Population genetics of three threatened catfish species in heterogeneous environments of the Cauca River, Colombia

Correspondence:
Edna Judith Márquez
ejmarque@unal.edu.co

Natalia Restrepo-Escobar[1], Anny Johanna Yepes-Acevedo[1] and Edna Judith Márquez[1]

Neotropical catfishes *Ageneiosus pardalis*, *Pimelodus grosskopfii* and *Sorubim cuspicaudus* are migratory fishes of commercial importance that exhibit decreasing populations due to overfishing and other anthropic interventions. This study used species–specific microsatellite loci to test the hypothesis that threatened fish populations show genetic vulnerability signs and are genetically structured in the middle and lower sections of the Cauca River. The studied species exhibit genetic diversity levels higher than the average values reported for Neotropical Siluriformes; however, they seem to have suffered recent bottlenecks and they present significant endogamy levels that are higher for the critically endangered catfish *P. grosskopfii*. Furthermore, both *Ageneiosus pardalis* and *S. cuspicaudus* are each formed by one genetic group, while *Pimelodus grosskopfii* comprises two coexisting genetic groups. The information obtained in this study is useful for the decision making in management plans that are appropriate for the sustainability of these three species populations within the proposal for the expansion of the hydroelectric development and other anthropic activities.

Keywords: Freshwater fish, Genetic diversity, Genetic structure, Microsatellites, Siluriformes.

1 Facultad de Ciencias, Escuela de Biociencias, Laboratorio de Biología Molecular y Celular, Universidad Nacional de Colombia, Sede Medellín, Carrera 65 No 59A, 110 Bloque 19A Laboratorio 310, Medellín 050034, Colombia. (NRE) nrestree@unal.edu.co; (AJYA) ajyepesa@unal.edu.co; (EJM) ejmarque@unal.edu.co, ejmarque@gmail.com (corresponding author).

Los bagres Neotropicales *Ageneiosus pardalis*, *Pimelodus grosskopfii* y *Sorubim cuspicaudus*, son peces migratorios de importancia comercial cuyas poblaciones han disminuido debido a la sobrepesca y otras intervenciones antrópicas. En este trabajo, se utilizaron loci microsatélites especie-específicos para contrastar la hipótesis de que las poblaciones de peces amenazadas muestran señales de vulnerabilidad genética y están genéticamente estructuradas en los sectores medio y bajo del río Cauca. Las especies estudiadas exhiben niveles de diversidad genética superiores a los promedios reportados para Siluriformes Neotropicales; sin embargo, parecen haber sufrido cuellos de botella recientes y presentan niveles significativos de endogamia que son más altos para el bagre en peligro crítico, *P. grosskopfii*. Además, *Ageneiosus pardalis* y *S. cuspicaudus* están conformados cada uno por un solo grupo genético, mientras que *Pimelodus grosskopfii* comprende dos grupos genéticos que coexisten. La información obtenida en este estudio es útil para la toma de decisiones en planes de manejo que sean adecuados para la sostenibilidad de las poblaciones de estas tres especies de bagre dentro de las propuestas para la expansión de desarrollo hidroeléctrico y otras actividades antrópicas.

Palabras clave: Diversidad genética, Estructura genética, Microsatélites, Pez dulceacuícola, Siluriformes.

INTRODUCTION

The Siluriformes order comprises 38 families, 15 of which are exclusively in Central and South America (Ferraris, 2007; Fricke *et al.*, 2020). Six of these families encompass the 86% of the 2347 catfish valid species of Neotropical distribution (Ferraris, 2007; Fricke *et al.*, 2020): Auchenipteridae (125), Callichthyidae (221), Heptapteridae (226), Loricariidae (1000), Pimelodidae (114), and Trichomycteridae (327). These species, known as catfishes, have commercial importance in different regions as nourish source, ornamental fishes or even sport fishing (Usma-Oviedo *et al.*, 2009; Nelson *et al.*, 2016).

Population genetic studies in Neotropical catfishes are centered in members of commercial interest of Pimelodidae (17) family, and in lower proportion, of Loricariidae (6), Trichomycteridae (1), and Heptateridae (2) families. To the date, evidences for gene flow have been found in species that perform medium and large distance migrations during their lifecycle, such as *Brachyplatystoma flavicans* (Coronel *et al.*, 2004), *Brachyplatystoma rousseauxii* (Batista, Alves-Gomes, 2006), *Pimelodus maculatus* (Almeida *et al.*, 2001, 2003; Ramella *et al.*, 2006; Ribolli *et al.*, 2012), *Pseudoplatystoma fasciatum* (Coronel *et al.*, 2004), *Pseudoplatystoma magdalenatium* (Gallo, Díaz-Sarmiento, 2003), *Pseudoplatystoma corruscans* (Dantas *et al.*, 2013; Vaini *et al.*, 2016; Prado *et al.*, 2018), and *Pseudoplatystoma reticulatum* (Vaini *et al.*, 2016; Prado *et al.*, 2018).

