



# Ten Complete Mitochondrial Genomes of Gymnocharacini (Stethaprioninae, Characiformes). Insights Into Evolutionary Relationships and a Repetitive Element in the Control Region (*D-loop*)

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## INTRODUCTION

Stethaprioninae, a subfamily of characiform fish, comprises small fishes popularly known as tetras. Species in this subfamily were initially assigned to the genus *Astyanax*, which has a broad geographical distribution ranging from the southern United States to northern Argentina (Ornelas-García et al., 2008). Some species of *Astyanax*, however, are hard to accurately identify based on the lack of diagnostic morphological characteristics (Weitzman and Malabarba, 1998). As noted by Weitzman and Malabarba (1998), the genus *Astyanax* is probably not monophyletic, and this thinking is reflected in the recent revision by Terán et al. (2020), who reassigned some species of *Astyanax* to six different genera. The species complexes *A. fasciatus* and *A. scabripinnis*, for instance, were placed in the genus *Psalidodon*, and *Astyanax* species from the coastal river basins of Brazil (e.g., *A. giton*) were placed in the genus *Deuterodon*. The only taxa remaining in the genus *Astyanax* were the species complex *A. bimaculatus* and North American species. The revision by Terán was predicted by previous studies on chromosomes and mitochondrial sequences. Molecular analysis of 16 species of *Astyanax* based on the mitochondrial gene ATPase 8 and chromosomal characteristics obtained four clades (Pazza et al., 2018), and based on a DNA barcoding approach, Rossini et al. (2016) identified five lineages of *Astyanax* separated by high levels of divergence.

Despite the great diversity and taxonomic issues, at least one species of the genus *Astyanax*, *A. mexicanus* (formerly cited as *Astyanax fasciatus*), has been used as a model to understand the development of its eyes and the evolution of complex traits (Borowsky, 2008; Jeffery, 2008). *Astyanax mexicanus* is represented by surface and troglobite populations that show similar traits for cave dwelling. According to Ornelas-García et al. (2008), populations from Brazil are not the same species as those seen farther north in middle America and Mexico. Currently, the valid name for tetras from Brazil is *Psalidodon fasciatus*, a taxon probably containing cryptic species

based on their chromosomal variation ranging from  $2n = 45$  to 49 plus B chromosomes and heterochromatin polymorphism (Pazza et al., 2006; Ferreira-Neto et al., 2012; Kavalco et al., 2013). These chromosomal variants display low molecular divergence (Kavalco et al., 2016). Morphologically, the populations of the São Francisco river basin are different from the specimens from Alto Paraná and Paraíba do Sul rivers (Melo, 2001) and the original morphological description is so broad that it certainly covers other species outside the complex (Melo and Buckup, 2006). In this case, *P. fasciatus* should be restricted to the specimens from the São Francisco river basin (original basin of the type species), while the others may be either considered cryptic species of the complex or even other species (Melo and Buckup, 2006). On the other hand, despite a clear genetic structure among populations from the Alto Paraná and São Francisco river basins, morphometric traits seem to be homoplasy (Pazza et al., 2017).

The *A. bimaculatus* complex is currently represented by *Astyanax bimaculatus* species along with others, such as *A. altiparanae* (considered as a junior synonym of *A. lacustris* by some authors), *A. lacustris*, *A. assuncionensis*, and *A. abramis*. With a preference for calm waters, these species inhabit mainly in the Alto Paraná, Paraguay, Iguassu, and São Francisco river basins (Domingues et al., 2007). Contrary to what has been observed in *P. fasciatus* and *P. scabripinnis*, *A. bimaculatus* shows a constant diploidy number in different populations,  $2n = 50$  chromosomes, which is considered a symplesiomorphic character in Gymnocharacini (Kavalco et al., 2011; Martinez et al., 2012; Fernandes et al., 2014). The diversity within the group refers to differences in its karyotypic formula, fundamental number, and general symmetry of the karyotypes (Kavalco et al., 2011; Fernandes et al., 2014). These cytogenetic data, associated with the molecular ones, suggest a relatively recent divergence, as well as the monophyletic status of this branch (Pazza et al., 2018).

