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## Effect commercial diets on growth, survival and chemical composition of the edible freshwater snail *Pomacea patula catemacensis*

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Vázquez Silva Gabriela, Talía Castro Barrera, Jorge Castro Mejía and Germán David Mendoza Martínez (2012) Effect commercial diets on growth, survival and chemical composition of the edible freshwater snail *Pomacea patula catemacensis*. Journal of Agricultural Technology 8(6):1901-1912.

Four commercial diets were studied to evaluate their effect on the growth, survival and chemical composition of *Pomacea patula catemacensis*. In total, 320 snails were fed with commercial diets of catfish, shrimp, tilapia and trout. Survival did not differ between foods, but there was a trend (Kruskal-Wallis,  $P=0.10$ ) to increase (94%) in snails fed with catfish diet. In a period of 195 days, snails showed similar growth with different foods ( $P>0.05$ ) with a slight trend in the food for trout with a final total weight of  $9.5 \pm 2.5$  g. Survival did not differ between food, but there was a trend of 94% in snails with food for catfish (Kruskal-Wallis,  $P=0.106$ ). The commercial diets modified the snails percentages of protein, fat and carbohydrates ( $P<0.05$ ), The commercial diets modified the snails percentages of protein, fat and carbohydrates ( $P<0.05$ ), being the highest concentrations of protein ( $49 \pm 3.3\%$ ) and fat ( $35.5 \pm 3.3\%$ ) in snails with trout food and carbohydrates with shrimp food ( $39 \pm 3.1\%$ ). The results show that the four diets produced a similar growth in *P. catemacensis patula*, however it is important to consider that food altered the chemical composition of the snail meat and its survival, which is important to consider for future practical cultivation in captivity.

**Key words:** commercial foods, composition chemical, growth, edible freshwater snail, *Pomacea patula catemacensis*, survival.

### Introduction

In Mexico, mollusks from freshwater, saltwater and brackish are part of the traditional diet in coastal communities (Baqueiro, 1984), in which highlights the endemic freshwater snail *Pomacea patula catemacensis* (Baker,

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1992), which inhabits the Catemaco Lake at the State of Veracruz, representing an important resource in community because its fishery is an important source of income (Naranjo and García, 1986). Unfortunately, very little conservation management has been practiced (Naranjo, 2003) and, as a result, the natural populations of this gastropod have drastically diminished in recent years. Conservation management of this species is limited to a restricted harvesting season during March and June, during which the minimum size of captured snails must be longer than 33 mm in length. This species has not been produced on a large scale, and their requirements in captivity for their growth and survival have not been studied in detail. In addition, basic information about *P. patula catemacensis* is scarce, especially regarding specific diets that could help to establish domestic culturing of this snail (Garcia-Ulloa *et al.*, 2007).

One approach that could be used for preserving this species without ignoring its demand in the area is the development of aquaculture techniques that would permit its breeding; this snail has shown great cultivation potential due to its large size, rapid growth, easy incubation and high hatching rate (Carreón *et al.*, 2003). In addition, snail growth can be optimized by using commercial diets rich in protein (Ramnarine, 2004; Tamaru *et al.*, 2005) and with equivalent percentages of animal and vegetable proteins (Mendoza *et al.*, 2002). The genus *Pomacea* has generally been considered as an herbivorous organism (Burlakova *et al.*, 2009); nevertheless, it is capable of adapting to artificial diets that include animal protein (Góngora *et al.*, 2004; Ruíz *et al.*, 2005; Tamaru *et al.*, 2005; Garr *et al.*, 2011). It is important to mention that there are no commercial diets available for this species of snail; therefore, a possible alternative for its feeding in captivity is diets designed for other aquatic species. The aim of this study was to compare the effect of four commercial diets (trout, catfish, shrimp and tilapia) on the growth, survival and chemical composition of *P. patula catemacensis* under laboratory conditions.

## **Materials and methods**

### ***Feeding Trial***

The experiment lasted 195 days, beginning from the hatching of eggs. In total, 320 snails of similar size were taken at 15 days post-hatching (initial total length  $0.4 \pm 0.05$  cm, initial width  $0.4 \pm 0.04$  cm, initial weight  $20 \pm 0.5$  mg; mean  $\pm$  SEM) and randomly distributed in four treatments or commercial diets: catfish, shrimp, tilapia and trout; with four replications (aquaria).

