Spatial arrangement of
Phyteuma nigrum
and insect visitation

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SPATIAL ARRANGEMENT OF *PHYTEUMA NIGRUM* AND INSECT VISITATION

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Contents

Abstract

How distances between plants and inflorescences of *Phyteuma nigrum* affect pollinator behaviour was examined in a natural situation as well as experimentally. Patches of *Phyteuma* in two densities were set up in a field in Drenthe. In the natural population flight distances of bumblebees and syrphids were examined. Migration of pollinators was demonstrated between two populations of *Phyteuma*, which in previous studies had seemed completely isolated. The largest distance between recaptures of bumblebees on *Phyteuma* on the same day was 200 m. In the experimental patches bumblebees made longer foraging bouts in high-density patches than in low-density patches. They stayed longer per inflorescence and visited more inflorescences per plant in low density, while the number of inflorescences visited per minute did not differ between densities. In low density bumblebees flew more often to nearest neighbours and visited more inflorescences per plant. Plants with four inflorescences were preferred to plants with one in both densities, and the preference was stronger in low density. Implications for the fitness of the plants are discussed.

1 INTRODUCTION

For many plant species the behaviour of pollinators is very important for the fitness of both plants and populations. A lot of plant species depend on pollination by insects for seed set. The dispersal of pollen from one individual plant to the other is to a great extent responsible for the gene flow of a colony (Ellstrand, 1992).

According to optimal foraging theory, the pollinators will tend to maximize their net energy gain (Pyke, 1981). Flying between flowers costs energy spent in flying and time in which nectar could have been collected. Especially social insects which have to provide for the nest as well as for themselves are compelled to forage economically (Heinrich, 1979a).

The spatial arrangement of plants is a very important factor for pollinator behaviour (Manning, 1956; Beattie, 1976) and for gene flow (e.g. Levin & Kerstner, 1969a; Levin & Kerstner, 1969b; Schmitt, 1983). It has been found that pollinators prefer high-density patches to sparse patches (Manning, 1956; Thomson, 1981) or plants with many flowers to plants with few (Augspurger, 1980). Distances between flowers will influence the insects' choice of host plants and their flower fidelity (Anatovnics & Levin, 1980).

Dark Rampion, *Phyteuma nigrum* F. W. Schmidt (Campanulaceae), is a rare species in the Netherlands and has been declining for the last two decades (Weeda, 1989; Van der Meijden, 1990). Most remaining populations are small and sparse. The species is self-incompatible. These factors make it vulnerable to further decline and the service of pollinators is essential for the survival of the populations. The species' main pollinators are bumblebees (Kwak, 1991a), though other insect species like syrphids visit it as well (Van den Brand, 1991).

The present study was designed to examine in what way insect visitation on *Phyteuma nigrum* is affected by various distances between plants and inflorescences.

The first question was, how distances between patches and physical barriers influence the rate of switching between patches of bumblebees. This part of the study was carried out in a natural population of *Phyteuma nigrum* in the nature reserve 'Stroomdallandschap Drentsche Aa'. Earlier work had

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been done in the same population in 1989 and 1990 (Kwak et al., 1991a; Kwak et al. 1991b; Kwak,1993; Van den Brand, 1989).

For the second part of the study experimental patches of *Phyteuma* were used. The following questions were asked: how density affects a) the number of insects, and specifically bumblebees, that visit the plants, b) the visitation rate, and c) the frequency of intraplant flights. Finally the difference in pollinator behaviour towards plants with few (1) and many (4) inflorescences was examined.

2 MATERIAL AND METHODS

2.1 Plant species

The Dark Rampion, *Phyteuma nigrum* (Campanulaceae), is a perennial, outbreeding species (further called *Phyteuma*). It flowers in May-June. The inflorescences carry on average about 40 flowers each (Van den Brand, 1989) and a plant may have up to twenty inflorescences at a time, but usually less. The flowers are protandrous with secondary pollen presentation. Nectar is mainly present in the male phase (Kwak et al, 1991a).

The species is self-incompatible and depends on insects for pollination. The insects observed visiting *Phyteuma nigrum* in the observation area are: honey-bees (*Apis mellifera*), bumblebees (Bombus species), syrphids (Syrphidae), and other flies, butterflies (Lepidoptera) and solitary bees (Van den Brand, 1989).

The insects visit the flowers primarily for nectar (Kwak et al, 1991). Bumblebees and solitary bees often gather pollen and nectar simultaneously. Syrphids eat pollen and drink nectar (among others Van den Brand, 1989). Kwak et al (1993b) have shown that bumblebees are more effective pollen-vectors than syrphids. Solitary bees hardly touch the stigma during their visits, so they are probably not very effective as pollinators (Van den Brand, 1989). Butterflies are few in numbers and do not visit many inflorescences sequentially, nor visit many per hour. So, probably bumblebees are the most important pollinators of *Phyteuma nigrum*.

The Netherlands are the northernmost border of the species' distribution. The number of populations in the Netherlands has declined seriously during the last two decennia (Van der Meijden, 1990; Weeda, 1989).

2.2 Study Site

The first part of the study was carried out in the nature reserve of the Drentsche Aa near Taarlo. The nature reserve harbours natural populations of *Phyteuma nigrum*. Two of the populations were used in this study: 'Populierenlaan' and 'Meander'. Populierenlaan (PPL) is a fairly large population of about 6000 inflorescences (1993).
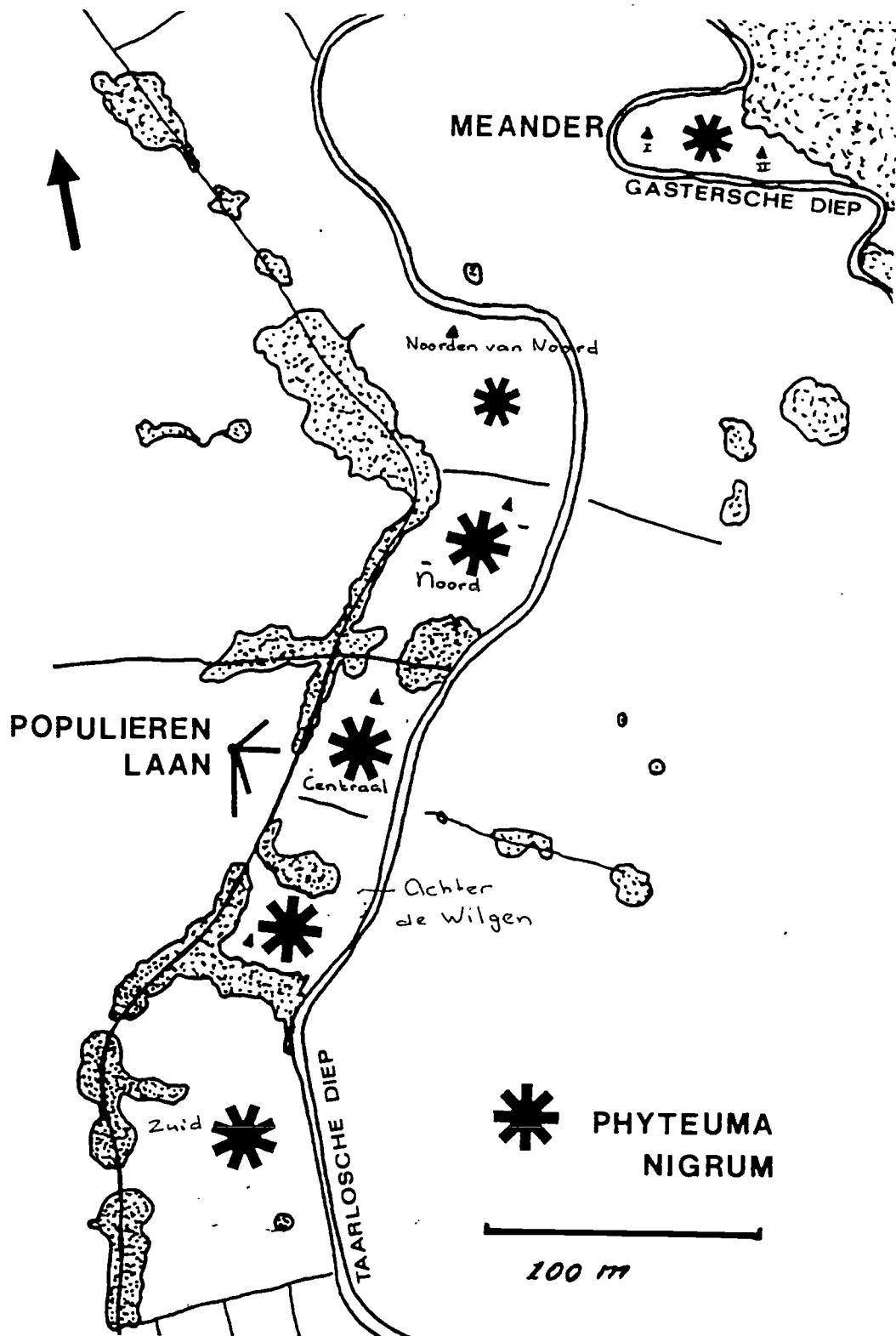
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Figure 1: Natural population of *Phyteuma nigrum* in nature reserve Stroomdallandschap Drentsche Aa near Taarlo. ▲ = location of quadrates for the counting of insects.
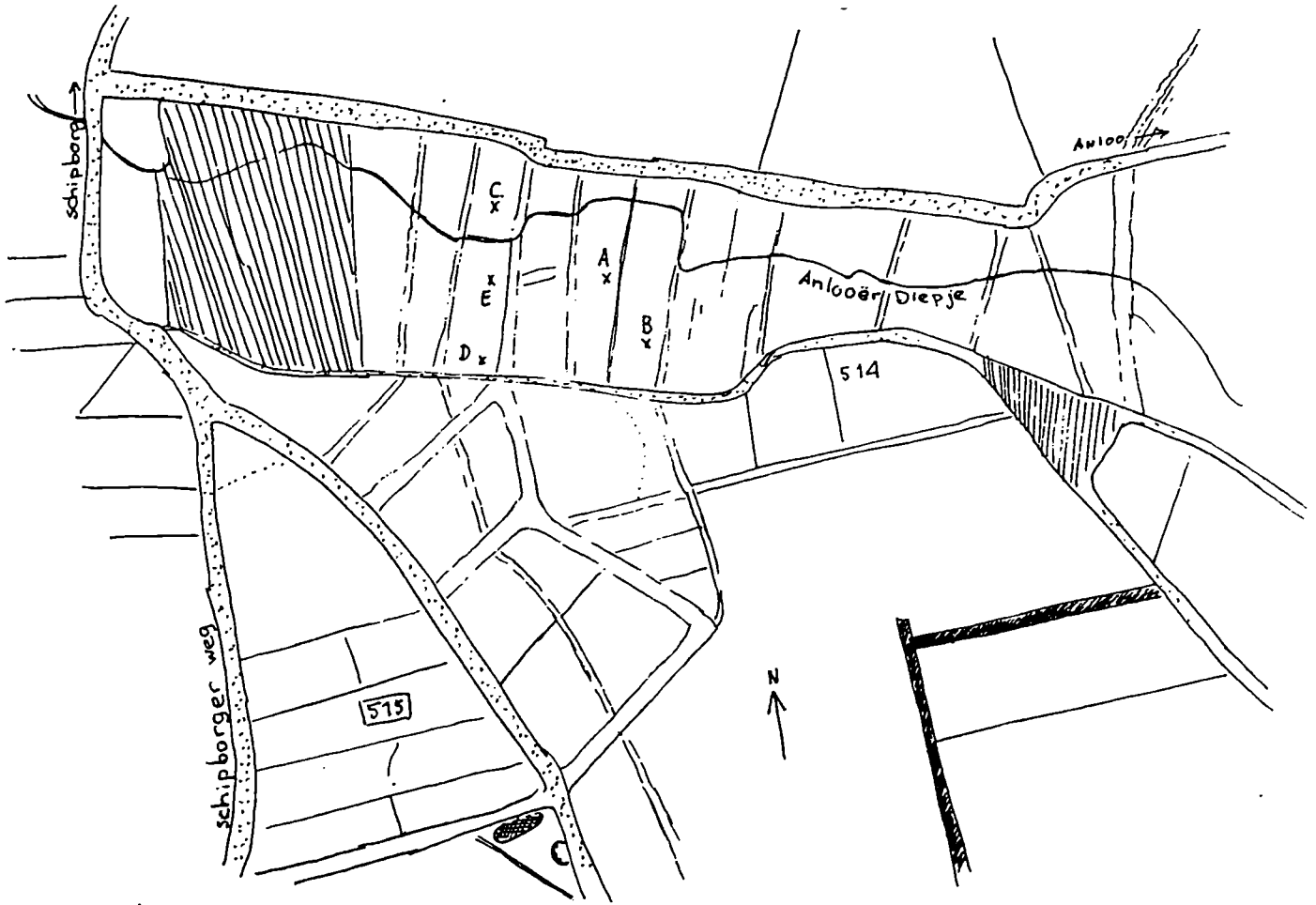
Figure 2. Position of experimental patches of *Phyteuma nigrum* in nature reserve Drentsche Aa near Anloo. A and C: high density, B and E: low density, D: intermediate density (See also Appendix 2).

