Phyllomedusa 20(1):75–88, 2021

ISSN 1519-1397 (print) / ISSN 2316-9079 (online)
doi: http://dx.doi.org/10.11606/issn.2316-9079.v20i1p75-88

Feeding habits of *Lithobates megapoda* (Anura: Ranidae), a threatened leopard frog used for human consumption, in Lake Chapala, Mexico

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Abstract

Feeding habits of *Lithobates megapoda* (Anura: Ranidae), a threatened leopard frog used for human consumption, in Lake Chapala, Mexico. The Big-footed Leopard Frog (*Lithobates megapoda*) is a threatened and endemic species from western Mexico. This species has aquatic habits and it is distinguished by its large size, particularly by the length of its legs, reason for which it is captured for human consumption. Also, knowledge about its natural history is scarce and incomplete. In this study, we analyzed the composition of the diet of *L. megapoda* on the shore of Lake Chapala, the largest lake in Mexico. A total of 69 adult individuals were collected lifeless in fishing nets, during the rainy season (June-October), of which 48 had stomach contents. A total of 96 prey items were identified, which correspond to 13 prey categories. Fish constituted the most dominant prey category in the diet in terms of number, volume, frequency of occurrence, and relative importance. No significant differences were found in the consumption by prey type (aquatic or terrestrial). However, the aquatic preys had a greater relative importance and were more voluminous than the terrestrial ones. The diversity of prey categories, in terms of prey volume, indicates males may have a higher dietary diversity than females, but we not evaluated possible bias. In addition, a significant effect was found in the interaction of size (SVL) of frogs with the average of prey volume. Females (that are larger than males) consumed prey within a wide volume range and, the larger they are, more voluminous are prey. In this way it is possible that intraspecific competition for trophic resources in the environment is reduced. This study helps us understand the trophic ecology of *L. megapoda*, a frog species that plays an important role in the food web where it lives, as a predator feeding on aquatic and terrestrial organisms.

Keywords: Amphibians, diet, fish, intersexual variation, trophic ecology.

Received 27 March 2020
Accepted 15 June 2021
Distributed June 2021

Resumen

Hábitos alimenticios de *Lithobates megapoda* (Anura: Ranidae), una rana leopardo amenazada utilizada para consumo humano, en el Lago de Chapala, México. La rana patona (*Lithobates megapoda*) es una especie endémica y amenazada del occidente de México. Esta especie es de hábitos acuáticos y se caracteriza por su gran tamaño, particularmente por la longitud de sus ancas, razón por la cual es capturada para consumo humano. Además, el conocimiento sobre su historia natural es escaso e incompleto. En este estudio, se analizó la composición de la dieta de *L. megapoda* en las inmediaciones del Lago de Chapala, el lago más grande de México. Un total de 69 individuos adultos fueron recolectados sin vida en redes de pesca, durante la época de lluvias (junio-octubre), de los cuales 48 tuvieron contenido estomacal. En total se identificaron 96 presas, las cuales corresponden a 13 categorías de presa. Los peces constituyeron la categoría más valiosa en la dieta en cuanto a número, volumen, frecuencia de ocurrencia e importancia relativa. No se encontraron diferencias significativas en el consumo por tipo de presa (acuática o terrestre). Sin embargo, las presas acuáticas tuvieron mayor importancia relativa y fueron más voluminosas que las terrestres. La diversidad de categorías de presa, en términos de volumen de presa, indica que los machos pueden tener una mayor diversidad dietaría que las hembras, pero no evaluamos un posible sesgo. Además, se encontró un efecto significativo en la interacción del tamaño (SVL) de las ranas con el volumen promedio de presa. Las hembras (más grandes que los machos) consumieron presas dentro de un rango de volumen amplio y, entre más grandes son, más voluminosas son las presas. De esta forma es posible que se reduzca la competencia intraespecífica por los recursos tróficos en el ambiente. Este estudio nos permite comprender la ecología trófica de *L. megapoda*, una especie que juega un papel importante en la red alimenticia donde vive, como depredador de organismos acuáticos y terrestres.

Palabras clave: anfibios, dieta, ecología trófica, peces, variación intersexual.

Resumo

Hábitos alimentares de *Lithobates megapoda* (Anura: Ranidae), uma rã-leopardo ameaçada utilizada para consumo humano, no Lago Chapala, México. A rã-leopardo-de-pés-grandes (*Lithobates megapoda*) é uma espécie ameaçada e endémica do México ocidental. Essa espécie tem hábitos aquáticos e distingue-se pelo seu grande tamanho, particularmente pelo comprimento das patas, razão pela qual é capturada para consumo humano. Além disso, o conhecimento sobre sua história natural é escasso e incompleto. Neste estudo, analisamos a composição da dieta de *L. megapoda* nas margens do Lago Chapala, o maior lago do México. Um total de 69 indivíduos adultos foram coletados mortos em redes de pesca durante a época das chuvas (Junho-Outubro), dos quais 48 apresentravam conteúdo estomacal. Foi identificado um total de 96 itens, o que corresponde a 13 categorias de presas. Os peixes constituíram a categoria de presas mais dominante na dieta em termos de número, volume, frequência de ocorrência e importância relativa. Não foram encontradas diferenças significativas no consumo por tipo de presa (aquática ou terrestre). No entanto, as presas aquáticas apresentaram maior importância relativa e eram mais volumosas que as terrestres. A diversidade de categorias de presas, em termos de volume de presas, indica que os machos podem exibir uma maior diversidade alimentar do que as fêmeas, mas não avaliamos possíveis vieses. Além disso, foi encontrado um efeito significativo na interacção do tamanho (SVL) das rãs com a média do volume de presas. As fêmeas (que são maiores do que os machos) consumiram presas dentro de uma vasta gama de volumes, e quanto maiores, mais volumosas as presas. Dessa forma, é possível que a competição intraespecífica por recursos alimentares no ambiente seja reduzida. Este estudo ajuda-nos a compreender a ecologia trófica de *L. megapoda,* uma espécie de anuro que desempenha um papel importante na teia alimentar do ambiente em que vive, como predador que se alimenta de organismos aquáticos e terrestres.