Nevertheless, genetic structure of populations of some species has been found and it seems to be explained by various types of behaviors: (1) sedentarism and short-distance migration range in *Steindachneridion parahybae* (Fonseca *et al.*, 2017), *Trichogenes longipinnis* (Zamudio *et al.*, 2009), *Iheringichthys labrosus*, and *Pimelodus* cf. *absconditus*

(Almeida *et al.*, 2001); (2) fidelity to breeding or spawning sites (*homing*) in *P. corruscans* (Pereira *et al.*, 2009), *P. reticulatum* (Abreu *et al.*, 2009), and *Rhamdia quelen* (Ríos *et al.*, 2019), and (3) presence of natural or artificial geographical barriers in *P. maculatus* in Paranapanema River (Almeida *et al.*, 2003), *P. corruscans* in Paraná River (Sekine *et al.*, 2002), and between Paraná and São Francisco rivers (Carvalho *et al.*, 2012), and *P. corruscans* and *P. reticulatum* in Paraná and Paraguay rivers (Prado *et al.*, 2018).

Contrary to the majority of population genetics studies in Amazonas and Paraná basins, very few have been performed in Colombian basins, most of which are found in grey literature (see Mancera-Rodríguez *et al.*, 2013; Márquez *et al.*, 2020). For the remaining freshwater catfishes in northwestern South America, there is still unawareness of genetic diversity, demography and population structure which limits the implementation of effective rules for their management and protection.

The latter is important since populations of these species have been affected by overfishing and other indirect effects of anthropogenic activities like mining, dam construction, gravel sand extraction, basins contamination and continuous water extraction for agricultural and livestock purposes, which have led to the decay and decrease of the populations of said species in all Colombian basins (Galvis, Mojica, 2007; Usma-Oviedo *et al.*, 2009; Barletta *et al.*, 2010; Jiménez-Segura *et al.*, 2016).

To contrast the hypothesis that threatened fish populations show genetic vulnerability signs in Cauca River sections exposed to anthropic activities, three catfish species were chosen, with differences in their standard length, migration range and reproductive strategies. *Ageneiosus pardalis* Lütken, 1874, is a medium size species (standard length between 12.83–42.98 cm; Ribeiro *et al.*, 2017), with short-distance migration range (< 100 km), marked sexual dimorphism (Dahl, 1971; Mojica *et al.*, 2012a), elaborated courtship and copulation (Galvis *et al.*, 1997) and distribution from south Panamá to north Venezuela; *Pimelodus grosskopfii* Steindachner, 1879, is a small size species (standard length between 10.17–32.12 cm; Villa-Navarro *et al.*, 2017), with medium migration range (100–500 km), external reproduction, endemic of the Magdalena-Cauca Colombian basin (Mojica *et al.*, 2006; Ortega-Lara *et al.*, 2006), and *Sorubim cuspicaudus* Littmann, Burr & Nass, 2000, a medium size species (can reach lengths greater than 80 cm; Littmann *et al.*, 2000), with medium-distance migration range (100-500 km), external reproduction and distribution in the rivers Catatumbo (Ortega-Lara *et al.*, 2012), Magdalena (Mojica *et al.*, 2006; Villa-Navarro *et al.*, 2006), Sinú and Cauca (Littmann *et al.*, 2000) in northwestern South America.

The appointed species are a good model for population genetics analysis since they are exposed to anthropic interventions such as loss of habitats, deforestation, river-course fragmentation, introduction of exotic fish species, contamination and overfishing (Barletta *et al.*, 2010; FAO, 2015; Jiménez-Segura *et al.*, 2016; AUNAP, 2020). The decreasing population density of the three species led to their classification as vulnerable in the Colombian red list of freshwater fishes (Mojica *et al.*, 2012b). Additionally, due to this decline in populations, *P. grosskopfii* was included as a critically endangered threatened species in the red list of threatened species of the International Union for Conservation of Nature – IUCN (Villa-Navarro *et al.*, 2016). Thus, the *a priori* expectation was that the species showed high values of inbreeding and recent bottlenecks, as they are exposed to different anthropic interventions that might affect the density of their populations.

Moreover, the studied area comprises the middle and lower sections of the Cauca River, which includes near 500 km of the main channel of the river, 110 km of torrents and 40.000 ha of swamps. This area also includes the Cauca River canyon, the steepest margin of the Antioqueño Plateau in the northern portion of the Central Cordillera (Restrepo-Moreno *et al.*, 2009), which has been considered a geographic barrier for many fish species (Dahl, 1971). Additionally, this area presents differences in habitats, water speeds, temperatures and it constitutes the influence zone of the Ituango hydroelectric project. In this context, this study also tested the hypothesis that populations of these species, collected before the beginning of the construction of the hydroelectric, were genetically structured. The *a priori* expectation was that the environmental heterogeneity of the zone caused genetic structuration. The answer to these queries generates information of interest to propose rules of management and conservation, suitable for the preservation of natural populations of these fish species.

MATERIAL AND METHODS

A total of 492 preserved in alcohol muscle tissues of *A. pardalis* (193), *P. grosskopfii* (170), and *S. cuspicaudus* (129) were analyzed. Said tissues were provided by Integral S.A through the scientific cooperation agreement CT–2013–002443, framed in the environmental license of Ministerio de Ambiente, Vivienda y Desarrollo Territorial # 0155 of January 30th, 2009. Samples were collected between 2011–2014 in the middle (sections S1–S4) and lower (sections S5–S8) sections of the Cauca River, located upstream (S1) and downstream (S2–S8) of the construction zone of Ituango hydroelectric project (Fig. 1; Tab. 1). The middle section of the Cauca River encompasses the Cauca River canyon, characterized by a steep topography with the presence of rapid zones (S1–S3) followed by a vast alluvial plain surrounded by mountains, with swamp ecosystems connected with the main river channel (S4). The section S5 includes the mouth of the Nechí River, a hydrologically monomodal river prone to flooding and scouring, and the remaining lower section (S6–S8) shows soft topography with flat and wavy surfaces wherein several swamps connected to rivers through canals are formed, conforming complex swampy systems (Mejía-Rivera *et al.*, 2007; Betancur-Vargas *et al.*, 2009).