Between different meadows in this part are ditches, usually overgrown with willow. Different meadows have been given their own names (Figure 1). Meander is a much smaller population: about 700 inflorescences (1993). It is enclosed in a loop of the brook Gastersche Diep. In a recent expansion of PPL, named 'Noorden van Noord' (NvN), there are about 165 inflorescences (1993). Their distribution here is rather sparse.

Meander is situated north of Populierenlaan. Between the two populations are two brooks and a field without pollinator-attracting flower species. The shortest distance between the two populations is about 100 metres (Figure 1).

The second part of the study was carried out in another part of the nature reserve of the Drentsche Aa, near Anloo (Figure 2). This part of the reserve comprises several former meadows, which are today extensively managed by StaatsBosBeheer. The dominant plants during our work there were *Rhinanthus angustifolius* and *Juncus acutiflorus*. *R.angustifolius* is found in different densities in the different meadows, due to differing periods of management by StaatsBosBeheer. *Rhinanthus angustifolius* is an important nectar and pollen providing plant for the bumblebees. *Phyteuma nigrum* is not found here.

2.3 Methods

2.3.1 Natural population

Observations in the natural populations were made from 27 April till 27 May 1993. Temperatures were between 18 and 25 °C and it was dry on most days.

The field was divided in 10m x 10m squares with small sticks on the corners. This provided the means to note accurately where observations were made.

To be able to follow the movements of individual bumblebees, bumblebees were marked with fast drying paint. New bumblebees were marked during the whole observation period. The species, caste, day, time, position in the field and plant species on which the bumblebee foraged were noted if a bumblebee was (re)captured.

At least three days per week observations were made from about nine a.m. till about six p.m.; in total more than 460 man-hours. Observations were made while the observers walked set routes through the field.

On 12 May individuals of *Rhingia campestris* (Syrphidae) were caught on *Phyteuma nigrum* in six of the 10x10 meter

squares. Three of these squares were located in PPL, one in
NvN and two in Meander (Figure 2). *Rhingia* caught in each
square were marked with a dot of fast drying paint and set
free. Each square got its own colour. Marking was repeated on
24 May with other colours in one of the squares in PPL and one
in Meander. The marking was done to obtain an indication of
the distances flown by *Rhingia*.

In population PPL two quadrates of 2m x 5m were
established, one of which in the section NvN, and one in
section Noord. In Meander two similar quadrates were situated.
In these four quadrates the number of flowering spikes of
Phyteuma nigrum were counted on observation days. Also the
total numbers of insects visiting *Phyteuma nigrum* in these
quadrates during five minutes were counted at least once per
observation day.

2.3.2 Experimental patches

Observations for the second part of the study were made
from 7 to 30 June 1993. A survey of the activities and the
weather can be found in Appendix 1.

Artificial patches of *Phyteuma nigrum* were constructed in
two densities *Rhinanthus angustifolius*. The higher density *R.
angustifolius* contained about 150 inflorescences of *R.
angustifolius* per m², the lower density about 50 at the top of
its flowering time.

In both densities *R. angustifolius* a high-density patch
and a low-density patch of *Phyteuma* were constructed. For some
experiments a third patch of intermediate density was
established in the lower density *R. angustifolius*. In the
higher density *R. angustifolius* the high-density patch of
Phyteuma, called A, was 80 m removed from the low-density
patch, called B. In the lower density *R. angustifolius* the
distance between the high-density patch (C) and the low-
density patch (E) was 55 m. The intermediate-density patch (D)
was 65 m removed from the low-density patch (Figure 2).

The experimental patches of *Phyteuma* consisted each of 36
Phyteuma plants in pots. These plants were grown in a
greenhouse. The patches were triangular and the pots were
uniformly distributed (Figure 3). In the high density, the
distance between pots, centre-centre, was 0.20 m. In the
intermediate density the distance between pots was 0.6 m, in
the low density 2.0 m. A survey of densities and locations of
the patches can be found in Appendix 2.

From 7 through 16 June plants were used with 3
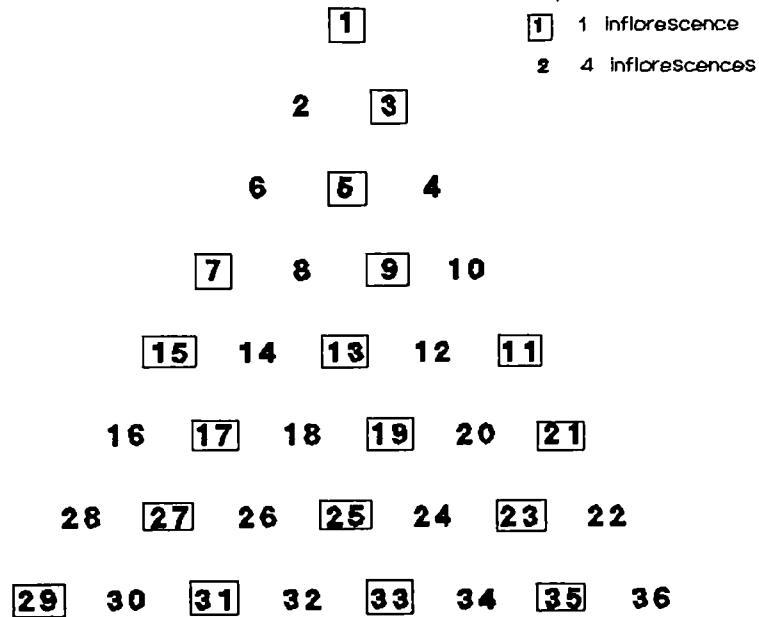inflorescences each, or two or one when inflorescences had

Figure 3. Position of *Phyteuma* plants within a patch. Distance
between pots is 2.00 m in low density and 0.20 m in high
density.

withered, so maximal 108 inflorescences were present. From 21
June on plants with one inflorescence (also called 'one-spiked
plants') were used at the odd numbers and plants with four
inflorescences ('four-spiked plants') at the even numbers (see
Figure 3). This way there were 72 inflorescences on four-
spiked plants and 18 inflorescences on one-spiked plants, in
total 90 inflorescences. Withered inflorescences were replaced
before observations started.

On 8, 10, 11, 16 and 30 June insects on *Phyteuma nigrum*
were counted in the different patches. On 8, 10 and 30 June
census was carried out in the patches situated in the lower
density *R. angustifolius*. On 11 and 16 June insects census was
carried out in the higher density *R. angustifolius*. The
counting was done following the principle of 'point
observation': each inflorescence was looked at for an instant
and the presence of insects was noted. For half an hour
insects were counted in one patch, then half an hour in the
other, doing eight rounds per half hour. Per observation day
seven sets of eight rounds were made per patch. In low-density
patches also the bumblebees on *R. angustifolius* were counted
for comparison.

On 21, 22, 28 and 29 June records were kept of the
movements of individual, marked bumblebees on *Phyteuma nigrum*
in the patches C and E (and D). Since all inflorescences were

numbered with small labels, track could be kept of all
inflorescences the insect visited. The total time from the
first visit till the insect left the patch was clocked.

In the low-density patch E, pollinators have to fly
longer between plants than in the high-density patch. This may
affect the visitation rate, so on 22 June we also measured how
long the bumblebees stayed at individual inflorescenses in
patches C and E.

2.4 Data analysis

Observations of marked bumblebees in the natural
population were ordered in a spreadsheet. Number of flights
between the subpopulations, between the fields etc were
counted without further statistical analysis.

The results from the census in the experimental patches
were analyzed using MANOVA.

