Freeze-dried artificial diets for three species of *Chilocorus* Ladybirds

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ABSTRACT

Artificial diets were developed for coccinellid beetles of the genus *Chilocorus*, which prey on armoured scale insects. Comparisons were made between 3 insect diet bases; bee brood, wasp brood and pupae of light brown apple moth, *Epiphyas postvittana*, with various additives. A freeze-dried wasp brood-based diet was found to be the most successful for rearing the larvae of these ladybirds. Adults of *C. infernalis* reared through the larval stages on scale insects, were able to oviposit normally when fed the artificial wasp brood diet, but *C. bipustulatus* and *C. cacti* did not oviposit readily unless fed the natural host scale insects.

Keywords: Freeze-dried artificial diet, *Chilocorus*, armoured scale, wasp brood, bee brood, *Epiphyas postvittana*.

INTRODUCTION

Beetles in the genus *Chilocorus* (Coleoptera: Coccinellidae) are generalist predators of armoured scale insects (Homoptera: Diaspididae). Three species of the genus, *Chilocorus bipustulatus* Linnaeus, *C. cacti* Linnaeus and *C. infernalis* Mulsant were reared in the laboratory for release as biological control agents against armoured scale insect pests. The host scale culture, (a mixture of greedy and latania scales, *Hemiberlesia rapax* (Comstock) and *H. lataniae* (Signoret)), normally used to rear the ladybirds was in short supply, so a supplementary artificial diet was sought. Initially, several agar-based diets were tested, but without success. Many researchers have used various species of pulverized insects for rearing ladybirds; for example, Okada & Nijima (1977) used bee brood (the larvae or pupae of honeybees *Apis mellifera* L.), blended, freeze dried and then pulverized as a diet for 17 species of ladybirds. Bain *et al.* (1984) used the larvae of potato tuber worm *Phthorimaea operculella* (Zeller) and Huhu grubs (larvae of *Prionoplus reticularis* White) as the bases for 2 freeze-dried diets, in the laboratory rearing of the predatory coccinellid *Cleobora mellyi* Mulsant for a biological control programme. Smirnoff (1958) varied a diet to suit coccinellid species with different feeding requirements, by including the natural prey as pulverized dry insects.

This paper reports investigations comparing 3 artificial diets, based on pupae of light brown apple moth *Epiphyas postvittana* Walker, pupae of the wasp *Vespula germanica* (F), and the larvae or pupae of honeybees *Apis mellifera* L. respectively. A regular supply of these 3 insects was available which was a factor in selecting them for our diet evaluations.

METHODS

Diet preparation and composition

For bee brood diets, mature larvae and young pupae with white to light purple eyes were selected as being most suitable and extracted whole from their combs. These contain the greatest amount of protein without having become substantially sclerotised. For wasp brood diets, only whole, newly formed pupae (pale and without sclerotised parts) were selected. Wasp larvae were avoided as they do not evacuate their gut contents until just prior to pupation and could introduce contaminants into the diet. For *E. postvittana* diets, young green to light brown pupae were selected. Brood or pupae were mashed in a blender, additives were calculated as a percentage of the wet weight of mashed brood, and the mixture was freeze dried. The resultant cake of freeze-dried artificial diet was pulverised into a powder and stored at 4°C.

There were four additives in each diet; (1) natural wheatgerm, reground, (2) Difco TC yeastolate®, yeast extract powder, (3) vitamin C (L-Ascorbic acid), (4) royal jelly, 100 mg capsules (contents only used).

Table 1: Composition of artificial diets.

				Additives	
	Insect Base	*Wheatgerm*	*Yeastolate*	*Vitamin C*	*Royal Jelly*
Diet 1	Bee Brood	10%	0.5%	0.2%	2%
Diet 2	*Epiphyas postvittana* pupae	10%	0.25%	0.2%	2%
Diet 3	Wasp brood	10%	0.25%	0.2%	2%

Larval rearing

Adult *Chilocorus bipustulatus* and *C. cacti* did not oviposit readily when fed artificial diets and were given instead the natural host scale culture (a mixture of latania and greedy scales) reared on pumpkins. Eggs were laid on small strips of double muslin gauze placed on the scale-infested pumpkins. These egg sheets were collected 3 times per week and kept in airtight pots until the larvae hatched, when they were transferred to scale-infested pumpkins. After a minimum of 2 days feeding on scales, larvae were then reared in Dixie® cups (unwaxed cardboard) with a water wick after the method of Bain *et al.* (1984) at

25°C and 60% relative humidity. Between 8 and 10 larvae were transferred to each cup, the floor of which had been scratched into grooves and onto which the powdered artificial diet was sprinkled. Crumpled tissue papers provided hiding places and moulting sites and helped to reduce cannibalism. The Dixie® cups were cleaned out at least once a week, the old diet powder being removed and replaced with fresh, and the tissues and water wick being removed, and replaced with clean ones. Larvae were allowed to pupate in the Dixie® cups and any remaining diet powder was carefully brushed out to avoid mould contamination.

Diet experiments

It was found that if newly hatched larvae were fed the natural host scale for at least 2 days, before being fed artificial diet, their survival and subsequent development was enhanced, compared with completely artificially fed larvae. Summaries are given for 1 test with *C. bipustulatus* larvae first fed on scale for 2 days before being fed the wasp and bee brood diets; and 1 test with *C. cacti* larvae first fed on scale up to 2nd and 3rd instar before being fed the 3 artificial diets (Tables 2 & 3). The percentage of adults obtained from the larvae in each treatment is given as the % yield, and the mean larval plus pupal period combined is given as MLP (± standard error) in days, from hatching of larvae to emergence of adults from pupae.

