J. ISSAAS Vol. 17, No. 2:29-37 (2011)

COMPOSITION OF SCALE INSECTS ON COFFEE IN DAKLAK, VIETNAM AND REPRODUCTIVE BIOLOGY OF JAPANESE MEALYBUG, *PLANOCOCCUS KRAUNHIAE* KUWANA (HEMIPTERA: PSEUDOCOCCIDAE)

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(Received: September 13, 2010; Accepted: August 21, 2011)

ABSTRACT

There were seven species of scale insects, including: 4 species belonging to the family Pseudococcidae, 2 species of Coccidae, and 1 species of Diaspididae, which infest coffee in Daklak, Vietnam and the Japanese mealybug, *Planococcus kraunhiae* Kuwana was the key insect pest. These were reared individually on coffee leaves at 27.82^{o}C and 28.76^{o}C and humidity of 79.53 % and 80.95% in the laboratory, to determine the development, life cycle, and fecundity at temperatures reflective of Daklak coffee field conditions. The duration of development from egg to adult ranged from 39.20 days and 34.40 days as temperature increased from 27.82^{o}C and 28.76^{o}C, respectively. Females laid an average of 146.8 and 153.2 eggs per female at the above temperatures, respectively. Likewise, the egg hatchability was 95.35% and 94.24 % at both temperatures, respectively. The male is holometabolic, develops through four stages: egg, larva (two instars), pupa and adult, while the female is hemimetabolic, which develops through only three stages: egg, larva (three instars) and adult. These data are very useful in predicting their emergence and damage so timely control measures can be implemented.

Key words: adult oviposition, development, hatchability, reproduction

INTRODUCTION

Coffee is one of the important industrial crops in Daklak province due to its high export potential and internal demand. Daklak has the largest area of coffee, occupying 85% of the coffee area in Vietnam. Within five years, the coffee production area in Daklak increased by more than 14,000 ha, from 170,403 ha in 2005 to 184,500 ha in 2009. Although the area increased rapidly, the yield was unstable, the average yield fluctuated from 0.7 to 0.8 tons ha^{-1} in 1980 to 3 to 4 tons ha^{-1} in 2008 (Dung and H'dok, 2009), then dropped to 2 to 2.5 tons ha^{-1} in 2009-2010, therefore the output decreased to about 415,500 tons in 2008-2009 and about 400,000 tons in 2009-2010.

One of the principal reasons for the unstable yield of coffee is pest infestation, especially that of scale insects. In Vietnam, scale insects were also recorded on potato, coffee and citrus trees (Lam, 1994). Scale insects are major economic pests, particularly on perennials such as fruit and nut trees, woody, forest trees and shrubs. Plant damage may occur directly by feeding, or by injecting toxins or plant pathogens causing stunted trees, defoliation, shedding of flowers when found in high density, while unchecked heavy infestation may kill trees (Plant Protection Daklak, 2004). In addition, the honeydew that is produced by many scale insects is an important source of food for ants and other foraging insects.

A pest outbreak in 2004 and 2005, severely damaged thousands of hectares of coffee, especially in Daklak. Daklak had 27,418 hectares infested with scale insects in 2004, of these 2,873 ha were damaged severely (Plant Protection Daklak, 2004).

The classification of scale insects is unsettled and is currently evolving. It has been based almost entirely on the morphology of the degenerate adult female. There are about 6,000 described species of scale insects, belonging to about 800 genera world wide. The number of families recognized by different coccidologists varies between 15 and 22 (Kosztarab and Kozars, 1988). According to a catalogue to be published shortly, the number of world mealybug species is about 2,000 (Cox and Ben-Dov, personal communication). At the beginning of the 20th century, only three species had been recorded in the South Pacific area (Fernald, 1903) however, half a century later, Dumbleton (1954) recorded 18 species.

From 1967 to 1968, 26 scale insects and mealybug species were recorded on citrus fruit trees, belonging to 15 genera and 4 families (PPRI, 1976). In 1994, there were 57 species on citrus fruit trees recorded, belonging to 24 genera, 4 families. Among these, the major species belonged to the family Diaspididae (27 species), while the other families Coccidae, Pseudococcidae and Margarodidae had 18, 8 and 4 species discovered, respectively (Lam, 1994). There are 41 species of scale insects that are associated with 23 fruit trees, 9 industrial and vegetable plants and 13 ornamental plants. These belong to 7 families, of which 13 species from the Pseudococcidae were the highest percentage of species collected (Chat et al, 2005).

