

# Biology and Rearing of *Diapetimorpha introita* (Cresson) (Hymenoptera: Ichneumonidae) on Host and Non-host Noctuid Pupae<sup>1</sup>

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J. Entomol. Sci. 30(4): 468-480 (October 1995)

**ABSTRACT** Laboratory studies were conducted to identify factors that influence *Diapetimorpha introita* (Cresson) reproduction and development rate on *Spodoptera* spp., *Helicoverpa zea* (Boddie), and *Heliothis virescens* (F.). Laboratory colonies of *D. introita* were successfully maintained on *S. frugiperda* (J. E. Smith) pupae placed in multicellular rearing units. Mated *D. introita* females produced an average of 81.3 eggs per individual over a mean lifespan of 23.6 days. Sex ratios were skewed with 40% more males than females produced from mated females. Unmated females produced all male progeny. *Diapetimorpha introita* females tended to select 3 to 5-day-old *S. frugiperda* pupae over younger or older pupae. *Diapetimorpha introita* successfully developed to the adult stage on host *Spodoptera* spp. and non-host *H. zea* and *H. virescens* pupae. Duration of larval stages was dependent upon host and temperature. Duration of pupal stages not exposed to diapause-inducing temperatures were dependent upon host, and adult weights were dependent upon host species, host weight, and temperature. Temperatures of 18 or 22°C resulted in ≥ 90% diapause regardless of host species. Generally, parasitoids from large host pupae developed more slowly and weighed more than those developing on smaller hosts. Successful utilization of *D. introita* as a candidate for augmentative releases against overwintering populations of *Spodoptera* spp. depends upon the advancement of *in vivo* mass rearing techniques.

**KEY WORDS** *Diapetimorpha introita*, pupal parasitoid, developmental biology, *Spodoptera* spp.

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Considerable information is available describing the biology of the parasitoids of noctuid larvae. However, little is known of the species that seek out and attack the pupal stage of noctuid pests, especially those that pupate beneath the soil surface. Pair and Gross (1984, 1989) first reported the existence and seasonal population dynamics of ichneumonid pupal parasitoids of fall armyworm, *Spodoptera frugiperda* (J. E. Smith). Identified as *Diapetimorpha introita* (Cresson) and *Cryptus albitarsis* (Cresson), females of these species locate and oviposit in *S. frugiperda* pupal cells. Upon hatching, larvae feed as ectoparasitoids until larval development is completed, whereupon cocoon

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<sup>1</sup> Accepted for publication 08 June 1995.

spinning occurs within the *S. frugiperda* pupal cell. Although *D. introita* attacks *S. frugiperda* as early as May in Georgia, the greatest frequency of *S. frugiperda* cell parasitism occurs during the late fall months in corn (Pair and Gross 1989).

Few reports exist for other parasitoids of lepidopteran pupae that employ reproductive strategies similar to that of *D. introita*. Ullyet (1949, 1950) described the life history of *Cryptus inornatus* (= *latigenalis*) Pratt, an ichneumonid parasitoid of the beet webworm, *Loxostege sticticalis* (L.). *Ichneumon* (= *Pterocormus*) *promissorius* (Erichson) and *Heteropelma scaposum* (Morley) are important pupal parasitoids of *Heliothis armigera* (Hübner) and *H. punctigera* Wallengren in Australia (Room 1979, Wilson 1983, Fitt and Daly 1990). Recently, *I. promissorius* was introduced into the United States for evaluation as a potential biocontrol agent of noctuid pests (Carpenter et al. 1993).

Most current biocontrol strategies for noctuid pests focus upon the larval stages or, in case of the adult stage, autocidal techniques. Suppression of pest species such as fall armyworm in its overwintering habitat prior to its emergence and subsequent dispersal has been suggested as a viable pest management strategy (Knipling 1980). Because there is a need to identify and utilize mortality factors of fall armyworm and other noctuid pupae, we examined the biology of *D. introita* in the laboratory.

