Journal of the Lepidopterists' Society
42(2), 1988, 120–131

BIOLOGY OF THE BLUEBERRY LEAFTIER *CROESIA CURVALANA* (KEARFOTT) (TORTRICIDAE): A FIELD AND LABORATORY STUDY

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ABSTRACT. Biology of *Croesia curvalana* (Kearfott) is described for the first time. Laboratory-laid eggs were white, later brown, 0.6 mm in diam., and were deposited singly under blueberry branches. Seventy-five percent hatched when given a 7-day chilling at 6°C followed by a 24-week cold treatment at 0°C. Four instars occurred during the 21-day larval development period at 21°C. Male pupal stage was 9 days, 2 days less than females. Field eggs were laid on surface litter under blueberry plants in July and August, and eggs overwintered. Flower buds were invaded by emerging larvae in the last part of April and early May, and pupation occurred during the first half of June. During four years of study, the flight season began at Blackville, New Brunswick, during the first week of July, and later at Pouch Cove, Newfoundland. Larvae from Pouch Cove were parasitized by *Chorinaeus excessorius* (Davies). In trapping experiments, virgin female *Croesia curvalana* attracted the largest proportion of males between 2200 h and 2400 h. Male *Croesia curvalana* were attracted to sex attractant lures and virgin *Choristoneura fumiferana* (Clem.) females between 2000 h and 0400 h.

Additional key words: Tortricidae, eggs, diel periodicity, trapping, *Vaccinium angustifolium.*

Croesia curvalana (Kearfott), commonly called blueberry leaftier (BBLT), are responsible for serious crop losses in Newfoundland, where lowbush blueberries, *Vaccinium angustifolium* (Ait.), are a two-million-pound export crop (Morris 1981). The insect was first recorded in Newfoundland in 1979, and subsequently reported to infest up to 30 percent of blueberry buds in 12 fields sampled in New Brunswick (G. Wood pers. comm.). Incidence of infestation is increasing due to change in blueberry cultivation practices: field burning every two years has been replaced by mowing because of rising oil prices and soil damage.

Kearfott (1907) described *Croesia curvalana,* as one of four "varieties" of *Tortrix albicomana* (Clemens) feeding an oak, rose, and huckleberry. MacKay (1962) described larval morphology based on a probable last instar. She placed it in the tribe Tortricini, genus *Argyrotoza.* Larval host plants were said to be Vacciniaceae, with distribution from Nova Scotia to British Columbia. Subsequently Powell (1964) and Hodges et al. (1983) listed the insect as *Croesia curvalana.*

In 1979 we discovered that adult male *C. curvalana* were attracted to virgin spruce budworm, *Choristoneura fumiferana* (Clemens), (SBW) adult females. Trapped moths were identified by the Forest Insect and Disease Survey (FIDS) of Environment Canada, and the Biosystematic Research Centre, Agriculture Canada. Initial trap capture of *C. curvalana* occurred in traps hung at 1.5 m above ground in balsam fir

stands. This finding suggested that some components of spruce budworm sex pheromone were also BBLT sex attractants. Sanders and Weatherston (1976) identified the primary components of SBW sex pheromone as (E) and (Z)-11-tetradecenal (96:4), and Silk et al. (1980) found traces of tetradecanal and E-11-tetradecenyl-acetate in the effluvia.

Little is known of BBLT biology. This paper describes laboratory studies from 1980 to 1983 concerning duration of egg diapause, larval, and pupal development; suitability of oviposition substrates, and fecundity. Field studies are also presented which explore the BBLT life cycle and examine diel periodicity of male attraction to calling females.

