

# Morphometric growth characteristics and body composition of bullfrog tadpoles in captivity

## Características do crescimento morfométrico e composição corporal de girinos de rã-touro em cativeiro

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### Abstract

Feed management needs to be improved in frog farming to reduce the indirect effects of inadequate feeding and, consequently, to increase growth rates and nutrient deposition, obtaining better quality animals. The objective of this study was to establish morphometric growth curves for bullfrog tadpoles (*Lithobates catesbeianus*) and to determine nutrient deposition in the carcass. A total of 6,480 bullfrogs (Gosner stage 25) received an experimental diet (26.23% digestible protein and 32.68% crude protein) and a commercial diet (37.92% crude protein) *ad libitum*. A Gompertz model was used to describe the growth curve. Tadpoles fed the experimental diet presented higher final protein deposition. In addition, the sigmoidal curve was much more homogenous, indicating a more constant daily protein deposition rate. The Gompertz model provided an excellent fit of the data to describe the morphometric growth curve and carcass nutrient deposition of bullfrog tadpoles, showing that animals fed the experimental diet presented a better growth rate and nutrient deposition.

**Key words:** Frog farming, Gompertz model, growth curve, nutrient deposition

### Resumo

Melhorias no manejo alimentar devem ser implementadas na ranicultura, visando diminuir os efeitos indiretos da alimentação inadequada, resultando em melhores taxas de crescimento e deposição de nutrientes, consequentemente obtendo animais de melhor qualidade. O objetivo foi estabelecer curvas de crescimento morfométrico de girinos de rã-touro e sua deposição de nutrientes na carcaça. Foram utilizados 6.480 girinos de rã-touro no estágio 25 de Gosner, alimentados com dieta experimental (26,23% PD e 32,68% PB) e comercial (37,92% PB), oferecida *ad libitum*. O modelo utilizado para descrever a curva de crescimento foi de Gompertz. Os girinos alimentados com a dieta experimental, além de apresentarem uma deposição protéica final maior, o modelo sigmoidal apresentou-se muito mais homogêneo, mostrando uma taxa de deposição protéica diária mais constante. O modelo de Gompertz apresentou um ótimo ajuste para descrição da curva de crescimento morfométrico e deposição de nutrientes na carcaça para girinos de rã-touro, mostrando que os girinos alimentados com a dieta experimental, apresentaram melhor taxa de crescimento e deposição de nutrientes na carcaça.

**Palavras-chave:** Ranicultura, modelo de Gompertz, curva de crescimento, deposição de nutrientes

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## Introduction

The bullfrogs (*Lithobates catesbeianus*) present a complex life cycle (SIMMONS; COSTA; GERSTEIN, 2004; ROBERTS et al., 2009). The first stage (tadpole stage) is essential for good functioning of the frog farm since the animals that emerge after metamorphosis will determine the conditions for frog culture. In this respect, an adequate diet is fundamental for frog rearing since mortality or abnormal development are classical signs of possible nutritional disorders (SEIXAS FILHO et al., 2008).

There are several studies designed to improve frog feeding and nutrition, but most of them only investigated the need for crude protein and energy (CARMONA-OSALDE et al., 1996; SEIXAS FILHO et al., 1998; BARBOSA; SILVEIRA; GOMIDE, 2005). The digestibility of protein and energy of some tadpole dietary components has also been evaluated (ALBINATI et al., 2000; SECCO; STÉFANI; VIDOTTI, 2005). However, the nutritional requirements of tadpoles remain unknown. Because of the lack of these data, diets of other species with different nutritional requirements, such as carnivorous fish, are generally administered to tadpoles (SEIXAS FILHO et al., 2008).

Studies developing growth models, taking into account the information provided by them, are important to implement feeding and genetic breeding programs, which are scarce in frog farming (SANTOS et al., 2007; MARCATO et al., 2010). Non-linear mathematical models describe the growth characteristics of individuals based on the response profile of some parameters over time, thus permitting to identify, for example, heavier animals at a younger age (MARCATO et al., 2010; SILVA et al., 2011). Non-linear models have been described for various domestic animals, such as commercial chicken (MARCATO et al., 2008), Santa Ines sheep (SARMENTO et al., 2006), beef cows (SILVA et al., 2011), Nile tilapia (SANTOS et al., 2007), postmetamorphic pepper frog

(AGOSTINHO et al., 1991) and postmetamorphic bullfrog (RODRIGUES et al., 2007), and have been shown to model the growth of these species with statistical accuracy. This tool has also been adopted to determine the nutritional requirements of animals since it contributes to define the ideal time necessary to reach maximum weight (WAFÁ; PIERRE; DANIEL, 2004). Non-linear models may therefore contribute to the development of a feeding program for bullfrog tadpoles designed to produce large numbers of high-quality froglets, which are of fundamental importance for frog farming (BARBOSA; SILVEIRA; GOMIDE, 2005).