The data from the records were used in several analyses.
The differences in bumblebee behaviour between densities with
respect to the lengths of foraging bouts, the number of
inflorescences visited per minute and number of plants visited
per minute were examined. Looking only at plants with four
inflorescences, the numbers of heads visited sequentially a
row per plant in high and low density were compared. MANOVA
was used for all analyses. The time spent at individual
inflorescences in high and low density was compared using
MANOVA as well.

In many papers it is assumed that the bumblebees will
minimize their energy costs while foraging (e.g. Pyke, 1981).
Since flying from plant to plant will cost more energy and
time in a low-density patch, it was examined whether the
bumblebees were more apt to visit the nearest neighbour in the
low-density patch than in the high-density patch. To calculate
the 'economy coefficient' the following method was used. All
flights between inflorescences were assigned a value. Flights
within the same plant received value 0, flights between
nearest neighbours 1 etcetera. For longer flights the smallest
number of plants passed to reach the target plant were
counted. For an example see Figure 4. These values were
averaged over the total number of flights between
inflorescences made during that bout. This was done for at
least 15 bouts in patch C and E each on 21 June as well as on
22 June. Wilcoxon Rank Sum Test was used for analysis. The
same was done neglecting flights between inflorescences on the
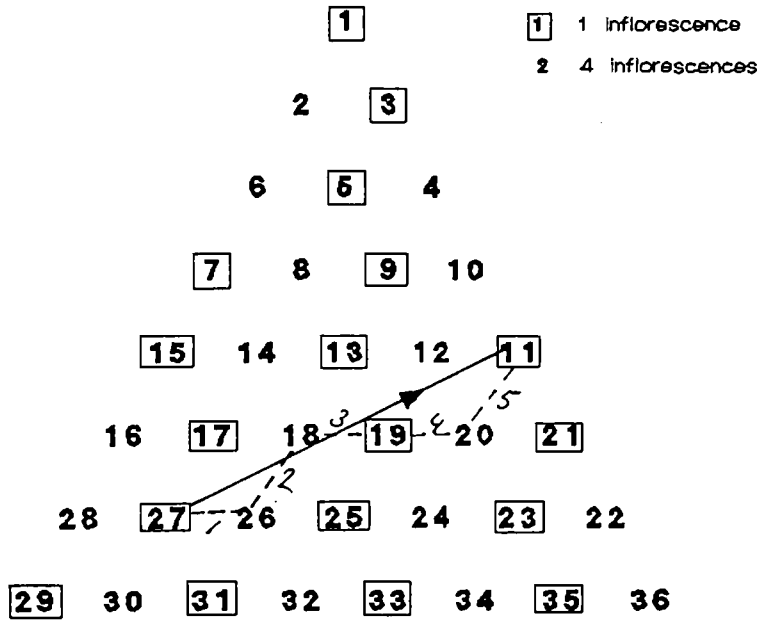same plant. This method overestimates the real flight

Figure 4. Calculation of 'economy coefficient'. ——— flight route, ----- steps in calculation. A flight from plant 27 to plant 11 gets the value 5, because it takes five imaginary steps.

distances, but is much simpler than the accurate method.

For the differences in behaviour towards plants with four and plants with one inflorescence, I compared the time spent at inflorescences on four-spiked plants and on one-spiked plants with MANOVA.

Probabilities less than 0.05 are assumed significant.

3 RESULTS

3.1 Flowering phenology and visitation of *Phyteuma nigrum*

The first plants of *Phyteuma nigrum* in the natural population near Taarlo flowered at about 4 May, the last around 30 May. The phenology in different parts of the population can be seen in Figure 5. The very sheltered part of subpopulation Meander had its peak at 14 May. The rest of Meander and Populierenlaan (PPL) had their peak almost a week later. During the first phase of flowering of *Phyteuma*, other plant species attracting insects were *Glechoma hederacea* and *Ajuga reptans*, at the end *Lychnis flos-cuculi* and *Rhinanthus angustifolius*. In Meander there was a patch of *Lamium album* which attracted many bumblebees.

Phyteuma was the most favoured plant by bumblebees during the period of the observations. Its share in the total visits can be seen in Figure 6 together with the shares of *Ajuga reptans* and *Rhinanthus angustifolius*. The relation between the share *Phyteuma* had in the visits and its phenology is depicted in Figure 7. In this figure it can be seen that the 'popularity peak' precedes the flowering peak. The same was found in the counts of all insects, not only bumblebees, in the quadrates, as significantly more insects visited the inflorescences before than after the flowering peak in the observed quadrate (t-test, $p < 0.05$).

3.2 Recaptures in the natural population

In Populierenlaan a total of 352 bumblebees was marked, in Meander 52. These bumblebees had been captured on *Phyteuma* as well as on other plant species. From the bumblebees marked in PPL 137 were seen more than once and from the bumblebees marked in Meander 19 were seen again. For both populations this is about 35%. More than 1800 observations were collected. From these observations about 800 were useless for this study, because some information was missing. Usually the identification number was missing, because the bumblebees brushed the mark off. 658 useful observations were left of bumblebees that were spotted more than once.

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Phyteuma nigrum
development of flowering
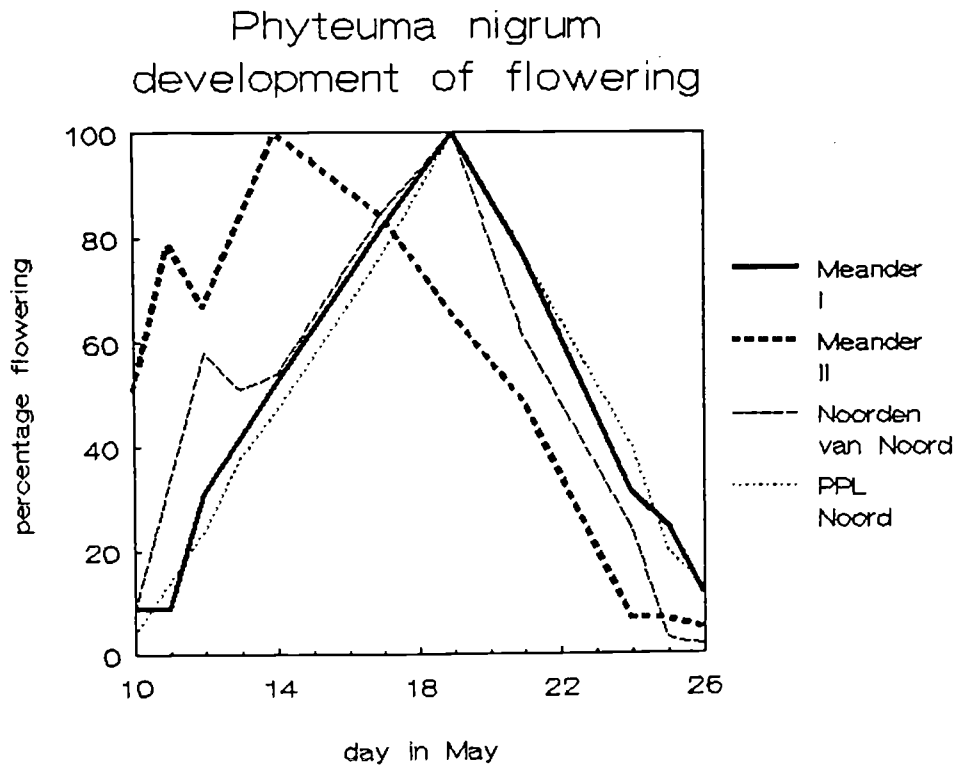
Figure 5. Flowering curve of *Phyteuma nigrum* in several parts of the population. May 1993.

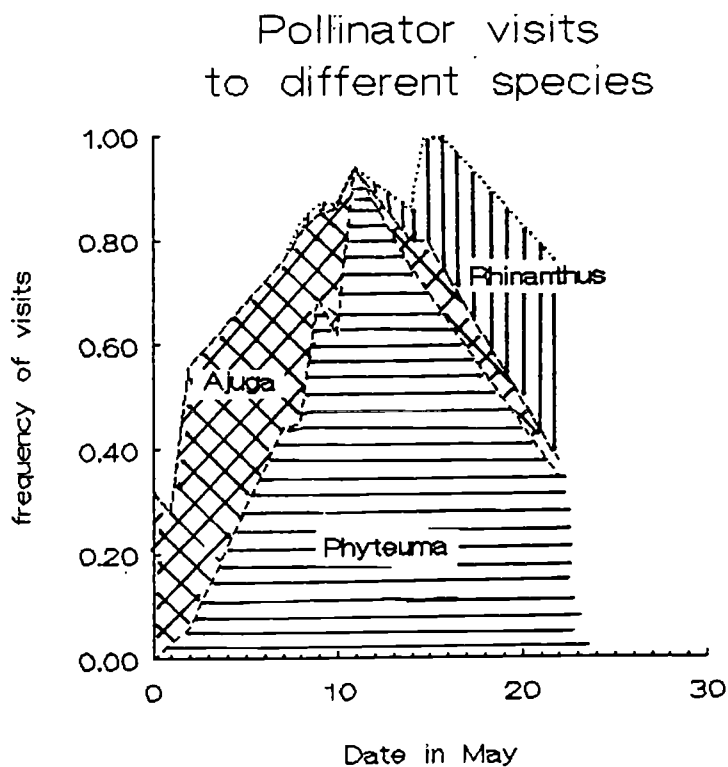## Pollinator visits
to different species

Figure 6. Share of *Phyteuma nigrum*, *Ajuga reptans* and *Rhinanthus angustifolius* in the visitations of bumblebees.

