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Study of the postembryonic development of *Idaea inquinata* under different abiotic factors

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Abstract

We investigated the biology of *Idaea inquinata* (Scopoli) (Lepidoptera Geometridae), the rusty wave moth, to determine the number and duration of larval instars and the duration of the pupal stage. The study was conducted at 21, 26, 29, and 34 ± 1 °C; for each temperature tests were conducted at 35 and 70 ± 5% relative humidity (RH), with a photoperiod of 16:8 (light:dark).

At 35% RH, five larval instars were observed at 26, 29, and 34 °C, whereas eight instars were found at 21 °C. At 21 °C and 70% RH, only one larva pupated after the fifth instar, two completed the sixth instar and one reached the seventh instar. At 21 °C and 35% RH an increase of mortality and in the number of larval instars was observed; the few larvae that reached the tenth instar did not survive. The shortest larval developmental periods were observed at 26 and 29 °C at 70% RH and were 31.9 ± 2.26 and 30.6 ± 3.12 days respectively. The longest developmental period was recorded at 21 °C and 35% RH, and was 172.5 ± 16.26 days. The pupal stage was longer at 21 °C at 35 and 70% RH, and lasted 22.5 ± 2.12 and 22 ± 3.65 days respectively. In all other conditions, the pupal periods lasted from 9 to 10 days.

The highest adult emergence was observed at 26 and 29 °C at 70% RH and corresponded to 90% and 83% respectively. *I. inquinata* did not complete development at 34 °C and 35% RH. At 34 °C and 70% RH, only 13% of the specimens reached the adult stage. At 26 °C and 35% RH there was 66% adult emergence but this declined to 30% at 29°C and 35% RH. Percentage of adult emergence at 21 °C and 35% and 70% RH were 6 and 13% respectively.

Considering the reduction in adult emergence at 21 °C, we recommend that a temperature of 18° C is maintained in air conditioned warehouses for the storage of dried medicinal plants to prevent *I. inquinata* infestations.

Key words: rusty wave moth, larval instars, temperature, relative humidity.

Introduction

The genus *Idaea* Treitschke includes species that feed on grasses, withered parts of plants and dried plants used in herbal medicine, particularly *Foeniculum*, *Silybym*, *Crataegus*, *Angelica, Matricaria*, *Malva* (Martinez and Coutin, 1985; Naves, 1995; Locatelli *et al*., 2005; Hausmann *et al.*, 2008) References on this genus are mainly on systematics and few concern the biology of species within this genus (Tusnadi and Meszaros, 1988; Vasilenko 1994; Naves, 1995; Hausmann *et al*., 2008).

The rusty wave moth, *Idaea inquinata* (Scopoli) (Lepidoptera Geometridae), develops on hay and on several dried plants (Candura, 1931a; 1931b; Tempel, 1941; Kratochvil, 1948; Locatelli *et al*., 2005), and can spread with the trade of dried aromatic plants or with vegetable packaging (Naves, 1995). It was recorded in warehouses of Southern Italy (Candura 1931a; 1931b) and the development from egg to adult lasts from 30 to 333 days, depending on temperature and diet (Candura 1931b; Limonta *et al*., 2010a). Larvae show negative phototropism and live in the lowest layer of the substrate feeding on apical leaves and flowers that are characterized by a high nutritional value. In a previous research on the development of *I. inquinata*, at 26 °C and 50 and 70% RH, and 29 °C and 50 and 70% RH, at different photoperiods, a lower mortality and a shorter development period were observed with the 0:24 (light:dark) photoperiod (Limonta *et al*., 2010a).

The rusty wave moth can be an important pest in warehouses where dried plants and cereal products are stored together, because it can also develop on bran, maize flour and wheat kernels (Locatelli *et al.*, 2005). Infestations of *I. inquinata* can be easily underestimated, because larvae are hidden in the substrate and adults have limited activity. Optimal environmental conditions, namely darkness, high temperatures and relative humidities facilitate rapid population growth of this pest and its potential damage to stored products (Limonta *et al*., 2010a; 2010b; 2010c).

In the present study, the number and duration of larval instars and of the duration of the pupal stage were investigated at four different temperatures and two relative humidities.

Materials and methods

Rearing

I. inquinata was collected on *Hypericum perforatum* L. in a warehouse in Milano, and has been reared on an artificial diet in a thermostatic chamber at 26 ± 1 °C, 70 ± 5% RH, and a photoperiod of 16:8 (light:dark) for 6 years. The artificial diet was prepared with 62 g bran, 8 g corn flour, 7 g wheat flour, 4 g wheat germ, 3 g dried yeast, 9 g glycerine, 7 g honey (Stampini and Locatelli, 2007).

