

AIA



Avances en Investigación Agropecuaria  
Universidad de Colima  
revaia@ucol.mx  
ISSN (Versión impresa): 0188-7890  
MÉXICO

2005

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JUVENILES FED INERT DIETS

*Avances en Investigación Agropecuaria*, septiembre, año/vol. 9, número 003

Universidad de Colima

Colima, México

pp. 49-59

Red de Revistas Científicas de América Latina y el Caribe, España y Portugal

Universidad Autónoma del Estado de México

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# Growth of angel fish *Pterophyllum scalare* [Gunther, 1862] juveniles fed inert diets

Crecimiento de juveniles del pez ángel *Pterophyllum scalare* [Gunther, 1862] alimentados con dietas inertes

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## Abstract

The growth, feed conversion ratio (FCR), survival and stress resistance of angel fish *Pterophyllum scalare* juveniles fed different diets (decapsulated *Artemia* cysts DAC, commercial flakes CF, commercial pellets CP, and a commercial starter diet for tilapia CSDT), were investigated. Diets were studied with three replicates and adjusted at 8% of daily feeding ratio. Fish had an average initial wet weight of 0.44 g. Diets showed a significant effect on fish growth performance from the first sampling day onwards. After 45 culture days, fish fed with the DAC diet showed the highest mean standard length, wet weight and specific growth rate ( $3.64 \pm 0.07$  cm,  $3.19 \pm 0.24$  g and  $4.36$  % body weight/day, respectively), compared with the rest of the treatments. The FCR fluctuated from  $1.26 \pm 0.01$  for the DAC group to  $2.01 \pm 0.17$  for the CP diet, showing significant differences ( $P < 0.05$ ) among treatments. Survival was similar for all diets. Fish fed with the DAC diet displayed higher stress resistance compared with the rest of the treatments. Although DAC seem to improve growth performance of angel fish juveniles, further trials are required to

## Resumen

Se investigó el crecimiento, conversión alimenticia (FCR), sobrevivencia y resistencia al estrés de juveniles del pez ángel *Pterophyllum scalare*, alimentados con diferentes dietas inertes (quistes decapsulados de *Artemia* DAC, hojuelas comerciales CF, pelets comerciales CP y una dieta comercial iniciadora para tilapia CSDT). Las dietas fueron estudiadas con tres réplicas y la ración alimenticia fue ajustada al 8% de la biomasa total. Los peces pesaron 0.44 g en promedio, al inicio. Las dietas mostraron un efecto significativo sobre el crecimiento de los peces desde la primera biometría. Después de 45 días de cultivo, los peces alimentados con DAC mostraron los valores promedio más altos en la longitud estándar, peso húmedo y crecimiento específico ( $3.64 \pm 0.07$  cm,  $3.19 \pm 0.24$  g y  $4.36$  % peso corporal/día, respectivamente), comparado con el resto de los tratamientos. El FCR fluctuó desde  $1.26 \pm 0.01$  para el grupo DAC, hasta  $2.01 \pm 0.17$  para la dieta CP, mostrando diferencias significativas ( $P < 0.05$ ) entre los tratamientos. La sobrevivencia fue similar para todas las dietas. Los peces alimentados con DAC mostraron la

identify other factors (nutritional biology, feeding techniques and strategies) associated with its most economic use.

### *Key words*

Ornamental fish, *Pterophyllum scalare*, nutrition.

mayor resistencia al estrés comparado con el resto de los tratamientos. Aunque la dieta DAC mejoró el crecimiento de los juveniles del pez ángel, se requieren de estudios posteriores para identificar algunos factores (biología nutricional, técnicas y estrategias de alimentación) asociados a su mejor uso económico.

### *Palabras clave*

Pez ornamental, *Pterophyllum scalare*, nutrición.