DNA extraction was performed with the commercial kit PureLink® Genomic DNA (ThermoFisher Scientific), following the instructions of the manufacturer. PCR conditions previously described by Landínez-García, Marquez (2016, 2018) were employed to amplify between 13 and 20 microsatellite loci for *A. pardalis* (Apar03, Apar04, Apar05, Apar11, Apar12, Apar14, Apar18, Apar19, Apar20, Apar21, Apar22, Apar23, Apar25, Apar27, Apar28, Apar30, Apar32, Apar34, Apar35, Apar36); *P. grosskopfii* (Pgrk01, Pgrk02, Pgrk03, Pgrk08, Pgrk10, Pgrk14, Pgrk15, Pgrk19, Pgrk24, Pgrk27, Pgrk28, Pgrk31, Pgrk40), and *S. cuspicaudus* (Scus03, Scus10, Scus12, Scus15, Scus16, Scus17, Scus18, Scus20, Scus21, Scus22, Scus23, Scus24, Scus25, Scus28, Scus32, Scus35, Scus39, Scus40, Scus41; Restrepo-Escobar, Marquez, 2020). PCR products were separated in an automatic sequencer ABI 3130 (Applied Biosystems, USA) using GeneScan-500 LIZ dye size standard (Applied Biosystems, USA) as internal marker and the allelic fragments were denoted according to their molecular size and scored using GeneMapper (Applied Biosystems, USA). Before the statistical analysis, the Micro-
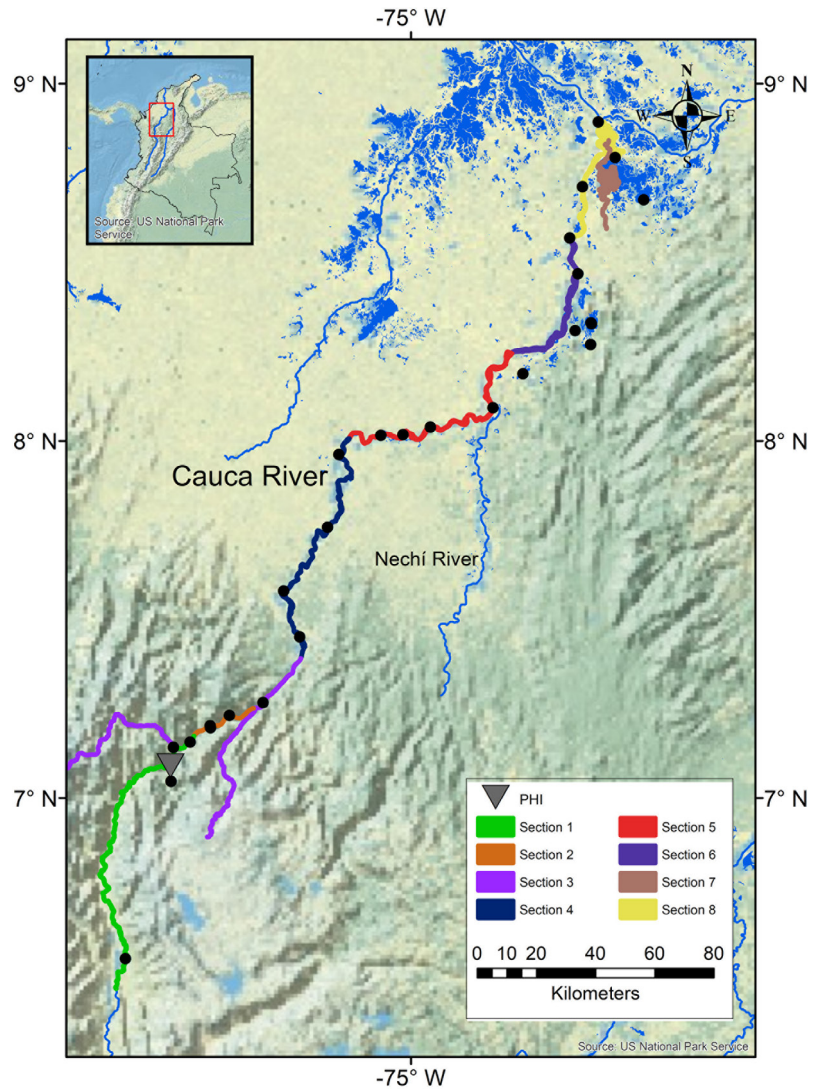
FIGURE 1 | Location of sampling sites of *Pimelodus grosskopfii*, *Sorubim cuspicaudus*, and *Ageneiosus pardalis* in the middle and lower sections of the Cauca River.

