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SOCIO-ECONOMIC EFFECTS ON COLONY SIZE IN
THE BUMBLE BEE BOMBUS TERRESTRIS.
(HYMENOPTERA: APIDAE)

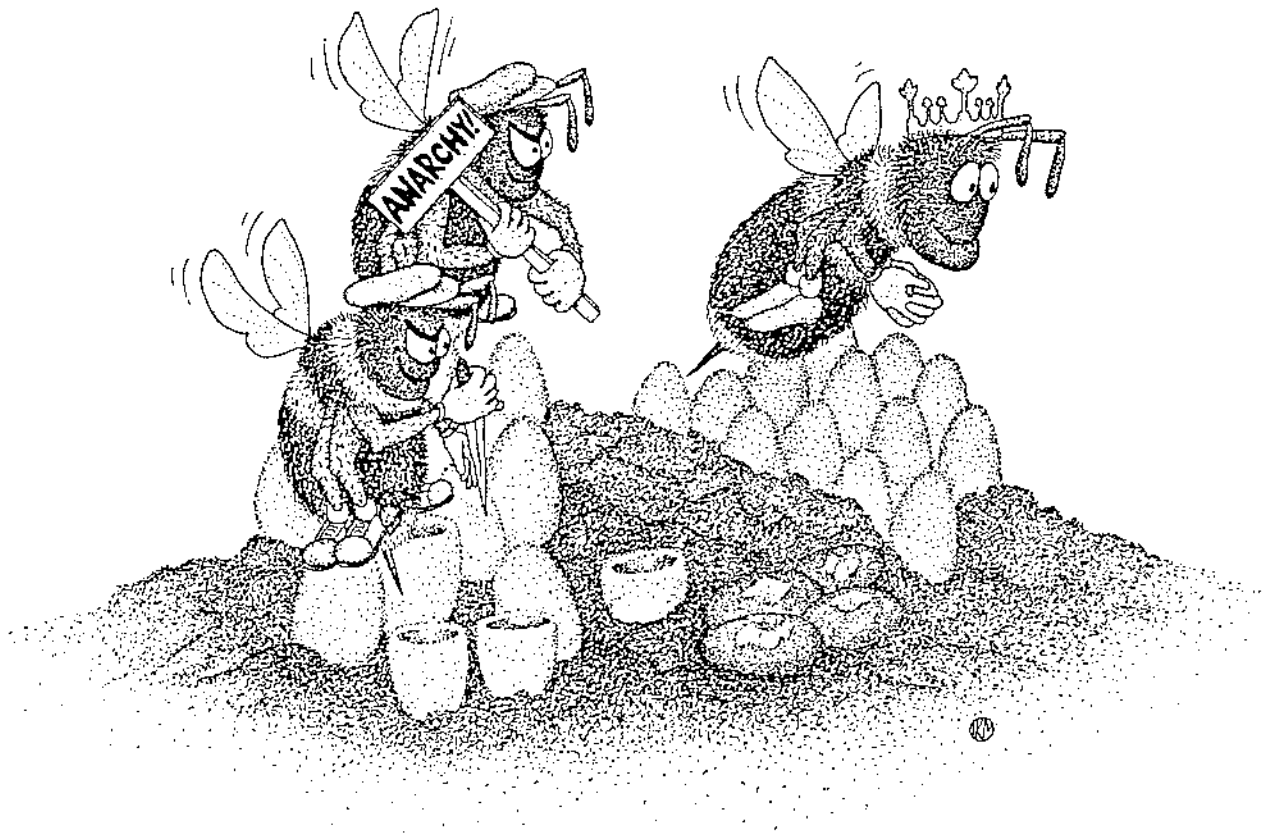
BY

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CONTENTS

	<u>Page</u>
LIST OF FIGURES	vi
LIST OF TABLES	vii
ACKNOWLEDGEMENTS	viii
ABSTRACT	ix
CHAPTER 1 INTRODUCTION	1
1.1 BUMBLE BEE NATURAL HISTORY	3
1.2 BUMBLE BEE DOMESTICATION	6
1.3 OBJECTIVES OF THE PRESENT STUDY	7
CHAPTER 2 THE EFFECT OF SUPPLEMENTARY FEEDING ON COLONY SIZE	9
2.1 EXPERIMENT 1	9
2.1.1 INTRODUCTION	9
2.1.2 METHODS	11
2.1.2.1 1984-1985 SEASON	11
2.1.2.2 1985-1986 SEASON	13
2.1.3 RESULTS	15
2.1.3.1 1984-1985 SEASON	15
2.1.3.2 1985-1986 SEASON	15
2.1.4 DISCUSSION	18
2.2 EXPERIMENT 2	19
2.2.1 INTRODUCTION	19
2.2.2 METHODS	20
2.2.3 RESULTS	22

2.2.4	DISCUSSION	28
2.3	GENERAL DISCUSSION	29
CHAPTER 3	DOMINANT WORKERS	31
3.1	INTRODUCTION	31
3.2	METHODS	32
3.3	RESULTS	34
3.4	DISCUSSION	36
CHAPTER 4	THE EFFECT OF SUPPLEMENTARY SUGAR FEEDING ON WORKER OVARY SIZE	38
4.1	INTRODUCTION	38
4.2	METHODS	39
4.3	RESULTS	40
4.4	DISCUSSION	42
CHAPTER 5	CROWDING	44
5.1	INTRODUCTION	44
5.2	METHODS	45
5.3	RESULTS	47
5.4	DISCUSSION	49
CHAPTER 6	GENERAL DISCUSSION	51
APPENDIX 1	FIELD HIVE CONSTRUCTION	61
APPENDIX 2	LIGHT BOX	64

APPENDIX 3 CORBICULAR POLLEN WEIGHTS	66
APPENDIX 4 INFLUENCE OF WEATHER CONDITIONS ON FORAGING	69
REFERENCES	72

List of Figures

	<u>Facing</u>	<u>Page</u>
FIGURE 1	(a) Softboard hive liner.	14
	(b) pumice concrete field hive.	14
FIGURE 2	Productivity of fed and non-fed colonies.	16
FIGURE 3	Colony productivity index versus date of queen capture.	17
FIGURE 4	Relationship between daily pollen income and area of growing brood.	23
FIGURE 5	Number of days from colony initiation to foundress queens' death in relation to artificial food supplementation.	25
FIGURE 6	Frequency distribution of oocyte lengths	41
FIGURE 7	Modified starter box.	46
FIGURE 8	Relation between day of first male emergence and day of first queen emergence.	48
FIGURE 9	Pumice concrete field hive with plastic hive liner.	62
FIGURE 10	Light box.	65
FIGURE 11	Amount of pollen (per unit brood area) collected by foragers under different weather conditions.	70

List of Tables

	<u>Page</u>
Table 1 Results of food supplementation experiment on in observation hives	26
Table 2 Results of worker age experiment on colonies in observation hives	35

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ABSTRACT

This study examined factors related to the development and eventual size of bumble bee colonies (Bombus terrestris (L).). Experiments were conducted over two seasons in both the laboratory and the field. In the first season the effect of supplementary feeding of sugar solution upon colony growth was examined. In the second season the effect of sugar feeding on worker dominance was studied.

The feeding of sugar syrup to bumble bee colonies resulted in a reduced colony size and (at the 56th day from colony initiation) greater mean ovarian development in workers. In laboratory colonies that were sugar-fed foundress queen longevity was reduced. These results were interpreted as suggesting that sugar feeding influenced colony size via an effect on the social structure of the colony. Conflict between workers and the foundress queen near the peak of colony development has been widely reported in bumble bees and my results suggested sugar feeding increased worker dominance over the queen and that this curtailed colony growth. The relationship between worker dominance and colony development was studied, using the assumption that the dominance of workers increases with their age. Colonies were manipulated by selective removal of either the oldest or youngest workers, to produce a difference in the average age of the worker populations. In colonies with a higher mean worker age there was an earlier change to sexual production and fewer workers were produced. Earlier sexual production also occurred where the number of workers in a nest was artificially increased. The small size of sugar-fed colonies was attributed to the early appearance of

reproductives due to a premature rise in the numbers of dominant workers. It was concluded that worker dominance was affected by trophic/economic effects such as the effort required to procure nectar, and that the provision of sugar solution in hives seems inadvisable in commercial management of bumble bees.

CHAPTER 1

INTRODUCTION

Bumble bees are widely recognised as effective pollinators of commercial crops (Free 1970a, 1970b), specifically forage legumes of which the most important are red clover and alfalfa (Bohart 1957, 1958, 1960; Akerberg and Stapel 1966; Peterson et al 1960). Bumble bees are considered to be especially tolerant of adverse weather conditions and are seen foraging at times when honey bees are not active (Heinrich 1972, 1979a, 1979b; Heinrich et al 1977; Pyke 1978; Wilson 1929). The failure to set seed of the first red clover (Trifolium pratense L.) crops in New Zealand was a result of the lack of any native or introduced bumble bees (Tillyard 1926). A dramatic increase in seed yield resulted from the introduction of bumble bees to the South Island in 1885 (Hopkins 1914). Bumble bees quickly became established and spread to the North Island (Thomson 1922). A second release was made in 1906. After a survey in 1953 Gurr described the presence and distribution of four species (Gurr 1957a, 1964, 1972). These four species were B. ruderatus (Fab.), B. hortorum (L.), B. subterraneus (L.) subsp latreillellus (Kirby) and B. terrestris (L.).

Seed yields began to decline again after a number of years. This was possibly because the increased acreage of red clover which was enthusiastically planted could not all be pollinated by a limited bumble bee population (Gurr 1975). As such there have been continual demands from farmers for the introduction of new species of bumble bees. This has instigated much research into the commercial management of bumble bees.

B. terrestris was the only short tongued species introduced to New Zealand. The workers of this species cannot reach the nectaries of red clover. They therefore resort to "robbing" the flowers by biting holes through the base of the corolla tube and collecting nectar without pollinating the plant. In New Zealand honey bees may become secondary "robbers" by the use of the holes made by the primary robbers, though in other countries many other bee species may fill this role (Holm 1966). Contrary to the opinion of many authors there is no evidence that robbing is detrimental to red clover seed yield (Hawkins 1961; Morrison 1961)

Nevertheless B. terrestris has been found to be an ideal pollinator of many fruit and berry crops, including kiwifruit. A research project is currently being undertaken at Massey University to discover methods by which B. terrestris colonies can be commercially produced primarily for Kiwifruit pollination. This species was therefore chosen as the object of this study.

Although bumble bee colonies are known to range in size from 50 to 500 and sometimes 1000 workers (personal observations), researchers attempting to raise colonies by allowing them to free forage have found that a surprisingly large percentage produce very small colonies. Initially the major purpose of this study was to investigate methods by which bumble bee colony size could be increased. As small colony size is a handicap to commercialisation, this would enable artificial colonies of a reasonable size to be produced for crop pollination.

1.1 BUMBLE BEE NATURAL HISTORY

Inseminated queen bumble bees hibernate over the winter period for, in the British Isles, "anything from six to eight or even nine months" (Alford, 1975). In the mild climate of Palmerston North, N.Z. this period can be substantially shorter (Pomeroy personal communication). They hibernate individually in small cavities at some 10cm or less below the surface in well drained soil (Sladen 1912; Bols 1937, 1939; Alford 1969). On emerging from hibernation in the spring the queens spend two or more weeks actively flying, foraging or sunning themselves (Heinrich 1979a). Once the queen's ovaries have begun to develop she begins to search for nest sites (Cumber 1949a; Miyamoto 1960).

After selecting a suitable site, usually an abandoned rodent hole, the queen will manipulate the available nest material to form a cavity in the centre in which she keeps her brood warm. At this point the presence of good thermally insulating material is important as a comb temperature of 28 to 30 degrees Celsius is to be maintained through the season (Hasselrot 1960; Wojtowski 1963).

The queen then collects pollen which forms a "clump" on the floor of the nest cavity. A batch of 8-14 eggs (Pomeroy 1979) is then laid into a cup shaped depression lined with wax (egg cell) which is then closed over with wax. After hatching, about 4 days after oviposition, the larvae grow rapidly, feeding upon the pollen lump below them and from regurgitated pollen/nectar mix from the queen through holes in their wax canopy. The larvae ultimately spin cocoons of silk and pupate inside them.

Based on my own observations of B. terrestris the first workers emerge 19-21 days after the eggs are laid. During the first 12 - 24 hours young bees (termed callows) perform no "duties" within the nest, during this stage they are still developing coat colouration and wing hardness (Pomeroy 1977). The first workers eventually take over all the foraging and most nest duties from the queen. Batches of 6 to 20 eggs are laid in egg cells on top of newly spun cocoons. Cumber (1949a) and Brian (1951) calculated that for B. agrorum less than 50% of eggs laid survived to adulthood. The edges of empty cocoons are extended with wax collars and reutilised as honey pots and pollen storage pots (Heinrich 1979a).

The comb develops upwards and outwards becoming roughly hemispherical, though this may differ in some species, and also depends on the shape of the hole in which the nest is located. The number of workers increases exponentially (Plowright 1966, Röseler 1967a) until there is a change to the production of males and queens. It is not uncommon that some colonies produce no queens at all, if male production is started at an early stage when the colony is small. Once the production of males starts it is rarely reversed, and the production of workers diminishes.

A division of labour among the workers often develops in relation to size and age. The youngest (1-2 days old) and smallest bees often stay within the nest while the larger workers do most of the foraging. After some time many of the "house-bees" may begin to develop their ovaries, the possible causes for which form the major part of this study. Much antagonism begins to occur within the nest between workers

with enlarged ovaries, and between these workers and the queen. The queen starts laying male eggs and soon after the workers also start laying eggs (Owen and Plowright 1980) (all worker eggs are unfertilised and therefore males). A large proportion of all the males may be worker produced. Owen and Plowright (1982) estimated 39% of males in 17 laboratory colonies of B. melanopygus to be produced by workers, and van Honk et al (1981) calculated that 80% of the males from a B. terrestris colony originated from worker laid eggs. At the start of male production any remaining female larvae are fed at an elevated rate and become new queens. Antagonism within the nest continues for some time and may increase to the point where the queen is killed or pushed off the comb by the most dominant workers. It should be pointed out that not all nests lose their queens in this way. Wild nests have been found in advanced stages of male and queen production in which the foundress queens seem to have retained their dominant position (personal observations).

After a young queen has emerged, in about mid summer, she stays within the nest for about 5 days before going on a mating flight (Cumber 1953a). She then returns to the colony and spends a period of time building up her fat bodies from the nest pollen and nectar store before going in search of a suitable hibernation area. During this time her ovaries would normally remain undeveloped (Cumber 1949b). Though in the few tropical Bombus species, eg. B. atratus, the young queens may not go into hibernation at all, but (after fertilization) will replace the old queen on the comb and start egg production (Sakagami et al 1967).