## Introduction

Freshwater ornamental fish trade is a million-dollar industry [Lim and Wong, 1997]. Among the most popular freshwater fish species in the aquarium trade industry, is the angel fish *Pterophyllum scalare* [Gunther, 1862], which is originated from a branch of the Amazonas River [Tapajoz River], at the north of Brazil [Soriano-Salazar and Hernández-Ocampo, 2002]. Due to its body coloration, shape and economical value [Luna-Figueroa, 2003], the angel fish represents one of the most important ornamental cichlid species, but amongst the main constraints to its optimal commercial production, is the lack of knowledge on proper diets for the different life stages [Luna-Figueroa *et al.*, 2000]. *P. scalare* is considered an omnivorous fish since in nature, its feeding is based on the consumption of plankton, larvae of insects and crustaceans, plants and worms [Soriano-Salazar and Hernández-Ocampo, 2002]. In captivity, the common live larval food used for growing of most ornamental fish is limited to macro-zooplankton such as *Moina*, *Daphnia* and *Artemia nauplii* [Lim and Wong, 1997]. Artificial diets, which are normally elaborated with dried live organisms that are processed in different presentations such as flakes, meals or small pellets, are also used. Although it is known that the angel fish accepts artificial diets [Luna-Figueroa *et al.*, 2000], lower growth and survival rates of *P. scalare* are commonly obtained when such diets are used as the sole feed [Luna-Figueroa, 1999], mainly during the fry and juvenile stages [Hofer, 1985].

In general, it is well accepted that fish nutritional requirements vary with some factors such as the relationship between the diet and life stage [Ricker, 1979] and therefore, the dietary content for feeding larvae is different to that needed for repro-



duction or for growth of juveniles. But as a common practice for culturing angel fish, on-growing diets are also used to promote its reproduction [Pérez *et al.*, 2002; Luna-Figueroa, 2003]. Thus, the indiscriminating use of diets for the different live stages of *P. scalare* is normally associated to different biological responses, as pointed out in few nutritional reports [Luna-Figueroa *et al.*, 2000; Soriano-Salazar and Hernández-Ocampo, 2002; Luna-Figueroa, 2003]. Since angel fish is valued by its individual characteristics such as the skin coloration, body shape and size, obtaining of the maximum expression of these phenotype traits by supplying the proper and optimum diet, should be a topic of growing interest and priority to producers. Therefore, the present work evaluates the effect of different commercial diets on growth and survival of *P. scalare* juveniles, under controlled conditions.

## Materials and Methods

Juvenile angel fish (250 fish with an average wet weight of  $0.30 \pm 0.02$  g) were obtained from a local supplier (Acuario Arboledas, Guadalajara, Jalisco, México) and transported to the lab in plastic bags filled with oxygen. Fish were acclimated by placing the bags in a 400 l fiber glass container until water temperature was equalized inside the bags at  $27 \pm 1$  C [Axelrod *et al.*, 1997]. The container was supplied with tap water passed through a cartridge filter ( $5 \mu\text{m}$  diameter), and gentle aeration was provided by two airstones. Water temperature in the container was kept constant ( $28^\circ\text{C}$ ) by introducing an electrical heater (100 W, Rena Submersible Heater, RH100, Apopka, FL, USA), and photoperiod was adjusted at 12 hours light and 12 hours dark during acclimating time. Fish were divided in four groups and kept in 4 containers for one week feeding them in excess with each experimental diet. Food ration was given three times at day (09:00, 13:00 and 17:00 h). Each day, 50% of the total water volume of the container was exchanged, extracting out feces and food waste by siphoning.

To evaluate the experimental diets, twelve 113 l glass aquaria were used. Each aquarium was provided with a self-filtration bottom system, consisted in a 3/8 inches PVC tubes net perforated at its bottom side with 1/16 inches diameter holes. The tubes net was covered, with a 0.3 cm diameter plastic mesh and then, with an 8 cm layer of small stones (0.5 cm diameter). Dissolved oxygen was maintained by a 3/8 inches PVC airlift standpipe connected to the bottom tubes net. This way, water was forced to flow down through the stones layer where suspended solids (feces and food waste) were retained.