Alongside with the *P. fasciatus* and *A. bimaculatus*, the third species complex explored in this work, *P. scabripinnis*, was proposed by Moreira-Filho and Bertollo (1991) based on morphological and chromosomal characteristics of specimens collected in the Paraná and São Francisco river basins. In a review of the *P. scabripinnis* group, Bertaco and Lucena (2006) pointed out the existence of 15 species, including *P. paranae* and *P. rivularis*. The species of this complex are known for their wide karyotypic diversity, with diploid numbers ranging from  $2n = 46$  to 50 chromosomes (Moreira-Filho and Bertollo, 1991; Fernandes and Martins-Santos, 2005). In recent studies

using molecular phylogeography and geometric morphometry, Rocha et al. (2019) reinforced the validity of *P. rivularis* and *P. paranae* as sister species of the complex, inhabiting the São Francisco and Paraná river basins, respectively. However, among the populations from the Alto Paranaíba river, the existence of a new species of the complex was observed due to morphometric and *mtDNA* data (Alves R de et al., 2020). This new species, called *Psalidodon rioparanaibanus*, was collected only in a small tributary of the Paranaíba river, surrounded by populations of *P. paranae*. Moreover, within the *P. paranae* and *P. rivularis* groups, karyotypic diversity is also present (Maistro et al., 1998) indicating that, even though delimited by individual lineages, these groups still constitute compilations of cryptic species.

Hundreds of studies describing mitogenomes have been published in the last few years. Despite being the most sequenced genome nowadays (Smith, 2015), to this date, just the mitogenomes of *A. mexicanus* (Nakatani et al., 2011), *P. paranae* (Silva et al., 2016), *Deuterodon giton* (Barreto et al., 2017), *P. fasciatus*, and *A. altiparanae* (Calegari et al., 2019) are published. In an attempt to fill this gap, we focused on the three species complex of the group (*A. bimaculatus*, *P. scabripinnis* and *P. fasciatus*) to present here the complete sequences of the mitochondrial genome of 10 species/cytotypes of *Astyanax/Psalidodon*. Such data could be very useful in further phylogeny studies and to understand the diversity of the group.

