

CULTURE OF TERRESTRIAL SLUGS AND SNAILS (GASTROPODA):  
ACCEPTANCE AND SUITABILITY OF SYNTHETIC INSECT DIETS

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## ABSTRACT

Natural products are usually used as a food source for the culture of terrestrial molluscs. Better uniformity, and perhaps economy, could be introduced to the culture of molluscs by using synthetic diets. I assessed acceptance by various terrestrial molluscs of several insect diets developed for culture of caterpillars, and suitability for mollusc growth. Diet selection (preference) studies were conducted with 2 species of snails (Cuban brown snail, *Zachrysia provisoria*; Asian tramp snail, *Bradybaena similaris*) and 4 species of slugs (meadow slug, *Deroceras laeve*; grey fieldslug, *Deroceras reticulatum*; dusky arion, *Arion subfuscus*; Carolina mantleslug, *Philomyces carolinianus*). The molluscs differed in their preference for insect diets, though tobacco hornworm diet and a bean-based diet often used for cutworms usually were not preferred, whereas gypsy moth, diamondback moth, and spruce budworm diets tended to be more preferred. Short-term (7-wk) developmental (weight gain) studies were conducted using *Z. provisoria*, *A. subfuscus*, *P. carolinianus* and Florida leatherleaf, *Leidyula floridana*. These same diets demonstrated great differences in suitability for mollusc growth, and although one or more diets were suitable for most species, *L. floridana* performed poorly. Relative preference displayed by the molluscs for insect diets generally corresponded to performance on those diets. Long-term developmental studies were conducted for growth of *Z. provisoria* on gypsy moth, spruce budworm, and diamondback moth diets, and compared to development on a natural diet, Romaine lettuce. Snail weight gain followed a typical logistic growth curve over the course of the developmental period. Survival of *Z. provisoria* was high on all diets, and eggs were produced by snails developing on all diets. *Zachrysia provisoria* fed diamondback moth diet produced significantly heavier snails than those fed gypsy moth and spruce budworm diets, and weights equivalent to snails fed lettuce. This study demonstrates that some synthetic insect diets are suitable for growth of some terrestrial molluscs, but that preference and suitability vary among species.

Key Words: gastropods, pest snails, pest slugs, artificial diet, *Zachrysia provisoria*

## RESUMEN

Se suelen utilizar productos naturales como fuente de alimento para la cría de moluscos terrestres. Una mayor uniformidad y tal vez un menor costo económico en la cría de moluscos podrían ser introducidos mediante el uso de dietas sintéticas. Se evaluó la aceptación de varios moluscos terrestres a diferentes dietas de insectos desarrolladas para la cultura de gusanos, y su potencial para el crecimiento de moluscos. Se realizó un estudio de selección de dieta (preferencia) utilizando 2 especies de caracoles (el caracol marrón de Cuba, *Zachrysia provisoria* y el caracol vagabundo asiático, *Bradybaena similaris*) y 4 especies de babosas (la babosa del prado, *Deroceras laeve*, la babosa gris de campo, *Deroceras reticulatum*; la babosa arion oscuro, *Arion subfuscus* y la babosa manto de Carolina, *Philomyces carolinianus*). Los moluscos se diferenciaron en su preferencia por las dietas de insectos, aunque la dieta del gusano del tabaco y la dieta a base de frijoles que se usa frecuente para los gusanos cortadores no fueron preferidas, mientras que tienden a ser más preferidas las dietas de la polilla gitana, la polilla de la col y el gusano del brote del abeto. Se realizaron estudios de corto plazo (7 semanas) sobre el desarrollo (aumento de peso) de *Z. provisoria*, *A. subfuscus*, *P. carolinianus* y *Leidyula floridana*. Las dietas para estas especies demostraron grandes diferencias en su idoneidad para el crecimiento de moluscos, y aunque una o más de las dietas eran apropiadas para la mayoría de las especies, la dieta del *L. floridana* dio un resultado pobre. En general, la preferencia relativa hacia las dietas de los insectos mostrada por los moluscos, correspondieron a su desempeño sobre cada dieta. Se realizaron estudios del desarrollo de largo plazo sobre el crecimiento de *Z. provisoria* alimentada sobre la dieta de la polilla gitana, la dieta de la polilla de col y del gusano del brote del abeto, en comparación con su desarrollo bajo una dieta natural de lechuga romana. El aumento en el peso del caracol siguió una curva típica de crecimiento logístico en el curso del periodo de desarrollo. La supervivencia de *Z. provisoria* fue alta en todas las dietas, y los huevos fueron producidos por los caracoles en todas las dietas. Los *Zachrysia provisoria* alimentados con la dieta de la

polilla de col produjeron caracoles significativamente más pesados que los alimentados sobre la dieta del gusano del brote del abeto, y pesos equivalentes a los caracoles alimentados con lechuga. Este estudio demuestra que algunas dietas sintéticas para insectos son adecuadas para el crecimiento de algunos moluscos terrestres, pero que la preferencia e idoneidad de las dietas varían entre las especies.