## Materials and Methods

**Rearing.** A laboratory colony of *D. introita* was established from field collections of parasitized *S. frugiperda* pupae in corn fields near Tifton, GA. *Diapetimorpha introita* cocoons were held individually in 27-ml plastic cups and placed in environmental chambers set at  $28 \pm 1^\circ\text{C}$  and a 12-h photophase. Relative humidity within the chambers varied from 60 to 80%. Unless stated otherwise, all adult holding and larval rearing studies were maintained under these conditions. Upon emergence, adults were transferred to plexiglass-topped plywood cages ( $33 \times 33 \times 18$  cm) (Gross and Young 1984). Adult *D. introita* were fed drops of honey placed on inverted portion cups placed on the cage floor; moisture was provided by an inverted 9-cm Petri dish that contained a sterile cotton pad soaked with distilled water. Both food and water were replenished twice weekly and adults were transferred to clean cages weekly. Generally, 12 to 15 pairs of *D. introita* were held in each cage.

Initially, rearing was accomplished by releasing 20 to 30 sixth-instar *S. frugiperda* larvae (ready to pupate) into a rectangular plastic pan ( $17.5 \times 12.5 \times 6$  cm) half-filled with moist autoclaved soil or sand. Pans that contained fall armyworm larvae were covered with a lid and placed in a dark area of the laboratory. By placing the larvae in complete darkness, pupation sites were more evenly distributed within the pan. Following pupation, pans containing 2- to 4-day-old *S. frugiperda* pupae were placed in cages containing up to 20 pairs of *D. introita* adults and were exposed for 2 to 3 days for oviposition by *D. introita* females. Pans were then removed from the cages and held at the same conditions as the adults. When the *D. introita* had spun cocoons (usually after 7 days), pupae were removed from the soil, washed in a 3% NaOCl solution, rinsed in distilled water, dried on paper toweling, and placed individually in

labelled plastic cups. *D. introita* pupae were held for emergence under the same conditions as developing larvae and adults. Using the bulk method described above in a soil medium proved unsatisfactory because of a high degree of larval and pupal mortality (approximately 50% or higher) due to bacterial and fungal contamination.

Although *D. introita* generally oviposit single eggs in field situations (Pair and Gross 1984), about 100 eggs were oviposited when a single *S. frugiperda* pupal cell was exposed to 10 to 20 females for 2 days within the cages. We devised a rearing protocol that utilized this behavior to increase both the quality and quantity of adult *D. introita* production in the laboratory. Single, late-instar *S. frugiperda* larvae were allowed to pupate in portion cups containing about 15 ml of soil. Depending upon the quantity of eggs desired, 2 to 3 cups containing 2- to 4-day-old *S. frugiperda* pupae were placed within each *D. introita* holding cage for 24 h. After exposure to *D. introita* females, the eggs were removed from each cell with a fine artist brush, washed in 3% NaOCl solution, rinsed in distilled water, and dried on paper toweling.

The highly cannibalistic behavior of *D. introita* larvae necessitated separation of eggs prior to hatching. When developmental data were required on individual eggs or when first-instar *D. introita* were needed for larval developmental data, eggs were confined individually for hatching after sterilization in a plexiglass brood chamber (10 × 4.3 × 0.5 cm). Vertical holes (5.0 mm diam; 3.0 mm deep) were drilled in the plexiglass block; each block accommodated up to 84 compartments. A plexiglass lid, the same size as the brood chamber except 0.3 cm thick, was clamped in place to prevent egg loss and movement of larvae from cell-to-cell. Thus, eggs could be observed directly for time of hatching, or neonates could be held temporarily until they were placed on host pupae.

It was noted that *D. introita* larvae desiccated rapidly when they were reared unconfined on their host. Therefore, a rectangular block of plexiglass (10 × 4.3 × 1.1 cm) was modified for use as a *D. introita* larval rearing chamber. In each block 20 compartments (1.9 × 0.8 × 1.0 cm) were milled to serve as individual rearing chambers. Single 2- to 4-day-old *S. frugiperda* pupae, surface sterilized as with the *D. introita* eggs, were placed in each compartment. Using an artist brush, single *D. introita* eggs were then placed upon each *S. frugiperda* pupa. To maintain humidity above 70% and to prevent entry of contaminants such as mold spores, etc., each block was wrapped with Saran Wrap™ (Dow Chemical Co., Indianapolis, IN 46268). Thus sealed, adequate relative humidity could be maintained within each rearing chamber. To prevent movement of neonate *D. introita* into adjacent compartments, a plexiglass lid (10 × 4.3 × 0.3 cm) was used to cover all compartments and was secured with rubber bands. Because no light penetrates the subterranean pupation site of its host, growth chambers containing developing *D. introita* were held in total darkness and at the same temperatures as adult *D. introita*. After pupation (cocoon formation), the *D. introita* cocoons were removed from the compartments using a small spatula, surface sterilized, placed in individual portion cups, and held for emergence under the same conditions as the adults. This system proved adequate for rearing moderate numbers of *D. introita* for experimental purposes; mortality rarely exceeded 10%.

