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3.5 SUPPRESSION OF SPRUCE BUDWORM POPULATIONS BY *TRICHOGRAMMA MINUTUM* RILEY, 1982–1986

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Abstract

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The ability of the egg parasitoid, *Trichogramma minutum* Riley, to suppress outbreak populations of the spruce budworm, *Choristoneura fumiferana* (Clemens), was studied annually near Hearst, Ont., from 1982 through 1986. Timing of broadcast parasitoid-releases was linked to spruce budworm moth emergence and oviposition. These phenological relationships were predicted from a regression based on larval development at least 2 weeks before expected emergence; this allowed sufficient time to regulate (program) parasitoid emergence during mass-rearing. Emergence of caged spruce budworm adults was used to monitor moth eclosion in the field. Pheromone traps provided daily information on the activity of male moths and helped to synchronize the parasitoid releases with spruce budworm oviposition. Information on parasitoid activity was obtained from sentinel (laboratory-reared) and naturally occurring spruce budworm egg masses. A curvilinear relationship between the rate of parasitoid release and parasitism of sentinel egg masses was developed. Two parasitoid releases, 1 week apart, early in the oviposition period of spruce budworm, significantly increased parasitism of host eggs by 14–83% and reduced larval populations correspondingly from 42 to 82%. Single releases were less effective and increased parasitism by 0.3–52% (single ground release, 1986). Two parasitoid releases, combined with a spring application of *Bacillus thuringiensis* Berliner to larval populations, was the most effective strategy and resulted in 83% egg parasitism and 93% larval reduction. Release rates greater than 12–16 × 10^6 ♀♀ *T. minutum* per hectare were not warranted based on impact and costs. The effects of release timing, weather, host density, and parasitoid quality on the future successful use of *T. minutum* are discussed.

Résumé

La capacité du parasitoïde de l'oeuf, *Trichogramma minutum* Riley, à supprimer les populations éruptives de la tordeuse des bourgeons de l'épinette, *Choristoneura fumiferana* (Clemens), a été étudiée annuellement, près de Hearst, Ont., à partir de l'année 1982 jusqu'à l'année 1986 comprise. Le réglage de relâchements de parasitoïdes à la volée a été lié à l'éclosion et la ponte de la tordeuse. Ces rapports phénologiques ont été prédits d'une régression basée sur le développement larvaire au moins 2 semaines avant l'éclosion attendue; ceci a donné du temps suffisant à régler (à mettre en marche) l'éclosion des adultes de la tordeuse dans les cages a été utilisée pour contrôler l'éclosion du Lépidoptère dans les champs. Les pièges de phéromones ont fourni des renseignements journaliers concernant l'activité des mâles adultes et ont aidé à synchroniser les relâchements de parasitoïdes avec la ponte de la tordeuse. Les renseignements concernant l'activité des parasitoïdes ont été obtenus des masses d'oeufs de référence (élevages du laboratoire) et de celles qui se présentaient en nature. Un rapport curviligne entre la vitesse de relâchement du parasitoïde et le parasitisme des masses d'oeufs de référence a été développé. Deux relâchements de parasitoïdes, séparés de 1 semaine, au début de la ponte de la tordeuse, ont augmenté significativement le parasitisme des oeufs de l'hôte de 14–83% et ont réduit les populations larvaires également de 42–82%. Les relâchements simples ont été moins efficaces et ont augmenté le parasitisme de 0,3–52% (relâchements terrestres simples en 1986). Deux relâchements de parasitoïdes, joints à un traitement printanier des populations larvaires avec *Bacillus thuringiensis* Berliner, se sont montrés le stratège le plus efficace et ont eu comme résultat un parasitisme d'oeufs de 83% et une réduction de larves de 93%. Les taux de relâchement plus extensifs que de 12–16 × 10^6 ♀♀ *T. minutum* par hectare n'ont pas été justifiés d'une base d'effet et de frais. Ont été discutés les effets du relâchement, du réglage, du temps, de la densité de l'hôte et de la qualité du parasitoïde sur l'utilisation réussie à l'avenir de *T. minutum*.

Introduction

The effect of egg parasitoids on populations of forest defoliating Lepidoptera is not completely understood (Anderson 1976), although many reports of their interactions have been published. In North American forests, the major families of egg parasitoids are Trichogrammatidae, Scelionidae, and Encyrtidae, and within these families *Trichogramma* Westwood is the most important genus associated with forest insects in northeastern North America (Houseweart *et al.* 1984*b*). Its seasonal distribution in North American forests has been described by Thorpe (1984).

Augmentative releases of native biological control agents against native pests has a long history in forest protection, although most experiments have been too fragmentary and short to be conclusive (Pschorn-Walcher 1977). Parasitoids such as *Trichogramma* generally are considered poor candidates for inundative releases because of their lack of selectivity or synchrony with specific host pests. In North America over the past 10 years, however, a concerted effort has been made to use a native species of egg parasitoid, *T. minutum* Riley, in inundative releases against a specific native pest, the spruce budworm, *Choristoneura fumiferana* (Clemens) (Houseweart *et al.* 1984*a*; Smith *et al.* 1987).

Initial studies showed little evidence that *T. minutum* could cause significant reductions in larval populations of spruce budworm. Houseweart *et al.* (1984*a*) concluded the following: (1) a native strain of *T. minutum* from a local area (Maine) performed better than a strain from California; (2) broadcast and multiple releases from the ground would be better than point releases; (3) closely timed aerial releases might significantly increase rates of parasitism; and (4) very high numbers of parasitoids ($>2\times10^6$ ♀♀ *T. minutum* per hectare) would be necessary to suppress natural populations of spruce budworm.

Recent research on ground releases of *T. minutum* has shown that two applications of ca. 12×10^6 ♀♀ per hectare, synchronized with the host's oviposition, could have a significant impact on overwintering larval populations (Smith *et al.* 1987). The extent to which such releases could reduce late larval feeding and protect foliage remains to be determined. As suggested by Van Hamburg and Hassell (1984), the relative success of reducing larval populations with an egg parasitoid will be dependent on the following: (1) the levels of egg parasitism achieved; (2) the subsequent level of early larval losses; and (3) the degree to which these two are density-dependent. Factors affecting the efficacy of *Trichogramma* following inundative release in agricultural crops have been dealt with by Lopez and Morrison (1985), Ridgway *et al.* (1981), Kot (1968, 1979), and Knipling (1977). Essentially no information is available on factors affecting inundative releases in the forest environment.

In 1982, the Ontario Ministry of Natural Resources developed an aerial application system for broadcast release of *T. minutum* on forested areas (Section 3.3). This aerial release system, combined with the large numbers of parasitoids made available through the rearing facility at the Biological Control Laboratory, University of Guelph, Guelph, Ont. (Section 2.0), allowed us to test the efficacy and feasibility of using aerial releases of *T. minutum* to suppress spruce budworm populations in northern Ontario. Specifically, the objectives were to determine the impact of inundative releases of *T. minutum* at varying rates on late larval populations of spruce budworm and to develop the operational use of such a strategy under forest conditions, in terms of aerial distribution, and host–parasitoid prediction and synchronization.

Materials and Methods

A description of the sample plots is provided in Section 3.1. As outlined in Section 3.3, plot sizes were 1.3 ha in 1982, 2.0 ha in 1983, 1.0 ha in 1984 and 1985, and 0.063 ha in 1986. Parasitoids were released aerially in all years except 1986 (see Section 3.3). Because studies in previous years showed that aerial release did not affect parasitoid quality (Section 3.4) and little parasitoid material was available in 1986, *T. minutum* was released from the ground. The design of this ground release simulated aerial release (Section 3.2).

Source of Parasitoid Material

Trichogramma minutum collected from Plummer Township (49°N, 86°W) in 1981 were reared in the production facility of the Biological Control Laboratory at Guelph, Ont., and used for all releases. New parasitoid material from Rogers Township (50°N, 87°W) was added to the stock colony held at Guelph in 1983 and in 1984, from recollected release material in Rogers Township. The parasitoid stock was maintained on factitious hosts during 1982: on the Mediterranean flour moth, *Ephestia kuehniella* (Zeller), at Guelph, Ont. (see Section 2.0), and on the Angoumois grain moth, *Sitotroga cerealella* (Olivier), at Biogenesis, Mathis, TX, USA. Rincon Vitova (Oak View, CA, USA) provided all parasitoids released during 1983 and one-third of those released in 1984. Parasitoids reared outside Canada were the original native strain (Plummer) collected from northern Ontario during 1981 and sent to facilities in the United States for mass-rearing. Parasitoids reared at the Biological Control Laboratory in Guelph, Ont., using the Angoumois grain moth as the host egg, comprised the remaining two-thirds of those released in 1984, as well as all parasitoids released in 1985 and 1986.

In 1982 and 1983, a sample (ca. 1000 parasitized eggs) was retained from each shipment prior to release to assess parasitoid quality, i.e. emergence, percentage of females, longevity, and fecundity. In 1984, 1985, and 1986, parasitoids were obtained after release by placing 26-cm funnels, with collection vials attached, in relatively open areas of the plots prior to release. Immediately following release, parasitized eggs were collected from these funnels and daily emergence, longevity, and fecundity of emergent female parasitoids determined. Because only female parasitoids are active in destroying host eggs, release rates were expressed as the number of female parasitoids per hectare per release. Longevity and fecundity were determined for 20 females from each rearing source. Each female was isolated in a 1.9-mL vial and provided with two fresh spruce budworm egg masses. Fecundity was determined from the number of host eggs parasitized and the number of progeny produced.

The parasitoids were shipped in bulk at about 10°C in Angoumois grain moth eggs and had been programmed during rearing so that >50% would emerge in 48 h (unless refrigerated) with the remainder emerging over 4–5 days. Parasitized material was held in cardboard containers in the dark at 5–10°C until required.

Timing of Parasitoid Releases

It was essential that the expected time of adult spruce budworm emergence and parasitoid release be accurately forecast because the Biological Control Laboratory could produce very large numbers of parasitoids for only a limited time. Thus, the releases had to coincide closely with the appearance of spruce budworm adults and oviposition; specifically, the first days and peak period of oviposition. For releases in early July, initial timing estimates were required by early March, to be followed by up-dating throughout the developmental period of the spruce budworm.

