Geographic and seasonal variation analysis of digestive morphology in the catfish *Iheringichthys labrosus* along lower Río Uruguay

Alejandro D'Anatro¹Nicolás Vidal²⁻⁴Iván González-Bergonzoni²⁻⁴Franco Teixeira de Mello⁴Jukka Tana⁵Daniel E Naya¹

¹Departamento de Ecología y Evolución, Facultad de Ciencias, Universidad de la República, Montevideo, Uruguay; ²Department of Bioscience, Aarhus University, Silkeborg, Denmark; ³Sino-Danish Centre for Education and Research (SDC), Beijing, People's Republic of China; ⁴Grupo de Ecología y Rehabilitación de Sistemas Acuáticos, Departamento de Ecología y Evolución, CURE-Facultad de Ciencias, Universidad de la República, Maldonado, Uruguay; ⁵ÅF-Consult Ltd, Environmental Consulting, Espoo, Finland

Abstract: The study of geographic variation of individual traits is an important component of evolutionary research, in that individual morphological features can be subjected to multiple selective pressures. The present work is aimed to analyze the geographic and seasonal variation in several digestive traits in *Iheringichthys labrosus* along three localities in the lower Río Uruguay. Statistically significant differences among locations and between seasons were found for all the variables assessed, the most noticeable of differences being observed were intestinal length, intestine weight, and liver weight. In part, these differences could be an adaptive response to changes in food availability and/or in the energetic costs of reproduction. Results obtained herein also suggest that individuals of this species spend enough time in each locality as to show a consistent response to local conditions in their digestive traits morphology.

Keywords: *Iheringichthys labrosus*, physiological flexibility, Río Uruguay

Introduction

Geographic analysis of variation in individual traits is an important component of evolutionary research since individual morphological features can be subjected to multiple selection pressures, often leading to tradeoffs in the optimization of their form–function relationship.¹ Because the gut represents the functional link between energy intake and metabolizable energy, it could be expected that gut size would be under strong selection pressures.² In this sense, reversible adjustments in the size of the digestive organs, ie, digestive flexibility, is a widespread phenomenon among vertebrates, and the congruence between empirical data and optimal digestion models strongly supports the idea that it has evolved by natural selection.^{3,4-8}

However, studies comparing digestive morphology among individuals belonging to different geographical sites, and having different trophic habits, are by far scarcer than studies analyzing digestive flexibility within populations. In any event, existing studies indicate (1) there is an important amount of variation in digestive features among populations and (2) there is agreement with digestion theory,² in the sense of there is a positive correlation between the size of the digestive organs and the amount of refractory material in the diet.⁹⁻¹⁴ Nevertheless, current information regarding the link between individual resource use and digestive trait morphology comes mainly from tetrapods, and only limited attention has been paid to fish populations,^{1,15-18} despite implications that fish represent an excellent model organism to study adaptive phenotypic plasticity.¹⁹

Correspondence: Alejandro D'Anatro
Departamento de Ecología y Evolución,
Facultad de Ciencias, Universidad de la
República. Montevideo 11400, Uruguay
Tel +598 2 525 8618 ext 143
Fax +598 2 525 8617
Email passer@fcien.edu.uy

Within this context, the present work is aimed at analyzing the geographic and seasonal variation in several digestive traits in *Iheringichthys labrosus* (Lütken 1874) along three localities in the lower Río Uruguay. Our focal species is one of the most abundant and widely distributed siluriform fish along Río Paraná and Río Uruguay basins,²⁰ and it has been recently proposed as a sentinel species for environmental impact assessments.²¹ Nevertheless, only a few studies considering this fish species were carried out in the Río Uruguay,^{22–24} an area where important agricultural (eg, soybeans monocultures), forest (eg, eucalyptus monocultures), and industrial (eg, pulp mill industries) activities have been increasing during the last decade.