TABLE 1 | Number of individuals of *Pimelodus grosskopfii*, *Sorubim cuspicaudus*, and *Ageneiosus pardalis* analyzed in the middle and lower basin of the Cauca River. S1–S4: Middle section of the Cauca River, S1 and S2–S4 are located upstream and downstream of the construction zone of the dam, respectively. S5–S8: Lower section of the Cauca River.

Sections	*P. grosskopfii*	*S. cuspicaudus*	*A. pardalis*
S1	44	–	–
S2/S3	25	19	–
S4	18	22	–
S5	26	43	45
S6	35	25	84
S7/S8	22	20	64
Total	**170**	**129**	**193**

Checker v2.2.1 software (Van Oosterhout *et al.*, 2004) was used to detect possible genotyping errors.

For samples of each studied section and genetic group, it was determined the genetic diversity calculating the expected and observed heterozygosities and the average number of alleles per locus with GenAlex v6.502 software (Peakall, Smouse, 2012). Additionally, the Arlequin v3.5.2.2 software (Excoffier, Lischer, 2010) was employed to test departures of the Hardy-Weinberg equilibrium, linkage equilibrium and calculate the inbreeding coefficient (F_{IS}) by population. In multiple comparisons, the statistical significance was adjusted by the Bonferroni sequential correction (Rice, 1989). Finally, two tests were used to determine whether the populations of these species have suffered a recent genetic bottleneck, the one-tailed test of Wilcoxon (Luikart, Cornuet, 1998), and M ratio of Garza, Williamson (2001) calculated respectively in Bottleneck v1.2.02 software (Piry *et al.*, 1999) and the Arlequin v3.5.2.2. program (Excoffier, Lischer, 2010).

To determine the number of possible genetic groups for *A. pardalis*, *P. grosskopfii*, and *S. cuspicaudus* in the middle and lower basins of the Cauca River, it was used different complementary approaches. The Bayesian clustering method with STRUCTURE v2.3.4 software (Pritchard *et al.*, 2000) used the following conditions: 100000 MCM, 10000 burn-in, admixture model, correlated allele frequency, and LOCPRIOR option. Simulations were repeated 20 times per each number of evaluated probable populations: 1–6 for *A. pardalis*, 1–8 for S. *cuspicaudus*, and 1–9 for *P. grosskopfii*. The most probable number of population was calculated using the ΔK method (Evanno *et al.*, 2005) and the estimators MedMedK, MedMeaK, MaxMedK, and MaxMeaK (Puechmaille, 2016) with the web-based software STRUCTURESELECTOR (Li, Liu, 2018). The STRUCTURE results were summarized using the integrated software CLUMPAK (Kopelman *et al.*, 2015) to generate a graphical representation of the results. Wherein STRUCTURE detected different genetic groups, the allocation of each individual to each group was determined by the coancestry coefficient from the Q-matrix generated by CLUMPAK (Kopelman *et al.*, 2015) from the 20 simulations produced by STRUCTURE. Moreover, the genetic differentiation of each of the species among sectors of the Cauca River was calculated with the Analysis of Molecular Variance AMOVA and the standardized estimators F'_{ST} (Meirmans, 2006) and Jost's Dest (Meirmans, Hedrick, 2011) using the GenAlex v6.502 software (Peakall, Smouse, 2012), and a Discriminant Analysis of Principal Components (DAPC) was performed using the package *adegenet* (Jombart, 2008) for R v3.4.0 program (R Development Core Team, 2017).

RESULTS

The sampling performed by Integral S.A. between 2011–2014 revealed that *A. pardalis*, *P. grosskopfii*, and *S. cuspicaudus* were differentially distributed in the middle and lower sections of the Cauca River. Namely, and despite having the same migration range, *P. grosskopfii* was found in all sampled sites, including upstream of the Cauca River canyon, whereas *S. cuspicaudus* was found in the downstream area of Cauca River canyon and the construction zone of the dam (S3–S8). In contrast, short-distance migration range *A. pardalis* was only captured in the lower basin of the river (S5–S8; Tab. 1).

Genetic diversity and population demographics. *Pimelodus grosskopfii* showed similar or nearly similar genetic diversity values in all evaluated sections (Na=10.308–13.308; He=0.849–0.866; Ho=0.607–0.682) and in the genetic groups proposed by the genetic structure analysis described below (Na=12.846–16.615; He=0.858–0.866; Ho=0.614–0.664; Tab. 2), further to statistical significance in heterozygosity deficit and inbreeding coefficients (F_{IS}=0.108–0.218; P<0.000; Tab. 2).

Likewise, *S. cuspicaudus* exhibited similar values of genetic diversity within the analyzed sections (Na=8.105–9.368; He=0.771–0.785; Ho=0.745–0.795; Tab. 2); however, in this case the genotypic frequencies behaved according to Hardy–Weinberg equilibrium except from S5 wherein the heterozygosity deficit was significant. Moreover, the inbreeding coefficient was significant in S5 (F_{IS}=0.045; P=0.026) and S8 (F_{IS}=0.058; P=0.032; Tab. 2).

In accordance with the other two species, *A. pardalis* showed similar genetic diversity values (Na=10.850–12.550; He=0.832–0.839; Ho=0.791–0.822; Tab. 2); statistical significance in heterozygosity deficits and coefficients of inbreeding in all the evaluated samples (F_{IS}=0.026–0.086; P<0.007) although in lesser magnitude than in *P. grosskopfii* (Tab. 2).