Due to the shortage of parasitized material, a single non-replicated release at peak budworm oviposition (maximum daily number of egg masses) was made in both 1982 and 1983. In the remaining years, based on studies by Smith *et al.* (1987), two releases were made early in the ovipositional curve (between the start and peak of oviposition) for each year. The number of parasitoids available determined the number of replicated plots that could be included.

Spruce Budworm Development

Development of spruce budworm was monitored by directly sampling egg masses, larvae, and pupae on branches (1982, 1985, and 1986) and catches of male moths in pheromone traps (1982–1986). Branch sampling (as described for each year of study) was conducted to monitor larval, pupal, and ovipositional stages and provide data that could be used to predict moth emergence. Sample trees, from which the branches were taken, were randomly selected within each plot. These trees were divided between balsam fir and white spruce according to the relative stocking density of each species. The surface area of each branch was measured (Sanders 1980) and counts made of all spruce budworm stages (larvae, pupae, and egg masses) as well as pupal emergence (%) and egg hatch (%) (McGugan 1954). Parasitized egg masses and pupae collected from these branches were held in a field laboratory to estimate parasitoid emergence and the onset and duration of adult budworm eclosion in the field. In 1982, 1985, and 1986, branch samples were collected every 3 days from mid-June until early August to assess pupal development and moth emergence.

In all years, larval development was used to predict moth emergence, 2–4 weeks before expected flight. One collection of branch samples (as described above) was specifically made in mid-June of each year, prior to pupation of spruce budworm. Ten years of data from Kapuskasing, Ont. (48.5°N, 82.5°W), suggested that the first adult moths would be collected in black-light traps at the earliest on 30 June and at the latest on 15 July (unpublished data). A process-oriented phenology and ovipositional model developed for spruce budworm by Régnière (1982, 1983) was used to refine this expected date of moth emergence taking into account site-specific information in each year. The model predicted first moth emergence from the percentage of fifth-instar or older larvae on a given sampling date by means of a regression equation. The equation followed the form $Y = a+bX$, where Y is the predicted date of first adult occurrence and X is the date on which the ith (0.05–0.9) proportion or greater of fifth-instar larvae were recorded. The regression coefficients, a and b, changed systematically as the proportion of fifth-instar larvae changed: "a" inversely with an increasing proportion and "b" directly. The regression coefficients themselves were regressed against the proportion of fifth-instar and older larvae and the resulting functions substituted back into the original function. This produced the equation used for updating the predicted date of first adult emergence:

$$Y = 72.474 + 0.693X + 0.23608XZ - 47.554Z \qquad [1]$$

where Y = the predicted date of first adult emergence; X = the larval sampling date (Julian date); and Z = the proportion of fifth-instar or older larvae on the sampling date (within the range 0.05–0.9).

Adult Moth Emergence

Pheromone traps baited with polyvinyl chloride cylinders (4 by 10 m) containing 0.03% of a 95% (E)-11-tetradecenal : 5% (Z)-11-tetradecenal mixture were used to monitor the emergence and activity of adult male spruce budworms (Sanders 1981). Both sticky traps (Pherocon 1CP®) and bucket-type Uni-traps (Sanders 1986) were used according to availability (see description for each year of study). The traps were spaced at least 40 m apart in each plot, 2–3 m above ground level. They remained in the field from mid-June until early August and were examined daily before 1000 hours EST for the number of male moths captured. When sticky traps were used, the bottom liners were changed daily to prevent saturation.

In 1985, the hourly activity of male moths in this area was monitored with an activity meter using the same type of pheromone lure at the centre of the trap. The meter contained a circular sticky card (75 cm diameter), divided into 24 equal parts, which was replaced daily. Each of the 24 sticky parts was exposed for only 1 h each day. The trap was run for 27 consecutive days (12 July to 5 August) during peak spruce budworm moth activity.

Assessment of Release Effect

The impact of *Trichogramma* releases on populations of spruce budworm was assessed using four criteria: in the year of release, (1) parasitism of sentinel egg masses and (2) parasitism of natural egg masses; and in the year following release, (3) density of spruce budworm larvae and (4) parasitism of natural egg masses. Data were compared between control and release plots using ANOVA with Duncan's new multiple range test (where applicable) and Ostle and Mensing's (1975) test for proportions with binomial distributions.

Sentinel egg masses: Sentinel egg masses were used to determine levels of natural parasitism on the non-release control plots as well as the extent and duration of parasitoid activity (temporal parasitism) on release plots. Sentinel eggs consisted of fresh spruce budworm egg masses laid on twigs of balsam fir by females reared in the laboratory on both artificial diet (young larvae) and natural foliage (older larvae) (Smith 1985). The egg clusters were shipped twice weekly from the laboratory in Sault Ste. Marie, Ont., to Hearst, Ont., in ice-cooled styrofoam boxes.

Sentinel egg masses were placed at varying heights in the crown of each sample tree by means of a pulley system (Smith 1985). The sample trees were balsam fir or white spruce, located in random clumps of two to four trees, at least 5 m away from the plot edge. Tree height, crown width, and condition were assessed yearly for each tree (see Section 3.1; Table 1). Because spruce budworm eggs are acceptable for parasitism by *T. minutum* only at a relatively young physiological age, before the head capsule of the embryo appears (Houseweart *et al.* 1982), the egg masses were changed every 3 days from mid-June until early August each year. This ensured a continuous supply of fresh susceptible egg masses for parasitism in the field. Following exposure of these egg masses to *Trichogramma* in the field, rates of parasitism were determined by holding them at the field laboratory in individual containers, usually size 00 gelatin capsules. Freshly laid egg masses of spruce budworm are bright green, but they turn gray as the head capsule of the developing larvae within the chorion becomes pigmented. Eggs parasitized and killed by *Trichogramma* turn shiny black within a few days at room temperature, thus making diagnosis of parasitism by *Trichogramma* relatively simple. For those egg masses parasitized, the number of eggs per egg mass that were either parasitized, missing, partially eaten, or not hatched were recorded as well as the date and sex ratio of emergents.

The number of egg masses parasitized out of the total number of viable egg masses placed in each plot constituted a measure of percentage egg mass parasitism for the 3-day period on that plot. Percentage egg parasitism was calculated by multiplying the proportion of egg masses parasitized by the proportion of viable eggs parasitized within each egg mass.

Natural egg masses: Parasitism of natural egg masses laid in the current year was determined by sampling annually at the end of the ovipositional period. Whole branches of balsam fir and white spruce were cut from the upper mid-crown of sample trees in each plot, one branch per tree (Dorais and Kettela 1982). Branch length and width were measured yearly, to calculate surface area (Sanders 1980), and in 1985 and 1986 each branch was weighed. Needles containing egg masses were removed from each branch and parasitism was calculated as for sentinel egg masses.

Carryover effect: The survival of *T. minutum* following inundative release was evaluated in those years following release. Throughout the period of oviposition by spruce

Table 1. Daily mean temperatures, sunshine, and wind speed, and total rainfall for the week following releases of *Trichogramma minutum* in Rogers Township from 1982 to 1986 inclusive

Year	Release date	Temperature (°C) Maximum	Temperature (°C) Minimum	Sunshine (h)	Wind speed (km/h)	Total rainfall (mm)
1982	14 July	23.0	10.0	5.7	9.5	19.6
1983	14 July	28.8	13.7	11.3	8.3	19.7
1984	10 July	24.6	12.1	7.3	4.9	25.5
	16 July	24.7	12.2	9.0	5.7	25.5
1985	9 July	20.9	9.1	6.7	7.4	32.8
	19 July	21.0	11.0	4.9	11.8	79.5
1986	5 July	24.1	10.2	10.5	3.4	0
	12 July	27.7	12.6	7.7	2.9	0

budworm, sentinel egg masses were placed on those plots that had received the highest release rate of *T. minutum* in the previous year. The egg masses were changed every 3 days, to ensure a constant supply of fresh, acceptable host eggs. Carryover effect was measured as the percentage of these egg masses that were parasitized.

Larval populations: To assess the impact of egg mass parasitism on larval populations of spruce budworm, 45-cm branch samples were collected in the spring of the year following each release. Samples were taken from balsam fir and white spruce in both the release plots and control areas. The branches were collected in mid-June when the majority of budworm were in the fourth to sixth larval instar. The number of larvae per branch was counted to compare larval populations on the release and control plots. The expected level of defoliation was then derived for each population according to the relationship outlined by Dorais and Kettela (1982). A measure of population reduction was calculated using a modified Abbott's formula to account for natural mortality in the control plots (Fleming and Retnakaran 1985).

ANNUAL RELEASES: METHODOLOGY AND RESULTS

1982 Releases

Trichogramma were released on only one 1.3-ha plot (plot 1) in 1982 because of the low number of parasitoids available. A control plot, similar to the release plot, was established ca. 3 km away (plot C1). Weather conditions during the sampling period in 1982 were relatively cool, overcast, windy, and wet (Table 1; see also Section 3.1).

To assess development of spruce budworm and estimate initial oviposition for timing the releases, whole branch samples of balsam fir and white spruce were taken from sample trees in 1982. One branch was taken from the lower, mid- and upper crown of each sample tree and the number and stage of each larva determined (McGugan 1954; Sanders 1980).

Larval development in 1982 was predicted by selecting three branches from each of 90 balsam fir and 30 white spruce on 9 June. This sampling indicated that the development of budworm larvae was further ahead on balsam fir (83% were fifth instar or older) than on white spruce (69% were fifth instar or older) (Table 2). Based on the regression equation, moth emergence was predicted for 25 June. Beginning on 7 July and on every 3rd day until 28 July, three branches from each of 180 balsam fir and 90 white spruce trees were taken to monitor spruce budworm phenology. Populations of pupae were highest on the first sample date, 7 July, and all adult moths had emerged by 28 July (Fig. 1). The daily activity of male moths was monitored with 30 sticky pheromone traps (Pherocon 1CP®), 15 in each of the release and control plots. The first male moth was captured on 4 July (Table 2), 9 days after the predicted date; relatively cool weather during June (daily mean = 12.7°C) delayed pupation and adult emergence. Moth flight peaked on 26 July, at about the same time as total adult emergence was reported (Fig. 2). Natural egg masses

Table 2. Phenological sampling of spruce budworm (SBW) on plots receiving inundative releases of *Trichogramma minutum* near Hearst, Ont., from 1982 to 1986 inclusive

Year	Sample date	Host tree species	No. SBW collected	SBW larval development (%) 5th instar	5th–6th instars	6th instar	Predicted date of emergence*	First empty pupal case	First ♂ moth in pheromone trap
1982	9 June	Balsam	921	73	—	10	25 June	10 July	4 July
		Spruce	912	42	—	27	25 June	10 July	
1983	16 June	Balsam	50	80	20	0	6 July	—	2 July
		Spruce	50	95	5	0	6 July	—	
1984	27 June	Balsam	396	7	82	11	12 July	—	9 July
		Spruce	408	7	76	17	12 July	—	
1985	29 June	Balsam	169	4	94	2	11 July	8 July	7 July
		Spruce	445	1	91	8	11 July	8 July	
1986	20 June	Balsam	154	4	95	1	3 July	29 June	30 June
		Spruce	136	5	93	2	3 July	2 July	

*Predicted from the equation $Y = 72.474 + 0.693X + 0.23608XZ - 47.554Z$ where Y = the predicted date of first moth emergence, X = the larval sampling date (Julian), and Z = the proportion of fifth-instar or older larvae on the sampling date (within the range 0.05–0.9).