Materials and methods

Study area and sample collection

The study area comprises three localities along the lower Río Uruguay (Figure 1): Fray Bentos, an area with putative anthropogenic impact; Nuevo Berlín, northwards to Fray Bentos; and Las Cañas, southwards to Fray Bentos locality, all situated within a Ramsar zone and the protected area of Parque Nacional Esteros de Farrapos e Islas del Río Uruguay. Surveys of these sites indicate that water physicochemical parameters such as temperature, dissolved oxygen, turbidity, conductivity, and pH did not differ among localities (Table 1). Individuals of the *I. labrosus* species were collected during November 2009 in Las Cañas (n = 17), and then during April 2010 in Las Cañas (n = 26), Nuevo Berlín (n = 21), and Fray Bentos (n = 29). For the purpose of collection we used eight standard Nordic gill nets (5.0, 6.25, 8.0, 10.0, 12.5, 15.5, 19.5, 24, 29, 35, 43 and 55 mm knot-to-knot, each of them 2.5 m

long); four of them were set parallel to the littoral zone and the remaining four were set parallel to each other in the pelagic area, from sunset to sunrise, (approximately 12 hours).

Morphological determinations and statistical analysis

Standard length of each fish was measured with a plastic rule (± 0.1 cm), and then stomach, intestine, liver, and gonads were removed. Stomach and intestine lengths were measured with a plastic rule (± 0.1 cm), then they were washed with running water, dried with paper towels, and weighed with an electronic balance (± 0.001 g). Total weight of liver and eviscerated body weight were also measured as described above.

Comparisons among collection sites were carried out with reference to samples obtained during April 2010, while comparisons among collection periods were conducted using individuals sampled in Las Cañas locality. In both cases, differences in length and weight of internal organs were separately studied by means of analyses of covariance, using standard length (for organ lengths) or eviscerated body weight (for organ masses) as covariate in order to remove the size-effect over the variables analyzed. Prior to all analysis of covariance (ANCOVA) analyses, we evaluated the interaction between the categorical and continuous predictors in order to choose the appropriate model. Post hoc comparisons were evaluated by means of Unequal N HSD tests. In order to obtain a proxy of the reproductive status of individuals between different times of sampling, we also analyzed the temporal variation in the gonadosomatic index ($[\text{gonad weight}/\text{total body weight}] \times 100$) for females from Las Cañas using an

Figure 1 Sampling localities of *Iheringichthys labrosus* analyzed in this work.

Notes: Approximate geographic coordinates: Nuevo Berlín 32°58'52"S, 58°04'06"W; Fray Bentos 33°06'35"S, 58°16'39"W; Las Cañas 33°09'48"S, 58°21'43"W.

Table 1 Descriptive values of water physicochemical parameters for each locality, measured monthly from July to December of 2009 and compared with Kruskal–Wallis test

	Nuevo Berlín		Fray Bentos		Las Cañas		$H_{(2, 18)}$	P-value
	Median	Range	Median	Range	Median	Range		
Oxygen (mg/L)	8.97	4.10	9.04	4.66	8.96	5.08	0.056	0.973
Conductivity (μ S/cm)	50.65	45.30	55.65	50.00	56.25	48.10	1.338	0.512
pH	7.53	1.22	7.35	0.79	7.40	1.24	0.667	0.716
Temperature ($^{\circ}$ C)	18.70	12.80	18.15	12.50	17.60	12.60	1.004	0.605

Notes: The parameter, $[H_{(2, 18)}]$ is related to the Kruskal–Wallis test, and is commonly associated with this analysis. The H subindices represents: 2 = degrees of freedom of the analysis and 18 = sample size used.

analysis of variance (ANOVA) test. All data analyzed were examined for assumptions of normality and homoscedasticity using Kolmogorov–Smirnov and Levene tests ($P < 0.05$), respectively. In all cases, data were log-transformed to meet the assumptions of the analyses.