Palavras-chave: anfíbios, dieta, ecologia trófica, peixes, variação intersexual.

Introduction

Amphibians, particularly anurans, are an important component of the trophic structure of ecosystems because they are abundant, show high biomass (Gibbons *et al*. 2006), and constitute a link between aquatic and terrestrial communities (Stebbins and Cohen 1995, Duré *et al*. 2009), acting both as predators (e.g., Luría-Manzano and Ramírez-Bautista 2017, 2019) and as preys of a wide variety of organisms (e.g., Fulgence *et al.* 2020, Nuñez-Escalante and Garro-Acuña 2021). Therefore, knowing the feeding habits of anurans help us to understand their role on the energy flow and on the interactions of the food webs in which they are involved (Duellman and Trueb 1994, Luría-Manzano and Ramírez-Bautista 2017). Besides ecological importance, dietary aspects of anurans may have implications for conservation. For example, knowledge of dietary diversity is necessary to detect those species with narrow trophic niches, which are potentially vulnerable to changes in the structure of the prey community they consume (Luría-Manzano and Ramírez-Bautista 2019).

Most anurans are considered generalist consumers because they feed on a wide prey range (Duellman and Trueb 1994, Solé and Rödder 2010). However, there is evidence that some of them have a narrow diet, showing a specialized consumption on certain biological groups (Agudelo-Cantero *et al*. 2015, Lopes *et al*. 2017, Araujo-Vieira *et al*. 2018). Differences in the diet composition have been related to different factors, which may be extrinsic, such as the site traits (Bonansea and Vaira 2007), competition (Duellman and Trueb 1994), or seasonality (Berazategui *et al.* 2007), which can offer different food resources, or intrinsic factors, such as ontogenetic changes (Whitfield and Donnelly 2006), reproductive behavior, and sex (Lamb 1984, Maneyro *et al.* 2004), which influence the nutritional requirements of anurans. Both extrinsic and intrinsic factors are related to the use of food resources and the variation in the diet composition between and within species (Lima and Magnusson 2000).

Lithobates megapoda (Taylor, 1942), commonly known as Big-footed Leopard Frog (Liner and Casas-Andreu 2008), is an endemic anuran species to western Mexico in the states of Nayarit, Jalisco, Guanajuato, and Michoacán (Webb 1996). Its large body size, with females reaching a snout–vent length of up to 157 mm (Webb 1996) and males of up to 117 mm (Taylor and Smith 1945), makes this species one of the largest anurans in Mexico. This frog is predominantly aquatic and inhabits and breeds in perennial water sources, in shrubland and pine-oak forest from 823 to 1,520 m a.s.l. (IUCN SSC Amphibian Specialist Group 2020). In addition, this species is threatened mainly due to habitat destruction and degradation, water pollution, droughts due to climate change, introduction of exotic species (IUCN SSC Amphibian Specialist Group 2020), and extraction for human consumption (Barragán-Ramírez *et al.* 2017, IUCN SSC Amphibian Specialist Group 2020). These impacts have led to this frog being currently protected by Mexican law NOM-059-SEMARNAT-2010 under the Special Protection category (Pr) (SEMARNAT 2010) and considered as Near Threatened by the IUCN (IUCN SSC Amphibian Specialist Group 2020). Despite this, basic information about its natural history is scarce.

In this study, we evaluated dietary composition of adult individuals of *L. megapoda* on the shore of Lake Chapala.

Materials and Methods

Study Site

Lake Chapala is the largest and most important lake in Mexico, it is located between the boundaries of the states of Jalisco and Michoacan (20º14' N, 103º00' W, elevation 1,524 m a.s.l., Figure 1). It has a surface area of 1,146 km^2 (28 km from north to south and 79 km from east to west), a maximum storage capacity of