Flowering curve and pollinator visits
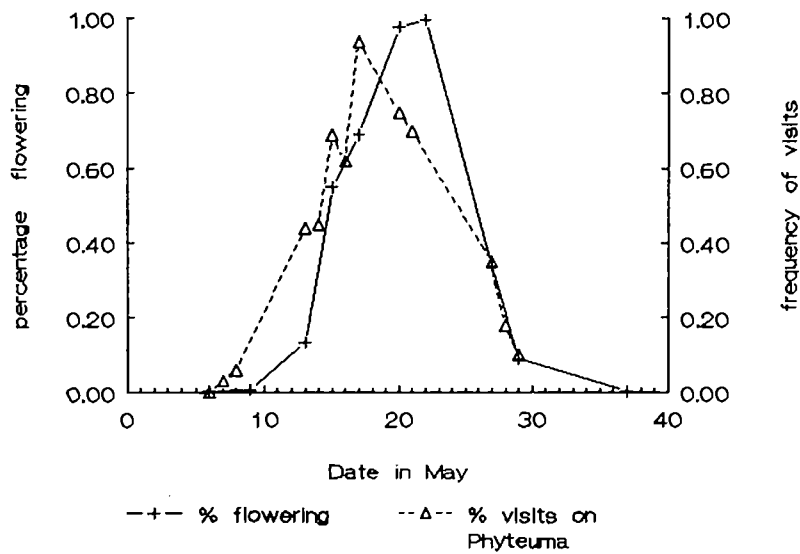## Phyteuma nigrum

Figure 7. Share of *Phyteuma nigrum* in the vistations of the bumblebees in relation to its flowering curve.

3.2.1 Bumblebee exchange between PPL and Meander

During earlier work in this area no bumblebees were found that visited both Meander and PPL. This year 14 individuals were found that did. Only one individual, a *B. pratorum* worker, was seen on the same day in both populations on *Phyteuma*. The distance between observations was 170 m. One more bumblebee was seen on *Phyteuma* in both populations but on different days. One *Bombus pascuorum* queen flew from PPL (Achter de Wilgen) to Meander and back on the same day. This trip was 430 metres there and 430 m back as the crow flies. In Meander this individual was seen on *Lamium album*, in PPL on *Rhinanthus angustifolius*. Another *B. pascuorum* queen was seen on the same day first in Meander on *Lamium album* and later in PPL on *Phyteuma*. From one spot to the other it was 350 m. Eight bumblebees were found on *Phyteuma* in PPL, but in Meander on *Lamium album* or *Lychnis flos-cuculi*. This was on different days. Three were found on *Phyteuma* in Meander, but on other species in PPL, like *Ajuga reptans* and *Symphytum officinale*. This too was on different days. One bumblebee was found on *Rhinanthus angustifolius* in both populations on the same day. Summarizing only one bumblebee out of all our observations was found that served both *Phyteuma* populations
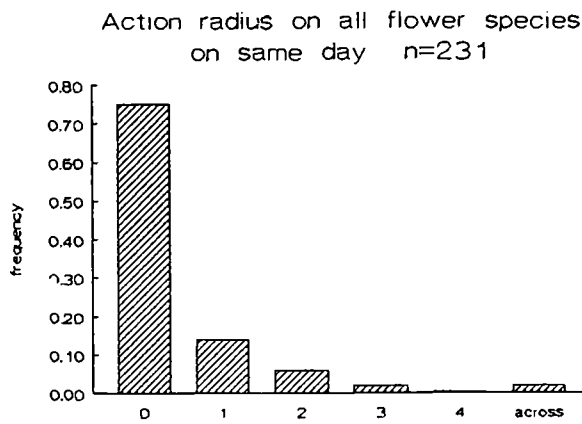
Action radius on all flower species
on same day   n=231

frequency

0.80
0.70
0.60
0.50
0.40
0.30
0.20
0.10
0.00

0   1   2   3   4   across

Action radius on Phyteuma
on same day   n=98

frequency

0.80
0.70
0.60
0.50
0.40
0.30
0.20
0.10
0.00

0   1   2   3   4   across

Figure 8. Number of fields
bumblebees moved on the
same day between recaptures on
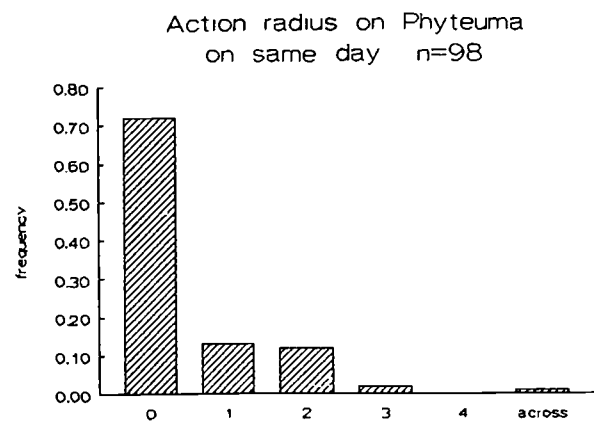áll flower species. 'Across'is
from Meander to PPL or back.

Figure 9. Number of fields
bumblebees moved between
recaptures on the same day on
Phyteuma. 'Across' is from
PPL to Meander or back.

on the same day and that might have cross-pollinated the
populations.

3.2.2 Bumblebees, action radius

There were 231 observations of bumblebees seen more than
once on the same day during the flowering season of Phyteuma.
In this study the shortest distance between two recaptures on
the same day has been called 'action radius' or 'trip'. The
bumblebee may of course have gone anywhere between the two
observations. Of the 231, 102 times recaptures on the same day
were between Phyteuma nigrum inflorescences, that is 44%. 27
were from Phyteuma to other flower species and 23 were from
other flower species to Phyteuma, together 22%. The 79 trips
left were between not-Phyteuma species. If these observations
reflect flight behaviour, about two thirds of the flights
involving Phyteuma is between conspecifics, and can result in
conspecific pollen transfer and achieve fertilization.

From the 231 trips on the same day 75% were within the
same field. The other trips were between fields and the
bumblebees had to cross one or more ditches with willow growth
(Figure 8). There were 17 trips within PPL on the same day
that exceeded 100 m. From these long trips eight were between
Phyteuma and three from another plant species to Phyteuma.
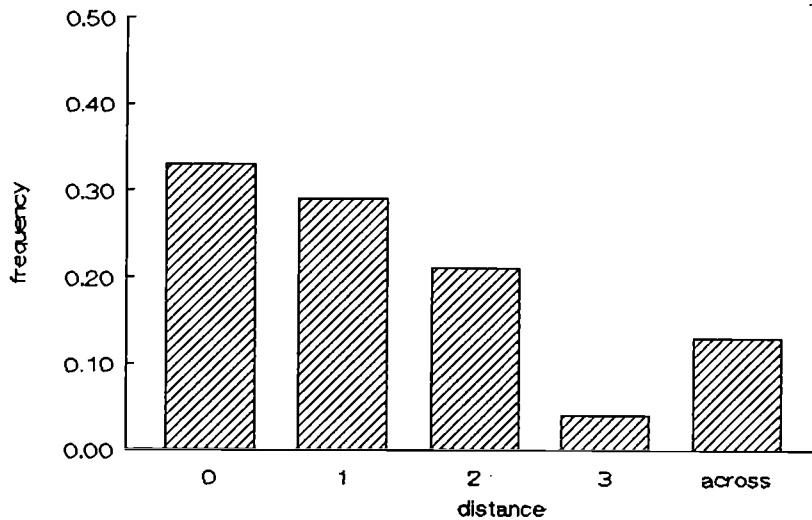
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action radius of Rhingia campestris
n=24

Figure 10. Number of fields *R. campestris* individuals moved between marking site and recapture. n=24.

Five of the trips were between PPL and Meander. The largest action radius on one day was in PPL and was 360 m. This trip was from *G. hederacea* to *Phyteuma*.

When we consider only trips involving solely *Phyteuma*, the same pattern of distances is found as for all trips (Figure 9). About 200 m was the largest action radius of bumblebees foraging on *Phyteuma*. Between days the largest distance between recaptures on *Phyteuma* was 390 m.

Because of too few observations of visits to *Phyteuma* after 15 May it is not possible to pronounce upon changes in time in the proportions of trips within and between fields. For the same reason nothing can be said about the fidelity of bumblebees to *Phyteuma* in time.

3.2.3 Syrphid exchange between Populierenlaan and Meander

On 12 May we marked 126 *Rhingia campestris* captured on *Phyteuma*. The syrphids received a colour code according to the 10 m x 10 m quadrate they were captured at (Appendix 3). Twenty-four times a marked syrphid was recaptured. On 14 May, two days after marking, twice an individual, marked in PPL (Noord) was seen in Meander on *Phyteuma*. The distance between the place where it was marked and where it was found in Meander is about 150 m. It had to fly across two brooks. A

syrphid marked in Meander was found in PPL (Centraal) on
Phyteuma on 24 May, that is 12 days after marking. The
distance between the two spots is 240 m.

3.2.4 Action radius of syrphids

The longest distance travelled by *Rhingia campestris* from
one day to the next was two fields, about 100 metre. In Figure
10 the it can be seen how many fields the syrphids shifted.
These are distances measured from the place of marking and
there may be several days between one observation and the
next. Moreover, because there were but 24 observations it may
not be representative for the behaviour of *Rhingia campestris*.
It seemed that the syrphids spread slowly from the site of
marking, but because there were but few observations this is
not sure.

On 24 May again eleven *Rhingia campestris* were marked,
but *Phyteuma* had almost finished flowering and most
recaptures of these syrphids were on *Glechoma hederacea* and
Lychnis flos-cuculi.

3.3 Experimental patches

3.3.1 Number of insects in different densities

The results of the census are summarized in Table 1.
The insects found were bumblebees (*B. pratorum*, *B.
lapidarius*, *B. jonellus*, *B. hortorum*, *B.pascuorum*, *B.
hypnorum*), small solitary bees (mainly *Andrena angustior*),
syrphids (*Rhingia campestris* and other species), butterflies
(Satyridae) and a few miscellaneous insects. Because the
experiment was done after the normal flowering time of
Phyteuma these may not be the usual visitors.

Combining the census in A and B, and C and E, no
significant difference in number of insects per round per
inflorescence between the patches with high and low density of
Phyteuma nigrum could be found (Manova, p=0.157). For the
patches in the lower density *R. angustifolius*, C and E, also
no significant difference in number of insects on *Phyteuma* was
found (Manova, p=0.263), nor for the patches in the higher
density of *R. angustifolius*, A and B (Manova, p=0.316).

The most important pollinators of *Phyteuma* are
bumblebees, so it may be interesting to look at these insects
separately. Considering all four patches together, no
significant difference in the number of bumblebees per round

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per inflorescence could be found between the dense and sparse patches (Manova, p=0.066). In the lower density *R. angustifolius*, patches C and E, also no significant difference between the densities could be found (Manova, p=0.423). In the higher density *R. angustifolius*, patches A and B, significantly more bumblebees were counted in the dense patch A (Manova, p<0.001).