The objective of the present study is to determine the growth curves and rates, as well as the carcass nutrient deposition (protein, fat, minerals, and water), of bullfrog tadpoles using a non-linear Gompertz model.

## Material and Methods

The experiment was conducted at the Laboratory of Aquatic Organism Nutrition, Aquaculture Center, São Paulo State University (UNESP), Jaboticabal, Brazil, between November 2010 and January 2011 (64 days).

### *Animals and experimental conditions*

A total of 6,480 bullfrog tadpoles (*Lithobates catesbeianus*) in stage 25 (GOSNER, 1960) of the same spawn, with an initial weight of  $0.044 \pm 0.001$  g, were obtained from the frog farm of the Aquaculture Center, UNESP. A completely randomized design with two treatments (experimental and commercial diet) and six repetitions was used, in which each experimental repetition consisted of three tanks. The animals were housed in thirty-six 100-L amiantus tanks containing 90 L of water at an initial density of 2 tadpoles/L. The tanks were supplied individually and drained directly through the bottom. The water obtained from a mine was chlorine free and was changed 100% at intervals of

24 h to prevent interferences with the feeding of the animals. Water flow was controlled (SCHMIDT; KNOWLES; SIMMONS, 2011).

For the maintenance of water quality, the tanks were siphoned out on alternate days to remove feces and uneaten food. The maximum and minimum temperatures of the environment and of the tank water were measured daily with a digital thermometer (Incoterm). Dissolved oxygen (YSI Professional Oxygen Meter), conductivity (PHTEK

Pocket Conductivity Meter, model CD-203), and pH (PHTEK Pocket pH Meter, model pH-100) were measured weekly.

#### *Diets and feed management*

The tadpoles were fed two ground diets: an experimental diet containing 26.23% digestible protein (32.68% crude protein) (Table 1) and a commercial diet containing 37.92% crude protein (Table 2).

**Table 1.** Formula and nutritional composition of the experimental diet.

Ingredient	(g.kg <sup>-1</sup> )
Fish meal	180.0
Soybean meal	205.0
Poultry by- product meal	100.0
Wheat meal	170.0
Corn meal	178.8
Corn Starch	100.0
Soybean oil	60.0
Mineral and vitamin premix *	6.0
BHT	0.2
<b>Composition</b>	
Crude protein (g.kg <sup>-1</sup> )	326.8
Digestible protein <sup>2</sup> (g.kg <sup>-1</sup> )	262.3
Gross energy (kcal/kg)	4434.34
Digestible energy (kcal/kg)**	3743.07
Crude fiber (g.kg <sup>-1</sup> )	24.5
Mineral matter (g.kg <sup>-1</sup> )	77.8
Ether extract (g.kg <sup>-1</sup> )	109.8
Nitrogen-free extract (g.kg <sup>-1</sup> )	381.3

\* Moisture content: 20.0 g.kg<sup>-1</sup>; ashes: 716.442 g.kg<sup>-1</sup>; choline: 30,000 mg.kg<sup>-1</sup>; magnesium: 0.0085%; sulfur: 1.1589%; iron: 25,714 mg.kg<sup>-1</sup>; copper: 1,960 mg.kg<sup>-1</sup>; manganese: 13,345 mg.kg<sup>-1</sup>; zinc: 30,000 mg.kg<sup>-1</sup>; iodine: 939 mg.kg<sup>-1</sup>; selenium: 30 mg.kg<sup>-1</sup>; vitamin A: 600,000 IU.kg<sup>-1</sup>; vitamin D3: 600,000 IU.kg<sup>-1</sup>; vitamin E: 12,000 mg.kg<sup>-1</sup>; vitamin K3: 631 mg.kg<sup>-1</sup>; thiamine (vitamin B1): 1,176 mg.kg<sup>-1</sup>; riboflavin (vitamin B2): 1,536 mg.kg<sup>-1</sup>; pyridoxine (vitamin B6): 1,274 mg.kg<sup>-1</sup>; vitamin B12: 4,000 µg.kg<sup>-1</sup>; niacin: 19,800 mg.kg<sup>-1</sup>; pantothenic acid (vitamin B3): 3,920 mg.kg<sup>-1</sup>; folic acid: 192 mg.kg<sup>-1</sup>; biotin: 20 mg.kg<sup>-1</sup>; vitamin C: 40,250 mg.kg<sup>-1</sup>.