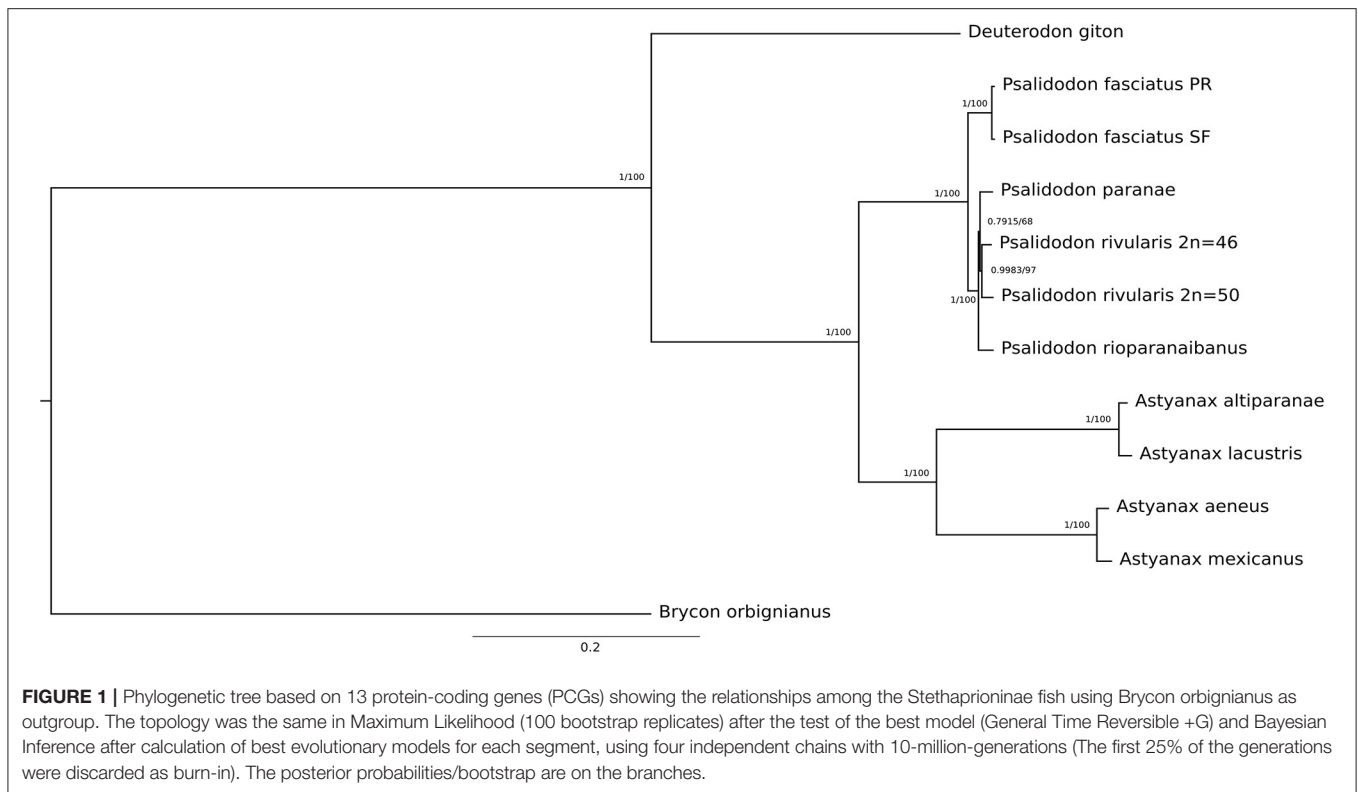
## MATERIALS AND METHODS

Specimens from the *Astyanax/Psalidodon* genus were analyzed. We collected the samples in different locations throughout the major Brazilian rivers and their vouchers are deposited in the ichthyological collection of the Laboratory of Ecological and Evolutionary Genetics at Federal University of Viçosa, campus Rio Paranaíba, Brazil (**Supplementary Table 1**). After sampling, we brought the living specimens to the laboratory, euthanized them in accordance with the ethical standards of CONCEA, the Brazilian Council for the Control of Animal Experimentation and CEUA/UFV—Ethics Committee on Animal Use/Federal University of Viçosa (760/2018). We performed the sampling with licenses provided by SISBIO/ICMBIO—Biodiversity Authorization and Information System (1938128) and SISGEN—National System for the Management of Genetic Heritage and Associated Traditional Knowledge (A9FE946).

We extracted the total genomic DNA from the liver and heart tissues of six specimens according to the instructions of Invitrogen's PureLink DNA extraction and purification kit. After quality checking using fluorometer Qubit (Thermo Fisher Scientific), the Whole Genome Sequencing was performed using Novaseq 6000 (Illumina, San Diego, CA) at Novogene, UK.

For broader comparisons, we also assembled the mitogenome of two *Astyanax/Psalidodon* species with raw reads available on ENA (European Nucleotide Archive): *P. fasciatus*, from the Alto Paraná river basin (SRR8476332) and *A. aeneus*, from Mexico (SRR1927238). Aiming to validate our methodology, we reassembled the mitogenomes of *P. paranae* (SRR5461470) and *A. mexicanus* (SRR2040423). In the Bayesian analysis, we also included the mitochondrial complete sequence of

**Abbreviations:** *ATP6/8*, ATP Synthase Membrane Subunit 6/8; BRIG, Blast Ring Image Generator; CEUA/UFV, Comissão de Ética no Uso de Animais/Universidade Federal de Viçosa (Ethics Committee on Animal Use/Federal University of Viçosa); CONCEA, Conselho Nacional de Controle de Experimentação Animal (Brazilian Council for the Control of Animal Experimentation); *COI*, Cytochrome c oxidase subunit I; *D-loop*, Displacement Loop; ENA, Europe Nucleotide Archive; ESS, Effective Sample Size; ESU, Evolutionarily Significant Unit; *mtDNA*, mitochondrial DNA; *ND6*, NADH-ubiquinone oxidoreductase chain 6; PCG, Protein-coding genes; SISBIO/ICMBIO, Sistema de Autorização e Informação em Biodiversidade/Instituto Chico Mendes de Conservação da Biodiversidade (Biodiversity Authorization and Information System/Chico Mendes Institute for Biodiversity Conservation); SISGEN, Sistema Nacional de Gestão do Patrimônio Genético e do Conhecimento Tradicional Associado (Brazilian National System for the Management of Genetic Heritage and Associated Traditional Knowledge).



*Deuterodon giton* (NC\_044970.1) and added *Brycon orbignianus* (KY825192.1) as outgroup.

We assembled the mitogenome from raw reads on Novoplasty v3.7 (Dierckxsens et al., 2017) in a parallel cluster computer (64 Gb RAM) using the mitogenome of *Psalidodon paranae* available on GenBank (SRR5461470) as seed. We chose this approach because it is fast and assemble the mitogenomes “*de novo*” from raw data using a single mitochondrial sequence as seed, without the bias of a reference. We annotated the sequences obtained on MitoAnnotator (Iwasaki et al., 2013) at MitoFish (<http://mitofish.aori.u-tokyo.ac.jp>).

We performed comparative genomics analysis by BLAST comparison of all the *Astyanax/Psalidodon* mitochondrial genomes against a reference (*Psalidodon paranae*) generated by Blast Ring Image Generator (BRIG) (Alikhan et al., 2011). To assess the repetitive region, we analyzed the mitochondrial sequences with Tandem Repeats Finder (Benson, 1999), following we isolate and duplicate the repeats of *D-loop*, and we aligned at ClustalW (Thompson et al., 1994) to find the repeat motif.

