42592593
C	4	S	0.28985507
C	4	S	0.52
C	4	S	0.06666667
C	4	CS	0
C	4	CS	0
C	4	CS	0.12698413
C	4	CC	0.06976744
C	4	CC	0.07575758
C	4	CC	0.42553191
C	5	S	0.41304348
C	5	S	0.60344828
C	5	S	0.73214286
C	5	CS	0.77419355
C	5	CS	0.54385965
C	5	CS	0.57575758
C	5	CC	0.075
C	5	CC	0
C	5	CC	0.38461538
C	6	S	0.81818182
C	6	S	0.75471698
C	6	S	0.5
C	6	CS	0.46666667
C	6	CS	0.64912281
C	6	CS	0.34210526
C	6	CC	0.03125
C	6	CC	0.14285714
C	6	CC	0

C	7	S	0.24074074
C	7	S	0.14583333
C	7	S	0.1754386
C	7	CS	0.02083333
C	7	CS	0.03846154
C	7	CS	0.06896552
C	7	CC	0.16666667
C	7	CC	0.21875
C	7	CC	0.06153846
C	8	S	0.23076923
C	8	S	0.1875
C	8	S	0.45454545
C	8	CS	0.18333333
C	8	CS	0
C	8	CS	0.06779661
C	8	CC	0.36842105
C	8	CC	0.05263158
C	8	CC	0.44827586
C	9	S	0.84
C	9	S	0.41538462
C	9	S	0.5483871
C	9	CS	0.1884058
C	9	CS	0
C	9	CS	0
C	9	CC	0
C	9	CC	0.52
C	9	CC	0
C	10	S	0.37096774
C	10	S	0.71052632
C	10	S	0.05
C	10	CS	0.16666667
C	10	CS	0.671875
C	10	CS	0.32352941
C	10	CC	0.14473684
C	10	CC	0.01515152
C	10	CC	0
C	11	S	0.53571429
C	11	S	0.06382979
C	11	S	0.48979592
C	11	CS	0.10204082
C	11	CS	0.525
C	11	CS	0.26785714
C	11	CC	0.43283582
C	11	CC	0.23809524
C	11	CC	0
C	12	S	0.15151515
C	12	S	0.27586207
C	12	S	0.08333333
C	12	CS	0.09090909
C	12	CS	0.07843137
C	12	CS	0.02

C	12	CC	0.18367347
C	12	CC	0.26666667
C	12	CC	0.18181818
C	13	S	0.11904762
C	13	S	0.07692308
C	13	S	0.17460317
C	13	CS	0.12
C	13	CS	0.14814815
C	13	CS	0.16666667
C	13	CC	0.16666667
C	13	CC	0.16
C	13	CC	0.23913043
C	14	S	0.16666667
C	14	S	0.30666667
C	14	S	0.31034483
C	14	CS	0.36666667
C	14	CS	0.0990099
C	14	CS	0.43617021
C	14	CC	0.10204082
C	14	CC	0.58461538
C	14	CC	0.36
C	15	S	0.32692308
C	15	S	0.2962963
C	15	S	0.36065574
C	15	CS	0.46774194
C	15	CS	0.36170213
C	15	CS	0.53658537
C	15	CC	0
C	15	CC	0.06976744
C	15	CC	0

Appendix 2

Compilation of data on flower density and pollinator visitation rates to *Dryas octopetala*

Site	Number of Dryas flowers	Number of visits to Dryas	Visitation rates to Dryas	Number of flowers (except Dryas)
A	33	4	0.121212121	35
A	8	0	0	43
A	87	9	0.103448276	29
A	23	7	0.304347826	90
A	33	6	0.181818182	20
A	19	0	0	14
A	71	10	0.14084507	0
A	27	4	0.148148148	100
A	6	2	0.333333333	99
A	93	1	0.010752688	24
A	27	0	0	0
A	98	1	0.010204082	17
A	106	8	0.075471698	1
A	18	6	0.333333333	20
A	68	29	0.426470588	13
A	11	1	0.090909091	18
A	28	1	0.035714286	24
A	32	19	0.59375	37
A	78	18	0.230769231	26
A	37	11	0.297297297	32
A	7	1	0.142857143	86
B	9	3	0.333333333	34
B	46	6	0.130434783	18
B	83	8	0.096385542	11
B	63	1	0.015873016	5
B	39	0	0	40
B	28	5	0.178571429	26
B	170	9	0.052941176	12
B	53	7	0.132075472	15
B	60	4	0.066666667	16
B	22	0	0	6
B	24	1	0.041666667	37
B	43	2	0.046511628	59
B	48	7	0.145833333	82
B	16	16	1	107
B	27	6	0.222222222	14
B	42	3	0.071428571	57
B	53	3	0.056603774	0
B	13	1	0.076923077	87
B	78	3	0.038461538	6
B	63	6	0.095238095	75
B	86	14	0.162790698	7
B	10	1	0.1	113
C	19	1	0.052631579	37
C	54	7	0.12962963	71

C	11	0	0	9
C	58	2	0.034482759	29
C	51	11	0.215686275	42
C	93	18	0.193548387	20
C	4	3	0.75	76
C	28	16	0.571428571	84
C	86	16	0.186046512	0
C	30	9	0.3	9
C	83	25	0.301204819	57
C	16	14	0.875	54
C	12	5	0.416666667	29
C	27	3	0.111111111	55
C	98	30	0.306122449	19
C	7	5	0.714285714	42
C	69	23	0.333333333	4
C	19	14	0.736842105	40
C	128	27	0.2109375	17
C	10	4	0.4	23
C	20	18	0.9	23
C	52	13	0.25	17

Appendix 3

Compilation of data on flower longevity from the three sites

Site	Flower longevity (in days)
A1	8
A1	9
A1	8
A2	8
A2	9
A2	8
A3	9
A3	8
A3	9
A4	9
A4	9
A4	9
A5	8
A5	8
A5	8
B1	9
B1	9
B1	9
B2	9
B2	9
B2	9
B3	10
B3	10
B3	9
B4	8
B4	8
B4	8
B5	10
B5	9
B5	9
C1	7
C1	7
C1	7
C2	6
C2	6
C2	6
C3	8
C3	6
C3	5
C4	6
C4	6
C4	6
C5	6
C5	6
C5	7