Males leave their nest when they are between two and four days old and normally never return. They generally live for three to four weeks spending their time foraging for their day to day food requirements from flowers, and inseminating young queens (Alford 1975).

1.2 BUMBLE BEE DOMESTICATION

As mentioned above, bumble bees are ideal pollinators of many crops. The pollination of these crops is ultimately dependent on the number of bees in the area (Jamieson 1950; Bird 1944), but the natural population is often too sparse and fluctuates from year to year. Many investigators have attempted to alleviate the problem of fluctuating numbers of bumble bees through propagation of the natural population (Frison 1926; Fye and Medler 1954 and Hobbs et al 1960). These and other authors have placed artificial domiciles of various designs in the field for attracting wild queens to initiate colonies. The most recent work in this area has been undertaken by Macfarlane et al (1983) and Donovan and Wier (1978) who had occupation rates of between 20 and 30 % (this is similar to the queen occupation rates of earlier researchers), and Pomeroy (1981a) who found that of 45 underground hives 93% were occupied by queens.

Laboratory initiation of colonies using wild-caught queens is another means by which researchers have attempted to raise bumble bees. Factors found to affect the success of this method were the physical design of the starter-boxes (Hasselrot 1952; Plath 1923), the food requirements and the way in which it is presented (Plowright and Jay 1966; Pomeroy personal communication.), heating requirements (early

researchers used various types of insulation material, but it was later found that this was not required if the temperature within the nest box was raised (Hasselrot 1960)) and social factors (both Medler 1957 and Plowright and Jay 1966 found that some social stimulation from other queens or workers produced better colony initiation than did single queen confinement). Current work in this field is being undertaken by Pomeroy (unpublished) in which large numbers of colonies are being started in small, two chambered, plastic moulded starter boxes maintained at an elevated temperature. This is the method of colony initiation used in this investigation.

1.3 OBJECTIVES OF THE PRESENT STUDY

The original aim of this project was to study the factors affecting the size of bumble bee colonies and to determine means by which final colony size could be artificially, and economically increased. The 1984-85 summer season was spent on an experiment attempting to increase the size of colonies by feeding them excess sugar syrup. As many authors have suggested that food is limiting (Cumber 1949a, 1953b; Medler 1958) the expectation was that they would increase in size. The results were contrary to this expectation and in fact showed that feeding sugar syrup had a debilitating effect upon the colonies. They also indicated that this might be due to social factors (specifically the presence of dominant workers) within the nest. Work by Free (1955a), Röseler (1967b) and van Honk and Hogweg (1981) has shown that the presence of dominant workers can have a disrupting effect upon the colony and that these workers may affect the timing of male production. As the final colony size is to some extent dependent on the timing of

male production (Pomeroy 1979) subsequent work was therefore directed towards determining the effect of sugar feeding on factors within the nest, particularly the preponderance of dominant workers, and the effect of these factors on colony size.

In the second season the aim of the first laboratory experiment was to discover whether the early presence of dominant workers in the colony affects the timing of male production and the consequent curtailment of colony growth (chapter 3). An experiment was also designed to determine whether sugar feeding affected the proportion of dominant workers within the colony.

Artificial feeding might be expected to reduce the colony's foraging effort resulting in more bees staying "at home" on the comb. Röseler (1967a) suggested an increased density of bees on the comb stimulates the foundress to produce male eggs, but Pomeroy and Plowright (1982) failed to demonstrate such an influence in an experimental situation. I examined this factor in the present study.

CHAPTER 2

THE EFFECT OF SUPPLEMENTARY FEEDING ON COLONY SIZE

2.1 EXPERIMENT 1

THE EFFECT OF FEEDING SUPPLEMENTARY SUGAR SOLUTION

ON FREE FORAGING BUMBLE BEE COLONIES

2.1.1 INTRODUCTION

Many authors have suggested that bumble bee colony size in the field is limited by the availability of food (Cumber 1949a, 1953b; Gurr 1957b; Medler 1958 and Plath 1934). Although there were few data available to support this, Plowright et al (1978) have shown improved forager profits in an area where the bumble bee density was reduced by aerial spraying of fenitrothion (for the control of spruce budworm). The pollen intake per unit foraging effort was five times greater for bumble bee colonies in the sprayed area than in nearby comparable unsprayed areas. They also state that their data indicate some plants "may depend upon the existence of competition for forage among bumble bees in order to secure a high visitation rate."

One of the most important factors related to colony growth is the amount of pollen brought into the hive. Pollen shortages would be expected to limit colony growth because pollen is a colony's only source of protein and therefore the only means by which larvae can increase in size. Pendrel and Plowright (1981) pointed out that sparsely fed larvae took longer to grow, while Plowright and Jay (1977)

suggested that a low rate of pollen intake may result in pupation at a smaller size, resulting in smaller adult workers. These small workers in turn would not collect as much pollen as large ones (Alford 1975). Yet another effect of low rates of pollen intake may be ejection of some larvae, resulting in fewer emergent adults (Pomeroy 1977).

If, as the above suggests, reduced pollen intake limits the rate of worker production, it is logical to assume that increasing the quantity of pollen in the hive will lead to greater bee production.

Two means by which the amount of pollen in the hive can be increased are;-

1/ Physically placing the pollen in the hive. This has some major drawbacks. Pollen deteriorates very quickly (a day or two) and becomes unpalatable to the bees (personal observations). This would mean pollen would have to be given in small amounts on a regular basis. For a commercial system this represents a high labour input and may be prohibitively expensive. There is also the problem that the pollen must be placed in close proximity to the comb. With many aggressive workers flying about this would be a delicate operation to say the least.

2/ Providing the colony with a sugar supply just outside the nest entrance. This can be easily replaced on a regular basis (every 1 or 2 weeks). A foraging bumble bee must divide its foraging time between the collection of nectar and the collection of pollen. Nectar is used by the bees as an energy source to fuel foragers for the collection of pollen and for heating the hive. Providing the colony with sugar solution should greatly reduce the need for nectar foraging and thus

"free up" forager effort for pollen collection. Free (1955b) pointed out that nectar collection takes priority over pollen collection. Easy access to a sugar source would therefore allow foragers to collect more pollen during breaks in bad weather, otherwise they would spend most of their time restocking empty nectar pots. Provision of sugar solution would hopefully decrease the chance of there being a pollen shortage, therefore increasing the growth rate and the final colony size.

The aim of experiment 1 was to test the effect on colony size of feeding supplementary sugar to bumble bee colonies. The hypothesis that supplementary sugar feeding would in fact increase pollen intake was tested in a simultaneous experiment (experiment 2, below).

2.1.2 METHODS

2.1.2.1 1984-1985 SEASON

Forty colonies were started in the laboratory for use in this experiment. Due to a lack of laying queens, brood clumps whose queens had died were fostered to other non-laying queens. In every case the recipient queen was induced to start laying. This occurred in eleven cases. When colonies had gained 8-12 workers, the starter boxes were taken to a field and placed in a pumice concrete hive (hive construction: appendix 1, figure 9). Half of the colonies were provided with sucrose solution (50% v/v) in their vestibules (treatment 1 : sugar fed). The other 20 colonies did not receive this supplement (treatment 2 : unfed).

Due to a low rate of queen capture at the beginning of the season, and a low colony starting rate, experimental colonies became available in small numbers over a long period of time (5/11/84 - 20/12/84). The batch of colonies put outdoors on a particular day comprised equal numbers of both treatments so that paired comparisons could be made independently of timing.

Hives were placed in a 0.5ha field situated on the flats on the north side of the Tiritea Stream which borders Massey University campus. Adjacent to this field was a 0.35ha field planted in Russell lupin, Fodder radish, Borage and Genista sp for the purpose of providing nectar and pollen for bumble bees.

The supplies of sugar solution in the vestibule were replaced weekly. All colonies were examined at this time.

When the queen had died and there were no more than three workers left on the comb (which by this time consisted largely of empty cocoons) and very little healthy brood, each colony was removed from the field and the combs were dismantled. The type (worker/male, queen, unemerged) and number of cocoons were noted and the productivity index (P.I.) for each hive was calculated as described by Pomeroy (1981a). His index was based on the total production of cocoons with queen-sized cocoons given a higher value to reflect the greater pollen investment in them. I used Pomeroy's formula for relating cocoon diameters to pollen investment, but took my own data to calculate a weighting factor for queen cocoons in relation to those of workers and males (indistinguishable). Fifty intact worker/male and fifty queen cocoons

were randomly chosen and their diameters measured using the method of Pomeroy (1979) to calculate the queen weighting factor (F). The P.I. for each colony was calculated as follows:

$$\text{P.I.} = (\text{no. of male/worker cocoons}) + F(\text{no. of queen cocoons})$$

F = 3.5 i.e. the mean pollen investment is estimated to be 3.5 times greater to raise queens than worker/males

2.1.2.2 1985-86 SEASON

The first season's comparison of sugar-fed and non-fed colonies was repeated with twelve colonies per treatment, but this time the primary aim was to examine the effect on worker ovarian development, therefore all colonies were sacrificed after 56 days from first egg laying for worker dissection. This aspect is dealt with in chapter 4. The outdoor hive was different from the first season and is illustrated in figures 1a and 1b. As the colonies were not allowed to complete their development the productivity indices recorded for the second season reflect only the growth up to the 56th day, and not the total potential of the colonies.

Figure 1.

A) Perspective view of softboard hive liner. The hatched surface indicates where half of the liner has been cut away to show the interior and starter box.

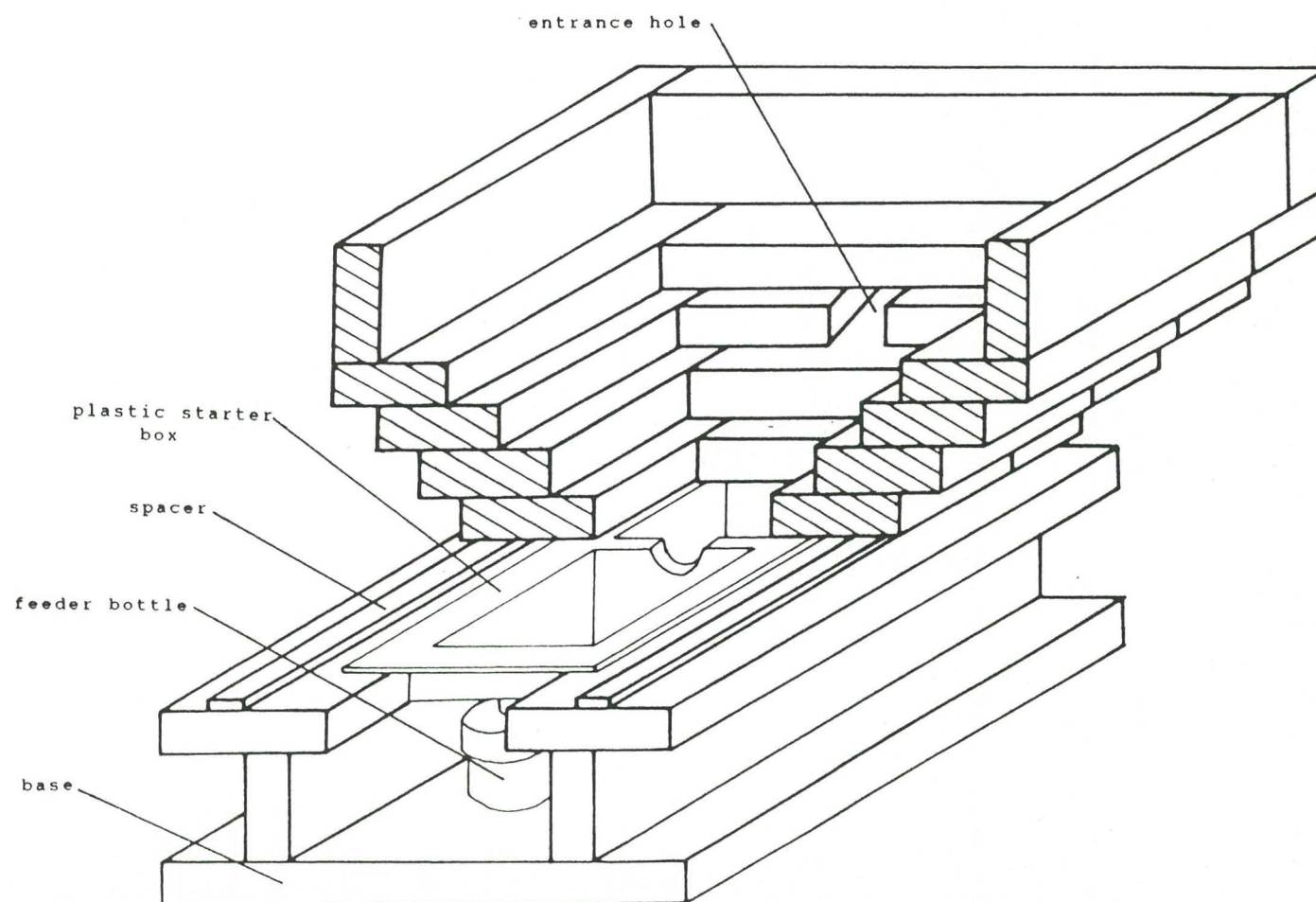
External dimensions; height = 180mm, width = 250mm

B) Mid-vertical section of pumice concrete field hive with softboard hive liner.