The snails were kept in glass aquaria (25 x 60 x 30 cm) at densities of 20 organisms per 40 l as recommended by Ruiz *et al.* (2005). The aquariums were subject to natural photoperiod and fitted with activated carbon filters, equipped

with heaters to maintain temperature at  $26 \pm 0.7$  ° C. Water pH was  $8.2 \pm 0.3$  and was measured weekly with a potentiometer (Hanna Instruments HI 98103;  $\pm 0.01$ ).

The protein content reported by the manufacturer (Agribands Purina Mexico, Cargill Company), for the foods was: catfish 28%, shrimp 25%, tilapia 25% and trout 40%. These diets were analyzed for content dry matter, ash, lipids (A.O.A.C., 2000), crude protein (Eberhard, 1991), neutral detergent fiber (Van Soest *et al.*, 1991), phosphorous (Harris and Popat, 1954) and gross energy was determined using an adiabatic oxygen bomb calorimeter model 1241 (Parr Instrument Company, Illinois, USA). The carbohydrates were estimated by difference between 100 and the sum of ash, crude protein, lipids and NDF, shown in Table 1. Food was provided at a rate of 1% of the total biomass in two portions (9:00 and 17:00 h). In order to maintain constant water quality, feces and uneaten food were removed daily replenishing the water level.

**Table 1.** Chemical analysis of the nutrients found in the four commercial diets fed to *P. patula catemacensis*

Contents (%)	Diet			
	Catfish	Shrimp	Tilapia	Trout
Dry material	91.7 $\pm$ 0.5	91.2 $\pm$ 0.4	91.3 $\pm$ 0.4	91.1 $\pm$ 0.3
Ash	8.5 $\pm$ 0.7	6.7 $\pm$ 0.03	9.1 $\pm$ 0.1	8.9 $\pm$ 0.1
Crude protein	31.0 $\pm$ 0.4	27.8 $\pm$ 0.2	28.7 $\pm$ 0.7	42.0 $\pm$ 0.1
Crude fat	9.1 $\pm$ 0.6	11.8 $\pm$ 0.9	1.70 $\pm$ 0.4	2.50 $\pm$ 0.9
Carbohydrates	24.0 $\pm$ 1.7	31.9 $\pm$ 1.0	34.3 $\pm$ 1.9	16.9 $\pm$ 0.6
FDN	27.3 $\pm$ 1.1	21.8 $\pm$ 4.1	26.2 $\pm$ 0.4	29.7 $\pm$ 1.3
Phosphorous	0.9 $\pm$ 0.1	1.0 $\pm$ 0.01	1.0 $\pm$ 0.01	1.1 $\pm$ 0.02
Gross energy (kcalg <sup>-1</sup> )	4.2 $\pm$ 0.6	4.3 $\pm$ 1.04	4.1 $\pm$ 1.3	4.40 $\pm$ 1.8

$\pm$  Standard deviation

### *Analysis of growth and survival*

The growth of the organisms was recorded every two weeks. The total length and width of the snails was measured according to the criteria of Burch and Cruz (1987), with digital calliper vernier ( $\pm 0.01$  mm) and live weight of the snails was recorded with an analytical balance ( $\pm 0.0001$  g). The weight of the shells and the shelled biomass produced was measured at the end of the experiment sacrificing the snails.

The average weight daily gain (WDG) was calculated by the difference between final weight ( $W_f$ ) and initial weight ( $W_i$ ) during the number of days ( $d$ ) as  $WDG$  (mg d<sup>-1</sup>) =  $(W_f - W_i)/d$ . Similarly, daily growth in length (DGL) was

obtained by difference between final length ( $L_f$ ) and initial length ( $L_i$ ) in days ( $d$ ) as DGL (cm d-1) =  $(L_f - L_i)/d$  as described by Hopkins (1992). The feed conversion ratio (FCR) or grams of food to obtain a gram of snail was estimated as described by You *et al.* (2008):  $FCR = [\text{dry feed intake (g)}]/[\text{wet weight gain (g)}]$ . Also were estimated the growth parameters of von Bertalanffy Model:  $L_t = L_\infty[1 - e^{-K(t-t_0)}]$  where  $L_\infty$  is the theoretical total maximal length that can be achieved by the organisms,  $L_t$  is the total length in age  $t$ ;  $K$  is the rate or speed of growth,  $t_0$  is the initial time at which the organism has a length of zero (Hopkins, 1992). The percentage of survival was calculated from the number of snails who completed the trial as a proportion of the number at the beginning of the experiment.