MATERIALS AND METHODS

Laboratory

Egg treatment during diapause. Eggs collected in August 1981 failed to hatch when held in the laboratory for 4 months at 21°C with temperature variations of ±2°C. A cold treatment was therefore provided for egg collections made the next year. Sequencing of temperature and photoperiod throughout diapause in the laboratory was similar to that used for SBW (Stehr 1954). Field-collected eggs laid on dead leaves under blueberry plants were stored in Petri dishes lined with dampened filter paper, sealed with parafilm, and held at 21 ± 2°C for 18 to 37 days. One batch of 1144 eggs was chilled at 6 ± 1°C for 7 days in a dark refrigerator. A 2nd batch of eggs was exposed to cold treatment of 0 ± 1°C in a freezer with no chilling. Samples from both batches were removed from the freezer after 18, 21 and 24 weeks. All eggs were placed in a refrigerator at 6 ± 1°C for 2 days before being exposed to a constant temperature of 21 ± 2°C in a 17L:7D photoperiod. Time required for eggs to hatch after removal from the freezer and percentage hatch were measured.

Larval and pupal rearing. After hatch, 994 larvae were transferred using a mohair brush to artificial-leaf-meal diet in plastic creamer cups (4 per cup) or to young foliage, and reared at 21 ± 2°C in a 17L:7D cycle at 70% RH. Diet was that developed for SBW (McMorran 1965) as modified by Grisdale (1973), to which was added dried blueberry leaf meal (50% v/v). The meal was produced by drying and grinding previously frozen leaves from June collections. The diet was allowed to dry for a day at room temperature, which made it draw away from cup sides and provide niches for larvae. Twenty larvae were reared singly from hatch to adult emergence to determine number of larval instars and time required for maturation. Exuvial head capsules were collected and widths measured.

Oviposition substrates. When neonate larvae were transferred from leaves to diet, mold spores also transferred immediately contaminated the diet. Therefore, a study was undertaken to examine suitability of other oviposition substrates. In 1983, 10 newly emerged virgin females were placed with males at a ratio of 1:1.5 in each of 8 screened cages measuring 30 cm^3. Moths were provided with a live blueberry branch, a 10% sucrose source, and a selection of oviposition substrates: parafilm, waxed paper, aluminum foil, filter paper strips, all 3 cm × 15 cm, and glass (2 bottles each of 21 cm surface area) which were placed on cage bottoms. In two cages, dried leaves were also offered as an oviposition substrate. Numbers and viability of eggs deposited on each substrate were determined. Cages were maintained in a 17L:7D light cycle at corresponding temperatures of 21 and 17°C with 70% RH for 3 weeks, after which eggs were counted. Eggs on the various substrates were given the cold treatment found effective in the initial diapause study, and emerging larvae counted.

Fecundity. Numbers of eggs produced by virgin and mated female moths under laboratory conditions were investigated. Single virgin females and male-female pairs were reared in 12-ml vials containing a 10% sucrose source. Seventy-one virgins 0 to 11 days old were dissected and their eggs counted. Females were dissected in a 5.5-cm Petri dish to expose the reproductive system. One or more drops of Shaeffer Script permanent blue-black ink diluted 50% with water was added to the preparation. Ovarioles were separated and eggs counted using a base-lit dissecting microscope at 160–400 magnification. Eggs were regarded as mature when they reached ca. 0.3 mm diam., the size at oviposition. Eggs, unfertilized and fertilized, were also counted from 31 mated 5- to 10-day-old females.

Field

Areas studied. Two geographically distinct regions were selected for study. The Pouch Cove, Newfoundland, blueberry barrens, which were used in 1984 and 1985, were rocky and windswept. An area near Blackville, New Brunswick, which was used from 1980 to 1984, had less open terrain, and blueberry plants were often interspersed with ferns, small trees, and bushes.

Life history. In late August 1981, two weeks after the flight season, whole live blueberry plants, surrounding vegetation, and surface litter were collected at Blackville. All materials were examined in the laboratory for BBLT eggs. Samples of leaf litter were again collected from the same area in October to check for egg hatch.