TABLE 2 | Genetic diversity, M-ratio estimator and bottleneck for populations and genetic groups of *Pimelodus grosskopfii*, *Sorubim cuspicaudus*, and *Ageneiosus pardalis* in the middle and lower sections of the Cauca River. N: Number of analyzed individuals, Na: Average number of alleles/locus, Ho: Observed heterozygosity, He: Expected heterozygosity, P: statistical significance of Hardy-Weinberg equilibrium; F_{IS}: inbreeding coefficient, M-ratio: M estimator of Garza, Williamson (2001), IAM: infinite alleles model, TPM: two phase model, SMM: stepwise model. Values in bold denote statistical significance.

	N	Na	Ho	He	P	F_{IS}	M-ratio	IAM	TPM	SMM
Pimelodus grosskopfii										
S1	44	13.308±1.184	0.682±0.020	0.863±0.015	**0.000**	**0.147**	0.217±0.049	**0.000**	**0.003**	0.342
S2/S3	25	11.615±0.675	0.679±0.062	0.860±0.012	**0.000**	**0.157**	0.224±0.057	**0.000**	**0.000**	0.317
S4	18	10.308±0.728	0.660±0.068	0.852±0.013	**0.000**	**0.178**	0.197±0.054	**0.000**	**0.000**	0.064
S5	26	12.462±0.991	0.618±0.061	0.866±0.011	**0.000**	**0.177**	0.201±0.058	**0.000**	**0.001**	0.527
S6	35	12.385±0.984	0.607±0.051	0.855±0.014	**0.000**	**0.186**	0.215±0.045	**0.000**	**0.000**	0.847
S7/S8	22	11.538±1.029	0.666±0.055	0.849±0.017	**0.000**	**0.109**	0.227±0.048	**0.000**	**0.040**	0.526
Genetic group 1	64	12.846±1.061	0.614±0.050	0.858±0.017	**0.000**	**0.218**	0.225±0.061	**0.000**	**0.000**	0.987
Genetic group 2	106	16.615±1.583	0.664±0.050	0.866±0.011	**0.000**	**0.108**	0.212±0.046	**0.000**	**0.000**	0.936
Sorubim cuspicaudus										
S3	19	8.105±0.501	0.767±0.029	0.774±0.020	0.814	0.018	0.229±0.053	**0.000**	**0.040**	0.928
S4	22	8.316±0.536	0.788±0.023	0.771±0.024	0.352	-0.065	0.248±0.068	**0.000**	0.176	0.922
S5	43	9.368±0.613	0.745±0.028	0.783±0.020	**0.000**	**0.045**	0.258±0.049	**0.000**	**0.020**	0.973
S6	25	8.684±0.484	0.795±0.024	0.783±0.019	0.412	-0.045	0.254±0.063	**0.000**	0.129	0.916
S7/S8	20	8.263±0.561	0.745±0.028	0.785±0.017	0.124	**0.058**	0.257±0.055	**0.001**	0.078	0.675
Ageneiosus pardalis										
S5	45	10.850±0.559	0.772±0.039	0.832±0.017	**0.000**	**0.086**	0.242±0.046	**0.000**	**0.007**	0.249
S6	84	12.550±0.755	0.791±0.027	0.839±0.015	**0.000**	**0.060**	0.240±0.042	**0.000**	**0.002**	0.607
S7/S8	64	11.950±0.630	0.822±0.021	0.839±0.014	**0.000**	**0.026**	0.245±0.043	**0.000**	**0.000**	0.892

The analysis for detecting bottleneck showed significant values for all the species in the evaluated sections under the IAM, while no significance was found under the SMM and the TPM showed significance in all or almost all of the evaluated sections in *A. pardalis*, *P. grosskopfii* (except S8), and *S. cuspicaudus* (except S4, S6, and S7/8). Additionally, the three species showed values of the M ratio of Garza, Williamson, (2001) lower than 0.680 (*P. grosskopfii*: 0.197–0.227; *S. cuspicaudus*: 0.229–0.258; *A. pardalis*: 0.242–0.245; Tab. 2), which has been proposed as limit value for inferring that the population has suffered a recent decline in population (Garza, Williamson, 2001).

Genetic structure analysis. In the distribution range of the evaluated samples in middle and lower sections of the Cauca River, the Bayesian analysis of STRUCTURE showed the homogeneous distribution of two coexisting genetic groups in *P. grosskopfii* (K=2; MedMedK=2; MedMeanK=2; MaxMedK=2; MaxMeanK=2; Fig. 2A), one genetic group in *S. cuspicaudus*, wherein each individual exhibits a similar genetic admixture of two biological groups (K=2; MedMedK=1; MedMeanK=1; MaxMedK=2; MaxMeanK=2; Fig. 2B), and one main genetic group with some genetic admixture in *A. pardalis* (ΔK=2; MedMedK=1; MedMeanK=1; MaxMedK=1; MaxMeanK=1; Fig. 2C).