How is it with the other insects? There are significantly more of them in the sparse patches, if all four patches are combined (Manova, p<0.001). This is also the case when A and B, and C and E are viewed independently (Manova, p<0.001 and p<0.01 respectively). Possibly these insects are more or less uniform distributed over space. Combined with a preference for *Phyteuma*, this might cause the observed differences between high and low density *Phyteuma*. A preference for *Phyteuma* is not unlikely since insects other than bumblebees cannot reach the nectar in *Rhinanthus angustifolius*.

Patches	Mean number of insects/round/inflorescence		Probability of equal means
	High density *P. nigrum*	Low density *P. nigrum*	
all insects			
A & B	0.0019	0.0022	p=0.316
C & E	0.0032	0.0035	p=0.263
All	0.0026	0.0029	p=0.157
bumblebees			
A & B	0.0024	0.0015	p<0.001 ***
C & E	0.0037	0.0032	p=0.423
All	0.0031	0.0025	p=0.066
not-bumblebees			
A & B	0.0015	0,0027	p=0.001 ***
C & E	0.0028	0.0037	p=0.010 *
All	0.0023	0.0033	p<0.001 ***

Table 1. Mean numbers of insects per round per inflorescence in two densities *P. nigrum*. For explanation of patches see text and Appendix 2. Manova.

Because census in higher and lower density *R. angustifolius* was not performed on the same days, the outcomes cannot be compared very well. This is because the numbers of insects differed significantly between days. The changes in

numbers of bumblebees are probably due to the phenology of *R. angustifolius* (see 3.3.2), the changes in numbers of other insect are probably due to changes in the weather conditions.

On 30 June insects were counted in three different densities *Phyteuma*, in patches C (high), D (intermediate) and E (low), following a slightly different procedure (Table 2). Again most insects were found in the lowest density (Manova, p<0.005). Between intermediate and high density no differences were found. Numbers of bumblebees did not differ significantly between the densities (Manova, p=0.054). Numbers of not-bumblebees were significantly higher in the low-density patch, but did not differ significantly between high density and intermediate density.

| | Mean number of insects/round/inflorescence | | |
	High	Inter-mediate	Low
all insects	0.0017	0.0017	0.0024
bumblebees	0.0016	0.0011	0.001
not-bumblebees	0.0019	0.0024	0.0038

Table 2. Number of insects per round per inflorescence on 30 June. Separate underlining means significantly different values.

3.3.2 Number of insects in time

The number of bumblebees on *R. angustifolius* in the low-density patches was counted simultaneously with the insects on *Phyteuma* (Figure 11). The flowering of *R. angustifolius* in the lower density declined from 54 inflorescences/m^2 with on average 3.5 flowers per inflorescence on 7 June to 31 inflorescences/m^2 with on average 1.8 flowers per inflorescence. This must have influenced the behaviour of bumblebees. The number of bumblebees on *R. angustifolius* declined as did the flowering of *R. angustifolius*. The number of bumblebees counted on *Phyteuma* declined also in time as can be seen in Figure 12. The number of other insects on *Phyteuma*
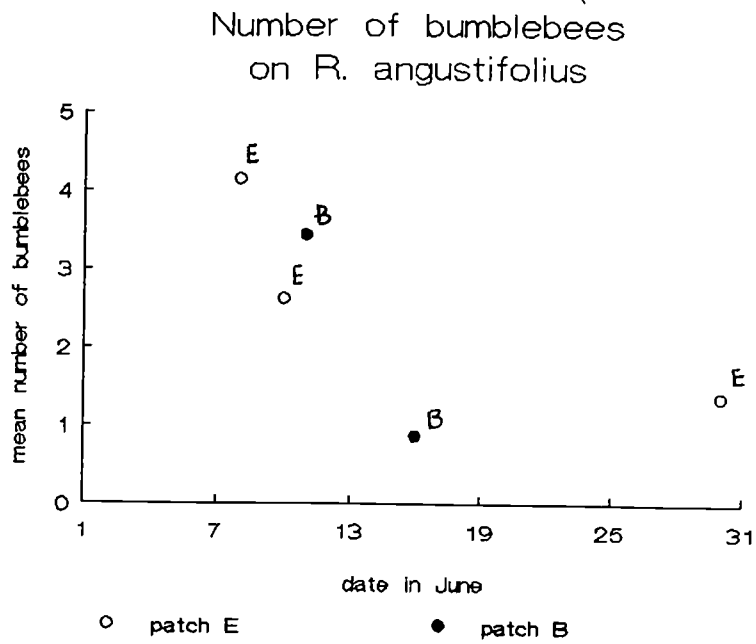
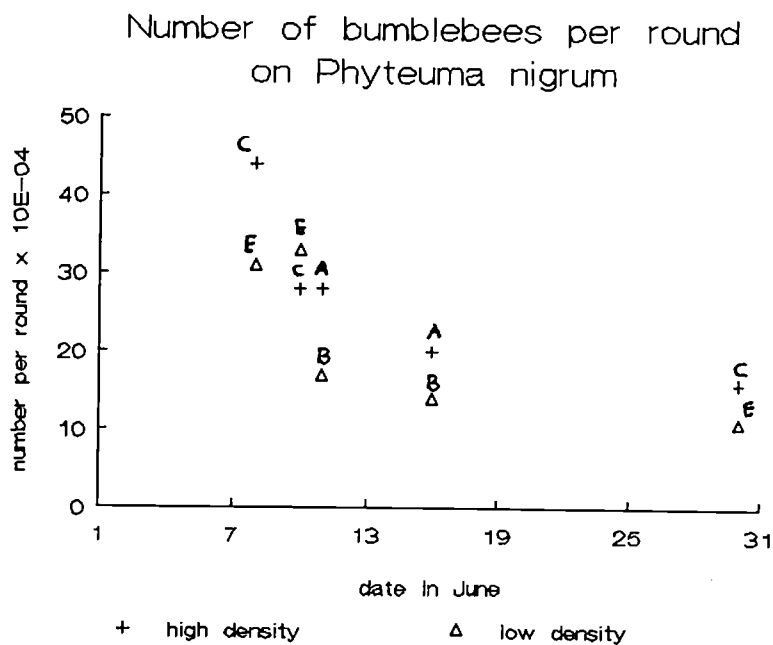Figure 11. Number of bumblebees counted on R. *angustifolius* in the low-density patches B and E.

Figure 12. Number of bumblebees counted on P. *nigrum*.

Number of not—bumblebees per round
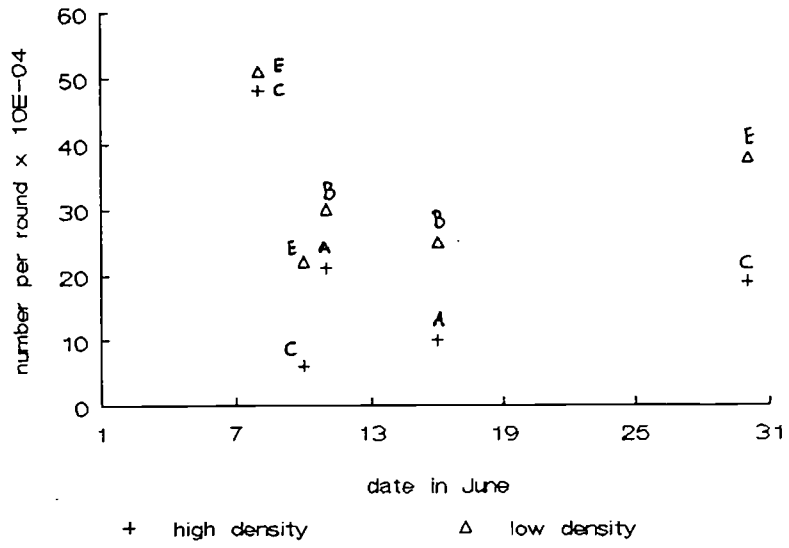## on Phyteuma nigrum

Figure 13. Number of not-bumblebee species counted on *P. nigrum*.

per inflorescence fluctuated more and showed no real decline or increase in time (Figure 13).

3.3.3 Length of foraging bouts

The time a bumblebee foraged in a patch of *Phyteuma* is called a foraging bout on *Phyteuma*. For comparison of boutlengths in high and low density *Phyteuma* only complete foraging bouts were used. Foraging bouts over all days were significantly longer in high density of *Phyteuma* (Manova, p<0.001 n=197). In high density the foraging bouts were on average 5'52"+5'21" (n=95). The longest bout was over 23 minutes and comprised 88 inflorescences. During this bout, one of the 90 inflorescences was visited seven times, and five not at all. In low density the length of a foraging bout was on average 2'27"+2'36" (n=102). Remarkable was that on 29 June the bouts in high density were very long, 10'20" on average, while the bouts in low density were very short, 0'34" on average. The lengths of foraging bouts differed significantly between days (Manova p<0.001).

3.3.4 Inflorescences and plants visited per minute

If the next inflorescence is far away, it may be economical for the bumblebee to stay longer on a not very rewarding inflorescence or plant. Also when much time is lost in flying, less inflorescences can be visited in the same time if time spent per inflorescence is equal. So density may influence the number of inflorescences and plants visited per unit of time.

Per foraging bout, I divided the length by the number of inflorescences visited or the number of plants visited. Bouts of less than three inflorescences were not taken into account, because a small error in time measuring will have grave consequences.

The number of inflorescences visited per minute foraging did not differ significantly between high and low density *Phyteuma* (Manova, p=0.072 n=156). The number of plants visited per minute foraging however did differ significantly (Manova, p=0.001 n=179). The number of plants visited per minute was higher in high density. This suggests that more inflorescences are visited per plant in low density. I will go into this later. The number of inflorescences and plants visited was significantly different between days (Table 3).

3.3.5 Time spent per inflorescence

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On 22 June we measured the time spent on inflorescences from the moment the bumblebee alighted till it took of. In low density the bumblebees stayed significantly longer on an inflorescence than in high density (Manova, p=0.005 n=207) (Table 3).

(When compared to the number of inflorescences visited per minute in low density in Table 3, the time spent per inflorescence is impossibly long. This is probably due to sampling errors.)

	High density (C)	Low density (E)	Probability
Inflorescences/minute	4.837	4.371	p=0.072
Plants/minute	3.365	2.580	p=0.001 ***
Seconds/inflorescence	10.37	16.62	p=0.005 **

Table 3. Visits per minute to inflorescences and whole plants. Time spent per inflorescence. All in patches C and E in lower density of *R. angustifolius*. Manova.

