

Biology of Tortoise Beetle (*Coleoptera: Chrysomelidae*) on Sweet Potato

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Abstract

The biology three tortoise beetle of *Aspidomorpha confinis* klug, 1835, *Aspidomorpha equatoriensis* Borowiec, 1986 and *Conchyloctenia punctata* Fabricius, 1787 which commonly feed on sweet potato in Wolaita, southern Ethiopia was studied under laboratory condition. Adults and larvae were reared separately on fresh sweet potato leaves in plastic containers (28cm x 20cm x 8 cm) under ambient condition (21-28°C). Females *A. confinis*, *A. equatoriensis* and *C. punctata* had 16.8 ± 0.6 , 22.8 ± 0.7 and 20.1 ± 1.0 days pre-oviposition, 44.6 ± 3.2 , 28.3 ± 1.8 and 32.3 ± 2.0 days oviposition, and 15.7 ± 1.9 , 17.8 ± 1.9 and 10.7 ± 1.4 days post oviposition periods, respectively. *A. equatoriensis* lived longer with the longest oviposition period than others. *C. punctata* and *A. equatoriensis* lay their eggs in golden brown ootheca having 10-40 and 3-35 distinct oothecal membranes, respectively. The ootheca of *A. confinis* is a simple translucent creamy parchment, enclosing a single ovum. A female of *A. equatoriensis* laid a total of 615 eggs, *C. punctata* 327 and *A. confinis* 157. The average numbers of eggs per female per day were 14.4 ± 0.9 , 16.2 ± 0.3 and 40.6 ± 0.6 for *A. confinis*, *A. equatoriensis* and *C. punctata*, respectively. The incubation period of eggs was 10.3 ± 0.1 , 9.4 ± 0.1 and 11.2 ± 0.4 days in *A. equatoriensis*, *C. punctata* and *A. confinis* beetles, respectively. Larval period lasts 18.4 ± 0.2 , 18.4 ± 0.2 and 18.8 ± 0.6 days for, *A. equatoriensis*, *C. punctata*, and *A. confinis*, respectively. Early instars of *A. equatoriensis* and *C. punctata* are gregarious whereas *A. confinis* were solitary. Pupation was completed in 6.2 ± 0.1 , 6.5 ± 0.3 and 8.8 ± 0.2 days for *A. equatoriensis*, *C. punctata* and *A. confinis*, respectively. All the three species suffered the highest mortality during the first larval instars period. Net reproductive rates (R_0) of 44, 22 and 18 were recorded for *A. equatoriensis*, *C. punctata* and *A. confinis*, respectively. *A. equatoriensis*, *C. punctata* and *A. confinis* had total generation period of 50.7, 57.6 and 58.7 days, respectively, showing that they can complete 6 to 7 generations per annum.

Introduction

In Ethiopia sweet potato is produced mainly for human consumption. Southern Ethiopia is the main sweet potato growing region of the country where about 3% of the total crop area is annually allotted for sweet potato and more than 16% of the total volume of production of the country comes from this region (CSA 2002). Since sweet potato is mostly produced in low input agricultural system, losses due to insects may often reach 60–100% (Chalafant *et al.* 1990; Sorensen 2009). Several species of tortoise and leaf beetles (Chrysomelidae), the sweet potato hornworm (*Herse convolvuli*) and the sweet potato butterfly (*Acraea acerata*) are defoliators of sweet potato (Hill, 1983; Smit *et al.* 1997). Defoliators of sweet potato have previously been considered as pests of minor importance (Azerefgne, 1999). However, the sweet potato tortoise beetles have been economically important pest in parts of Africa (Hill 1983; Smit *et al.* 1997).