**Reproductive Behavior.** Characteristics of *D. introita* mating and searching behavior were accomplished by confining several newly-emerged pairs in the large plywood cages under laboratory conditions and natural light. Fecundity, fertility, and sex ratio of progeny were determined by confining individual pairs of newly-emerged adult *D. introita* in 473-ml paper drink cups fitted with a plastic lid. A 5-cm-diam section of the lid was removed and replaced with screen cloth to allow air circulation and entry of light. Adult *D. introita* were provided drops of honey and water which were dispensed by a medicine dropper inserted through the side of the cage. Each day, two 15-ml cups each containing a single 2- to 4-day-old *S. frugiperda* pupae were placed in the cage. After 24 h of exposure to the *D. introita* female, the portion cups were removed and the eggs were counted and recorded for each female. Eggs were then surface sterilized with 1% NaOCl, rinsed with distilled water, and dried on paper towelling. Using a moistened camel hair brush, individual eggs were transferred to fall armyworm pupae in a plexiglass rearing chamber labelled specifically for each female and date of oviposition. Progeny from each female were then reared to the adult stage and the sex was recorded. Longevity of both sexes was recorded.

Studies were conducted to determine the age of *S. frugiperda* pupae that *D. introita* females preferred for oviposition. On six successive days, late-instar *S. frugiperda* larvae were placed in 27-ml cups containing about 15 ml of sterilized soil. In single holding cages described above, four replicates representing the six ages of *S. frugiperda* pupae (total of 24 pupae) were exposed to 10 previously-mated *D. introita* females for 24 h. Thirteen trials were conducted. Following exposure, cups containing *S. frugiperda* pupae were removed from the cages and the number of eggs deposited by *D. introita* females into the pupation cells were recorded for each age of pupae. The data were converted to  $\text{LOG}_{10+1}$  and analyzed with PROC GLM; mean numbers of eggs oviposited among ages of pupae were separated using Waller-Duncan k-ratio (SAS Institute 1989).

**Developmental Studies.** To determine *D. introita* egg hatchability, at least 20 newly-deposited eggs were removed from the adult holding cages, surfaced sterilized, and placed individually in an egg-holding chamber. The chambers containing eggs were placed in environmental cabinets set at 13, 18, 22, 28, or 33°C temperatures and 60-80 % RH. Eggs in each chamber were observed every 8 h for hatching.

Presently, *D. introita* is known to attack only *S. frugiperda* under field conditions. However, *D. introita* females readily oviposit in other *Spodoptera* spp. pupal cells but reject corn earworm, *Helicoverpa zea* (Boddie), and tobacco budworm, *Heliothis virescens* (F.), pupae in laboratory and cage studies (S. D. P., unpublished data). *Diapetimorpha introita* larval and pupal development was measured on the following noctuid pupae: fall armyworm, *S. frugiperda*; yellow-striped armyworm, *S. ornithogalli* (Guenee); beet armyworm, *S. exigua* (Hübner); *H. zea*; and *H. virescens*. Corn earworm and fall armyworm pupae were obtained from regularly maintained colonies while all other species that were tested originated from collections made near Tifton, GA. Tobacco budworm were reared according to the methods described for corn earworm (Burton 1969). All *Spodoptera* spp. were reared using the procedures for *S. frugiperda* (Perkins 1979).

The number of larval instars and influence of host pupal size on molting behavior were determined by transferring neonate *D. introita* to beet armyworm, fall armyworm, and corn earworm pupae placed in brood chambers and held at 28° C. These three species represented, respectively, small, intermediate, and large host pupae. The chambers were uncovered twice daily and each cell was examined under magnification for the presence of *D. introita* larval head capsules. Each head capsule was considered as a molt. The number of molts was recorded for each larva until cocoon formation was initiated.