Eggs were obtained by placing new emerged adults in a glass jar closed with tulle, turned upside down in a Petri dish with filter paper. Eggs were collected daily and incubated at 26 ± 1 °C and 70 ± 5% RH. Newly emerged larvae were transferred for the tests with a fine paint brush.

Table 1. Mean developmental period (±S.E.) of the larval instars of *I. inquinata* reared at 21, 26, 29, and 34 ± 1 °C, at 35 and 70 ± 5% RH, that developed to adult. Brackets indicate minimum and maximum values.

T (°C)	RH (%)	No.	Mean developmental period (days) ±SD Larval instars I	II	III	IV	V	VI	VII	VIII
21	35	2	15.5 ± 1.50ab (14-17)	18 ± 4.00a (14-22)	15.5 ± 0.50a (15-16)	17.5 ± 1.50a (16-19)	18.5 ± 1.50a (17-20)	21.5 ± 2.50 (19-24)	25 ± 5.00 (20-30)	40 ± 5.00 (35-45)
	70	4*	14.0 ± 1.08ab (12-17)	15.5 ± 0.96a 14-18	14.7 ± 0.85a 13-17	17.5 ± 1.84a 14-22	19 ± 1.91a 16-24	15.5 ± 8.50 7-24	24	
26	35	20	15.7 ± 0.87a (7-20)	8.0 ± 0.70bc (4-18)	8.2 ± 0.64c (3-15)	11.0 ± 1.55b (5-32)	11.1 ± 1.00b (5-25)			
	70	27	4.6 ± 0.11d (4-6)	6.5 ± 0.45bc (3-14)	6.5 ± 0.44cd (2-10)	9.9 ± 0.63bc (2-16)	4.2 ± 0.47c (2-12)			
29	35	9	13.8 ± 0.83ab (12-18)	6.1 ± 0.54bc (3-9)	5.7 ± 0.37cd (4-7)	5.7 ± 0.60cd (2-8)	12.9 ± 2.29b (4-25)			
	70	25	9.7 ± 0.23c (8-13)	5.6 ± 0.35c (3-11)	4.8 ± 0.28d (2-8)	4.6 ± 0.36d (2-8)	5.9 ± 0.47c (1-11)			
34	70	4	12.7 ± 1.25b (9-14)	9.0 ± 1.08b (7-12)	11.5 ± 0.87b (10-13)	9.0 ± 1.22bcd (6-11)	19.2 ± 4.85a (8-30)			

* One larva pupated after the fourth moult, two after the fifth moult, and one after the sixth moult.

One-way Anova: first instar $F_{6,84} = 58.527$ $P < 0.001$; second instar $F_{6,84} = 17.817$ $P < 0.001$; third instar $F_{6,84} = 24.762$ $P < 0.001$; fourth instar $F_{6,84} = 11.365$ $P < 0.001$; fifth instar $F_{6,84} = 20.971$ $P < 0.001$. Means followed by different letters in the same column are significantly different according to Duncan's multiple range test.

Table 2. Number of larvae of *I. inquinata* that did not complete development and mean developmental period (S.E.) of each larval instar at 21, 26, 29, and 34 ± 1 °C, at 35 and 70 ± 5% RH.

T (°C)	RH (%)	Mean developmental period (days) ±SD Larval instars No.	I	No.	II	No.	III	No.	IV	No.	V	No.	VI	No.	VII	No.	VIII	No.	IX
21	35	13	20.8 (0.69)	9	18.1 (1.82)	8	15.0 (1.14)	7	21.1 (1.18)	6	22.8 (5.52)	6	25.5 (5.54)	4	42.5 (7.03)	3	22.3 (1.33)	2	19.5 (5.50)
	70	7	15.8 (0.74)	5	17.8 (0.49)	4	15.0 (1.35)	4	20.2 (1.93)	4	18.0 (1.78)	4	30.5 (2.25)	3	22.7 (7.53)				
26	35	10	13.6 (1.49)	2	7.5 (0.50)	1	8												
	70	3	4.7 (0.33)																
29	35	21	9.5 (0.96)	6	4.5 (1.26)	4	7.0 (1.47)	3	0.3 (0.3)										
	70	5	9.0 (0.32)	1	7	1	4	1	11										
34	35	30	13.1 (0.91)	29	10.2 (1.00)	24	8.2 (0.69)	12	7.2 (0.64)	4	3.7 (1.18)								
	70	6	12.2 (0.62)	20	9.7 (0.79)	14	10.8 (0.99)	1	14										

Development of *Idaea inquinata*

In each test the development of 30 newly emerged larvae was monitored. First instar larvae that were 12-24 hours old were individually placed in glass jars (diameter 3.8 cm; height 2.5 cm). The jars contained 0.1 g of artificial diet that was replenished as needed and were closed with gauze nets to allow gaseous exchange. Individuals were checked daily and the number of moults and mortality were recorded.