Over twelve species of insects have been recorded on sweet potato in Ethiopia. The most important among these are: the sweet potato leaf miner (*Bedellia somnulentella*), sweet potato weevil (*Cylas spp.*), sweet potato tortoise beetles (*Aspidomorpha spp.*), and sweet potato butterfly (*Acraea acerata*) (Adhanom *et al.*, 1985). So far nine species of tortoise beetles have been recorded on sweet potato in Ethiopia. These are: *Aspidomorpha apicalis* (Klug), *Aspidomorpha areata* Klug, *Aspidomorpha areata var nigripennis*, *Aspidomorpha cincta* Fabricius, *Aspidomorpha quadrimaculata* (Oliver), *Aspidomorpha tecta* (Beheman), conchylo tortoise beetles; (*Conchyloctenia hybrida* (Beheman), *Conchyloctenia illota* (Beheman), and *Conchyloctenia punctata* (Ferdu *et al.* 2009).

According to Ferdu *et al.* (2009) the importance of this beetle was minor, however during the survey period (2008) of this study it was a big problem even for Msc students who work their research on sweet potato under field conditions and they

sprayed more than two times. This is may be due to the sporadic nature of the insect. Adults are metallic and very colorful, ranging from gold to green to yellow (Ghidiu, 2006). Both larvae and adults of tortoise beetles are leaf eaters.

Tortoise beetles cause serious damage sporadically (Ghidiu, 2006). Newly set plants are most susceptible, and can be severely damaged or killed by feeding of this pest (Shepard *et al.* 1999; Ghidiu 2006; Bwana 2008). However, life history studies have not been conducted on any of the Ethiopian cassidinae species and little is known about their importance. Thus the study was conducted to understand the biology of three common species of tortoise beetles, *Aspidomorpha confinis*, *Aspidomorpha equatoriensis*, and *Conchyloctenia punctata* which commonly feed on sweet potato in southern Ethiopia. There is no related work on the biology of these species in other countries. The absence of this information may be due to lack of biological study about the species.

Material and Methods

Biology of *A. equatoriensis*, *A. confinis* and *C. punctata* was studied at Crop Protection Laboratory of Hawassa University, Ethiopia in 2008–2009. The adults and larvae of the three tortoise beetles were separately collected from sweet potato fields in Wolaita, southern Ethiopia where sweet potato is intensively cultivated in January 2008. Samples of adult tortoise beetles were preserved in 70% ethanol and sent to Zoological Institute, University of Wroclaw in Poland for identification. The remaining adults and larva collection were reared separately in plastic containers (measured 28 x 20 x 8 cm³) under ambient condition (temperature range 21–28°C). Complete randomized design was used for the experiment with three replication. The insects were fed on fresh leaves of sweet potato and checked daily for oviposition. Larvae of the three species were fed separately on leaves of sweet potato until they complete their life cycle.

Eggs laid on a particular day on leaves of sweet potato were kept separately in Petri-dishes (Ø9 cm) and the number of days needed for hatching was recorded. The development of the larvae was checked daily for molting to identify the number of instars and stadium by observing the insect every day. Duration of pupae was determined by monitoring daily emergence of adults. And the pupation took place in petri -dishes. The adult beetles of the three species which emerged on the same date were put together to get mating couple for subsequent studies. The beetles were frequently checked within a day to note the start of mating. The numbers of adult males and females were recorded separately for the tree species to determine the sex ratio. The sexes were identified while they mate, males of the tree species are relatively smaller in size which was clearly visible. Males of *C. punctata* have 15-17 and females 19-23 small black spots. The number of insects which survived at each stage was recorded for each cohort and the survivorship of each species was determined. The number of cohort used for each species and different parameters was different as indicated in the result part in the table for each parameter.

Paired Couple of the newly emerged adults were placed in Petri dishes (9mmØ) with fresh leaves of sweet potato and data were collected on mating, pre-oviposition, oviposition, post-oviposition, longevity and fecundity. Egg masses were collected and counted daily from each pair and kept for hatching. The adults were kept and offered leaves of sweet potato until they die. The number of larvae hatched from each egg batch was recorded and percent hatchability was determined (larvae/ootheca). When hatching ceased, all

ootheca were dissected under a microscope to determine the total; number of eggs per ootheca, and the number of fertile and unfertile eggs, respectively. Eggs with larval head capsules were classified as fertile (Jimenez-Perez et al.2004).