Palabras Clave: gasterópodos, plagas caracoles, plagas babosas, dieta artificial, *Zachrysia provisoria*

Natural or semi-natural foods have long been the predominant means of providing nutrition for culture of terrestrial molluscs under laboratory conditions (Godan 1983; South 1992). Items commonly used for food include carrot (*Daucus carota* L.; Apiales: Apiaceae) (Stephensen 1961; Rollo 1988); cabbage (*Brassica oleracea* var. *capitata* L.; Brassicales: Brassicaceae) (Walker et al. 1999; Zotin 2007); lettuce (*Lactuca sativa* L.; Asterales: Asteraceae), bean (*Phaseolus vulgaris* L.; Fabales: Fabaceae), cabbage and potato (*Solanum tuberosum* L.; Solanales: Solanaceae) (Raut & Panigrahi 1988); alfalfa (*Medicago sativa* L.; Fabales: Fabaceae) and clover (*Trifolium* sp.; Fabales: Fabaceae) (Byers & Bierlein 1982); germinating wheat (*Triticum* spp.; Poales: Poaceae) (South 1982); and many other plants, particularly vegetables. Processed and partly defined diet media include guinea pig pellets (Rueda 1989), rabbit food (Faber et al. 2006), poultry feed (Carvalho et al. 2008), or natural products supplemented with casein or dried milk or fish food, and mineral supplements (especially lime) (Wright 1973; Stanislawski & Becker 1979; Cowie & Cain 1983; Godan 1983). Godan (1983) describes culture of snails and slugs in containers with soil and peat, and creation of near-natural environments including nooks and crannies for egg-laying. One possible exception of dependency on natural or seminatural diets is the work of Lazaridou-Dimitriadou et al. (1998), who successfully used a dehydrated diet for culture of *Cornu aspersum* (Müller 1774) (as *Helix aspera* (Müller); Pulmonata: Helicidae). Most of these techniques, while adequate for small-scale maintenance and culture, are not particularly conducive for large-scale culture, or experimentation under uniform conditions. Research could benefit commercial production of terrestrial gastropods (e.g., *Cornu aspersum* (Müller) [Pulmonata: Helicidae]; *Helix pomatia* L.) used as escargot, or could help advance mollusc pest management research. Chemical management and integrated pest management research projects often benefit from access to a reliable supply of test organisms, and synthetic diets may support such efforts.

Many synthetic diets have been developed for phytophagous insects, including some that are commercially available, pre-mixed, and easily prepared. It seems reasonable that some might be useful for mollusc culture. Here I describe studies to assess the acceptance and suitability by ter-

restrial slugs and snails of some synthetic diets developed for plant-feeding Lepidoptera.

Most molluscs are marine-dwelling, but some in the class Gastropoda (clade Stylommatophora, snails and slugs) have made the evolutionary transition to land. Though generally not plant pests, those that feed on higher plants tend to be defoliators, causing damage much like that which is caused by caterpillars. Similarly, damaging populations tend to be managed much like insects, using chemical pesticides (molluscicides). In many other respects, however, molluscs tend to differ from insects. In contrast to insects, they lack the external mouthparts and legs, have two pairs of antennae, bear their eyes on antennae, secrete mucus, grow almost continuously, and are hermaphroditic.

Snails and slugs, though superficially quite different from one another, are really quite similar to one another morphologically, anatomically, and ecologically. Slugs are simply snails with the shell reduced in size, or existing only internally, or absent. Both snails and slugs possess a finely toothed radula that serves to rasp away plant tissue. They use the muscular 'foot' as a means of locomotion. Their epidermis produces copious amounts of mucus, which is important in enhancing mobility. Growth is almost continuous until maturity (and sometimes after maturity), with no molting to interrupt their development. Their eyes are attached to the dorsal pair of antennae associated with the head. Most are simultaneously hermaphroditic, each individual possessing functional male and female sex organs simultaneously (South 1992; Barker 2001).