Timing of insect development at Blackville was investigated in 1981. First invasion of flower buds by larvae was monitored by microscopic

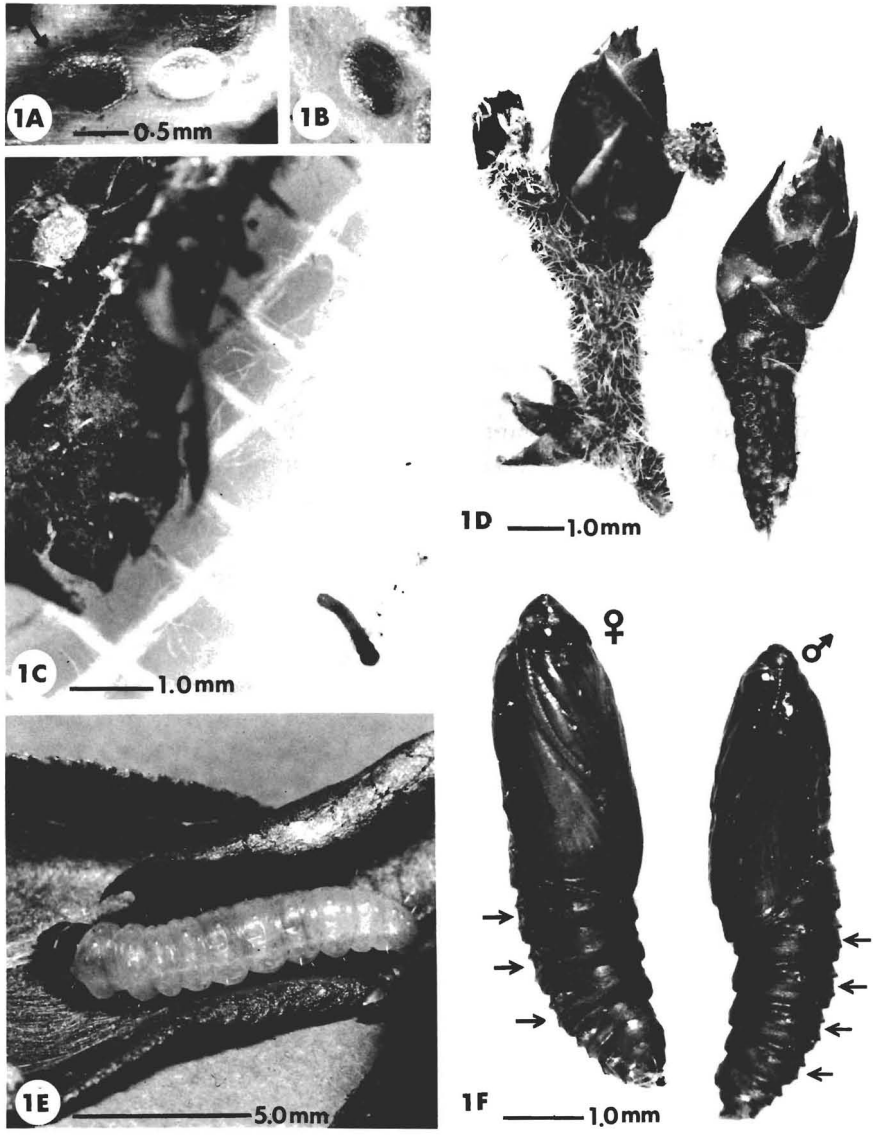

FIG. 1. *Croesia curvalana* life stages and injury to host. **A,** Fertile (arrow) and infertile eggs; **B,** Fully mature egg two days before eclosion; **C,** Newly emerged larva with empty egg; **D,** First-instar entry holes in blueberry buds; **E,** Fourth instar; **F,** Male and female pupae (arrows indicate moveable abdominal segments).

TABLE 1. Effect of chilling at 6 ± 1°C and duration in freezer at 0 ± 1°C on timing and success of *Croesia curvalana* egg hatch. All eggs received a post-freezer period of 2 days in refrigerator at 6 ± 1°C before being incubated at 21 ± 2°C. N = 2100.

No. chilling days in refrigerator	No. weeks in freezer	Percent of hatching eggs after removal from freezer				Percent hatch of eggs subjected to treatment
		5–11 days	12–18 days	19–25 days	26–32 days	
7	24	45	44	9	2	75
7	21	4	66	28	2	57
7	18	0	64	35	1	55
0	24	8	55	31	6	49
0	21	0	55	45	0	20
0	18	0	36	47	17	31

examination of blueberry plant clippings beginning in early April. Foliage was subsequently clipped for examination at two-week intervals for observations of larval and pupal development.