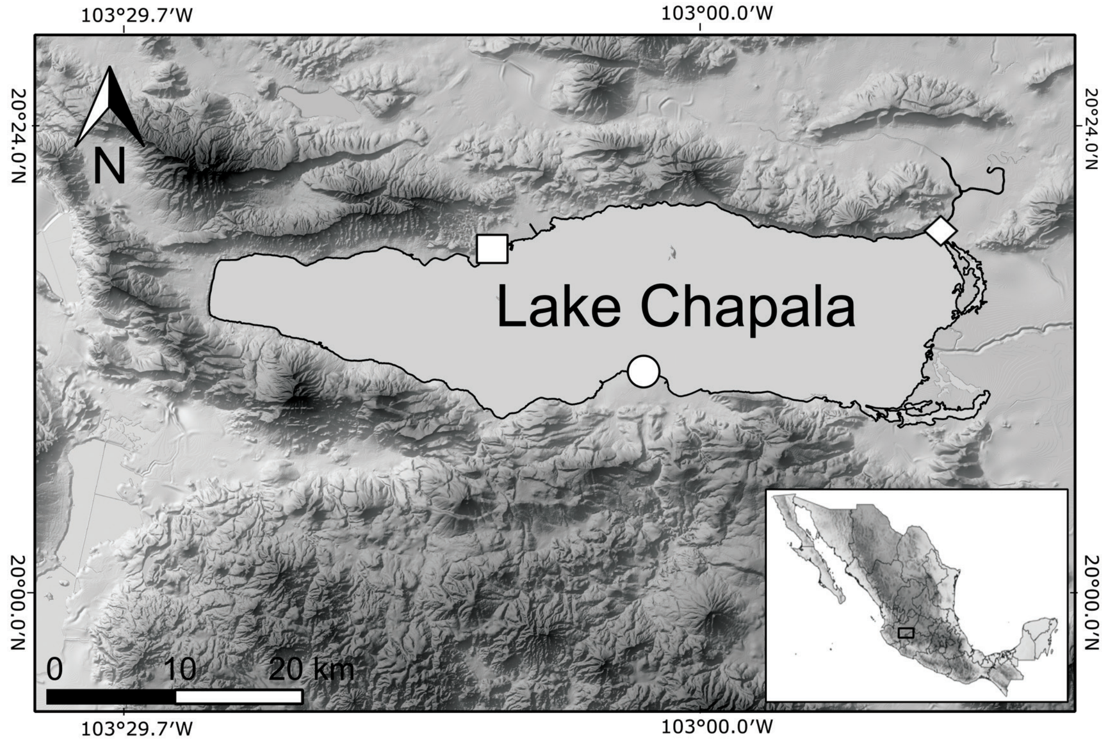

Figure 1. Map showing the sampled areas of Chapala (square), Tizapan El Alto (circle) and Ocotlan (diamond) along the Lake Chapala, state of Jalisco, Mexico.

7,897 m^3 and a maximum of 8 m of depth (CEA Jalisco 2020). Mean annual temperature in the region is 19.9ºC and varies from 9 to 30ºC. Annual rainfall is estimated at 735 mm (Filonov *et al.* 2001), with most rains occurring between June and September.

Data Collection

Sixty-nine adult individuals of *Lithobates megapoda* were examined for the present study. All individuals were found dead in fishing nets during the rainy season (June-October) in different years. Eleven frogs were collected in Ocotlán (20°19'06" N, 102°47'20" W) between September-October 2020; forty-seven frogs collected in the vicinity of Chapala (20°17'38" N, 103°10'40" W) between June and October 2016, and the rest of the individuals were captured between June and September 2015: seven in Tizapán El Alto (20°09'04" N, 103°02' 48" W) and four from Ocotlán (Figure 1). All frogs were collected using a special permit SGPA/DGVS/03444/15 issued by Mexican Ministry of Environment (Secretaría de Medio Ambiente y Recursos Naturales). Specimens were fixed in 10% formalin solution and preserved in 70% alcohol after data collection and deposited in a vertebrate scientific collection at Centro de Estudios en Zoología, Universidad de Guadalajara (CZUG).

Each individual was measured, weighed and its sex was identified. A Mitutoyo® digital caliper was used to record snout–vent length

(SVL) (to the nearest 0.01 mm). Body mass was measured using an Ohaus® digital balance (± 0.1 g of accuracy). Values are presented as mean ± standard deviation (SD). Maturity and sex of individuals were determined either by secondary sexual characters (presence of vocal sacs and nuptial pads in males; absent in females) or by gonadal identification after dissection. We removed the stomach of each individual and the content was placed in a Petri-dish and examined under a stereomicroscope (ZEISS Stemi DV4). Prey items were classified taxonomically to the Order level (prey category) using the keys of Coronado and Márquez (1977) and Triplehorn and Johnson (2005); when possible, items were identified to the genus and/or species level. Only in vertebrate prey, taxonomic Class level is equal to "prey category". Later, we counted and classified each prey according to its habits: aquatic or terrestrial. Prey volume was estimated by fluid displacement to the nearest 0.1 mL (Magnusson *et al.* 2003). Subsequently, the units in mL were converted to cm^3. Stomach contents were preserved in 70% ethanol and stored in the entomological collection at Centro de Estudios en Zoología of the Universidad de Guadalajara (CZUG). Furthermore, those prey items that were too fragmented or that could not be identified due to advanced stage of digestion, were included in the "undetermined remains" category.

Data Analyses

In order to determine the contribution of each prey category, we calculated an index of relative importance using the following formula I = (F% + N% + V%)/3, where F%, N% and V% are the percentages of frequency, number of prey and prey volume, respectively (Biavati *et al.* 2004). Differences in consumption per prey type (aquatic vs. terrestrial) and volume of prey consumed were analyzed with Mann-Whitney U-test. To determine differences in diet composition between the sexes by prey categories in terms of abundance and volume prey, we constructed contingency tables and a chi-square test (χ^2) was applied.

