SPERMATOPHORE PERSISTENCE AND MATING DETERMINATION IN THE GYPSY MOTH (LYMANTRIIDAE)¹

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ABSTRACT. Spermatophores were detectable in all female gypsy moths dissected within 1.5 h following inception of copulation. After 1.5 h, the percentage of detectable spermatophores decreased with time; by 4.5 h, no spermatophore could be detected in any mated female moth. The percentages of detectable spermatophores did not differ significantly among three gypsy moth populations (laboratory-reared, high and moderate density natural populations) for intervals timed from inception of copulation. Examination of the bursa copulatrix for the presence of a spermatophore can be useful for rapid determination of female gypsy moth mating success.

The spermatophore of the gypsy moth, Lymantria dispar (L.), is formed within the female bursa copulatrix during the first 10 min of copulation (Klatt, 1920; Leonard, 1981). It consists of an oval sperm sac with a tapered neck that extends into the ductus bursae and a proteinaceous mass secreted by the male accessory glands. Proteolytic enzymes produced by the female begin to dissolve the spermatophore shortly after its formation (Chapman, 1971; Engelmann, 1970).

However, little is known of the fate of the gypsy moth spermatophore between formation and disintegration. Taylor (1967) reported that the spermatophore disintegrates within one or two hours of copulation but did not state whether this is time accrued from inception or termination of copulation. The distinction is essential since copulation averages 60–73 min (range = 20–198 min) (Forbush and Fernald, 1896; Doane, 1968; Waldvogel et al., 1981). Because the gypsy moth spermatophore is not persistent, determination of female mating success relies on examining eggs for embryonation several weeks after deposition or examining the female reproductive system for the presence of sperm (Stark et al., 1974). This paper presents, for the first time, data on the persistence of the gypsy moth spermatophore, with implications for rapid determination of female mating success.

MATERIALS AND METHODS

Laboratory-reared virgin gypsy moths were mated, uninterrupted, in arenas described by Waldvogel et al. (1981). The time *in copula* was recorded for each pair. To obtain data on the persistence of the

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Hours following inception of copulation	% spermatophores detectable			
	Laboratory-reared	High density	Moderate density	Total
1.5	100.0 (11)ª	100.0 (9)	100.0 (10)	100.0 (30)
2.0	81.8 (11)	66.6 (9)	70.0(10)	73.3 (30)
2.5	70.8(24)	50.0 (10)	40.0 (10)	59.1(44)
3.0	16.7(24)	44.4 (9)	30.0 (10)	25.6(43)
3.5	0.0(19)	11.1(9)	10.0 (10)	5.3 (38)
4.0	— (0)	0.0(9)	10.0 (10)	5.3 (19)
4.5	-(0)	— (0)	0.0 (9)	0.0 (9)

TABLE 1. Percentages of spermatophores detectable at intervals timed from inception of copulation for three gypsy moth populations: laboratory-reared, and high and moderate density natural populations.

* Values in parentheses are numbers of mated female moths dissected. Percentages did not differ significantly (Chi-square test, Fisher's exact test; P > 0.05) among populations at each time interval.

spermatophore, females were dissected under a microscope at $30 \times$ magnification, at intervals timed from inception of copulation. A medial incision through the abdominal terga provided access to the bursa copulatrix. The bursa copulatrix was then dissected *in situ* and its contents compared with those of an unmated female. All matings and dissections were performed at room temperature. These procedures were repeated with virgin moths that emerged from pupae collected from moderate density (ca. 3000 egg masses/ha) and high density (ca. 70,000 egg masses/ha) natural populations in Clearfield County, Pennsylvania. Egg mass densities were estimated by the method of Wilson and Fontaine (1978).

RESULTS AND DISCUSSION

Duration of copulation averaged 87 ± 2.3 min for all mated pairs (n = 213, range = 22-218 min). The percentages of spermatophores that remained detectable at intervals timed from inception of copulation are presented in Table 1. For each time interval, the percentages of detectable spermatophores did not differ significantly among populations (Chi-square test, Fisher's exact test; P > 0.05). Within 1.5 h following inception of copulation, 100% of the spermatophores in all populations could be detected. During this period, the shiny white spermatophore was visible through the wall of the bursa copulatrix. After 1.5 h, the percentage of detectable spermatophores decreased with time; the spermatophore was rarely visible through the bursa copulatrix wall, and dissection was necessary to determine its presence. At 3.5 h following inception of copulation, the spermatophore was detectable in less than 12% of the moths examined from any population. By 4.5 h, the contents of the bursa copulatrix of all mated females were indistinguishable from those of an unmated moth.

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These data eliminate the ambiguity arising from Taylor's (1967) report. His observations, if timed from termination of copulation, roughly agree with our findings. In other species of Lepidoptera, where the spermatophore may persist for several days or more, the bursa copulatrix can be examined for the presence of a spermatophore to determine whether a female has mated (Burns, 1968; Snow and Carlysle, 1967; Taylor, 1967). Although the gypsy moth spermatophore is not persistent, it can be useful for rapid determination of female mating success, which may be required in some precopulatory behavioral studies. Examination of the bursa copulatrix for a spermatophore is highly reliable within 1.5 h following inception of copulation. The presence of a spermatophore indicates female mating success and establishes that mating occurred less than 4.5 h prior to examination. Unfortunately, the absence of a spermatophore does not establish that the female gypsy moth is unmated. When no spermatophore is detectable, the most immediate recourse is examination of the spermatheca for the presence of sperm (Stark et al., 1974).

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