adults of the 3rd generation reared at 25°C are best for mark-recapture experiments in summer, and those of the 3rd generation reared at 15°C are appropriate for autumn experiments (Table 3). However, when the longevity and flight ability are not markedly different between laboratory-reared and wild adults, reared adults could be used in mark-recapture experiments, without distorting the estimates of population density or dispersal.

For practical purposes, laboratory colonies reared from one to three generations, showing similar longevity and flight duration to wild adults can be used in mark-recapture experiments. Adults reared at about 25°–30°C are best for summer experiments, and those reared at about 15°–20°C are best for autumn experiments.

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Effect of Pollen-Substitute Feeding Site on Brood Rearing in Honeybee Colonies¹

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Honey and pollen are basic foods for honeybee colonies. Pollen is collected from a number of floral sources and is consumed to satisfy the dietary requirement for proteins, lipids, minerals and

vitamins. Since pollen is a limiting factor in colony development, beekeepers are concerned with providing an effective pollen substitute or supplement when natural pollen supplies are insufficient.

Pollen supplements contain honeybee-collected pollen (usually 5%–10%) mixed with other protein sources such as yeast, soybean flour, and/or other materials; pollen substitutes replace pollen totally with nutritionally-adequate materials (SHIMANUKI and HERBET, 1985).

When a moist patty of pollen substitute is placed inside a hive either on the top-bars or in the brood area, bees consume it, but never stored as bee bread. A part of ingested substitute is regurgitated and transferred to nestmates but majority is consumed by own and utilized for own nutrients and for royal jelly synthesis (ZAITOUN, 1990). The effect of the hive feeding site on diet consumption and production of royal jelly and queens

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Fig. 1. Application of diet in each feeding method. Accessible area to the diet (shadowed at the top row) in BAF-1 was equalized to space between frames in TBF (two 1×18 cm open area). The diets were set as bottom row: it was hung by wire between frames in the middle of combs.

has been studied previously (ZAYTOON et al., 1988).

The purpose of this study was to determine the effect of the pollen-substitute feeding site on hive activities based on the following criteria.

1. Dependency of pollen substitute consumption on feeding method (site).
2. Rate of sealed brood development with different feeding methods.

MATERIALS AND METHODS

Bees. Egg-laying sister queens obtained through routine rearing were used for the experiment. Three outdoor flight cages ($2 \times 2 \times 2$ m) were prepared, and a Langstroth hive with four empty frames, 1 kg (ca 10,000 individuals) of mixed aged workerbees and a sister egg-laying queen.

Diet and feeding methods. A commercial pollen substitute (Beehatcher; Nippon Formula Feed Manufacturing Co. Ltd.) was fed by three methods (Fig. 1): 1. Top-bar feeding (TBF)—the plastic dish ($10 \times 18 \times 1.5$ cm) of Beehatcher was inverted on the top bars of the frames as indicated by the usage instructions; 2. Brood-area feeding-1 (BAF-1)—the dish of Beehatcher with the same accessible area as the TBF was hung as illustrated in Fig. 1; 3. Brood-area feeding-2 (BAF-2)—the Beehatcher diet was spread on a mesh feeder (ZAYTOON et al., 1988) with a 10×18 cm surface area for both sides, and hung near the brood like in BAF-1. The total accessible area was ten times

that of TBF and BAF-1. The diet was renewed every week.

Measurement of diet consumptions and sealed brood. Weekly diet consumption was calculated as a sum of daily weight decrease of the diet. Degree of brood rearing was evaluated by direct counting of the number of sealed brood with aid of 5×5 cm wire grid. Measurements were made every week for ten weeks from July to September 1992.

RESULTS AND DISCUSSION

Table 1 shows the results. BAF-1 showed twice the consumption of Beehatcher diet compared with TBF. When the accessible area was increased 10 times, consumption of the diet further doubled. The site effect of ZAYTOON et al. (1988) was confirmed, but in their case they found a 1.5 times increase between BAF (BAF-2 in our study) and TBF. With TBF, the distance from the feeding site to the brood area is relatively large compared to BAF. Newly-emerged workers stay mostly in the brood area, so they can feed easily on the diet supplied by BAF. Young workers frequently visit diet in the brood area but diet on the top bars was visited by older worker bees that range far from the brood site (ZAYTOON et al., 1988).