Diet diversity was estimated with the Shannon index (Shannon and Weaver 1949) using the number of items and volume of each prey category as diversity attributes: $H' = -\sum_{i=1}^{S} p_i \ln p_i$, where p_i is the proportion of the resource *i* (prey category) in the diet and *S* is the total number of prey categories. The exponential to Shannon's diversity values was calculated in order to represent the results in terms of effective species number (true diversity), sensu Jost (2006) : ${}^1D$ = exp (H′), where ${}^1D$ represented the dietary diversity observed (${}^1D_{obs}$). Additionally, we estimated dietary diversity (${}^1D_{est}$) with a Jacknife estimator (Zahl 1977) and statistically compared it among sexes by calculating 95% confidence intervals.

The relationship SVL-average of prey volume, SVL-total prey volume, and SVL-prey number were analyzed under a Covariance Analysis structure (ANCOVA) where a Generalized Linear Model (GLM) was used with a Gamma distribution as error and a square-root link function. Volume and prey number were selected as dependent variables and the sex as covariate. All statistical analyses were performed using R software (R Core Team 2017).

Results

Diet Composition

We obtained 69 adult individuals of *Lithobates megapoda*, 40 females and 29 males. Females had a snout–vent length (SVL) of 123.30 ± 18.52 mm and a body mass of 289.0 ± 118.4 g. Males had a SVL of 96.67 ± 9.33 mm and body mass 132.7 ± 35.1 g. Females were larger (Mann-Whitney U = 458, $p < 0.001$) and heavier (Mann-Whitney U = 382, $p < 0.001$) than males, with more than twice the mass of males on average (Table 1).

A total of 48 individuals had stomach content (29 females and 19 males); 42 with at least one identifiable prey item and six with only plant

Table 1. Snout–vent length (SVL), body mass and dietary parameters for females and males of *Lithobates megapoda.*

	Females		Males	
	Mean ± SD (range)	*N*	Mean ± SD (range)	*N*
SVL (mm)	123.30 ± 18.52 (77.10–150.88)	40	96.67 ± 9.33 (80.02–112.76)	29
Body mass (g)	289.0 ± 118.4 (57.0–567.0)	40	132.7 ± 35.1 (65.0–208.0)	29
Number of prey per stomach	2.4 ± 2.0 (1–8)	27	2.1 ± 1.9 (1–8)	15
Prey volume (cm^3)	0.81 ± 1.41 (0.02–7.86)	65	0.19 ± 0.31 (0.01–1.52)	31
Stomach content volume (cm^3)	2.06 ± 2.12 (0.09–7.86)	29	0.57 ± 0.47 (0.03–1.70)	19

material and non-identifiable remains. We identified 96 prey items belonging to 13 prey categories: 11 invertebrates and only two vertebrates (anurans and Actinopterygii fishes) (Table 2). The mean number of prey items per stomach was 2.00 ± 1.95 (range 1–8). We found a range of 0.01 to 7.86 cm^3 of prey volume per stomach, and a range of 0.03 to 7.86 cm^3 of volume of stomach contents. Fish were the most dominant prey category in terms of frequency (50.1%), number (36.5%) and volume (57.3%), which comprised two taxonomic orders: Cyprinodontiformes and Perciformes. Also, this food category was the most important in the diet of *L. megapoda*, $I = 48.0$ (Table 2, Figure 2). Araneae was the second most important prey category ($I = 12.5$) and Hemiptera was the third one ($I = 7.8$). Plant material and undetermined remains were also part of the diet, these were found in six (12.5%) and 25 (52.1%) stomachs and they constitute 2.6% and 14.3% of the total volume ingested, respectively (Table 2).

The consumption by prey type, aquatic and terrestrial, comprised 43.8% and 56.2% of the total number of prey items, respectively (Table 2). Frogs with only identifiable prey items in their stomach ($N = 42$) showed a range of 0–7 terrestrial prey per stomach, followed by a range of 0–5 aquatic prey per stomach, with no differences between the consumption of both prey types (Mann-Whitney U = 875, $p = 0.95$). Regarding the total volume of ingested prey, aquatic preys (76.4%) surpassed the terrestrial ones (23.6%). The volume range of aquatic prey consumed was 0.04–7.86 cm^3 ($N = 42$), while that of the terrestrial prey was 0.01–3.00 cm^3 ($N = 54$). Terrestrial preys were smaller (volumetrically) than the aquatic ones (Mann-Whitney U = 1868, $p < 0.001$). The index of relative importance showed that aquatic preys were more important than terrestrial in the diet of *L. megapoda* (Table 2).