Aquaria were stocked with 15 fish each, randomly selected from the initial group. The initial mean wet weight was  $0.44 \pm 0.08$  g. Four diets were tested with three

replicates: Commercial flakes (CF) with 43% crude protein (wardley total tropical, USA); commercial pellets (CP) with 45% crude protein (azoo, 9 in 1 drawf cichlid pellet, China); decapsulated *Artemia* cysts (DAC) with 54% crude protein (*Artemia* shell-free, inve aquaculture, USA); and a commercial starter diet for tilapia (CSDT) with 40% crude protein (API-ABA, MaltaCleyton de México, S. A de C. V., México, D. F.). The proximal composition of experimental diets is shown in Table 1. Feeding ration was adjusted at 8% of the total biomass/day [García-Ulloa, 2004] and given at same hours than acclimation time. The experimental diets were tested for 45 days.

Table 1. Proximal composition of tested diets (% dry weight) given by the food suppliers.

	DAC	CF
Dry matter	95	91
Crude protein	54	43
Lipid	9	5
Fiber	6	4
Ash	4	—*
Nitrogen-free extract	—	—

DAC = Decapsulated *Artemia* cysts, shell-free inve aquaculture, USA; CF = Commercial flakes, wardley total tropical, USA; CP = Commercial pellets, AZOO, 9 in 1 drawf cichlid pellet, China; CSDT = Commercial starter diet for tilapia, API-ABA, MaltaCleyton de México.

\* Information not given by the producer.

All fish from each replicate were individually measured and weighted at the beginning and every two weeks until the end of the experiment. Wet weight (g) and standard length (cm) were determined at each sampling day, with an electronic balance (Navigator, Scout, 100 g x 0.1 g, Apopka, FL, USA) and a domestic scale, respectively. Prior of weighing, fish were placed on absorbent paper to remove excess of water. The total weight gain (TWG) was calculated as final fish weight-initial fish weight. The daily growth rate (DGR) was calculated as  $DGR = TWG / \text{culture days}$ . The specific

growth rate (SGR, % body weight/d) was calculated as  $SGR = 100 (\ln W_f - \ln W_i) / t$ , where  $W_f$  = mean weight at the end of sampling,  $W_i$  = mean weight at the beginning of the sampling, and  $t$  = time in days of the sampling period [Ricker, 1979]. The feed conversion ratio (FCR) was calculated as  $FCR = \text{total weight of dry feed given} / \text{TWG}$ . Survival was obtained by counting total animals per diet at the end of the experiment. All data were tested for normal distribution and homogeneity of variance before ANOVA was performed (Sokal and Rohlf, 1969). An analysis of covariance was used with initial wet weights as covariate to detect possible differences among the diets at the beginning of the experiment [Johnson, 1976].

The percentages of fish survival values were arcsine transformed to make the variance independent of the mean [Reyes, 1982]. Figures present untransformed data of mean percentage  $\pm$  standard deviation. A stress test was used to evaluate the stress resistance of the angel fish fed with the different diets, according to the procedure described by Lim *et al.* [2000]. The test consisted in the exposition of the fish to osmotic shock in a saline solution, and the cumulative mortality of the fish was monitored at 3-min intervals over a 2-h period. After a previous test to determine the optimal saline solution for the stress test (30 ‰), five fish from each replicate were exposed to the stress salinity concentration to obtain the cumulative mortality per treatment expressed as stress index. ANOVA analysis was performed and differences between means were compared for significance ( $\alpha = 0.05$ ) using the Tukey's multiple range test [Reyes, 1982]. The statistical analyses were performed using Jandel SigmaStat 2.0 statistical software (Jandel Co., USA).