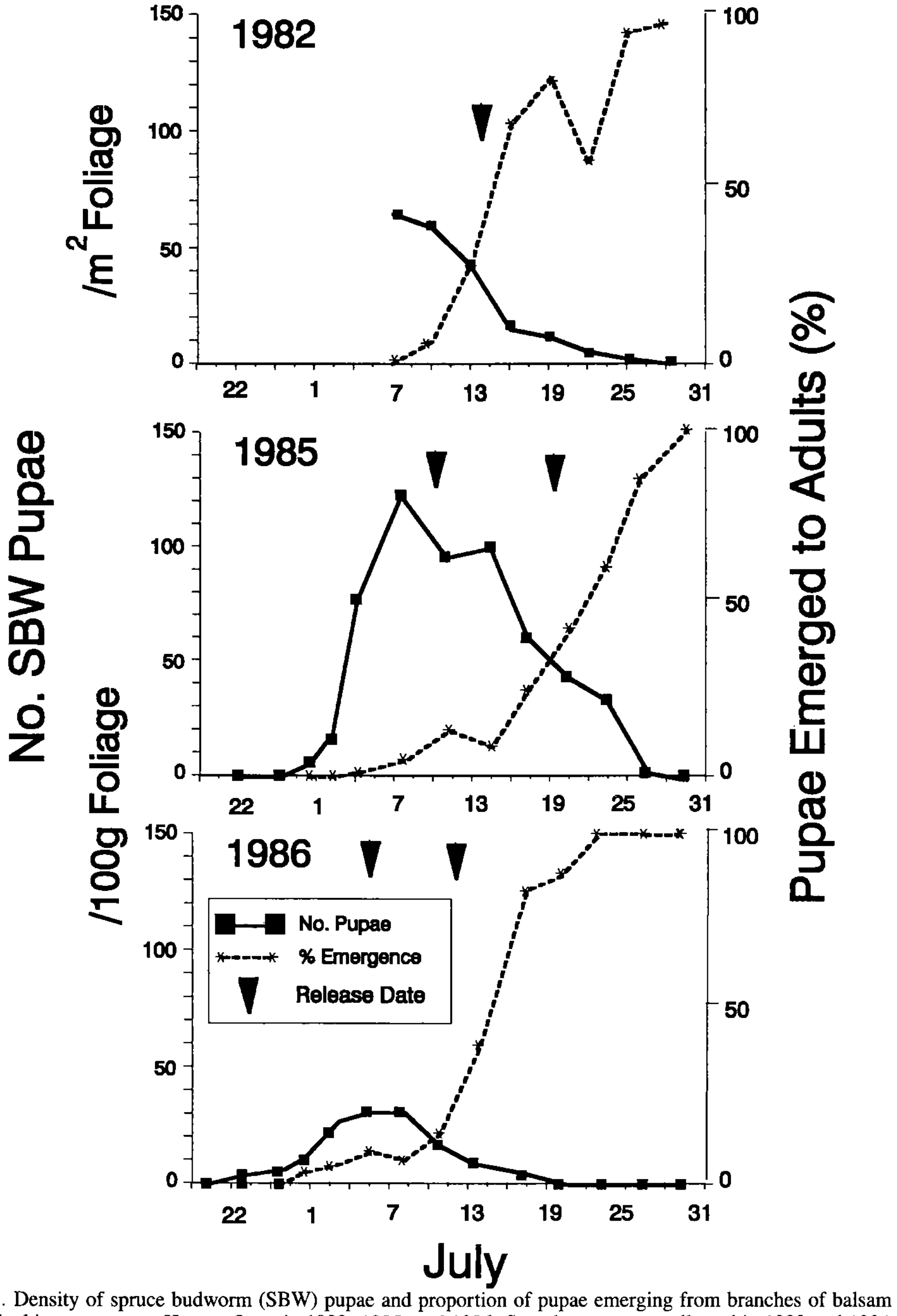

FIG. 1. Density of spruce budworm (SBW) pupae and proportion of pupae emerging from branches of balsam fir and white spruce near Hearst, Ont., in 1982, 1985, and 1986. Samples were not collected in 1983 and 1984.

were first laid on 16 July (Fig. 3) with peak oviposition occurring 9 days after egg-laying started on 25 July. Eggs began to hatch on 16 July and further releases of *Trichogramma* were not warranted.

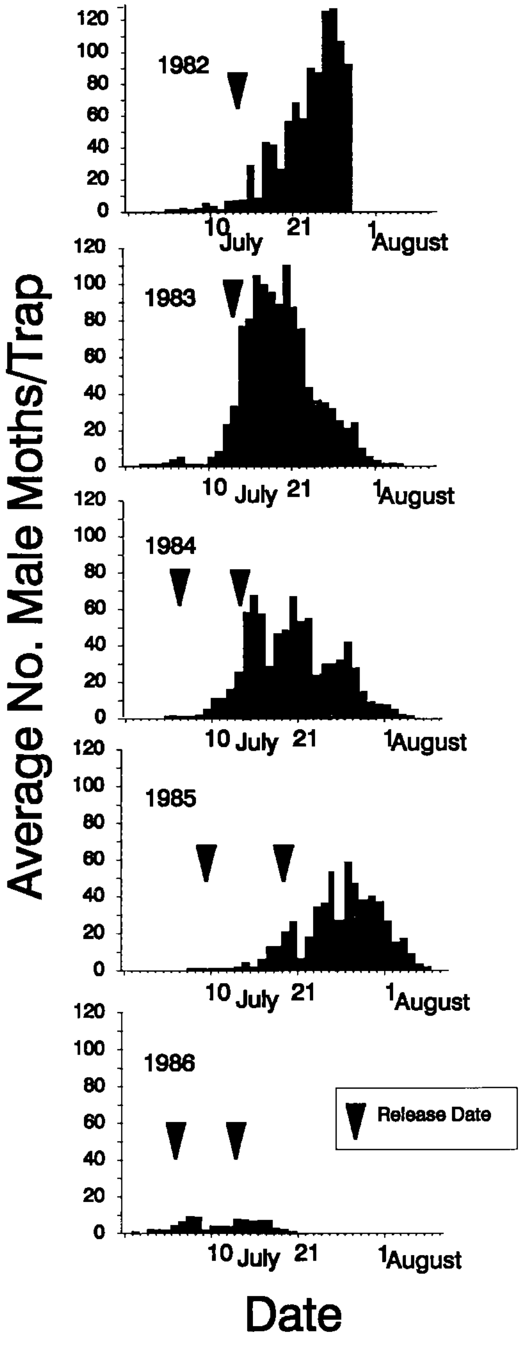

FIG. 2. Mean number of male spruce budworm (SBW) moths collected in pheromone traps near Hearst, Ont., from 1982 to 1986.

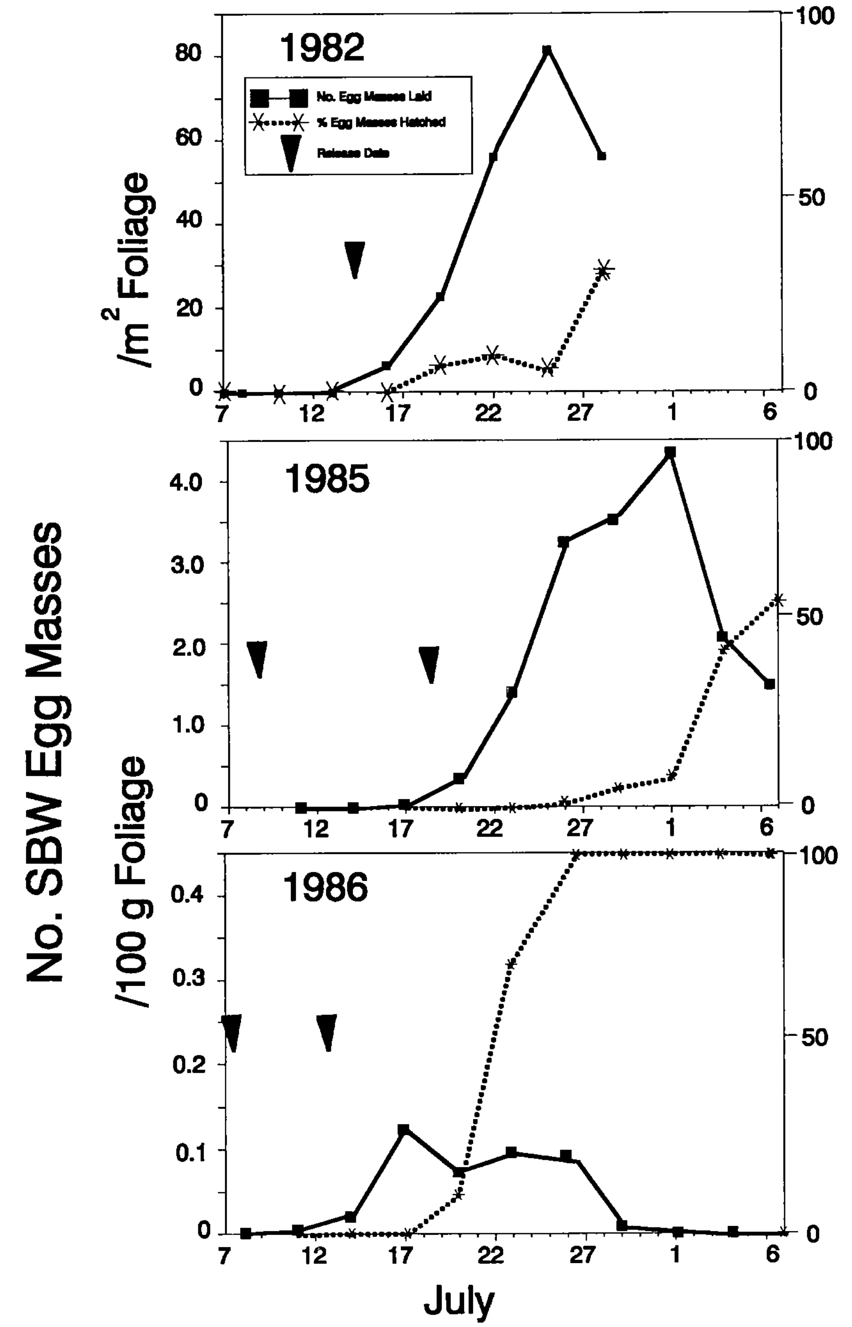

FIG. 3. Density of spruce budworm (SBW) egg masses and proportion of egg masses hatching on branches of balsam fir and white spruce near Hearst, Ont., in 1982, 1985, and 1986. Samples were not collected in 1983 and 1984.