Diet Variation

Analyzing the prey consumption data by sex, we found differences in the prey volume (Mann-Whitney U = 1542, $p = -0.001$) and in the volume of stomach content (Mann-Whitney U = 431, $p = -0.001$). Females consumed bulky prey and showed a greater volume of stomach content than males (Table 1). Also, we found differences in the numerical ($X^2 = 45.95$, df = 12, $p < 0.001$) and volumetric ($X^2 = 60.53$, df = 12, $p < 0.001$) proportions of prey categories consumed between the sexes. Fish (Actinopterygii) were the most important prey category in the diet of females ($I = 52\%$) and males ($I = 25.4\%$) (Table 3). Dietary diversity (^1D) observed and estimated, using the abundance of prey categories, was similar in both sexes (females $^1D_{obs} = 6.68$, $^1D_{est} = 7.72$; males $^1D_{obs} = 6.68$, $^1D_{est} = 7.72$). However, when analyzing dietary diversity using prey volume, we found that males show greater diversity ($^1D_{obs} = 5.23$, $^1D_{est} = 12.14$) than females ($^1D_{obs} = 2.81$, $^1D_{est} = 3.22$) (Figure 3).

Table 2. Diet composition of *Lithobates megapoda* (*N* = 48) on the shore of Lake Chapala, Jalisco, Mexico. *N*, Number of individuals; *V*, Prey volume (cm³); *F*, Frequency of occurrence of each prey item; *I*, Index of relative importance. *Endemic to Lake Chapala basin and the surrounding area; **Introduced to Lake Chapala.

Taxonomic Class/ Prey category	**Prey type**	***N***	***N%***	***F***	***F%***	***V***	***V%***	***I***
INVERTEBRATES								
Arachnida								
Araneae		14	14.6	10	20.8	1.45	2.1	12.5
Lycosidae	T	6	6.3	5	10.4	1.16	1.6	6.1
Tetragnathidae	T	6	6.3	4	8.3	0.26	0.4	5.0
Unidentified	T	2	2.1	2	4.2	0.03	0.0	2.1
Opiliones								
Sclerosomatidae								
Leiobunum sp.	T	1	1.0	1	2.1	0.10	0.1	1.1
Diplopoda								
Polydesmida								
Paradoxosomatidae	T	5	5.2	2	4.2	0.39	0.6	3.3
Hexapoda								
Coleoptera		7	7.3	5	10.4	1.55	2.2	6.6
Chrysomelidae								
Disonycha sp.	T	1	1.0	1	2.1	0.01	<0.1	1.0
Omophoita sp.	T	1	1.0	1	2.1	0.01	<0.1	1.0
Scarabaeidae								
Phyllophaga sp.	T	1	1.0	1	2.1	0.26	0.4	1.2
Xyloryctes sp.	T	2	2.1	2	4.2	1.13	1.6	2.6
Staphylinidae								
Platydracus sp.	T	1	1.0	1	2.1	0.04	0.1	1.1
Unidentified	T	1	1.0	1	2.1	0.10	0.1	1.1
Dermaptera								
Forficulidae								
Doru sp.	T	9	9.4	3	6.3	1.07	1.5	5.7
Hemiptera		6	6.3	6	12.5	3.38	4.8	7.8
Belostomatidae (adult)	A	2	2.1	2	4.2	2.24	3.2	3.1
Belostomatidae (nymph)	A	2	2.1	2	4.2	0.84	1.2	2.5
Notonectidae	A	1	1.0	1	2.1	0.10	0.1	1.1
Unidentified	T	1	1.0	1	2.1	0.20	0.3	1.1
Hymenoptera		3	3.1	3	6.3	0.15	0.2	3.2
Apidae								
Apis mellifera	T	2	2.1	2	4.2	0.12	0.2	2.1
Formicidae								
Atta mexicana	T	1	1.0	1	2.1	0.03	< 0.1	1.1

Table 2. *Continued.*

Taxonomic Class/ Prey category	**Prey type**	***N***	***N%***	***F***	***F%***	***V***	***V%***	***I***
Lepidoptera								
Unidentified (larvae)	T	2	2.1	2	4.2	0.12	0.2	2.1
Odonata		7	7.3	6	12.5	2.22	3.1	7.6
Coenagrionidae								
Argia sp.	T	2	2.1	2	4.2	0.09	0.1	2.1
Libellulidae	T	2	2.1	1	2.1	0.48	0.7	1.6
Unidentified (adult)	T	2	2.1	2	4.2	1.38	2.0	2.7
Unidentified (nymph)	A	1	1.0	1	2.1	0.27	0.4	1.2
Orthoptera								
Unidentified	T	4	4.2	3	6.3	6.76	9.6	6.7
Crustacea								
Isopoda								
Armadillidiidae	T	2	2.1	1	2.1	0.11	0.2	1.4
VERTEBRATES								
Amphibia								
Ranidae								
Lithobates sp. (larvae)	A	1	1.0	1	2.1	0.91	1.3	1.5
Actinopterygii (Fishes)		35	36.5	24	50.1	40.43	57.3	48.0
Cyprinodontiformes		29	30.2	21	43.8	26.32	37.3	37.1
Goodeidae								
*Chapalichthys encaustus**	A	1	1.0	1	2.1	0.07	0.1	1.1
Goodea atripinnis	A	1	1.0	1	2.1	3.95	5.6	2.9
Unidentified	A	3	3.1	3	6.3	4.51	6.4	5.3
Poeciliidae								
Poecilia sp.**	A	7	7.3	6	12.5	12.34	17.5	12.4
Poeciliopsis infans	A	3	3.1	3	6.3	0.38	0.5	3.3
Non-identified	A	14	14.6	9	18.8	5.07	7.2	13.5
Perciformes		6	6.3	3	6.3	14.11	20.0	10.8
Cichlidae								
Oreochromis sp.**	A	5	5.2	2	4.2	12.91	18.3	9.2
Non-identified	A	1	1.0	1	2.1	1.20	1.7	1.6
Aquatic preys (A)	-	42	43.8	29	60.4	44.79	63.5	55.9
Terrestrial preys (T)	-	54	56.2	24	50.0	13.85	19.6	42.0
Plant material	-	-	-	6	12.5	1.86	2.6	-
Undetermined remains	-	-	-	25	52.1	10.07	14.3	-
Totals	-	**96**	**100**	**48**	**100**	**70.57**	**100**	-