Results

Statistically significant differences among localities were found for all the variables assessed, with the most noticeable differences observed for intestinal length, intestine weight, and liver weight (Table 2). In almost all variables, individuals from Nuevo Berlín showed the greatest values, followed by individuals from Las Cañas, and then by individuals from Fray Bentos (Table 2). Temporal analysis also revealed significant differences for intestinal length and liver weight, with individuals collected in November 2009 showing lower values than those collected in April 2010 (Table 3). Finally, gonadosomatic index also showed statistically significant differences between sampling periods, but in this case, individuals collected in November 2009 showed greater gonadosomatic index values than those sampled during April 2010 (Table 3).

Discussion

Fish have been known to exhibit developmental plasticity in response to prey type, and the consequences of this plasticity have been highlighted in several studies.^{19,25–29} For instance,

it is accepted that this developmental plasticity would allow species to become morphologically specialized in response to local environmental conditions.¹⁹ In addition, more recent work demonstrates that reversible, flexible changes in digestive traits during adult life of individuals could be achieved over a period of few days in response to nutritional changes.^{16,30–32} In line with this idea, results obtained here showed a clear differentiation in the morphology of the digestive traits among collection sites as well as between collection periods, particularly considering the intestinal length and liver weight.

Regarding the spatial analysis, variation in digestive traits among sites agreed with previous studies that considered *I. labrosus* as a generalist feeder, with a wide feeding spectrum,^{33,34} which would enable the species to adapt its digestive system to the most abundant, or available, prey items in each locality. However, the recorded pattern of variation for digestive traits did not clearly match existing data on *I. labrosus* diet for the same three sampled localities. In this sense, Masdeu,³³ based on a correspondence analysis, proposed that adult individuals from Las Cañas were mainly associated with the consumption of Bivalvia, while individuals from the other two localities were mainly associated with the consumption of Diptera and Ostracoda. By contrast, the digestive features recorded here indicate that individuals from all the three sites differ among them to a similar extent.

Table 2 Descriptive statistics of the traits analyzed in different localities

Variable	Nuevo Berlín			Fray Bentos			Las Cañas		
	Mean	SD	N	Mean	SD	N	Mean	SD	N
SL (cm)	15.8 ^a	3.7	21	12.6 ^b	2.3	29	15.4 ^a	3.7	26
EWB (g)	59.866 ^a	39.084	21	27.975 ^b	14.796	29	51.054 ^a	26.622	26
ESTL (cm)	1.2 ^a	0.2	21	1.3 ^b	0.2	29	1.2 ^a	0.2	26
IL (cm)	18.1 ^a	2.8	21	17.7 ^b	3.4	29	17.8 ^c	2.7	26
SW (g)	0.242 ^a	0.051	21	0.234 ^b	0.077	29	0.240 ^c	0.048	26
IW (g)	0.309 ^a	0.087	21	0.271 ^b	0.131	29	0.262 ^c	0.083	26
LW (g)	0.310 ^a	0.054	21	0.327 ^b	0.082	29	0.293 ^c	0.052	26

Notes: ^{a–c}indicate statistically significant differences among groups ($P < 0.05$ after a Tukey HSD test).

Abbreviations: Mean, absolute mean (for SL and EWB) or least squares-adjusted means (for remaining variables); N, sample size; SD, standard deviation; SL, standard length; EWB, eviscerated body weight; ESTL, stomach length; IL, intestinal length; SW, stomach weight; IW, intestinal weight; LW, liver weight.

Table 3 Descriptive statistics of the temporal analyses of traits

Variable	2009			2010			P-value
	Mean	SD	N	Mean	SD	N	
SL (cm)	13.5	4.3	17	15.4	3.7	26	0.288
EWB (g)	1.265	0.457	17	51.054	26.622	26	0.127
ESTL (cm)	1.3	0.2	17	1.2	0.2	26	0.284
IL (cm)	19.4	2.5	17	17.5	2.5	26	0.026
SW (g)	0.264	0.056	17	0.240	0.056	26	0.106
IW (g)	0.394	0.092	17	0.262	0.092	26	0.103
LW (g)	0.302	0.061	17	0.294	0.062	26	0.022
GSI	0.473	0.095	8	0.333	0.095	21	0.002

Notes: Values in bold indicate statistically significant differences among groups ($P < 0.05$ after a Tukey HSD test).