3.3.6 Economy of flight

Since the distances between nearest neighbours in low density were 10 times as large as in high density, bumblebees were expected to visit nearest neighbours more often in low density. 'Economy coefficient' was calculated per individual bout. Bouts in high and low density were compared. Visits to the same plant (0), the nearest neighbour (1), the next nearest (2) etc. were distributed as depicted in Figure 14a. The bumblebees flew more economically in low density *Phyteuma* (0.66+0.51, n=31) than in high density (1.34+0.41, n=35) as expected (Wilcoxon Rank Sum, p<0.001). When the flights between inflorescences on the same plant were left out of the calculations, ergo only flights between different plants were taken into account, the 'economy coefficient' was still significantly lower for the low density (1.38+0.36) than for the high density (1.87+0.44) (WRS, p<0.001, Figure 14b).

3.3.7 Visits to plants with one and four inflorescences

The number of inflorescences per plant may also influence the behaviour of the bumblebees. If a plant has more inflorescences the bumblebee may collect more nectar without having to fly to a next plant. In our experimental patches
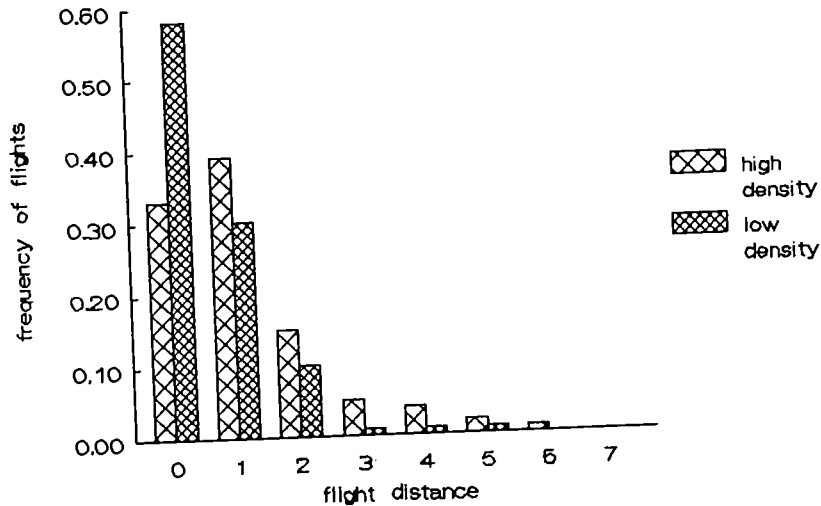
Figure 14a. Standardized flight distances of bumblebees in low and high density of *P. nigrum* on 21 and 22 June. Flights within plants included. Maximum distance is 7. See text.
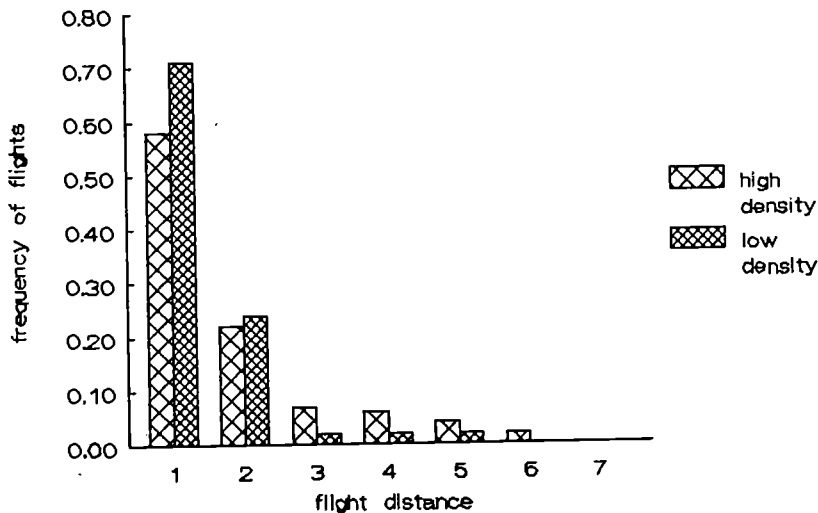
Figure 14b. Standardized flight distances of bumblebees in low and high density of *P. nigrum* on 21 and 22 June. Only flights between plants included. See text.

half of the plants had one inflorescence and half had four inflorescences (Figure 3). The number of visits to inflorescences of four-spiked plants and of one-spiked plants were counted. To see whether bumblebees visited plants with one inflorescence more or less often than plants with four, also the number of visits to whole plants were counted. Detailed results are in Table 4a and b. Inflorescences on four-spiked plants received 0.48 as many visits as

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inflorescences on one-spiked plants in high density (n=2173).
In low density the inflorescences on four-spiked plants
received 0.92 as many visits as inflorescences on one-spiked
plants.

Date	High density		Low density	
	1 head	4 heads	1 head	4 heads
21 June	167	385	89	376
22 June	190	420	72	198
28 June	158	187	55	179
29 June	230	436	39	90
	—	—	—	—
Total	745	1428	255	943

Table 4a. Number of observed visits to inflorescences on one-
and four-spiked plants.

Date	High density		Low density	
	1 head	4 heads	1 head	4 heads
21 June	168	229	84	131
22 June	196	222	68	84
28 June	156	112	58	69
29 June	272	302	38	35
	—	—	—	—
Total	792	865	248	319

Table 4b. Number of observed visits to plants with one and
four inflorescences.

In high density bumblebees had a small, but significant
'preference' for four-spiked plants (testing a proportion,
$p(z) < 0.05$ n=1657). The four-spiked plants received 1.09 as
many visits as the one-spiked plants. In low density the
bumblebees also had a preference for four-spiked plants. The
four-spiked plants received 1.29 as many visits. This
preference is also significant ($p(z) < 0.001$ n=567). The
preference for four-spiked plants in low density is
significantly stronger than in high density ($p(z) = 0.045$, one-
sided). Recapitulating, plants with four inflorescences
receive more visits than plants with one inflorescence. The
inflorescences on four-spiked plants individually receive less
visits than on one-spiked plants, though.
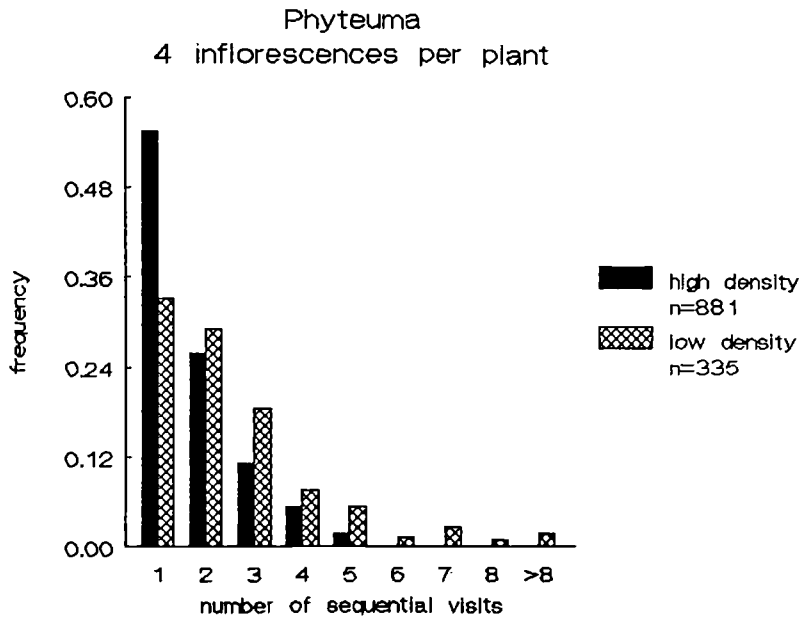
Phyteuma
4 inflorescences per plant

Figure 15. Number of inflorescences visited sequentially per plant with four inflorescences in two densities of *Phyteuma*.

3.3.8 Number of inflorescences visited per plant

Since *Phyteuma* is self-incompatible, visits between inflorescences on the same plant will be not effective or less effective than visits between inflorescences on different plants. How many inflorescences are visited in a sequence on the same four-spiked plant is therefore an interesting question.
In the high-density patch of *Phyteuma* 1.73 inflorescences on average are visited in a row on the same four-spiked plant. In the low-density patch 2.57 inflorescences are visited sequentially on average (Figure 15). The distribution of number of visited inflorescences is leptokurtic.

3.3.9 Time spent per inflorescence (2)

On one-spiked plants the bumblebees stayed 15.5 seconds per inflorescence per visit on average, on four-spiked plants 11.9 seconds. The difference is not significant (Manova, p=0.059, n=227).

3.3.10 A model for foraging behaviour

With the measured proportions of bumblebee visits to one- and four-spiked plants and the proportion of flights between inflorescences on the same plant a model for foraging behaviour can be constructed. With this model it is possible

to check for internal contradictions in the data.

definitions:

Proportion of visits to four-spiked plants	x
Proportion of visits to one-spiked plants	1-x
Proportion of flights within a plant	p

help variables:

Number of plants visited	N
Number of flights	F

Number of four-spiked plants	xN
Number of flights between plants	$N = (1-p)F$
Number of flights within a plant	$pF = N*p/(1-p)$

Number of inflorescences visited per four-spiked plant
$$1 + p/(x(1-p))$$

Total number of inflorescences visited on four-spiked plants
$$\{1 + p/(x(1-p))\} * xN$$

Total number of inflorescences visited on one-spiked plants
$$(1-x)N$$

Ratio of visits per inflorescence on four-spiked and one-spiked plants is

$$\frac{x+p/(1-p)}{4}/(1-x)$$

If the model is right the answer is the same as the proportion found by dividing the total number of visited inflorescences on four-spiked plants by the total number of visited four-spiked plants.

In high density $x = 0,52$ (Table 4b) and $p = 0,295$ (as in Figure 14a, but for all observation days). The number of visited inflorescences per four-spiked plant would be 1.80. This is the same as the value found by dividing all visited inflorescences on four-spiked plants by all visited four-spiked plants. The counted number of visited inflorescences per four-spiked plant is however 1.73 (3.3.8).

   The ratio of visits is 0.49 according to the model and 0.48 (3.3.7) when calculated directly. For some of the

calculated values the records of 17 June were left out because of some trouble in interpreting them. This may have caused the differences in outcomes. Especially so, because of the significant differences in behaviour between days.