### ***Chemical composition of the snail***

In order to have a reference on chemical composition of *P. patula catemacensis*, wild organisms were collected with similar weight and size (total length  $3.5 \pm 0.3$  cm,  $8.5 \pm 1.1$  g, mean  $\pm$  SEM) characteristics to those from the experiment. Biomass of organisms obtained in each aquarium and that of wild snails were oven dried to determine dry matter, ash, lipids (A.O.A.C., 2000), crude protein (Eberhard, 1991) and gross energy was determined using an adiabatic oxygen bomb calorimeter model 1241 (Parr Instrument Company, Illinois, USA). The carbohydrates were estimated by difference between 100 and the sum of ash, crude protein and lipids.

### ***Statistical Analysis***

Statistical tests were performed using the SYSTAT 8.0 computer program (Systat Software Inc.). A one-way analysis of variance (ANOVA) and a confidence level of 95% were applied to the results of total length, shell width, weight, weight of the shell, weight daily gain, daily growth in length, feed conversion ratio and chemical composition of the snail. Only the means of snails fed the commercial diets were compared. Tukey tests of comparison of multiple averages were used to determine significant differences between the treatments. Survival of the organisms was compared using a Kruskal-Wallis test ( $\alpha = 0.05$ ) (Zar, 1999).

## Results

### *Growth and Survival*

There were no differences in the biometric measurements (final length, width, final weight and weight of the shell) between treatments ( $P>0.05$ ). The growth in total length of *P. patula catemacensis*, the average size after 195 days ranged between was 3.0 to 3.2 cm for the four diets (Table 2), however the final body weight showed a trend with food for trout ( $9.5 \pm 2.5$  g). The diets did not exert a significant effect on WDG ( $P=0.70$ ), the DGL ( $P=0.76$ ) nor in the FCR (Table 2), But in the best efficiency numerically was observed with the catfish diet (1.5).

The diets used in the experiment had no significant effect on the survival of snails (74%, Kruskal-Wallis=6,126,  $P=0.10$ ,  $df=3$ ), however, there was a tendency to be higher (94%) with the catfish diet, while the lowest survival was obtained with tilapia diet 74% (Table 2).

The parameters of the model indicated that the snail showed an allometric growth response in all treatments. The  $L_{\infty}$  values showed a range of 3.4 to 3.9 cm between the catfish and the tilapia diets respectively. The model of Von Bertalanffy showed a determination coefficient of 0.99 in the catfish and tilapia diets, whereas the diets of shrimp and trout were 0.98 (Table 3).

**Table 2.** Growth and survival values of the snail *Pomacea patula catemacensis* after being fed different commercial diets

Parameters	Diet			
	Catfish	Shrimp	Tilapia	Trout
n	75	69	59	65
Final total length (cm)	3.1 $\pm$ 0.3	3.0 $\pm$ 0.2	3.0 $\pm$ 0.4	3.2 $\pm$ 0.3
Final width (cm)	2.9 $\pm$ 0.2	2.8 $\pm$ 0.2	2.7 $\pm$ 0.4	3.0 $\pm$ 0.3
Final body weight (g)	8.7 $\pm$ 2.1	7.9 $\pm$ 1.1	8.1 $\pm$ 2.6	9.5 $\pm$ 2.5
Final weight of shell (g)	1.3 $\pm$ 0.5	1.3 $\pm$ 0.5	1.4 $\pm$ 0.5	1.4 $\pm$ 0.5
WDG (mg/day)	45 $\pm$ 0.1	40 $\pm$ 0.1	42 $\pm$ 0.1	49 $\pm$ 0.1
DGL (cm/day)	0.013 $\pm$ 0.02	0.013 $\pm$ 0.01	0.012 $\pm$ 0.02	0.014 $\pm$ 0.01
FCR (g food/g snail gain)	1.5 $\pm$ 0.2	1.7 $\pm$ 0.2	2.1 $\pm$ 0.4	1.6 $\pm$ 0.5
Snail biomass (g)	79.5 $\pm$ 27.5	75.2 $\pm$ 5.3	64 $\pm$ 26.6	76.1 $\pm$ 24.9
Snail biomass without shell (g)	54.7 $\pm$ 21.5	51.9 $\pm$ 5.2	42.7 $\pm$ 24.62	52.9 $\pm$ 17.7
Survival (%)	94 $\pm$ 6	86 $\pm$ 13	74 $\pm$ 11	81 $\pm$ 13