Abbreviations: Mean, absolute mean (for SL, EWB and GSI) or SL-adjusted means (for remaining variables); SD, standard deviation; N, sample size; SL, standard length; EWB, eviscerated body weight; ESTL, stomach length; IL, intestinal length; SW, stomach weight; IW, intestinal weight; LW, liver weight; GSI, gonadosomatic index.

Moreover, individuals from Las Cañas occupied an intermediate position – in the morphological space – in relation to individuals from the other two collection sites. Given that the study of Masdeu³³ was not conducted at the same time as our study, temporal change in food availability and consumption among sites cannot be disregarded.

On the other hand, temporal analysis showed that there is a clear seasonal variation in digestive traits and also in the gonadosomatic index. Previous work analyzing the reproduction period of this species in Brazil had shown a peak of maturity from October to January.^{35,36} If this is also the case for *I. labrosus* of lower Río Uruguay, the sampling scheme of this work would correspond with the beginning and the end of the spawning season of this species. The clear reduction in the gonadal size between November 2009 and April 2010 is also in line with this idea (see Table 3). Thus, it is possible that the increase in gut length and liver weight that occurred during April 2010 was associated with the major energy demands linked to reproductive efforts in this species, as is the case of other fish³⁷ and other ectothermic vertebrates³⁸.

In summary, *I. labrosus* showed geographical and temporal variation in digestive trait morphology, which could be, in part, an adaptive response to changes in food availability and/or in the energetic costs of reproduction. The results obtained here also suggest that individuals of this species spend enough time in each locality to show a consistent response to local conditions in the morphology of their digestive trait. Such a finding is remarkable, taking into account the limited geographic area covered in this study. Finally, further analyses are needed in order to elucidate the factors driving the trophic and morphological differences exhibited by *I. labrosus* sampled in these localities.

Acknowledgments

We extend thanks to Diego Larrea for field assistance, to Carolina Abud and two anonymous referees for valuable comments on the draft, and to Gervasio González of UPM SA for providing environmental data of Río Uruguay, as well as logistical support during the development of this study. AD, DEN, and FTM are supported by Agencia Nacional de Investigación e Innovación (ANII, Uruguay).

Disclosure

The authors report no conflicts of interest in this work.

References

- Schaack S, Chapman LJ. Foraging ecology of the African cyprinid, *Barbus neumayeri*: the role of hypoxia in intraspecific variation. *Environ Biol Fishes*. 2004;70:95–105.
- Sibly RM. Strategies of digestion and defecation. In: Townsend C, Calow P, editors. *Physiological Ecology: an Evolutionary Approach to Resource Use*. Oxford: Blackwell Scientific Publications; 1981:109–139.
- Starck JM. Structural flexibility of gastro-intestinal tract of vertebrate – implications for evolutionary morphology. *Zoologischer Anzeiger*. 1999;238:87–101.
- McWilliams SR, Karasov WH. Phenotypic flexibility in digestive system structure and function in migratory birds and its ecological significance. *Comp Biochem Physiol A Mol Integr Physiol*. 2001; 128(3):579–593.
- Naya DE, Bozinovic F. Digestive Phenotypic flexibility in post-metamorphic amphibians: studies on a model organism. *Biol Res*. 2004;37(3):365–370.
- Karasov WH, McWilliams SR. Digestive constraint in mammalian and avian ecology. In: Starck JM, Wang T, editors. *Physiological and Ecological Consequences of Feeding in Vertebrates*. Enfield, NH: Science Publishers; 2005:88–112.
- Starck JM. Structural flexibility of the digestive system of tetrapods – patterns and processes at the cellular and tissue level. In: *Physiological and Ecological Consequences of Feeding in Vertebrates*. Starck JM, Wang T, editors. Enfield, NH: Science Publishers; 2005:175–200.
- Karasov WH, Martínez del Río C, Caviedes-Vidal E. Ecological physiology of diet and digestive systems. *Annu Rev Physiol*. 2011;73: 69–93.
- Bozinovic F, Rojas JM, Maldonado K, Sabat P, Naya DE. Between-population differences in digestive flexibility in the olivaceous field mouse. *Zoology (Jena)*. 2010;113(6):373–377.
- Corp N, Gorman ML, Speakman JR. Apparent absorption efficiency and gut morphometry of wood mice, *Apodemus sylvaticus*, from two distinct populations with different diets. *Physiol Zool*. 1997;70(6): 610–614.
- Hansson L. Geographic differences in bank voles *Clethrionomys glareolus* in relation to ecogeographical rules and possible demographic and nutritive strategies. *Ann Zool Fennici*. 1985;22:319–328.
- Hansson L, Jaarola M. Body size related to cyclicity in microtines: dominance behaviour or digestive efficiency? *OIKOS*. 1989;55:356–364.
- Naya DE, Veloso C, Bozinovic F. Interpopulation variation in the digestive tract of *Bufo spinulosus* along an altitudinal (and dietary) gradient. *Ann Zool Fennici*. 2009;46:16–20.
- Sassi PL, Borghi CE, Bozinovic F. Spatial and seasonal plasticity in digestive morphology of cavies (*Microcavia australis*) inhabiting habitats with different plant qualities. *J Mammal*. 2007;88(1):165–172.
- Horn MH, Gawlicka AK, German DP, Logothetis EA, Cavanagh JW, Boyle KS. Structure and function of the stomachless digestive system in three related species of New World silverside fishes (Atherinopsidae) representing herbivory, omnivory, and carnivory. *Mar Biol*. 2006;149:1237–1245.

