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Assessment of temperature effects on the development and fecundity of *Pullus mediterraneus* (Col., Coccinellidae) and consumption of *Saissetia oleae* eggs (Hom., Coccoida)

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Abstract: Eggs, larval and nymphal periods and fecundity of *Pullus mediterraneus* were examined under 16 h light : 8 h dark combined with six constant temperatures: 15, 20, 25, 30, 35 and 40°C. Eggs of *Saissetia oleae* were used as prey. The developmental time at 15, 20, 25, 30 and 35°C was 17.23, 4.5, 2.64, 1.67, 1.28 days for eggs and 98.47, 68.88, 53.94, 28.96, 36.51 days for larval–pupal duration, respectively. At 7°C no eggs hatched, and at 40°C all the stages died after 36 h of maximum exposure except the three last stages. The fecundity of females rearing at different temperatures ranged between 1.7 eggs at 15°C and 601.86 eggs at 30°C. The pre-oviposition period ranged between 23.75 days at 15°C and 3.47 days at 35°C. The consumption of *S. oleae* eggs by the larvae reached 597.69 eggs during the pre-imaginal development. Females attacked more eggs than males averaging 77.69 ± 22.34 eggs per 4 day period compared with 46.97 ± 10.12 eggs per 4 day period for males.

1 Introduction

Saissetia oleae Olivier (Hom., Coccoidae) is a major pest of olive trees in the south of Morocco (Haouz). Resistance of this pest to all available chemical and environmental contamination caused by insecticides used for its control (Civantos, 1995), have resulted in the development of effective biological control agents.

There are many natural enemies, belonging to the Hymenoptera and Coleoptera (Coccinellidae), that are associated with this pest. Several studies have already shown that some parasites of the genus *Metaphycus* could be good agents to control black scale on olive trees (Panis, 1974, 1978; Argyriou and Katsoyannos, 1976; Katsoyannos, 1976; Blumberg and Swirski, 1977; Paraskakis et al., 1980; Viggiani and Mazzone, 1981) and the real potential of the Coccinellidae is still unknown. Among these, *Pullus mediterraneus* Fabre appears to be a particularly good polyphagous predator (Panis, 1977). The presence of this species was reported for the first time in Morocco by Smirnoff in 1956. It is widely found in the orchards of the Haouz region (south of Morocco) (Chemseddine, 1988) and its numerical strength and time of occurrence are closely related with those of *S. oleae* (Ba M'Hamed, 1993).

Studies of the biological characteristics of an insect must be undertaken prior to its mass-production and release as a biological control agent. The purpose of the present study was to determine the developmental duration of *Pullus mediterraneus* from eggs to adults and the fertility of the females under constant temperatures. The voracity of larvae and adults on eggs of *S. oleae* was also studied.

2 Material and methods

Adults of *Pullus mediterraneus* were collected at various sites in the Haouz. Three series of experiments were carried out.

In the first series, five batches of 120 newly laid (0 to 24 h old) *P. mediterraneus* eggs were placed on humidifid filter paper in a Petri dishes and each batch was kept at one of the seven temperatures: 7, 15, 20, 25, 30, 35 or 40°C. The development of the eggs was examined every day. Newly hatched first instar larvae were confined individually in Petri dishes and reared at one of six temperatures (15, 20, 25, 30, 35 and 40°C). A layer of female *S. oleae* that had been collected directly from orchards were given to the larvae and renewed every day. The dates of the moults into successive instars (visible exuvia was used as the evidence of moulting) and emergence of adults were recorded daily.

In the second series of experiments, pairs (one male and one female) of newly emerged adults (0 to 24 h old) were confined in Petri dishes, the bottoms of which were covered with filter paper. The black scale carapaces, which were either empty or full of eggs were placed in the Petri dishes. For each female, kept at one of the different temperatures, the date of the first laying was recorded and the oviposition activity was observed daily. All experiments were conducted in temperature-controlled hood (± 2°C), under a 16 h light : 8 h dark photoperiod and 70–80% of relative humidity. To prevent a fall of humidity which occurred at temperatures of 30, 35 and 40°C, large pans of water were placed in the etuve.