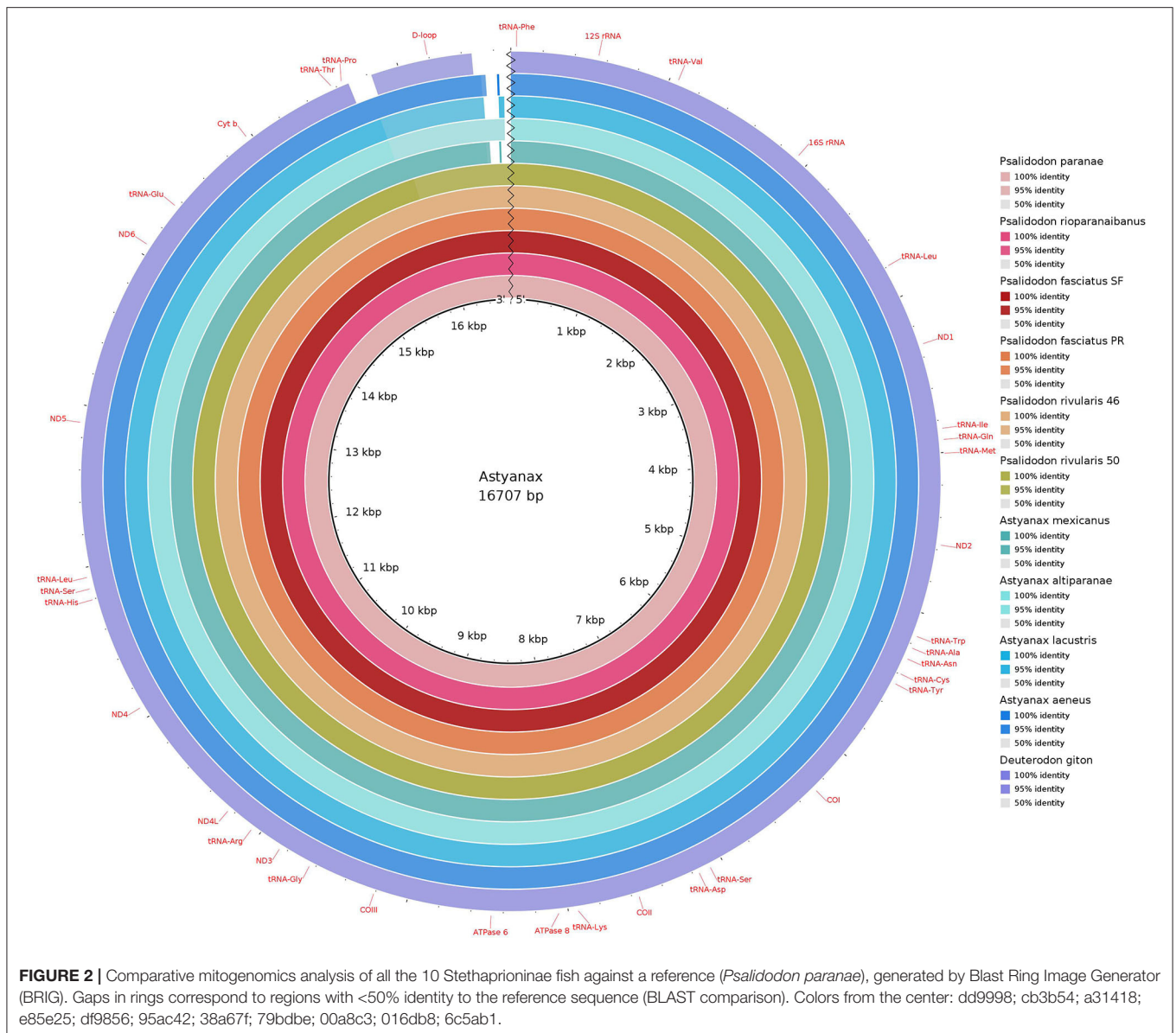
To validate the mitogenome as a tool for understanding the phylogenetic relationships among the samples, we aligned Fasta sequences with ClustalW and calculated the p-distance with MEGA X software (Kumar et al., 2018). We used the 13 protein-coding genes (PCGs), extracted by hand from fasta file produced by Mitoannotator, in Bayesian phylogenetic inference with MrBayes 3.2.7 (Roquist et al., 2012) after calculating the best evolutionary models for each segment with Partition Finder 2.1.1 (Lanfear et al., 2016). All PCGs used on phylogeny was tested by saturation (Xia et al., 2003) on DAMBE v7. (Xia,

2018). Bayesian analyses were performed using four independent chains with 10-million-generations and the effective sample size (ESS) and strand convergence were, posteriorly, verified in the software Tracer 1.7 (Rambaut et al., 2018). The first 25% of the generations were discarded as burn-in. For the Maximum Likelihood analysis, we used the concatenated 13 protein-coding genes (PCGs) after testing for the best model in the software MEGA X (Kumar et al., 2018).

