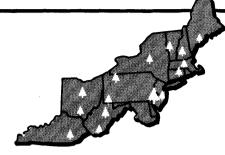
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### A TECHNIQUE FOR MARKING FIRST-STAGE LARVAE OF THE GYPSY MOTH FOR DISPERSAL STUDIES

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Abstract.—Zinc cadmium sulfide fluorescent particles can be used to mark first stage larvae of the gypsy moth, Porthetria dispar (L.), without effecting changes in their development and behavior. Marked larvae dispersed readily; so the technique could be used to correlate dispersed larvae with any particular source point.

Wind dispersal of newly-hatched larvae of the gypsy moth, *Porthetria dispar* (L.), is a major factor in the geographical spread of this pest. Although this means of distribution was noted in the early 1900s, only recently have studies been initiated to identify the morphological and meteorological characteristics associated with airborne distribution of the tiny first-stage larva (McManus 1973).

One of the problems associated with identifying the characteristics of the aerobiological pathways of dispersal was the lack of an adequate technique for correlating dispersed larvae with any particular point source. Development of such a technique requires an appropriate tagging or tracing element that will not adversely affect the relatively fragile insect and can be detected easily in the field.

Various physical, chemical, and radioactive techniques have been used to mark insects for use in release-recapture studies. Each requires a certain amount of handling of the study insect; most can be conveniently used with large sturdy species. Because fluorescent powders have been used successfully with a variety of insects (Turner and Gerhardt 1965, Vail and others 1966, Medley and Aherns 1968, Bennett and Smith 1968, Holbrooks and others 1970) and are readily available, they were tested for tagging gypsy moth larvae.

Zinc cadmium sulfide fluorescent particles (FP) are readily distinguishable from al-

most all naturally-occurring particulates by their color, size, and intensity (*Himel and Moore 1967*). The fluorescent properties of FP are physically stable under exposure to sunlight, and the particles are insoluble in water. Also, FP characteristically clings to hairs and setae, a definite advantage for this particular problem.

In the spring of 1973, studies were begun to determine: (1) the effect of FP on the development and behavior of the first-stage larvae; (2) if the marked insects would readily disperse; and (3) if identification of the marked larvae could be made at subsequent recapture intervals.

#### **Methods and Materials**

Samples of yellow and green fluorescent particle tracer material, each with a mean particle size of 3.5 microns, were obtained from Metronics Associates, Inc., Palo Alto CA. (Mention of brand-name materials should not be construed as endorsement by the U.S. Department of Agriculture or the Forest Service.)

Gypsy moth egg masses were collected near Easton CT in November 1972 and held at 2 to 3°C until April 1973. The eggs were then cleaned of hair and placed in Saran screen packets. Eggs and larvae were maintained in a Sherer-Gillett Mobile Greenhouse at 26°-27°C and 16:8 LD photoperiod. All laboratory bioassays were conducted under the same conditions. Larvae were reared on a diet described by ODell and Rollinson (1967).

To determine FP effect on egg hatch, three packets of eggs were brushed with FP and three packets were left untreated. Recently emerged first-stage larvae were individually marked by flicking a small amount of fluorescent powder on the dorsal surface of the abdomen. All larvae were checked with a short-wave ultra-violet lamp to ensure that they were satisfactorily marked. In some cases FP was easily identified without the lamp.

Twenty-five marked larvae were placed in each of ten  $15 \ge 100$ -mm plastic petri dishes,

and diet was provided. The dishes were sealed with masking tape and placed in the environmental chamber. Similarly, 226 unmarked larvae were set up in nine plastic petri dishes. Larvae were inspected every 48 hours for the next 8 days.

On 30 May 1973, as part of an investigation of the meteorological parameters affecting dispersal of the gypsy moth, field tests were held at the Saltonstall Experimental Area in Branford CT. A mass of about 180,000 newly emerged gypsy moth larvae were dusted with yellow FP and allowed to disperse from a box attached about 20 feet up in the lower crown of an American elm (vonLindern 1973). Butcher's paper streaked with Tac-Trap was laid on the ground, starting at the crown drop line and running downwind for 350 feet. The trap paper was inspected every 2 hours for larvae stuck in the Tac-Trap. Larvae crawling free on the paper, between the sticky streaked areas, were collected on masking tape. Counts and collections were made five times on each of 2 days. The larvae collected on masking tape were examined under the ultra-violet lamp for FP.

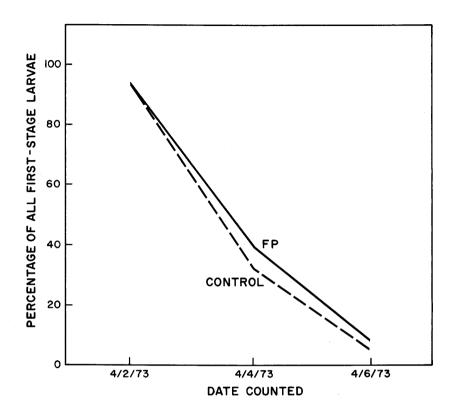
#### Results

Egg hatch, development and, behavior. — The first hatch of larvae was observed on the sixth day after treatment on both treated and control replicates. Daily emergence counts and total emergence were similar for all packets. Development (molting) and

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ticles	on	the	develop	ment	and	survival	of	gypsy	moth
larvae									

	Number of larvae in instar—							Mortality	
Date	I		II		III				
	$\mathbf{FP}$	С	FP	С	FP	С	FP	С	
3/29/73	250	226	0	0	0	0	0	0	
3/31/73	250	226	0	0	0	0	0	0	
4/2/73	238	215	3	5	0	0	9	6	
4/4/73	93	72	146	147	0	0	2	1	
4/6/73	17	10	165	160	31	25	26	24	

Figure I.—First-stage larvae counted in each 48-hour period, expressed as a percentage of the total number of larvae.



mortality were recorded and summarized (table 1 and fig. 1). There was no observable difference in molting time or behavior. Survival of FP-treated larvae was 85.2 percent compared to 86.3 percent survival of controls. FP was picked up easily on cast skins with the black light. This may be useful for monitoring dispersal after first-stage larvae have settled and molted.

Field test. — We were interested only in whether or not marked larvae could be picked up at all distances monitored in the test; that is, if dispersal was affected by the marking procedure, and if the marked larvae could be identified easily. Approximately 30 percent of all larvae collected were marked. The ratio of marked to unmarked larvae was about the same in each 100-foot section of the paper.

The ratio of marked to unmarked larvae was influenced by several factors, which may be important in future use of fluorescent particles: (1) the collected larvae were not examined under the microscope, and to see the smaller FP, magnification is required; (2) many larvae in the middle and at the bottom of the large dispersing mass were probably not marked; (3) the resident gypsy moth population was dispersing on both release days.

Table 2.—Number of			
tape during 48-hour	dispersal	period a	nd examined
for presence of FP			

a 11	. D	ay 1	Day 2		
Collection	FP	No FP	FP	No FP	
1	11	20	0	24	
2	18	48	0	17	
3	<b>24</b>	<b>45</b>	8	32	
4	33	64	28	48	
5	27	53	61	142	
Total	113	230	97	263	
Percent	33	67	27	73	

#### Conclusion

The results indicate that fluorescent particles can be used to mark first-stage gypsy moth larvae without effecting changes in development and dispersal behavior and thus could be used for identifying point sources of dispersing gypsy moth populations.

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