The Japanese mealybug, *Planococcus kraunhiae* Kuwana, produces sooty mold on the surface of fruits and leaves (Ueno, 1963; Oomasa, 1990) and is distributed in Japan, China, Eritrea and North America (Kawai, 1980). In Japan, it is known to be a euryphagous pest of fruit trees such as pears, citruses, grapes and persimmons (Kawai,1980). Recently, the damage caused by *P. kraunhiae* is increasing on citruses, persimmons and figs (Oomasa, 1990; Tsutsumi, 1997; Shibao and Tanaka, 2000). This might be due to the development of resurgence that occurred when natural enemies populations decreased with the use of synthetic pyrethroid insecticides against pests such as stink bugs, aphids and the citrus leaf miners, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae) (Oomasa, 1990; Tsutsumi, 1997).

Although synthetic pyrethroid insecticides were found to be less effective than organophosphorus insecticides on *P. kraunhiae* in a fig field, there was no study on the influence of these chemicals on natural enemies or reproduction of mealybugs (Shibao and Tanaka, 2000). In recent years, damage has increased and it is reported that it is the most serious pest on Japanese persimmon in Japan. A study on the natural enemy complex of *P. kraunhiae* in Fukuoka revealed 8 parasitoid species and 4 predator species, among which 5 parasitoid species and 4 predator species were newly recorded on *P. kraunhiae*. Species such as *Allotropa subclavata, Anagyrus fujikona* and *Trisopsis incisa* were abundant and considered to be important species for integrated pest management (IPM) in Japanese persimmon production (Teshiba and Tsutsumi, 2004).

Many food sources have been used as alternative food for rearing mealybugs, such as Japanese pumpkin (Murakami, 1965; Ueno, 1977; Izawa and Utida, 1991), bleached potato sprouts (Murakami, 1965), sprouted potatoes (Tsugawa, 1972) and potted citrus (Arai, 1996). Various plants and plant parts such as rooted leaves, detached leaves, fruits and sprouts have been used as alternative food sources to rear various soft scale insect cultures for laboratory and mass rearing (Rose and Stauffer, 1997). Methods using broad bean seeds for rearing some polyphagous insects such as aphids, and thrips and oviposition substrate in mass rearing of predatory bugs were devised by Murai (1991), Murai and Loomans (2001), and Murai and co-workers (2001).

The studies on scale insects and mealybugs on coffee in Daklak are still quite limited. So far, studies on species *P. kraunhiae* have not yet been conducted.

In this paper, we investigated species composition of scale insects on coffee in Daklak, Vietnam and reared Japanese mealybug (*P. kraunhiae*) on coffee leaves to determine some biological characteristics such as: development duration of larval instars, life cycle and fecundity, which can be used in developing effective measures for monitoring and control.

MATERIALS AND METHODS

Scale insects were collected every month from 5 major coffee districts which usually had high density of scale insects and severe damage in Daklak province, namely: Krong Buk, Krong Pak, Krong Ana, Buon Ma Thuot, Cu Mga districts from 2005- 2008. The collection was conducted on 45 trees at 9 points located as a diagonal line across each of the 3 plantations for each district, with a minimum area of 6 ha. Scale insect specimens were collected from all parts of the coffee tree such as leaves, flowers, branches, berries, root and tree-trunk. Many samples had to be collected for the verification because it is based on characteristics of the adult female bugs. Samples collected were stored in clear nylon bags with dehumidifying papers and mealybug specimens were mounted on slides for identification following the methods explained by Watson (2000). Verification documents were based on mealybug identifications of ETI (2002). The importance of the damage for each species used the following criteria:

Ranking scale	Distribution	Number of trees damaged	Percent of tree canopy damaged
-	Present but not important	> 0% - 5%	> 0% - 5%
+	Important locally	> 5% - 20%	>5% - 20%
++	Widespread and important	> 20% - 50%	> 20% - 50%
+++	Very widespread and very important	> 50 %	> 50 %

Mealybugs (*P. kraunhiae)* were reared individually in the laboratory at room temperature from 27.82^oC to 28.76^oC and 79.53% to 80.95% relative humidity, which are the same conditions in coffee plantations, during the experimental period in 2007. Each mealybug was reared on coffee leaves in a plastic container (7 cm diameter, 5 cm depth) and leaves were replaced every day, as a general rule. The developmental period of eggs, larvae, prepupae and pupae, and pre-oviposition stage were determined by observation at 24 h intervals. More than 30 individuals were observed at each temperature. Exuvia were discarded after every observation. The number of eggs laid and the period from egg deposition to adult first oviposition (life cycle) were determined by observation at 1–3 day intervals. More than 20 females were observed at each temperature. Eggs laid were eliminated after each observation period.

