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S. SAVOLDELLI

Survival of *Plodia interpunctella* (Hübner), *Cadra cautella* (Walker), *Ephestia kuehniella* Zeller, *Corcyra cephalonica* (Stainton) (Lepidoptera Pyralidae) larvae to starvation

Abstract - The survival skills of *Plodia interpunctella*, *Cadra cautella*, *Ephestia kuehniella*, *Corcyra cephalonica* larvae have been investigated in laboratory trials. Tests were carried out at three different environmental conditions. *C. cephalonica* larvae were the most resistant to starvation. Second instar larvae anticipated mould; third instar larvae of the considered species anticipated metamorphosis and in few cases they became adults.

Riassunto - *Sopravvivenza al digiuno delle larve di* Plodia interpunctella *(Hübner),* Cadra cautella *(Walker),* Ephestia kuehniella *Zeller,* Corcyra cephalonica *(Stainton) (Lepidoptera Pyralidae).*

La capacità di sopravvivenza delle larve di I, II, III età di *Plodia interpunctella*, *Cadra cautella*, *Ephestia kuehniella*, *Corcyra cephalonica* in condizioni di digiuno è stata valutata in laboratorio, a tre diverse condizioni ambientali. Le più resistenti al digiuno sono risultate le larve di *C. cephalonica*. Le larve di II età delle 4 specie hanno anticipato la muta, mentre quelle di III età hanno anticipato l'impupamento, con sviluppo di alcuni adulti.

Key words: Lepidoptera Pyralidae, larval survival, larval behaviour, starvation, moths of stored products.

INTRODUCTION

Studies about food requirements of insects are numerous and have been developing from a long time. They allowed to clarify the influence of every nourishment on their life and to identify the most adapted food for rearing. Like every living creature, insects require a balanced diet (Lecato, 1977; Rodriguez-Menendez *et al.*, 1988), because the lack or the excess of some nutrients can interfere with their development, can cause death or can make them more susceptible to micro-organisms.

Researches about larval behaviour during dietary stress or abstinence of food are few. In Italy, Candura (1928) made the first observations on the survival of *Ephestia kuehniella* and *Plodia interpunctella* larvae to starvation. He observed that the more temperature was low, the more they survived to starvation. In another paper Candura (1930) reported that *E. kuehniella* and *P. interpunctella* larvae were able to resist to starvation during autumn and winter: he probably referred to "diapause" that is well known for Lepidoptera larvae (Tzanakakis, 1959; Morére & Le Berre, 1967; Bell *et al.*, 1979, Bell *et al.*, 1983).

A few data can be found in a paper of Cline (1978), who relegated "large and small larvae" of *P. interpunctella*, *Cadra cautella* and *Corcyra cephalonica* in presence and absence of food. He noticed that small larvae of *P. interpunctella* and *C. cautella* were able to survive without food for 4 days, those of *Corcyra cephalonica* for 6-8 days, but he didn't specify neither larval instar nor temperature. He also observed that larvae of all species generally showed a much greater propensity to penetrate flexible packaging materials when confined without food.

Further news about starvation of larvae are reported by other Authors. Imura (1982) reported that a starvation period of more than 3 days could increase significantly pigmentation of *E. kuehniella* larvae. Starvation of mid and late final-instar larvae of *C. cephalonica* for 24, 48 or 72 hrs prevented the synthesis of the two most important protein of haemolymph (LHP49 and LHP46), generally produced during the penultimate and last larval instars (Ismail & Dutta-Gupta 1988).

Some Authors have studied the relationship between a balanced diet and the incidence of micro-organisms diseases, but the results disagreed. Boots & Begon (1994) report that diet quality had no effect on the individual susceptibility of *P. interpunctella* larvae to a viral pathogen; on the contrary Benz (1987) and Mc Vean *et al.* (2002) support the hypothesis that diet quality affect the susceptibility of insects to virus infections.