Table 3. Emergence, longevity, and fecundity of *Trichogramma minutum* released near Hearst, Ont., from 1982 to 1986 inclusive

			Emergence					Fecundity	
Year	Source	Release date	%	Duration (days)	% in first 3 days	Sex ratio (% ♀♀)	Female longevity (days)	No. SBW* eggs parasitized per ♀	No. progeny per parasitized SBW egg
1982†	BCL‡ and Biogenesis	14 July	98	8	53	54	3.0	33.1‖	—
1983§	Rincon Vitova	14 July	80	—	—	51	1.6‖	4.0‖	—
1984§	BCL	10 July	74	4	94	58	2.5	5.9	1.2
		16 July	85	6	85	58	2.3	5.8	1.0
	Rincon Vitova	10 July	63	—	—	51	2.0	0.6	0.2
		16 July	71	—	—	67	1.6	4.1	0.7
1985§	BCL	9 July	91	10	45	62	1.3	4.5	2.2
		19 July	53	16	30	60	1.5	4.3	1.7
1986§	BCL	5 July	75	10	75	46	1.8	5.4	0.6
		12 July	87	8	79	52	1.6	5.3	0.6

*SBW = spruce budworm.
†Measurements based on a pre-release sample.
‡BCL = Biological Control Laboratory, University of Guelph, Guelph, Ont.
§Measurements based on a post-release sample.
‖Values based on 15 females from ground releases made on the same date using the same source of material sampled before release where emergence was 82% and the sex ratio was 53% females (Smith 1985).

Parasitoids were released on 14 July in three overlapping swaths across the release plot (1.3 ha) at a rate of 0.6×10^6 ♀♀ *T. minutum* per hectare. Deposit cards (see Section 3.4) were placed in groups of five at each of three stations along the length of the release plot; five cards were located ca. 20 m on each side of the release plot (20 cards total) to monitor horizontal drift of parasitized eggs. Within the release plot, on average, 0.28×10^6 parasitized eggs per card (625 cm^2) were deposited and no parasitized eggs were found on cards outside the plot. Pre-release sampling showed that the parasitoids had high percentage emergence, and normal sex ratio, longevity, and fecundity (Table 3). Parasitoids emerged over 8 days in the middle of the ovipositional period.

Sentinel egg masses were placed on 15 sample trees within the release area, two egg masses at the upper and mid-crown on each tree for each sampling date. Similarly, on each sample date, two egg masses on each of 75 sample trees were used to monitor parasitism in the control plot. Extremely low parasitism (1.0% of viable eggs) was observed on the release plot in 1982 (Table 4), probably due to the low application rate and poor weather conditions following release (Table 1). Maximum parasitism of sentinel egg masses in this plot was 2.3% (N = 43 egg masses) on 20 July compared with a maximum of 0.5% (N = 5413 egg masses) in the control plot. A second peak in parasitism (ca. 1.0%) of sentinel egg masses was observed on 6 August, suggesting the appearance of a second generation of *Trichogramma*.

Parasitism of egg masses laid naturally was assessed on 16 August by collecting 45-cm branch tips of balsam fir and spruce: 36 branches on the release plot and 43 branches on the control plot (divided among balsam fir, white spruce, and black spruce). All egg masses found on these branches were recorded. Of 684 egg masses collected from the release plot, only 5.1% were parasitized compared with 2.6% on the control plot (N = 648 egg masses). In terms of viable eggs, only 0.3% were parasitized on the control plot versus 1.0% on the release plot (Table 4). Because of the low level of parasitism, larval density and, thus, population reduction were not assessed the following spring.

1983 Releases

For the 1983 release, a 2.0-ha plot was established directly south of plot 1, because parasitism was low in plot 1 during 1982. The control plot used in 1982 was retained in 1983 (plot C1). During the releases in 1983, the weather was warmer and sunnier than in any other study year (Table 1).

As in 1982, larval development was assessed by taking a pre-release sample of ten 45-cm branch tips, equally divided between balsam fir and white spruce, on 16 June (Table 2). In contrast with 1982, only one 45-cm branch tip was collected from the upper mid-crown of each tree. From the proportion of fifth-instar or older larvae in the sample, the predicted date of emergence was 6 July. Pupal collections were not made in 1983. Nine sticky pheromone traps (as in 1982) placed in the field between 26 June and 4 August were used to assess moth emergence. The first male moth was captured on 2 July (Fig. 2). Moth densities peaked on 20 July with no activity observed after 3 August.

A single release of 8.2×10^6 ♀♀ *T. minutum* per hectare was applied just prior to peak moth activity on 14 July (Table 3). The date and rate of application were selected to coincide with ground releases made on other plots in the same area (Smith *et al.* 1987). Twenty deposit cards placed randomly in a 0.5-ha area at the centre of the plot indicated that the parasitoids were deposited relatively uniformly (X = 37.0 ± 11.6 parasitized eggs per card). No parasitized eggs were observed on cards placed outside the release plot. Based on the number of parasitized eggs released, an emergence of 80%, and a sex ratio of 1:1 (Table 3), ca. 8.8×10^6 ♀♀ per hectare were applied in 1983. The progeny of these parasitoids started to emerge in the field laboratory 12 days later, on 26 July, with emergence continuing for 25 days.

Table 4. Parasitism of natural spruce budworm (SBW) eggs and reductions in mature larval populations on plots receiving various rates of *Trichogramma minutum* near Hearst, Ont., from 1982 to 1985 inclusive

				Parasitoid release						
Year	Treatment	Plot	Parasite source	Date	Total (10^6 ♀♀ per hectare)*	No. SBW eggs per 45-cm branch†	Viable eggs parasitized (%)	No. SBW larvae per 45-cm branch	Population reduction (%)‡	Expected defoliation (%)§
1982	Control	C1	—	—	—	240.5a¶	0.3	—	—	—
	Release	1	BCL and Biogenesis	14 July	0.6 (1)	228.6a	1.0	—	—	—
1983‖	Control	C1	—	—	—	247.7a	0.1	22.4a¶	—	80
	Release	1	Rincon Vitova	14 July	8.8 (1)	168.2b	15.9	7.6b	50	45
1984	Control	C2	—	—	—	78.0a	1.1	34.9a	—	94
	Release	2	BCL	10 and 16 July	22.7 (2)	75.4a	79.5	6.1b	82	41
		3**	BCL	10 and 16 July	22.7 (2)	80.5a	83.4	2.4c	93	20
		4	Rincon Vitova	10 and 16 July	30.9 (2)	12.0b	14.1	—	—	—
1985	Control	C3and C4	—	—	—	100.7a	0.2	11.9a	—	60
	Release	5 and 6	BCL	9 and 19 July	4.2 (2)	94.9a	15.6	12.0a	0	60
		7 and 8	BCL	9 and 19 July	8.4 (2)	119.7a	22.2	8.2b	42	47
		9	BCL	9 and 19 July	16.9 (2)	87.6a	30.4	5.7b	45	40

*Total number of female *T. minutum* released; number of releases is indicated in parentheses.

†Based on unpublished data from the site, includes conversion for a mean of 14.6 eggs in each SBW egg mass (Smith 1985).

‡Reduction $= 1 - \left(\frac{\text{post-release density in treatment}}{\text{pre-release density in treatment}}\right) \times \left(\frac{\text{pre-release density in control}}{\text{post-release density in control}}\right) \times 100$ (from Fleming and Retnakaran 1985).

§Predicted level of defoliation based on 130 ± 23 buds per branch (field data) and the relationship between the number of fourth-instar spruce budworm larvae per bud per 45-cm branch described by Dorais and Kettela (1982).

‖Data provided by the Forest Insect and Disease Survey, Forestry Canada, Sault Ste. Marie, Ont.

¶Means followed by the same letter within each year and column are not significantly different at the $p \leq 0.05$ level (Duncan's new multiple range test).

***B.t.* used in this plot against third and fourth larval instars in the spring following release of parasitoids.

Sentinel egg masses were not used to assess parasitism in 1983, but instead, parasitism was examined in egg masses laid naturally. A sample of twenty-five 45-cm branch tips of balsam fir and white spruce was taken on 10–11 August in each of the release and control plots. All egg masses on a minimum of 10 mid-crown branches from each area were examined and those egg masses parasitized were categorized into four groups by the proportion of eggs parasitized: 0–25%, 26–50%, 51–75%, and 76–100%. On balsam fir, 28% of 204 egg masses were parasitized on the release plot compared with 0% of 347 egg masses on the control plot (significantly different; $\chi^2 = 16.17$; df = 1; $p = 0.05$). Similarly, on white spruce, 22% of 228 egg masses were parasitized on the release plot versus 0.6% of 336 egg masses on the control plot (significantly different; $\chi^2 = 9.04$; df = 1; $p = 0.05$). Of those egg masses parasitized, over 65% had greater than 50% of the eggs within each egg mass parasitized providing an estimated total level of viable egg parasitism of 15.9% (Table 4).