## MATERIALS AND METHODS

### Diet Material

Four commercially prepared agar-based insect diets developed for caterpillars of different insect families and dietary habits were purchased and compared to a very simple and commonly used agar diet containing beans. Three diets were acquired from Bio-Serve (Frenchtown, New Jersey, USA): diamondback moth, *Plutella xylostella* (Linnaeus) (Plutellidae); gypsy moth, *Lymantria dispar* (Linnaeus) (Lymantriidae), and spruce budworm, (*Choristoneura* spp.) (Tortricidae). Diet for tobacco hornworm, *Manduca sexta* (L.) (Sphin-

gidae), was acquired from Carolina Biological Supply, Burlington, North Carolina, USA. All contain wheat germ as the predominant component, but BioServe does not divulge the exact proportions so detailed comparisons are not possible. The bean diet was made according to the formula given by Shorey (1963) and differed from the wheat germ-based diets in using lima beans and dried brewers' yeast as the basic ingredients. The latter is commonly used for the culture of many caterpillars, particularly cutworms (Noctuidae). Diet was mixed according to directions, poured into flat pans, covered and stored at 7 °C until needed. In some studies, these synthetic diets were compared to a natural diet, Romaine lettuce (*Lactuca sativa* L. var. *longifolia*; Asterales: Asteraceae). In studies where Romaine lettuce was used as a dietary item, it was purchased twice per wk from a grocery store and refrigerated at 7 °C until used.

#### Preference

For preference tests, 2 species of snails [Cuban brown snail, *Zachrysis provisoria* (Pfeiffer, 1858) (Pleurodontidae); Asian tramp snail, *Bradybaena similis* (Férussac, 1821) (Bradybaenidae)] and 4 species of slugs [meadow slug, *Deroceras laeve* (O.F. Müller, 1774) (Agrolimacidae); grey field-slug, *Deroceras reticulatum* (O.F. Müller, 1774) (Agrolimacidae); dusky arion, *Arion subfuscus* (Draparnaud, 1805) (Arionidae); and Carolina mantleslug, *Philomycus carolinianus* (Bosc, 1802)] were evaluated. All specimens were field collected and cultured on raw Romaine lettuce at 22 °C until evaluation. The offspring of some easily cultured species (*Z. provisoria*, *D. laeve*, *P. carolinianus*) were also used, but were not fed synthetic diets.

Preference tests were performed by simultaneously providing disks of the aforementioned synthetic diets to the test animals in test arenas. The diet disks were punched from the diet pans using a 1 cm diam cork borer. Each disk typically weighed about 1.5 g, but varied slightly. One disk of each diet type was weighed and randomly positioned within the test arena on moist filter paper. The test arena consisted of a cylindrical transparent plastic container with a flat top and bottom. The top was tight fitting and there was almost no moisture loss from these containers. The test arena measured 8 cm high and 18 cm in diam. The disks were arranged in a circle within the test area, and were approximately equidistant from each other and the sides of the container. Preliminary tests demonstrated greatly different rates of consumption by the different mollusc species evaluated, so I adjusted the number of animals tested per container to allow for measurable consumption without depletion of diet material during the evaluation period of 7 d. The number

of animals evaluated per test arena was 1 for *P. carolinianus*, 2 for *Z. provisoria* and *A. subfuscus*, 6 for *B. similis*, and 10 for both *Deroceras* spp. After the period of evaluation, the diet disks were removed and individually re-weighed. Each preference assessment replicate consisted of 3 individual containers (samples) containing diet and test animals, and 3 control (check) containers consisting of diet disks arranged identically but without the test animals. Replicate tests were conducted over time, with each replicate separated by a wk and using different animals. The number of replicates varied among species, according to availability of test animals. The number of replicates was 4 for *A. subfuscus*, 6 for *D. reticulatum*, and 7 for each of the other species.

Diet preference was assessed based on consumption, and consumption was determined by change in weight from the initial weight. Weights were determined with a Mettler Toledo AL 104 balance. Weight change of the test diet disks was adjusted for weight change in the control disks using the formula:

$$\text{Corrected \% consumption} = \frac{P_t \pm P_{ck}^*}{100 \pm P_{ck}} 100$$

where  $P_t$  = % diet weight change in the presence of molluscs, calculated from the difference in weight after 7 d;  $P_{ck}$  = % diet weight change in the absence of molluscs for the same period of time; and where the sign was positive (+) when the control diet disk increased in weight and negative (–) when it decreased.

The percent consumption values were transformed to arcsin square root after being adjusted for weight changes of control diet. Adjustments never exceeded 10%, and usually were less than 5%. Consumption data were analyzed with two-way analysis of variance using GraphPad Prism software (GraphPad Software, Inc., San Diego California, USA) with mollusc species and diets as the sources of variation. Diet consumption within each mollusc species was compared statistically using Bonferroni's Multiple Comparison Test (GraphPad). Graphic presentation of consumption data uses untransformed data.

#### Weight Gain

Short-term studies were conducted to assess the potential of various terrestrial molluscs to develop satisfactorily on synthetic insect diets, with diet suitability being determined only by weight gain. Four species (*P. carolinianus*, *Z. provisoria*, *A. subfuscus*, and Florida leatherleaf [*Leidyula floridana* (Leidy, 1851)]) were reared on diets for 7 wk to assess growth potential. I reared 10 molluscs of each species on each of the 5 aforementioned synthetic diets, except in the case of *A. subfuscus*, where 2 diets were deleted because not enough slugs were available. The molluscs

of each species were approximately equal in age (weight) at the start of the study, and each individual was reared separately in a vented cylindrical 200 ml plastic container containing moist filter paper and a 1 cm<sup>3</sup> cube of synthetic diet. Food was replaced weekly, with the molluscs cultured at 22-24 °C and 14:10 h L:D. The molluscs were weighed weekly with a Mettler Toledo AL 104 balance.