Among stored product pests, larvae or adults of Coleoptera are able to resist for several months without food (Stein, 1986). For Lepidoptera there are not data about survival skill of larvae at fixed environmental conditions; so the aim of this paper is to fill up the lack of data on Pyralid moths of stored products. These data are an essential presupposition to evaluate the possibility of infestation of stored products by larvae of moths.

MATERIALS AND METHODS

Larvae of *Plodia interpunctella* (Hübner), *Cadra cautella* (Walker), *Ephestia kuehniella* Zeller, *Corcyra cephalonica* (Stainton), used in the tests, were obtained from Institute of Agricultural Entomology, University of Milan. They are reared in a climatic room, at 27±1°C, 70±5% RH and light/dark 16:8 hrs, on an artificial diet composed of bran, corn meal, wheat meal, glycerine, wheat germ, honey and yeast (Locatelli & Limonta, 2004).

Fifty larvae of first, second and third instar of each species have been isolated.

Third-instar larvae of *P. interpunctella*, *C. cautella*, *E. kuehniella* have been distinguished in males and females, thanks to the presence of a dark spot on male's IX-abdominal tergum.

Every larva was put in Petri's dishes (Ø: 5,5 cm), without food. Tests were carried out in three different environmental conditions:
– (18 ± 1)°C and (65 ± 6)% RH;
– (24 ± 1)°C and (63 ± 7)% RH;
– (28 ± 1)°C and (70 ± 5)% RH.

The behaviour of first instar larvae was observed every two hrs, those of second instar larvae three time a day, those of third instar daily, untill death or birth of the adult.

Results were submitted to ANOVA and Duncan's multiple range test ($P<0.05$). The tests were performed using the SPSS for Windows version 12.0.

RESULTS

First instar larvae. Results about first instar larvae are shown in Table 1. It is evident that *C. cephalonica* larvae are able to survive without food more time if compared to other species, in particular if compared to *P. interpunctella* larvae, which are the less resistant. In fact, at 28°C they survive not even 24 hrs. It was noticed that *P. interpunctella* larvae were very dynamic and they incessantly moved, untill death. On the contrary, *E. kuehniella* and *C. cephalonica* larvae moved little or nothing and it was necessary to spur them with a tweezers, to verify if they were alive or dead.

Table 1 - Survival time (hrs) of first instar larvae, confined without food, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05)

Temperature (°C)	*P. interpunctella* (hrs ± S.D.)	*C. cautella* (hrs ± S.D.)	*E. kuehniella* (hrs ± S.D.)	*C. cephalonica* (hrs ± S.D.)
18	78.72 ± 18.72 a	79.68 ± 16.32 ab	81.12 ± 16.80 ab	86.40 ± 13.68 b
24	39.84 ± 7.44 a	37.20 ± 9.12 a	39.60 ± 8.16 a	44.40 ± 7.68 b
28	21.84 ± 3.84 a	25.68 ± 6.72 b	27.36 ± 9.36 bc	28.80 ± 3.84 c

Table 2 - Survival time (hrs) of second instar larvae, confined without food, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05).

Temperature (°C)	*P. interpunctella* (hrs ± S.D.)	*C. cautella* (hrs ± S.D.)	*E. kuehniella* (hrs ± S.D.)	*C. cephalonica* (hrs ± S.D.)
18	139.20 ± 31.44 a	160.32 ± 17.76 b	161.28 ± 19.44 b	176.64 ± 22.56 c
24	90.00 ± 20.16 ab	84.96 ± 22.80 a	95.04 ± 18.00 b	115.20 ± 16.32 c
28	60.48 ± 27.60 a	63.84 ± 20.40 ab	71.52 ± 14.88 b	95.04 ± 18.24 c

Second instar larvae. Results about second instar larvae are shown in Table 2. Also second instar larvae of *C. cephalonica* survive to starvation more than other species. As one would expect, all second instar larvae are able to survive longer than first instar larvae of some species. At 24°C, in the first 48-72 hrs from the beginning of starvation, it was observed that second instar larvae of all species carried out a moult in advance.