Spring larval populations in the release and control plots were assessed on 10 June 1984; >90% of the larvae were in the fifth or sixth instars. Twenty-five 45-cm branch tips of balsam fir and white spruce were taken from each plot. Significantly fewer larvae were found on the release plot (X = 7.6 larvae per branch) compared with the control plot (X = 22.4 larvae per branch) (Table 4). Based on the previous level of egg masses per 45-cm branch tip in these two plots (control = 247.7 and release = 168.2), this represented a 50% reduction in spruce budworm populations. Projected defoliation for the release plot was 45% versus 80% for the control plot. Suppression measures using *Trichogramma*, therefore, reduced populations of spruce budworm below the currently accepted economic threshold of 50% defoliation (Dorais and Kettela 1982).

1984 Releases

During 1984, three plots (plots 2, 3, and 4), each 1.0 ha in size, were established at least 3 km from the 1982–1983 study sites. A control plot (plot C2), ca. 0.25 ha, was located 2 km from the nearest release plot. Plots 2 and C2 had never been treated with insecticides for suppression of spruce budworm. Plot 3 was located in a 80-ha block sprayed at 30 BIU per hectare with *Bacillus thuringiensis* (*B.t.*) in the spring of 1984, and plot 4 was established in a 100-ha block which had been sprayed each spring with aminocarb (Matacil®) from 1981 to 1984. Intermediate weather conditions were recorded in 1984 (Table 1).

To assess larval development, a pre-release sample of 20 branches was taken, as in 1983. One 45-cm branch tip was taken from the upper mid-crown of 10 balsam fir and 10 white spruce in each release plot on 27 June. Due to the larvicide treatments in early June, development and population levels of spruce budworm varied among the plots. On plots 2, 3, and 4, there were 19, 12, and 2 per 1000 cm^2 foliage, respectively, with corresponding developmental indices (Dorais and Kettela 1982) of 5.9, 5.4, and 5.1, respectively. As in 1983, no pupal sample was taken. Using the proportion of each larval instar in the regression model, the predicted date of first moth emergence was 12 July (Table 2).

The first male moth was collected on 8 July in Uni-trap pheromone traps (Sanders 1986), three of which were placed in each plot (plots C2, 2, 3, and 4). The traps were in the field from 1 July to 5 August with maximum trap catches on 15 and 20 July (Fig. 2). Moth flight was completed by 4 August after three distinct peaks in activity.

Studies using ground releases of *T. minutum* against the spruce budworm indicated that two releases, 1 week apart, would significantly improve parasitism (Smith *et al.* 1987). In 1984, therefore, with more parasitized material available, two aerial releases were made at the beginning of the ovipositional period of the spruce budworm. The material was deposited relatively uniformly over each plot with essentially no drift beyond 25 m (see fig. 2 and table 4 in Section 3.4).

Release rates for *T. minutum* reared at Rincon Vitova were relatively high because the parasitoids were not cooled properly during shipment and emergence had begun by the time of release. The quality of these parasitoids was comparatively poor, with lower emergence, longevity, and fecundity than *T. minutum* produced at the Biological Control Laboratory, Guelph, Ont. (Table 3). On 10 and 16 July, 10.2 and 12.5×10^6 ♀♀ per hectare, respectively, reared at Guelph, were applied on each of plot 2 and plot 3, and 7.9 and 23.0×10^6 ♀♀ per hectare, respectively, reared at Rincon Vitova, were released on plot 4 (Table 4). In the field laboratory, the parasitoids did not emerge simultaneously, but over a 4- to 6-day period. Parasitoids reared at Guelph, Ont., parasitized 5.9 spruce budworm eggs per female and produced 1.2 progeny per parasitized budworm egg (Table 3). These progeny produced offspring with a sex ratio of 1.6 ♀♀:1.0 ♂♂, emerging over a period of 25 days beginning on 22 July. A continuous supply of *T. minutum*, therefore, was present in the field throughout the ovipositional period of spruce budworm.

To monitor temporal parasitism, three sentinel budworm egg masses per tree were placed in the upper, mid-, and lower canopy on 15 trees in each plot (divided between balsam fir and white spruce). Sentinel parasitism was monitored from 29 June to 1 August by replacing these egg masses every 3 days. Parasitism was very low in the control plot (1.1%), but parasitism was observed in all three release plots (Fig. 4). Over a period of ca. 15 days, the rate of egg mass parasitism increased sharply to a maximum of 50% on plot 4 (Fig. 4*a*) and 89% (SE = 4%) on plots 2 and 3 (Fig. 4*b*). Parasitism of sentinel egg masses by the next generation of *Trichogramma* appeared at the beginning of August (57% ± 21%), 20–23 days after the first release.

On plot 4, parasitism continued to decline after the first release, becoming indistinguishable from the control plot by 24 July. Overall parasitism was significantly lower on plot 4 than plots 2 and 3 (Fig. 4; Table 4). This low parasitism may have resulted from several factors: (1) plot 4 had relatively low larval populations of spruce budworm due to the aminocarb treatment in the spring: (2) development of spruce budworm on plot 4 was comparatively delayed and protracted due to insecticide treatment; and (3) the emergence of parasitoids reared at Rincon Vitova (released on plot 4 only) was lower because of improper shipping.

Parasitism in plots 2 and 3 (parasitoids reared at the Biological Control Laboratory, University of Guelph) was similar (i.e. small standard errors) except for the last sample date, 1 August (Fig. 4). This variability may be associated with the difference in development of spruce budworm between plots: larval development in plot 3 was behind that in plot 2 because of a *B.t.* application in the spring. This, in turn, delayed moth emergence (plot 2: <10 ♂ moths per trap on 10 July versus plot 3: <10 ♂ moths per trap on 15 July) and, thus, oviposition. Parasitism observed on the last sampling date was attributable to the progeny of *T. minutum* released on 10 July. This second generation parasitism was more apparent in plot 2 ($\bar{X}$ = 92.7% egg masses parasitized) than plot 3 ($\bar{X}$ = 21.4% egg masses parasitized), likely because of the more rapid development of spruce budworm and, thus, greater abundance of eggs at the time of the first release on plot 2 than on plot 3.

Parasitism of naturally laid egg masses of spruce budworm was assessed by collecting 36 whole branches per plot, 18 each of balsam fir and white spruce, on 5 August. Each branch was sequentially sampled until five egg masses were found (Dorais and Kettela 1982). The egg masses were classified as parasitized if at least one egg per egg mass turned black. In all three release plots, parasitism of viable eggs was significantly higher than in the control plot (Table 4). In general, parasitism of viable egg masses and eggs was higher on balsam fir (83–88%) than white spruce (74–84% eggs parasitized) although this was only significant on plot 3 (Table 5). Despite a parasitism rate of over 80% on plots 2 and 3 in 1984, there was no carryover of this parasitism to 1985; <1% of the 20 sentinel egg masses placed in plot C2 every 3 days during spruce budworm oviposition in 1985 were parasitized.

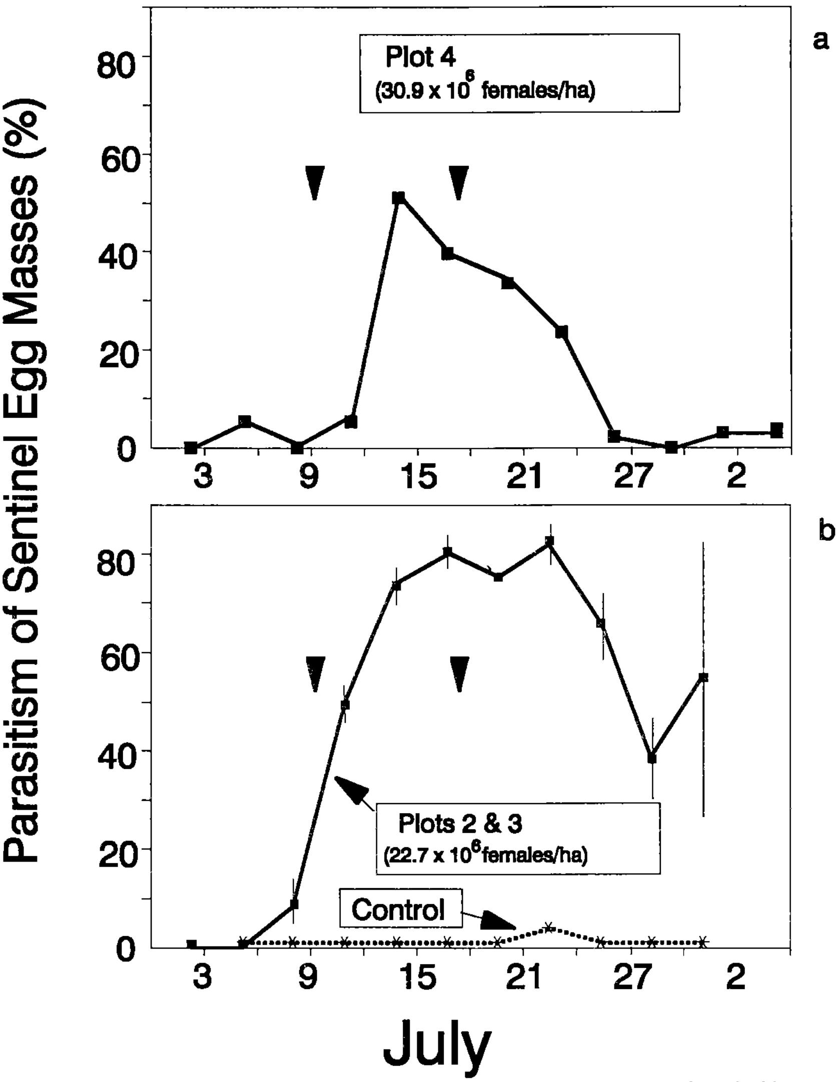

FIG. 4. Parasitism of sentinel spruce budworm (SBW) egg masses attached to balsam fir and white spruce trees on plots receiving *Trichogramma minutum* near Hearst, Ont., in 1984. Vertical lines at each sample point represent standard error for two plots.

Larval populations in the control plot and on those two plots receiving releases of *T. minutum* reared in Guelph, Ont., were measured in 1985. Thirty-six 45-cm branch tips, 18 each of balsam fir and white spruce, were taken from the upper mid-crown of trees in each plot on 18 June 1985. Plot 2 was not treated in the spring of 1985 and plot 3 was within a ca. 100-ha area which received an aerial application of *B.t.* prior to sampling, on ca. 10 June. Development of spruce budworm in plot 3, where both *T. minutum* and *B.t.* were applied, was delayed over that in plot 2 (*Trichogramma* release alone) or in the control plot (plot C2); developmental indices of spruce budworm for plots 2, 3, and C2 were 4.2, 3.7, and 4.2, respectively.