In an experiment to test the effects of larval diet on adult weight (Table 4), 3 groups of *C. cacti* larvae were fed different proportions of artificial diet and/or natural host scale. One group was reared entirely on scale, 1 group was reared entirely on artificial diet (the wasp brood diet), and the third group was fed on scale from hatching until second, third or early 4th instar, then transferred to artificial wasp brood diet and reared to pupation. The adults obtained from these larvae were weighed after emergence from their pupae and as soon as they had hardened.

Adult *C. infernalis* reared on scale insects as larvae, were fed the wasp brood diet when first imported and held in quarantine. Between 25 and 44 adults were kept in each of 3 boxes containing a water wick, the wasp brood diet sprinkled on the floor, and double muslin gauze egg laying strips. Eggs were collected every second or third day over a 2 week oviposition period.

RESULTS

The comparison of the wasp brood diet with the bee brood diet in Table 2 shows a greater yield of adults from larvae on the wasp brood diet, than from the bee brood diet, although the mean larval period is nearly the same for both diets.

The comparison of the 3 diets in Table 3, however, shows a marked difference in yields of adults. The wasp brood is clearly the best diet tested with a yield of 75% live adults, while the bee brood diet produced only 35% live adults, despite the early larval feeding on scale insects. The *E. postvittana* diet produced a yield of 55% live adults.

Results from the experiment to determine the effect of diet on weight of *C. cacti* (Table 4) showed that both males and females were significantly heavier when reared on the natural scales than on either artificial diet alone or on a proportion of scale and of artificial diet.

The best diet, wasp brood, is obviously not a complete substitute for armoured scale insects, and beetles of the 2 species reared through the larval stages on artificial diet and fed on it as adults did not lay any eggs. However, when *C. infernalis* adults were imported into quarantine and fed the wasp brood diet, they produced sufficient numbers of eggs to successfully start a colony of this species. These adults had been reared on scale as larvae. The mean rate of eggs/beetle/day of 0.4 over 13 days for the 3 boxes compares with 1.07 mean eggs/beetle/day collected from 120-150 beetles reared on scale and fed scale during oviposition period of 53 days. During the peak of this latter oviposition period, 3 eggs/beetle/day were recorded.

Table 2: Yield and mean larval plus pupal period (MLP) from the bee diet 1 compared with the wasp diet 3 using 1st instar *Chilocorus bipustulatus* larvae initially fed on scale for 2 days.

		Number of replicates	*larvae*	*pupae*	*adults*	*% yield*	*MLP*
Diet 1	Bee brood	3	28	8	8	28%	27 ± 1.1
Diet 3	Wasp brood	3	25	10	10	40%	28 ± 0.9

Table 3: Yield from diets with 3 different insect bases using *Chilocorus cacti* larvae first fed on scale up to 2nd and 3rd instar.

		Number of replicates	*larvae*	*pupae*	*adults*	*% yield*
Diet 1	Bee brood	2	20	7	7	35%
Diet 2	*E. postvittana*	2	20	11	11	55%
Diet 3	Wasp brood	2	20	15	15	75%

Table 4: Comparison of weights of male and female adult *Chilocorus cacti* fed on different proportions of artificial diet and host scale insects as larvae.

Number of males:females	*Type of diet*	*Mean weight males (SE)*	*% wt* reduction*	*Mean weight female (SE)*	*% wt* reduction*
27:29	Totally scale	14.6 mg (0.6)		15.4 mg (0.6)	
43:51	Scale + Artificial	10.6 mg (0.5)	27.4%	12.8 mg (0.5)	16.9%
33:43	Totally Artificial	11.0 mg (0.4)	24.6%	10.1 mg (0.5)	34.4%

* % weight reduction compared with totally scale fed
SE = standard error

DISCUSSION

There have been many attempts to find suitable artificial diets for rearing insects, and in particular for coccinellid beetles required for biological control releases. Okada & Nijima (1977) used their bee brood diet to rear 17 species of mostly aphid-feeding coccinellids. The 2 species of *Chilocorus, C, rubidus* and *C. kuwanae*, included in their report, were reared on the bee brood diet only through the larval stages. Smirnoff (1958) used an agar-based diet in which were included sugars, royal jelly, and pulverized dry insects, (the natural prey of the species), and reported 20 species reared, including *C. bipustulatus*. As we found agar-based diets unsuitable under our rearing conditions, we incorported the royal jelly in our freeze-dried diets.

Diets we tested earlier included a greater variety of additives, such as honey, sugar, trehalose sugar, linseed oil, and Wesson's salts; these were all eventually excluded as having no marked beneficial effect on the larval duration, survival and subsequent adult weight.

Wheatgerm and various forms of yeast have been incorporated in many different diets (Singh 1977; 1985), although not generally in diets for ladybirds. The use of wasp brood as a diet base has not been reported previously. We had a ready supply of wasp brood from wild nests. In addition, mass rearing of beetles appeared to yield better results with wasp brood, rather than bee brood or *E. postvittana* diets.

This work was carried out at a time when we were unable to raise sufficient armoured scale insects to feed our *Chilocorus* cultures. Fortunately, with the success of the diet we were able to continue rearing these ladybirds. The wasp brood diet has proved to be a useful supplementary diet with potential for extending the live-host scale cultures in rearing programmes for scale-feeding coccinellids.

ACKNOWLEDGMENTS

We would like to thank Dr P. Singh for his suggestions on the use of lepidopteran pupae for artificial diets.

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