Tests were carried out at four temperatures, namely, 21, 26, 29, and 34 ± 1 °C. For each temperature tests were conducted at 35 and 70 ± 5% RH, with a photoperiod of 16:8 (light:dark).

Data were analysed using one-way analysis of variance (ANOVA) and Duncan's multiple range test (SPSS 17.0).

Results

Developmental times of larval instars that developed to adult were reported (table 1). At temperatures of 26, 29, and 34 °C and 35 and 70% RH, *I. inquinata* had five larval instars. However, it had eight and six instars at 21 °C and 35 and 70% RH respectively. At 21 °C and 70% RH eight instars were completed before pupation and only one larva pupated after the fifth instar, two larvae completed the sixth, and one pupated after the seventh instar. With the exception of 21 °C, the first larval instar period was longer at 35% RH than at 70% RH, whereas in the second to the fifth larval instars the periods were statistically equal at 35 and 70% RH. At 21 °C and 35% RH, developmental times of the sixth, seventh and eighth larval instars (21.5 ± 3.53; 25 ± 7.07; 40 ± 7.07 respectively) were relatively longer. The developmental period of the different instars was significantly longer at 26 and 29 °C at 35% RH. Only four larvae developed to adult at 34 °C and 70% RH, whereas none developed to adult at 35% RH.

Developmental times of larval instars that did not develop to adult were reported (table 2). A gradual decrease of the specimens and an increase of the larval instars were observed at 21 °C and 35% RH, where two larvae reached the tenth instar but did not survive. The number of larval instar increased and three specimens reached the eighth instar but did not survive even at 21 °C and 70% RH. Larvae, that do not complete development died before the fourth moult at 26 °C and 35% RH, but died before the second moult at 70% RH. Mortality increased with number of moults and larval mortality happened before the fourth moult at 29 and 34 °C, and 35 and 70% RH.

Developmental times of larvae and pupae, and number of individuals that developed to adult at the different temperatures and relative humidity values are reported (table 3). The shortest larval developmental periods were observed at 26 and 29 °C at 70% RH, 31.9 ± 2.26 and were 30.6 ± 3.12 days, respectively, while the longest developmental period was 172.5 ± 16.26 days, recorded at 21 °C and 35% RH. Pupal developmental periods were longer at 21 °C at both 35 and 70% RH, and were 22.5 ± 2.12 days and 22 ± 3.65 days respectively. Pupal developmental periods were similar in all the other conditions and lasted from 9 to 10 days. The shortest postembryonic periods were observed at 26 and 29 °C, and were 41.3 ± 1.9 and 39.5 ± 3.2 days, respectively, at 70% RH. The longest postembryonic period was recorded at 21 °C and 35% RH, and was 195 ± 18.34 days. Adult emergence was higher at 70% RH for all temperatures tested. The highest numbers of emerged adults were observed at 26 and 29 °C and 70% RH and corresponded to 90% and 83% adult emergence, respectively. No individuals completed development at 34 °C and 35% RH. At 26 °C and 35% RH, 66.6% of speci-

Table 3. Number of emerged adults, mean developmental period (±S.E.) of larvae and pupae of *I. inquinata* reared at 21, 26, 29, and 34 ± 1 °C, at 35 and 70 ± 5% RH. Brackets indicate minimum and maximum values.

T (°C)	RH (%)	Emerged adults	Developmental period		
			Larvae	Pupae	Larva and pupa
21	35	2	172.5 ± 11.50a (161-184)	22.5 ± 1.50a (21-24)	195.0 ± 13.00a (182-208)
	70	4	90.5 ± 17.56b (56-139)	22 ± 1.82a (18-26)	112.5 ± 19.25b (74-165)
26	35	20	54.2 ± 11.14c (40-62)	10.6 ± 0.70b (7-17)	64.8 ± 1.18c (48-70)
	70	27	31.9 ± 0.43e (28-36)	9.4 ± 0.29b (7-12)	41.3 ± 0.36e (38-47)
29	35	9	44.1 ± 2.43d (35-58)	9.5 ± 0.80b (7-13)	53.7 ± 2.54d (42-65)
	70	25	30.6 ± 0.62e (27-35)	8.9 ± 0.77b (5-17)	39.5 ± 0.64e (34-44)
34	35	0	-	-	-
	70	4	61.5 ± 5.04c (50-73)	9.7 ± 3.82b (3-19)	71.2 ± 1.93c (69-77)

One-way Anova: Larvae $F_{6,84} = 134.67$ $P < 0.001$; Pupae $F_{6,84} = 14.747$ $P < 0.001$; postembryonic period $F_{6,84} = 153.279$ $P < 0.001$. Means followed by different letters in the same column are significantly different according to Duncan's multiple range test.

mens emerged as adults, whereas 30% emerged as adults at 29 °C and 35% RH. At 21 °C and 35% RH, 6% of the individuals emerged as adults, whereas 13% emerged as adults at 21 °C and 70% RH.