The duration of larval and pupal stages of male and female *D. introita* was determined on fall armyworm, corn earworm, tobacco budworm, yellow-striped armyworm, and beet armyworm pupae held at 18, 22, and 28°C and 60-80% RH. Twenty pupae, 2 to 4 days old, of each species were used in each test and tests were repeated at least three times, except with yellow striped armyworm (twice) due to loss of the culture. In each test a single *D. introita* neonate was placed upon each pupa within the brood chamber and maintained in total darkness except for brief periods when the environmental cabinet was opened for inspection. Total darkness was chosen because light does not normally penetrate host pupal cells under natural conditions.

Effects of host species, temperature, and *D. introita* sex upon larval and pupal developmental rates and adult weights were analyzed using PROC GLM (SAS Institute 1989). Waller-Duncan k-ratio *t* test was used to separate mean differences ( $P = 0.05$ ) among main effects and their interactions.

## Results

**Reproductive Behavior.** *Diapetimorpha introita* females appeared to mate only once while males were capable of multiple mating when offered several receptive females. Virgin females were aggressively pursued by males which exhibited wing-fanning behavior in an apparent attempt to gain acceptance by the female. Copulation generally lasted from 20 to 30 seconds and usually occurred during early-to-mid morning hours. *Diapetimorpha introita* appear to be totally diurnal in habit; searching and/or other locomotory activities ceased when cages were transferred to low light or to darkened conditions.

Female *D. introita* host-searching behavior suggested that a sequence of olfactory, auditory, and physical cues were used to detect and accept a host pupa. In the search mode, females walked about on the soil surface harboring host pupae while rapidly drumming their fully-extended antennae upon the surface. This behavior likely allows the female to detect kairomones associated with the host pupa through chemical-receptor sites located on the antennae segments. It may also allow *D. introita* females to echo locate hollow cavities (pupal cells) in the soil. Echo location of soil-dwelling prey has been reported for other ichneumonids (Henaut and Guerdoux 1982). Upon location of a pupal cell, the antennae were retracted somewhat, forming a convex posture, and the female delineated the site by rapidly drumming with the most distal antennal segment. Upon acceptance, the female would then probe with her ovipositor to determine the presence of an actual tunnel under the cell cap. If acceptable, oviposition would ensue, however, if the tunnel was plugged with soil the site was rejected even when the pupae remained. Females oviposited in host pupal

tunnels even when the pupa was removed without disturbing the cell cap or if the host pupa was replaced with non-host pupa such as corn earworm. Thus, physical factors played a primary role in the final acceptability of a cell.

Virgin *D. introita* females lived about 6 days longer and oviposited 16% fewer eggs than females exposed to males (Table 1). Hatchability of eggs was similar for females from both mating classes. Virgin females produced all male progeny as is typical of the haplo-diploid mechanism of sex determination commonly found in Hymenoptera. However, mated females produced 40% more male (1.4:1) than female progeny.

**Table 1. Longevity and reproductive characteristics of female *D. introita*.**

Mating status	n	Days lived ± SD	No. eggs/ female ± SD	% hatch	Sex ratio m:f
virgin	5	30.2 ± 4.7	68.6 ± 25.1	81.7	1:0
mated	14	23.6 ± 9.6	81.3 ± 25.4	87.3	1.4:1

**Developmental Studies.** Hatchability of *D. introita* eggs exposed to 18 or 33° C was greatly reduced compared to the > 90% eclosion of eggs observed at 22 or 28°C (Table 2). None of the eggs incubated at 13°C hatched. Duration of the egg stage was shortest at the two highest temperatures while those exposed to 18°C required 4.1 days. Although the lower and upper lethal temperature limits for hatching of *D. introita* eggs were not determined, they apparently lie between 13 and 18°C, and above 33° C.