Data are presented as means  $\pm$  standard error (SEM) of the four replicates

WDG= Weight Daily Gain

DGL= Daily Growth in Length

FCR= Feed Conversion Ratio

**Table 3.** Growth parameters of Von Bertalanffy's Model for *Pomacea patula catemacensis* fed different diets

Parameters	Diet			
	Catfish	Shrimp	Tilapia	Trout
$L_{\infty}$	3.9	3.5	3.4	3.7
K	0.11	0.13	0.14	0.12
$t_0$	-0.05	-0.11	-0.08	-0.11

$L_{\infty}$  = theoretical total maximal length that can be achieved by the organisms

K = rate or speed of growth

$t_0$  = initial time at which the organism has a length of zero

### Chemical composition

The chemical compositions of snails fed with the balanced diets did not differ in dry material, ash and gross energy (Table 4). The highest concentration of protein ( $P < 0.05$ ) was observed in snails fed the trout diet ( $49.7 \pm 3.3\%$ ). The trout and tilapia diets promoted higher levels of fat deposition in the snails, whereas the catfish diet resulted in minor levels of fat deposition ( $P < 0.05$ ).

**Table 4.** Comparison of the chemical composition of wild and cultivated *Pomacea patula catemacensis*

Content (%)	Diet				Wild snail
	Catfish	Shrimp	Tilapia	Trout	
n	75	69	59	65	
Dry material	19.5 $\pm$ 2.0	21.7 $\pm$ 1.9	19.0 $\pm$ 3.7	20.5 $\pm$ 3.3	24.7 $\pm$ 10.9
Ash	8.1 $\pm$ 0.6	6.1 $\pm$ 1.8	9.0 $\pm$ 2.3	8.1 $\pm$ 0.4	13.0 $\pm$ 3.3
Protein	46.9 $\pm$ 1.6 <sup>ab</sup>	41.1 $\pm$ 3.7 <sup>b</sup>	47.7 $\pm$ 6.4 <sup>a</sup>	49.7 $\pm$ 3.3 <sup>a</sup>	45.2 $\pm$ 15.7
Crude fat	8.2 $\pm$ 2.1 <sup>b</sup>	13.8 $\pm$ 1.8 <sup>ab</sup>	17.2 $\pm$ 3.7 <sup>a</sup>	35.2 $\pm$ 3.3 <sup>a</sup>	2.6 $\pm$ 0.8
Carbohydrates	38.5 $\pm$ 1.7	39.0 $\pm$ 3.1 <sup>a</sup>	26.0 $\pm$ 3.4 <sup>ab</sup>	7.0 $\pm$ 3.1 <sup>b</sup>	41.8 $\pm$ 0.7
Gross energy (kcalg <sup>-1</sup> )	4.4 $\pm$ 0.2	4.6 $\pm$ 0.2	4.3 $\pm$ 0.3	4.4 $\pm$ 0.2	4.4 $\pm$ 0.04

Data are presented as means  $\pm$  standard error (SEM) of the four replicates

Means in a row with different superscript are different ( $P < 0.05$ )

Carbohydrates showed highest variation in values, with snails fed the catfish and shrimp diets having the highest carbohydrate levels, intermediate levels for snails fed the tilapia diet and the lowest levels for snails fed the trout diet. Despite of the differences in nutrient concentrations, the calorific values of the snails were similar between all of the diets ( $P > 0.05$ ). The wild snails presented lower amounts of fat, but similar protein, dry matter, ash and gross energy contents.