The purpose of the third series of experiments was to evaluate the consumption of *S. oleae* eggs by larvae and

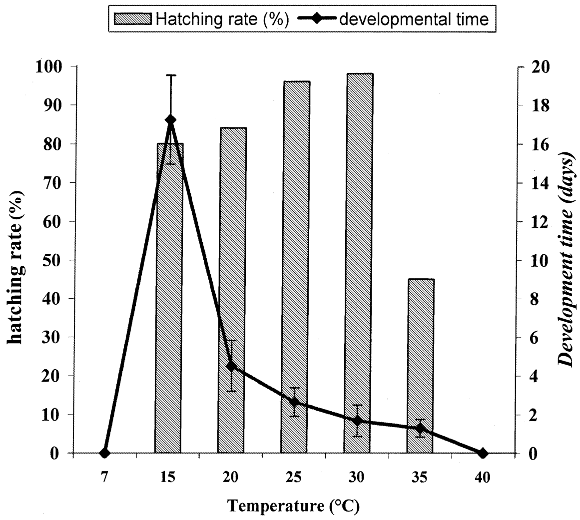

Fig. 1. The relationships between the temperature and the developmental time and hatching rate for Pullus mediterraneus *eggs at 16 h light : 8 h dark and 70–80% relative humidity*

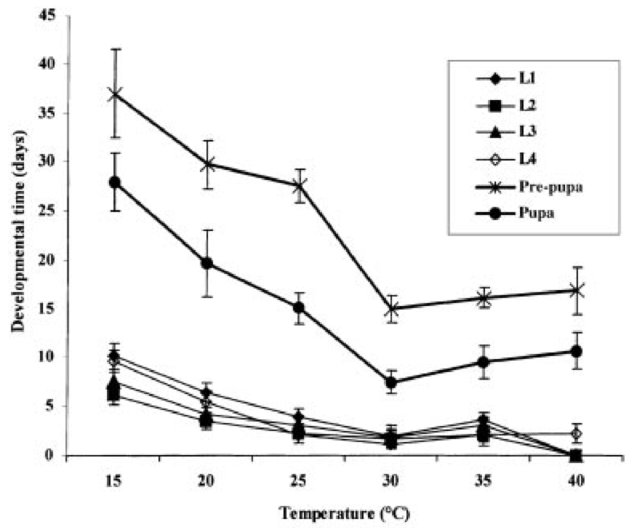

Fig. 2. The relationships between the constant temperature and developmental time for each of the pre-imaginal stages (first larval stage) in Pullus mediterraneus *reared on* Saissetia oleae *eggs*

adult of *P. mediterraneus*. The tests were carried out in the laboratory in which the temperature fluctuated between 25 and 30°C and the relative humidity between 40 and 60%. Larvae and adults of *P. mediterraneus* were pre-conditioned before testing to standardize their levels of hunger. The larvae were collected at least 8 h after the last moult, isolated and starved for 12 h. The adults were individually starved for 24 h. The larvae and adults (males and females) were supplied with high and determined quantities of *S. oleae* eggs placed on black paper in the Petri dishes. The number of eggs eaten daily was recorded for each individual and replaced by new quantities. This experiment was carried out on 37, 32 and 29 larvae at the L1, L2 and L3 stages, respectively, and 28 at the (L4 + pre-nymphal) stage. Regardless of their longevity, the number of eggs consumed by the adults was only recorded for 4 days in 42 males and 42 females.