Analysis of weight gain was determined by calculating % weight gain for each mollusc species and diet between the start of the study and wk 7. One-way ANOVA was used to assess the differences among diets provided to each mollusc species, with mean weight gain compared statistically using Bonferroni's Multiple Comparison Test from GraphPad Prism software (GraphPad Software, Inc., San Diego California, USA).

More detailed and prolonged assessment of weight gain was determined using *Z. provisorio* as a model organism and by measuring snail weight gain on each of 4 diets for the individual molluscs from hatch to maturity. The 3 synthetic diets most preferred by *Z. provisorio* (gypsy moth, diamondback moth, and spruce budworm) were compared to a diet of Romaine lettuce, a food almost universally accepted by plant feeding molluscs. As a check, 20 snails were also confined to containers with moist soil, but no test diet, because organic material (humic acid) in the soil could be a source of nutrition.

Long-term weight gain tests were conducted by culturing individual snails in vented cylindrical 500 mL plastic containers containing about 200 g of moist soil plus the test diet. Diets were presented in the form of 1 cm<sup>3</sup> cubes of synthetic diet, or 25-50 cm<sup>2</sup> of lettuce foliage. Diet material normally was changed at 3-4 d intervals, and the soil was replaced at monthly intervals. The snails were weighed at 3-4 d intervals with a Mettler Toledo AL 104 balance. The snails were cultured for 130 d at 22-24°C and 14 h photophase, until their weight seemed to stabilize, suggesting maturity. Each treatment consisted of 20 separate containers, each with one snail, and commenced with 2 d-old hatchlings.

Weight gain curves were plotted for each diet, and the mean weights were determined for each diet and compared using one-way ANOVA, with means compared statistically using Tukey's Multiple Comparison Test from GraphPad Prism software [GraphPad Software, Inc., San Diego California, USA]. Regression analyses were conducted on some weight gain curves, also using GraphPad Prism. Spearman rank correlation coefficients were calculated at several points in time to assess the association between snail weights over the course of their development, also using GraphPad Prism.

## Observations on Adults

After 130 d, about half of the snails ( $n = 8$  or 10) from each diet treatment, were paired with snails of similar size and confined in 500 mL cups ( $n = 4$  or 5 per diet) with moist soil about 5 cm deep. They continued to be fed the diet on which they were initially cultured, and checked twice per wk for egg production. The remaining snails ( $n = 9-10$  per diet) were placed individually into cups and treated in the same manner. These adult snails were maintained for 6 wk at 22-24 °C and a photophase of 14 h to determine if they would produce eggs.

## RESULTS

### Preference

Some synthetic insect diets were consumed significantly more by the molluscs ( $F = 42.76$ ;  $df = 4, 160$ ;  $P < 0.0001$ ). The pattern of consumption (Fig. 1) also differed significantly among mollusc species ( $F = 11.44$ ;  $df = 5, 160$ ;  $P < 0.0001$ ). The diet and species interaction was significant ( $F = 7.14$ ;  $df = 20, 160$ ;  $P < 0.0001$ ), so the results are discussed by species.

*Arion subfuscus* displayed strong dietary preferences, consuming significantly more spruce budworm diet than the tobacco hornworm, bean, and diamondback moth diets ( $P < 0.0001$  in each case). Consumption of the gypsy moth diet was intermediate. Gypsy moth diet consumption was significantly ( $P < 0.01$ ) less than spruce budworm

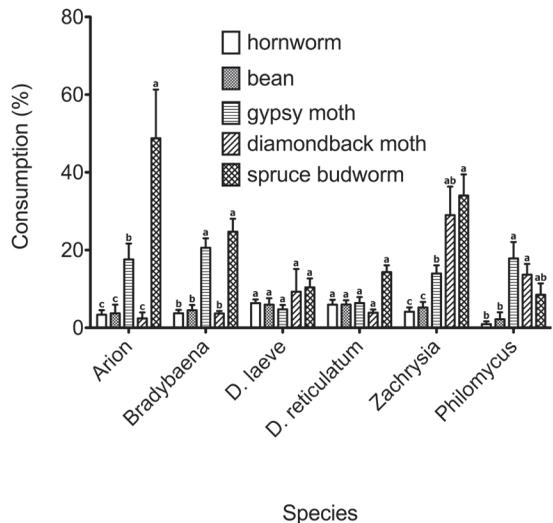


Fig. 1. Preference of 6 species of slugs and snails for 5 synthetic insect diets provided simultaneously in choice tests. Values shown are mean levels of diet consumption + SE. Mean values within a species topped by the same letter are not significantly different ( $P < 0.05$ ).

diet consumption, but significantly greater than tobacco hornworm and diamondback moth diet consumption ( $P < 0.01$ ) and bean diet consumption ( $P < 0.05$ ).