**Table 2. Hatchability and duration of *D. introita* egg stage at five temperatures.**

Temp °C	n	% hatch	$\bar{x}$ days ± SD
13	20	0.0	—
18	20	65.0	4.1 ± 0.10
22	20	90.0	2.3 ± 0.40
28	29	96.7	1.5 ± 0.05
33	20	75.0	1.6 ± 0.30

Typically, eggs oviposited by *D. introita* reared on *S. frugiperda* are elliptical in shape and measure about 1.4 mm long and 0.35 mm diam and are white to opaque in color. Prior to eclosion, the eggs take on a slightly yellowish hue. Interestingly, we observed that egg size appeared to be correlated with size of the female, i. e., females reared on small *S. exigua* pupae oviposited eggs roughly one-third the size of those laid by females which were reared on the 5X larger *H. zea* pupae. We did not pursue the apparent relationship between egg size and host; however, the host upon which an individual female developed did not seem to influence fecundity.

Ovipositing *D. introita* females accepted a wide range of fall armyworm pupal ages (Table 3). However, 4-day-old pupae were preferred over 1, 2, or 6-day-old pupae. It does appear that pupal age is not critical until 6 days when moth development is advanced to the stage (wing pads visible) that *D. introita* larvae could not survive.

**Table 3. Ovipositional response of *D. introita* females to *S. frugiperda* pupae of different ages.**

Pupal age (days)	Avg. no eggs/pupa*	% of total eggs
1	0.82 bc	12.6
2	0.89 bc	13.7
3	1.19 abc	18.3
4	1.69 a	26.0
5	1.25 ab	19.2
6	0.66 c	10.1

\* Means within a column followed by the same letter are not significantly different ( $P > 0.05$ ); Waller-Duncan k-ratio test (SAS Institute 1989).

*Diapetimorpha introita* larval development at three temperatures and on five host pupae is noted in Table 4. At 18°C, *D. introita* larvae developed upon *H. zea* significantly slower than on *S. frugiperda* ( $F = 16.7$ ;  $df = 1,56$ ;  $P < 0.0001$ ). Similarly, *D. introita* larvae developed significantly slower on *H. zea*, and significantly faster on *S. exigua* at 22°C ( $F = 8.2$ ;  $df = 4,163$ ;  $P < 0.0001$ ) and at 28°C ( $F = 41.9$ ;  $df = 4, 184$ ;  $P < 0.0001$ ) than on all other hosts. Development was a function of host size as the slowest development occurred on larger hosts such as corn earworm and fastest on the much smaller beet armyworm pupae. There were significant differences ( $F = 633.9$ ; 4, 410  $df$ ;  $P < .01$ ) between the average pupal weights of each noctuid species tested with *H. zea* > *S. ornithogalli* > *H. virescens* > *S. frugiperda* > *S. exigua* (data not shown).

**Table 4. Influence of host species and temperature on the duration of *D. introita* larval stages.**

Host*	n	18 °C**	$\bar{x}$ No. days ( $\pm$ SD)			
			n	22 °C†	n	28 °C†
CEW	27	16.7 $\pm$ 2.1 a	38	9.3 $\pm$ 1.6 a	38	5.7 $\pm$ 0.9 a
YSA			31	8.6 $\pm$ 0.9 b	7	5.4 $\pm$ 0.5 b
TBW			20	8.4 $\pm$ 1.1 b	38	5.2 $\pm$ 0.7 b
FAW	37	13.8 $\pm$ 3.2 b	55	8.4 $\pm$ 1.3 b	72	5.4 $\pm$ 0.8 b
BAW			36	7.6 $\pm$ 1.1 c	47	3.9 $\pm$ 0.5 c

\* Key to species: CEW = *H. zea*, YSA = *S. ornithigalli*, TBW = *H. virescens*, FAW = *S. frugiperda*, and BAW = *S. exigua*.

\*\* Means within a column followed by the same letter are not significantly different ( $F = 16.71$ ; 1,56 df;  $P > 0.05$  (SAS Institute, 1989).

† Means within a column followed by the same letter are not significantly different ( $P > 0.05$ ); Waller-Duncan k-ratio test (SAS Institute 1989).