## Results

All experimental diets were eagerly consumed by angel fish. Mean body wet weight and standard length were significantly different ( $P < 0.05$ ) among diets from the first sampling day (day 15) onwards (Figure 1). At the end of the experiment, fish fed with the CP diet obtained the lowest mean standard length ( $2.71 \pm 0.01$  cm) and weight ( $1.49 \pm 0.15$  g), meanwhile the DAC group showed the highest growth values ( $3.64 \pm 0.07$  cm and  $3.19 \pm 0.24$  g, for the standard length and wet weight, respectively). Growth responses were significantly different ( $P < 0.05$ ) for TWG, DGR and SGR among the treatments (Table 2).

Angel fish from the DAC diet obtained the highest mean final TWG ( $2.75 \pm 0.24$  g), DGR ( $0.061 \pm 0.005$  g/d) and SGR ( $4.36 \pm 0.17\%$  BW/d). The FCR among the treatments showed differences, and ranged from  $1.26 \pm 0.10$  for the DAC group to  $2.01 \pm 0.17$  for the CP diet. The mean final survival of juvenile angel fish (Table 2), fluctuated from 88.88% for the DAC treatment, to 97.77 % obtained for

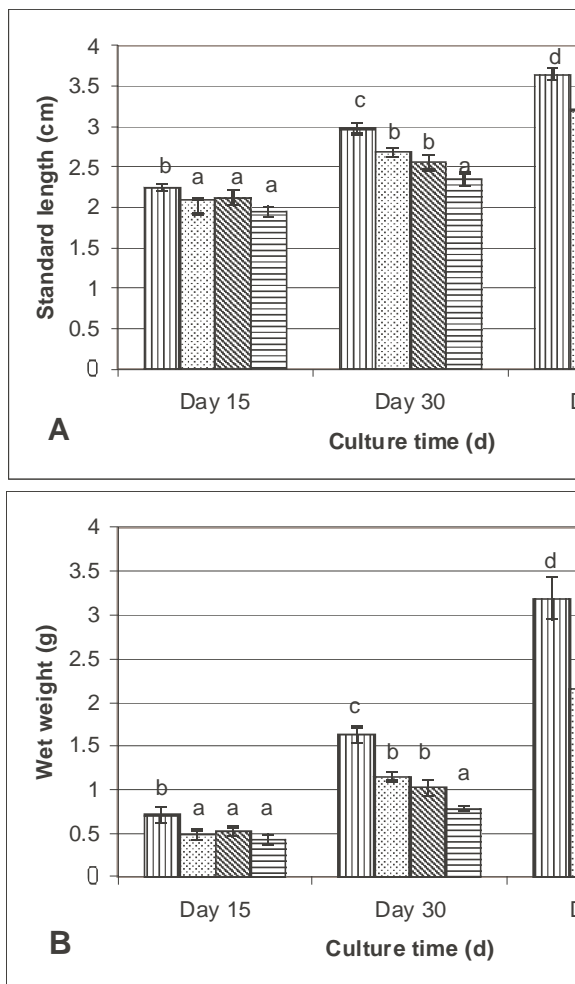
the CF diet, after 45 culture days. After two hours of osmotic stress challenge, there were not significant differences ( $P > 0.05$ ) for the cumulative mortality among the dietary groups (Table 2).

Table 2. Mean final total weight gain (TWG), daily growth rate (DGR), specific growth rate (SGR), food conversion ratio (FCR), survival and cumulative mortality of angel fish juveniles fed with different diets.