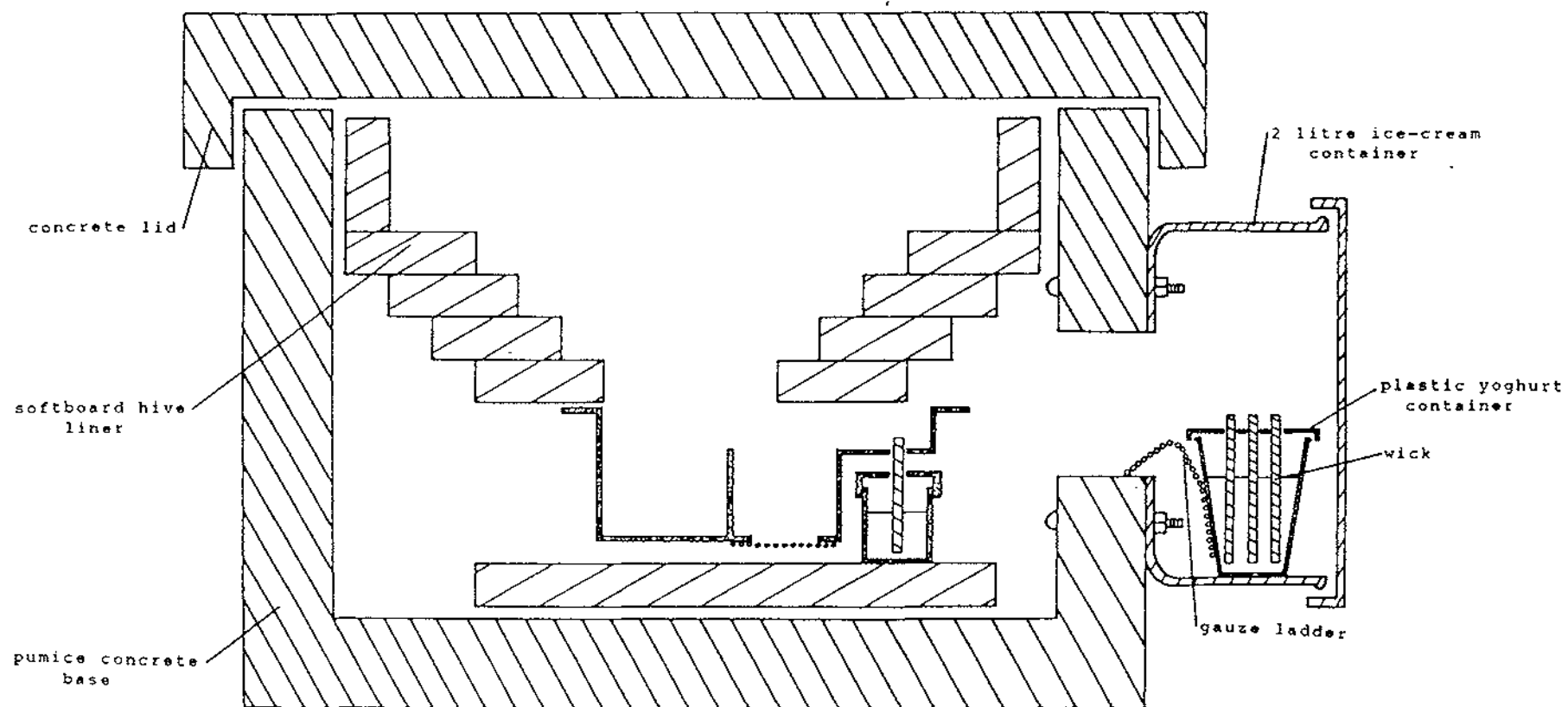
External dimensions; height = 250mm, width = 330mm

Drawn by I. McGee

A



B



2.1.3 RESULTS

2.1.3.1 1984-85 SEASON

On two separate occasions sheep found their way into the study field and on two occasions the grass in the field was mown by a tractor. Due to these factors two hives were destroyed and one was damaged to the point where it was discarded.

The mean productivity index for the colonies in the fed treatment (101.8) was significantly smaller than that of the control (198.3), ($P = 0.02$, Mann-Whitney U-test) (see figure 2).

The effect of fostering, differing insulation and the date at which the colonies were put out in the field were shown to have little effect on the productivity index. The date at which the queens were caught had a significant effect ($P < 0.02$, analysis of covariance), but this effect was independent of the experimental treatment (see figure 3).

2.1.3.2 1985-86 SEASON

Figure 2 shows the productivity indices for the 1985-86 season field hives. As expected the mean productivity index was greater in the control (mean = 626) than in the sugar treatment (mean = 252.8), ($P = 0.051$, Mann-Whitney U-test, 1-tailed).

Figure 2.

Productivity of colonies in sugar fed and non-fed (control) treatments from season 1984-85 and season 1985-86. Colonies are pooled within intervals of 50 P.I. units.

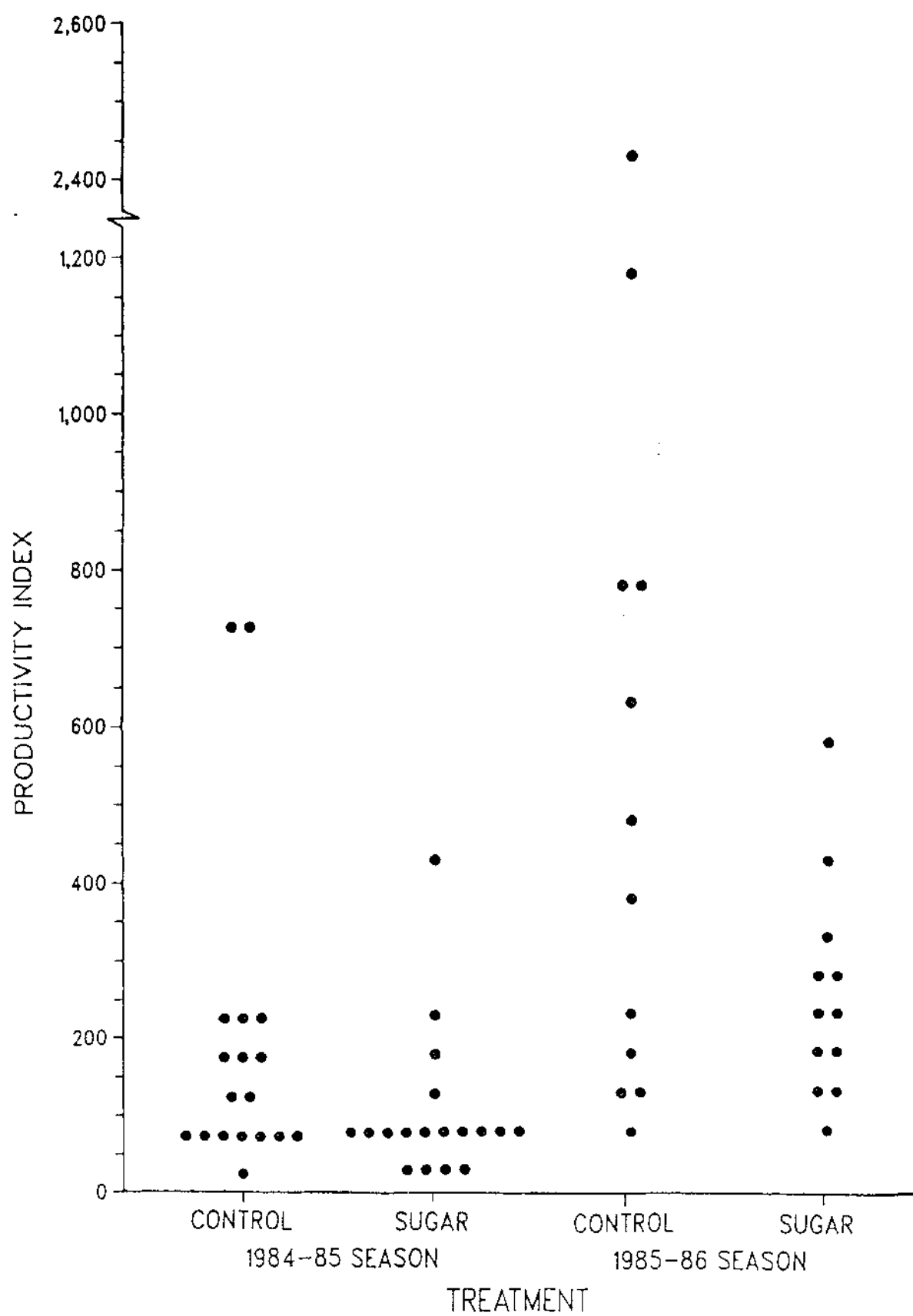
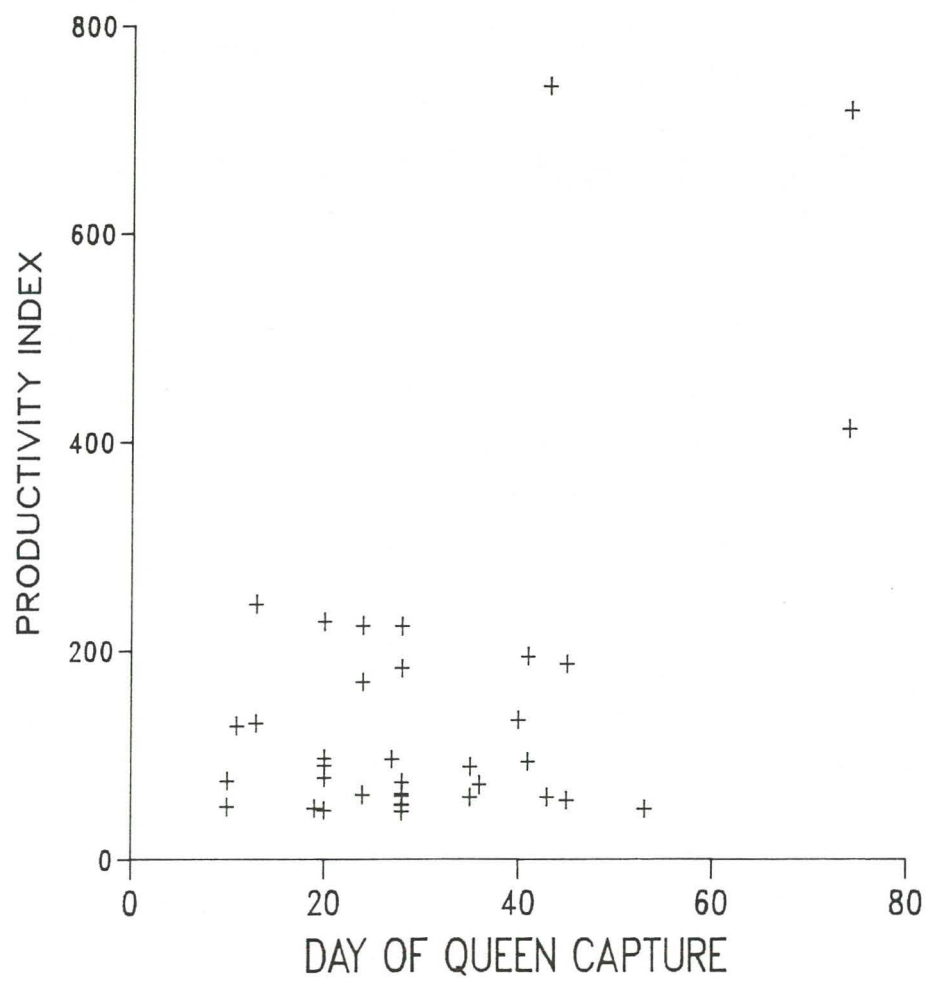


Figure 3.

Colony Productivity Index (P.I.) versus the date of queen capture
(day 0 represents 31 August 1984)



2.1.4 DISCUSSION

These results do not support the hypothesis that feeding sugar solution to bumble bee colonies will result in an increased final colony size. In fact feeding bumble bee colonies with sugar solution would seem to be detrimental to their growth. One possible explanation was suggested by Cumber (1953b). Although he stresses the importance of copious spring forage for incipient colonies if they are going to be successful, he also suggests that an over abundance of food during the summer months may reduce their numbers. If the food supplied by the workers increases in relation to the number of eggs in the colony, there may be a change to the production of the sexual forms earlier in the colony life, before the nest has reached a large size (many authors have stated that food intake is an important factor stimulating the production of new queens in the bumble bee colony (Cumber 1949a; Plowright and Jay 1968 and others)). Cumber suggests that the long tongued species present in New Zealand "may find a deep seated supply of nectar for which there is little competition" and that this will increase worker efficiency. As pollen is the only protein source for larval growth, Cumber seems to be suggesting that this extra nectar supply leads to increased pollen intake by larvae, which leads to the production of early reproductives. Experiment 2 (next section) was designed, in part, to determine whether colonies supplemented with sugar solution did in fact collect more pollen.

2.2 Experiment 2.

THE EFFECT OF FEEDING ON FORAGING AND COLONY DEVELOPMENT

2.2.1 INTRODUCTION

Whereas the experiment 1 colonies were outdoors, and undisturbed except for the unobtrusive provisioning with sugar syrup, the experiment 2 colonies, although still free foraging, were in indoor, heated, observation hives so that daily addition of pollen and detailed comb observations were possible. This enabled me to see what effect sugar supplementation actually had on pollen intake by foragers, and also to supplement some colonies directly with only pollen to study the effect on colony development.

As mentioned in the general introduction, there is wide concurrence with the view that trophic conditions (specifically worker:larvae ratio and larval feeding rate) affect queen rearing (Cumber 1949a, Free 1955c, Röseler 1967a, Plowright and Jay 1968). Therefore the secondary aim of this experiment was to investigate the effect of supplementary feeding on male and queen production.

2.2.2 METHODS

Fifteen colonies were started and placed in conically shaped, heated observation hives as designed by Pomeroy (1977), and ducted to the exterior of the building through the floor. The exit holes were made inconspicuous to avoid bees returning to the wrong colony (bees that become lost or disorientated may return to the colony with the most prominent entrance hole. This is called "drift" in apiculture and is described in Free (1958)) but the surrounding areas were "decorated" with various distinctive-looking debris to increase the chances of foragers returning to their own hive.

Each exit tunnel passed through a "light box" (appendix 2) which photoelectrically detected returning foragers and emitted an audible "beep". This was so that foragers could be examined as they returned without the observer having to watch all tunnels continuously.

Five colonies were randomly allocated to each of 3 treatments (These treatments were spread between three tables so that each table had at least one hive from each treatment.)

The treatments were:

1. Excess sugar was provided as in the field experiments. The wicks of the sugar reservoirs were cut at a 45 degree angle and pressed against the gauze tunnels leading from the vestibule to the light box.
2. Excess pollen was provided. Small plastic vials (volume=4.4ml) containing pollen, mixed to a doughy consistency with sugar syrup, were placed inside the hive and were replaced daily.

3. Control: no extra sugar or pollen.

Pollen-intake data were collected one day a week, for the first 30 minutes of every hour from sunrise to sunset, from 16/11/84 to 13/2/85. There were large numbers of workers wandering within the tunnels without actually foraging in the field, and it was impossible to distinguish these from foragers bearing only nectar. Therefore only pollen bearing bees were recorded. For each returning forager the size of its corbicular pollen lumps was estimated. Pollen loads where present were scored on a scale of 1 to 4.

Samples of corbicular loads were taken from bees on 20/11/84, 22/12/84 and 8/1/85. On each of these three days ten pollen bearing bees for each of the four pollen lump classes (ie., 40 on each day) were captured, when returning to their nest, and their corbicular pollen weighed (see appendix 3). In this way I could determine the average weights of the corbicular pollen lumps for each of the 4 different size classes.