Similarly, *Bradybaena similaris* preferred the spruce budworm and gypsy moth diets. Spruce budworm diet consumption was significantly greater than bean and tobacco hornworm diet ( $P < 0.0001$  each) and diamondback moth diet ( $P < 0.001$ ) consumption. Gypsy moth diet consumption also was significantly greater than bean diet and tobacco hornworm diet ( $P < 0.001$  each) and diamondback moth diet ( $P < 0.01$ ) consumption. The consumption levels of the spruce budworm and gypsy moth diets did not differ significantly from one another.

No statistically significant differences were detected in the selection of diets by the 2 *Deroceras* spp., although as with *A. subfuscus* and *B. similaris*, consumption of spruce budworm diet averaged higher than consumption of any other diet.

*Zachrysis provisoria*, like the other molluscs, seemed to favor spruce budworm diet, and consumed significantly more than the tobacco hornworm and bean diets ( $P < 0.0001$  each) and the gypsy moth diet ( $P < 0.01$ ). Diamondback moth diet also was consumed readily, and did not differ significantly from consumption of either spruce budworm or gypsy moth diet, although consumption of diamondback moth diet significantly surpassed consumption of the bean and tobacco hornworm diets ( $P < 0.001$ ). Consumption of gypsy moth diet was significantly greater than tobacco hornworm diet ( $P < 0.05$ ), but not bean diet ( $P > 0.05$ ).

*Philomycus carolinianus* preferred the gypsy moth and diamondback moth diets over bean and hornworm diets ( $P < 0.001$ ). Spruce budworm diet was intermediate in preference, and consumption did not differ significantly from gypsy moth and diamondback moth diet, nor from bean and hornworm diet. On average, the preferences displayed by *P. carolinianus* were similar to most of the other molluscs, especially *Z. provisoria*.

#### Weight Gain

In 7-wk weight gain studies, all 4 mollusc spp. displayed statistically significant differences in

growth potential when provided with synthetic insect diets. Weight gains were much greater on some diets (Table 1). Although gypsy moth, spruce budworm, and diamondback moth diets generally maximized weight gain, none of the diets was very suitable for *L. floridana*; most of these slugs lost weight or died. There was some correlation between preference and short-term performance, but there was not an exact correspondence. For example, *A. subfuscus* consumed significantly more spruce budworm diet than gypsy moth diet in preference tests, but performed significantly better on gypsy moth diet. On the other hand, in both the preference and growth studies, these 2 diets exceeded the other 3 diets.

In long-term studies, survival of *Z. provisoria* was over 90% for each test diet. This was not the case with snail hatchlings that were denied test diet and provided access only to moist soil (Fig. 2). Initially they gained weight, apparently due to imbibition of water and organic matter (their excreta initially was dark), but soon displayed a steady loss of weight. This portion of the study was discontinued after 21 d because the snails began to perish. These data are well represented by a centered fourth order polynomial equation ( $R^2 = 0.1809$ ), which was superior to first and second degree polynomial equations.

In sharp contrast to the soil-only study, hatchling *Z. provisoria* snails provided with test diets gained weight rapidly and for a protracted period of time. Fig. 3 shows the weight change per d for snails fed diamondback moth diet over the 130 d of the study, expressed both in grams and percentage. The patterns were the similar for all diets, so data for only 1 diet are presented. Weight gain was rapid initially, averaging about 10% weight gain/d for the first 40 d of growth. Some individuals increased in weight by 50-100% in 3-4 d. As the snails became larger, their rate of weight gain decreased. Also, they seemingly underwent alternate periods of feeding and rest, resulting in erratic weight gain, and in some cases, periodic weight loss.

Increase in mean snail weights on all diets followed a typical logistic growth form (Fig. 4), though attaining different mean maximum weights (Table 2). I analyzed mean weights at d 120 and 130 because there is considerable varia-

TABLE 1. MEAN INCREASE IN WEIGHT (%) OF 4 MOLLUSC SPECIES AFTER FEEDING FOR 7 WEEKS ON VARIOUS SYNTHETIC INSECT DIETS. MEANS WITHIN A ROW FOLLOWED BY THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT ( $P < 0.05$ ).