Parasites. In 1984, 528 late instars collected at Blackville and 102 from Pouch Cove were reared singly in the laboratory on foliage to determine incidence of parasitization.

Trap height. Height of male flight and trap height for optimal male capture were investigated at Blackville in mid-August 1980. Initial capture of male BBLT in SBW-baited traps had occurred at a height of 1.5 m. Four Pherocon® 1C sticky traps were each baited with 2 spruce budworm virgin females 0 to 24 h old in small screen cages, and 4 Pherocon® traps were left empty. Two moth-baited traps, and two empty Pherocon® 1C traps were hung 1.5 m above the blueberry canopy and 2 of each at 10 cm above the canopy. Traps were 30 m apart. Captured moths were counted each day for 10 days and the SBW virgins were replaced every 2 days.

Flight season. Onset and duration of the flight season was studied during four seasons in Blackville and two seasons at Pouch Cove by sweep netting and by capturing males in sticky traps. Traps were placed at canopy level in advance of the flight season and monitored every 48 h. They were baited with polyvinyl chloride (PVC) lures (Fitzgerald et al. 1973) formulated by G. Lonergan, Department of Chemistry, University of New Brunswick, to release (E) and (Z)-11-tetradecenal (95:5) with small amounts of (E)-11-tetradecanyl-acetate (0.2) at the rate of 1 SBW equivalent (Sanders 1981). These had attracted leaftier males in previous experiments (Ponder unpubl.).

Periodicity of sexual activity. Diel periodicity of sexual activity under field conditions was observed at Blackville between 13 and 16 July 1982 in a 96-h trapping study. Pherocon® 1C traps were each baited with one of the following: two virgin SBW females, two virgin BBLT

females, a PVC lure releasing a sex attractant at the rate of one SBW equivalent, or a blank PVC formulated without attractants. Each trap type was replicated three times within the array. Virgin SBW females were included in this experiment as lures because of their proven success in the capture of male leaftiers. Traps were separated by 30 m and their initial positions in the 12-placement array were selected by random numbers. Trapped moths were counted and traps were moved foward by 1 position every 2 h because the population was not uniformly distributed.

RESULTS AND DISCUSSION

Laboratory

Egg treatment during diapause. Three to 4 days after removal from 18 to 24 weeks in the freezer, and 2 to 3 days before hatch, a black head and larval outline could be observed inside eggs (Fig. 1B).

Significant decreases in egg mortality occurred with acclimation. Chilling eggs at 6°C in the refrigerator for one week before putting them in the freezer enhanced hatch (Table 1). The longer period of 24 weeks in the freezer resulted in significantly increased egg hatch (2-way ANOVA w/o replication, $P < 0.05$). Seventy-five percent of eggs hatched if given a 24-week freezer treatment after 7 days of chilling (Table 1).

Larval and pupal rearing. Larvae matured through 4 instars to pupation in 21 (SD ± 3, N = 20) days at 21 ± 2°C. Mean head capsule widths (mm ± SD) progressing through instars were 0.25 ± 0.03, 0.35 ± 0.04, 0.57 ± 0.05, and 1.22 ± 0.04. Hatchlings were 1.2 mm long, cream colored, with a dark thoracic shield and black head (Fig. 1C). Second and third instars remained cream colored, had black heads and thoracic shields, and dark anal shields. Fourth instars (Fig. 1E) became yellow, and the head changed to cinnamon brown; the thoracic shield was cinnamon brown medially, shading to dark brown laterally. Male gonads in the fifth abdominal segment were maroon, simplifying larval sexing. Exuvial head capsules appeared slightly lighter in color than the head in the last two instars.