The hypothesis that sugar feeding would increase pollen intake could not be tested by measuring the intake alone because this quantity is strongly influenced by the demand for larval feeding (Pomeroy 1977). Pendrel and Plowright (1981) found that pollen intake and consumption fluctuates greatly in accordance with larval area.

To record larval area for those days on which pollen collecting data were being recorded, each colony was photographed at about 12 noon. The outlines of larval clumps were traced from the photographs onto

acetate sheets which were then cut out and weighed. Larval clump area was then calculated using the standard area:weight ratio of acetate. Pollen intake per unit larval area was related to the feeding treatment of the colonies.

Other factors which may have been influenced by the above feeding regimes were also measured. These include the productivity index, the duration of both the queen's life and the life of the colony as a whole, and the date of first-male and queen production.

The effect of weather conditions on foraging was also noted, the results for this are presented and discussed in appendix 4.

2.2.3 RESULTS

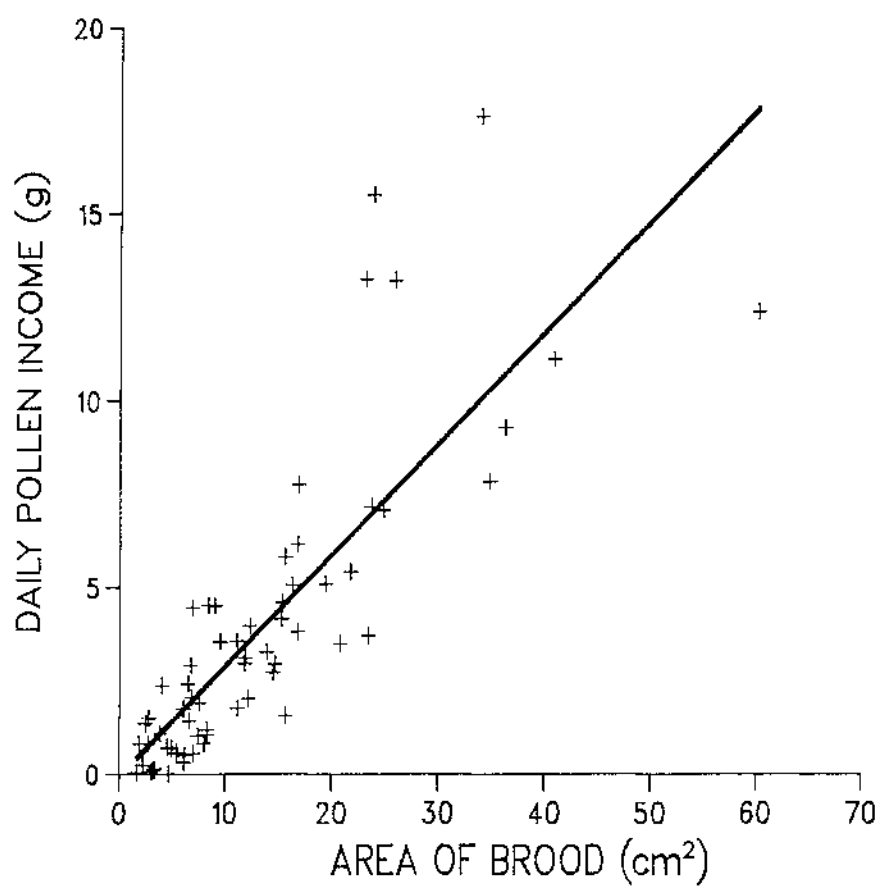
A two sample t-test was used on all results unless otherwise specified. The amount of pollen brought into the hive per unit area of growing brood did not differ for the 3 treatments (anova, $F_{2,10} = 1.874$, n.s.). The amount of pollen brought into the colony for the sugar treatment and the control colonies, in dry weather, was highly correlated with the square area of brood in the nest ($r = 0.817$, $P < 0.0001$) (see figure 4).

Figure 4.

Relationship between daily pollen income and area of growing brood.
Data pooled for sugar treatment and control.

Regression equation: $X = -0.08 + 0.298Y$

X = area of brood, Y = daily pollen income



In the hives with the pollen treatment, the bees made little use of the pollen given to them, preferring to collect their own. However on days of heavy rain when collecting was reduced (see appendix 4) the amount of provided pollen that was consumed did increase ($P = 0.01$).

Figure 5 shows the date of queen death for each of the three treatments. The length of the foundress queens' life was found to be significantly shorter in the sugar treatment than in the control ($P < .0001$), there was also a marginally significant difference between the sugar and pollen treatments ($P = 0.079$).

The mean day on which colonies died was earlier for the sugar fed colonies than for the control or pollen fed colonies, although this was only marginally significant ($P = 0.086$). There was no significant difference between the three treatments for the productivity index ($P = 0.55$) or the day on which the first males emerged ($P = 0.13$). Queens were only produced in four colonies; two within the pollen treatment and one each in the sugar treatment and the control. A summary of the main results is given in table 1.

Figure 5.

Number of days from colony initiation to foundress queens' death in
relation to artificial food supplementation

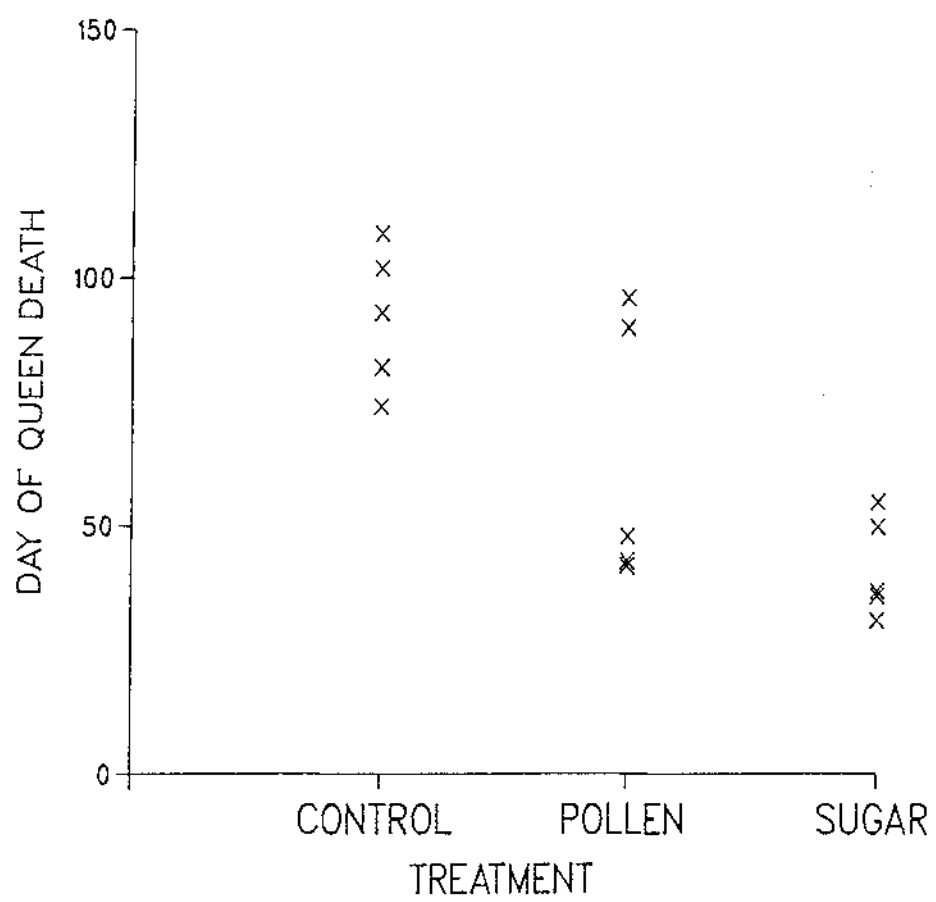


Table 1 Results of food supplementation experiment on colonies
in observation hives.

Treatment	Number of days from colony initiation to day of:-					Mean daily pollen income/brood area	
	Hive no.	Queen Death	First Male	First Queen	Colony Death	(g/cm ²)	P.I.
Sugar fed (S)	1	51	62	67	109	0.237	95
	2	31	29	-	47	0.154	864.5
	3	55	62	-	88	0.226	417.5
	4	33	28	-	49	0.221	137
	5	34	45	-	65	0.155	184
	mean	40.8	45.2	-	71.6	0.199	339.6
Pollen fed (P)	1	56	47	60	116	0.161	88
	2	100	80	-	110	0.182	361
	3	48	39	-	51	0.174	173
	4	90	51	-	104	0.186	449
	5	46	29	33	46	0.057	83
	mean	68	49.2	-	85.4	0.152	230.8

(table 1 cont.)

	1	97	71	—	97	0.204	855
	2	77	48	63	90	0.226	216
Control	3	82	59	—	99	0.205	436
(C)	4	102	55	—	102	0.154	246
	5	109	68	—	109	0.175	513
	mean	93.4	60.2	—	99.4	0.193	453.2

S-C	***	N.S.	—	*	N.S.	N.S.
S-P	*	N.S.	—	*	N.S.	N.S.
C-P	*	N.S.	—	N.S.	N.S.	N.S.

*, $P < 0.10$

**, $P < 0.05$

***, $P < 0.01$

N.S., Not significant

2.2.4 DISCUSSION

Pollen is the only protein source for brood growth, and since sugar supplementation failed to increase pollen collection (in relation to larval area and presumed demand) it is difficult to see how increasing nectar availability alone could advance sexual production as suggested by Cumber (1953). And indeed male production was not significantly advanced by sugar or pollen supplementation, although my number of replicates was rather small to provide a definitive answer. Cumber's statements were more concerned with queen production however, since these consume much more food as larvae than either males or workers. Unfortunately too few of my colonies produced queens for conclusions to be drawn as to its relative timing between the treatments.

The correlation between the amount of foraged pollen and the brood area agrees with the results of Pendrel and Plowright (1981) who found that the overall rate of pollen consumption is correlated with total area. This shows that the rate of pollen foraging is closely related to larval pollen demand.

The bees in the pollen treatment colonies made little use of the extra pollen provided, except on days of bad weather when their own pollen stocks may have run low. This may be explained by the fact that the pollen used for this treatment was provided by a commercial (Hastings) apiarist. The pollen, as received, was heat dried and had to be moistened before it was used. It may be that during the drying process some of the properties which make pollen attractive to bumble bees are lost. Although bumble bees do store pollen, it is likely that

they add some form of preservative which may also act as a signal to the bees on whether the pollen is "good" (bumble bees are less likely to eat pollen which has been exposed to the air for a few days than fresh pollen; personal observations))

Another possible explanation is that workers will continue to forage regardless of the amount of pollen available in the nest. Although my own results show that foraging is correlated with larval area (in agreement with Pendrel and Plowright (1981), Sladen (1912) and Brian (1952) have noted that pollen sometimes continues to be brought into, and accumulates in, colonies which no longer have any larvae to feed.

2.3 GENERAL DISCUSSION

As stated above, feeding sugar solution to bumble bee colonies is detrimental to their growth. Although this effect is difficult to account for by trophic-economic reasoning, there may be an important social (perhaps more strictly "socioeconomic") factor, that of worker "dominance" i.e. the tendency for some workers to withdraw from the "work" force and pursue their own reproductive ends. Röseler (1967b) suggested that if there are no or few larvae in the colony to be fed or if there are many younger workers to take over the feeding, the corpora allata and ovaries of older workers will begin to develop. This supports a statement made by Free and Butler (1959) that the presence of plenty of food, together with lack of strenuous activity, is one of the factors in the development of the ovaries of worker bees. The feeding of larvae and egg development are mutually exclusive (Röseler 1967b).

Dominant bees are highly aggressive towards the queen. Their presence in increasing numbers often leads to the production of male eggs, and eventually to the death of the queen. Once male eggs are laid, the colony begins its decline (Pomeroy 1977).

In colonies fed with sugar solution the pressure on the workers to keep the colony supplied with nectar would have been reduced. As the workers did not respond to this by increasing their pollen intake, it can be assumed that pollen was not a limiting factor to colony growth. Therefore the total foraging work load will have been reduced. This reduction in work load may have allowed many of the older bees to divert their efforts away from colony upkeep and towards the development of their own ovaries. The sooner aggressive (dominant) workers appear, the sooner their effect upon the queen will show. This is supported by the results from experiment two, where the queens' death came much earlier in the fed colonies than in the control. Though significant only at the 10 percent level, the length of colony duration is consistent with this hypotheses, i.e. colonies that were fed sugar syrup died sooner than those in the other treatments.

The above theory would predict earlier male production in sugar-fed colonies but experiment two failed to demonstrate this effect. Male production was examined in more detail in later experiments (chapters 3-5)

CHAPTER 3
DOMINANT WORKERS

3.1 INTRODUCTION

As stated above (chapter 2) the early decline of those 1984-85 season colonies which were supplemented with sugar solution may have been due to the early appearance of dominant workers in the hive. Van Honk et al (1980) showed that these bees are generally the older bees in the hive and are less affected by the queen's dominance pheromones. Attacks from these workers would presumably have some debilitating effect upon the queen. Röseler (unpublished, cited by van Honk and Hogweg, 1981) suggested that the stress of these attacks may induce her to begin producing unfertilised (male) eggs

In this experiment I attempted to test the hypothesis that the presence of dominant workers in the nest has an effect upon the queen, which results in earlier male production, and consequent curtailment of colony growth.

From the above statement of van Honk et al (1980) I equated worker age with degree of dominance. I therefore manipulated two sets of colonies to create a difference in the average age of their worker populations.

3.2 METHODS

Fourteen colonies were confined in heated conical hives (see section 2.2.2) immediately after the emergence of their first worker. Free foraging was not permitted. These hives were each ducted via a 6cm tunnel of wire mesh and cardboard to a vestibule comprising a vertical cylinder (11cm diameter, 20cm high) of wire mesh (3 strands/cm) with a margarine container lid on top with a small rock to hold it in place, and filter paper on the bottom (to absorb excess faecal moisture; replaced weekly).

Colonies were fed sugar solution from a plastic container (volume = 100ml) with a wick protruding through a hole in the lid, which was pressed up against the wire vestibule allowing the bees to lick the sugar solution from the end of the wick. Pollen (fresh-frozen from honey bee pollen traps) was packed dry, in small vials (volume = 4.4ml). These were taped to U-shaped lengths of wire which were hooked inside the rims of the conical hives. The vials were replaced daily, the number in each hive depending upon recent consumption rates.

Seven colonies were allocated to each of the two treatments, designated "old" and "young". Seven days after the first worker in each colony had emerged the worker population was reduced to five. Once a week, all bees, except for the foundress queen, were removed under red light and transferred to a jar. They were then placed in a refrigerator (7°C) for half an hour. The bees became slow and torpid, and were thus easily handled.