Third instar larvae. Tables 3-8 refer to results of 50 third instar larvae (25 males and 25 females) confined without food. Some larvae dead of hunger (Tables 3, 6), others have changed into pupae but they didn't become adult (Tables 4, 7), others

Table 3 - Number of third instar larvae dead for starvation and survival time (days), at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05).

	Plodia interpunctella		*Cadra cautella*		*Ephestia kuehniella*		*Corcyra cephalonica*	
T (°C)	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.
18	37	12.03±3.14 ab	36	11.92±1.96 ab	29	13.07±2.10 b	39	17.79±5.88 c
24	23	4.46±1.34 a	32	4.84±1.35 ab	30	5.51±1.27 ab	24	10.5±2.55 c
28	3	4.33±0.58 a	25	5.48±1.36 ab	15	5.07±0.80 ab	16	6.94±1.84 b

Table 4 - Number of third instar larvae change into pupa and mean time in days, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05).

	Plodia interpunctella		*Cadra cautella*		*Ephestia kuehniella*		*Corcyra cephalonica*	
T (°C)	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.
18	10	12.40±2.07 a	14	13.64±1.60 a	19	13.42±2.19 a	11	22.91±2.59 b
24	16	6.12±1.20 a	9	7.33±1.73 b	12	6.25±0.97 a	19	9.00±1.15 c
28	25	3.60±0.58 a	13	3.92±0.64 a	17	3.88±0.78 a	19	5.42±0.69 b

Table 5 - Number of adults and mean time of development (days) from third instar larva to adult, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05). (*) One adult was born with deformed wings; (□) two adults, (Δ) three adults, (○) five adults were born with deformed wings.

	Plodia interpunctella		*Cadra cautella*		*Ephestia kuehniella*		*Corcyra cephalonica*	
T (°C)	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.	N.	days ± S.D.
18	3*	52.00±3.61	0	-	2*	56.50±2.12	0	-
24	11	17.73±2.53 a	9	18.33±1.22 ab	8*	19.75±1.04 c	9$^{\Delta}$	22.57±1.72 d
28	22	11.27±1.91 a	12	14.33±1.37 b	18$^{\square}$	14.56±1.38 b	15°	17.40±1.12 c

Table 6 - Number of third instar larvae, males and females, dead for starvation and survival time in days, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05).

T	*Plodia interpunctella* days ± S.D.				*Cadra cautella* days ± S.D.				*Ephestia kuehniella* days ± S.D.			
°C	N.	♂	N.	♀	N.	♂	N.	♀	N.	♂	N.	♀
18	20	11.17±2.64 a	19	12.84±3.91 a	17	12.06±1.71 a	19	11.79±2.20 a	14	13.07±1.64 a	15	13.07±2.52 a
24	12	4.37±1.32 a	11	4.45±1.49 a	18	5.18±1.13 a	15	4.47±1.51 a	13	5.23±1.30 a	17	5.65±1.27 a
28	0	-	3	4.33±0.58 a	11	5.27±1.55 a	14	5.64±1.22 a	10	5.20±0.84 a	10	4.50±0.71 a

Table 7 - Number of third instar larvae, males and females, change into pupa and mean time in days, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05).

T	*Plodia interpunctella* days ± S.D.				*Cadra cautella* days ± S.D.				*Ephestia kuehniella* days ± S.D.			
°C	N.	♂	N.	♀	N.	♂	N.	♀	N.	♂	N.	♀
18	3	11.00±1.00 a	5	13.80±1.92 a	8	13.00±1.51 b	6	14.40±1.52 a	9	11.77±1.39 ab	10	14.90±1.66 a
24	7	5.57±0.98 a	9	6.56 ± 1.24 a	4	6.80±1.30 b	4	8.00±2.16 a	7	6.14 ± 0.90 ab	5	6.40±1.14 a
28	12	3.58±0.51 a	13	3.61±0.65 a	8	3.62±0.52 a	5	4.40±0.55 b	9	4.22±0.67 b	8	3.50±0.76 a

Table 8 - Number of adults, males and females, and mean time of development (days) from third instar larva to adult, at different temperatures. Means followed by the same letter are not significantly different (Duncan's test; P>0.05). (*) One adult was born with deformed wings.