16. Olsson J, Quevedo M, Colson C, Svanbäck R. Gut length plasticity in Eurasian perch: into the bowels of resource polymorphisms. *Biol J Linnean Soc.* 2007;90(3):517–523.
17. Abelha MCF, Agostinho AA, Goulart E. Plasticidade trófica em peixes de água doce. *Acta Scientiarum.* 2001;23(2):425–434. Portuguese.
18. D'Anatro A, Naya DE, Lessa EP, Defeo O. Contrasting patterns of morphological variation with dietary preferences in Micropogonias furnieri: insights from stable-isotopes and digestive-trait analyses. *J Fish Biol.* 2013;82(5):1641–1658.
19. Wainwright PC. Ecomorphology of prey capture in fishes. In: Saksena E, editor. *Advances in Ichthyological Research.* Gwalior, India: Jiwaji University Press; 1999:375–387.
20. Burges WE. *An Atlas of Freshwater and Marine Catfishes: A Preliminary Survey of the Siluriformes.* Neptune, NJ: T. F. H. Publications; 1989.
21. Saizar C, Lucia B, Clemente J, et al. Línea de base para evaluar el impacto de una planta de celulosa en el Río Uruguay. *INNOTEC.* 2010;5:11–22. Spanish.
22. Tellechea JS, Teixeira-de Mello F, González-Bergonzoni I, Vidal N. Sound production and pectoral spine locking in a neotropical catfish (*Iheringichthys labrosus*, Pimelodidae). *Neotropical Ichthyology.* 2011;9(4):889–894.
23. Masdeu M, Teixeira-de Mello F, Loureiro M, Arim M. Feeding habits and morphometry of *Iheringichthys labrosus* (Lütken, 1874) in the Uruguay River (Uruguay). *Neotropical Ichthyology.* 2011;9(3):657–664.
24. Teixeira de Mello F, Vidal N, Gonzalez-Bergonzoni I, Iglesias C. Length-weight relationships of eight fish species from the lower section of the Uruguay River (Río Negro, Uruguay). *J Appl Ichthyol.* 2009;25(1):128–129.
25. Buddington RK, Chen JW, Diamond J. Genetic and phenotypic adaptation of intestinal nutrient transport to diet in fish. *J Physiol.* 1987;393:261–281.
26. Benavides AG, Cancino JM, Ojeda FP. Ontogenetic changes in gut dimensions and macroalgal digestibility in the marine herbivorous fish, *Aplodactylus punctatus*. *Funct Ecol.* 1994;8:46–51.
27. Muñoz AA, Ojeda FP. Ontogenetic changes in the diet of the herbivorous *Scartichthys viridis* in a rocky intertidal zone in central Chile. *J Fish Biol.* 2000;56(4):986–998.
28. Drewe KE, Horn MH, Dickson KA, Gawlicka A. Insectivore to frugivore: ontogenetic changes in gut morphology and digestive enzyme activity in the characid fish *Brycon guatemalensis* from Costa Rican rain forest streams. *J Fish Biol.* 2004;64(4):890–902.
29. Teixeira de Mello F, Iglesias C, Borthagaray AI, et al. Ontogenetic allometric coefficient changes: implications of diet shift and morphometric traits in *Hoplias malabaricus* (Bloch) (Characiforme, Erythrinidae). *J Fish Biol.* 2006;69(6):1770–1778.