All bees were counted, and reproductives (males and young queens) were discarded. All new workers were marked by having one or both wings clipped, a coloured spot on the thorax (typist's correcting fluid mixed with ball point pen ink), or a combination of both. This mark was the same for all workers that emerged on any one week. By this method I could establish the age, within a week, for every worker.

A proportion of the workers were removed weekly to simulate the natural attrition which would have occurred if the colonies had been free-foraging. Observations of the 1984-85 season's colonies suggested an attrition rate of 40% of all new workers emerging weekly. This percentage was removed initially but after four weeks of data collection the colonies were becoming unnaturally crowded, therefore the removal rate was increased to 60%. As stated in the introduction, the bees in the "old" treatment hives should have an average older age than the bees in the "young" treatment hives. Therefore the colonies in the "old" treatment had their bees removed from the youngest age group whereas the colonies in the "young" treatment had their bees removed from the oldest age groups.

The date on which males and queens first emerged was recorded, also the date on which the foundress queen died. Observations of egg eating and agonistic interaction between workers and the queen were recorded when noticed.

The experiment was terminated after 15 weeks. All the young females had emerged by that time although males continued to be raised in some colonies.

3.3 RESULTS

Two colonies from the "old" treatment produced males in the third week after the start of the experiment. As it takes approximately 20 days from oviposition to emergence for a male, the male eggs in these two colonies were laid before the treatments differed and were not included in analyses. Two-sample t-tests were used to compare the treatments. A summary of the results is given in table 2.

Males appeared earlier in the "old" colonies than in the "young" colonies ($P = 0.029$). It is not known whether the first males that emerged were from eggs laid by workers or the queen. It is therefore possible that the date on which the last female egg was laid is a better indicator of the debilitating effect on the queen by the workers. Although if the workers are laying eggs, this indicates that the queen is losing her dominant status. The day on which the last female egg was laid was significantly earlier in the "old" treatment ($P = 0.045$)

Fewer workers were produced in the "old" treatment (mean = 87.3) than in the "young" treatment (mean = 227; $P = 0.013$). Although not enough of the foundress queens died during the duration of the experiment to justify statistical analysis, the results seen in table 2 show that 4 queens from the "old" treatment died before the end of the experiment. Whereas only 2 from the "young" treatment died early.

Table 2 Results of worker age experiment on colonies in observation hives.

Treatment	Hive no.	Number of weeks from colony initiation to day of:-				Total no. of Queens Emerged	Total no. of Workers Emerged
		First Male	First Queen	Last Female	Queen Death	Queens Emerged	Workers Emerged
Old	1	5	-	5	>17	0	128
	2	5	5	6	>17	6	100
	3	-	-	4	7	0	34
	4	-	7	8	7	9	93
	5	5	6	8	12	9	141
	6	4	9	9	>17	3	72
	7	4	-	5	10	0	43
	mean	4.6	6.7	6.4	-	3.9	87.3
Young	1	5	11	11	>17	31	216
	2	7	-	8	>17	0	200
	3	4	-	7	16	0	79
	4	7	7	9	8	16	396
	5	7	6	8	>17	16	141
	6	10	-	10	>17	0	304
	7	10	-	9	>17	0	252
	mean	7.14	8	8.8	-	9	226
P =		0.029	-	0.045	-	-	0.013

Aggressive interactions between the queen and the workers were noted on 14 different occasions. Only 3 of these were in "young" treatment hives. Aggressive interactions were also noted between workers on 8 different occasions, three of these were from "young" treatment hives. On 5 occasions egg eating by the queen was observed and on 2 occasions egg eating by workers was observed, all of these were in old treatment hives.

3.4 DISCUSSION

The results show that a population of old workers in a colony is more likely to result in the early production of males than a colony comprised of workers with a younger average age. This also resulted in a smaller number of workers being produced. If the assumption that increasing average worker age is equivalent to increasing dominance is accepted, the results are consistent with the hypothesis that the presence of dominant worker bees within the colony has a debilitating effect upon the queen. Pomeroy (1981b) observed that queens of B. impatiens confined with only five workers which were always less than one week old, continued to produce female eggs, and survived, for longer than queens in normal colonies. Similarly, my results, show that workers continue to be produced in colonies with few or no dominant workers long after they have ceased being produced in colonies with many dominant workers.

The early death of the four old treatment (table 2) queens may be explained as the result of worker dominance behaviour. Workers whose ovaries are increasing in size become a threat to the queen's dominant

status in the colony. She must therefore modify her behaviour to resist these dominant workers. She frequently attacks these workers, pushing them off the comb and destroying any egg cells they may have constructed (van Honk et al, 1981). Soon after this the dominant workers become openly aggressive, destroying the queen's egg cells, pulling and biting her until she leaves the nest and dies (van Honk et al 1981). My observations of aggressive and egg eating behaviour within the nest support this. They suggest that queens in the "old" treatment colonies may have been subject to more agonistic interactions than queens in the "young" treatment colonies. They also indicate that there may be increased worker-worker interactions within the "old" treatment colonies.

CHAPTER 4

THE EFFECT OF SUPPLEMENTARY SUGAR FEEDING ON WORKER OVARY SIZE

4.1 INTRODUCTION

The primary purpose of this experiment was to determine one of the factors which may have led to the early decline of colonies seen in chapter 2.

Free and Butler (1959) stated that the presence of plenty of food together with a lack of strenuous activity is one of the factors contributing to the development of worker ovaries. These bees become highly aggressive towards the queen to the extent that she may lose her dominant status within the colony (Free 1955a).

The 1984-85 season results show that colonies that are fed sugar solution are smaller than those that are not (see chapter 2). If this is due to the early appearance of dominant workers within the colony, a greater percentage of workers with large ovaries would be expected in the fed treatment than in the non-fed treatment.

The aim of this experiment was to determine whether feeding sugar solution to bumble bee colonies resulted in increased ovary sizes for the workers within those colonies.

4.2 METHODS

Twenty four small colonies were placed in softboard hive liners (figures 1a and 1b). These were put into pumice concrete boxes in the study area beside the Tiritea Stream. In contrast to the method used in chapter 2, only one concrete box was used for each colony. Fibreglass Batts were used as insulation. Each colony was provided with a small bottle (volume = 15ml) of sugar syrup and a vial (volume = 4.4ml) of pollen to last them until they started to collect their own. Colonies were not put out until they had produced 8-12 workers. They were placed in the field in two groups, with an even number of hives in each. The colonies in each group were split evenly into two treatments. These treatments were the same as in the 1984-85 field experiment i.e. the provision or not of a continuous supply of sugar solution (see chapter 2).

After fifty six days in the field, all colonies were killed by blocking up the entrance holes with cloth, then placing three teaspoons of ethyl-acetate beneath the lid. This was performed at night when the majority of the foragers would be in the hive. The colonies were then frozen. The night after the colonies were killed I went back to the field and picked up any bees I found inside the empty concrete boxes. These were also frozen with their appropriate colonies.

All the workers were dissected under a stereo dissecting microscope, and the length of the longest oocyte from each worker was measured.

4.3 RESULTS

After fifty six days two colonies (one from each treatment) had died out completely, and three colonies (two from the sugar treatment) had lost their queens.

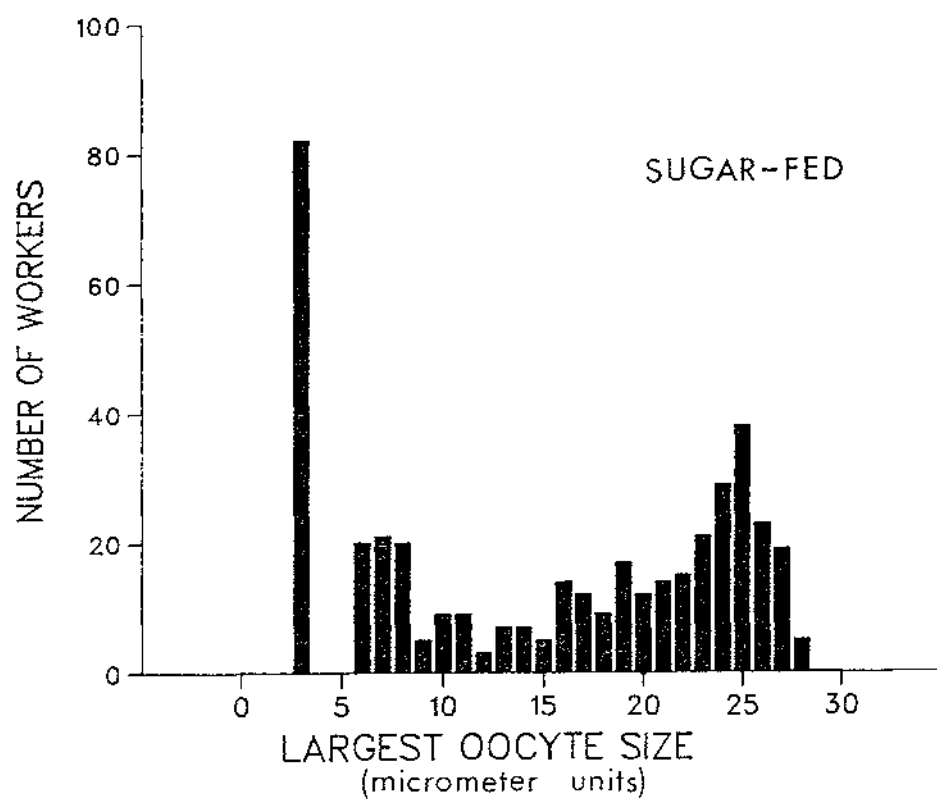
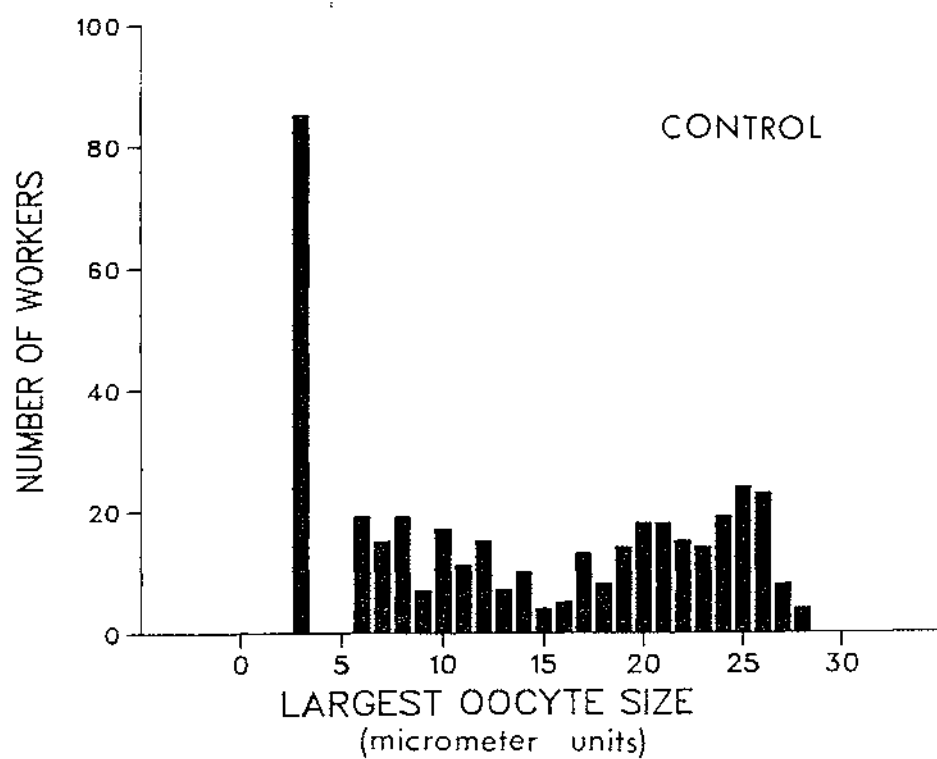
I had originally hoped to "catch" the colonies at a point where the most "advanced" were just starting to produce males. But instead all the colonies in both treatments were producing reproductives at the time they were discontinued. They also all had workers with large ovaries capable of egg production.

I had also expected the oocyte-size data to fall into two distinctly separate categories; i.e. dominant workers with large oocyte and non-dominant workers with small oocytes. I would therefore be able to discern the proportion of dominant bees within each hive. Unexpectedly the distribution of ovary sizes was more continuous (see figure 6). I therefore took the mean oocyte size for workers in each hive. This was larger in the fed treatment (mean = 1.87mm) than the non-fed treatment (mean = 1.65mm, $P = 0.0302$, Mann-Whitney, 1-tailed).

It seems likely that a large number of these colonies had reached the equilibrium position at which there is no further travel up the dominance spectrum. As such many of the other colonies may have "closed the gap" between the average ovary sizes.

Figure 6.

Frequency distributions of the lengths of the largest oocyte per worker, with all workers pooled from all colonies in each treatment. Oocyte "size" is the length in ocular micrometer graduations (one graduation = 0.12mm).



4.4 DISCUSSION

The results show that feeding sugar syrup to bumble bee colonies leads to an increased average ovary size for the workers within those colonies. This suggests that dominant bees were present in the nest earlier in sugar treatment colonies than in control colonies. As the dominant bees aggression towards the queen (mentioned above) increases, this may induce her to start producing male eggs (Röseler 1967b). These dominant workers also begin to lay male eggs (Katayama, 1971, 1974), though, the foundress is generally the first to do this (Pomeroy 1977).

As male production is generally irreversible and the production of new workers soon ceases, its timing becomes a major factor affecting the length of a colony's life (Pomeroy 1977, 1979). This in turn (depending upon the growth rate of the colony) has an effect upon its final size. As the feeding of sugar syrup to bumble bee colonies induced the growth of worker ovaries sooner than normal, the early decline of the colony can therefore be expected. This was seen in the 1984-85 field experiment (see chapter 2).

As the older workers with large ovary sizes gain dominant positions within the nest, they, like the queen, begin to produce queen (dominance) pheromone (van Honk 1982). As more bees reach the dominant position the younger bees must therefore come under increased pressure to remain submissive. Most colonies probably reach an equilibrium point where a set percentage of workers obtain a dominant position and when this is reached it does not increase. Van Honk and Hogweg (1981)

suggest that 25% of the workers are in the dominant group.