Data analysis

Basal statistical parameters were analyzed using a statistical software DPS (data processing system) developed by Tang and Feng (2002) and Analysis of Variance (ANOVA) for the experimental data was done by IRRISTAT 4.0.

When: + T_{theory} (T_t) < $T_{stat\ analysis}$ ($T_{stat.}$) at α = 5% (P =95%): significant difference
+ F_{theory} (F_t) < $F_{stat\ analysis}$ ($F_{stat.}$) at α = 5% (P =95%): significant difference
+ T_{theory} (T_t) > $T_{stat\ analysis}$ ($T_{stat.}$) at α = 5% (P =95%): not significant
+ F_{theory} (F_t) > $F_{stat\ analysis}$ ($F_{stat.}$) at α = 5% (P =95%): not significant

RESULTS AND DISCUSSION

Field populations of scale insects in coffee in Daklak, Vietnam

Seven species of scale insects belonging to 3 major families were collected from coffee in Daklak province, Vietnam from 2005-2008. There were 4 Pseudococcidae species: *P. kraunhiae*; *Ferrisia virgata* Cockerell; *Planococcus lilacinus* Cockerell, and *Planococcus* sp., Coccidae had 2 species (*Coccus viridis* Green and *Saissetia coffeae* Walker), and Diaspididae had 1 (*Aulacaspis* sp.). *P. kraunhiae* is the key insect pest which damages leaves, flowers and berries (Table 1) resulting in decreased coffee yield and quality.

In Vietnam, the Plant Protection Research Institute (1976 and 1999) reported 6 species of scale insects on coffee, namely *S. coffeae*, *C. viridis*, *C. celatus* De Lotto, *Hemiberlesia palmae* Cockerell, *Ischnaspis longirostris* Signoret, and *Pseudaulacaspis dendrobii* Kuwana. Chat (2005) reported 10 species of scale insects on coffee such as *C. viridis*, *F. virgata*, *Pseudococcus citri* Risso, *Planococcus comstocki* Kuwana, *Rastrococcus* sp., *Pseudococcus citriculus* Green, *Pseudococcus longispinus* Targioni-Tozzetti, *Icerya seychelarum* West, *S. coffeae* and *P. dendrobii*.

In all the collection sites however, there were no differences in the population density of the scale insect species, which was always high for the common species.

Table 1. Species composition and plant parts damaged by scale insects found in coffee in Daklak, Vietnam.

Species	Damaged parts	Importance
Planococcus kraunhiae Kuwana	Leaves, flowers and berries	+++
Ferrisia virgata Cockerell	Leaves, flowers and berries	+ +
Aulacaspis sp.	Leaves, flowers and berries	+
Planococcus lilacinus Cockerell	Root	+
Coccus viridis Green	Leaves, flowers and berries	+
Saissetia coffeae Walker.	Leaves, flowers and berries	+
Planococcus sp.	Base trunk	-

Notes: +++: very widespread and very important +: important locally
++: widespread and important - : present but not important

Biological characteristics of mealybug (*P. kraunhiae*)

Development period

The mealybug undergoes two types of metamorphosis. The female is hemimetabolic and has three developmental stages i.e. egg, larva (three instars), and adult. The male is holometabolic and has four development stages i.e. egg, larva (two instars), pupa, and adult.

Under laboratory conditions, the mean developmental time of each stage, pre-oviposition period, nymph stage life cycle at temperature of 27.82^{o}C and 28.76^{o}C and relative humidity of 79.53 % and 80.95% with coffee leaves as food source are shown in Table 2. Differences between the two conditions were not detected in the egg, first instar and second instar stages. Significant mean differences were found in the next stages such as: the third instar was 7.4 days and 6.58 days, the pre-oviposition period was 13.8 days and 10.42 days, the nymph stage was 21.6 days and 20.08 days, and life cycle was 39.20 and 34.40 days at 27.82^{o}C at 28.76^{o}C, respectively.

