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Influence of Nosematosis on the Hoarding Behavior of the Honeybee

Nosematosis, caused by *Nosema apis* in the honeybee, *Apis mellifera,* generally results in decreased honey yields (C. L. Farrar, *J. Econ. Entomol.* **49,** 333–338, 1947). Part of these decreased yields are related specifically to reduced nectar collection by parasitized bees that have been infected long enough to contain *Nosema* spores (O. Hammer and E. Karmo, *Schweiz. Bienen Ztg.* **4,** 190–194, 1947). The effects, if any, of the infectious process prior to sporulation on nectar gathering have not been established. The possibility was considered that an accurate test of hoarding behavior (J. M. Kulinčević and W. C. Rothenbuhler, *J. Apicult. Res.* **12,** 179–182, 1973) might be conducted on bees the first week after they were fed *Nosema apis* spores. Such a procedure would permit a measurement of hoarding behavior and also a measurement of response to *Nosema* infection on the same group of bees. The experiments reported here were designed to explore the effect of the early infectious process of *Nosema apis* in bees on the expression of their hoarding behavior.

Bees for the two experiments were obtained from different source colonies. Adult worker bees, 0–24 hr old, were emerged from brood combs in a 35°C incubator. The bees were collected, denied access to food for 1 hr, and administered an experimental treatment. This treatment consisted of individually feeding a 5-μl droplet of either a 33% sucrose solution to control bees or a 33% sucrose solution with *Nosema* spores to experimental bees by the method of T. E. Rinderer (*J. Econ. Entomol.* **69,** 489–491) but without using CO_2.

The *Nosema* spores were obtained by removing the midguts from laboratory-infected bees, grinding them in either 2 or 3 ml of water, and filtering the resulting suspension through a coarse screen. Spore concentrations were determined with the use of a hemacytometer, and the suspension was adjusted with 50% sucrose solution to obtain the appropriate concentration of sucrose. Experimental bees received 3.5×10^6 spores in the first experiment and 1.6×10^5 spores in the second experiment.

After the feeding treatment, groups of 50 bees were placed in cages similar to those described by J. M. Kulinčević and W. C. Rothenbuhler (loc. cit.). The first experiment consisted of three cages of bees fed *Nosema* spores and three cages of bees fed sucrose solution. The second experiment consisted of six replicate cages of bees for each of the two treatments. All cages were fitted with a section of comb to provide a place for storage of the sugar solution by the bees. Also each cage was provided with two gravity feeders. One feeder contained 20 ml of a 50% sucrose solution; the other contained deionized water.

All cages were held at 32°C and inspected daily. Dead bees were removed, and the volume of sucrose solution remaining was measured. These numbers were used to compute the average removal of sucrose solution from the gravity feeders (hoarding behavior) per bee day for the first 7 days of the experiment. Thereafter, the cages were maintained and observed for an additional 7 days. At the end of this period, the midguts of 10 or 5 bees per cage for experiment 1 or 2, respectively, were removed and observed for the presence of spores. Measurements of hoarding behavior for each experiment were analyzed by a *t* test.

Nosematosis led to a reduction in hoarding behavior (Table 1). The 35% reduction in hoarding behavior in experiment 1 was significant ($P < 0.05$), and the 22% reduction in experiment 2 was highly sig-

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TABLE 1

AVERAGE HOARDING BEHAVIOR PER BEE DAY FOR THE FIRST 7 DAYS OF EXPERIMENTS WITH GROUPS OF BEES FED OR NOT FED *NOSEMA* SPORES

Experiment	Number of spores fed/bee	Number of replicates[a]	Infection[b]		Hoarding[c] ($\bar{X} \pm$ SD)
			Average percentage ($\bar{X} \pm$ SD)	Average number of spores	
1	3.5×10^6	3	100 ± 0	—[d]	$0.064 \pm 0.016^{e,*}$
	0	3	40 ± 10	—[d]	$0.098 \pm 0.019^{e,*}$
2	1.6×10^5	6	100 ± 0	1.8×10^7	$0.059 \pm 0.006^{e,**}$
	0	6	0 ± 0	0	$0.076 \pm 0.005^{e,**}$

[a] Bees/replicate = 50.

[b] Based on the examination of 10 or 5 bees per replicate for experiment 1 or 2, respectively, on Day 14 of the experiment.

[c] Expressed as milliliters/bee/day for the first 7 days of the experiment.

[d] Not counted.

[e] Significantly different from the other mean in the same experiment: *, $P < 0.05$; **, $P < 0.01$.

nificant ($P < 0.01$). The effect of nosematosis on hoarding behavior began on the first day of both experiments. Differences continued through the experiment and tended to be largest through the second to fourth day when both control and experimental bees displayed the highest level of hoarding. In the first experiment, 40% of the control bees were naturally infected with *Nosema*. It seems reasonable that this infection influenced the hoarding behavior expression adversely in the control cages and led to a lower level of significance. We conclude that early intracellular nosematosis, prior to sporulation, substantially reduces hoarding behavior.

It is apparent from this experiment that nosematosis debilitates bees from a very early point in the infectious process. This early debilitation conceivably has far-reaching consequences on the overall production of a colony.

The difference in reduction of hoarding behavior between the two experiments, each established from a different source colony, suggests the possibility that bees may vary substantially in the behavioral effects of nosematosis. Such variation could preclude using the same group of bees to measure both the hoarding behavior and the response to *Nosema* of a source colony with the intent of ranking source colonies for breeding purposes. An experiment is in progress to explore this question further.

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