CHAPTER 5

CROWDING

5.1 INTRODUCTION

Many researchers have pointed to the worker/larva ratio or larval food intake as important factors stimulating the production of new queens in the bumble bee colony (Frison 1926; Cumber 1949a; Free 1955c; Plowright and Jay 1968, 1977). Röseler (1967a) suggested that once the bees reach a certain density on the comb, the queen changes to the production of male eggs. He then suggests that it is the presence of these male larvae which stimulates the workers to discard excess larvae until the appropriate worker : larvae ratio is reached. The remaining female larvae receive the optimal feeding rate and develop into queens.

In testing Röseler's hypothesis, Pomeroy and Plowright (1982) found no difference between the timing of male production in colonies with different densities of workers on the comb. Though they did report a slight non-significant "trend" towards shorter pre-male periods where more workers were present, they associated this trend with slight differences in the mean colony-starting dates.

When starting new colonies for experiments in the laboratory, I noticed that if they are allowed to produce more than 8-12 workers within the small starter boxes, many of the foundress queens began to lay male eggs and sometimes queens start being produced. If they were shifted to a larger container, male production did not occur so soon.

The following experiment was designed to test the hypothesis that worker density in a hive affects the timing of male egg production.

5.2 METHODS

Twenty-six colonies, each with a naturally gained complement of four workers, were randomly allocated to two treatments, thirteen in each. In one treatment an additional eleven callow workers were given to each colony to bring the total number to fifteen. In the second treatment worker numbers remained at four. All workers had their wings clipped.

The starter boxes were modified so as to increase the height of the brood chamber by taping four plywood walls with a sliding perspex lid to the top of the brood chamber (see figure 7). Additional plywood walls were added if the colony became too big for the first box.

From Monday to Friday of each week, between 9:00 and 11:00am all new workers were removed. These were identified by the presence of unclipped wings. All "old" (wing clipped) workers that had died were replaced with callows (wings clipped). Males and queens were also removed. The number of new workers produced and the date the first male and queen emerged was noted. All records were terminated when the colonies had ceased raising any females.

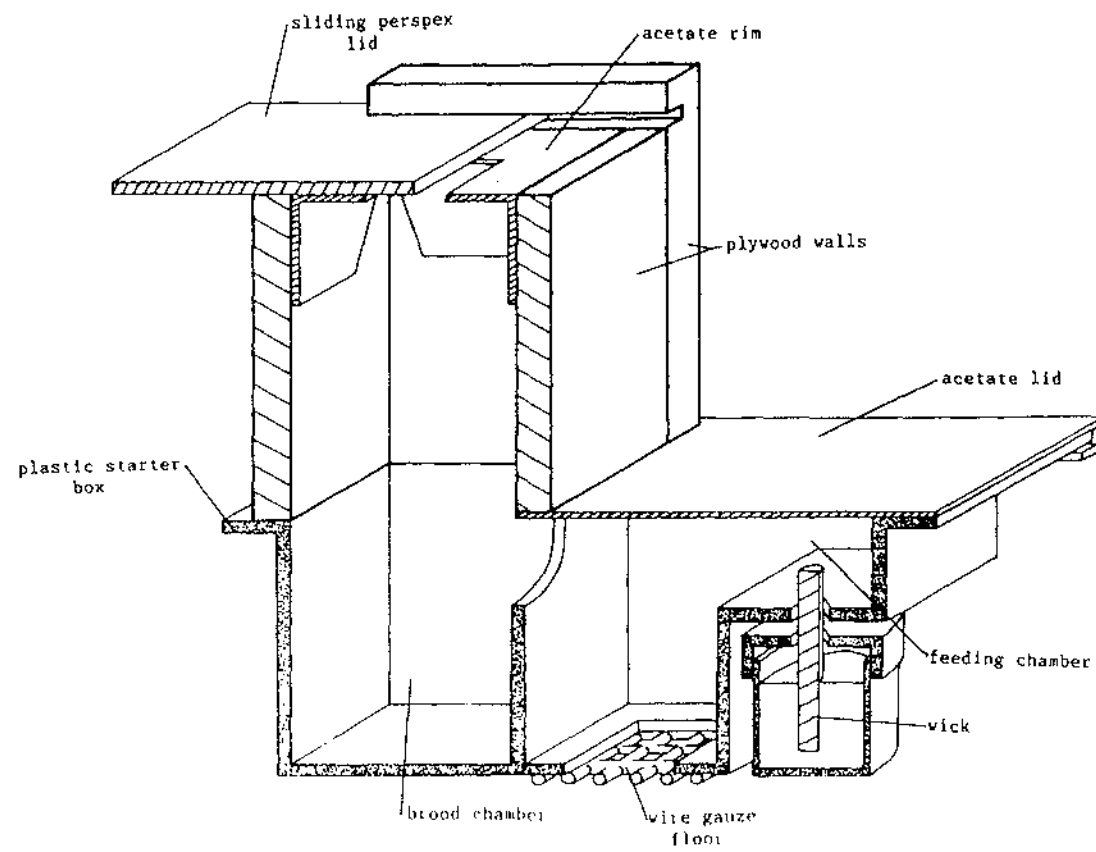
Figure 7.

Perspective cut away view of modified starter box. The acetate rim stopped bees from escaping when the perspex lid was removed during data collection (bees were removed with tweezers through the square hole in the acetate).

External dimensions; height = 115mm, width = 75mm, length = 170mm

The starter box has been drawn diagrammatically to avoid infringing a provisional patent application.

Drawn by I. McGee



5.3 RESULTS

In two colonies, one from each treatment, males appeared within fourteen days from the beginning of the experiment. As males take twenty days from egg laying to emergence, these males hatched from eggs layed before the treatments were imposed. One foundress queen died, from a fifteen worker treatment, four days after the beginning of the experiment. These three colonies were discarded.

The mean number of days from colony initiation to emergence of the first male was earlier for the fifteen-worker treatment (mean = 44.55) than the four-worker treatment (mean = 57.42) ($P = 0.03$, Mann-Witney U-test).

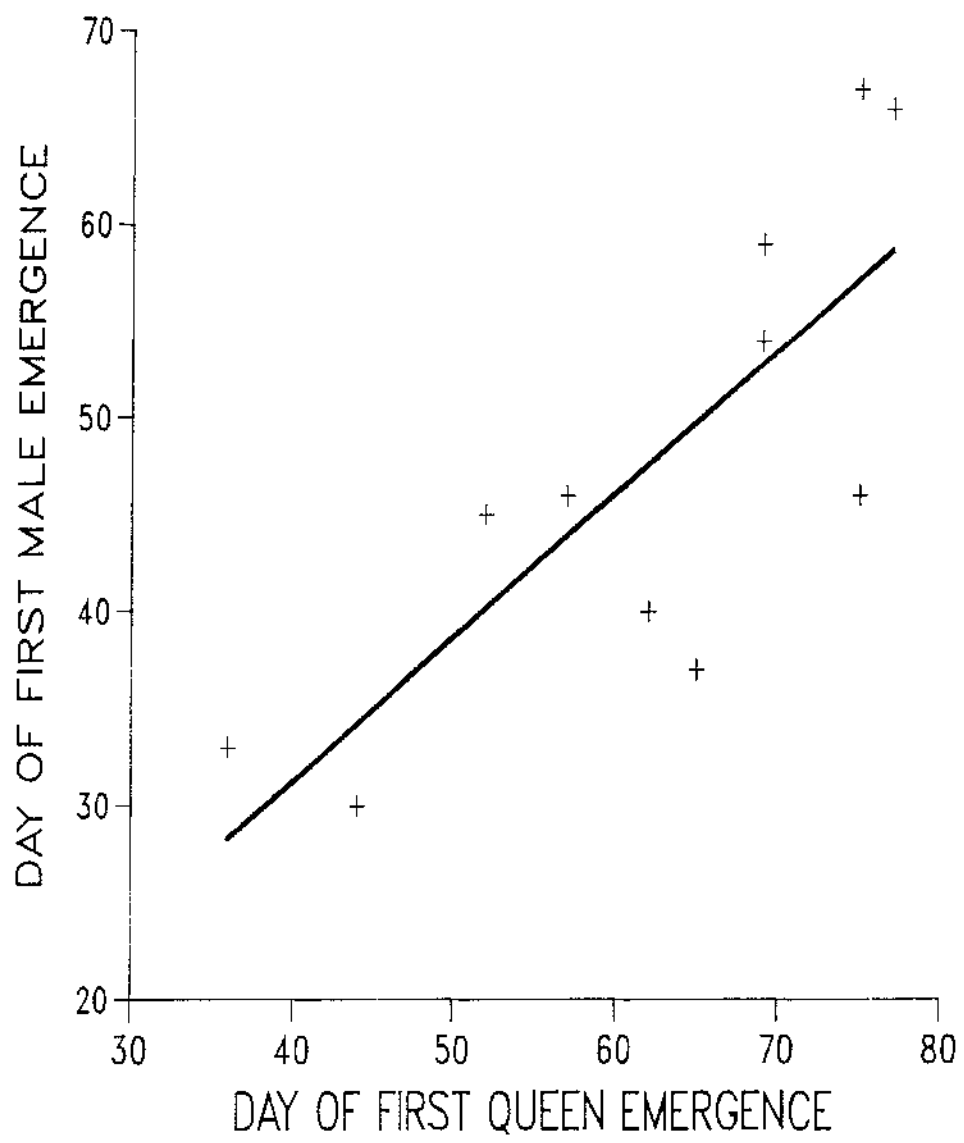
Queens were produced in 6 colonies from the four-worker treatments and in 5 colonies from the fifteen-worker treatment. There was a correlation between the lengths of the periods from initiation to first male, and initiation to first queen emergence ($n=11$, $r = 0.794$, $P > 0.005$) (figure 8). There was no effect of worker numbers on the timing of queen production ($P = 0.583$, Mann-Witney U-test). As expected there was a significant difference between the mean number of workers produced 4-worker treatment (mean = 98.8) and that of the 15-worker treatment (mean = 84.64) ($P = 0.058$, Mann-Witney U-test, 1-tailed).

Figure 8.

Relation between day of first male emergence and day of first queen emergence. Day 0 is the day of colony initiation.

Regression equation: $X = 22.1 + 0.836Y$

X = day of first queen emergence Y = day of first male emergence



5.4 DISCUSSION

Contrary to Pomeroy and Plowright (1982) the results obtained above show that the density of workers in a hive can effect the timing of male production. This is in agreement with Röseler's hypothesis (1967a), but it is possible that worker density per se is not the main factor which affected the queens. The accumulative dominance effect of workers, coupled with the amount of time and the number of such workers may be of greater importance.

Bombus (Pyrobombus) perplexus Cresson was chosen by Pomeroy and Plowright as their experimental species because of its similarity to Bombus (Pyrobombus) hypnorum used in Röseler's (1967a) study. B. perplexus colonies have a very short growth phase before the production of reproductives. As queens and males emerged from the third brood of their colonies, the eggs from which they hatched would have been laid on the unemerged second brood cocoons. The type of eggs (female or male) would therefore have been determined while only the first brood workers had been in the nest for three weeks. In the above experiment male eggs were determined after the workers had been in the nest for an average of 4 to 5 weeks.

Two possible explanations for the different results present themselves. The first is that in Pomeroy and Plowright's experiments there was not enough difference in the density of workers between the treatments (2, 4 and 8 workers in each treatment). Whereas in my own experiment an increase of 11 extra workers may have been enough to affect the queen.

The second explanation is that it may not be the effect of the number of workers per se which induces the queen to lay male eggs but the number of dominant workers. Pomeroy 1981 showed that in the presence of a queen, worker ovary weight increases with age. The workers in Pomeroy and Plowright's (1982) experiment were not old enough to have become very dominant at the time male eggs were being laid (approximately three weeks) whereas the workers in my experiment were much older at this time (approximately five weeks). With more time in which to become dominant, they therefore would have had a greater debilitating effect upon the queen.

The initiation of queen production seems to be associated with the timing of male production. This agrees with the findings of both Pomeroy and Plowright (1982) and Röseler (1967a), and lends support to Röseler's hypothesis that the presence of males stimulates the production of queens.

CHAPTER 6

GENERAL DISCUSSION

Although it has been suggested by Cumber (1949a), Gurr (1957a) and others, that food requirements play a primary role in determining the size to which bumble bee colonies develop, the experiments described in chapter 2 show that this may not necessarily be the case, and that social factors may play a more important role. The results show that feeding pollen to bumble bee colonies has no effect upon the size of the colonies. This indicates that in the year of the experiment pollen was not a limiting factor, though it should be pointed out that in bad seasons when there is little forage available pollen feeding may be advantageous to the colony. The results of the sugar feeding trials showed that sugar feeding has a debilitating effect upon the colony. The sugar trials also showed that the length of the queen's life (from the colony initiation date) was shorter in sugar fed colonies than in non-fed colonies. This indicates that social factors may play an important role in the size determination of the colony. Most of this thesis has been devoted to studying how one of these social factors, dominant workers, affects the size to which a colony grows, and how some external factors, specifically ease of food gathering, affects the proportion of these workers in the colony.

The timing of male production is highly important in the regulation of colony size because the appearance of males in the nest often marks the end of worker production (Pomeroy 1979). bumble bee males are haploid and develop from unfertilised eggs (Crozier 1975). The fertilisation of eggs is under the control of the queen (Snodgrass

1956; Flanders 1946; Wigglesworth 1972). In many other bee genera egg fertilisation can be affected by oviposition site stimulus (Knerer 1980; Butler 1954; Michener 1974), or environmental factors (Knerer and Plateaux-Quenu 1967), though there is no evidence to suggest that either of these factors stimulates male production in bumble bees. Depletion of sperm is not a factor in male production (Röseler 1974). Röseler (1967a) suggested that social factors, specifically worker density, stresses the queen such that she ceases to be able to fertilise eggs. My experiment in which the density of workers on the comb was manipulated (chapter 5) confirmed Röseler's suggestion. Pomeroy and Plowright (1982) failed to support Röseler's worker density hypothesis. The difference between my results and those of Pomeroy and Plowright may be accounted for by the fact that their B. perplexus colonies all produced males early, when their workers were still very young. In my own experiment the workers were much older when males were produced. As, even in the presence of a queen, worker ovary weight increases with age (Pomeroy 1981b); and by implication dominance increases; it is possible that dominance plays a role in the timing of male production. A high density of dominant workers may therefore have a greater debilitating effect upon the queen than a low density of dominant workers, or even a high density of submissive workers.

Van Honk and Hogweg (1981) described the existence and formation of what they termed the "elite group", to which they ascribe the following characteristics (I have termed this "decadent" behaviour):-

"1/ The elite group distinguishes itself from the 'common workers' by a higher 'activity index' i.e. the members interact more frequently with all the other members of the nest.