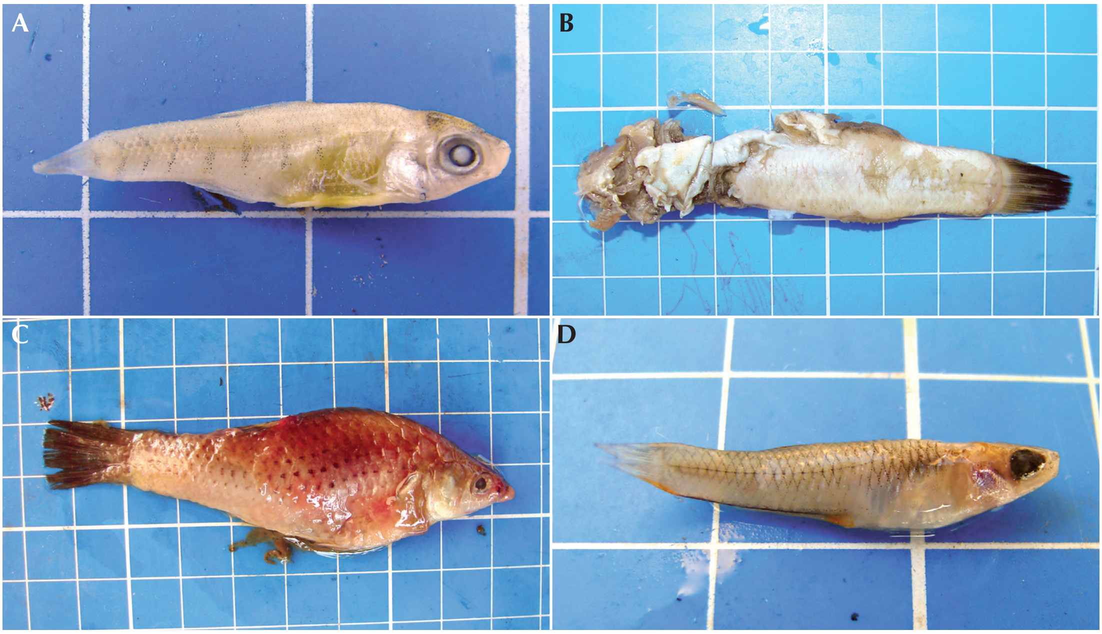

Figure 2. Some fish samples extracted from the stomach contents of *Lithobates megapoda*. Goodeidae: (**A**) *Chapalichthys encaustus*, (**B**) *Goodea atripinnis*. Poecilidae: (**C**) *Poecilia* sp., (**D**) *Poeciliopsis infans*. Scale: each square = 1 cm².

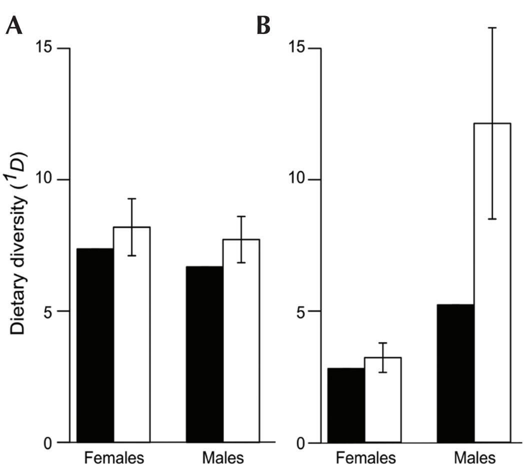

Figure 3. Dietary diversity (¹D) with abundance (**A**), and prey volume (**B**) in males and females of *Lithobates megapoda*. Dark bars = observed dietary diversity; white bars = estimated dietary diversity. Error bars are 95% confidence intervals.

We found correlation between the average of prey volume and SVL ($F_{1,40}$ = 60.009; $p < 0.001$), which was different between the sexes ($F_{1,38}$ = 54.1; $p < 0.001$), with a negative trend in males and a positive trend in females (Figure 4A). We found differences between total prey volume per SVL ($F_{1,46}$ = 57.61; $p < 0.001$) and sex ($F_{1,45}$ = 46.98; $p < 0.001$), with females presenting larger volumes of prey in their stomach than males of similar size (Figure 4B). No differences were found in the correlation of number of prey and SVL ($F_{1,40}$ = 52.38; p = 0.54) and sex ($F_{1,39}$ = 50.77; p = 0.2) (Figure 4C).

Discussion

The results of this study indicate that the diet of *Lithobates megapoda* on the shore of Lake Chapala, during the rainy season, is composed of a wide variety of prey, as occurs with other

Table 3. Intersexual diet composition of *Lithobates megapoda* (*N* = 48) on the shore of Lake Chapala, Jalisco, Mexico. *N*, Number of individuals; *V*, Total volume of preys (cm^3); *F*, Frequency of occurrence of each prey item; *I*, Index of relative importance.