Mortality was high in the 994 larvae fed leaf-meal diet; only 22% survived compared with 50% on fresh foliage under the same conditions, though maturation time was approximately equal. Mortality of diet-fed larvae could be attributed in part to mold transferred with hatchlings from leaves to diet. No attempt was made to surface-sterilize eggs. Larvae did not feed on previously frozen blueberry foliage unless mixed with SBW diet. An attempt had been made in 1980 to rear 230 larvae on SBW diet without addition of blueberry meal. Three larvae survived

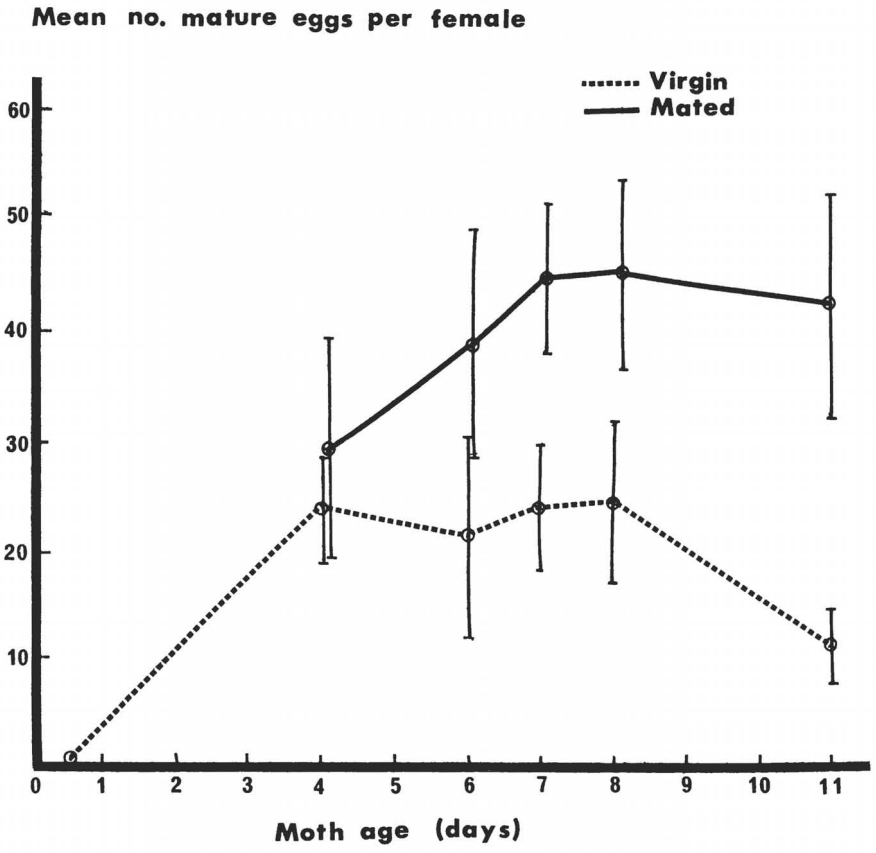

FIG. 2. Numbers of mature eggs in mated (N = 31) and virgin (N = 71) female *Croesia curvalana* in relation to age. Vertical bars indicate SD.

to pupation, and larval development time was 75 to 80 days. Newly hatched larvae seldom burrowed into flower buds (Fig. 1D) on diet, but rather spun nests between the diet and creamer cup wall.

Pupation occurred either in spun nests between diet and creamer cup wall, or under the lid. Males pupated before females, and mean male pupal duration was 9.2 (SD ± 0.8, N = 20) days which was shorter by 2 days than mean female pupal duration of 11.5 (SD ± 1.3, N = 20) days. Male pupae could be distinguished by presence of a fourth moveable abdominal segment (Fig. 1F), females having only three moveable segments.

Oviposition substrates. Few eggs were deposited by BBLT females in the laboratory. Such eggs were scattered singly on materials placed in cage bottoms. Eggs were white, flattened, convex, 0.6 mm in diam.,

and had a clear pebbled surface. Within a week they changed to reddish brown as the enclosed embryo matured (Fig. 1A). In total, 1061 eggs were collected in 1983 on synthetic substrates, 44% on waxed paper, 37% on parafilm, 8% on aluminum foil, 7% on filter paper, and 4% on glass. No preference was shown for dried leaves as an oviposition substrate. Egg maturation and hatch after diapause was 66% on waxed paper, and 64% on filter paper, but only 32% on parafilm. Other substrates resulted in less hatch.