Although temperature and host size influenced *D. introita* larval growth rates, these factors apparently did not affect the number of instars attained during *D. introita* larval development. Thus, growth rates were inversely proportional to the amount of food available. Observations revealed that larvae generally underwent four molts although 2.5 and 11.8%, respectively, of the larvae placed on *S. exigua* and *S. frugiperda* pupae molted five times and thus attained the sixth instar. None of the *D. introita* larvae placed on *H. zea* pupae were observed to molt more than four times prior to spinning a cocoon. The larvae seemingly possess the capability to determine the quantity of food available and then regulate growth accordingly. Indeed, 95% of the *D. introita* larvae placed upon *S. exigua* pupae advanced to the fourth instar on day 2 compared with 79.4 and 0.0%, respectively, of those placed upon *S. frugiperda* or *H. zea* pupae. On day 2, only 41.7% of the *D. introita* larvae on *H. zea* pupae had attained the third instar.

Table 5 summarizes pupation success, survival to adult stage, and diapause incidence of *D. introita* reared on five noctuid hosts and at two or three temperature regimes. In general, survival was greatest when *D. introita* developed at the higher temperatures and when *S. frugiperda* or *S. exigua* served as the hosts. Temperatures of 22°C or less resulted in > 90% diapause. However, the percentage of diapausing *D. introita* ranged from 10 (*S. frugiperda*) to 53.8% (*S. ornithigalli*) even when reared at 28°C. Critical limits of temperatures necessary to induce diapause or larval mortality were not determined. It should be noted that *D. introita* were also reared to the adult stage on southern armyworm, *S. eridania*, pupae (data not shown).

**Table 5. Influence of host and temperature upon survival rates and the incidence of diapause in *D. introita*.**

Host*	°C	n	% pupation	% emergence	% diapause
CEW	18	60	45.0	92.6	100.0
	22	60	61.7	89.2	94.6
	28	60	81.7	87.7	35.7
YSA	22	40	77.5	58.1	100.0
	28	60	21.7	92.3	53.8
TBW	22	60	33.3	80.0	100.0
	28	60	70.0	92.8	16.7
FAW	18	60	61.7	91.9	100.0
	22	60	91.7	87.2	96.5
	28	100	80.0	100.0	10.0
BAW	22	60	60.0	80.5	91.4
	28	60	93.3	96.4	19.6

\* Key to species: CEW = *H. zea*, YSA = *S. ornithogalli*, TBW = *H. virescens*, FAW = *S. frugiperda*, and BAW = *S. exigua*.

There was a significant two-way interaction between host and temperature on the duration of *D. introita* pupal stages ( $F = 3.14$ ; 5, 365 df;  $P = 0.0087$ ). Irrespective of host, *D. introita* reared at 18 or 22°C entered diapause (Table 6). Although *D. introita* reared at 18°C on *H. zea* had significantly longer pupal stage durations than those reared on *S. frugiperda*, there was no influence due to host in pupal weights of *D. introita* reared at 22°C. However, at 28°C, parasitoids reared on *H. zea* developed significantly slower ( $F = 31.61$ ; 4, 81 df;  $P = 0.0001$ ) than those reared on other hosts. Similar to results derived from *D. introita* larval development studies, *D. introita* adults reared on *S. exigua* emerged 4 days sooner than those reared on *H. zea*. Thus, host size affected not only the rate of larval development but the duration of pupal stages as well when they were exposed to non-diapause inducing temperatures.

When *D. introita* entered a diapause state, the duration of pupal stage was highly variable. *Diapetimorpha introita* diapauses as a last-instar larva inside the silken cocoon. One female that had been reared on fall armyworm pupae held at 22°C remained in diapause for 3 years. Other cocoons of a similar age were dissected and live larvae were found which eventually died due to probable desiccation. When diapausing *D. introita* were exposed to temperatures of about 0°C for 2-3 wks, development was initiated and most of the wasps emerged within 4-6 wks.

**Table 6. Influence of host species and temperature on the duration of *D. introita* pupal stages.**

Host*	n	18 °C**	$\bar{x}$ No. days ( $\pm$ SD)			
			n	22 °C	n	28 °C†
CEW	25	164.2 $\pm$ 112.9 a	34	148.4 $\pm$ 100.3	32	13.8 $\pm$ 2.7 a
YSA			18	125.4 $\pm$ 77.0	6	12.0 $\pm$ 0.9 b
TBW			15	172.5 $\pm$ 78.5	35	10.7 $\pm$ 1.6 c
FAW	34	102.3 $\pm$ 55.7 b	49	162.2 $\pm$ 95.2	72	11.4 $\pm$ 1.5 bc
BAW			28	129.8 $\pm$ 85.9	45	9.8 $\pm$ 0.7 d

\* Key to species: CEW = *H. zea*, YSA = *S. ornithigalli*, TBW = *H. virescens*, FAW = *S. frugiperda*, and BAW = *S. exigua*.