2/ They show less submissiveness towards the queen than do the common workers.

3/ They show much greater aggression towards workers 'in transition' from the common worker-group towards the elite group.

4/ Members of the elite group lay eggs.

5/ Elite workers (because of their stage of ovary development) are less susceptible to the inhibitory effect of the queen's pheromones and can thus endure her proximity."

Van Honk et al (1981) showed that these decadent workers actively seek the immediate proximity of the queen. They follow her all over the comb and take up positions at her head whenever she stops and repeatedly touch her with their antennae. It seems contradictory that those workers that have the greatest exposure to the queen's pheromones are the first to lay eggs. Van Honk and Hogweg (1981) and Röseler and Van Honk (1985) have suggested that these workers become less susceptible to the inhibitory effect of the queen's pheromones because of their advanced stage of ovarian development. Van Doorn and Heringa (1986) disagree with this, pointing out that these workers sought the proximity of the queen from their second day of life. Pomeroy (1981b) has shown that physical contact is the main avenue of dominance communication and that gaseous pheromone is a minor factor in the operation of the dominance signal. These decadent workers are therefore the only ones in continuous contact with the dominance pheromone. It may be that sporadic contact with the dominance

pheromone (as the queen moves over the comb) is most important for effective inhibition of worker ovary development. Continuous contact may have no inhibiting effect at all upon the workers. The fact that the queen (and egg laying workers) does not inhibit herself, though she is presumably saturated with dominance pheromone, lends support to this argument.

The experiment described in chapter 3 shows that males are produced earlier in colonies which have a higher proportion of older (and presumably dominant) workers. The number of workers produced was substantially smaller in these colonies. This supports the statement made above that the timing of male production is important to the regulation of colony size. These results also indicate that there is a debilitating effect upon the queens from dominant workers. This agrees with the results of Van Honk et al (1981) who showed that the decadent workers compete with the queen for opportunities to lay eggs to the point where they become openly aggressive. This can lead to the queen being pushed off the comb or to her death. A dominant bumble bee female is characterised by occupying a preferred territory in the nest e.g., egg batches or clumps of young larvae. When a queen has lost her dominant status she frequently rests on the periphery of the nest and sometimes on empty cocoons, and often she hides between the combs (Röseler and Röseler 1977). The most dominant worker will then take on the position of a pseudoqueen. These dominant workers, like the queen, inhibit the development of other workers' ovaries through the production of pheromones (van Honk (1982) suggested that these pheromones are similar to those produced by the queen). The earlier an elite group forms the earlier it will, presumably, affect the queen.

As stated above decadent workers may kill or push a queen off the comb but before this happens the queen is presumably put under considerable stress from increasing aggression and starts to produce males.

Why do decadent workers appear earlier in some colonies than in others? One possible explanation is the inherent characteristics of the queen. Voveikov (1953) suggests that the ability to raise a colony differs between queens of different sizes. He says that small queens are able to start colonies easier than large queens, but that they break down sooner due to a lower fecundity. This small size may limit the queens ability to suppress worker oogenesis. If so, small queens may suffer an earlier appearance of decadent workers.

Many authors have stated that control over the workers by the queen declines with age (Sladen 1912, Free and Butler 1959, Röseler and Röseler 1977). However, Pomeroy's work (1981a) shows that queens have the potential to exert an undiminished dominance signal for much longer than the signal actually remains effective in real colonies. He suggests the appearance of egg laying workers earlier in the colony cycle could be caused by three factors:-

"1/ expansion of the brood comb to the extent that workers fail to encounter the queen often enough to be suppressed by her.

2/ the cumulative effect over time of a low level of oogenesis due to queen's effecting incomplete suppression.

3/ inhibition of dominance signal output from the queen due to the influence on her of workers which achieved enlarged ovaries via mechanisms 1 and 2".

He also suggests another possible factor being the degree of pollen and/or "labour" surplus. Free (1955a) and Free and Butler (1959) have also suggested that surplus pollen in the colony at the peak of the season, may encourage workers to undergo ovarian enlargement. Free (1955a) showed that an absence of pollen prohibited oogenesis. However the converse, i.e. a surplus of pollen, may not necessarily promote oogenesis. This is supported by my laboratory results (chapter 2), which show that in colonies that were fed supplementary pollen the effects of decadent behaviour, decadence being a result of increased oogenesis, were not seen any sooner than in control colonies.

Pomeroy (1981b) showed that under conditions of pollen surplus the act of feeding larvae failed to reduce worker oogenesis. Pomeroy's colonies were totally enclosed, and the workers were not required to forage for food. The number of larvae requiring to be fed was small (5-10 larvae in his two worker experiment and 10-20 larvae in his five worker experiment: Pomeroy personal communication), this indicates a low labour demand on the workers. I suggest that it is this low labour demand which encourages ovary development in workers. The experiments described in chapter 2 were designed to reduce the demand for nectar collection and to transfer this labour to pollen collection. My experiment in which the oocyte sizes of workers were measured shows

that colonies that are supplied with surplus sugar have a larger proportion of their workers with enlarged ovaries than those colonies without surplus sugar. I suggest that the extra "bee time" created by the reduced labour demand was in fact taken up by a select group of workers (who later formed the "elite group") and was spent on ovary enlargement. In this way workers had an opportunity to become dominant and therefore lay eggs earlier than in colonies which were not supplied with an easily accessible supply of surplus sugar.

The major controversy related to the theories of the evolution of sociality has been whether the workers of eusocial Hymenoptera were "initially altruistic or oppressed" (Michener and Brothers 1974). Darwin (1860) attributed the altruistic properties of Hymenopteran workers to individual selection on the queens. Alexander (1974) agreed with this stating that the workers' reduced fecundity is caused by the queens manipulation.

Hamilton (1964, 1972), Trivers and Hare (1976), West Eberhard (1975), and Dawson (1976) have suggested that the supposed altruism of Hymenopteran workers is in fact selfish behaviour. They point to the unique haplodiploid sex determination in Hymenoptera. Sisters, of a monogamous outbred mating, share an average of $3/4$ of their genes by descent, whereas only $1/2$ of their genes are shared with their own offspring. This unusual relatedness makes it more in a worker's interest to help the queen produce more sisters than to produce her own female offspring.

In bumble bees the queen "dominates" the workers via a queen pheromone which is picked up by worker antennal sensillae and inhibits the activity of the corpora allata and hence the production of gonadotrophic hormone (Röseler et al 1981). Though as Pomeroy (1981b) pointed out "the worker has to have been party to the evolution of this communication system, i.e. the queen cannot impose reproductive restraint on a worker without involving the worker's sensory/transduction mechanisms". It would therefore seem likely that bumble bee workers voluntarily allow their reproductive abilities to be suppressed by the queen for the benefit of increasing their genetic input into the next generation via their sisters (young queens).

Towards the end of the colony's life, when males are being produced, this benefit becomes reduced. Workers are only related to their brothers by $1/4$ whereas they are related to their own sons by $1/2$. Therefore at this point it is in the workers' interest to stop the queen from laying, and to start producing their own male eggs.

Since decadent behaviour is observed before the production of male eggs by the queen, some other signal (other than the presence of male eggs and larvae) must be detected by the workers that indicates the imminent change to the production of reproductives. Colonies seem to change to reproductive production once they have reached a "point of prosperity". It may be that some factor related to a colony's prosperity acts as a "Prosperity Signal" (eg. a reduced labour demand) to the workers to "switch off" or reduce their receptivity to the queen's pheromones.

The social system within the colony then appears to revert to "parental manipulation" where the queen attempts to suppress ovarian development in workers through aggression (Wilson 1971; Michener and Brothers 1974). Not all workers can become egg layers for who would then forage for the colony? Those bees that do not gain dominant laying positions are better off foraging to raise nephews. But what is the mechanism which allows some workers to become dominant and lay eggs while the queen is still in the nest (producing dominance pheromones), but stops other workers from doing so? Older workers have been shown to have slightly larger ovaries than younger workers when the colony is still early in its growth phase (Pomeroy 1981b). Because of their different physiological state these bees (which are in a higher dominance position) may be able to detect the Prosperity Signal, whereas bees lower down the dominance spectrum may not. Combined with this there may be a feed back system where, as the nest becomes less prosperous, the Prosperity Signal may be reduced to an undetectable level. The reduced nest prosperity is created by a loss in forager numbers. This loss is caused by natural attrition in the wild, with no replacements (with no queen to lay fertilised eggs, only males are being produced), and also by the general upward movement of workers upon the dominance ladder.

Nevertheless these workers end up better off than when the queen was dominant because their relatedness to their nephews is $3/8$ whereas it is only $1/4$ to their brothers.

How do the results of the present study relate to the commercial management of bumble bees for crop pollination?. They have shown that the direct feeding of bumble bee colonies with sugar solution has a debilitating effect upon colony size. Though as has been pointed out by Heinrich (1979a), competition for food from flowers may be so high as to be equally harmful. A compromise between these two problems would be to grow "bumble bee flower" crops beside which colonies are placed to develop until ready to be transported to the crop requiring pollination. This might allow a readily available food source for the bees without making it so easily available that the proportion of dominant workers in the colony is increased. Inevitably the wild population of bees will increase around any bumble bee farm of this sort and again competition may occur. Feeding stations may be a better solution (Pomeroy personal communication). This being an unlimited source of sucrose some distance from the hive with a distinctive flavouring that the bees recognise from their laboratory feeding.

Another possible way in which dominance behaviour in the colony can be kept low is the use of artificially synthesised queen pheromones. This pheromone is known to inhibit the maturation of worker ovaries (Röseler et al 1981) and van Honk (1982) has suggested that its structure is very similar to that of the honey bee queen pheromone. Research is presently being undertaken to identify it (D. Greenwood personal communication).

APPENDIX 1

CONSTRUCTION OF 1984-85 FIELD HIVE

When the colonies in the laboratory starter boxes had gained a compliment of 8-12 workers, the starter boxes were placed into the bottom of plastic hive-liners (see figure 9). The acetate lid was removed from the starter boxes and insulation material was placed on top of the comb. Two forms of insulation material were used. The first twenty-two hives introduced to the field utilised fibreglass batts. The last eighteen used carpet underfelt. The plastic liner was then placed in a pumice concrete "box" with another such box inverted on top, this having an entrance hole 5cm in diameter, in one side. A solid concrete lid was placed on top to prevent rain from percolating through the porous concrete beneath. These lids were painted with various shapes and colours so as to enable the bees to distinguish their own hives from their neighbours'. On the front of each concrete hive, over the entrance hole, was a plastic vestibule (two-litre ice-cream container).

A 1cm square hole was cut into the side of the ice-cream container to act as an access hole for bees. Into each hive was also placed a small container of pollen (4.4ml) and a small 15ml jar of sugar solution with a wick to aid the bees in their transition from the laboratory to the field.

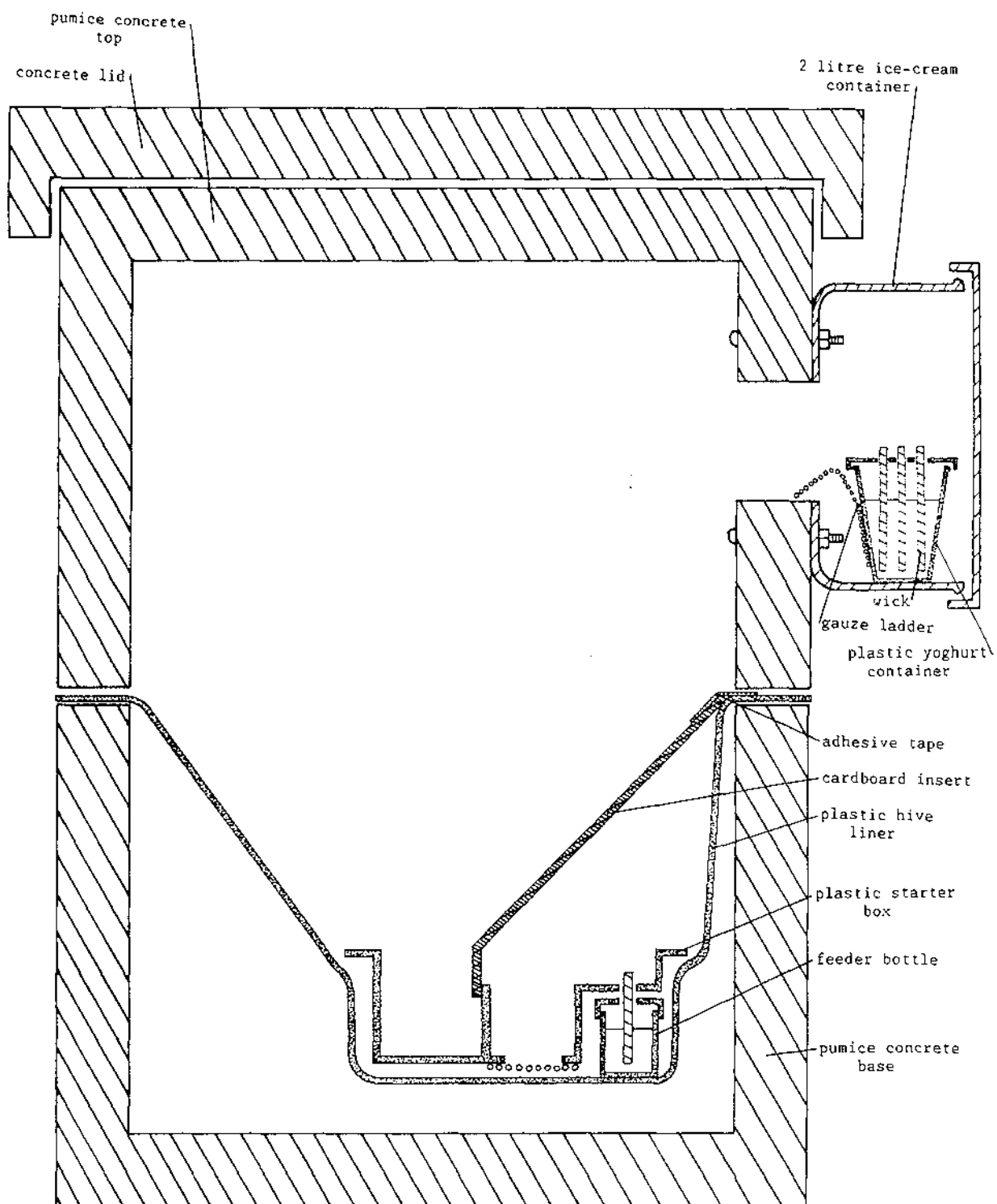
Figure 9.