For each of the three species their fecundity (the total number of eggs laid per adult female), the mean number of eggs per female per days, fertility (the proportion of fertile eggs), and longevity (duration from emergence to death) were determined. The mean longevity of males and females was calculated, assuming that an individual died the day before it was found dead in the Petri dish (Devis and Van Lenteren, 2002). The net reproductive rate (R_0 , mean number of female offspring/female) $\sum l_x m_x$ were computed for the three beetles following the method used by Birch (1948).

Result and Discussion

Description of the life stages

Adult

Adults of *A. equatoriensis* have flattened, oval, and convex shaped bodies. They are gold or bronze in color. *C. punctata* adult are brown and larger in size than the other two species investigated. The elytra contain black spots on the transparent extension of the elytra and 19-24 small black spots for the female and 15-19 for the male. The adults of *A. confinis* are green and the smallest of the three. Female body size tends to be larger than male in all the three species (Figure 1).



Figure 1. Adults of *A. equatoriensis* (top left), *C. punctata* (below) and *A. confinis* (top right)

The elytra ground color of the three species changed remarkably with age. Though the changes were gradual, three color phases in *A. confinis* and *C. punctata* beetles and four in *A. equatoriensis* were recognized (Table 1). In the three species the last color phase coincides with sexual maturity. Knowledge on the color change can provide a rough measure for determining the age of adult in the field and can be used to take management actions before the beetles start mating and oviposit because mating was took place at the last color phase. *A. confinis* females started mating 12.8 ± 1.3 (N=44) days, *A. equatoriensis* 10.5 ± 0.5 (N=72) and 15.1 ± 0.3 (N=132) days for *C. punctata* after emergence. Females of *A. equatoriensis*, *C. punctata* and *A. confinis* first laid eggs at 4.5 ± 4.7 (N=72), 8.5 ± 0.5 (N=132) and 7.2 ± 0.6 (N=44) days after mating. Males and females of the three species repeatedly mate and spend long hours in copula.

Table 1. Change in ground color of elytra in *A. equatoriensis*, *C. punctata* and *A. confinis*

Species	Age (days after emergence)	Phase	Color
<i>A. equatoriensis</i>	0-2	I	White
	2-6	II	yellow-brown
	6-11	III	Dark brown
	11 and above	IV	Golden (bronze)
<i>C. punctata</i>	1-2	I	White
	2-5	II	Light yellow
	5 and above	III	Brown
<i>A. confinis</i>	0-2	I	White
	2-6	II	Light green
	6 and above	III	Green

The sex ratio (female to male) for *A. equatoriensis*, *C. punctata* and *A. confinis* were 1:1.5(N=61), 1:1.4(N=101) and 1:1.7(N=66), respectively.

The three species differed in their mean longevity. The highest and significantly ($P < 0.0001$, LSD) different female longevity were recorded for *A. equatoriensis* 75.6 ± 2.7 followed by *C. punctata* 69.2 ± 2.4 and *A. confinis* 63.1 ± 2.0 days. Significant ($P < 0.0001$) differences were also observed in longevity of the three male Tortoise beetles. *A. equatoriensis* beetle had the highest male longevity 71.5 ± 3.3 days followed by *C. punctata* 62.1 ± 2.5 and *A. confinis* 50.8 ± 2.3 (Table 2). Nakamura et al. (1989) also reported 63.8-88.4 days and 83.3-87.9 days of mean longevity for male and female