Both treatment strategies had significantly fewer larval spruce budworm than the control plot ($F=22.51$; df$=107$; $p=0.05$) although plot 3 (*B.t.* and *Trichogramma*) had a

Table 5. Parasitism of viable spruce budworm egg masses and eggs laid naturally on balsam fir and white spruce on plots receiving releases of *Trichogramma minutum* near Hearst, Ont., in 1984 and 1985

Year	Plot	No. × 10⁶ ♀♀ *T. minutum* released	Percentage viable* egg masses parasitized: Balsam fir (*n*)	Percentage viable* egg masses parasitized: White spruce (*n*)	Percentage viable* eggs parasitized: Balsam fir	Percentage viable* eggs parasitized: White spruce
1984	2	22.7	95 (84)	91 (72)	83	84
	3	22.7	93 (79)†	81 (56)	88†	74
	4	30.9	40 (70)	33 (55)	15	12
	C2	—	3 (70)	0 (66)	0.2	0
1985	5	4.2	51 (123)†	39 (151)	21†	13
	6	4.2	49 (101)	38 (95)	15	11
	7	8.4	34 (133)†	46 (213)	9†	18
	8	8.4	65 (104)	64 (138)	41	42
	9	16.9	73 (80)†	50 (136)	40†	24
	C3	—	4 (229)	3 (264)	0.1	0.2

*Viable represents those egg masses and eggs that remained after missing, partially eaten, or infertile eggs were subtracted.
†Values between host trees that are significantly different at the $P < 0.05$ level ($\chi^2 = 6.41$; test for proportions with binomial distribution, Ostle and Mensing [1975]).

significantly greater reduction in larval populations than did plot 2 (*Trichogramma* alone) (93% vs. 82%) ($t = 7.83$; df $= 71$; $p = 0.05$) (Table 4). A reduction of over 90% in populations of spruce budworm was observed on both plots 2 and 3, based on the density of budworm eggs per 45-cm branch tip and subsequent larval populations. Projected defoliation for the plots was 20 and 41% versus 94% on the control plot. Both suppression measures, therefore, reduced populations of spruce budworm below the currently accepted economic threshold of 50% defoliation, with the combined strategy being most effective.

1985 Releases

In 1985, five release plots (1.0 ha each; plots 5–9) and two control plots (1.0 ha each, plots C3 and C4) were established. A minimum of 200 m separated all plots to reduce the chance of drift during application and dispersal by parasitoids. Releases were made when wind speeds were less than 5 km/h in the plots. The weather following each release in 1985 was the coolest and wettest of all study years with generally overcast and windy conditions (Table 1).

As in 1982, spruce budworm phenology was followed from 19 June to 6 August by taking one 45-cm branch tip every 3 days, from the upper mid-crown of each sample tree: 16 balsam fir and eight white spruce. On 29 June, the majority of spruce budworms were at the fifth larval instar and the regression equation predicted first moth emergence on 11 July (Table 2). Pupae appeared on the branch samples of both host trees on 30 June (Fig. 1). Pupal development peaked on 8 July and was completed by 29 July. The first empty pupal case was found on 8 July. Pupae maintained in the field laboratory began emerging on 10 July. Emergence in the laboratory was similar to that observed in the field, suggesting that this method was reliable for assessing emergence. The cool temperatures in 1985 delayed development of spruce budworm, including egg-laying (Fig. 3). The first egg masses were not laid until 17 July with densities peaking between 29 July and 1 August. At the time of the last sample on 6 August, 50.6% of the egg masses had hatched.

Adult moth activity was monitored with 16 pheromone traps: eight sticky traps (as in 1982 and 1983) and eight Uni-trap buckets (as in 1984). One of each type of trap was placed within a sample plot, each at least 40 m apart. The first male moth was collected in a sticky trap on 7 July, 4 days before the predicted date (Table 2). Adult activity peaked between 25 and 27 July and was completed by 7 August (Fig. 2) with populations lower

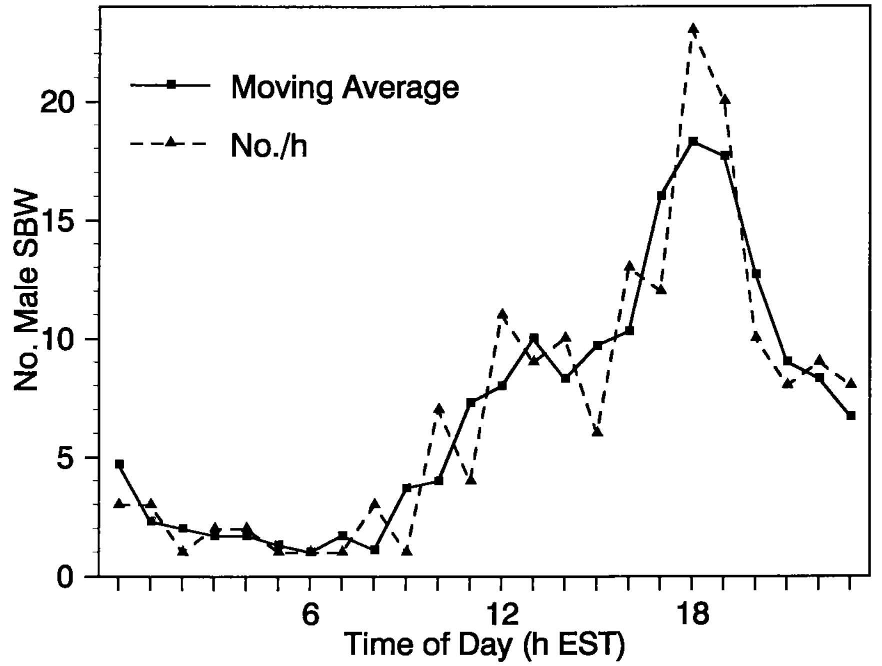

FIG. 5. Hourly catch of male spruce budworm moths in an activity meter with a pheromone lure in a forest stand near Hearst, Ont., during 1985. The moving average is a mean of three consecutive hourly catches.

than in previous years. The hourly activity of male moths was monitored between 14 July and 3 August using an activity meter with a pheromone lure identical to those in the daily traps (Fig. 5). Moth activity began after 0900 hours EST and peaked between 1700 and 1900 hours. Moths were relatively inactive between midnight and mid-morning suggesting that the best time to change pheromone traps for an accurate estimate of daily moth activity was before 0900 hours.

Parasitoids reared at the Biological Control Laboratory in Guelph were released from aircraft at three different rates in 1985 (Table 4). The two lowest rates, using double releases, were replicated twice. As in 1984, deposit of parasitized eggs on these plots was monitored (see fig. 2 and table 4 in Section 3.4). Deposit was uniform on each plot, averaging 334, 440, and 864 parasitized host eggs per card on plots 5 and 6, plots 7 and 8, and plot 9, respectively. The original design was to release 3, 6, and 12×10^6 ♀♀ per hectare per release on plots 5 and 6, plots 7 and 8, and plot 9, respectively; however, because of the cool wet weather, particularly following the release on 9 July, emergence of parasitoids in the field was reduced and the actual rates of release were 2.7, 5.4, and 10.9×10^6 ♀♀ per hectare on 9 July and 1.5, 3.0, and 6.0×10^6 ♀♀ per hectare, respectively, on 19 July.

The weather extended and delayed parasitoid emergence (Table 3). Emergence of *T. minutum* occurred over 10–16 days with only 30–45% of the sample emerging within the first 3 days of each release. This was considerably fewer than in other years; however, those parasitoids that did emerge had normal sex ratio, longevity, and fecundity. The progeny from the releases began emerging on 23 July, peaked 28 and 31 July, and continued until 20 August. These females produced, on average, 1.9 progeny per parasitized host egg with a sex ratio of 2 ♀♀:1 ♂. A continual, relatively low supply of *T. minutum* was thus provided in the field during oviposition of spruce budworm.

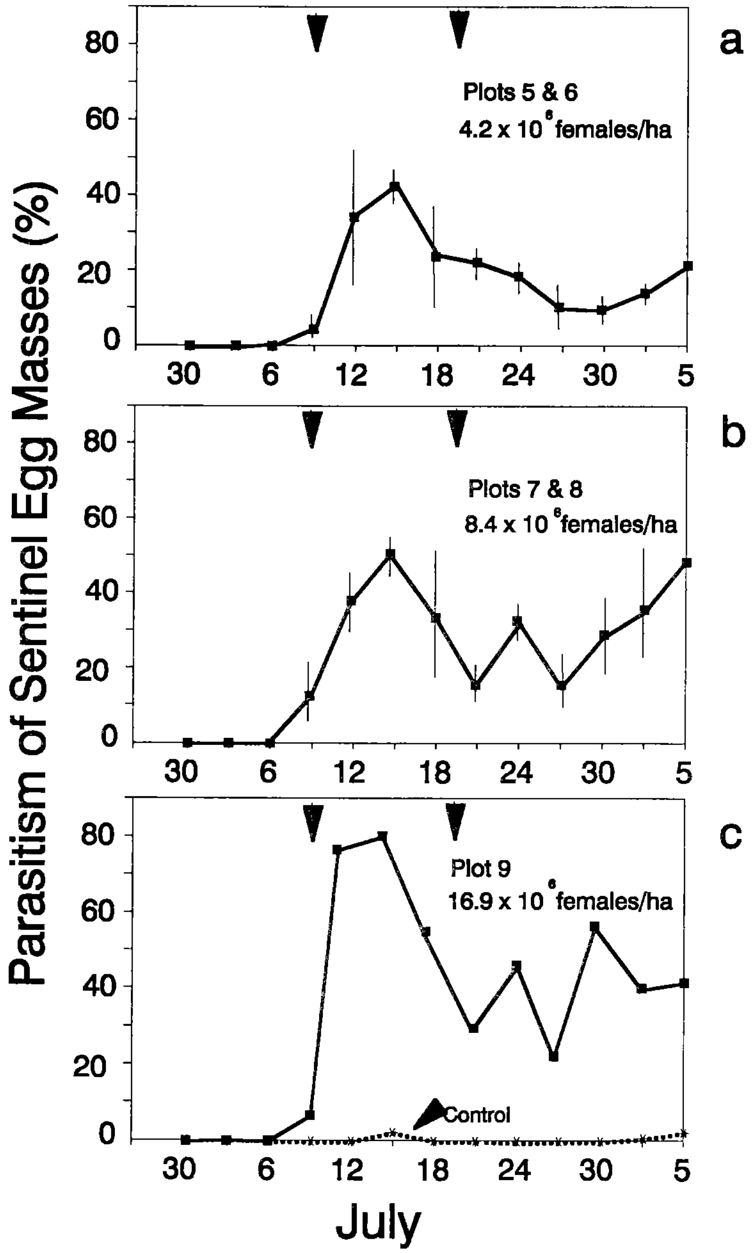

FIG. 6. Parasitism of sentinel egg masses attached to balsam fir and white spruce trees on plots receiving releases of *Trichogramma minutum* near Hearst, Ont., in 1985. Vertical lines at each sample point represent standard errors for two plots.