Mid-vertical section of pumice concrete field hive with plastic hive liner and starter box. With the triangular shaped cardboard wall in place, an inverted, four sided pyramid shape was created to support the upward and outward growing brood.

External dimensions; height = 450mm, width = 330mm

The starter box has been drawn diagrammatically to avoid infringing a provisional patent application.

Drawn by I. McGee



Sugar solution (for the sugar fed treatment) or water (for the control) was contained in a 225ml yoghurt container with three wicks protruding through holes in the top (these are 5mm diameter felt pen filters for water soluble ink).

After the first hives had been in the field for three weeks it became obvious that excessive faecal moisture was collecting in the bottom of the plastic liners to the extent that a distinct pool was being formed. To drain this I pierced holes in the base of both the starter boxes and liners. All subsequent hives were provided with drainage holes.

APPENDIX 2

LIGHT BOX

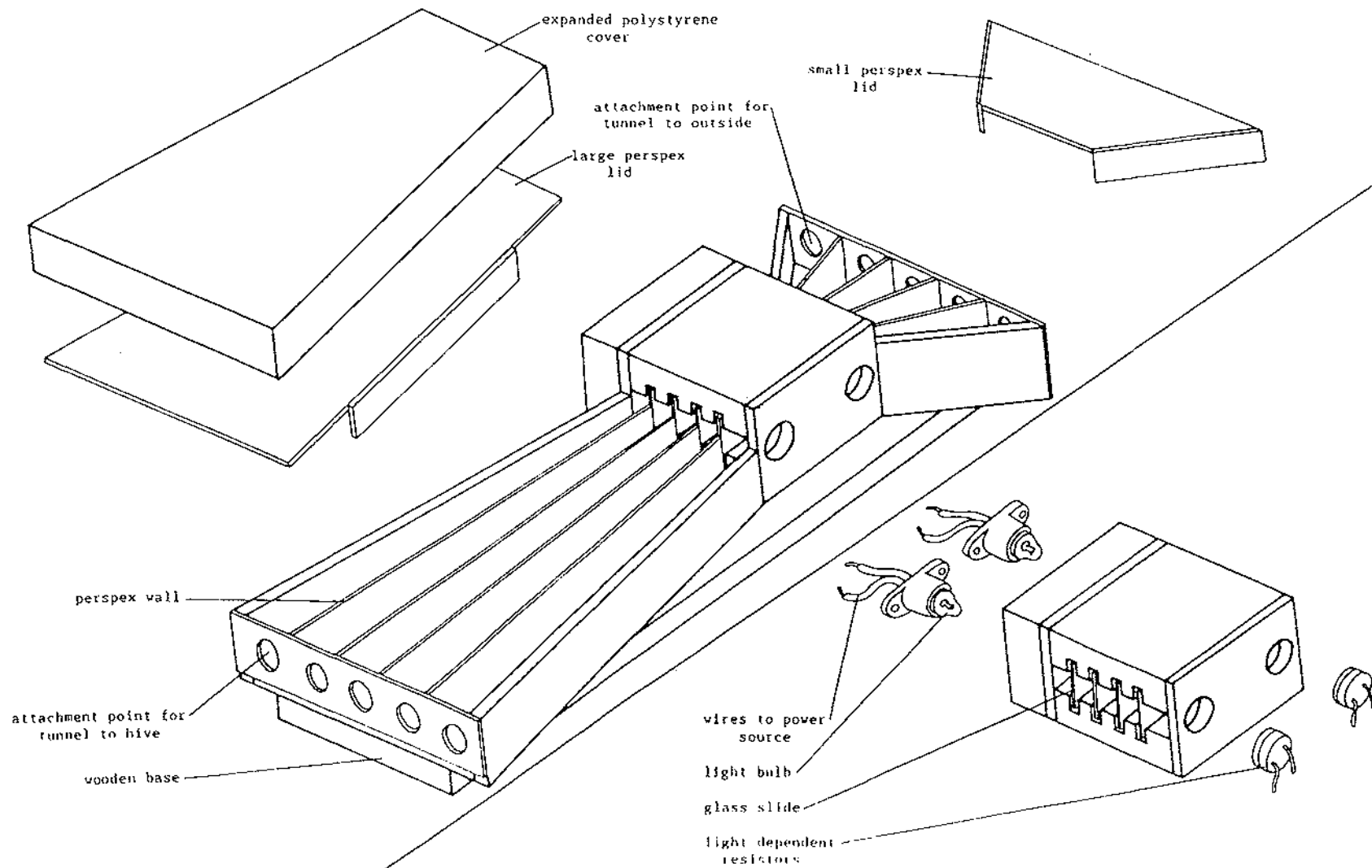
The light box was a wooden and perspex apparatus (see figure 10) at which five tunnels converged. While going through the central portion of the light box, bees would pass through two light beams which were emitted by a pair of lens-end bulbs shining through red cellophane (as bumble bees do not detect red light they therefore would not be confused by light from the bulbs). When a bee interrupted the light beams while going towards the exit, nothing would happen. If the beams were interrupted by a bee heading towards the hive, an electronic circuit (designed and built by G. Baker and G. Nicholls) would sense them and produce a high pitch sound or "beep". A light would also indicate which light box was emitting the signal. Using this apparatus I could simultaneously monitor bees entering the three groups of 5 hives.

Figure 10.

Perspective view of Light Box with lids raised (A). The central portion is drawn separately (B). Each "lens end" light bulb, which was wrapped in red cellophane and attached to the side of the light box, sent a beam of red light through the glass slides (these were painted black except for two clear circles to allow the light through) onto the light-dependent resistors. The small perspex lid was covered with adhesive masking tape to stop light from entering the tunnels and thus preventing the bees from trying to get out.

External dimensions; height = 48mm, width = 140mm, length 320mm

Drawn by I. McGee



A

B

APPENDIX 3

CORBICULAR POLLEN WEIGHTS FROM INCOMING FORAGERS

Size catagory				
Date	1	2	3	4
20/11/84	0.0085	0.0124	0.0151	0.0227
	0.0084	0.0124	0.0152	0.0228
	0.0038	0.012	0.0172	0.0212
	0.0035	0.012	0.017	0.0211
	0.0033	0.01	0.0188	0.021
	0.004	0.0103	0.0186	0.0215
	0.0044	0.0108	0.0174	0.0222
	0.0043	0.0107	0.0175	0.0221
	0.0067	0.0103	0.0202	0.0238
	0.0073	0.0104	0.0206	0.0234
	0.0057	0.0147	0.0209	0.0258
	0.0053	0.0144	0.021	0.0258
	0.0073	0.0126	0.0179	0.0245
	0.0071	0.0131	0.018	0.0242
	0.0084	0.0151	0.0194	0.0263
	0.0087	0.0147	0.0192	0.0265
	0.0077	0.0122	0.0156	0.0226
	0.0075	0.0126	0.0156	0.0221
	0.0097	0.013	0.0166	0.0247
	0.0094	0.0131	0.0161	0.0261

Mean	0.0065	0.0123	0.0179	0.0235
22/12/84	0.0095	0.0105	0.0209	0.0252
	0.0092	0.0102	0.0206	0.0217
	0.0057	0.0104	0.0206	0.0218
	0.0052	0.0107	0.0206	0.0218
	0.0035	0.0126	0.0186	0.0268
	0.0037	0.0129	0.0186	0.0269
	0.0042	0.0138	0.019	0.0225
	0.0096	0.0139	0.019	0.0225
	0.0077	0.0096	0.0192	0.0239
	0.0088	0.01	0.0194	0.0238
	0.0074	0.0142	0.0166	0.0266
	0.0076	0.0134	0.0166	0.0265
	0.0029	0.0113	0.0189	0.0273
	0.0027	0.0114	0.0188	0.0274
	0.0064	0.0133	0.0171	0.0246
	0.0067	0.0141	0.0178	0.0241
	0.0047	0.0103	0.0209	0.0227
	0.0046	0.0096	0.0199	0.0231
	0.0055	0.015	0.0185	0.0208
	0.0052	0.0152	0.0185	0.0203
Mean	0.0060	0.0121	0.019	0.024
8/1/85	0.0088	0.0118	0.0204	0.0226
	0.0089	0.0117	0.0204	0.0225
	0.0094	0.014	0.0207	0.0213

	0.0094	0.0149	0.0201	0.0211
	0.0045	0.0125	0.0189	0.0254
	0.0043	0.0127	0.0183	0.0235
	0.0091	0.0097	0.0158	0.0261
	0.0096	0.0102	0.0157	0.0261
	0.0049	0.0148	0.0151	0.0247
	0.0047	0.0148	0.0156	0.0246
	0.0088	0.0134	0.018	0.0232
	0.0051	0.0134	0.0201	0.0233
	0.0066	0.0115	0.0163	0.0221
	0.0074	0.0116	0.0163	0.0221
	0.0087	0.0108	0.0195	0.0264
	0.0083	0.0094	0.0197	0.0269
	0.003	0.0151	0.0168	0.0274
	0.0041	0.015	0.0171	0.0283
	0.0052	0.0123	0.015	0.0219
	0.0044	0.0153	0.0152	0.0219
Mean	0.0067	0.0115	0.0177	0.024
Grand Mean	0.0064	0.0119	0.0182	0.0239

APPENDIX 4

THE EFFECT OF WEATHER CONDITIONS ON FORAGING

In an attempt to determine the influence of weather on bumble bee foraging during the first season's laboratory experiments, I made a note of the weather type on each data collection day. The type of weather for any one day was placed into one of three broad categories:-

- 1/ generally fine weather with no rain
- 2/ showers and drizzle (light rain)
- 3/ heavy rain

Pollen intake per unit larval area was related to weather type.

Weather conditions made a significant difference to the amount of pollen brought in to the hives ($P < 0.001$, Mann-Whitney U test). Workers did very little foraging during heavy rain. Drizzle and showers also reduced the amount of foraging but to a smaller extent (see figure 11)

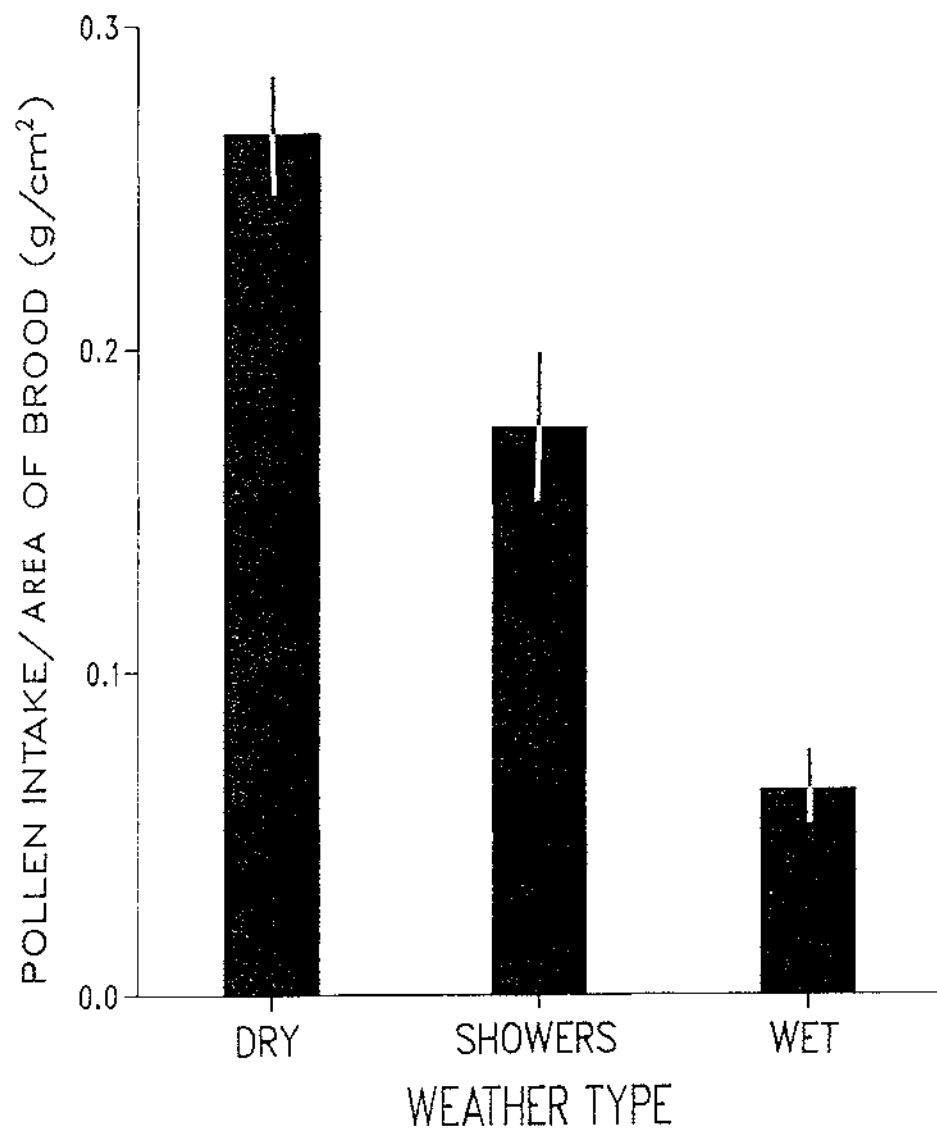
Bumble bees have gained a reputation for foraging under a wide range of weather conditions. In arctic regions they have been observed flying when air temperatures are below freezing (Longstaff, 1932; Richards, 1973). Wilson (1929) showed that bumble bees will continue to forage during dull and overcast weather, cold winds and gales, heavy rains and even during snow and hail showers. Though Free and Butler

Figure 11.

Amount of pollen (per unit brood area) collected by foragers under different weather conditions.

Vertical lines indicate 95% confidence limits.

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(1959) pointed out that most of Wilson's observations were of young queens (which, being larger and heavier than workers, can tolerate the above weather conditions to a higher degree). My own observations have shown there to be reduced pollen foraging during rainy weather. This is supported by observations made by Løken (1949), Miyomoto (1957, 1960) and Sladen (1912).

Apart from the physical difficulty of flying in rainy weather (though I have seen this on a number of occasions) a possible explanation for the reduced foraging in wet weather may be a lack of available forage. When the relative humidity of the atmosphere is high, some flowers fail to open (Salisbury and Ross, 1978) and, furthermore, pollen does not become available in others that do open (Free and Butler, 1959). Workers would have no reason to leave the nest if there was little or no pollen available to collect. On a number of occasions I observed returning foragers who had been carrying pollen loads and caught in the open during a heavy shower. Large amounts of pollen had been washed away and what remained was very watery making it difficult to unload. This may also act as a deterrent to foraging in wet weather.

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