Aspidomorpha miliaris and *Aspidomorpha sanctaecrucis*, respectively. The three beetles differed significantly ($p < 0.05$) in the total number of days in ovipositional period (Table 3) *A. equatoriensis* had the longest oviposition period than *A. confinis* and *C. punctata*. The incubation periods of eggs were 10.3 ± 0.1 , 9.4 ± 0.1 and 11.2 ± 0.4 days for *A. equatoriensis*, *C. punctata* and *A. confinis* eggs, respectively and significantly different ($p < 0.05$) (Table 2). Incubation periods of cassidinae eggs had been shown to vary from species to species. For example *Aspidomorpha maculatissima* eggs hatched within 6-10 days (Hawkeswood, 1982), *Chrysomela scripta* 4-5 days (Neel et al. 1972) and *Melasoma populi* in about a week (Kadu et al. 1980).

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Table 2. Mean $\pm$ SE of the duration (days) of life stages and longevity of the three species under laboratory conditions at Hawassa University, 2008.

Name of Species	Stage and periods									Total Generation
	Adult longevity		Pre-oviposition	oviposition	Post-oviposition	Incubation period	Larval period	Pupal period	Developmental period	
	Male	Female								
<i>A. equatoriensis</i>	71.5 $\pm$ 3.3 (N=68)	75.6 $\pm$ 2.7 (N=68)	16.8 $\pm$ 0.6 (N=132)	44.6 $\pm$ 3.2 (N=69)	15.7 $\pm$ 1.9 (N=132)	10.3 $\pm$ 0.1(N=207)	18.4 $\pm$ 0.2 (N=246)	6.2 $\pm$ 0.1 (N=27)	33.9 $\pm$ 0.2 (N=225)	50.7
<i>C. punctata</i>	62.1 $\pm$ 2.5 (N=61)	69.2 $\pm$ 2.4 (N=61)	22.8 $\pm$ 0.7 (N=72)	28.3 $\pm$ 1.8 (N=60)	17.8 $\pm$ 1.9 (N=72)	9.4 $\pm$ 0.1 (N=246)	18.4 $\pm$ 0.2 (N=209)	6.5 $\pm$ 0.3 (N=194)	34.8 $\pm$ 0.3 (N=201)	57.6
<i>A. confinis</i>	50.8 $\pm$ 2.3 (N=54)	63.1 $\pm$ 2.0 (N=54)	20.1 $\pm$ 1.0 (N=44)	32.3 $\pm$ 2.0 (N=52)	10.7 $\pm$ 1.4 (N=44)	11.2 $\pm$ 0.4 (N=129)	18.8 $\pm$ 0.6 (N=135)	8.8 $\pm$ 0.2 (N=126)	38.6 $\pm$ 0.7 (N=128)	58.7

± = standard error of the mean

Table 3. Mean± of the duration (days) of life stages and longevity of the three species under laboratory condition at Hawassa University, 2008.

Name of specie	Stages and								
	Adult longevity		Pre oviposition	oviposition	Post oviposition	ation	Larvae period	Pupae period	Total developmental period
	Male	Female							
<i>A. equatoriensis</i>	71.5±3.3a	75.6±2.7a	16.8 ±0.6a	44.6±3.2a	15.7±1.9a	10.3±0.1a	18.4±0.2a	6.2±0.1a	33.9±0.2a
<i>C. punctata</i>	62.1±2.5b	69.2±2.4b	22.8±0.7b	28.3±1.8ab	17.8±1.9b	9.4±0.1b	18.4±0.2a	6.5±0.3ab	34.8±0.3ab
<i>A. confinis</i>	50.8±2.3c	63.1±2.0bc	20.1±1.0c	32.3±2.0c	10.7±1.4bc	11.2±0.4c	18.8±0.6a	8.8±0.2c	38.6±0.7c

* Means within column followed by the same letter are not significantly different at P = 0.005 (LSD)

Egg

C. punctata and *A. equatoriensis* laid their eggs enclosed in a translucent golden brown ootheca (Figure 2). There were 10-40 and 3-35 distinct oothecal membranes in *C. punctata* and *A. equatoriensis*, respectively. Each oothecal membrane contains four eggs in *C. punctata* and

three in *A. equatoriensis*. The total number of oothecae laid by *A. equatoriensis* ranged 10-130 per female while 1-24 by *C. punctata*. Females of *C. punctata* produced ootheca per day while it was 1.4 for *A. equatoriensis*. *A. confinis*, however, laid single eggs in an ootheca covered with translucent creamy parchment-like membrane.