The homogeneous distribution of the genetic groups of *P. grosskopfii* (F_{ST}: 0.004, P: 0.075), *S. cuspicaudus* (F_{ST}: 0.003, P: 0.084) and *A. pardalis* (F_{ST}: 0.001, P: 0.066) in the middle and lower sections of the Cauca River was corroborated by AMOVA, the pairwise comparisons with standardized estimators F'_{ST} and Dest (Tab. 3), and DAPC (Figs. 2D-F).
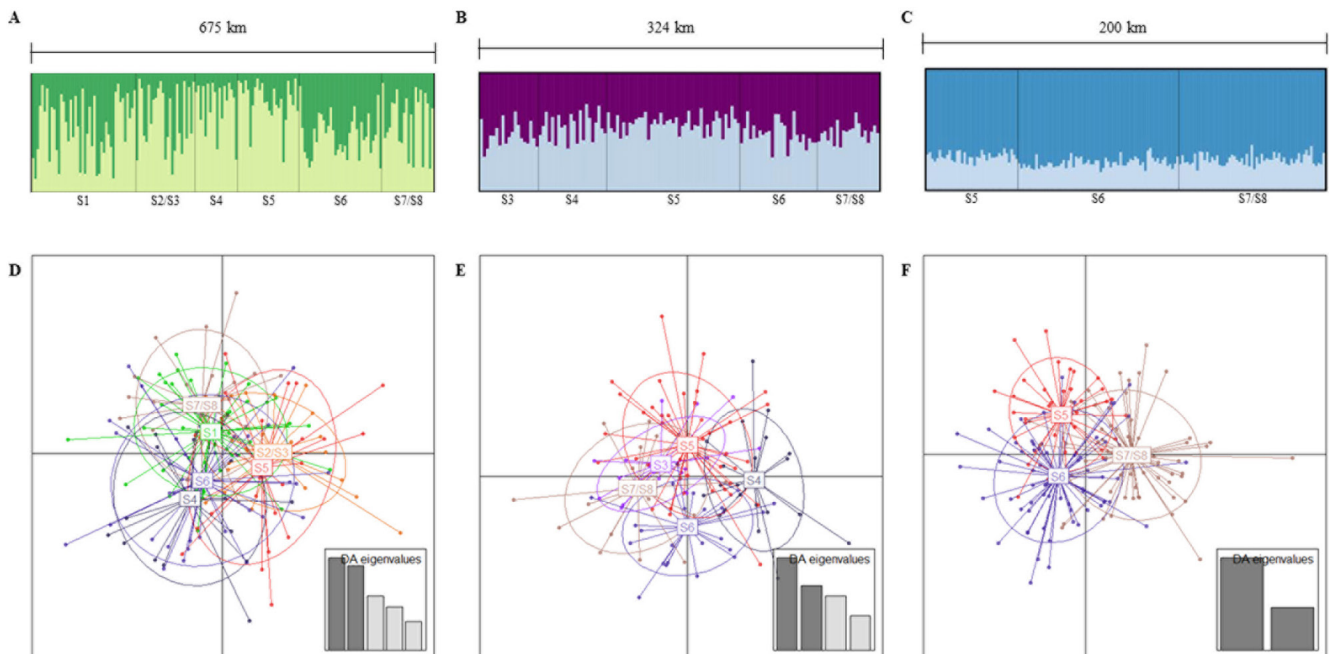
FIGURE 2 | Population structure suggested by STRUCTURE (**A–C**) and the Discriminant Analysis of the Principal Components (**D–F**) for *Pimelodus grosskopfii* (**A, D**), *Sorubim cuspicaudus* (**B, E**), and *Ageneiosus pardalis* (**C, F**).

TABLE 3 | Statistical values F'$_{ST}$ and Dest by pairs of the *Pimelodus grosskopfii*, *Sorubim cuspicaudus*, and *Ageneiosus pardalis* populations in middle and lower sections of the Cauca River.

	F'$_{ST}$ (P)	D$_{ST}$ (P)
Pimelodus grosskopfii		
S1–S2/S3	0.010 (0.502)	-0.004 (0.543)
S1–S4	0.013 (0.432)	-0.002 (0.504)
S1–S5	0.012 (0.095)	0.027 (0.098)
S1–S6	0.009 (0.276)	0.008 (0.284)
S1–S7/S8	0.013 (0.123)	0.027 (0.123)
S2/S3–S4	0.018 (0.332)	0.010 (0.332)
S2/S3–S5	0.012 (0.831)	-0.028 (0.829)
S2/S3–S6	0.013 (0.215)	0.019 (0.219)
S2/S3–S7/S8	0.018 (0.041)	0.055 (0.014)
S4–S5	0.015 (0.649)	-0.018 (0.666)
S4–S6	0.014 (0.635)	-0.013 (0.656)
S4–S7/S8	0.019 (0.169)	0.033 (0.177)
S5–S6	0.012 (0.361)	0.007 (0.352)
S5–S7/S8	0.017 (0.037)	0.052 (0.037)
S6–S7/S8	0.016 (0.037)	0.050 (0.036)
Sorubim cuspicaudus		
S3–S4	0.015 (0.180)	0.014 (0.186)
S3–S5	0.010 (0.531)	-0.003 (0.555)
S3–S6	0.014 (0.208)	0.010 (0.217)
S3–S7/S8	0.012 (0.837)	-0.017 (0.838)
S4–S5	0.009 (0.562)	-0.002 (0.546)
S4–S6	0.012 (0.298)	0.007 (0.298)
S4–S7/S8	0.015 (0.146)	0.017 (0.150)
S5–S6	0.010 (0.159)	0.010 (0.159)
S5–S7/S8	0.009 (0.665)	-0.006 (0.679)
S6–S7/S8	0.011 (0.774)	-0.011 (0.785)
Ageneiosus pardalis		
S5–S6	0.005 (0.615)	-0.003 (0.624)
S5–S7/S8	0.007 (0.120)	0.008 (0.127)
S6–S7/S8	0.005 (0.029)	0.011 (0.029)