	DAC	CF	CP	CSDT
<b>TWG (g)</b>	2.75 (0.24) <sup>b</sup>	1.71 (0.02) <sup>ab</sup>	1.71 (0.02) <sup>ab</sup>	1.71 (0.02) <sup>ab</sup>
<b>DGR (g/d)</b>	0.061 (0.005) <sup>b</sup>	0.037 (0.001) <sup>ab</sup>	0.037 (0.001) <sup>ab</sup>	0.037 (0.001) <sup>ab</sup>
<b>SGR (%BW/d)</b>	4.36 (0.17) <sup>b</sup>	3.50 (0.02) <sup>ab</sup>	3.50 (0.02) <sup>ab</sup>	3.50 (0.02) <sup>ab</sup>
<b>FCR</b>	1.26 (0.01) <sup>a</sup>	1.44 (0.03) <sup>b</sup>	1.44 (0.03) <sup>b</sup>	1.44 (0.03) <sup>b</sup>
<b>Survival (%)</b>	88.88 (6.28)	97.77 (3.14)	97.77 (3.14)	97.77 (3.14)
<b>Cumulative mortality (stress index)</b>	170.00 (1.63) <sup>a</sup>	178.33 (1.69) <sup>b</sup>	178.33 (1.69) <sup>b</sup>	178.33 (1.69) <sup>b</sup>

(DAC = Decapsulated *Artemia* cysts, shell-free inve aquaculture, USA; CF = Commercial flakes, wardley total tropical, USA; CP = Commercial pellets, azoo, 9 in 1 drawf cichlid pellet, China; CSDT = Commercial starter diet for tilapia, API-ABA, MaltaCleyton de México). Values in parenthesis indicate  $\pm$  standard deviation. Mean in each line with same superscript are not significantly different ( $P > 0.05$ ).

Figure 1. Mean standard length (A) and wet weight (B) of *Pterophyllum scalare* juveniles (mean  $\pm$  standard deviation of three replicates) fed with different diets.



(DAC = Decapsulated *Artemia* cysts, shell-free invertebrate aquaculture, USA; CF = Commercial flakes, Wardley total tropical, USA; CP = Commercial pellets, azoo, 9 in 1 drawf cichlid pellet, China; CSDT = Commercial starter diet for tilapia, API-ABA, MaltaCleyton de México). For each sampling day, different letters at the top of the bars indicate a significant difference ( $P < 0.05$ ) between treatments.

## Discussion

For selecting an adequate fish diet, some physical and chemical criteria related to feed intake should be carefully reviewed [Léger *et al.*, 1987]. For the former, the ingestion of food by fish is probably affected by its size and palatability. Size is thereby considered as one of the most important aspect. As food preference is closely related to the match between food and mouth size, the selected food changes with the growth of fish [Verreth, 1994]. In this study, it was observed that despite of the differences in size and presentation, all tested diets were eagerly consumed by fish at the different sampling times registered along the experiment, and consequently, at different growing stages (from  $1.65 \pm 0.21$  mm, to  $3.12 \pm 0.34$  mm of mean initial and final standard lengths, respectively). In the case of the latest, the relationship between digestibility and biochemical composition of diet is considered as a crucial selecting criterion. Due to its high affinity to be metabolized and retained, protein is considered the most important energy component for fish growth [Halver, 1972]. In the case of cultured carnivorous fish, dietary protein requirement usually accounts for 40 to 50% of feed fry matter [NRC, 1993].

Although there is evidence that high protein diets promote good feed utilization and growth in different fish species, mostly in human consumption species [Morais *et al.*, 2001; Ruohonen *et al.*, 2003], information about the protein requirements on angel fish growth rate is scarce. Soriano-Salazar and Hernández-Ocampo [2002], evaluated the use of live food and two inert commercial diets on the growth of *P. scalare*, concluding that live food (*Daphnia pulex*) with the higher protein content, produced better fish growth performance and survival, which coincides with observations made by Luna-Figueroa [2003], who compared two live foods (*D. pulex* and *Culex quinquefasciatus* larvae, with 50.15 and 40.18% crude protein, respectively) and three commercial flakes with different protein levels (45, 43 and 27% crude protein) for on-growing angel fish juveniles. In both cases, there was a direct relationship between the fish growth performance and the dietary protein content. In this experiment, proximal composition of tested diets given by the food suppliers showed comparable protein content (from 40 % for the CSDT group, to 54% for the DAC dietary treatment) with the above mentioned works, but growth of *P. scalare* juveniles did not show a direct relationship with the protein level of the tested diets.