	Insect diet				
	Gypsy moth	Spruce budworm	Diamondback moth	Bean	Tobacco hornworm
<i>Philomycus carolinianus</i>	493 ab	614 a	390 b	121 c	381 bc
<i>Zachrysis provisoria</i>	48 a	47 a	54 a	12 b	9 b
<i>Leidyula floridana</i>	1 a	9 a	0 b	0.1 b	0 b
<i>Arion subfuscus</i>	92 a	25 b	40 ab		

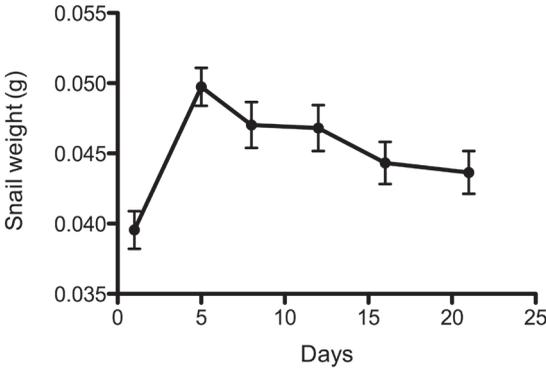


Fig. 2. Weights of *Zachrysia provisoria* hatchlings (mean + SD) for 21 d when provided moist soil but no lettuce or test diets for nutrition.

tion in attainment of maximum weight, and also analyzed the highest means weights attained by individual snails irrespective of date of attainment. There were significant differences in snail weight at d 120 ( $F = 19.4$ ;  $df = 3,75$ ;  $P < 0.0001$ ), at d 130 ( $F = 17.4$ ;  $df = 3,75$ ;  $P < 0.0001$ ), and individual snail maximum weights ( $F = 19.2$ ;  $df$

= 3,75;  $P < 0.0001$ ) attributable to diet. Snails fed Romaine lettuce attained a mean maximum weight of 8.07 g (+ 0.18; SD), with gypsy moth diet-fed snails attaining 7.73 g (+ 0.85; SD), diamondback moth diet-fed snails attaining 9.06 g (+ 0.81; SD), and spruce budworm diet-fed snails attaining 6.03 g (+ 0.85; SD). Mean weights on d 120 and 130 were slightly less (Table 3) due to asynchronous attainment of maximum weight by individual snails (Table 2). Using any of the 3 measures of weight assessment, diamondback moth diet produced significantly ( $P < 0.05$ ) heavier snails than the gypsy moth and spruce budworm diets. Lettuce was intermediate between diamondback moth and gypsy moth diets in terms of weight gain, usually not differing significantly from either. Spruce budworm diet was least suitable for snail weight gain, producing significantly ( $P < 0.05$ ) lower weights than the other diets.

Assessment of snail weights through time (Table 3) suggested that *Z. provisoria* size later in life was not strongly determined by weight early in life. For example, snail weight at d 2 was significantly correlated with weights at d 9 and 33, but not later in development. However, by the mid point of development (d 58), size at maturity seemed to be largely determined, as there were strong correlations between weights of snails at this point and thereafter.

Observations on Adults

Paired adult snails cultured on all diets produced eggs after mating. The sample size was too small to adequately assess the suitability of the different diets for egg production, but no eggs were produced by any snails held singly, and on average, 55% ( $\pm 34$ ; SD) of the snail pairs produced eggs within 6 wk of apparent maturity. Some of the paired snails produced a second batch of eggs, though it is not certain whether a single snail produced more than 1 clutch, or (more likely) whether each of the 2 snails produced a clutch. Thus, although some hermaphroditic molluscs self-fertilize, cross-fertilization in *Z. provisoria* is apparently advantageous, and probably necessary. Snails did not deposit eggs unless provided with soil.

DISCUSSION

It is not surprising, considering the different plant feeding habits and the natural geographic ranges inhabited by these molluscs, that they would have somewhat different feeding preferences when presented with synthetic insect diets. What is somewhat surprising, however, is that these diverse species displayed a relatively high degree of similarity in their preference for synthetic insect diets: spruce budworm and gypsy moth diets were almost always among the most

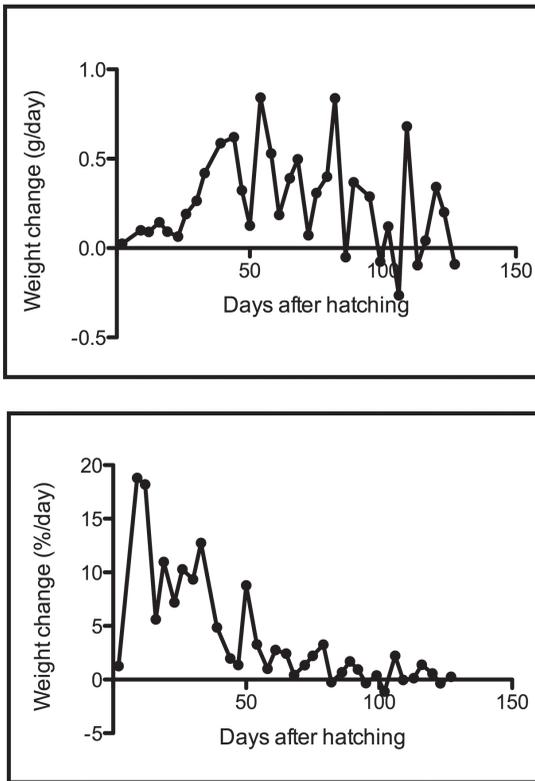


Fig. 3. Rate of weight change in *Zachrysia provisoria* fed diamondback moth diet from hatch to maturity, expressed in grams (upper panel) and percent (lower panel).