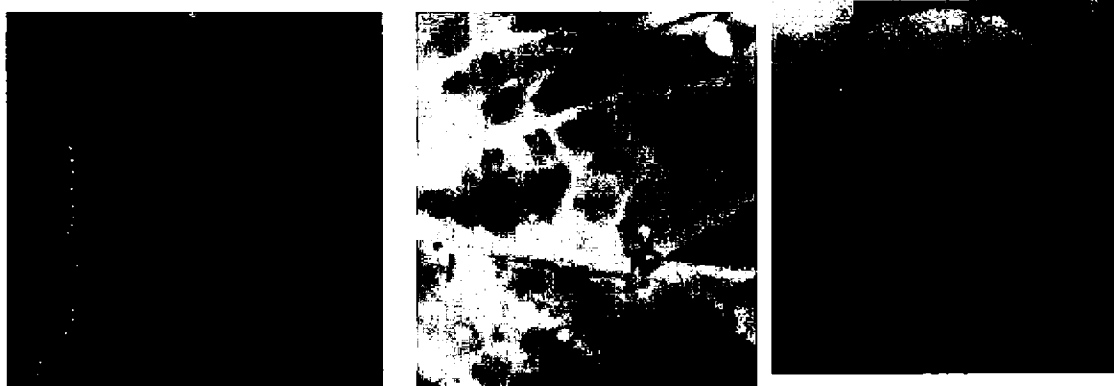


Figure 2. Ootheca of *C. punctata* (left), egg of ootheca of *A. equatoriensis* (middle) and egg of *A. confinis* (right)

Larvae

C. punctata larvae are light brown with black spines. *A. equatoriensis* larvae are flat, light green and possess lateral fleshy spine. The larva of *A. confinis* are green, flattened, with branched lateral fleshy green spines per segment (Figure 3). All the three species undergo 5 larval instars (Figure 4) that was determined by inspection of molting. Larvae of *A. equatoriensis* and *C. punctata* are gregarious at earlier instars. *A. confinis* larvae are strictly solitary. *Aspidimorpha areata* which lays single eggs in an ootheca is similar to *A. confinis* that is also solitary during their larval periods (Heron,

2007). The earlier larval instars (1st to 3rd), feed by scraping the surface of the leaf, whereas older instars (4th to 5th) consume the entire leaf, and create irregular holes between the leaf veins excluding the midribs. The holes formed by the larvae and adults are similar.

Larval periods were similar of the three species. The larval periods were within the range reported by Sanders et. al. (2003) for the cassidinae beetles of *Metritona bicolor* that lasted 17-23 and 17-26 days of *Cassida circumdata* (Vasquez and Sajise, 2002).



Figure 3. Larvae of *C. punctata* (left) *A. equatoriensis* (middle) *A. confinis* (right)