## DATA DESCRIPTION

Our results have shown that all mitogenomes content and gene order were identical (**Figure 1**), with 13 PCGs, 22 *tRNA* genes, and 2 *rRNA* genes, as already described for Characiformes mitogenomes. The same is true for other *Astyanax/Psalidodon/Deuterodon* species, such as *A. mexicanus* (Nakatani et al., 2011), *P. paranae* (Silva et al., 2016), and *D. giton* (Barreto et al., 2017). All PCGs, except the *ND6* gene, are on the heavy chain. All but 8 *tRNAs* are on the heavy chain as well.

All the new sequences are deposited at GenBank (**Supplementary Table 2**). The length of mitochondrial sequences range from 16,626 bp in the *Psalidodon fasciatus* from the São Francisco river basin to 16,812 bp in *Psalidodon rivularis* 2n = 50. The average length of *D-loop* was 1,061 bp, ranging from 951 bp in *Psalidodon fasciatus* from São Francisco river basin to 1,136 bp in *Psalidodon rivularis* with 2n = 50 chromosomes. No differences between deposited mitogenome of *P. paranae* and our reassembling could be seen. On the other side, our pipeline could extend the *D-loop* region of *A. mexicanus*, a problematic



region in the deposited sequence (named “almost complete” on the GenBank entry). The difference in the size of *D-loop* was due to a repeat of 35 bp in all *D-loops*, except in *Deuterodon giton* (Supplementary Table 3). For the alignment, we got a repeated motif (TATGTATTAGTACATATATGCATAATTATACATA) slightly variable in some species.

Deepening the knowledge on the mitogenome control region, called *D-loop*, can play a fundamental role in understanding the evolutionary history in the *Astyanax* and *Psalidodon* genera. In this work, we observed that the size variation among different *Astyanax/Psalidodon* mitogenomes occurs mainly due to the extension of the *D-loop*. Neglecting this region in the reconstruction of mitogenomes can result in a valuable loss of information since, in addition to the variation in size, we found a repetitive sequence of 35 bp in nine of the 10 mitogenomes studied (Supplementary Table 3). On the other side, studies

based on *D-loop* sequence must be aware of this kind of feature that can bias the analysis.

*D. giton* was the only species that did not present the repetitive sequence in the *D-loop*. In addition to occupying a sister group position in the recuperated phylogeny (Figure 2) when describing the mitogenome of the species, Barreto et al. (2017) observed in their phylogeny of Characidae that the species *Grundulus bogotensis* was closer to the *Astyanax/Psalidodon* group than to *D. giton*. Therefore, we not only confirm *D. giton* as a species outside the genus *Astyanax* as suggested by the taxonomic review (Terán et al., 2020) but also that the repetitive sequence found in the *D-loop* may correspond to a synapomorphy absent in the *Deuterodon* group. However, it is necessary to reassemble the genome of *D. giton* under our methodology and conduct complementary studies on species of the *Deuterodon* group to clarify this issue.

The genetic distance among species (**Supplementary Table 4**) is reflected in both the Maximum Likelihood and the Bayesian tree (**Figure 2**), which shows strong construction with high bootstrap value and posterior probabilities, respectively. Once no gene sequence show saturation, we did the phylogeny with all 13 PCGs. The Partition Finder analysis results in five subsets (**Supplementary Table 5**) used in bayesian phylogeny.

The topology of the tree is congruent with those inferred by Rossini et al. (2016) and Pazza et al. (2018), except for the North American clade, which appears as a sister group of *A. altiparanae* and *A. lacustris* here. Besides, our study reinforces the taxonomic review by Terán et al. (2020), meanwhile disagree with Lucena and Soares (2016) that describe *A. altiparanae* as a new junior synonyms of *A. lacustris*.

The methodology used to reconstruct the mitochondrial genome proved to be satisfactory and enabled the assessment of the length of this type of genome, plus the composition and nature of the *D-loop*, solving possible gaps in previous methodologies (Silva et al., 2016; Barreto et al., 2017; Calegari et al., 2019). Besides, the study of the complete mitochondrial genome proves to be a tool with the potential to solve taxonomic problems and to help understand the evolutionary relationships in species complexes, such as *A. bimaculatus*, *P. fasciatus*, and *P. scabripinnis*.