3 Results

The embryonic developmental times of *P. mediterraneus* under the seven different temperatures (7, 15, 20, 25, 30, 35 and 40°C) are shown in fig. 1. The mean duration decreased with ascending temperature between 15 and 35°C. The average developmental time of the eggs was 17.23 ± 2.28, 4.5 ± 1.31, 2.64 ± 0.74, 1.67 ± 0.81 and 1.28 ± 0.45 days at 15, 20, 25, 30 and 35°C, respectively. At 7°C no egg hatched after 30 days of incubation, and only 66% remained in good aspect (turgescent and yellow in colour, which meant that there had been no development). When those eggs were placed at 25°C, hatching occurred in the normal way. At 40°C, all the eggs dehydrated completely and died after 12 h (eggs with dead embryos become dark). The eggs hatching rate shown in fig. 1 indicates that the ratio were high and ranging between 80% at 15°C and 98% at 30°C. The exposure of eggs to a constant temperature of 35°C decreased the hatching rate to nearly one-half and 55% of the eggs dried and died after 24 h.

The effect of the six constant temperatures on larval and pupal developmental time is summarized in fig. 2. The prepupal and pupal periods showed the longest duration of development. The duration of development was shortest for all stages at 30°C. Development from hatched egg to adult was fastest at this temperature, with a developmental time of 28.96 days. All the stages exhibited a slackening of development at 35°C and the development duration recorded was 36.51 days. No larvae of the three first stages (L1, L2 and L3) was able to develop at 40°C; however, the fourth, prepupal and pupal stages were more resistant to dryness and continued their development, although less rapidly in comparison with those reared at 30 or 35°C. Figure 3 presents the mortality ratio during the pre-imaginal development according to the different

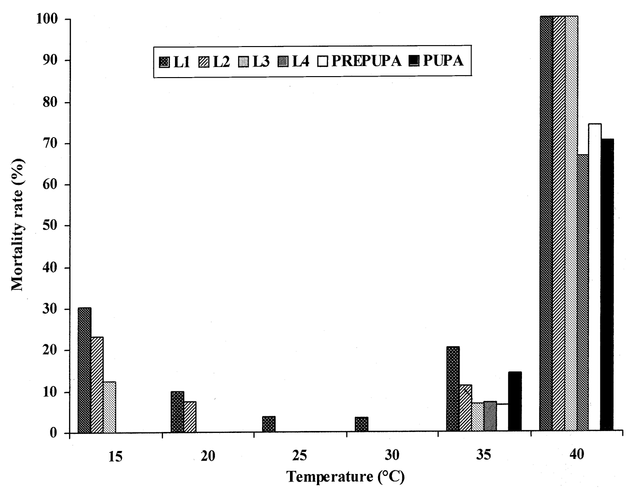

Fig. 3. Stage-specific mortality of Pullus mediterraneus *under six constant temperatures and held at a photoperiod of 16 h light : 8 h dark and 70–80% relative humidity*

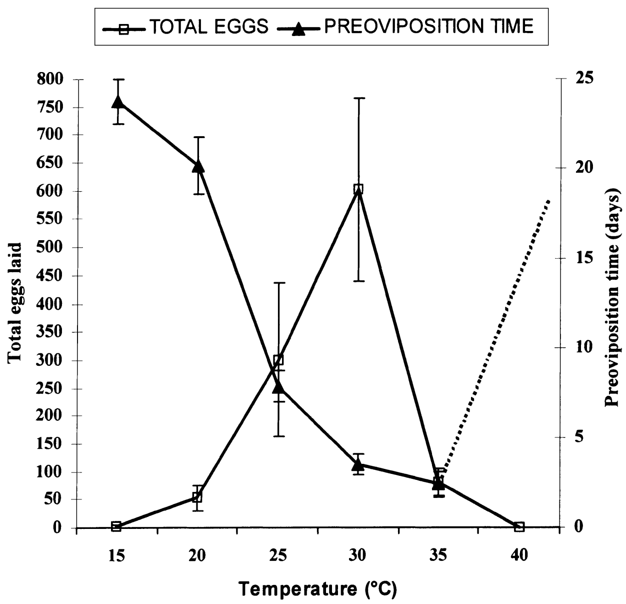

Fig. 4. *Total number of eggs laid and pre-oviposition duration at six temperatures for* Pullus mediterraneus

