

CHEMICAL ANALYSIS OF CERTAIN CONSTITUENTS OF THREE PREY SPECIES OF THE PREDATOR, *CRYPTOLAEMUS MONTROUZIERI*

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Abstract

The predator, *Cryptolaemus montrouzieri* Mulsant, (Coleoptera: Coccinellidae) is an important predator of mealybugs (Hemiptera: Pseudococcidae). It may be fairly, considered a valuable biocontrol agent against mealybugs because other biological control methods proved ineffective due to the waxy coat that covers insect body in addition to its feeding habits. To shed light on the effect of the chemical constituents of a prey on the biology of its predator, certain chemical analyses were carried out on the tested preys to estimate the protein, carbohydrate, lipids, sodium, potassium, and calcium contents. The present work dealt with the host preference of the predator, *C. montrouzieri* by using chemical analysis of certain constituents (protein, carbohydrates, lipids, calcium, sodium together with the Potassium) of *Planococcus citri* (Risso) (Hemiptera : Pseudococcidae) eggs, *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) eggs and *Schizaphis graminum* (Rondani) (Hemiptera: Aphididae) nymphs. Chemical analysis of certain constituents of the tested preys of the predator *C. montrouzieri* showed that *P. citri* eggs had the highest total carbohydrates (31.37mg/g), total lipids (8.81 mg/g) and potassium content (1.3179 /mg), compared to the other two prey species (*E. kuehniella* eggs and *S. graminum* nymphs). It could be finally concluded that the greatest constituents of carbohydrates, lipids and potassium in eggs of *P. citri* are mostly, the reason of its suitability for rearing the predator *C. montrouzieri*.

INTRODUCTION

Biological control with the coccinellid predator, *Cryptolaemus montrouzieri* Mulsant (Coleoptera: Coccinellidae) proved very successful in suppressing populations of the mealybugs (Hemiptera : Pseudococcidae) . A single predator can consume 900-1500 eggs or 300 nymphs of the mealybug *Maconellicoccus hirsutus* (Green) (Hemiptera : Pseudococcidae) during its development. This coccinellid could be reared in large numbers on *M. hirsutus* infesting pumpkin fruits. Releases of 1000-1500 predator adults /acre gave effective control within 2 months (Mani, 1988).

Baskaran *et. al.* (1999), studied the biology and predation of *C. montrouzieri* (at 29.4 - 32.1°C and 65 - 71% RH.) in association with two types of prey [*Planococcus citri* (Risso) (Hemiptera : Pseudococcidae) and *Dactylopius tomentosus* (Lamarck) (Hemiptera: Dactylopiidae)]. They found that *C. montrouzieri* Mulsant completed the development successfully with both prey species, but preferred *P. citri* to *D. tomentosus*. Feeding preference by *C. montrouzieri* on *P. citri* was twice that with *D. tomentosus* (150.83 and 72 mg, respectively).

Prabal and Balasubramanian (2000) found that, the larva of *C. montrouzieri* period was prolonged when reared on nymphs of aphids compared to mealybug nymphs. The larval period of *C. montrouzieri* was completed in 21.20, 20.00, 23.20 and 13.92 days when reared on *Aphis gossypii* Glover (Hemiptera : Aphididae) on cotton, okra, cowpea aphid and mealybugs, respectively.

Abd Alla (2011) found that, the adult of *C. montrouzieri* fed on average of 246 ± 13.09 eggs of *P. citri*, 132 ± 6.63 eggs of *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae), 7.9 ± 0.65 nymphs of *P. citri* or 5.8 ± 0.74 nymphs of *Schizaphis graminum* (Rondani) (Hemiptera: Aphididae). *S. graminum*. The order of predatory preference of adult predator on different prey species was *P.citri* eggs ($61.85 \pm 0.01\%$) > *E. kuehniella* eggs ($33.66 \pm 0.01\%$) > *P. citri* nymphs (2.06%) > *S. graminum* nymphs (1.54%). Therefore, it is quite safe to state that eggs of *P. citri* were the most preferable prey for *C. montrouzieri*, and the nymphs of *S. graminum* were the least preferred.

The aim of the present work was to study the prey preference of the predator, *C. montrouzi* relation to the chemical constituents (protein, carbohydrates, lipids, calcium, sodium together with the Potassium) in three prey species (*P.citri* eggs, *E. kuehniella* eggs and *S. graminum* nymphs) .