## Discussion

Although there has been much debate regarding the nutritional habits of species of the genus *Pomacea*, such as herbivory and polyphagia, the results confirmed that these snails accept other types of diet (Asiain and Olguín, 1995; Estebenet, 1995; Estebenet and Martín, 2002). Experiments performed by Fellerhoff (2002) demonstrated that *P. lineata*, in addition to consuming vegetables, also accepts diets of organic matter and compost from livestock.

Other investigations of this genus showed that balanced diets of fish or frogs (Góngora *et al.*, 2004) are accepted and allow the growth of these snails. In the present experiment it was found that *P. patula catemacensis* readily accepted feed designed for other aquaculture species. In some studies that did not report the chemical compositions of the feeds (Lagunes, 1997; Asiain and Olguín, 1995), the diets were based on *ad libitum Ipomoea aquatica*, which contains around 7.5% protein (Garcés *et al.*, 2006). In the assays by Mendoza *et al.* (2002) all of the diets included 25% protein.

Protein requirements for the optimal growth of *P. patula catemacensis* are still unknown and further research is required. Growth estimated on the biometric values indicated that these snails are able to metabolize a wide range of nutrient concentrations and compositions, suggesting that they may be able to adjust their metabolism; however, it was not clear if this might be a factor that affects survival.

## Growth and survival

The values recorded in the feeding trial with *P. patula catemacensis* are similar to those reported by Carreón *et al.* (2003), who reported that over a 6-month period, males and females of the species reached sizes of 3.8 and 3.3 cm, respectively, on a mixed diet of 40% gelified microalgae *Scenedesmus incrassulatus* and 60% trout feed (40% protein).

With respect to the weight of this gastropod, the four balanced diets used in the present study produced a weight of between 4.2 and 5.2 g on day 135 of the experiment, with a density of one snail 2 l. Asiain and Olguín (1995) obtained an average weight of 1.37 g in *P. patula* fed a diet of *I. aquatica* over 133 days with a density of 22 organisms per l, which shows that diet and density are important factors in the weight of *Pomacea*. Ruíz *et al.* (2005) recorded similar values for the weight of juvenile *P. patula* as those observed in the present experiment (7.9-9.5 g in 195 days); in their study, snails on a diet of carp feed and *Calothrix* sp cyanobacteria weighed 8.02 and 8.94 g (14 snails per 2.1 l), respectively, over a 181 day period.

In an experiment with *P. canaliculata*, Tamaru *et al.* (2005) found greater weight gain (2.3 g) with feeds for catfish, compared with diets for chicken (0.5 g), a combination of catfish and chicken foods (1.3) and a control without food (0.4); maintaining a density of 37.5 snails per l in 30 days of the experiment.

The values of shell weight reported in this experiment differ from those found by Lobo (1986), who reported those weights under wild conditions, *P. flagellata* individuals with a total weight of 9.80 g and a size of 3.40 cm achieved an average shell weight of 6.10 g. In addition, Rojas (1988) found similar values in wild *P. costaricana* snails with an average shell weight of 5.2 g and a size and weight of 3.25 cm and 9.91 g, respectively. It is important to consider that the values of shell weight obtained in this study were lower than those reported by other authors, these results are more related to the variations in size and morphology, which are inherent in species of this genus, that factors which may limit the development of the shell such as calcium and pH (Glass and Darby, 2009).

It is assumed that diets offered in this experiment contained minerals in appropriate concentrations and available sources for *P. patula* (calcium carbonate, monocalcium phosphate and monosodium, potassium and sodium chloride, iron sulphate, zinc sulphate and copper sulphate) and the water pH (8.2) had an alkalinity adequate for the shell development. When calcium is limited and the pH is acidified, the shell growth can be affected. Glass and Darby (2009) reported that at concentrations less than 3.6 mg of  $\text{Ca}^{2+}$  and pH below 6.5, the shells of *P. paludosa* showed a lower weight and size and erosions on the surface.

The daily growth in length (DGL) of *P. patula catemacensis* (0.014 cm per day) was lower than that reported for *P. padulosa* by Garr *et al.* (2011) of 0.049  $\text{cm d}^{-1}$  when used catfish food and a diet of macroalgae *Ulva* sp.