## DATA AVAILABILITY STATEMENT

The datasets presented in this study can be found in online repositories. The names of the repository/repositories and

accession number(s) can be found below: <https://www.ncbi.nlm.nih.gov/genbank/>, BK013055 MT428072 MT428067 BK013062 BK013061 MT428071 MT428068 MT428069 MT428070.

## ETHICS STATEMENT

The animal study was reviewed and approved by Brazilian Council for the Control of Animal Experimentation and CEUA/UFV—Ethics Committee on Animal Use/Federal University of Viçosa (760/2018).

## AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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## SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fevo.2021.650783/full#supplementary-material>

## REFERENCES

- Alikhan, N. F., Petty, N. K., Zakour, N. L. B., and Beatson, S. A. (2011). BLAST Ring Image Generator (BRIG): simple prokaryote genome comparisons. *BMC Genomics* 12:402. doi: 10.1186/1471-2164-12-402
- Alves R de, M., Oliveira, I. H. R., Pasa, R., and Kavalco, K. F. (2020). A new Psalidodon species related to the *P. paranae* complex (Characiformes, Characidae) from Upper Paranaíba river basin, Brazil, supported by genetic and morphometric data. *Biodivers. J.* 11, 807–814. doi: 10.31396/Biodiv.Jour.2020.11.4.807.814
- Barreto, C. A. V., Granja, M. M. C., Vidigal, P. M. P., Carmo, A. O., and Dergam, J. A. (2017). Complete mitochondrial genome sequence of neotropical fish *Astyanax giton* Eigenmann 1908 (Ostariophysi; Characidae). *Mitochondrial DNA B* 2, 839–840. doi: 10.1080/23802359.2017.1403869
- Benson, G. (1999). Tandem repeats finder: a program to analyze DNA sequences. *Nucleic Acid Res.* 27, 573–580. doi: 10.1093/nar/27.2.573
- Bertaco, V. A., and Lucena, C. A. (2006). Two new species of *Astyanax* (Ostariophysi: Characiformes: Characidae) from eastern Brazil, with a synopsis of the *Astyanax scabripinnis* species complex. *Neotrop. Ichthyol.* 4, 53–60. doi: 10.1590/S1679-62252006000100004
- Borowsky, R. (2008). *Astyanax mexicanus*, the blind Mexican cave fish: a model for studies in development and morphology. *Cold Spring Harb. Protoc.* 11: pdb-emo107. doi: 10.1101/pdb.emo107
- Calegari, R. M., Rodrigues, P. H. M., Santos, R. Z. D., Foresti, F., Utsunomia, R., and Porto-Foresti, F. (2019). The complete mitochondrial genome sequences of five Otophysi species (Vertebrata, Teleostei). *Mitochondrial DNA B* 4, 4198–4199. doi: 10.1080/23802359.2019.1693294
- Dierckxens, N., Mardulyn, P., and Smits, G. (2017). NOVOPlasty: de novo assembly of organelle genomes from whole genome data. *Nucleic Acids Res.* 45:e18. doi: 10.1093/nar/gkw955
- Domingues, M. S., Vicari, M. R., Abilhoa, V., Wamser, J. P., Cestari, M. M., Bertollo, L. A. C., et al. (2007). Cytogenetic and comparative morphology of two allopatric populations of *Astyanax altiparanae* Garutti and Britski, 2000 (Teleostei: Characidae) from upper rio Paraná basin. *Neotrop. Ichthyol.* 5, 37–44. doi: 10.1590/S1679-62252007000100005
- Fernandes, C. A., da Rocha, R. H., Bailly, D., Guterres, Z. R., Soares, D., and Martins-Santos, I. C. (2014). Cytogenetic and Morphometric Analysis in the Species *Astyanax altiparanae* Garutti and Britski, 2000 (Teleostei, Characidae) from the Iguatemi River Basin, Brazil. *Folia Biol.* 62, 193–201. doi: 10.3409/fb62\_3.193
- Fernandes, C. A., and Martins-Santos, I. C. (2005). Sympatric occurrence of three cytotypes and four morphological types of B chromosomes of *Astyanax scabripinnis* (Pisces, Characiformes) in the River Ivaí Basin, state of Paraná, Brazil. *Genetica* 124, 301–306. doi: 10.1007/s10709-005-4751-7
- Ferreira-Neto, M., Artoni, R. F., Vicari, M. R., Moreira-Filho, O., Camacho, J. P. M., Bakkali, M., et al. (2012). Three sympatric karyomorphs in the fish *Astyanax fasciatus* (Teleostei, Characidae) do not seem to hybridize in natural populations. *Comp. Cytogenet.* 6, 29–40. doi: 10.3897/compcytogen.v6i1.2151
- Iwasaki, W., Fukunaga, T., Isagozawa, R., Yamada, K., Maeda, Y., Satoh, T. P., et al. (2013). MitoFish and MitoAnnotator: a mitochondrial genome database of fish with an accurate and automatic annotation pipeline. *Mol. Biol. Evol.* 30, 2531–2540. doi: 10.1093/molbev/mst141
- Jeffery, W. R. (2008). Emerging model systems in evo-devo: cavefish and microevolution of development. *Evol. Dev.* 10, 265–272. doi: 10.1111/j.1525-142X.2008.00235.x
- Kavalco, K. F., Pazza, R., Brandão, K. D. O., and de Almeida-Toledo, L. F. (2013). Biogeographic patterns in the chromosomal distribution of a satellite DNA in the banded tetra *Astyanax fasciatus* (Teleostei: Characiformes). *Org. Divers. Evol.* 13, 67–76. doi: 10.1007/s13127-012-0100-8