\*\* Values calculated based on the digestibility coefficient proposed by Secco, Stéfani and Vidotti (2005).

**Source:** Elaboration of the authors.

Bromatological analysis of the components of the experimental diet was performed at the Laboratory of Aquatic Organism Nutrition,

Aquaculture Center, UNESP, and at the Laboratory of Animal Nutrition, Department of Animal Sciences, FCAV, UNESP.

**Table 2.** Centesimal composition analyzed of the commercial diet.

	Centesimal composition
Crude protein (g.kg <sup>-1</sup> )	379.2
Ether extract (g.kg <sup>-1</sup> )	75.3
Cross energy (kcal/kg)	4156.34
Crude fiber (g.kg <sup>-1</sup> )	33.5
Mineral matter (g.kg <sup>-1</sup> )	109.5

Basic diet composition: ground whole corn, soybean meal, corn gluten meal – 60, meat and bone meal, hydrolyzed feather meal, blood meal, stabilized vegetable fat, sodium chloride (common salt), choline chloride, and limestone. Eventual substitutes: ground whole grain sorghum, broken rice, corn meal, corn gluten meal, wheat bran, rice bran, and dry sugar cane yeast.

Premix (minimum): Vitamin A (min) 35,000 IU; vitamin D3 (min) 2,000 IU; vitamin E (min) 120 IU; vitamin K3 (min) 800 mg; folic acid (min) 10 mg; biotin (min) 10 mg; thiamine (B1) (min) 25 mg; riboflavin (B2) (min) 35 mg; pyridoxine (B6) (min) 40 mg; vitamin 12 (min) 100 µg; niacin (min) 350 mg; pantothenic acid (min) 150 mg; choline (min) 2,500 mg; copper (min) 25 mg; iron (min) 150 mg; manganese (min) 75 mg; selenium (min) 1 mg; zinc (min) 140 mg; mannan oligosaccharide (min) 60 mg.

**Source:** Elaboration of the authors.

The animals were fed *ad libitum* three times per day, avoiding leftovers in such a way the quantity supplied corresponded to the quantity consumed (SOLOMON; TARUWA, 2011).

#### *Variables analyzed*

For the calculation of mean weight and weight gain (final weight – initial weight), 10% of the tadpoles of each experimental tank were randomly selected and weighed individually on a digital electronic balance to the nearest 0.01 g. In addition, the total length (from snout to tail tip) and partial length (snout to tail base) of the tadpoles were measured with a digital caliper. These measurements were obtained on days 1, 13, 23, 33, 42, 55 and 64, last day of the experiment and onset of metamorphic climax (WRIGHT; RICHARDSON; BIGOS, 2011).

Food intake in each experimental tank was quantified by the calculation of apparent feed conversion (food intake/weight gain). Before transfer to the experimental tanks, a batch of tadpoles ( $\pm$  35 g live weight tadpoles) was killed for the analysis of initial body composition (protein crude, ether extract, dry matter, and ash). In the subsequent evaluations (days 13, 23, 33, 42, 55 and 64), samples of 10 g live weight tadpoles were collected from each of three experimental tanks, corresponding to an experimental repetition. The tadpoles selected from the three tanks were transferred to a container with water for 24 h for the elimination of gastrointestinal tract content. Next, the animals were placed on ice for stunning, killed, stored in a plastic container, and frozen for subsequent processing and preparation of laboratory samples.

#### *Sample processing and laboratory analysis*

For analysis of carcass nutrients, the frozen tadpoles were ground in a food processor to obtain homogenous samples. The samples were then transferred to disposable plastic Petri dishes and lyophilized at -50°C in a Thermo VLP200 lyophilizer to obtain pre-dried material. Next, the samples were ground in a ball mill and sent to the laboratory for analysis of protein (ETHERIDGE; PESTI; FOSTER, 1998), ether extract, dry matter, and ash (SILVA; QUEIROZ, 2002).