MATERIALS AND METHODS

1. Rearing of prey species

a. Mealybug, *Planococcus citri*

The prey was obtained from infested croton ornamental plants, *Codiaeum variegatum* (L.) at Giza region and reared on pumpkin fruits under laboratory conditions ($27 \pm 2^\circ\text{C}$ and 60-70% R.H.) in the *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) mass production laboratory, Faculty of Agriculture, Cairo University. The pumpkins were selected with ridges, grooves and with a small stalk which makes handling very easy. Fruits were cleaned with running water to get rid of dust. Ovisacs of the mealybug were placed over the pumpkin for about 48 hours.

Infested pumpkins were kept on a plastic stand in wooden cages, with glass sliding front and cloth on other sides. After emergence from ovisacs, crawlers settled on all sides of fruits and developed into fully mature mealybugs in 30-40days. This technique of propagating mealybug culture on pumpkin fruits has been standardized by Chacko *et. al.* (1978) and Singh (1978).

b. *Ephestia kuehniella*

Eggs of *E. kuehniella* were obtained from the stock culture of *Chrysopa* Mass Rearing Unit, Faculty of Agriculture, Cairo University. The pyralid, *E. kuehniella* was mass reared at 23°C. Batches of 900 eggs were distributed upon the rearing tray containing crushed wheat. Rearing started in the laboratory at 23°C for 30 days, then at 10°C for 30 days and at 23°C for the last 5 days until moths' emergence. Newly emerged adults were collected daily, knocking them down by passing carbondioxide gas. The recovery was quite rapid and allowed to put adults in reproduction cylinder. Eggs produced by this colony of adults were divided into two groups, one group used for maintaining the colony, and the other as food supply for the predator.

c. *Schizaphis graminum*

Nymphs of *S. graminum* were obtained from the stock culture of "*Chrysopa* Mass Rearing Unit", Faculty of Agriculture, Cairo University. Rearing was carried out in chambers conditioned at 19°C and 16h light/day. Wheat seedlings were grown in Plexiglas boxes (30×13×10cm) containing humid pine tree sawdust (previously soaked in water for 24hours).The use of pine tree sawdust previously soaked was to avoid fermentation of material that could inhibit wheat growth. The humid sawdust was placed and compacted to half the height of the box so as to provide the future plants with an adequate substrate for rooting, the remaining area of the box was covered with a non compacted layer of wet sawdust to facilitate germination of seeds and penetration of seedlings. For germination, dry seeds were placed in water for 24 hours under the laboratory conditions mentioned above and then planted between the two sawdust layers. The boxes were kept at 19°C and germination occurred one week after seeding. When the young plants reached 2 cm hight, those were artificially infested with aphids through cut offs. Graminum infested of older green pea plants.. One box from the colony allowed infestation of 2 to 2.5 new boxes that were then placed in muslin cages to prevent any other infestation.

Chemical analysis

The prey's contents of protein, carbohydrates, lipids, calcium, sodium and potassium were analyzed as follows:

- 1- Total soluble proteins (mg/g body weight): determined by the method of Bradford (1976).

- 2- Total soluble carbohydrates (mg/g body weight): determined by the method previously followed by Singh and Sinha (1977).
- 3- Total soluble lipids (mg/g body weight): estimated according to Knight *et. al.* (1972), by using phosphor aniline reagent.
- 4- Calcium, Sodium and Potassium contents: Samples were digested using advanced microwave digestion system. ETHOS1 and the measurement on the device ICP Spectrometer (ICAP 600 series, Thermo Scientific.)

Statistical analysis

Data were analyzed by multivariable analysis (ANOVA) using Mstat-C program. When significant differences ($P < 0.05$) were calculated, the least significant difference (LSD) test was used to separate the mean values according to Steel and Torrie (1981).

RESULTS AND DISCUSSION

Data revealed that *E. kuehnielle* eggs contained the greatest total amount of protein (8.7mg/g), followed by *P. citri* eggs (7.69mg/g), and lastly by nymphs of *S. graminum* (4.69mg/g) (Table 1). Statically, significant deviations existed between the protein contents estimated in the different tested preys while, the highest carbohydrate and lipid contents were estimated in *P. citri* eggs, (31.37 and 8.81 mg./g body weight), being significantly higher than those in *E. kuehniella* eggs (6.96 and 3.32 mg./g) and also in bodies of *S. graminum* nymphs (14.75 and 1.22mg./g body weight, respectively) (Table, 1).

As for calcium, potassium and sodium contents in the three prey species, those were nonsignificantly different in cases of calcium (0.50, 0.458 and 0.537 g/100g body weight of *P. citri* eggs, *E. kuehniella* eggs and *S. graminum* nymphs, respectively) and sodium (0.212, 0.234 and 0.424g/100g, respectively) the highest potassium content (1.317g/100g) was estimated in *P. citri* eggs, being no significantly higher than that estimated in *E. kuehniella* eggs (1.034g/100g) and significantly higher than that estimated in *S. graminum* nymphs (0.738g/100g body weight) (Table, 2).