Prey class/Prey order	**Females (*N* = 29)**				**Males (*N* = 19)**			
	N (%)	*FO (%)*	*V (%)*	*I (%)*	*N (%)*	*FO (%)*	*V (%)*	*I (%)*
Arachnida	7 (10.7)	7 (24.1)	0.72 (1.1)	12.0 (10.9)				
Araneae	6 (9.2)	6 (20.7)	0.62 (1.0)	10.3 (9.1)	8 (25.8)	4 (21.1)	0.83 (7.7)	18.2 (20.6)
Opiliones	1 (1.5)	1 (3.4)	0.10 (0.1)	1.7 (1.5)	-	-	-	-
Diplopoda (Polydesmida)	5 (7.7)	2 (6.9)	0.39 (0.7)	5.1 (4.6)	-	-	-	-
Hexapoda	21 (32.4)	12 (41.4)	12.00 (20.0)	31.3 (28.5)	17 (54.8)	11 (57.9)	3.25 (30.0)	47.6 (54.0)
Coleoptera	4 (6.2)	3 (10.3)	1.43 (2.4)	6.3 (5.6)	3 (9.7)	2 (10.5)	0.12 (1.1)	7.1 (7.7)
Dermaptera	4 (6.2)	1 (3.4)	0.96 (1.6)	3.7 (3.3)	5 (16.1)	2 (10.5)	0.11 (1.0)	9.2 (10.0)
Hemiptera	3 (4.6)	3 (10.3)	1.62 (2.7)	5.9 (5.2)	3 (9.7)	3 (15.8)	1.76 (16.2)	13.9 (15.2)
Hymenoptera	2 (3.1)	2 (6.9)	0.12 (0.2)	3.4 (3.0)	1 (3.2)	1 (5.3)	0.03 (0.3)	2.9 (3.2)
Lepidoptera	2 (3.1)	2 (6.9)	0.12 (0.2)	3.4 (3.0)	-	-	-	-
Odonata	3 (4.6)	2 (6.9)	1.52 (2.5)	4.7 (4.1)	4 (12.9)	4 (21.1)	0.7 (6.5)	13.5 (14.7)
Orthoptera	3 (4.6)	2 (6.9)	6.23 (10.4)	7.3 (6.4)	1 (3.2)	1 (5.3)	0.53 (4.9)	4.5 (4.9)
Crustacea (Isopoda)	2 (3.1)	1 (3.4)	0.11 (0.2)	2.2 (2.0)	-	-	-	-
Anura	1 (1.5)	1 (3.4)	0.91 (1.5)	2.1 (1.9)	-	-	-	-
Actinopterygii	29 (44.6)	18 (62.0)	38.68 (64.7)	57.1 (52.0)				
Cyprinodontiformes	23 (35.4)	15 (51.7)	24.57 (41.1)	42.7 (37.7)	6 (19.4)	6 (31.6)	1.75 (16.1)	22.4 (25.4)
Perciformes	6 (9.2)	3 (10.3)	14.11 (23.6)	14.4 (12.7)	-	-	-	-
Plant material	-	4 (13.8)	1.05 (1.8)	-	-	2 (10.5)	0.81 (7.5)	-
Undetermined remains	-	12 (41.4)	5.87 (9.8)	-	-	13 (68.4)	4.2 (38.7)	-
Totals	**65 (100)**	**29 (100)**	**59.73 (100)**	-	**31 (100)**	**19 (100)**	**10.84 (100)**	-