#### *Estimation of the growth curve and statistical analysis*

A Gompertz model was used to describe the growth curve and body composition (MANSANO et al., 2012), (protein crude, fat, water, and ash) of bullfrog tadpoles:  $W_t = W_m \times \exp \times (- \exp \times (- b \times (t - t^*)))$ , where  $W_t$  = nutrient weight (g)

of the animal at time  $t$ , expressed as a function of  $W_m$ ;  $W_m$  = nutrient weight (g) at maturity of the animal;  $b$  = maturation rate (per day);  $t^*$  = time (days) when the growth rate is maximal. On the basis of the estimated equation, growth rates (g/day) were calculated as a function of time ( $t$ ) by the derivative  $dW_t / dt = b \cdot W_t \cdot \exp(-b \cdot (t - t^*))$  of the equation described by Winsor (1932). When the parameters were adjusted, we used the NLIN procedure of SAS (2001), and the parameter estimates were obtained by iterative modified Gauss-Newton method, developed by Hartley (1961), for non-linear models.

The parameters indicated in the equations of the non-linear mathematical models and feed conversion (observed and estimated) were submitted to F test using procedure of the SAS software (2001).

The experimental procedures were conducted in accordance with the guidelines of the Brazilian College of Animal Experimentation (COBEA) and were approved by the Ethics Committee on Animal Use of São Paulo State University (protocol n° 025000/10).

## Results

### *Physical and chemical characteristics of the water*

The minimum and maximum temperatures of the tank water during the experiment were  $24.2 \pm$

$1.4$  and  $26.0 \pm 1.2^\circ\text{C}$ , respectively.

The mean dissolved oxygen content of the water was  $3.07 \pm 0.92 \text{ mg.L}^{-1}$ , the electrical conductivity of the tank water was  $38 \pm 0.26 \mu\text{S/cm}$  and the mean pH of the tank water was  $6.17 \pm 0.34$ .

### *Growth*

Table 3 shows the parameter estimates obtained with the Gompertz equation for live weight, total length, partial length, food intake, protein intake, and body composition (protein, water, fat, and ashes) of tadpoles fed the different diets.

Tadpoles fed the experimental diet reached a higher final live weight estimated with the Gompertz equation than those receiving the commercial diet (Table 3). However, these differences in live weight were not as marked at the beginning of the experiment, becoming more prominent after day 23 (Figure 1A).

Tadpoles fed the experimental diet (26.23% digestible protein and 32.68% crude protein) presented better daily weight gain (Figure 2A).

The total length of bullfrog tadpoles was not influenced either diet (Table 3 and Figure 1B). In contrast, there was a significant difference in partial length, with the best result being obtained for animals fed the experimental diet which also presented a higher final live weight (Table 3 and Figure 1C).



**Table 3.** Parameter estimates obtained with the Gompertz equation for live weight, food and protein intake, total and partial lengths and nutrient deposition of bullfrog tadpoles fed the experimental (ED) and commercial (CD) diets.

Variable	Diet	Parameter		
		Pm	b (per day)	t*
Live weight (g)	ED	10.66±1.0517a	0.0558±0.0088	38.195±2.2956
	CD	9.54±0.4174b	0.0590±0.0044	37.571±0.9918
P value		0.0028	0.3628	0.3020
Total length (mm)	ED	120.0±3.8715	0.0394±0.0022	21.813±1.0297
	CD	122.1±3.1691	0.0371±0.0016	23.516±0.8630
P value		0.3124	0.2764	0.1046
Partial length (mm)	ED	37.26±1.0098a	0.0415±0.0023	16.465±0.8371
	CD	35.56±0.8304b	0.0425±0.0021	15.978±0.7135
P value		0.0199	0.5618	0.4519
Cumulative food intake (g)	ED	15.19±0.6551	0.0482±0.0026	42.563±1.0919
	CD	15.33±0.5732	0.0485±0.0023	42.656±0.9413
P value		0.5828	0.7863	0.5979
Cumulative protein intake (g)	ED	4.56±0.1970b	0.0482±0.0026	42.563±1.0919
	CD	5.42±0.5732a	0.0485±0.0023	42.655±0.9413
P value		0.0001	0.7863	0.8405
Total body protein (mg)	ED	873.8±0.1837a	0.0478±0.0122	43.759±2.3173
	CD	697.0±0.0373b	0.0672±0.0062	41.271±1.0896
P value		0.0265	0.0817	0.2525
Total body water (mg)	ED	9.103.8±0.8588a	0.0564±0.0088	37.461±2.2084
	CD	8.168.8±0.3603b	0.0599±0.0048	36.467±1.0097
P value		0.0028	0.5940	0.1574
Total body fat (mg)	ED	469.4±0.0864	0.0568±0.0154	43.961±3.9850
	CD	421.5±0.0330	0.0592±0.0061	46.103±1.6829
P value		0.6612	0.4787	0.1197
Total body ash (mg)	ED	195.6±0.0444	0.0443±0.0105	48.064±2.932
	CD	169.6±0.0124	0.0528±0.0043	47.024±1.706
P value		0.1044	0.0545	0.7943

Pm = weight or length at maturity; b (per day) = maturation rate; t\* (days) = time of maximum growth rate. Means in the same column followed by different superscript letters differ significantly ( $P < 0.05$ , F test).