The FCR values of the snails within the treatments in this study are similar to that reported by Ramnarine (2004) who found FCRs in *P. urceus* of 1.46 and 1.77 in diets with 15% and 20% protein, respectively. Mendoza *et al.* (2002) found that for *P. bridgesi*, the highest rate of feed conversion (between 4 and 5) was observed in diets formulated with 75% animal protein and 25% vegetable protein. Tamaru *et al.* (2005) obtained a similar feed conversion rate (1.03) for *P. canaliculata* with catfish feed with 32% crude protein. These values demonstrate that the snails of the *Pomacea* genus can assimilate high protein content, whether it is animal or vegetable in origin.

The survival values for *P. patula catemacensis* fed the catfish diet were higher (94%) than those reported for *Pomacea* sp. by Lagunes (1997) which varied between 68% and 76% on a diet of *I. aquatica* under semi-natural conditions. On the other hand, Asiain and Olguin (1995) who also utilized *I.*

*aquatica*, obtained a survival rate of 93% in controlled conditions. This indicates that survival is not only influenced by the diet but also by cultivation management. Mendoza *et al.* (2002) recorded a survival rate of 100% in *P. bridgesi* using experimental diets with different levels of vegetable and animal proteins.

According to the Von Bertalanffy model, the growth of the snail *P. patula catemacensis* is described with high precision, in contrast with Rojas (1988) who reported a lack of fit for the growth of *P. costaricana* with the same model ( $r^2 = 0.57$ ), although the  $L_\infty$  values obtained for *P. costaricana* were larger ( $L_\infty$  6.75 cm) than those found in this study in a growth trial lasting 240 days. Those differences reflect inherent characteristics of the specie in addition to food quality and test duration.

### ***Chemical Composition***

Differences in snail composition reflected differences in the metabolism of nutrients and the sources of feeds used to formulate the commercial diets, none of which were specifically balanced for this snail. The crude protein content of snails in this experiment is similar to that observed in *P. costaricana* snails, which were found to contain 44.9% protein (Rojas, 1988). On the other hand, wild *P. flagellata* (Lobo, 1986) and *P. canaliculata* (Bombero-Tuburan *et al.*, 1995) were richer in protein than *P. patula*, as these contained 59% and 54%, respectively. These values indicate that the protein value may vary between species and that each species has a potential limit for the utilization of protein independent of the protein level found in the diet (Ramnarine, 2004).

With regard to the fat content, the diets with higher fat concentrations did not result in higher levels of fat deposition. Also, the wild snails had the lowest fat values, suggesting that energy expenditure is lower in captivity and that *P. patula* stored more energy with the experimental diets. Values reported for the fat concentration in snails are usually low: Lobo (1986), Rojas (1988) and Bombero-Tuburan *et al.* (1995), found 3.67%, 3.14% and 1.4% fat, respectively, in other *Pomacea* species.

In terms of the carbohydrate content, *P. patula catemacensis* showed a range of 7% to 38.5%, with 41.8% for wild snails, while levels of 17.5% were reported for *P. flagellata* (Lobo, 1986) and 16.83% for wild *P. costaricana* (Rojas, 1988) in the natural environment with herbivorous diets.

Chemical analysis of the four diets used in the present experiment presented formulations for different species consider different nutritional requirements and also diverse feed types; therefore, there were differences in most of the concentrations of the feed components, such as protein (range 27.8 to 42), fat (range 1.7 to 9.5) and carbohydrates (16.9 to 34.3), but the Neutral

Detergent Fiber (NDF) values indicated that the different diets had similar concentrations of vegetable sources, that the phosphorous levels were basically the same and that the energy levels were similar, which explains the similar biometric values obtained in the snails.

## Conclusion

The results indicated that growth of the *P. patula catemacensis* snail was similar on the four balanced diets with different protein levels (catfish, shrimp, tilapia and trout); however, the catfish diet was found to favored survival. In a relative comparison, the trout food improved the protein content in the meat snail regarding to the wild snails but increased the lipid percentage. Results from this experiment are useful to plan feeding programs and the cultivation of *P. patula catemacensis* in captivity.

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(Received 17 April 2012; accepted 30 October 2012)