30. Gawlicka AK, Horn MH. Storage and absorption in the digestive system of carnivorous and herbivorous pricklyback fishes (Teleostei: Stichaeidae): ontogenetic, dietary, and phylogenetic effects. *Physiol Biochem Zool.* 2005;78(6):879–892.
31. German DP, Neuberger DT, Callahan MN, Lizardo NR, Evans DH. Feast to famine: the effects of food quality and quantity on the gut structure and function of a detritivorous catfish (Teleostei: Loricariidae). *Comp Biochem Physiol A Mol Integr Physiol.* 2010;155(3):281–293.
32. Gaucher L, Vidal N, D'Anatro A, Naya DE. Digestive flexibility during fasting in the characid fish *Hyphessobrycon luetkenii*. *J Morphol.* 2012;273(1):49–56.
33. Masdeu M. *Hábitos alimenticios de Iheringichthys labrosus (Siluriformes, Pimelodidae) en el Río Uruguay (Río Negro, Uruguay)* [undergraduate thesis]. Montevideo: Universidad de la República; 2007. Spanish.
34. da Silva Abes S, Agostinho AA, Kiyoshi Okada E, Gomes LC. Diet of *Iheringichthys labrosus* (Pimelodidae, Siluriformes) in the Itaipu Reservoir, Paraná River, Brazil-Paraguay. *Braz Arch Biol Technol.* 2001;44(1):101–105.
35. dos Santos JE, Bazzoli N, Rizzo E, Santos GB. Reproduction of the catfish *Iheringichthys labrosus* (Lütken) (Pisces, Siluriformes) in Furnas reservoir, Minas Gerais, Brazil. *Rev Bras Zool.* 2004;21:193–200.
36. Holzbach AJ, Gubiani EA, Éder A, Baumgartner G. *Iheringichthys labrosus* (Siluriformes: Pimelodidae) in the Piquiri River, Paraná, Brazil: population structure and some aspects of its reproductive biology. *Neotropical Ichthyology.* 2009;7(1):55–64.
37. Schwalme K, Chouinard GA. Seasonal dynamics in feeding, organ weights, and reproductive maturation of Atlantic cod (*Gadus morhua*) in the southern Gulf of St Lawrence. *ICES J Mar Sci.* 1999;56:303–319.
38. Naya DE, Veloso C, Bozinovic F. Physiological flexibility in the Andean lizard *Liolaemus bellii*: seasonal changes in energy acquisition, storage and expenditure. *J Comp Physiol B.* 2008;178(8):1007–1015.

Open Access Animal Physiology

Publish your work in this journal

Open Access Animal Physiology is an international, peer-reviewed, open access journal publishing original research, reports, reviews and commentaries on all areas of animal physiology. The manuscript management system is completely online and includes a very

quick and fair peer-review system. Visit <http://www.dovepress.com/testimonials.php> to read real quotes from published authors.

Submit your manuscript here: <http://www.dovepress.com/open-access-animal-physiology-journal>

Dovepress