- Kavalco, K. F., Paza, R., Brandão, K. D. O., Garcia, C., and Almeida-Toledo, L. F. (2011). Comparative cytogenetics and molecular phylogeography in the group *Astyanax altiparanae*–*Astyanax* aff. *bimaculatus* (Teleostei, Characidae). *Cytogenet Genome Res.* 134, 108–119. doi: 10.1159/000325539
- Kavalco, K. F., Paza, R., Brandão, K. D. O., Garcia, C., Bertollo, L. A. C., and Almeida-Toledo, L. F. (2016). Chromosomal diversification higher than molecular variation in *Astyanax* aff. *fasciatus* (Teleostei, Characidae). *Zebrafish* 13, 345–353. doi: 10.1089/zeb.2016.1272
- Kumar, S., Stecher, G., Li, M., Knyaz, C., and Tamura, K. (2018). MEGA X: molecular evolutionary genetics analysis across computing platforms. *Mol. Biol. Evol.* 35, 1547–1549. doi: 10.1093/molbev/msy096
- Lanfear, R., Frandsen, P. B., Wright, A. P., Senfeld, T., and Calcott, B. (2016). PartitionFinder 2: new methods for selecting partitioned models of evolution for molecular and morphological phylogenetic analyses. *Mol. Biol. Evol.* 34, 772–773. doi: 10.1093/molbev/msw260
- Lucena, C. A. S., and Soares, H. G. (2016). Review of species of the *Astyanax bimaculatus* “caudal peduncle spot” subgroup sensu Garutti and Langeani (Characiformes, Characidae) from the rio La Plata and rio São Francisco drainages and coastal systems of southern Brazil and Uruguay. *Zootaxa* 4072, 101–125. doi: 10.11646/zootaxa.4072.1.5
- Maistro, E. L., Oliveira, C., and Foresti, F. (1998). Comparative cytogenetic and morphological analysis of *Astyanax scabripinnis paranae* (Pisces, Characidae, Tetragonopterinae). *Genet Mol Biol.* 21, 201–206. doi: 10.1590/S1415-47571998000200005
- Martinez, E. R., Alves, A. L., Silveira, S. M., Foresti, F., and Oliveira, C. (2012). Cytogenetic analysis in the incertae sedis species *Astyanax altiparanae* Garutti and Britzki, 2000 and *Hyphessobrycon eques* Steindachner, 1882 (Characiformes, Characidae) from the upper Paraná river basin. *Comp. Cytogenet.* 6, 41–51. doi: 10.3897/compcytogen.v6i1.1873
- Melo, F. A. G. (2001). Revisão taxonômica das espécies do gênero *Astyanax* Baird e Girard 1854 (Teleostei, Characiformes, Characidae) da região da Serra dos Órgãos. *Arq Mus Nac Rio de Janeiro.* 59, 1–46.
- Melo, F. A. G., and Buckup, P. A. (2006). *Astyanax henseli*, a new name for *Tetragonopterus aeneus* Hensel (1870), from Southern Brazil (Teleostei, Characiformes). *Neotrop. Ichthyol.* 4, 45–52. doi: 10.1590/S1679-62252006000100003
- Moreira-Filho, O., Bertollo, L. A. C. (1991). *Astyanax scabripinnis* (Pisces, Characidae) a species complex. *Rev. Brasil Genet.* 142, 331–357.
- Nakatani, M., Miya, M., Mabuchi, K., Saitoh, K., and Nishida, M. (2011). Evolutionary history of Otophysi (Teleostei), a major clade of the modern freshwater fishes: Pangaean origin and Mesozoic radiation. *BMC Evol. Biol.* 11:177. doi: 10.1186/1471-2148-11-177
- Ornelas-Garcia, C. P., Dominguez-Dominguez, O., and Doadrio, I. (2008). Evolutionary history of the fish genus *Astyanax* Baird and Girard (1854) (Actinopterygii, Characidae) in Mesoamerica reveals multiple morphological homoplasies. *BMC Evol. Biol.* 8:340. doi: 10.1186/1471-2148-8-340