**Source:** Elaboration of the authors.

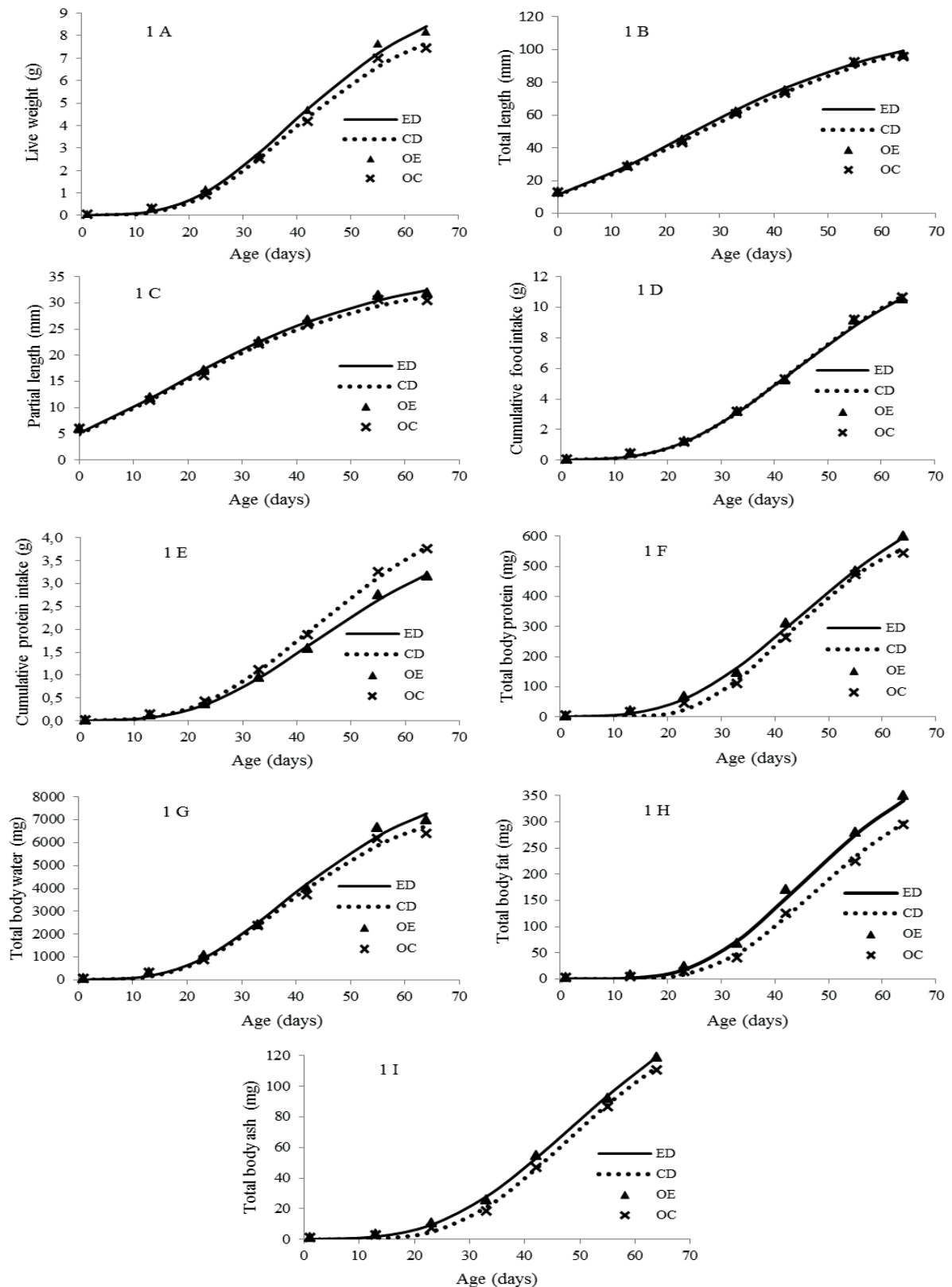
### *Feed intake*

No significant difference in food intake was observed between tadpoles receiving the experimental and commercial diets (Table 3). Estimation of food intake using the Gompertz model provided values similar to those observed (Figure 1D).