On basis of the aforementioned data, it could be stated that the greatest constituents of carbohydrates, lipids and potassium in eggs of *P. citri* compared to other tested preys, may be considered as the main reason of its suitability for mass-rearing of the predator *C. montrouzieri*.

According to data recorded by Attia *et al* (2011), the eggs of *P. citri* were found the most adequate prey for rearing *C. montrouzieri* predator. In association with this prey,

adult female deposited the greatest number of eggs (1049.7 eggs/female), coincided with the highest daily rate of reproduction (10.86 eggs / day/female). On this prey, the adult female lived a comparatively longest period (104.5 days), of which 97.9 days was reported as an oviposition period. The larval stage lasted 20.27 days with lowest rate of mortality (6.0%). By feeding on *E. kuehniella* eggs, the larval stage lasted a shorter period, (16.70 days). Larval mortality was higher (38.0%). Females produced lower amount of eggs (695.7eggs/female), throughout a longer oviposition period of 104.4 days. Fewer daily rate of reproduction was reported (6.7 eggs/ female) . When using *S. graminum* nymphs as a prey, the same authors found that the longevity of adult female was the shortest (45.30 days). Female produced also the smallest amount of eggs (89.50 eggs) throughout the shortest oviposition period (34.30 days).While the larval mortality was the highest (42%). In the same trend, Oonincx and Dierenfeld (2011) studied the chemical composition of eight invertebrate species and evaluated their suitability as alternative preys. Dry matter (DM), crude protein, crude fat, neutral detergent fiber, acid detergent fiber, ash, macro and trace minerals, vitamins A and E, and carotenoid concentrations were quantified. Significant differences were found between species. Crude protein content ranged from 38 to 76% DM, fat from 14 to 54% DM, and ash from 2 to 8% DM. In most species, calcium:phosphorus was low (0.08–0.30:1). Vitamin E content was low for most species (6–16 mg/kg DM). The retinol content, as a measure of vitamin A activity, was low in all specimens, but varied greatly among samples (0.670–886mg/kg DM). The data presented can be used to alter diets to better suit the estimated requirements of insectivores in captivity.

Table 1. Proteins, carbohydrates and lipids contents (mg/g body weight) in different tested preys.

Chemical Constituents	Preys		
	<i>P. citri</i> Eggs	<i>E. kuehniella</i> eggs	<i>S. graminum</i> nymphs
Total proteins	7.69 ± 0.13 b	8.70 ± 0.03 a	4.69 ± 0.10 c
Total carbohydrates	31.37 ± 0.13 a	6.96 ± 0.01 c	14.75 ± 0.05 b
Total lipids	8.81 ± 0.01 a	3.32 ± 0.01 b	1.22 ± 0.01 c

Averages having the same letter in the same row are not significantly different at 0.05 % probability level.

Table 2. Calcium, potassium and sodium contents (g/100g) in different tested preys

Chemical Constituents	Preys		
	<i>P. citri</i> eggs	<i>E. kuehniella</i> eggs	<i>S. graminum</i> nymphs
Calcium	0.500 a	0.458 a	0.537 a
Potassium	1.317 a	1.034 ab	0.738 b
Sodium	0.212 a	0.234 a	0.424 a

Averages having the same letter in the same row are not significantly different at 0.05 % probability level.

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التحليل الكيمياءى للمكونات الأساسية لثلاث أنواع من فرائس المفترس كريبتوليمس مونتروزيرى

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تم إجراء تحليل كيمائى لثلاثة أنواع من الفرائس المختلفة للمفترس الحشرى كريبتوليمس مونتروزيرى وهى بيض بق الموالح الدقيقى، بيض فراش دقيق البحر المتوسط وحوريات من الغلال لتقدير كمية البروتين، الكربوهيدرات، الدهون الصوديوم، الكالسيوم والبوتاسيوم فى مجمع المعامل النوعية بكلية الزراعة جامعة القاهرة ومعهد بحوث وقاية النباتات فى محاولة لتفسير تفضيل المفترس كريبتوليمس لبيض البق الدقيقى مقارنة بباقى الفرائس. وقد أوضحت التحليلات الكيمائية للفرائس المستخدمة أن بيض البق الدقيقى يحتوى أعلى كمية من الكوبوهيدرات (31.37مجم/جم) ، الدهون(8.81مجم/جم) والبوتاسيوم (1.317جم/100جم) وقد يكون هذا أحد العوامل الأساسية لتفضيل المفترس لهذه الفريسة، مقارنة ببيض فراش دقيق البحر المتوسط وحوريات من الغلال.