- Paza, R., Cruvinel, L. M., and Kavalco, K. F. (2017). Parallel evolution evidenced by molecular data in the banded-tetra (*Astyanax fasciatus*). *Biochem. Syst.* 70, 141–146. doi: 10.1016/j.bse.2016.10.024
- Paza, R., Dergam, J. A., and Kavalco, K. F. (2018). Trends in Karyotype Evolution in *Astyanax* (Teleostei, Characiformes, Characidae): Insights From Molecular Data. *Front. Genet.* 9:131. doi: 10.3389/fgene.2018.00131
- Paza, R., Kavalco, K. F., and Bertollo, L. A. C. (2006). Chromosome polymorphism in *Astyanax fasciatus* (Teleostei, Characidae). 1. Karyotype analysis, Ag-NORs and mapping of the 18S and 5S ribosomal genes in sympatric karyotypes and their possible hybrid forms. *Cytogenet Genome Res.* 112, 313–319. doi: 10.1159/000089886
- Rambaut, A., Drummond, A. J., Xie, D., Baele, G., and Suchard, M. A. (2018). Posterior summarization in Bayesian phylogenetics using tracer 1.7. *Syst Biol.* 67, 901–904. doi: 10.1093/sysbio/syy032
- Rocha, R. R., Alves R de, M., Pasa, R., and Kavalco, K. F. (2019). Morphological and genetic structure of two equivalent *Astyanax* species (Characiformes: Characidae) in the region of Paranaíba Arc. *Sci World J.* 4:650795. doi: 10.1155/2019/6507954
- Roquist, F., Teslenko, M., van der Mark, P., Ayres, D. L., Darling, A., Höhna, S., et al. (2012). MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. *Syst Biol.* 61, 539–542. doi: 10.1093/sysbio/sys029
- Rossini, B. C., Oliveira, C. A., Melo, F. A., Bertaco, V. A., Astarloa, J. M. D., Rosso, J. J., et al. (2016). Highlighting *Astyanax* Species Diversity through DNA Barcoding. *PLoS ONE.* 11:e0167203. doi: 10.1371/journal.pone.0167203
- Silva, D. M. D. A., Utsunomia, R., Ruiz-Ruano, F. J., Oliveira, C., and Foresti, F. (2016). The complete mitochondrial genome of *Astyanax paranae* (Teleostei: characiformes). *Mitochondrial DNA B* 1, 586–587. doi: 10.1080/23802359.2016.1222251
- Smith, D. R. (2015). The past, present and future of mitochondrial genomics: have we sequenced enough mtDNAs? *Brief. Funct. Genom.* 15, 47–54. doi: 10.1093/bfpg/elv027
- Terán, G. E., Benitez, M. F., and Mirande, J. M. (2020). Opening the Trojan horse: phylogeny of *Astyanax*, two new genera and resurrection of *Psalidodon* (Teleostei: Characidae). *Zool. J. Linnean Soc.* 190, 1217–1234. doi: 10.1093/zoolinnean/zlaa019
- Thompson, J. D., Higgins, D. G., and Gibson, T. J. (1994). CLUSTALW: improving the sensitivity of progressive multiple sequence through weighing, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res.* 22, 4673–4680. doi: 10.1093/nar/22.22.4673
- Weitzman, S. H., and Malabarba, L. (1998). “Perspectives about the phylogeny and classification of the Characidae (Teleostei: Characiformes),” in *Phylogeny and classification of Neotropical fishes* edn, eds L.R. Malabarba, R. E. Reis, R. P. Vari, Z. M. S. Lucena, C. A. S. Lucena, (Edipucrs, Porto Alegre), 161–170.
- Xia, X. (2018). DAMBE7: New and improved tools for data analysis in molecular biology and evolution. *Mol. Biol. Evol.* 35, 1550–1552. doi: 10.1093/molbev/msy073
- Xia, X., Xie, Z., Salemi, M., Chen, L., and Wang, Y. (2003). An index of substitution saturation and its application. *Mol. Phylogen. Evol.* 26, 1–7. doi: 10.1016/S1055-7903(02)00326-3

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