### *Feed conversion*

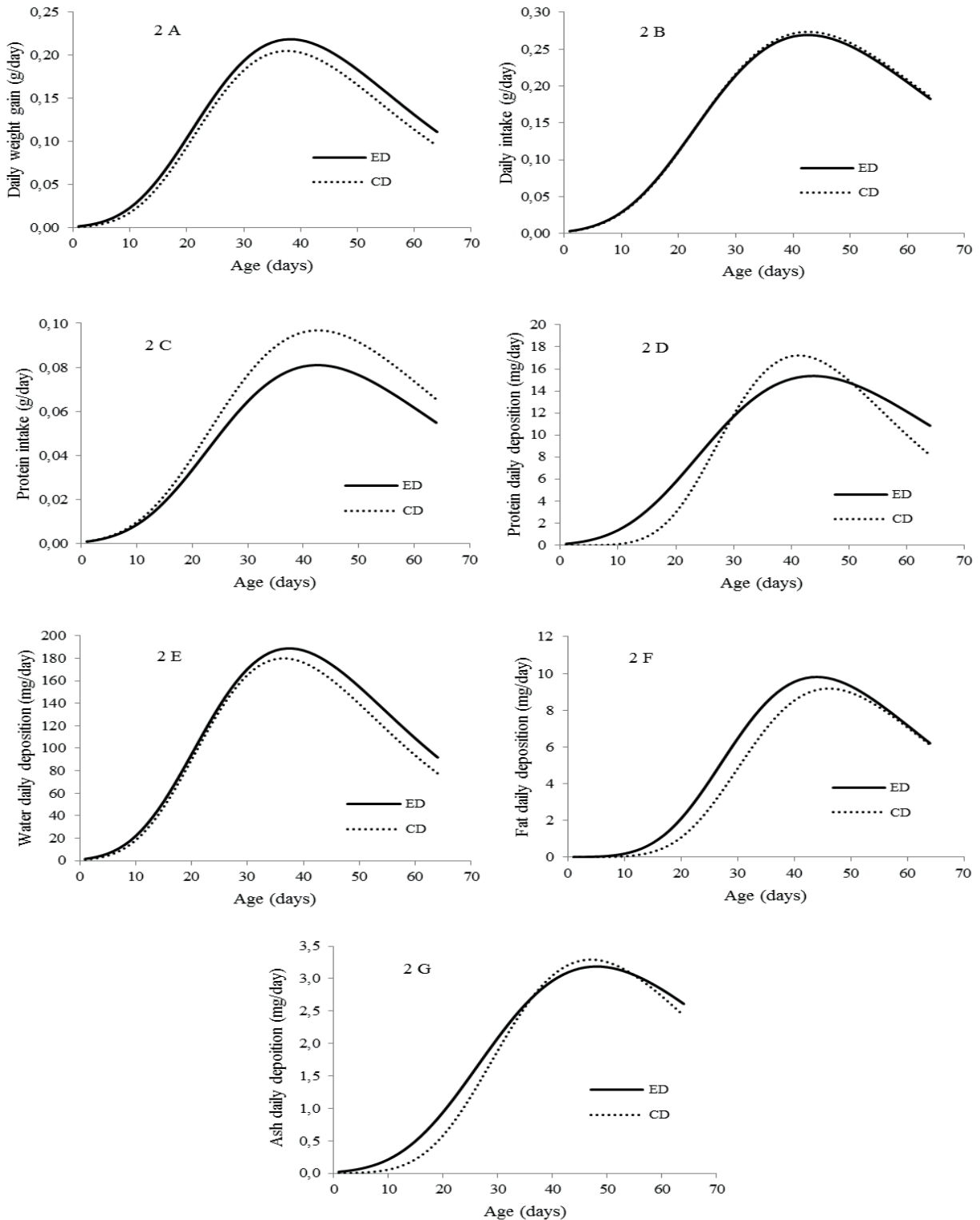
Tadpoles receiving the experimental diet presented better feed conversion than those fed the commercial diet despite the lower protein content of the former diet ( $p < 0.05$ ) (Table 4).

**Figure 1.** Daily weight gain (A), daily food (B) and protein (C) intake, and daily deposition of protein (D), water (E), fat (F) and ash (G) of bullfrog tadpoles fed the experimental (ED) and commercial (CD) diets.



Source: Elaboration of the authors.