In low density $x = 0.57$ and $p = 0.53$. The number of inflorescences visited per four-spiked plant is 2.98 according to the model. Direct calculations as described before give a value of 2.57 which is the same as the counted number (3.3.8). Here the difference between the model and the direct calculations is somewhat larger.

The ratio of visits is 0.98 according to the model and 0.92 (3.3.7) according to direct calculations. All in all the different measurements do not contradict each other too much according to this model.

If we neglect the differences in outcomes, we can summarize the bumblebees' foraging behaviour as follows. Not because the values in themselves mean much -they will vary from situation to situation-, but because it is the easiest way to survey the preceding.

In high density, the bumblebees visited on average 19.7 plants per bout (not shown, see Table 3). 52% of these visits was to plants with four inflorescences. Once the bumblebee had chosen a plant with four inflorescences, it visited on average 1.7 (or 1.8) inflorescences on that plant. Per inflorescence it spent on average 10 or 12 seconds (Table 3).

In low density the bumblebees visited 6.3 plants per bout on average. From these visits 56% were to plants with four inflorescences. From plants with four inflorescences 2.6 (or 3.0) inflorescences were visited on average. Per inflorescence the bumblebee stayed on average 14 (17) seconds.

One should keep in mind that the number of inflorescences visited per plant with four inflorescences was not normally distributed, but leptokurtic. From many plants only one inflorescence was visited, but on some occasions the bumblebee visited 10 or 15 inflorescences, revisiting the same inflorescence many times.

4 DISCUSSION

4.1 Natural population

4.1.1 Share of *Phyteuma nigrum* in insect visits

In this study *Phyteuma nigrum* had its peak in 'popularity' with the bumblebees before its peak in flowering (Figure 7). When *Phyteuma* reached its peak in flowering its share in the bumblebees' visits already declined. On the other hand the bumblebees already favoured *Phyteuma* when it was still relatively scarce. This is the same pattern as was found in earlier studies in the same population (Kwak et al. 1991b)

In studies with other plants (Stephenson, 1982; Schmitt, 1983), the opposite was found. In the raising phase of the flowering curve, few insects visited a flower species, at the top many visitors were attracted and in the declining phase still many pollinators visited the species. They might have become conditioned to it. These authors argue that in the declining phase maximum outcrossing (Stephenson) or gene flow (Schmitt) takes place. Because many visitors visit few flowers, each flower contains but little nectar and pollinators are compelled to visit many flowers to collect enough nectar. The flowers will often be at a greater distance from each other and thus less related. Maximum outcrossing in *Phyteuma* may have occurred in the early phase of flowering. Kwak et al. 1991a found that early and late in the season bumblebees flew relatively more between patches than at the top. The present study could not affirm this account, but this would also increase outcrossing early in the season.

The changes in 'preference' found in this study may derive from a changing composition of the pollinator force in the course of the blooming (Schmitt, 1983). The composition indeed changed from mainly *Bombus pascuorum* queens in the beginning to *B. pratorum* workers and *B. terrestris* workers later. All these bumblebee species however showed the same behaviour. From around 12 May they foraged a lot on *Phyteuma*, but from about 23 May they all turned there attention to other species, like *Rhinanthus angustifolius*, *Trifolium pratense* and *Rubus*. Gene flow however may have been influenced by the change in the composition of the pollinators, since *B. pascuorom* queens are less site constant than the other species

(Kwak et al, 1991).

Probably the main factor influencing the preference of the bumblebees besides the flowering curve of *Phyteuma* is the presence of other flower species as was suggested by Kwak et al. (1991b). *Phyteuma* is in its upward phase, when *Ajuga reptans*, an important bumblebee plant is at its top and bumblebees switch to *Phyteuma*. When *Phyteuma* is near its top three other important flower species are coming up: *Trifolium pratense*, *Lychnis flos-cuculi* and *Rhinanthus angustifolius*. Bumblebees may decide to start serving these instead of *Phyteuma* (See Heinrich, 1979a for information on flower choice by bumblebees.)

It is clear that the density of *Phyteuma* (in time) is not the only factor influencing pollinator behaviour and consequently pollen flow. In this study much attention is given to the role of density (in space), but it should be kept in mind that for instance the phenology of other bumblebee visited species may influence the visitation of the studied species. This is also shown in the experimental part of this study, as phenology of *R. angustifolius* probably affected the visitation of *Phyteuma*.

4.1.2 Pollen exchange between Populierenlaan and Meander

In 1990 the distance between the *Phyteuma* subpopulations Populierenlaan and Meander was over 150 metres. Thanks to a recent expansion to the north, this distance was about 100 metres in 1993. In the expansion *Phyteuma* distribution was very sparse. According to Levin & Kerstner (1969a, 1969b) low density is the optimal situation to serve as a bridge for gene flow between two populations.

In 1989 and 1990 no bumblebees were found that visited both the sites Populierenlaan and Meander, but this year 14 individuals were found that went across the brooks. This may be due to the expansion of the population Populierenlaan to the north which decreased the distance. Another explanation is that a different season, with different weather and flowering phenology may have affected the behaviour of the bumblebees. Studies in future years may explain the role of the expansion NvN if it continues to grow.

The smallest distance between PPL and Meander is 100 m. According to Heinrich (1979) average flight speed is 15 km/h, so it will take a bumblebee about 24 seconds to cross this. Bumblebees brush pollen from their body into the corbiculae during flight (Thomson & Plowright, 1980) and this may diminish the chance of pollen carryover over such distances.

A few successful fertilizations however would be enough to maintain gene flow between the patches (Wright, 1931).

-Losing 24 seconds involve no great cost if a bumblebee visits 4.5 inflorescences per minute, as found in the experiments.-

Rhingia campestris have no nest to return to, so they can roam from one site to another. From the 126 marked syrphids at least one crossed from PPL to Meander and one vice versa. Whether they will contribute to gene flow is doubtful, but not impossible. *Rhingia* are much less effective pollinators of *Phyteuma* than bumblebees, but they are able to pollinate *Phyteuma* (Kwak, 1993). Of course it may have been coincidence that 2 out of 126 individuals (and 3 out of 24 recaptures) were seen to have flown from one *Phyteuma* population to the other. If this reflects true proportions, *Rhingia* may still be important for pollen exchange between the sites. In other years (Kwak, 1993) as well as this year, *Rhingia* were more abundant visitors on *Phyteuma* than bumblebees.

An experiment with dye as a pollen analog might establish whether there really is pollen exchange between the sites. Considering the number of observations it took to demonstrate that insects do fly across, such an experiment would involve quite a lot of dyed flowers and need a bit of luck.

4.1.3 Gene flow between colonies

In recent literature much attention is paid to the consequences of inbreeding and outcrossing for plant populations. Some authors claim outcrossing between separated populations or even patches is harmful because local adaptation can be disrupted (Ellstrand, 1992). Other side authors claim that inbreeding is usually harmful to a population (Frankel & Soulé, 1981, Van Treuren, 1993). Disruption of local adaptation is hard to proof in natural situations and so is the negative influence of self-fertilization or breeding with close relatives. In greenhouse experiments, though, it has been found that inbred progeny was less vigorous (Van Treuren, 1993; Dudash, 1990). There are indications in literature that natural populations thrive by outbreeding: in some field studies it has been found that individuals with a high level of heterozygosity perform better (Schaal & Levin, 1976, Frankel & Soulé, 1981 and references there).

The local conditions in the *Phyteuma* populations Meander and PPL are different. If gene flow between both populations does exist, this may prevent local adaptation.

The population Meander is rather small. About 700
inflorescences were counted which means there were possibly
less than 500 flowering plants. Van den Brand (1989) estimated
a population size of only 150 inflorescences in 1989. If the
population size fluctuates like that, effective population
size is much smaller than 500 (e.g. Hartl & Clarke, 1989). In
such a small population the risks of inbreeding are probably
greater than the advantages of local adaptation (Frankel &
Soulé, 1981). Gene flow from the larger population PPL would
diminish the risks of extinction.

4.1.4 Flight distances between meadows

The largest distance between two recaptures of a
bumblebee on *Phyteuma* on the same day was 80 m in 1989/1990
(Kwak et al, 1991b), the largest distance between days 250 m.
Both were much longer in this study, 200 m and 390 m
respectively. When other plant species are included, the
largest distance in this study was 430 m, and that was on one
day. This exceeds the distance between Meander and PPL by far.

In the population PPL 17 trips of bumblebees on the same
day were observed that were longer than 100 m, and eight of
these trips involved solely *Phyteuma*. From PPL to Meander or
back, which is about 100 m, five trips on the same day were
observed of which one solely between *Phyteuma*. It seems that
the flowerless field and the brooks presented some obstacle
for the bumblebees. Whether the willow growths between meadows
were a hindrance for the bumblebees is not clear from this
study.

As in most studies on the subject, (e.g Beattie, 1976)
the distribution of flight distances was leptokurtic.

4.2 Experimental patches

4.2.1 Plant density and numbers of bumblebees

If the number of bumblebees did not respond significantly
to the density of Phyteuma, it did respond to the density of
Rhinanthus angustifolius. Or so it seemed, because the numbers
of bumblebees and *R. angustifolius* both decreased in time. The
not-bumblebees did not decrease in time, but they do not feed
on *R. angustifolius*. Their numbers fluctuated significantly
from one day to another and this was probably because these
species are sensible to the weather conditions.

Maybe the insensitivity of bumblebees to *Phyteuma* density
was an artifact of the experimental design. The patches

consisted of but 36 plants and covered only a very small area, the high-density patch being smaller than 1 m². It may be that both patches were saturated with pollinators. Bumblebees also react to areal size of patches (Thomson, 1981; Sih & Baltus, 1987). This may have interfered with the experiment.

4.2.2 Time spent per inflorescence

Bumblebees visited equal numbers of inflorescences per minute in high and low density. Yet they spent more time per inflorescence in low density. Though it is not unexpected that a bumblebee would stay longer on an inflorescence if the next one is farther away (see later), it seems odd that the bumblebees still visited equal numbers per minute in high and low density. It may be that the statistical test used was not powerful enough to prove a difference in numbers of inflorescences visited per minute. Another possibility is that the bumblebees lost less time in flying in low densities. This seems counter-intuitive, but in low density bumblebees visited more inflorescences per plant and visited more often nearest neighbours than in high density. This reduces flying time. Also I had the impression that in low density it happened less often that bumblebees flew to an inflorescence and rejected it at the last moment (See also Manning, 1956; Heinrich, 1979b). For practical reasons, in low density taller plants were used. If these had larger inflorescences that may have biased the measurements of time spent per inflorescence (Pyke, 1981).