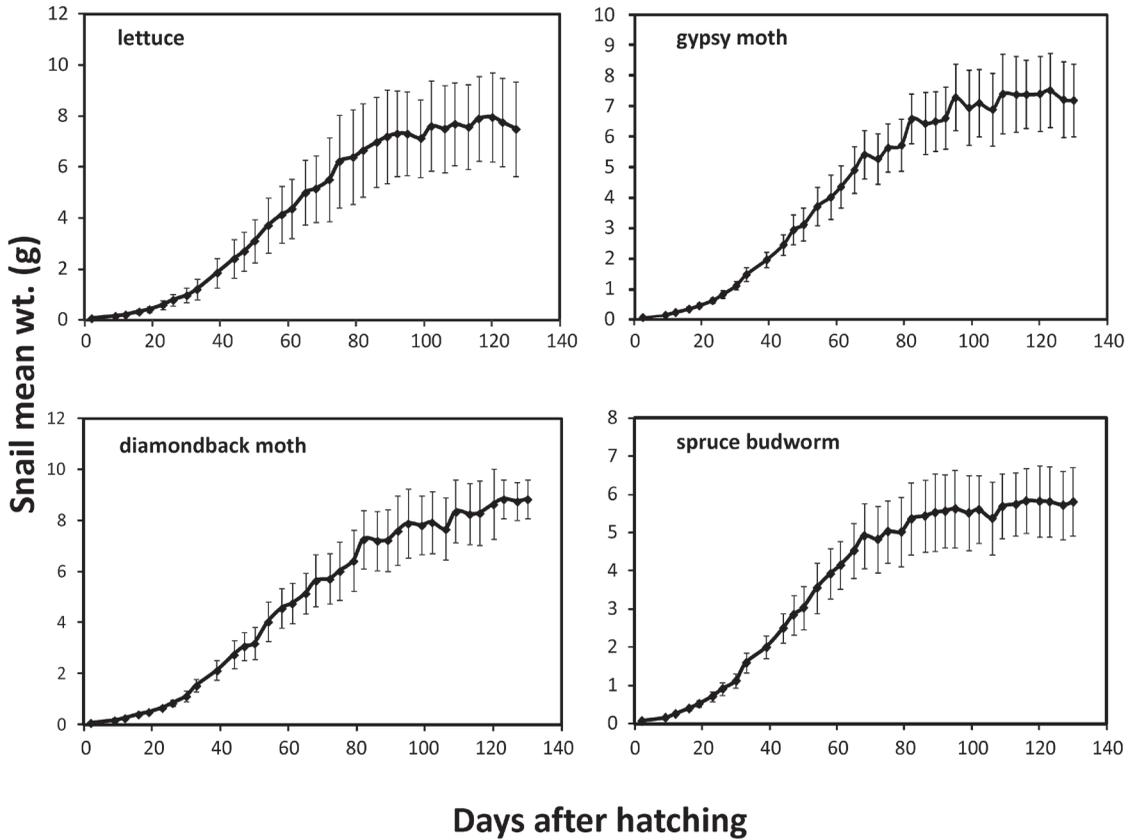


Fig. 4. Patterns of weight gain (mean + SD) by *Zachrysis provisoria* during a 130 d trial period when provided with lettuce or 1 of 3 preferred synthetic insect diets.

preferred diets. Even *Z. provisoria* and *P. carolinianus*, which are relatively anomalous in their ready acceptance of diamondback moth diet, also readily consumed spruce budworm and gypsy moth diets. Interestingly, these molluscs display quite different dietary preferences in nature, as *Z. provisoria* is a folivore, whereas *P. carolinianus* is considered to be fungivorous (Pilsbry 1948; Ingram 1949).