DISCUSSION

This study employed species–specific microsatellite *loci* to acknowledge the level of genetic diversity, demographic status and genetic structure degree of three species of Colombian endemic Neotropical catfishes that coexist in the middle and lower sections of the Cauca River basin. The *a priori* expectation was that the species showed high values of inbreeding and recent bottlenecks as they are subject of different anthropic interventions that might affect the density of their populations. Furthermore, it was expected a population genetic structure concordant with some heterogeneous features of the basin.

Genetic diversity and population demography. The three studied species showed higher levels of genetic diversity than the average values reported for Neotropical catfishes (Na=7.470, He=0.609; revision of Hilsdorf, Hallerman, 2017) although *S. cuspicaudus* showed lower diversity (Na=8.547, He=0.779) than *A. pardalis* (Na=11.783, He=0.835) and *P. grosskopfii* (Na=11.143, He=0.847).

The average expected heterozygosity values in the three species were higher than in other Neotropical catfishes such as *T. longipinnis* (He=0.393, Zamudio *et al.*, 2009), *S. parahybae* (He=0.470, Fonseca *et al.*, 2017), *P. reticulatum* (He=0.498–0.751, Abreu *et al.*, 2009; Vaini *et al.*, 2016; Prado *et al.*, 2017), and *P. corruscans* (He=0.709–0.760, Pereira *et al.*, 2009; Dantas *et al.*, 2013; Vaini *et al.*, 2016). Nonetheless, the genetic diversity observed in this study was lower than those found in *R. quelen* (He=0.614–0.909, Ríos *et al.*, 2017; 2019), *P. punctifer* (He=0.783, Telles *et al.*, 2014), *P. corruscans* (He=0.862, Prado *et al.*, 2017), and *P. maculatus* (He=0.871, Ribolli *et al.*, 2012).

In *S. cuspicaudus*, genotypic frequencies were distributed according to Hardy-Weinberg equilibrium and the inbreeding values were not significant save for sections S5 and S8 in which values lower than 10% suggest a careful management to prevent adverse effects regarding fitness (Frankham *et al.*, 2014). On the contrary, and despite their high genetic diversity, *A. pardalis* and *P. grosskopfii* exhibited a heterozygosity deficit that cannot be explained by the Wahlund effect in the only genetic group formed by *A. pardalis* nor in each of the two genetic groups of *P. grosskopfii* that co-migrate in the studied area, wherein the heterozygosity deficit stayed significant.

Unlike *P. grosskopfii*, which has external reproduction (Loir *et al.*, 1989), the assortative mating can explain the heterozygosity deficit in *A. pardalis* since it has internal insemination and elaborated courtships (Galvis *et al.*, 1997) and the females store spermatozoa inside their ovaries (Contreras *et al.*, 2012). However, to verify this hypothesis, it remains necessary to further research the reproductive behavior of these species.

Alternatively, the heterozygosity deficit could be explained by inbreeding as *A. pardalis* (2.600–8.600%) and *P. grosskopfii* (10.800–21.800%) showed significant F_{IS} values. This effect may result from different anthropic activities like overfishing due to diminishments in captures since 1994 (Jiménez-Segura,Villa-Navarro, 2011) and their presence in the first 10 species captured for the Magdalena-Cauca basin (AUNAP, 2020). Other factor that could negatively contribute to the studied catfish species is the contamination in basins caused by mining as in the studied zone there is gold, silver, and platinum extraction (UPME, 2019), and there is evidence of mercury bioaccumulation, a residual contaminant of this activity, in several species that cohabit in the zone (Marrugo-Negrete *et al.*, 2008; Álvarez *et al.*, 2012).

All those factors above mentioned may explain the recent decline in populations suggested by bottlenecks in the three studied species, supporting the hypothesis of genetic vulnerability of fishes due to anthropic interventions. A non-excluding alternative considers the climate factor as an additional influence of the demographic aspects of the populations of these species. Extreme changes in the Cauca River flow as consequences of the climatic phenomenon *El niño-Southern Oscillation* (ENSO) presented in the region in the prior years, wherein since 2007 there has been extreme NIÑA (2007–2008 and 2010–2011) and NIÑO (2009–2010, Hoyos *et al.*, 2013) phenomena, could have caused bottlenecks as described in the same region for *Curimata mivartii* (Landínez-

García, Marquez, 2018). This climate factor could contribute to bottlenecks phenomena associated to overfishing, which has been described for other Neotropical catfishes as *P. corruscans*, *P. reticulatum*, and *B. flavicans* (Coronel *et al.*, 2004; Prado *et al.*, 2018).