Fifteen trees were selected on each plot to monitor temporal parasitism. Three sentinel egg masses were attached at upper, mid-, and lower crown level to each tree between 1 July and 4 August. Immediately following the first release, parasitoid activity was evident on all the release plots (Fig. 6). The level of parasitism increased with the release rate, and parasitoid activity persisted throughout the sampling period. Over a period of 8 days, the rate of parasitism increased sharply to a maximum of 42, 58, and 80% on plots receiving 4.2, 8.4, and 16.9×10^6 ♀♀ per hectare, respectively. Parasitism by the next generation of *Trichogramma* appeared around 24 July on the plots receiving the highest two rates and continued at 20–40% throughout the remainder of the season.

Parasitism was lower following the second release than the first, due to the extension of parasitoid emergence and the low numbers emerging (Table 3). Trends in parasitism were similar on replicated plots. On the two control plots, very low parasitoid activity (<4%) was observed. The carryover effect to the following year was assessed by placing 10 sentinel egg masses on five sample trees every 3 days (two egg masses per tree) in plot 9 between 25 June and 4 August 1986. Essentially no carryover was observed on this plot (receiving the highest release rate in 1985); <3.1% of the sentinel egg masses were parasitized.

Parasitism of egg masses laid naturally was assessed in 1985 by taking thirty-six 45-cm branch tips from each release plot and one control plot on 8 August. At the time of sampling, at least 50% of the egg masses had hatched (Fig. 3) and the remainder, based on mean daily temperatures in the field, would have been unacceptable to parasitism by *T. minutum* (Houseweart *et al.* 1982). The average density was seven egg masses per branch. Parasitism of viable spruce budworm eggs in every release plot was significantly higher than in the control plots ($F = 14.67$; df = 1511; $p = 0.05$) (Table 4). In two of five release plots, parasitism of natural viable eggs was higher on balsam fir (21 and 40%) than on white spruce (13 and 24%) regardless of the release rate (Table 5). On plot 7, however, significantly more parasitism was evident on white spruce than on balsam fir.

On 11 June 1986, larval populations were assessed by taking thirty 45-cm branch tips, 15 each of balsam fir and white spruce, in each release plot and one control plot. Significant reductions in larval populations were found in plots receiving the two higher release rates, 8.4 and 16.9×10^6 ♀♀ per hectare, and no difference was apparent at the lowest rate (Table 4). Based on egg densities per 45-cm branch tip, the releases resulted in 87.4–93.5% reductions in populations. This was equivalent to 0, 42, and 45% control of budworm populations following releases of 4.2, 8.4, and 16.9×10^6 ♀♀ per hectare, respectively. Projected defoliation for the three release plots was 40, 47, and 60% (at decreasing release rates) versus 60% for the control plot. Although the natural populations of spruce budworm were generally low (plots C3 and C4), the two highest rates of *Trichogramma* release reduced the population of spruce budworm slightly below the economic threshold of 50% defoliation.

1986 Releases

The annual studies to 1986 showed that parasitism by *T. minutum* was not carried over beyond the year of release. Similar conclusions were reached by Smith *et al.* (1987) for ground releases of *T. minutum*. Due to this lack of carryover, the limited supply of parasitoid material, and the fact that the aerial releases did not reduce parasitoid quality (Section 3.4), three plots from 1985 (plots 5, 6, and 7) were used for ground release of *T. minutum* in 1986. Four smaller plots (25 by 25 m) were established within each plot (each separated by 50 m). Seven of the 12 plots were used to examine the effect of releasing various densities of *T. minutum* from the ground and the remaining five were used as non-release control plots. Weather conditions were average with no rainfall reported following the releases and relatively low wind speeds (Table 1).

From 16 June to 3 August, the phenology of spruce budworm was monitored by taking 45-cm branch tips from the upper mid-crown (of 22 balsam fir and 29 white spruce) within the 12 plots. Development of spruce budworm was accelerated by warm temperatures in the spring of 1986. On 20 June, over 95% of the larvae were fifth instar or older and the regression equation predicted moth emergence on 3 July (Table 2). Pupae appeared on 23 June with the first empty pupal case present 6–9 days later. Pupal populations peaked on 5 and 8 July and by 23 July, all pupae had emerged (Fig. 1). Similar emergence was observed in the field laboratory although it was extended more toward the end of July.

Twelve sticky pheromone traps (Pherocon 1CP®), one in each plot, were used to monitor adult activity from 28 June to 3 August. The first male moth was collected on 30 June, 3 days before the predicted date (Table 2). Throughout 1986, very low numbers of males were trapped with the mean never exceeding seven moths per trap (Fig. 2). Two distinct peaks were observed, one on 8 July and one on 14 July. Because of these low populations, the number of egg masses present in 1986 was extremely low with the first egg mass found on 11 July (Fig. 3; note change in vertical scale). All fresh egg masses had been laid by 23 July with the majority hatched by 30 July.

Parasitoids were released by ground application (see Section 3.2) on each plot at rates of 2.1, 4.3, 8.6, and 12.9×10^6 ♀♀ per hectare on 5 July and 2.7, 5.3, 10.6, and 15.9×10^6 ♀♀ per hectare on 12 July, for a total of 4.8, 9.6, 19.2, and 28.8×10^6 ♀♀ per hectare. The lowest two release rates were replicated twice and the highest two were applied only on single plots. One plot received a single rate of 12.9×10^6 ♀♀ per hectare on 5 July. Parasitoids emerged over a period of 8–10 days with 75% having emerged 3 days after release (Table 3). The parasitoids had average longevity and fecundity but produced a low number of progeny per parasitized host egg. The progeny of these parasitoids began emerging in the field laboratory on 18 July. Emergence of this second generation peaked on 23 and 25 July, thereby providing a continual supply of *T. minutum* in the field during oviposition.

Two sentinel egg masses were attached in the upper and mid-crown of 96 sample trees, eight trees in each plot (seven release plots and five control plots) between 22 June and 4 August. Parasitism of these egg masses on the control plots was always less than 2.1%. In the release areas, parasitism increased slowly after the first release (over 11 days) possibly due to the low number of egg masses on these plots (Fig. 7). Maximum parasitism was observed after the second release, even on the plot receiving the single release at the highest rate (12.9×10^6 ♀♀ per hectare) (Fig. 7*d*). This delay was not evident in parasitoid emergence (Table 3); therefore, it probably reflected an actual delay in parasitoid oviposition. On these plots, maximum egg mass parasitism of 52.8, 66.7, 53.3, 84.2, and 85.7% was achieved with total releases of 4.8, 9.6, 12.9, 19.2, and 28.8×10^6 ♀♀ per hectare, respectively. Parasitism by the second generation of *Trichogramma* peaked on 24–25 July, 10–14 days after the initial peak in sentinel parasitism.

The curvilinear relationship between maximum parasitism of sentinel egg masses and the rate of application for releases made in 1985 and 1986 is shown in Figure 8. This relationship was similar for both years and suggests that increasing rates of application above $12–16\times10^6$ ♀♀ per hectare will not significantly improve parasitism of sentinel egg masses. It also shows that the maximum parasitism of acceptable spruce budworm egg masses that can be expected with *Trichogramma* releases alone is 80–85%.

Parasitism of natural egg masses was assessed by collecting one whole branch from the upper mid-crown of 100 sample trees on each of the seven release plots and one control plot. Collections were made on 6 August when all egg masses had hatched (Fig. 3). Only 14 egg masses, laid in 1986, could be found. This provided an inadequate sample size for further analysis of parasitism: less than two egg masses per plot. The low populations of spruce budworm at the end of the field season in 1986 precluded the collection of larval samples on these plots the following spring to determine the level of population reduction.

Discussion and Conclusions

We consider that timing of inundative releases with *Trichogramma* relative to oviposition by spruce budworm is one of the most important components affecting efficacy of the parasitoids in our study. A similar conclusion was reached by King *et al.* (1985) using releases of *T. pretiosum* against *Heliothis* spp. In our study, both the first release in

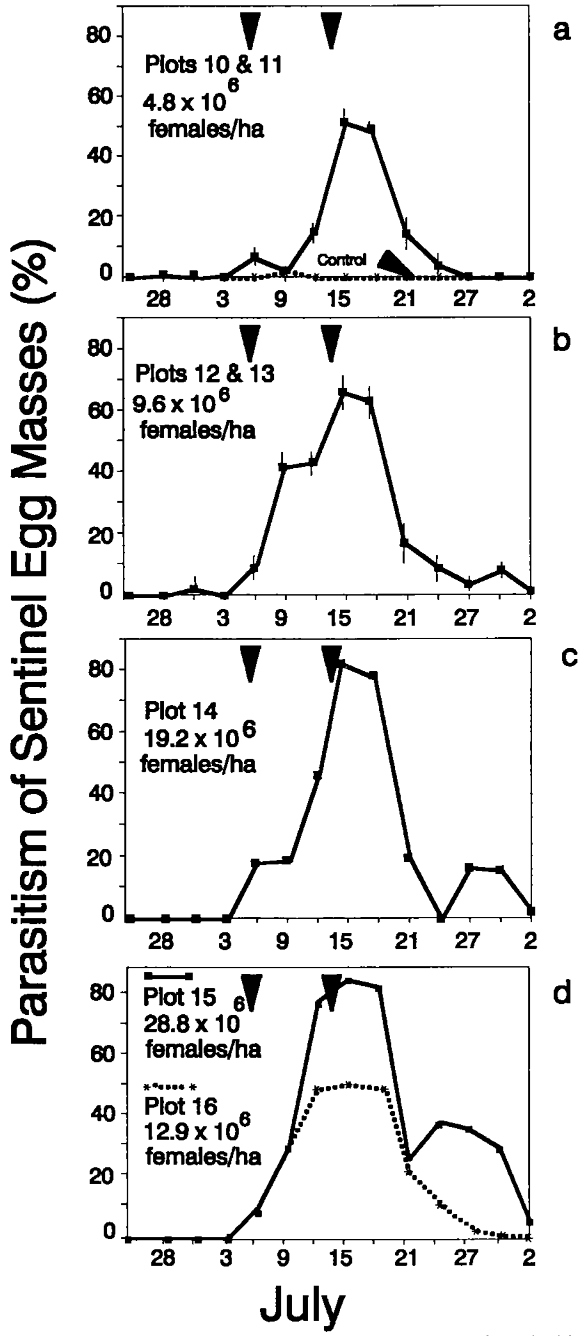

FIG. 7. Parasitism of sentinel spruce budworm egg masses attached to balsam fir and white spruce trees on plots receiving different rates of *Trichogramma minutum* near Hearst, Ont., in 1986. Vertical lines at each sample point represent standard errors for two plots.