**Figure 2.** Curve of live weight (A), total length (B), partial length (C), cumulative food (D) and protein (E) intake, total protein (F), total water (G), total fat (H), and total ash (I) obtained for bullfrog tadpoles fed the experimental (ED) and commercial (CD) diets. Observed values for the experimental (OE) and commercial (OC) diets.



Source: Elaboration of the authors.



**Table 4.** Apparent feed conversion observed and estimated by equation of Gompertz, for bullfrog tadpoles fed the experimental and commercial diets.

Diet	Mean feed conversion	
	Observed	Estimated
Experimental	1.28 ± 0.07 <sup>a</sup>	1.26 ± 0.04 <sup>a</sup>
Commercial	1.43 ± 0.05 <sup>b</sup>	1.40 ± 0.03 <sup>b</sup>

Means in the same column followed by different superscript letters differ significantly ( $P < 0.05$ , F test).

**Source:** Elaboration of the authors.

#### *Protein intake and carcass nutrient deposition*

Tadpoles fed the commercial diet presented significantly higher protein intake since the diet contained higher amounts of crude protein (Table 3 and Figure 1E). However, daily protein intake rate (b) and time of maximum growth ( $t^*$ ) were the same for the two diets (Table 3). Although tadpoles receiving the commercial diet had consumed more crude protein, protein deposition was significantly higher in the carcass of animals fed the experimental diet (Table 3 and Figure 1F). This finding indicates that, despite the lower protein content of the experimental diet (26.23% digestible protein and 32.68% crude protein) compared to the commercial diet (37.92% crude protein), the protein of the former was better utilized by the animals, suggesting that it is good quality protein.

On the basis of Figure 1E and 1F, a correlation was observed between protein intake and total carcass protein, with protein retention being lower in tadpoles fed the commercial diet (Figure 2D), although these animals had consumed larger daily amounts of protein (Figure 2C). In addition, the sigmoidal model was much more homogenous for tadpoles fed the experimental diet despite higher final protein deposition, indicating more constant daily protein deposition (Figure 2D).

No significant difference in carcass fat deposition was observed between tadpoles fed the different diets (Table 3). The gradual increase in fat deposition was due mainly to the growth of the fat body during

tadpole development. As can be seen in Figure 1H, fat deposition in the carcass occurred later than protein and water deposition. Although tadpoles fed the experimental diet presented an increase in daily fat deposition at the beginning of the experiment, no significant difference was observed between groups (Table 3) and the rate was similar at the end of the experiment (Figure 2F).

## **Discussion**

### *Physical and chemical characteristics of the water*

These values are within the range considered to be optimal for tadpole rearing (24.5 to 29.1°C) (LIMA; CASALI; AGOSTINHO, 2003; BAMBOZZI et al., 2004; HAYASHI et al., 2004; SEIXAS FILHO et al., 2008). The water temperature interferes directly with the metabolism of the animals since tadpoles are ectotherms, with favorable temperature conditions increasing the growth rate and body weight gain of the animals (HOFFMANN; LEBOUTE; SOUZA, 1989).

The mean dissolved oxygen content of the water ( $3.07 \pm 0.92$  mg/L) was within the acceptable range proposed by Hailey et al. (2006) for tadpole rearing. The electrical conductivity of the tank water ( $38 \pm 0.26$   $\mu$ S/cm) was within the range recommended by Sipaúba-Tavares (1994) for the culture of aquatic organisms in ponds (23.0 to 71.0  $\mu$ S/cm). The mean pH of the tank water ( $6.17 \pm 0.34$ ) was close to the 6.5 reported by Albinati, Lima and Donzele (2001).

The water quality was adequate for tadpole rearing since organic matter deposited at the bottom of the tanks was removed and food supply was controlled to prevent excess residues at the bottom of the tank.

### *Growth*

Weight at maturity ( $W_m$ ) is a parameter that expresses genetically the potential of development and the interactions of genes that determine growth,

with the asymptotic measurement becoming a parameter resulting from previous growth stages (SILVA et al., 2004).

The protein content of this diet differs from the 55% crude protein reported by Camona-Osalde et al. (1996), 45 and 55% crude protein used by Seixas Filho et al. (2010), and 36.8% digestible protein reported by Hayashi et al. (2004). Dietary protein content affects the growth rate of tadpoles and the metamorphic climax can only be achieved when the tadpole fulfills its nutritional requirements, permitting them to transform into a new and completely different animal (CARMONA-OSALDE et al., 1996). This nutritional dependence has also been reported by Martínez, Herráez and Álvarez (1994) for *Rana perezi*. The authors observed that inadequate feeding prolonged the larval period.