temperatures. The first stages, regardless of their small size and their high sensitivity were especially affected by mortality at the all of the test temperatures. The mortality ratio were weak at 25 and 30°C (3.57 and 3.4%, respectively). The highest mortality ratio were recorded at 35 and 40°C for all stages. A temperature of 40°C produced 100% mortality of the first three stages after 36 h of exposure, and only 33.4, 29.63 and 25.93% of the L4, prepupal and pupal stages, respectively, survived.

The fertility of the females varied substantially over the six temperatures, from a mean of 601.86 ± 162.66 eggs/female at 30°C to a mean of 1.75 ± 0.95 eggs/female at 15°C (fig. 4). The total number of eggs laid per individual within a temperature also varied substantially. The maximum oviposition recorded was 831 eggs at 30°C. The differences in total fertility could be attributed to differences in longevity and daily fertility at the six temperatures. The pre-oviposition period of

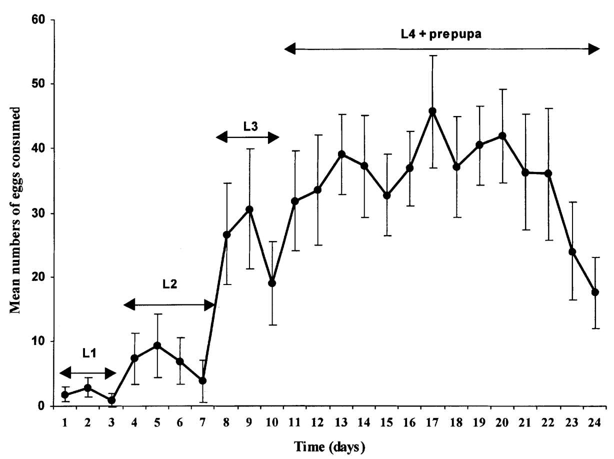

Fig. 5. *Mean number of* Saissetia oleae *eggs attacked daily by each of the larval stages held at 25–30°C and 40–60% relative humidity*

P. mediterraneus females varied according to the temperatures. It ranged from 23.75 days at 15°C to 2.43 days at 30°C. Although the females survived at 40°C, no eggs were laid.

The mean number of *S. oleae* eggs consumed daily by the selected stages of *P. mediterraneus* are presented in fig. 5. The figure shows an identical pattern of consumption in the four stages. The maximal quantities of eggs attacked were recorded at the beginning of each stage and this quantity reduced considerably at the approach of the moults. A heterogeneity in food intake rhythms was observed at the different stages of development and it was reflected by the important standard deviation. The consumption of larvae was very important at the fourth stage, it represented 81% of the total consumption during the pre-imaginal stages. Females attacked more eggs (77.69 ± 22.34 eggs per 4 days) than males (46.97 ± 10.12 eggs per 4 days).

4 Discussion

Differences in the value of biocontrol agents in the field can be predicted with caution, from measurements of biological parameters used in quality monitoring. Hattingh and Samways (1994) indicated that there may be a large selective advantage if coccinellids have rapid larval development, thereby reducing the duration of this highly vulnerable life stage. The experimental results obtained in our study indicate that the developmental and reproductive biology of *P. mediterraneus* is strongly affected by exposure to a constant temperature in the range 15 to 40°C, with 35°C probably approaching the physiological limit for this coccinellid. The developmental rate was maximal at 30°C and it was lowest at 15°C, probably due to simple poikilothermic reduction in developmental physiology. The development tend to be slower at 35°C than at 30°C. This could be explained by the fact that at this temperature all the metabolic and physiological functions were modified in order to resist the high temperature that would cause drying of the insect. The same phenomena was reported in *Clitostethus arcuatus* (Bellows et al., 1992). Development from eggs to adults ranged between 86.75 days at 15°C to 28.92 days at 30°C. These data suggest that the pattern of development for each life stage of *P. mediterraneus* is lower than that of larvae and pupa of the other species of the same genus, such as *Scymnus frontalis* (Naranjo et al., 1990) and that of temperate zone aphidophagous coccinellids at 15°C (Miller, 1992; Miller and Lamana, 1995).