4.2.3 Implications of bumblebee behaviour for plant fitness

Because *Phyteuma* is self-incompatible it needs to be pollinated with pollen from another conspecific individual to set seed. Pollinator behaviour affects directly a very important fitness component, seed set. In this study we have investigated the consequences of number of plants per m² and number of inflorescences per plant on pollinator behaviour.

In lower density the foraging bouts are shorter than in high density. From each bout the first inflorescence does not receive conspecific pollen and pollen from the last inflorescence will not fertilize another *Phyteuma*. The first and possibly some following inflorescences as well may even receive strange pollen which interferes negatively with conspecific pollen. When foraging bouts are shorter, relatively more visits will involve improper pollen transfer (sensu Rathke, 1983).

Foraging bumblebees visit more inflorescences on the same plant sequentially in lower density. Because *Phyteuma* is self-

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incompatible this means that in lower density more visits bring incompatible pollen. When a pollinator switches between inflorescences on the same plant, most of the pollen the second inflorescence receives will be coming from its 'brother' (though see Thomson & Plowright, 1980). This pollen cannot fertilize the ovula and is wasted and may prevent compatible pollen to fertilize the ovula.

The time spent per inflorescence was longer in lower density. This probably means the pollinator probed more flowers per inflorescence in lower density. In a study on bumblebee behaviour on clover, Heinrich (1979b) also found that bumblebees visited more florets per inflorescence in low densities. Though pollen removal is often correlated with the time spent on a flower (e.g. Mitchell, 1993) it may be unadvantageous for plants if pollinators spend much time on an inflorescence and probe many flowers. A plant's pollen dispersal may be maximized if each pollinator removes a little pollen and leaves pollen for a next pollinator. If the first pollinator removes all pollen, it may be deposited on only a few other flowers (Heinrich & Raven, 1972; Harder & Barrett, 1992). Of course this is only the case if the amount of pollen is limiting and this view takes only the male fitness into account.

Pyke (1981) on the other hand, argues that plant fitness is an asymptotically increasing function of the number of flowers probed per pollinator visit to the plant, because the longer an insect remains on a plant, the more pollen it deposits. He does not take into account that when a pollinator probes many flowers on an inflorescence, the last flowers may receive a lot of incompatible pollen. When pollen is limited, the waste of the pollen would imply a decrease of fitness. If pollen is limited and/or interferes with other pollen, Pyke's curve should have a maximum and bend down again. Pyke takes mainly the female fitness into account.

If the number of pollinators is limiting, and thus it is not sure whether there actually will be a next visitor, Pyke's model may be the more accurate. The fitness value of longer stays per inflorescence cannot be judged without further study.

According to this study, if all other things are equal, seed set in low density will probably be less than in high density. Of course in natural situations all other things may not be equal. For instance, plants in low density may suffer less from parasites (Anatovnics & Levin, 1980). This may affect plant characteristics that ultimately increase seed

set, like the number of inflorescences. Still, a low density is probably disadvantageous for seed set in *Phyteuma*.

Plants with many inflorescences were preferred by the bumblebees. Per inflorescence however these plants received less visits. How does this affect the plant's fitness? Let's assume that inflorescences on a plant with many inflorescences are equal to inflorescences on plants with one inflorescence in all respects. Plants with four inflorescences receive more visits than plants with one inflorescence. Even if only the visit to the first inflorescence results in fertilization, the plants with many inflorescences will set more seed than plants with one, because they receive more visits. All subsequent visits to inflorescences on the same plant are bonus.

There may however be a trade-off between the number of inflorescences a plant produces and their quality. In that case plants with many inflorescences need more visits to achieve equal seed set. There may be an optimal number of inflorescences that depends on plant density. Since density may fluctuate from one year to the next, selection on this trait is hard to imagine. A trade-off between quality and quantity of spikes of *Phyteuma*, however, has so far not been demonstrated (Boerrigter & Kwak, unpublished results).

Pollinator behaviour also affects another fitness component, genetic structure of the population. The present study does not allow pronouncements upon this aspect (but see Levin & Kerstner, 1969a & 1969b).

4.2.4 Optimal foraging?

Optimal foraging theory assumes animals to forage in such a way as to maximize their fitness. For nectar feeding animals the currency of fitness is believed to be the net rate of energy intake. So if the bumblebees maximize their net energy intake, they maximize their fitness (Pyke, 1981).

Bumblebees visited more inflorescences per plant when the plants stood further apart. Intuitively this is the most economical strategy. If a bumblebee visits more inflorescences on a plant, the chance of revisitation increases, assuming the bumblebee does not remember which inflorescences it already visited. The expected reward decreases with increasing number of visited inflorescences. However, if the next plant is at greater distance it is rewarding to stay longer on a plant, even if the reward decreases. Otherwise too much time will be spent in flying and the yield per minute decreases. The same reasoning goes for the number of flowers visited per inflorescence (See mathematical model Box 1).

When density was lower the bumblebees chose relatively shorter routes from one plant to the next. Since flying takes time and energy, this conserves energy. If a bumblebee always chooses a near neighbour plant, the chance of revisitation will increase. But when density is low, bumblebees remember better where the plants are and which they already visited (Manning, 1956).

The behaviour of the bumblebees in this study did not contradict optimal foraging theory.

BOX 1. Simple model for optimal foraging in different densities
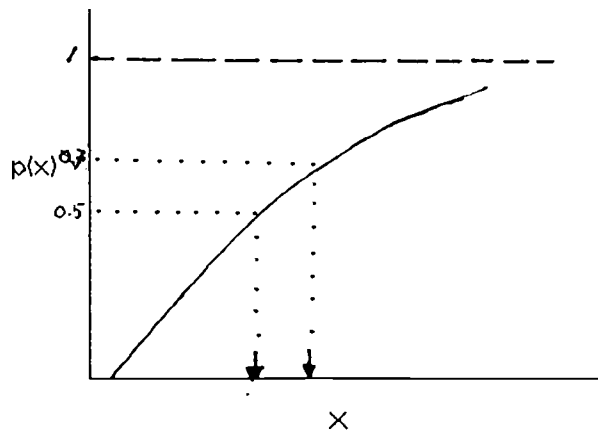
x   : number of visited flowers
p(x)   : probability of revisit is increasing function of x
nectar : amount of nectar in unvisited flowers.
The actual form of p(x) depends on the number of flowers on the plant and
their arrangement and the way the insect moves.

Expected yield per flower:

$$nectar*(1-p(x))$$

Expected cumulative yield after X flowers:

$$nectar*(X-\sum_{x=1}^{X} p(x))$$

1) Suppose the bee decides to leave after X flowers. It has to fly for the
duration of one visit to reach another plant. Advantage is: this plant has
not been visited. If it has visited one flower on the new plant, a bee that
stayed on the old plant would have collected:

$$nectar*(X+2-\sum_{x=1}^{X+2} p(x))$$

The bee that migrated has visited one flower less, but has less 'costs'.
Its yield is:

$$nectar*(X+1-\sum_{x=1}^{X} p(x))$$

The difference is:

2) If the next inflorescence is so far away a bee has to fly for a period

$$nectar*(1-\sum_{X+1}^{X+2} p(x))$$

as long as two visits, the difference between staying and leaving is after one visit to a new flower:

$$nectar*(2-\sum_{X+1}^{X+3} p(x))$$

The bee should leave if p(X) is over 0.67. Since p(x) is an increasing function this means the bee should stay longer on the old plant when the next plant is further away.(Figure).
In general, if flying takes as long as visiting n flowers, the difference is:

$$nectar*(X+n-\sum_{X+1}^{X+1+n} p(x))$$

This means that the right moment to leave is later if the distance between flowers is larger.
    This is of course a very crude model. One should include visits to other flowers on the new plant. Distances are of course not as discrete as in this model and the energetic costs of flying are not included (but see Heinrich, 1979a) etc. As far as I can see, such refinements do not change the general principal that the larger the distances between plants, the more flowers should be visited for optimal foraging, provided p(x) is an increasing function.

Acknowledgements

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Appendix

Appendix 1.
Survey of activities and weather. For explanation A, B, C, D,
E see Appendix 2.

Date	Experiment	Weather ☼
06/6/93	plants set out	mainly sunny, 22°C, little wind
07/6/93	plants set out, preliminary census A & B	as 06/6/93
08/6/93	census C & E	some clouds, 23°C, little wind
10/6/93	census C & E	sunny, 30°C, no wind
11/6/93	census A & B	mainly cloudy, 25°C, windy
16/6/93	census A & B	overcast, 18°C, windy
17/6/93	records kept in C & E	overcast, 17°C, windy
21/6/93	records kept in C & E	cloudy, 17°C, little wind
22/6/93	records kept in C & E and time per inflorescence measured	cloudy, 17°C, little wind
28/6/93	records kept in C, D & E	overcast, 18°C, no wind
29/6/93	records kept in C, D & E	fog, later sunny, 23°C, little wind
30/6/93	census in C, D & E	sunny, 18°C, little wind

Appendix 2.

Characteristics of experimental patches *Phyteuma nigrum* in
fields covered by *Rhinanthus angustifolius*. Experimental
patches contained 36 plants each. The plants were arranged in
a triangular form and uniformly distributed. From 6 till 16
June plants had 3 infloresences each, from 17 June on, half of
the plants had 4 inflorescences and half of the plants had 1
inflorescence.

Patch	Density *Phyteuma*	Density *Rhinanthus*
A	high: 43 plants/m²	high
B	low : 0.4 plants/m²	high
C	high: 43 plants/m²	low
D	intermediate: 4.7/m²	low
E	low : 0.4 plants/m²	low

Appendix 3.

Marked *Rhingia campestris* on 12 May. The differences in number of males and females are not significant (Wilcoxon Signed Rank). For whereabouts of the quadrates see map, Figure 1.

Quadrate	colour	♀	♀	♀+♂
Meander I	white	0	1	1
Meander II	red	8	4	12
PPL (NvN)	blue	4	1	5
PPL (Noord)	yellow	5	6	11
PPL (Centraal)	green	20	11	31
PPL (Achter de Wilgen)	pink	47	19	66
TOTAL		84	42	126