In all the three hives, the uptake of sugar solution (HFCS) was highest 5,700–6,000 ml in the first week. In particular, for the first two days, the bees hoarded 1,600–1,700 ml each day. Such a large HFCS uptake was caused by the fact that the colony started with four empty frames. In the second week, the amount was 1,065–1,600 ml, and then decreased over the next three weeks by almost half in all three hives. After the fifth week, the HFCS decreased rapidly until the end of the experiment due to the weakening of the colonies.

The number of dead bees was fairly constant throughout the experiment in all the three hives indicating no specific abnormality. The colonies weakened after the fifth week because fewer new bees were emerging than were dying (sealed brood reflects the number of emerging bees after 1 week).

There were significant differences between the numbers of sealed brood produced with TBF, BAF-1 and BAF-2 (Table 1). There was least sealed brood with TBF, although the bee populations and other factors were initially the same. The only difference was the consumption of diet, which was always lowest in TBF compared with

Table 1. Consumption of diet and HFCS, brood rearing and dead honeybees per week for three feeding methods in caged honeybee colonies^a

Week	T B F				B A F - 1				B A F - 2			
	DC ^b (g)	HFCS (ml)	SB ^b (No.)	DB (No.)	DC ^b (g)	HFCS (ml)	SB ^b (No.)	DB (No.)	DC ^b (g)	HFCS (ml)	SB ^b (No.)	DB (No.)
1	80	6,100	0	750	150	5,700	0	680	265	5,900	0	710
2	40	1,065	500	882	100	1,480	700	934	210	600	800	856
3	15	750	800	790	20	500	850	790	80	700	1,070	740
4	25	650	82	625	45	600	120	640	110	700	260	630
5	15	790	163	650	30	810	241	600	60	910	383	700
6	10	210	72	700	20	200	100	650	40	250	160	750
7	5	200	21	590	10	100	33	580	15	200	100	760
8	1	50	15	510	2	60	40	500	3	70	60	550
9	0	20	0	525	0	25	0	510	0	30	36	560
10	0	0	0	560	0	0	0	590	0	0	0	520
Total	191	9,835	1,653	6,584	377	9,475	2,084	6,474	783	9,760	2,869	6,776

^a TBF, top-bar feeding; BAF, brood-area feeding; BAF-1 with same accessible area as TBF; BAF-2 with surface of 10 times wide; DC, diet consumption; HFCS, HFCS consumption; SB, number of sealed brood; DB, number of dead bees.

^b Every difference between diet consumptions, and between numbers of sealed brood in each treatment was significant by WILCOXON test ($p < 0.05$).

BAF-1 and BAF-2 (differences were significant among each treatment, Table 1). Comparing the diet consumption and sealed brood production, one finds that the higher consumption of diet produced greater numbers of sealed brood. If one would calculate the efficiency, however, by sealed brood production per diet consumption, the value for BAF-2 (3.66) was less than that in TBF (8.65). The BAF-1 value was intermediate (5.53). These values are abnormally big when considering the balance between nitrogen content of bees and the diet. Bees may offset poor nutrition by exhausting their body protein for their brood especially in TBF condition. The fact indicates a need for better diets and better feeding methods. Still though, the observed stimulation (increased worker bees) is important in colony building in the spring and during the period with insufficient food resources, even if the efficiency was low.

After the fifth week, the numbers of sealed brood gradually decreased until the end of the experiment. HERBERT and SHIMANUKI (1977) reported that bees in such a confined condition are not able to rear brood successfully when the diet contains 5% or 10% protein. They claim diets should contain

23% to 30% protein to allow successful brood rearing for a period of ten weeks. The Beehatcher diet has been developed for reinforcing colony in the field and contains 8.8% protein. Since confined condition such as our experiment is rather unnatural, we might have to increase the protein level for colony maintenance. Further improvement concerning diet protein content and quality is progressing.

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