Eggs, larval and pupal mortality differed according to temperature. No larvae hatched from eggs held at 7 or 40°C. At 7°C the embryonic development was blocked (the eggs remained yellow). The eggs could be conserved at least, for 30 days at this temperature. In contrast, at 40°C all the eggs died (eggs have a dark colour). Important larval mortality was found in the first three instars at 15°C (65.70%). Previous studies on larval–pupal survival in temperate zone coccinellids have reported a relatively high death rate in larvae at temperature below 18°C. Miller (1992) and Rodriguez-Saona and Miller (1999) observed up to 83% mortality at 17°C for *Hippodamnia convergens* Guerin-Meneville. Orr and Obrycki (1990) noted that *H. parenthesis* exhibited 67% mortality at 14°C. Similarly, *Scymnus frontalis* experienced 74% mortality at 15°C (Naranjo et al., 1990). The aforementioned studies also recorded that the mortality of coccinellids ranged between 85 and 100% at temperatures below 13°C. However, an exceptional species, *Eriopis connexa* Mulsant, exhibited 33 and 4.8% mortality at 14 and 18°C, respectively (Miller and Paustian, 1992). In contrast, Obrycki and Tauber (1982) and Michels and Behle (1991) reported that high mortality at the lower temperatures occurred in the later stages of development. Our results are consistent with those found in *Rhyzobius forestieri* in which pupa were more resistant to the heat than the younger stages (Katsoyannos, 1983).

The fertility of *P. mediterraneus* females was greater at 30°C, lower at 15°C and absent at 40°C. The preoviposition period was shorter for females reared at 35°C in comparison with those reared in the temperature range 25–30°C. The capacity of *P. mediterraneus* to have a high rate of reproduction at high temperatures in the laboratory (30 and 35°C) agrees with the data of the ovarian activity study in the orchard (Ba M'Hamed, 1993). *Pullus mediterraneus* females showed a high ovarian activity during the summer. This confers upon the predator an important role in the regulation of the *S. oleae* population during the aestivate period when coccidiphagous coccinellids such as *Exochumus quadripustulatus* are in diapause in most olive groves (Katsoyannos, 1983).

During all the pre-imaginal periods one larvae of *P. mediterraneus* could attack 597.69 eggs of *S. oleae*. This consumption, because of the small size of this coccinellid, is moderate in comparison with other coccidiphagous coccinellids (Katsoyannos, 1984). However, it is close to that of *Pharoscymnus semiglobolus*, which showed similar characteristics to *P. mediterraneus* (size and larval developmental time) (Iperti et al., 1972). Larvae attacked eggs when they were continuously available in excess of needs. At high prey densities, encounters were more frequent; a predator stays in an area where prey aggregates and continues to feed until satiation or even beyond (Frazer and Gill, 1981). Any estimation of the predation capacity of the *P. mediterraneus* population should include consideration of sex ratio, because males attacked less often than females.

In conclusion, the basic biological measurements in the laboratory were in favour of improving the biocontrol predictions of *P. mediterraneus*. The present results provide data for establishing suitable conditions for rearing. Mass culture methods could be enhanced by selecting temperature conditions for either rapid development or high fecundity. With respect to biological potential and voracity, *P. mediterraneus* shows a good adaptation to the climatic conditions of the south of Morocco, where the summer temperatures are high and often in excess of 30°C and when *S. oleae* females show a greater reproductive activity.

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