T	*Plodia interpunctella* days ± S.D.				*Cadra cautella* days ± S.D.				*Ephestia kuehniella* days ± S.D.			
°C	N.	♂	N.	♀	N.	♂	N.	♀	N.	♂	N.	♀
18	2*	50,00±1,41	1	56,00±0,00	0	-	0	-	2	56,50±2,12	0	-
24	6	16,50±2,66 a	5	19,20±1,48 a	3	17,00±1,00ab	6	19,00±0,63 a	5	19,60±1,14 bc	3*	20,00±1,00 a
28	13	10,15±1,41 a	9	12,89±1,27 a	6	13,33±1,03 b	6	15,33±0,82 b	11*	13,80±1,40b	7*	15,43±0,53 b

became adults (Tables 5, 8). Third instar larvae are able to survive more time if compared to younger larvae, because they have accumulated more supply of food. A number of larvae, different for each species, is able to change into pupa and in few cases to change into adult. In particular, the number of pupae is lower at 18°C, than at 28°C. At 18°C no *E. kuehniella* and *C. cephalonica* larva was able to become adult. Some adults of *P. interpunctella*, *E. kuehniella* and *C. cephalonica* were born with deformed wings perhaps for the lack of some nutrient. At 28°C all male larvae of *P. interpunctella* are able to change into pupae, unlike females (Table 6).

It is possible to observe that, except for one case (*C. cautella* at 24°C), adults males are more numerous than females (Table 8). Generally, males have their size smaller than females and they have a faster development (Podoler, 1974), while females become adults later. It is also noticed that the number of deformed adults of *E. kuehniella* and *C. cephalonica*, which have their size smaller than other species, is higher than those of *P. interpunctella* (one case), or *C. cautella* (no one) (Table 5).

CONCLUSIONS

The survival skill of *Plodia interpunctella*, *Cadra cautella*, *Ephestia kuehniella*, *Corcyra cephalonica* larvae increases when temperature goes down, as was observed by Candura (1928), probably on quiescent larvae of *P. interpunctella* and *E. kuehniella* larvae. *C. cephalonica* larvae are the most resistant to starvation, if compared to larvae of the same age of other species.

The larval survival of these Pyralid moths is similar to survival time of *Lasioderma serricorne* larvae (Stein, 1986).

Some adults with deformed wings were born from third instar larvae. They probably ingested an insufficient quantity of fatty acids, which are essential for right development of insect (Fraenkel & Blewett, 1946). It is also highlighted that starvation prevented the synthesis of the two most important proteins of haemolymph that could have interfered with their development (Ismail & Dutta-Gupta, 1988).

Larvae which were able to complete development, have probably accumulated a sufficient quantity of nutrients, in particular lipids - as linolenic acid - essential for metamorphosis (Dadd, 1973).

In conclusion, it is possible to summarize the consequences of starvation on larvae of *P. interpunctella*, *C. cautella*, *E. kuehniella*, *C. cephalonica* as follows:

- death of first instar larvae after 1-4 days of starvation, depending on tested temperature and species (Table 1);
- anticipation of mould for second instar larvae;
- anticipation of metamorphosis for third instar larvae;
- birth of some adults with deformed wings, probably for a low quantity of fatty acids;
- birth of a number of males higher than females, thanks to their smaller size.

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Dr Sara Savoldelli - Istituto di Entomologia agraria, Università degli Studi di Milano, Via Celoria 2, I-20133 Milano (Italy). E-mail: sara.savoldelli@unimi.it

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