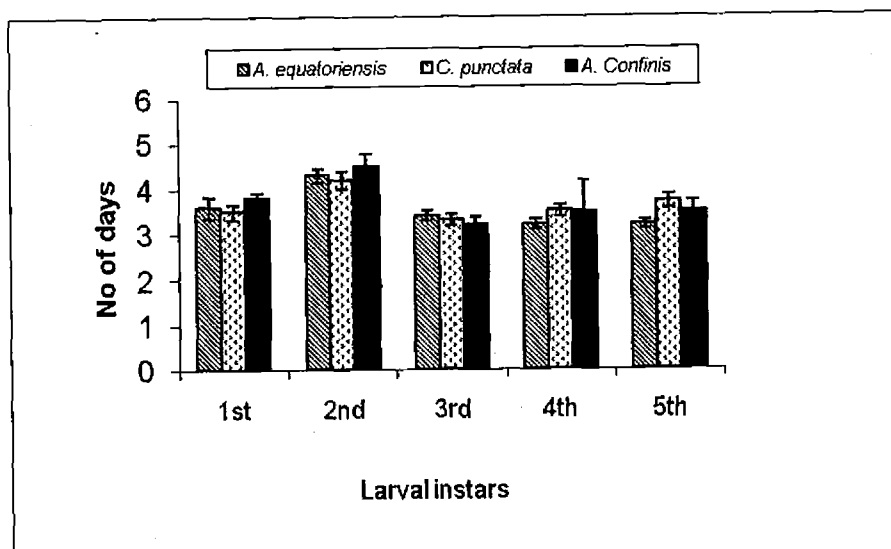


Figure 4. Duration of different larval instars of the three species tortoise beetle reared on sweet potato.

Pupae

Pupa of *C. punctata* is yellow with darker shade along the body margin. While that of Pupa of *A. equatoriensis* are light green. As it was the case

with larvae, the pupae of *A. equatoriensis* and *C. punctata* bear spines and cast exuviae of larva remain attached to the pupa. Pupa of *A. confinis* is green, oval and flattened (Figure 5).



Figure 5. Pupal stage of *C. punctata* (left), *A. equatoriensis* (middle) and *A. confinis* (right)

Pupal period lasted 6.2 ± 0.1 , 6.5 ± 0.3 and 8.8 ± 0.2 days for *A. equatoriensis*, *C. punctata* and *A. confinis*, respectively. The pupation periods are within the range reported for other cassidinae. Golden tortoise beetle, *Charidotella bicolor* completes pupation between 7-15 days (Ghidui, 2006) *circundata* and *miliaris* 4-8 and 7 days, respectively (Vasquez and Sajise, 2002).

The total life cycle (egg to adult) lasted 33.9 ± 0.2 , 34.8 ± 0.3 and 38.6 ± 0.7 days for *A. equatoriensis*, *C. punctata* and *A. confinis*, respectively, which were comparable to other cassidinae. *Charidotella* (= *Metriorhina*) *bicolor* required about 40 days to develop from egg to adult (Capinera, 2007) and *Aspidomorpha areata* 37-42 days (Heron 2007). Total generation times (egg to egg) were about 51 for *A. equatoriensis*, 58 for *C. punctata* and 59 days

for *A. confinis* which suggested that the three beetles can complete about 6 generations per annum if food and other environmental conditions are not limiting.

Survivorship

The three cassidinae beetles differed in their survival rate in which *A. confinis* had the highest (30%) (Figure 6). *C. punctata* and *A. equatoriensis* had almost similar survival. For all the three species the highest mortality was recorded during the first larval instars period. The survivorship follow type III curve characterized by high infant/juvenile mortality (Begon *et al.* 1990). This survivorship curve shows at what age death rates are high and low.