Genetic structure analysis. This study did not find significant differences among individuals of middle and lower sections of the Cauca River, which indicates that the environmental heterogeneity of the basin and, in particular, the steep slope of the Cauca River canyon is not a barrier for the gene flow of *P. grosskopfii*. However, such slope seems to limit of dispersion of *S. cuspicaudus* and *A. pardalis* in the middle section of the Cauca River since no individuals of these species were captured in the higher sections of the studied area in the sampled period (2011–2014). This result is not surprising for a short migrator like *A. pardalis*; nonetheless, the spatial distribution differed in the middle range migrators *S. cuspicaudus* and *P. grosskopfii*, suggesting that besides from their migration range, other factors as body conformation, habitat preferences and behavior, can play an important role in the observed spatial distribution, which is an important approach for future researches. A strong limitation to spatial distribution by the Cauca River canyon is also observed in *Pseudopimelodus atricaudus*, a new species described in the lower section of the Cauca River that diverged about 16 mya from its congener *P. magnus* distributed upstream the Cauca River canyon (Rangel-Medrano *et al.*, 2020; Restrepo-Gómez *et al.*, 2020).

In the spatial distribution analyzed for each species, the Bayesian analysis of STRUCTURE, the DAPC, the AMOVA and the pairwise distances showed absence of geographic population structure in the studied species. These results concord with the ones described for other Neotropical catfishes that perform medium and large distance migrations as *P. maculatus* (Almeida *et al.*, 2001, 2003; Ribolli *et al.*, 2012), *B. flavicans* (Coronel *et al.*, 2004), *B. rousseauxii* (Batista, Alves-Gomes, 2006), *P. corruscans* and *P. reticulatum* (Vaini *et al.*, 2016; Prado *et al.*, 2018). Nevertheless, our results with *A. pardalis* and *S. cuspicaudus* contrast with the results for *P. corruscans* in the basin of rivers Paraguay and Paraná wherein the authors hypothesize a possible homing behavior for the species (Pereira *et al.*, 2009; Dantas *et al.*, 2013).

Distinct from *A. pardalis* and *S. cuspicaudus*, each formed by one genetic group, *P. grosskopfii* represented two coexisting genetic groups in the evaluated sections of the Cauca River. This result in *P. grosskopfii* could reflect a possible spatial reproductive isolation, considering the preference for reproductive sites (*homing*) described for other species of Pimelodidae family as *B. rousseauxii* (Batista, Alves-Gomes, 2006), *P. corruscans* (Pereira *et al.*, 2009; Dantas *et al.*, 2013), and *P. reticulatum* (Abreu *et al.*, 2009). Another non-excluding alternative is that *P. grosskopfii* experiences temporary reproductive isolation as the Cauca River presents two hydrological periods of shallow and deep waters that could benefit two reproductive periods of the fish species that habit this basin (Jiménez-Segura *et al.*, 2010; López-Casas *et al.*, 2016). This explanation is supported in the observation of a greater number of individuals of *P. grosskopfii* participating in the migration during the first period compared to the second hydrological period of the Magdalena River (López-Casas *et al.*, 2016). Despite the latter, to the date, there is no detailed information as per the reproductive periods and behaviors of *P. grosskopfii* that allows to elucidate the origin of the genetic groups.

Under this context, it is recommended that the information obtained in this study

regarding the structure and diversity of three coexisting species of catfishes in the middle and lower sections of the Cauca River is considered within the expansion proposals of the hydroelectric development for Colombia (Angarita *et al.*, 2018) and of other anthropic activities. The higher values of the inbreeding coefficient were found for *P. grosskopfii*, included in 2016 as a critical danger species in the red list of the International Union for Conservation of Nature – IUCN (Villa-Navarro *et al.*, 2016), for which it is necessary to enforce the proposed measures for the recovery and conservation of natural populations of the species. Despite the inbreeding values found for *S. cuspicaudus* and *A. pardalis* are lower than 10%, which is the suggested limit to avoid substantial losses in the reproductive features (Soulé, 1980), it is advisable to continue monitoring these populations to evaluate the changes of the species over time since it has been indicated that inbreeding values higher than cero ($F_{IS}>0$) can cause negative effects in the fitness of natural populations (Frankham *et al.*, 2014).

In conclusion, employing species-specific microsatellite loci, this study determined the diversity and possible genetic structure in the middle and lower sections of the Cauca River for the Neotropical catfishes *A. pardalis*, *P. grosskopfii* and *S. cuspicaudus*. The genetic diversity levels of the three species (expected heterozygosity and number of alleles/*locus*) are high; however, all the studied species seem to have suffered recent bottlenecks and exhibit significant inbreeding values, which are greater for *P. grosskopfii*, currently in a higher risk category. Furthermore, *P. grosskopfii* shows the coexistence of two genetic groups while *A. pardalis* and *S. cuspicaudus* are mainly represented by a genetic group. Contrary to the expectations, none of the evaluated species showed population structure related to some heterogeneous features of the basin. It was also evidenced that the cannon of the Cauca River in this zone does not represent a barrier to the gene flow of *P. grosskopfii* but it seems to limit the dispersion of *A. pardalis* and *S. cuspicaudus*.

ACKNOWLEDGMENTS

REFERENCES

• **Abreu MM, Pereira LHG, Vila VB, Foresti F, Oliveira C**. Genetic variability of two populations of *Pseudoplatystoma reticulatum* from the Upper Paraguay River Basin. Genet Mol Biol. 2009; 32(4):868–73. https://doi.org/10.1590/S1415-47572009005000075

• **Almeida FS, Sodré LMK, Contel EPB**. Population structure analysis of *Pimelodus maculatus* (Pisces, