

Determination of suitable host plant for rearing of *Coccus hesperidum* (Homoptera:Coccidae)

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Brown soft scale, *Coccus hesperidum* Linnaeus (Homoptera: Coccidae) is an economically important pest on wide range of horticultural and agricultural crops. A study was carried out to determine the suitability of three host plants, black nightshade (*Solanum nigrum*), mandarin (*Citrus reticulata*) and cotton (*Gossypium barbadense*), for mass rearing of *C. hesperidum* under laboratory conditions of 25 ± 2 °C, $65\pm 5\%$ RH and photoperiod of 14D:10L. *C. hesperidum* was able to develop and reproduce on all three host plants. However, according to developmental period, longevity and fecundity, mandarin was the best among the three host plants.

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1. Introduction

The family Coccidae comprises many species, some of which are among the most serious horticultural and agricultural pests in tropical regions (Talhok 1975, Ben-Dov 1993, Paolo *et al.* 2004). This is typical to the brown soft scale, *Coccus hesperidum* Linnaeus, which is increasingly drawing major attention. This insect is well known as the main coccid pest of citrus with an extremely wide host range such as olive, avocado, cotton, mango, cocoa, ficus, hibiscus, oleander, palm, fern and orchid (Ben-Dov & Hodgson 1997, Malais & Ravensberg 2003).

Scale insects cause crop losses by severely depleting plant cell nutrients resulting in a reduction of photosynthetic capacity. Damages caused to plants include loss of sap, clogging of leaf or

fruit surfaces with honeydew, on which sooty mould subsequently grows (Ben-Dov & Hodgson 1997). *C. hesperidum* reproduces parthenogenetically. Sexual reproduction occurs in some circumstances but the male scales are rarely found (Copland & Ibrahim 1985).

A fertilized female of this coccid carries its eggs in a large brood chamber where newly emerged larvae remain for a few hours before crawling to other host parts. Crawlers move over the plants to find a suitable feeding site typically along a leaf vein (Annecke 1959, Gill 1988, Kosztarab 1996, Johnson 2002). The second and third nymphal stages are distinguished by the size of the scale. Annecke (1959) distinguished second stage nymphs to be less than 1/12 inch long and third stage nymphs being greater than 1/12 inch.

For multi-crop insect pests, knowing the plant on which they feed may aid to discover control strategies. Accurate information regarding the life history traits of the target has the potential to generate a better understanding of the factors governing pest population development. Such information is also important for a chemical-based control program (Van Rijn *et al.* 1995) and for a large-scale production of high quality laboratory-reared insects for biological control purposes (Conlong & Mugoya 1996). Clearly, rearing insects on a suitable host plant is central to these issues (Zalucki *et al.* 1986).

Given the increasing economic importance and the limited information on the laboratory rearing of *C. hesperidum*, a screening study was carried out with this pest on three different types of plants. The main objective of the current study was to compare developmental parameters, reproduction rate and longevity of *C. hesperidum* on these important hosts for biological control purposes.

2. Materials and methods

2.1. Laboratory rearing of brown soft scale

The laboratory colony of *Coccus hesperidum* used in this study was maintained at the Department of Plant Protection, Faculty of Agriculture, Guilan University. It was derived from the leaves collected from *Citrus sinensis* plants in Rasht, Iran. These leaves were placed in close contact with the butternut squash, *Cucurbita moschata* in a climatic chamber at 25 ± 2 °C, $65\pm 5\%$ RH and 14L:10D photoperiod in the laboratory. Crawlers were allowed to infest the squashes. Samples of squashes were washed in a 5% bleach solution to prevent fungal growth. They were then rinsed twice, dried and used for experiments.

2.2. Experimental host plants

Three host plants of *C. hesperidum*, cotton (*Gossypium barbadense*), black nightshade (*Solanum nigrum*) and mandarin (*Citrus reticulata*), were used as experimental hosts. These plants were heavily infested with *C. hesperidum* in our

region. Individuals of the two first plants were grown from seeds in plastic pots under greenhouse conditions of 25 ± 2 °C and $65\pm 5\%$ RH at the Faculty of Agriculture, Guilan University. The vent openings of the greenhouse were covered with insect-proof nets to avoid undesired insect infestations. The mandarin plants were provided by Ramsar Citrus Research Institute. The plants were watered as required and fertilized with stern's Miracle-Gro on a weekly basis.

2.3. Determination of crawler's establishment rate on different host plants

For each host plant species, ten leaves of similar age and size from ten individuals of each plant species were collected for the experiment. Each leaf was individually inserted into a 200 ml plastic container filled with water, through a hole on the lid so that the petioles were down-oriented. The gap between the lid hole and the petiole was filled with cotton to prevent drowning of the released crawlers. Ten crawlers were transferred singly to each leaf by a fine hair brush made of a single hair due to the delicate and small body of the crawlers. The 30 plastic containers holding crawlers were then kept in a 24L-climatic chamber. As there is a possibility that responses vary with light conditions, a similar set of plastic containers treated as explained above were kept in a 24D-climatic chamber. Both chambers had otherwise similar environmental conditions (25 ± 2 °C, $65\pm 5\%$ RH). After 24 hours, the 60 leaves were examined for the presence of established crawlers. The numbers of established crawlers were recorded for each plant species and used to calculate the related establishment rates.

2.4. Development of brown soft scale on different plants

Ten newly hatched crawlers from the females reared on the squashes were transferred to the leaves of ten individuals of each host plant raised in pots. After 24 hours, five crawlers were kept and allowed to develop on the leaf substrates and the remaining ones removed. The potted plant was kept separately in a net cage (30 cm×30

cm×40 cm) covered with fine muslin to prevent undesired infestations. Each crawler was numbered (from 1 to 5) on each host plant using an ink pen. Development of the crawlers was checked daily using a stereomicroscope and nymphal stages determined based on morphological differences.

2.5. Reproduction rate and longevity of brown soft scale on the host plants

The crawlers were reared up to adult stage on the plants mentioned above. When they became mature, females were checked daily to record the onset of reproduction period and lifetime fecundity. Non-drying glue was covered in one centimeter distance on the leaf substrates around each female so that when the crawlers emerged, they moved and stuck on the glue. The stuck crawlers were removed daily and their numbers recorded and used to calculate reproductive parameters: pre-oviposition, oviposition, post-oviposition periods, the daily number of crawlers per female, the number of crawlers laid per female during the whole lifetime and parent longevity.

2.6. Statistical analysis

Data (mean value of individuals within each replicate) of developmental time of immature stages, fecundity, daily crawler production per female, duration of pre-oviposition, oviposition, post-oviposition and longevity were analyzed by analysis of variance (ANOVA). Mean values were separated using the Tukey's test when significant differences were indicated ($P < 0.05$). Paired t-test was used to compare establishment of crawlers under dark and light conditions (SPSS Inc 2004).

3. Results

3.1. Determination of crawler's establishment rate on different host plants

Establishment rate of crawlers on all plants species was significantly higher in dark than in light (paired t-test, $P < 0.05$). Mean percentage of

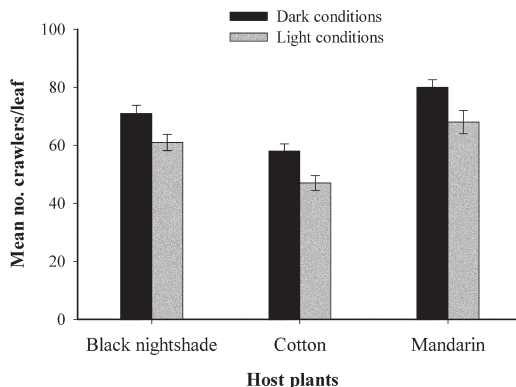


Fig. 1. Mean (\pm S.E.) establishment of crawlers on different host plants under dark and light conditions.

crawler's establishment on the three host plants varied from 47% on cotton to 68% on mandarin under light and 58% on cotton to 80% on mandarin under dark conditions, respectively (Fig. 1).

Under both photoperiods, the establishment of crawlers on mandarin and black nightshade were similar and significantly higher than recorded on cotton (dark condition: $F_{2,27}=17.9$, $P < 0.001$; light condition: $F_{2,27}=8.6$, $P < 0.001$).

3.2. Development of brown soft scale on different plants

Plant species significantly affected the developmental period of *C. hesperidum*. Individuals maintained on cotton had longer developmental periods than their counterparts kept on mandarin and black nightshade. However, development on mandarin was on a par with black nightshade (Table 1).

3.3. Reproduction and adult longevity

Tukey's test revealed that the pre-oviposition period was similar on the three plants but the oviposition period was significantly the longest on the mandarin leaves (Table 2). There was significant difference in fecundity among the host plants. The highest fecundity of *C. hesperidum* was observed on mandarin, which was approximately 1.75-fold higher than recorded on cotton (Table 2). Mean crawler production on all the

Table 1. Duration of developmental periods (Mean ± S.E.) of immature stages of *Coccus hesperidum* on three host plants.