\*\* Means within a column followed by the same letter are not significantly different ( $F = 6.48$ ; 1,53 df;  $P > 0.05$ ).

† Means within a column followed by the same letter are not significantly different ( $P > 0.05$ ); Waller-Duncan k-ratio test (SAS Institute 1989).

Female *D. introita* were generally 2 to 3X larger than males, however, adult weights of male and female *D. introita* were significantly influenced by temperature ( $F = 14.05$ ; 2, 356 df;  $P = 0.0001$ ) and host species ( $F = 25.32$ ; 4, 356 df;  $P = 0.0001$ ). Male and female *D. introita* reared at 28°C were significantly heavier ( $F = 9.2$ ; 2,183 df;  $P < 0.0001$  and  $F = 14.91$ ; 2,187 df;  $P < 0.0001$ , respectively) than cohorts reared at lower temperatures (Table 7). As stated above, host species, i.e. host pupal size, had a profound effect upon resultant adult *D. introita* weights. Weights of *D. introita* males and females, respectively, ranged from 19.7 and 30.8 mg (*S. exigua*) to 32.5 and 62.6 mg (*H. zea*). Regression analysis showed that there was a significant linear correlation between the host species tested, i.e., host pupal size, and adult *D. introita* weight except for wasps reared upon *S. ornithigalli* ( $P = 0.2818$ ). Analysis of covariance (PROC GLM) revealed that slopes of male and female *D. introita* weights were common on all hosts except when they were reared upon *H. virescens* ( $P = 0.0412$ ). Therefore, host adult *D. introita* weight may be a function of both size and species of the host pupa.

Fig. 1 illustrates the relationship of host weights for male and female *D. introita* reared on pooled and the individual host species. The effect of reduced host weight on resultant *D. introita* weight was most apparent for those reared on *S. exigua*. The relationship between host size and female *D. introita* size and fecundity was not fully investigated. However, it was noted that small *D. introita* females (reared from *S. exigua*) appeared to be as fecund as those reared from larger hosts such as *H. zea*.