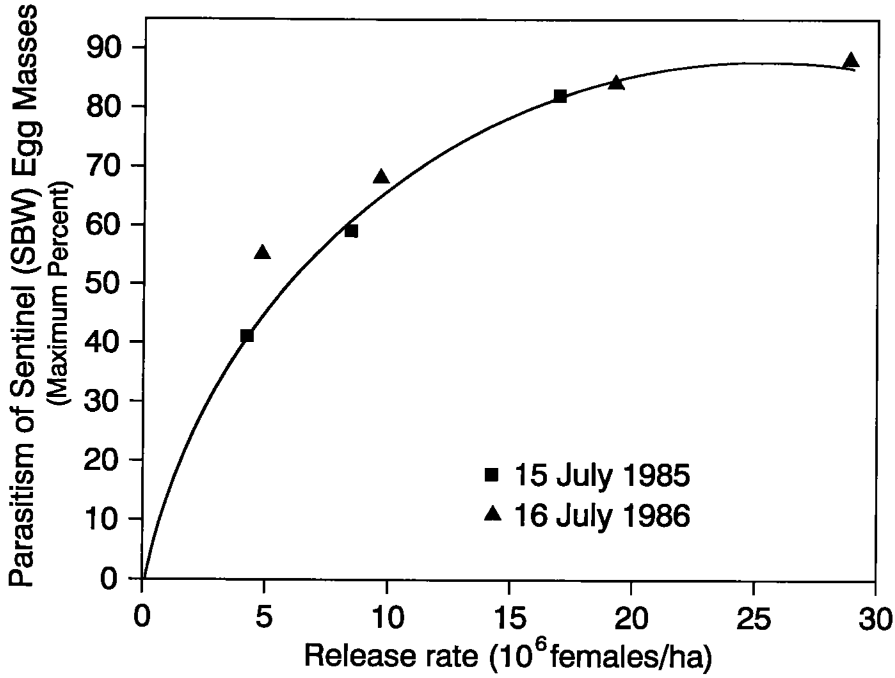

FIG. 8. The effect of release rate of *Trichogramma minutum* on the maximum parasitism level achieved on sentinel egg masses of spruce budworm (SBW) attached to balsam fir and white spruce trees near Hearst, Ont., in 1985 and 1986.

1985 and those releases made using parasitoids produced at Rincon Vitova in 1984 occurred before oviposition by spruce budworm. Although *T. minutum* were active in the field, they had little effect on parasitism of eggs laid naturally. For a univoltine species such as spruce budworm, eggs are present in the field at only one time each year. These eggs are laid over a period of about 3 weeks and are only acceptable to parasitism by *T. minutum* for 3–6 days following oviposition (Houseweart *et al.* 1982). From the present study and work by Smith *et al.* (1987), we suggest two parasitoid releases between the beginning and peak of egg-laying will provide optimal results. Obviously, reliable information on the temporal pattern of budworm oviposition is required for such a strategy.

The regression model, incorporating the proportion of spruce budworm at fifth larval instar or older to estimate moth emergence, generally provided a reliable method for predicting the onset of egg-laying. Larval samples taken at least 2 weeks before the expected onset of oviposition predicted moth activity in the pheromone traps within 3–4 days (except in 1982 where exceptionally cool weather during June delayed oviposition by 9 days). In all cases, this predictive regression allowed the rearing facility sufficient time to produce large numbers of parasitoids programmed for a specific date of emergence. As suggested by Lawrence *et al.* (1985) and Witz *et al.* (1985), pheromone traps examined before 0900 hours were good indicators of moth activity and, thus, oviposition. Although some variability will always be present in estimating moth emergence with the regression model because of the uncertainty of weather conditions, further studies quantifying the relationship between larval development and moth emergence could improve the model's accuracy.

Two releases of *T. minutum* during the oviposition period of spruce budworm provided better control than a single release. Unless the timing of a single release is exact and emergence of these parasitoids is extended over at least 10 days, insufficient *T. minutum* will be continuously present to ensure high parasitism of freshly laid host eggs. In 1983, the single application of 14.1×10^6 ♀♀ per hectare was well synchronized with oviposition and yet parasitism of naturally laid eggs was only 15.9%. In 1984, however, following two releases at ca. 11.3×10^6 ♀♀ per hectare per release, mean parasitism of naturally laid eggs was 81.5%. Because female fecundity for *Trichogramma* is greatest during the first days of emergence (Houseweart *et al.* 1982; Smith and Hubbes 1985), two releases, 1 week apart, will be most effective if the parasitoids are programmed to emerge over a 4- to 7-day period. This will provide highly fecund females each day of oviposition by spruce budworm. As suggested by Lawrence *et al.* (1985) and confirmed by Smith *et al.* (1987), using three releases, 1 week apart, would be redundant and costly because the progeny of parasitoids from the first release would be active in the field at the same time as parasitoids from a third release. In each year of our study, the second generation of parasitoids began emerging before *T. minutum* from the second release had stopped parasitizing host eggs. When emergence from the release was not extended beyond 7 days, two releases maintained high levels of parasitism (>80%) throughout the ovipositional period of the spruce budworm.

Abundance of host eggs in the release area is thought to influence the level of parasitism following inundative release (Ridgway *et al.* 1981). Alternate hosts used by polyphagous *T. minutum* may be present on forest stands prior to spruce budworm and used as reservoirs (Houseweart *et al.* 1984*a*). In the young spruce–fir stands of our study, this effect was never observed; there was no detectable parasitism by *T. minutum* on the control plots after oviposition by spruce budworm. Any sustained parasitism in the ovipositional period of spruce budworm, therefore, was due solely to development of parasitoids on populations of spruce budworm. As observed by Houseweart *et al.* (1984*a*), we also found no evidence that densities of host eggs affected parasitism by *T. minutum*. The supply of naturally laid spruce budworm eggs declined significantly during 1984 and 1986 (see Section 3.1; Table 2) for Rogers Township, yet the level of maximum parasitism observed on sentinel egg masses following the release of about 12×10^6 ♀♀ per hectare each year remained similar. An independent functional response for *T. minutum* on egg masses of spruce budworm has been observed by Smith *et al.* (1986) and Smith and Hubbes (1985). Although further information is required on the functional response of *T. minutum* to spruce budworm under varying stand conditions, the abundance of host eggs appears to be less important to the success of inundative releases than their temporal distribution.

Weather had a significant impact on parasitoid activity. Cool wet conditions during the second release in 1985 reduced and extended parasitoid emergence, thus lowering the effectiveness of the releases. Smith *et al.* (1986) observed a similar effect with ground releases in 1982. It is suggested, then, that *T. minutum* produced in eggs of *S. cerealella* not be released during cool rainy conditions. When such conditions exist, it may be best to hold the parasitoids at cool temperatures until weather conditions improve. Before such a strategy can be implemented, however, information is needed as to the effect of such holding on later parasitoid emergence, longevity, and fecundity. In this way, losses in parasitoid quality due to holding can be weighed against expected losses in the field under rainy conditions.

In all years of our study, parasitoid quality was identified as an important factor in the success of the releases. Lopez and Morrison (1985) came to a similar conclusion when examining parasitoid activity on eggs of *Heliothis* species. The components previously discussed were influential because of their effect on parasitoid emergence, longevity, ability to locate hosts, and fecundity. The production, programming, and shipment systems for *T. minutum* must ensure a parasitoid with high percentage emergence, proportion of females, longevity, and fecundity. If any of these three steps is mishandled, then parasitoid quality will be lowered and the proportion of runts (brachypterous adults) increased. Such an effect was observed in 1984 when *T. minutum* were shipped from California without proper cooling; parasitoid emergence began before release, resulting in extremely low parasitism. Future studies must ensure that these steps are carefully controlled to achieve consistently high results following release.

Of prime consideration in parasitoid quality is the continual rearing of parasitoid colonies for release. After 1983, the parasitoid material in the present study was not infused with new strains of *T. minutum* from the field but only with other strains reared equally as long in the laboratory. With ca. one generation completed every 2 weeks, the *T. minutum* strain released in 1986 had been reared for more than 100 generations in the laboratory. Although emergence and sex ratio remained relatively unchanged over this period, studies in the field showed that longevity and the number of progeny surviving per parasitized host egg declined. In contrast, maximum parasitism of sentinel egg masses at similar application rates remained the same over this period. The effect of continual mass production on efficacy has been seldom reported in the literature; if the use of *Trichogramma* is to become feasible, future research must address this potential problem.

The number of parasitoids required for release will depend naturally on many factors, but principally cost and the desired level of spruce budworm reduction (i.e. stand protection). Application rates over 12×10^6 ♀♀ per hectare per release do not justify the increased cost because the increase in parasitism is only marginal. In our study, the reduction in larval populations was proportional to the rate of parasitoid release; a rate of 4.2×10^6 ♀♀ per hectare did not lower budworm populations and, therefore, was too low to provide stand protection. Higher rates of $8.4–22.7 \times 10^6$ ♀♀ per hectare, however, did substantially reduce larval populations (42–82%) below the level that results in 50% defoliation and at which suppression measures are implemented. This is the first time that aerial releases of *T. minutum* have been shown to increase parasitism of spruce budworm eggs significantly and cause a corresponding reduction in larval populations. Unfortunately, the natural collapse of budworm populations in the study area during 1986 prevented us from obtaining another year of data to confirm this relationship between egg parasitism and larval reduction.

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