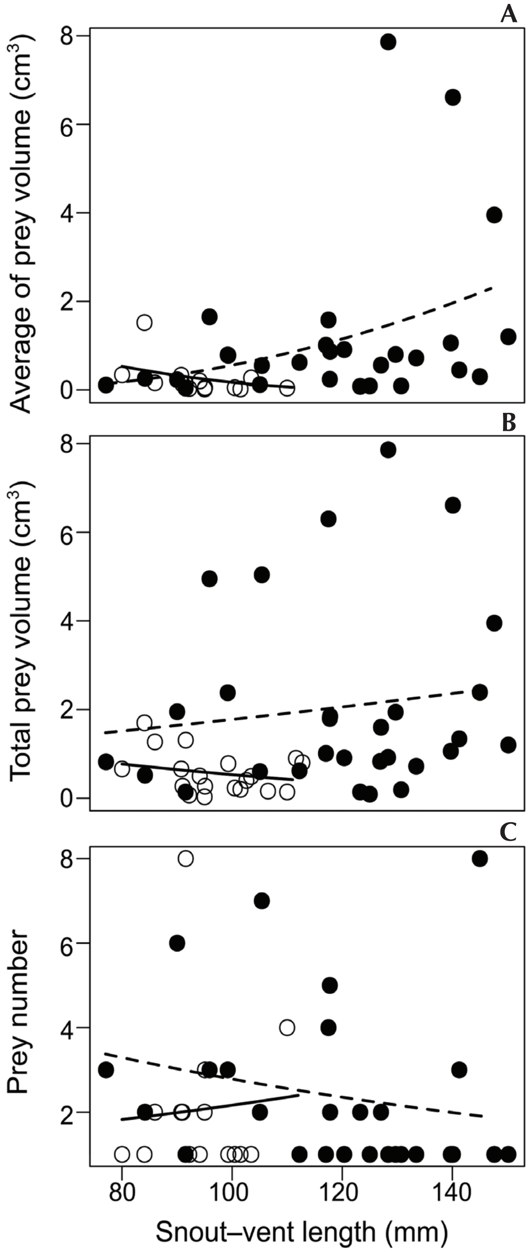

Figure 4. Correlation between snout–vent length (SVL) of adults *Lithobates megapoda* and (**A**) average of prey volume, (**B**) total prey volume, and (**C**) number of prey consumed. Open circles and trend lines represent males and solid circles and dashed lines represent females.

ranids that are considered generalist-opportunistic predators (Hirai and Matsui 1999, Wu *et al.* 2005). However, *L. megapoda* shows a preference for fish consumption, which according to the relative importance index is the most valuable prey category in the diet of this frog. Because its diet includes both aquatic and terrestrial organisms, this frog species represents a link between aquatic and terrestrial communities where it plays an important role in the food web.

Unlike other frogs that can feed on fish, *Lithobates megapoda* shows a high percentage of frequency of fish consumption (~ 50%), which is above that reported in other ranids in Mexico (with less than 5%) as *L. berlandieri* (Baird, 1859) and *L. johni* (Blair, 1965) (Hernández-Austria *et al.* 2019), *L. brownorum* (Sanders, 1973) (Ramírez-Bautista and Lemos-Espinal 2004), *L. vaillanti* (Brocchi, 1877) (Ramirez *et al.* 1998, Ramírez-Bautista and Lemos-Espinal 2004, Luría-Manzano and Ramírez-Bautista 2019), and *L. zweifeli* (Hillis, Frost, and Webb, 1984) (Mendoza-Estrada *et al.* 2008). We need further studies to evaluate the high contribution of fish as food source for *L. megapoda*. It can be related to different factors, such as opportunistic habits (Premo and Atmowidjojo 1987, Hirai and Matsui 1999, Dietl *et al.* 2009), digestibility (Secor *et al.* 2007) promoting greater nutritional contribution (Das 1996), especially during reproductive period due to the energy expenditure involved (Lamb 1984, Ryser 1989), or high availability in the habitat (Parker and Goldstein 2004, López *et al.* 2009).

We recorded at least five species of fishes in the diet of *L. megapoda*, three are native to Lake Chapala: *Chapalichthys encaustus* (Jordan and Snyder, 1899) (endemic to Lake Chapala basin), *Goodea atripinnis* Jordan, 1880, and *Poeciliopsis infans* (Woolman, 1894). The other two are non-native species, *Poecilia* sp. and *Oreochromis* sp., the latter known as "Tilapia" and native to Africa (Miller *et al.* 2009, Moncayo-Estrada *et al.* 2012). All these fishes are frequent in areas with aquatic vegetation (Miller *et al.* 2009), where *L. megapoda* is commonly found. Aquatic prey, which were more valuable than terrestrial prey according to the relative importance index, included along with fish, organisms such as belostomatids, damselfly nymph, and a tadpole, indicating the ability to capture prey underwater, as already suggested for other ranids (Stewart and Sandison 1972). The presence of a congeneric tadpole in one of the stomachs indicates the occurrence of cannibalism, reinforcing the opportunistic behavior of this frog. Similar cases have been widely reported in other ranids (e.g., Mendoza-Estrada *et al.* 2008, Silva *et al.* 2009, Alvarez 2013).

The presence of terrestrial preys in the diet of *L. megapoda* suggests that individuals forage near the lake margins, a behavior also documented to other ranids (e.g., Ramírez-Bautista and Lemos-Espinal 2004). The floating and aquatic vegetation constitute an important habitat for different invertebrates, like spiders and dragonflies (Akamatsu *et al.* 2004), which were important prey in the diet of *L. megapoda* after fish.

Considering the low percentage of occurrence and volume in the stomachs, the ingestion of plant material is probably accidental, as reported for other ranids (Hedeen 1972, Kramek 1972, Mendoza-Estrada *et al.* 2008).

Dietary diversity based on abundance seems similar between sexes, as has happened in other species of anurans (e.g., Hirai and Matsui 1999, Silva *et al.* 2009, Luría-Manzano and Gutiérrez-Mayén 2014), but females seem to consume prey with a wider volume range, usually larger ones when compared to males of similar size. Unfortunately, we did not access prey availability or sex or size variation among the three different collection sites within the Lake Chapala, and the results should be considered cautiously, since sample biases are possible. The effect of sex and size of the frogs may suggest larger intraspecific competition between large mature males and smaller young females. Additionally, foraging behavior of males and females may vary. An interesting future approach would be to compare the diet composition of adults out of the rainy season (which is also the breeding season) to see if differences remain.

Acknowledgments

We thank Agustín Camacho-Rodríguez for its valuable support in the fish identification and the CONACyT for support of the PhD scholarship to J. L. Barragán-Ramírez. We are grateful to the anonymous reviewers for their comments and suggestions to improve the manuscript.

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Editor: Vanessa K. Verdade