Table 2. Development durations of female *P. kraunhiae* on coffee leaves

Developmental stage		Number of individuals	Mean developmental duration (days ± SE) at respective temperature and relative humidity		Statistical results	
			27.82°C and 79.53%	28.76°C and 80.95%	T_t	$T_{stat.}$
Egg		50	3.80 ± 0.25a	3.92 ± 0.29a	2.00	1.76
Larvae	1st instar	50	7.80 ± 0.71a	7.58 ± 0.29a	2.00	1.14
	2nd instar	50	6.40 ± 0.37a	5.92 ± 0.36a	2.00	1.18
	3rd instar	45	7.40 ± 0.40a	6.58 ± 0.36b	2.015	3.68
Pre-oviposition		43	13.80 ± 0.40a	10.42 ± 0.98b	2.018	8.64
Nymph stage		43	21.60 ± 0.88a	20.08 ± 0.51b	2.018	3.37
Life cycle		43	39.20 ± 1.50a	34.40 ± 0.76b	2.018	7.61
Adult longevity		43	24.91 ± 0.59a	21.02 ± 0.49b	2.018	4.66

+ Means ± SE are given. Within the same rows, means followed by different letters are significantly different by the t-test at α = 5%. For pairs of means with the same letters are not significantly different by the t-test at α = 5% (Tang & Feng 2002).

The results of individually reared *P. kraunhiae* on other food sources were reported by Arai (1996) who noted that the egg stadium, the total larval developmental times of female, and the pre-oviposition period of *P. kraunhiae* on citrus leaves at 25°C were 6.8, 29.6, and 18.7 days, respectively. The egg period, the total larval development times of female, and the pre-oviposition period of *P. kraunhiae* on Japanese pumpkin fruit at 25°C were 10 to 11 days, 27 to 35 days, and 9 to 13 days, respectively (Ueno,1977). Narai and Murai (2002) reported that the egg period, the total larval development times of female, and the pre-oviposition period of *P. kraunhiae* on germinated broad bean seeds were 11.7, 38.1, and 16.6 days, respectively, at 20°C and were 8.0, 25.7, and 11.6, respectively at 24°C.

Our study was conducted at a higher temperature than the above studies. The total larval development times of female on coffee leaves and the egg period at 28.76°C was shorter than those grown on citrus leaves, Japanese pumpkin fruit and germinated broad bean seeds. Insects are poikilothermic animals, so their body temperature closely depends on ambient temperature conditions. Each species has a different temperature range suitable for growth, and in temperatures that are appropriate. When the temperature is high, the development duration of phases shortens and when the temperature is low, these are longer. In our experiment, the temperature condition was higher, so the development duration of the phases was shorter.

The other stages for the male were significantly different under the same conditions of temperature and humidity except for the egg stage (Table 3). The first instar was 8.8 days and 8.2 days, the second instar was 8.55 days and 9.32 days , the pupal stage was 5.05 days and 4.16 days, at 27.82°C and 28.76°C, respectively. The adult period for both male and female were 22.40 days and 21.28 days, respectively. The mean longevity for the males was considerable shorter, 4.01 ± 0.47 days and 3.40 ± 0.78 days, at 27.82°C and 28.76°C, respectively.

Table 3. Development durations of the male *P. kraunhiae* on coffee leaves

Developmental stage		Number of individuals	Mean developmental duration (days ± SE) at respective temperature and humidity		Statistical result	
			27.82°C and 79.53%	28.76°C and 80.95%	T_t	$T_{stat.}$
Egg		50	3.80 ± 0.25a	3.92 ± 0.29a	2.00	1.76
Larvae	1st instar	50	8.80 ± 0.22a	8.20 ± 0.20b	2.00	2.75
	2nd instar	48	8.55 ± 0.31a	9.32 ± 0.23b	2.01	3.15
Pupa		40	5.05 ± 0.17a	4.16 ± 0.23b	2.02	3.45
Nymph stage		35	22.40 ± 0.49a	21.28 ± 0.42b	2.03	2.46
Adult longevity		35	4.01 ± 0.47a	3.40 ± 0.78b	2.03	3.12

+ Means ± SE are given. Within the same rows, means followed by different letters are significantly different by the t-test at α = 5%. For pairs of means with the same letters are not significantly different by the t-test at α = 5%.