Growth rates are highly flexible when environmental interactions influence the size of the animals, with specific growth and mortality being the fundamental factors that characterize the growth of a population (WERNER; GILLIAM, 1984). It is widely accepted that amphibians and reptiles exhibit indeterminate growth and that body size and age are therefore positively correlated (DUELLMAN; TRUEB, 1994).

The pattern of the tadpole growth curve differs markedly from that of postmetamorphic animals. Tadpoles have a rapid growth rate (MONELLO et al., 2006) and the exponential function of the Gompertz model fits the data well (SOCKMAN et al., 2008). The parametric variables of the model can be generated by various biological processes, such as the supply, distribution and access of the animal to food, variations in feed quality, or animal heterogeneity causing competition, factors that interfere with the growth curve and deposition of nutrients (HOTA, 1994).

#### *Feed intake*

Figure 2B illustrates the daily food intake of bullfrog tadpoles. A marked decline in food intake was observed after day 42 as a result of the onset of metamorphic climax. This phase is especially stressful for tadpoles and is characterized by complex anatomophysiological modifications (WRIGHT; RICHARDSON; BIGOS, 2011), including the change from omnivorous to carnivorous feeding habits. During this stage, the tadpole stops eating and feeds on body fat and tail reserves (OLIVEIRA-BAHIA, 2007).

#### *Feed conversion*

This finding demonstrates that the experimental diet is better for tadpoles as indicated by the higher weight gain of these animals. Since tadpoles are omnivores (OLIVEIRA-BAHIA, 2007), high-protein diets are not required for better development.

The present results indicate clear improvement of productivity when compared to the data reported by Lima and Agostinho (1992) and Lima, Casali and Agostinho (2003) (mean feed conversion ratio of 1.5), with the experimental diet providing a mean feed conversion ratio of 1.28. These results may contribute to a reduction in production costs, which correspond to 57% of total costs of frog feeding (LIMA; AGOSTINHO, 1992).

#### *Protein intake and carcass nutrient deposition*

According to Reginatto et al. (2000), the higher and longer the plateau of protein deposition in an animal, the more efficient is the meat production and the better its body composition. Tadpole metamorphosis should not occur before the animals have stored a minimum amount of energy. Under conditions of high tadpole density, the animals are likely to spend more energy to prevent physical

encounters and other associated stresses. In addition, less energy is probably stored if reserves are necessary to combat growth inhibitors under conditions of high density, for example, in intensive commercial tadpole culture systems (GRUMP, 1981). Even if there is a superabundance of food, competition is expected and a balanced diet is therefore necessary to meet the nutritional needs of tadpoles (MARTÍNEZ; ÁLVAREZ; HERRÁEZ, 1996).

Diets elaborated for animal feeding should contain a balance of protein and energy so that the animal grows without converting excess protein into energy or depositing large amounts of adipose tissue in the carcass (ALBINATI et al., 2001). However, protein and energy sources represent the largest cost of diet production and any imbalance can lead to economic losses.

The deposition rate of body water is intimately related to the rate of body protein deposition (REGINATTO et al., 2000), demonstrating that tadpoles start to retain more water during this period as a result of higher protein synthesis. Water deposition was significantly higher in tadpoles fed the experimental diet (Table 3 and Figure 1G), a finding that is directly related to the higher protein deposition observed in this group.

An association between water and fat deposition rates was observed at the time when deposition of the two nutrients declined (Figure 2E and 2F), in agreement with the findings of Silva; Albino and Nascimento (2003). The decline in water deposition was much more critical, i.e., the animal deposits more fat than water during the final phase, showing that the higher the quantity of body water, the less fat the animal will present.

Maximum ash deposition was the last to occur when compared to the other nutrients, with no significant difference in carcass ash deposition between tadpoles fed the two diets (Table 3). This finding demonstrates similar bone formation in the two groups (Figures 2G and 1I) and synchronous

growth. Muscle tissue growth occurs before adipose tissue growth which, in turn, is more accelerated than bone tissue growth since in tadpoles the hind limbs and forelimbs develop during the final stage of metamorphosis. As a consequence, the highest mineral deposition in bone tissue is observed at the end of growth (GARTNER; HIATT, 2007).

## Conclusion

The Gompertz model provided a good fit of the data to describe the morphometric growth curve and carcass nutrient deposition of bullfrog tadpoles. A higher growth rate and nutrient deposition were observed for tadpoles receiving the experimental diet (26.23% digestible protein).

## Acknowledgements

We thank the state funding agency Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP) for financial support.

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