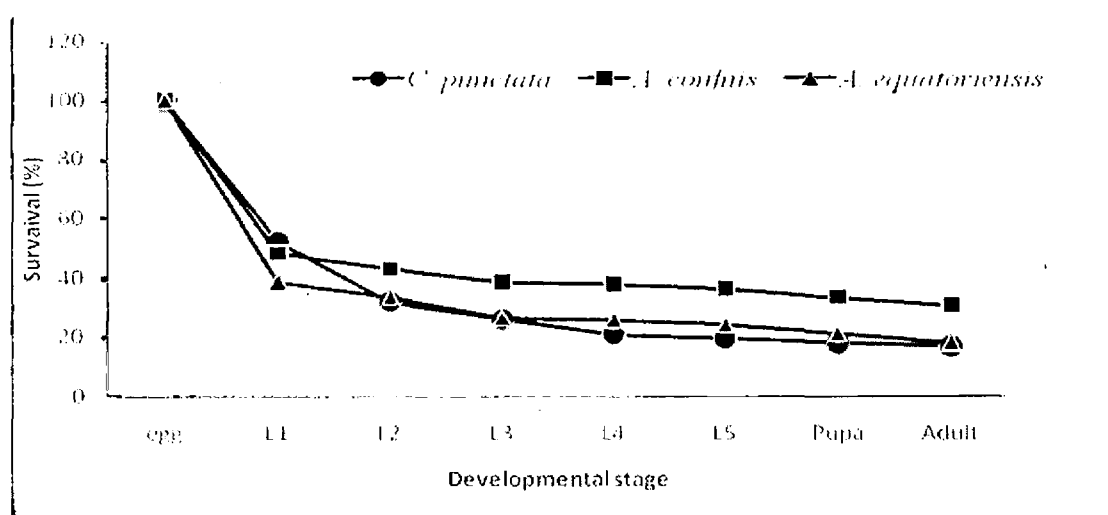


Figure 6. Survivorship of the three tortoise beetle species reared on sweet potato leaves under laboratory conditions.

Population attributes

The gross fecundity (total number of eggs laid per female) differed considerably ($p < 0.0001$) among the three tortoise beetles species (Table 4). A female of *A. equatoriensis* laid a total of 615.5 ± 48.6 , *C. punctata* 327.1 ± 24.7 and *A. confinis* 157.1 ± 13.6 eggs. The average numbers of eggs

per female per day were 14.4 ± 0.9 , 16.2 ± 0.3 and 40.6 ± 0.6 for *A. confinis*, *A. equatoriensis* and *C. punctata*, respectively (Table 4). The study showed that fecundity is negatively correlated with longevity in *A. equatoriensis* ($r = -0.5$, $P < 0.0001$) and *A. confinis* ($r = -0.7$, $P < 0.0001$).

Table 4. Reproductive potential of the three tortoise beetles reared on sweet potato leaves under laboratory conditions.

Attributes	<i>A. equatoriensis</i>	<i>C. punctata</i>	<i>A. confinis</i>
Total number of oothecae per female	53.2 ± 4.2 (N=56)	8.0 ± 0.60 (N=55)	157.1 ± 13.6 (N=61)
No of oothecae per day	1.4 ± 0.03 (N=81)	1.0 ± 0.0 (N=55)	
No. of eggs /ootheca	11.5 ± 0.2 (N=1824)	40.6 ± 0.6 (N=895)	
Total no. of eggs /female	615.5 ± 48.6 (N=56)	327.1 ± 24.7 (N=55)	157.1 ± 13.6 (N=61)
No. of egg /female/day	16.2 ± 0.3 (N=56)	40.6 ± 0.6 (N=55) (N= 61)	14.4 ± 0.9 (N= 61)

Eggs of *C. punctata* had the highest hatchability (51.9%) followed by *A. confinis* (48%) and *A. equatoriensis* (38.3%). Dissection of all the ootheca masses revealed that *A. equatoriensis* and *C. punctata* had some fertile eggs in the oothecae that failed to hatch. As a result, the fertility of *A. equatoriensis* (52.9±0.7%) was slightly higher than the hatchability while the increase was very high for *C. punctata* (81.8±0.9%). Fertility and hatchability did not differ in case of *A. confinis*.

The beetles had high net reproductive rate (Ro) of 44, 22 and 18 for *A. equatoriensis*, *C. punctata* and *A. confinis*, respectively. The capacity of multiple generation per annum coupled with the high biotic potentials indicate that the three species could build population densities of damaging levels with a couple of generations in absence of limiting factors like natural enemies, food and weather.

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