Host plant	First instar	Second instar	Third instar
Black nightshade	24.7±0.26 a	13.1±0.16 a	8.9±0.14 a
Mandarin	23.9±0.28 a	13.0±0.25 a	8.4±0.16 a
Cotton	28.8±0.19 b	15.0±0.19 b	10.2±0.1 b
<i>F</i> _{2,27}	110.1	32.1	42.9
<i>P</i> -value	<0.001	<0.001	<0.001

Mean values within the same column followed by different letters are significantly different (*P*<0.05).

Table 2. Pre-, post- and oviposition periods and adult longevity (Mean ± S.E.) of *Coccus hesperidum* on three host plants.

Host plant	Pre-ovip. period	Ovip. period	Post-ovip. period	Fecundity	Longevity
Black nightshade	2.2±0.16 a	26.6±0.23 a	6.8±0.39 a	309.9±4.29 a	35.1±0.52 a
Mandarin	2.1±0.15 a	31.8±0.19 b	8.6±0.48 b	380.3±3.27 b	41.7±0.39 b
Cotton	2.2±0.12 a	21.9±0.11 c	4.3±0.30 c	218.6±4.05 c	29.0±0.14 c
<i>F</i> _{2,27}	0.8	714.0	29.9	718.9	275.9
<i>P</i>	0.47	<0.001	<0.001	<0.001	<0.001

Mean values within the same column followed by different letters are significantly different (*P*<0.05).

three plants peaked during the second week of the reproduction period with 26.16±0.32, 25.62±0.24, 18.52±0.20 crawlers produced on mandarin, black night shade and cotton leaves, respectively. Daily crawler production on mandarin, black nightshade and cotton was 11.21±1.27, 10.26±1.33, 9.02±1.08 crawlers, respectively (Fig. 2). *C. hesperidum* reared on the mandarin leaves lived the longest among the host plants (Table 2).

4. Discussion

The culture and colonization of herbivorous pests is fundamental to the understanding of their population dynamics and is a practice of great importance for the design of sound control strategies. Related to this importance, the search for suitable host plants for insect pests has been an especially active field. These include scale insect species, some of which are among the world's most serious horticultural and agricultural pests (Paolo *et al.* 2004). Attempts have been made to colonize coccids using various plant species. The

oleander scale, *Aspidiotus nerii* Bouche successfully survive, reproduce and mass produce on butternut pumpkins, *Cucurbita moschata* Duchesne (Papacel & Smith 1985). It was also found that when detached potato sprouts were used as host, the hemispherical scale, *Saissetia coffeae* could be maintained in the laboratory as did *C. hesperidum* when fed squash fruits, oleander plants, butternut and hubbard squash (Blumberg 1976, Ponsonby & Copland 2007). The latter species has been shown to reproduce on citron mel-

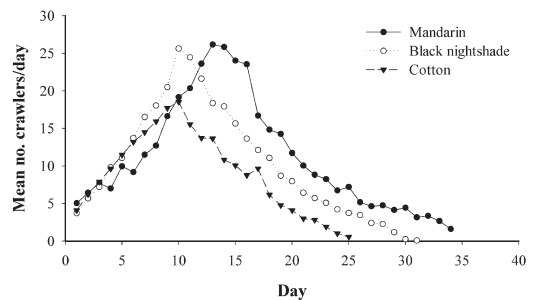


Fig. 2. Mean number of crawlers produced by females per day on different host plants.

ons (Reed *et al.* 1968). However, the cost of growing this host was considered very high and labor intensive.

The quality of food affects longevity, feeding, growth and developmental time of immature brown soft scale, *C. hesperidum* (Annecke 1966). In this study, *C. hesperidum* could complete its life cycle on all the three plants. However, some biological attributes, such as developmental duration and fecundity, were significantly different among these three host plants. Rearing of *C. hesperidum* on the mandarin leaves resulted in the shortest developmental period and the highest fecundity. This result could be explained by the fact that *C. hesperidum* population originated from the closely related plant species (*Citrus sinensis*). The faster developmental rates and the higher fecundity of insects on host plants show a better suitability of the host plant (Van Lenteren & Noldus 1990).

Saakyan Baranova (1964) argued that the type of host plant is an affective factor in establishment of *C. hesperidum* crawlers. The lower establishment rate of *C. hesperidum* on cotton leaves was mainly due to the morphological attributes of this plant with pubescent leaves that makes the crawlers difficult to establish. In the present study, the crawlers showed more movement under light condition due to their positive phototactism. Similar observations were reported also by Ben Dov and Hodgson (1997) and Washburn and Frankie (1981) for scale insects, *C. hesperidum* and *Pulvinariella mesembryanthemi*, respectively. Based on the present study it could be concluded that among the three host plants tested, mandarin is the most suitable host for rearing of *Coccus hesperidum*.

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