Reproductive capacity

There was no statistical difference between the number of eggs laid per female and the oviposition period for the temperatures and humidity conditions studied (Table 4). The eggs of mealybugs are laid in clusters which are wrapped by wax. The number of eggs laid per female was 146.80 ± 13.78 eggs and 153.20 ± 16.58 eggs, at 27.82°C and 28.76°C respectively while the oviposition period lasted 11.1 days at 27.82°C and 10.6 days at 28.76°C.

Table 4. Reproductive capacity and oviposition periods of *P. kraunhiae* on coffee leaves.

Temperature (°C)	Humidity (%)	Number of individuals observed	Number of Eggs per female (mean ± SE)	Oviposition period (days ± SE)
27.82	79.53	35	146.80 ± 13.78a	11.1 ± 0.58a
28.76	80.95	33	153.20 ± 16.58a	10.6 ± 0.64a
$F_{t=1.79}$			$F_{stat.} = 1.44$	$F_{stat.} = 1.34$

+ Means ± SE are given. Within the same column, means followed by different letters are significantly different by the F-test at α = 5%. For pairs of means with the same letters are not significantly different by the F-test at α = 5%.

Narai and Murai (2002) reported that when rearing *P. kraunhiae* on germinated broad bean seeds, the number of eggs laid at 24°C was 965 eggs or 1.6 times more than that at 20°C. Adult longevity at 20°C and 24°C was 32 and 26 days, respectively, the pre-oviposition period lasted 16.6 days at 20°C and 11.6 days at 24°C. The number of eggs laid on germinated broad bean seeds at 24°C is 18.3 times as many as that on citrus leaves at 25°C (Arai, 1996) and 1.8–2.3 times as many as that on Japanese pumpkin. Our present study was conducted at a higher temperature than these studies, the number of eggs laid was lower than those on citrus leaves and germinated broad bean seeds, such that the number of eggs laid at 27.82°C was 146.80 eggs (Table 4) or 6.57 times lower than those on germinated broad bean seeds. The pre-oviposition periods on coffee (13.8 and 10.4 days, Table 2) were slightly shorter than on germinated broad bean seeds (16.6 days at 20°C and 11.6 days at 24°C).

Egg hatchability

The percent egg hatchability was higher than 94% at both temperatures, however the rate of hatchability was very different (Table 5). Eggs began hatching from the second to the seventh day and the peak was attained on the fourth day with 30.80% at 27.82°C and 29.08% at 28.76°C. The lowest number of eggs hatched was on the seventh day, only 0.54% at 27.82°C and 0.26% at 28.76°C. There were no more eggs hatched after eight days.

Table 5. Daily egg hatchability of *P. kraunhiae*

Temp. (°C)	Hum. (%)	Number of eggs Observed	Daily egg hatchability (%) 2nd day	3rd day	4th day	5th day	6th day	7th day	Total egg hatchability (%)
27.82	79.53	1500	23.41	25.83	30.80	12.21	2.56	0.54	95.35a
28.76	80.95	1230	20.62	26.95	29.08	15.84	1.49	0.26	94.24a
LSD (5%)									3.43

+ Within the same column, means followed by same letters are not significantly different (P=0.05, LSD).

Our results are similar as those of To (2000) which studied *Pseudococcus citri* on coffee in Daklak and Gialai provinces. The egg hatchability was from 90.5% to 97.1%. Eggs began hatching on the second day, peaked on the fourth day, and stopped after the seventh day.

SUMMARY AND CONCLUSIONS

There were seven species of scale insects which include: 4 species belonging to the family Pseudococcidae, 2 species belonging to Coccidae and 1 species of Diaspididae, attacking coffee in Daklak province, Vietnam where the mealybug (*P. kraunhiae)* was the key insect pest. The life cycle of the mealybugs was 39.20 days at 27.82°C and 34.40 days at 28.76°C. Its oviposition capacity was 146.80 and 153.20 eggs per female and the percentage of egg hatchability was 95.35% and 94.24% under the conditions of average temperature of 27.82°C and 28.76°C, respectively. These data are very useful in predicting their emergence and damage so timely control measures can be implemented.

ACKNOWLEDGEMENT

The authors thank Prof. Qiao Ge-Xia, Group of Aphids Systematics and Evolution, Key Laboratory of Animal Systemtics and Evolution, Institute of Zoology, Chinese Academy of Sciences, for identifying the mealybugs, and Dr. Khanh, Entomological Division of Plant Protection Research Institute, Vietnam, for assistance in rearing the mealybugs. In addition, funding from the national project of science that was sponsored by Ministry of Science and Technology of Vietnam is acknowledged.

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