Fecundity. No mature eggs were found when 5 newly emerged virgins were dissected; however, a mean of 90.8 (SD ± 11.4) immature eggs were counted. In mated females, ca. ⅓ of eggs matured by the 4th day (Fig. 2), and up to 50% matured by the 7th day after emergence. Twenty-six percent matured by the 7th day in virgins. Both mated and virgin females laid few eggs in vials. A total of 4 unfertilized eggs laid by the 71 virgins remained white. Fifty-five fertile eggs which changed from white to brown were laid by the 31 mated females.

Decrease in numbers of eggs in virgin females between days 8 and 11 (Fig. 2) suggested that their eggs were resorbed. Most moths (79%) died between 11 and 12 days after emergence; however, some lived 15 days.

Many factors contributed to the difficulty of rearing this insect on artificial-meal diet. Fecundity was low. The preferred oviposition substrate was ill-defined, and many substrates proved unsuitable for complete egg maturation. Hatchlings were small, delicate, and difficult to locate even with a microscope. Year-round rearing of BBLT using fresh vegetation would be impractical without light- and temperature-controlled greenhouse and refrigeration for continuous propagation of blueberry plants.

Field

Life history. Eggs were laid singly on dried leaf litter under blueberry plants, and were difficult to locate even with a microscope. No eggs were located on living plant leaves, stems, or branches. October collections of eggs indicated that hatch had not yet occurred. Seventy-five percent of field-collected eggs hatched successfully in the laboratory when given a treatment of 1 week at 6°C and 24 weeks at 0°C. These findings indicate that eggs overwinter. Blueberry cultivation practices that incorporate field mowing rather than burning may therefore result in increased leaftier populations.

Foliage clipped weekly from April to mid-June at Blackville indicated that infestation of flower buds by first-instars occurred during the last two weeks of April (Table 2). Larvae burrowed into closed flower buds leaving a round hole marked by an accumulation of yellow frass. Fre-

TABLE 2. Development of *Croesia curvalana* larvae, Blackville, N.B., 1981.

Date	Stage of blueberry foliage	No. larvae collected	Instar no. or stage
18 April	Closed buds	0	
24 April	Closed buds	6	1
6 May	Expanded flower buds	327	1
12 May	Expanded leaf buds	121	1
		129	2
22 May	Young leaves	62	2
		102	3
2 June	Expanded leaves and flowers	247	3
		153	4
12 July	Flowers and immature fruit	2	4
		187	pupae
		15	pupal cases

quently, two larvae were found feeding in the same bud. Larvae subsequently fed on swelling leaf buds. Numbers of buds infested with larvae increased to mid-May, indicating a three-week period of egg hatch. Visible plant damage peaked just before larvae pupated. At this time, terminal leaf growth was webbed and eaten, and larger leaves were folded or webbed together to form shelters. Increased numbers of abandoned shelters during late-instar development suggested that larvae moved frequently.

First appearance of pupae in the field at Blackville ranged from the first to third weeks in June. Males pupated before females. The dark brown pupae could be found sandwiched in shelters or occasionally hanging freely by the cremaster from blueberry twigs.

The moths were 5 to 7 mm long, and were of a yellowish hue with forewing markings of rust and yellow. Toward the end of the flight season, spent moths lost many wing scales, which made them appear cream colored. First male moth emergence at Blackville, as established by trap capture, occurred during the first week of July in all four years of study. It occurred thus regardless of differences in weather during the larval and pupal stages. Sweep-net collections indicated that, as in the laboratory, males emerged before females. Sweep-net collections were achieved two to three days after first male trap capture. First sweep-net collections had a male : female ratio per sweep of 0.116:0.044, which changed to 0.009:0.008 by the end of the flight season. At Pouch Cove where the population was higher, the ratio changed from 0.23:0.02 to 0.22:0.16 by the end of the flight season. Moth location may have had a bearing on results. At Blackville where mean day time temperatures were 5°C higher than Pouch Cove, moths preferred shad-

TABLE 3. Trap capture of male *Croesia curvalana* in Pherocon® 1C traps hung at different heights in 10 days, Blackville, N.B., 1980. Number of traps = 8.

Trap height above blueberry canopy	Mean 24-h catch per trap by virgin spruce budworm	Mean 24-h catch per unbaited trap
1.5 m	6.0	0.1
10 cm	17.6	0.7

ed areas, while at Pouch Cove where the barrens had mean wind velocities of 21.6 km/h and mean RH of 82%, moths were located in sheltered areas of deep vegetation. Female moths may have been at a lower stratum or beneath vegetation during oviposition, making sampling for females by sweep-net unreliable.

Length of flight season in the 4 years of study at Blackville ranged from 30 to 47 days; at Pouch Cove, it began in 1984 on 12 July and lasted 35 days in 1984, and in 1985 on 19 July and lasted 28 days.

Parasites. Larvae collected at Blackville were parasitized 10% by tachinid flies which emerged as larvae from their hosts. Tachinid puparia were held in the laboratory for eight months without a cold period. One fly emerged in too poor condition to identify. No tachinids were found in larvae from Pouch Cove. Two ichneumonids emerged as adults from pupae collected as larvae at Pouch Cove. These were identified by the Biosystematic Research Centre, Agricultre Canada, as *Chorinaeus excessorius* Davies. This parasite has not been reported previously from BBLT.

Trap height. Significantly more male BBLT were captured in traps hung at 10 cm than at 1.5 m above the foliage in both unbaited and virgin SBW baited traps $\chi^2_c = 109.3$, $P \ll 0.001$, df = 1) (Table 3) which confirmed visual observations that moths flew immediately above the foliage.

Croesia semipurpurana (Kft.), a species morphologically similar to *C. curvalana*, is attracted to traps hung at 1.5 m baited with components (Grant et al. 1981) which are also part of the spruce budworm sex pheromone bouquet.

Periodicity of sexual activity. The largest proportion of BBLT males trapped by all bait types was between 2200 and 0200 h (Table 4). Virgin BBLT females captured the largest proportion of BBLT males from 2200 to 2400 h ($P = 0.05$, Chi square for multiple proportion, Zar 1984) indicating that female sex pheromone release (calling) took place during these hours. PVCs which released attractant continuously over a 24-h period attracted BBLT males consistently between 2000 and 0400 h, suggesting that male flight period and attraction to lures may extend

TABLE 4. Proportion of 96-h trap capture of male *Croesia curvalana* by 2-h intervals at Blackville, N.B., 1982. Baits in Pherocon® 1C traps were replicated 3 times (N = 1513). No moths were captured between 1000 and 1400 h.

Bait	Trapping interval (h) (AST)									
	1400–1600	1600–1800	1800–2000	2000–2200	2200–2400	2400–0200	0200–0400	0400–0600	0600–0800	0800–1000
Two virgin BBLT	0.00	0.00	0.00	0.08	0.55	0.27	0.02	0.02	0.02	0.05
Blank PVC	0.11	0.00	0.00	0.04	0.44	0.33	0.04	0.00	0.04	0.00
PVC sex	0.01	0.01	0.01	0.26	0.30	0.30	0.11	0.01	0.01	0.00
Two virgin SBW	0.00	0.00	0.00	0.23	0.57	0.16	0.03	0.00	0.00	0.01

on either side of the virgin female BBLT calling period. This was confirmed by the finding that virgin female SBW, which would have been calling from 2000 h to 2400 h (Palaniswamy & Seabrook 1985), also attracted BBLT males before the BBLT female calling period.

ACKNOWLEDGMENTS

We thank Agriculture Canada in St. John's, Newfoundland for providing laboratory space and personnel, and George Wood and R. F. Morris for advice and encouragement. We are indebted to the Government of Newfoundland and Labrador for providing a truck for fieldwork in 1984 and 1985, and to Paul Hendrickson, Small Fruit Specialist, Government of Newfoundland, for aid in selecting infested areas. We thank L. J. Dyer and D. C. Eidt for helpful criticisms of the manuscript, and L. R. Kipp for assistance with numerical testing. Financial support for this study was received by W. D. Seabrook from Agriculture Canada and the Natural Sciences and Engineering Research Council.

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Received for publication 28 August 1987; accepted 27 January 1988.