After 45 culture days, angel fish from the DAC group showed the highest TWG and DGR values, and the better FCR compared to the other treatments (Table 2), being similar to those reported by Soriano-Salazar and Hernández-Ocampo [2002] and Luna-Figueroa [2003], feeding *P. scalare* juveniles with live food (*Daphnia pulex* or mosquito larvae) as sole diet. The SGR values obtained for all tested diets in this

experiment fluctuated from 2.67 %BW/d for the CP diet, to 4.36 %BW/d for the DAC group, lower to those reported by Soriano-Salazar and Hernández-Ocampo [2002] and Luna-Figueroa [2003], for the live diets, but similar to the obtained with the inert diets. It has been established that the SGR decreases when the fish size increases [Verreth and Den Bieman, 1987], since fish metabolism is reduced when body size increases. Soriano-Salazar and Hernández-Ocampo [2002] and Luna-Figueroa [2003], used smaller fish at initial stocking (0.1 and 0.058g, respectively) compared to the mean weight at the beginning of the present experiment (0.44 g), which could partially explain the differences in the SGR. For all groups, final survival were above 88.88% (Table 2), similar to the obtained by Luna-Figueroa *et al.* [2000], who fed the angel fish juveniles with *D. pulex*, but higher compared with the values reported with the use of inert diets.

Lim *et al.* [2002] mention that fish resistance to stress can be affected by sub-optimal physiological conditions arising from factors such as infection diseases, poor water quality and nutritional deficiencies, among others. In our study, the higher stress resistance was observed for the fish fed with the DAC diet, suggesting that its nutritional composition covered the physiological requirements at the fish age and salinity concentration evaluated.

Since water parameters (7.2-7.8 pH,  $27 \pm 1^\circ\text{C}$  and  $> 4$  mg/L DO) were constants and proper for the angel fish [Axelrod *et al.*, 1997] throughout the experiment, and diets were given at same daily ration and readily consumed by fish, it is suggested that overall results in this study were influenced by the nutritional quality of the tested diets. The nutritional value of one food for a species is related to the degree of correspondence between food biochemical composition and nutritional requirements of that species. This correspondence can change depending on the individual target species, its developmental stage [Walker *et al.*, 1998], or physiological condition, and as a function of the fish growth phase and culture conditions. The better growth results with DAC suggest that its composition is more related to the nutritional requirements of angel fish juveniles, but the gross biochemical composition of the diets tested does not properly explain these results.

Sorgeloos *et al.* [1986] and Vanhaecke *et al.* [1990], mentioned that DAC are suitable as unique food for some fish species at early life stages, because they are able to combine the advantages of live and inert diets, being a more hygienic and highly nutritional feed that can be processed for long-term storage, and compared with the use of live food, direct feeding of DAC would represent a labor saving strategy [Lim *et al.*, 2002]. Besides, Verreth *et al.* [1987], concluded that the differences in physical properties and ingestion of the feed can be eliminated when DAC are used as sole food, which was corroborated in this study since the cyst diet was eagerly consumed by

angel fish juveniles throughout the 45 experimental days. On the other hand, the nutritional value of DAC can be compared with that found for newly-hatched *Artemia*. García-Ortega *et al.* [1998], mentioned that both nauplii larvae and DAC have similar biochemical composition regarding all major nutrients, concluding that there is no difference in feeding *Artemia* cysts or nauplii to fish.

Inferring, DAC seem to improve growth performance of *P. scalare* juveniles, however much research is still required on the fish nutritional biology related to its feeding techniques and strategies using such diet.

## Conclusions

The overall results show that the DAC diet improved the angel fish growth performance at the cultured conditions described, nevertheless, much attention should be still paid on its nutrition, as well as economic considerations, in order to optimize the utilization of DAC at commercial level.

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Recibido: Agosto 12, 2005.

Acceptedo: Octubre 12, 2005.