NOTE

Thoracic Leg Coloration of Soybean Looper and Cabbage Looper Larvae (Lepidoptera: Noctuidae): an Unreliable Field Identification Character for Species Separation[1]

Douglas J. Jost[2] and Henry N. Pitre

Department of Entomology and Plant Pathology, Mississippi State University, Mississippi State, MS 39762 USA

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One or more applications of insecticide are often required to protect late-season cotton and soybean from insect defoliators such as soybean looper, *Pseudoplusia includens* (Walker), and cabbage looper, *Trichoplusia ni* (Hübner) (Lepidoptera: Noctuidae), larvae. The soybean looper has become resistant to some frequently used insecticides including organophosphate and carbamate compounds (Newsom et al., 1980, pp. 51–98, *In* C. B. Huffaker (ed.), New Technology of Pest Control, Wiley and Sons, NY), as well as pyrethroids (Felland et al., 1990, J. Econ. Entomol., 83: 35–40; Leonard et al., 1990, J. Econ. Entomol., 83: 27–34; Portillo et al., 1993, Florida. Entomol., 76: 577–584), while the cabbage looper remains more susceptible to these chemicals. During 1995 and 1996, we collected looper larvae in cotton and soybean production fields during the later part of the crop growing season in Mississippi. Loopers were reared to moths and moths were identified based on adult characteristics (Lafontaine and Poole, 1991, The Moths of America North of Mexico Including Greenland, fasc. 25.1.). Two species were present on both crops.

Due to variation in looper susceptibility to insecticides, proper larval identification is necessary for adequate looper management in both cotton and soybean. Thoracic leg color of larvae often is used to identify loopers on cotton and soybean and may serve as the basis for pest management recommendations. The common practice is to identify loopers with black thoracic legs as soybean loopers (often referred to as the "black legged looper") and those with green thoracic legs as cabbage loopers (Hensley et al., 1964, J. Econ. Entomol., 57: 1006–1007; Miss. Coop. Ext. Serv., 1994, Info. Sheet 1400). However, we have found no published data to support using this phenotypic characteristic to positively identify these two looper species.

As this looper identification practice has not been verified, thoracic leg color of soybean looper and cabbage looper larvae was examined, and larvae were reared to

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[2]Present address: Monsanto Leland Agronomy Center, P.O. Box 388, Stoneville, MS 38756.

adults for identification to determine relative frequencies for each species in cotton and soybean. In addition, the commonly held belief that increased population density may result in a change of phenotypic coloration in some insects, the characteristic black-legged trait of soybean looper was evaluated using larvae collected from soybean during 1996. On 15 August 1995 and during the sampling period 10 July through 14 August 1996, a total of 286 and 473 first through fifth instars, respectively, were collected using a beat sheet from cotton and soybean at two locations in Holmes Co. Sixth instars were not collected due to observed variation of thoracic leg color during the prepupal stage. The locations were separated by approximately 13 km. 'Terra C-40' cotton and 'Terra Cajun' (maturity group VI) soybean were planted on 28 April and 19 May 1995, respectively, in location 1. 'Deltapine 90' (DPL-90) cotton and Terra Cajun soybean were planted on 26 May and 4 May 1995, respectively, in location 2. In 1996, 'Deltapine 50' (DPL-50) cotton and 'Pioneer 9592' (V) soybean were planted on 3 May and 10 May, respectively, in location 1. DPL-50 and 'Hornbeck 49' (IV) soybean were planted on 5 May in location 2. All larvae collected were placed individually into empty 30-ml clear plastic diet cups, capped, placed in an ice chest, and transported to the laboratory. In the laboratory, each larva was placed in a separate 30-ml plastic diet cup containing flash-sterilized wheat germ agar diet (King et al., 1985, J. Econ. Entomol, 78: 1166–1172.) and cups were capped. Approximately 12.5 ml of raw linseed oil was added per liter diet. Sterile corn cob grit (0.15 g) treated with 0.30 g Phaltan and 0.40 g Griseofulvin per 1,000 g grit was added to the surface of the diet in each diet cup to prevent or delay fungal contamination. Larvae were reared at 31°C and 70% RH under a photoperiod of 15:9 (L:D) h. Because leg color may change immediately after molting due to melanization (Chapman, 1982, The Insects Structure and Function, 3rd ed., Harvard University Press, Cambridge, MA), thoracic leg color was observed daily throughout larval growth stages. Thoracic leg color was recorded as black or green, and adult loopers were correctly identified after adult eclosion. Statistical analysis of the fractions of looper larvae with black thoracic legs collected from soybean in 1996 included z test ($P > 0.01$) (Steel and Torrie, 1980, Principles and Procedures of Statistics, McGraw-Hill, NY, pp. 167–170). Number of insects collected from sampling days 11 July, 18 July, 24 July, and 1 August were pooled due to low larval density and compared to soybean looper larvae collected from soybean on sampling dates 7 and 14 August.

The occurrence of black or green thoracic leg color of soybean looper and cabbage looper larvae varied based on host crop plant (cotton or soybean) and year (1995, 1996) (Table 1). In 1995, most soybean looper larvae collected from cotton (69/79, 87%) or soybean (188/190, 99%) had black thoracic legs, whereas 31% of the cabbage looper larvae collected from cotton (5/16) had black thoracic legs. The only cabbage looper larva collected from soybean had black thoracic legs. In 1996, the percentage of soybean loopers collected from cotton (14/28, 50%) and soybean (225/407, 55%) with black thoracic legs was much lower than in 1995. Very few cabbage looper larvae collected from cotton (1/25, 4%) had black thoracic legs, whereas cabbage looper larvae collected from soybean (6/13, 46%) were as likely to have black thoracic legs as green thoracic legs.

The sampling period differed between years, with collections of larvae occurring on 15 August in 1995 and throughout the sampling period in 1996. During the sample period in 1995, larval density on soybean steadily increased (observations noted, but date not recorded). The increased soybean looper population density may have influenced morphological color change of thoracic legs. However, no observations on

Table 1. Identification of *Pseudoplusia includens* (SBL) and *Trichoplusia ni* (CL) from cotton and soybean based on thoracic leg color

Year	Crop	Insect	n	Fraction with black legs	SE
1995	Cotton	SBL	79	0.87	0.038
	Soybean	SBL	190	0.99	0.007
	Cotton	CL	16	0.31	0.116
	Soybean	CL	1	1.00	—
1996	Cotton	SBL	28	0.50	0.095
	Soybean	SBL	407	0.55	0.025
	Cotton	CL	25	0.04	0.039
	Soybean	CL	13	0.46	0.138

thoracic leg color were made prior to the 15 August sample date. Larvae of other lepidopterous species including *Anticarsia gemmatalis* Hübner (Hammond and Fescemyer, 1987, Insect Sci. Applic., 8: 581–589; Fescemyer and Hammond, 1988, J. Insect Physiol., 34: 29–35), *Spodoptera litura* (F.) (Morita et al., 1988, J. Insect Physiol., 34: 751–758), and *S. littoralis* (Boisduval) (Rivnay and Meisner, 1966, Bull. Entomol. Res., 56: 623–634) have shown morphological color variation in crowded conditions.

Thoracic leg color of loopers was determined in 1996 during the time when larval population density was steadily increasing. A significantly higher fraction of black-legged soybean looper larvae was collected from soybean on 14 August compared to the other sample dates (Table 2). Prior to this sample date, no significant differences in thoracic leg color were observed. In 1995, soybean looper larval population density

Table 2. Identification of *Pseudoplusia includens* from soybean based on thoracic leg color, 1996

Julian date	n	Fraction with black legs	Date	n	Fraction with black legs*	SE
194	1	1.00				
201	2	0.0				
207	12	0.17				
215	6	0.0	215**	21	0.14a	0.076
221	38	0.21	221	38	0.21a	0.066
228	348	0.62	228	348	0.62b	0.026

* Fractions with same letters do not differ significantly ($P > 0.01$), z test.

** Pooled data; July 11, dates: 194, 201, 207, and 215.

was monitored on soybean during the 2 wks prior to the 15 August sampling date, with soybean looper density increasing from 1,857 to 37,185 larvae/ha. Although looper thoracic leg color was only recorded for larvae collected on 15 August, the substantial increase in soybean looper population density late in the crop growing season may have contributed to increased incidence of black-legged soybean loopers. This crowding effect would support observations of increased incidence of black-legged soybean loopers collected from soybean in 1996. However, further studies comparing soybean loopers in crowded and uncrowded conditions should be conducted to support this hypothesis. Similar studies should be conducted with cabbage looper larvae.

These observations indicate that cabbage looper larvae collected from cotton or soybean may have black thoracic legs, which contradicts common belief (Hensley et al., 1964, J. Econ. Entomol., 57: 1006–1007; Miss. Coop. Ext. Serv., 1994, Info. Sheet 1400). In addition, thoracic leg color of soybean looper larvae may change from green to black due to crowded conditions or other factors (i.e., environmental conditions, nutritional quality of the host, or combination of factors) unknown at present. Using thoracic leg color as the single method for field identification and separation of soybean looper and cabbage looper larvae may lead to incorrect estimates of looper populations in the field.

The results of this study further suggests that population ratios of soybean looper to cabbage looper larvae in soybean and cotton may not be consistent from one year to the next. However, because soybean loopers appeared to be the most prevalent of the two species in soybean, the widely accepted method of identifying soybean loopers in soybean using the black-legged phenotypic character may remain a useful generalization for insect pest management practitioners. And because the looper population in cotton may be predominately soybean loopers or a complex near equal numbers of soybean and cabbage loopers, depending upon time during the growing season and the year, this same generalization could apply in considering looper populations in cotton.

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