Conclusions

Increase in the number of larval instars can be caused by different factors, such as extreme temperatures, short photoperiod, poor food quality, and low relative humidity and many studies concern intraspecific variability in insects (Chippendale and Yin, 1973; Kfir, 1991; Tammaru, 1998; Berthiaume *et al*., 2007; Esperck *et al.*, 2007). In our research, at 21 °C and 34 °C an increase in the number of the larval instars of *I. inquinata* was observed).

At temperatures of 26, 29 and 34 °C and 35 and 70% RH *I. inquinata* had five larval instars while Sannino and Espinosa (2002) described four larval instars in *Idaea seriata* (Schrank) but they did not mentioned temperature.

At 34 °C and 70% RH only the 13% of the individuals of *I. inquinata* emerged as adults, whereas at 35% RH only the 24% completed four instars but died before the fourth moult. In a previous study it was observed that, at 36 °C, less than the 9% of eggs hatched at 35% RH, while at 70% RH 16% of eggs hatched, but in both cases the larvae died before the first moult (Limonta *et al.*, 2010c). At 21 °C six instars at 70% RH and eight instars at 35% RH were observed. Also Berndt *et al.* (2004) observed six larval instars at 23 °C in *Pseudocoremia suavis* Butler, and in *Corcyra cephalonica* (Stainton), a moth pest of stored products, the number of larval instars increased when relative humidity was lowered, even at 28 °C (Russell *et al*., 1980).

Larval development of *I. inquinata* was completed in about 30 days at 26 and 29 °C and 70% RH, while at 21 °C and 70% RH it lasted 90 days, and at 35% RH it lasted 172 days. Developmental time of stored products pyralid moths are shorter (Allotey and Azalekor, 2000; Brindley, 1930; Burges and Haskins, 1965; Siddiqui and Barlow, 1973). For example *Plodia interpunctella* (Huebner), at 25 °C and 70% RH, completed five larval instars in 24 days (Amaral Filho and Praca Neto, 1984) and in *Cadra calidella* (Guenee) postembrional development lasted 52 days at 20 °C (Alrubeai, 1987).

At 35 and 70% RH, the pupal duration of *I. inquinata* lasted 8 to 10 days at 26, 29 and 34 °C, while at 21 °C it lasted 22 days. Hausmann *et al.* (2008) observed that in *Idaea omari* Hausmann et Blasius the pupal development time was 11 days at 25 °C "or slightly longer at 21 °C". An increase in the pupal developmental period with low temperature was also observed in *Cadra cautella* (Walker) where it lasted 16 days at 20 °C and 70% RH, and decreased to 6 days at 35 °C and 70% RH (Burges and Haskins, 1965). In *P. interpunctella* it was 12.46 days at 25 °C and 70% RH (Amaral Filho and Habib, 1991), 12.7 days at 22 °C and increased to 28 days at 18 °C (Savov, 1973).

In the present study the optimal conditions for the development of *I. inquinata* were 26 and 29 °C and 70% RH. At this condition, mortality is low and postembryonic development is short. Incubation period was also short under these optimal conditions with eggs hatching in 6-9 days (Limonta *et al*., 2010b; 2010c). The lower and upper embryonic developmental thresholds at 35% RH were 13 and 38 °C and these values at 70% RH were 15 and 36 °C (Limonta *et al*., 2010c).

The postembryonic development at 26 °C and 35% RH lasted 65 days, and it was similar to the findings of Hausmann *et al.* (2008) in *I. omari*. At 21 °C the postembryonic development in *I. inquinata* was considerably prolonged. In fact at 21 °C and 70% RH it lasted 112 days whereas it lasted 195 days at 35% RH. Mortalities at 35 and 70% RH were 87% and 94% respectively. At the same temperature *I. omari* completed development in about two months (Hausmann *et al*., 2008).

Considering the reduction in adult emergence at 21 °C, and the long developmental period of the combined immature stages of more than six month at 35% RH, we recommend that a temperature of 18 °C is maintained in air conditioned warehouses to prevent *I. inquinata* infestations.

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