Erratic weight change in molluscs has long been documented. Howes & Wells (1934a, 1934b) reported that *H. pomatia* snails could display up to 50% weight change in a single d. They attribut-

ed this to changes in hydration, and demonstrated that the changes in amplitude were greater when animals had ready access to both food and water, and less pronounced if denied either food or water. These authors also demonstrated that up to half the weight change could be due to changes in environmental humidity. In this study, the *Z. provisoria* snails were continuously provided with opportunity to be in contact with moist soil so they were not likely to be stressed by low humidity. The intermittent increases and decreases in weight (Fig. 3) are more likely due to irregular periods of feeding, defecation and rest. Indeed,

TABLE 2. MEAN WEIGHTS (G) ACHIEVED BY *ZACHRYSLA PROVISORIA* DURING A 130 DAY TRIAL PERIOD WHEN PROVIDED WITH LETTUCE OR 1 OF 3 PREFERRED SYNTHETIC INSECT DIETS. ANALYSES WERE CONDUCTED OF MEAN WEIGHTS ON DAYS 120, 130, AND GREATEST WEIGHT ATTAINED BY INDIVIDUAL SNAILS. MEAN VALUES WITHIN A ROW FOLLOWED BY THE SAME LETTER ARE NOT SIGNIFICANTLY DIFFERENT ( $P < 0.05$ ).

Weight	Insect diet			
	Diamondback moth	Spruce budworm	Gypsy moth	Lettuce
120 d old	8.8 a	5.8 c	7.4 b	7.9 a,b
130 d old	8.8 a	5.8 c	7.2 b	7.4 b
Individual maximums	9.1 a	6.0 c	7.7 b	8.1 a,b

TABLE 3. SPEARMAN CORRELATION MATRIX FOR *ZACHRYSLA PROVISORIA* SNAILS OF VARIOUS AGES (DAYS AFTER HATCHING). VALUES SHOWN ARE R VALUES, WITH STATISTICAL SIGNIFICANCE SHOWN AS \*\* FOR  $P < 0.01$ , AND \*\*\* FOR  $P < 0.001$ .

Day	9	33	58	79	120	130
2	.497**	.461***	.187	.018	-.107	-.129
9		.394***	.133	.015	-.116	-.170
33			.601***	.353***	.125	.123
58				.802**	.598***	.542***
79					.778**	.708***
120						.948***

adult *Z. provisorio* snails commonly defecate 0.3-0.4 g per d, so that factor alone could account for considerable variation in weight. The presence of excreta and mucus also inhibits mollusc activity (Dan & Bailey 1982; Baur 1988; Garcia et al. 2006), and although there was only a single snail per container, the presence of mucus may have inhibited feeding. I could detect no decrease in feeding over the course of the 3-4 d between diet changes, however. The amplitude of the changes, though seeming large when portrayed as in Fig. 3, appears much less when plotted against the weight of the animal over time. Thus, we can see in Fig. 4 that there was a relatively consistent increase in weight until maturity. The 3 periods of growth found in many (but not all) slugs have been described as the infantile phase (initial period of rapid growth), juvenile phase (a subsequent period of slow growth), and the mature phase (a period of little or no growth, and punctuated by egg laying)(South 1992). The patterns of growth observed with *Z. provisorio* certainly correspond with this model of development.

The time to maturity of *Z. provisorio* fed different diets was similar despite different diets; there was little additional weight gain after about 100 d. However, the diets produced mature snails of different final mean weights, and diamondback moth diet promoted the greatest weight gain. Although there certainly was individual variation in development rate and eventual size, I did not see evidence of distinctly different groups of fast-, intermediate-, and slow-growing molluscs, as reported by Lazaridou-Dimitriadou et al. (1998) and Zotin (2007) in *Cornu aspersum* (Muller, 1774) (as *Helix aspera*) and *Deroceras reticulatum*, respectively.

Growth of some molluscs is thought to be affected by egg size, or by size at hatching (South 1992; Moran & Emlet 2001). Egg and juvenile offspring size is sometimes correlated with adult size and reproductive capacity, or other biological parameters such as survival, and is thought to be an adaptive strategy for dealing with unpredictable environments (Marshall et al. 2008). With *Z. provisorio*, however, there was not a strong relationship between initial and final size.

Snails, though not slugs, require ready access to calcium sources if shell development is to be

normal (Speiser 2001; Beeby & Richmond 2007). I provided *Z. provisorio* with continuous access to soil so they would have adequate calcium, regardless of the insect test diet. The adequacy of the insect diets as sources of calcium thus remains uncertain, and should be a priority for additional research, because inclusion of soil introduced a significant source of bacteria, fungi, and mites that could cause deterioration of diets. If soil could be eliminated from the rearing process it might be possible to reduce the frequency of diet change, increasing rearing efficiency.

Acceptability of synthetic diets has long been known for terrestrial molluscs, but the diets developed expressly for molluscs have been used mostly to evaluate feeding behavior and the nutritional value of additives. Clearly, many molluscs also accept insect diets, and some display satisfactory growth and development when fed these diets. Thus, some synthetic diets offer potential for improving mass production of molluscs, whether used for research or other purposes. However, suitability of insect diets varies greatly among molluscs.

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