**Table 7. Influence of temperature upon resultant adult *D. introita* weight.**

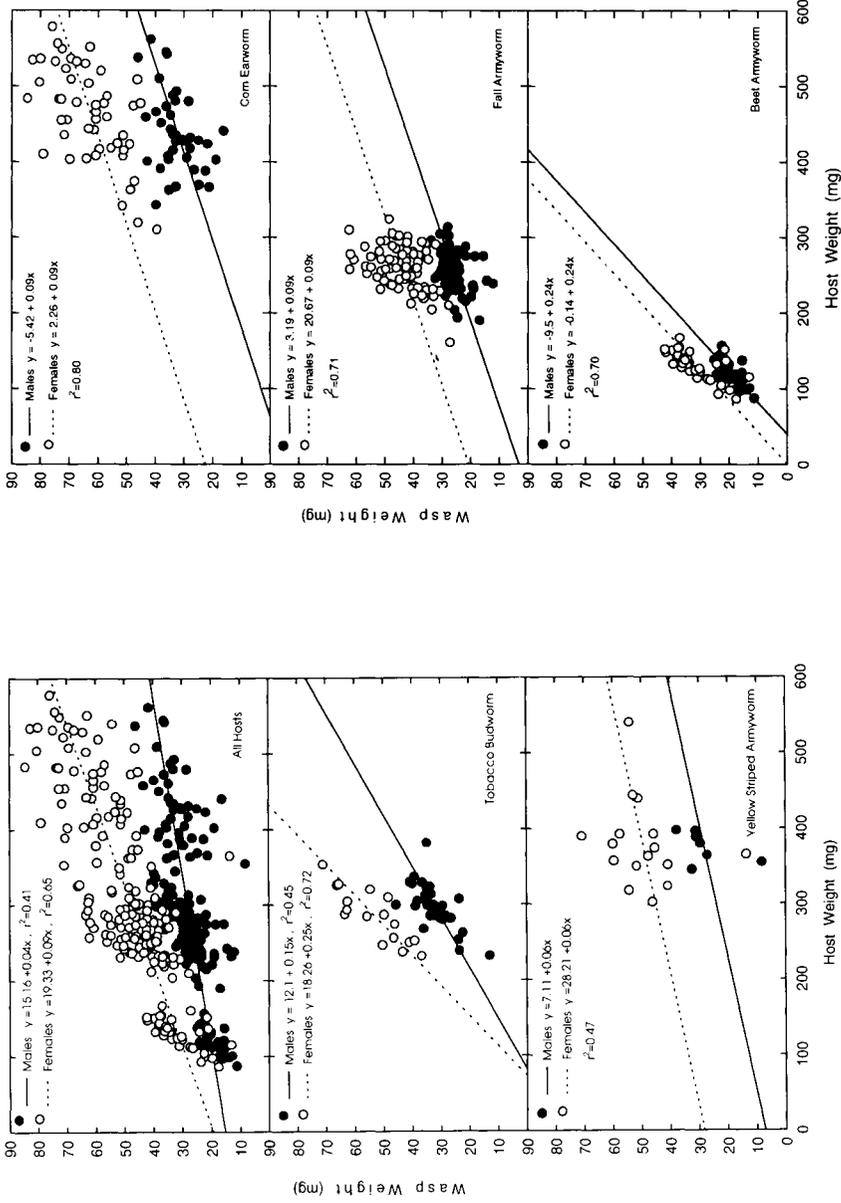
Temp $\pm$ 1°C	♂		♀	
	n	$\bar{x} \pm SD^*$ (mg)	n	$x \pm SD^*$ (mg)
18	40	24.0 $\pm$ 6.8 b	48	42.1 $\pm$ 9.1 b
22	59	25.5 $\pm$ 7.9 b	75	43.1 $\pm$ 12.0 b
28	100	29.3 $\pm$ 6.4 a	90	52.4 $\pm$ 15.6 a

\* Means within a column followed by the same letter are not significantly different ( $P > 0.05$ ); Waller-Duncan k-ratio test (SAS Institute 1989).

### Discussion

The results of this study indicate that *D. introita* can be successfully reared in the laboratory on a wide variety of noctuid pupae with the possible exception of *S. ornithogalli*. Because *S. frugiperda* and *S. exigua* larvae exhibit much less cannibalism and appear to be the more preferred hosts than *H. zea* or *H. virescens*, they would be likely candidates for rearing *D. introita in vivo* with the least expense. However, the cannibalistic behavior of wasp larvae, coupled with the necessity of rearing hosts and the low fecundity of *D. introita* females, could hamper efforts to rear this species on a large scale for biocontrol studies of fall armyworm. The development of a satisfactory artificial diet allowing *in vitro* rearing would greatly facilitate greater production of *D. introita* for evaluation in field release situations. Successful development of artificial diets for ectoparasitoids such as *D. introita* should be less complicated because the living host substrate does not have to be duplicated. Indeed, *D. introita* larvae kill their host pupae within 2 days (Pair and Gross 1984).

More intensive field studies are clearly needed to accurately determine the potential role of *D. introita* as a biocontrol agent of *Spodoptera*. Its greatest value as a control agent, however, may be as components of a multi-faceted approach involving the augmentative releases of r-strategist larval parasitoids such as *Archytas marmoratus* (Townsend) (Gross and Pair 1986). As such, their greatest potential may result from augmentative releases against low density populations of *Spodoptera* spp. in overwintering areas. The lower peninsula of south Florida is known to harbor overwintering populations of fall, beet, and southern armyworm. However, the success of such an endeavor would likely be limited unless sufficient numbers of *D. introita* could be released. Indeed, Knipling (1977) stated that until the number of parasitoids produced by artificial means are increased 10 to 50 times over the population that normally occurred, the results of such releases would largely remain unknown.



**Fig. 1.** Adult weights of male and female *D. introita* as a function of overall host size, and of host size among host species.

### Acknowledgments

The author gratefully acknowledges R. Caldwell, Biological Technician, for his assistance in rearing and data collection, and of R. Layton, Computer Program Analyst-SAA, and T. Popham, Biometrician-SPA for their statistical services.

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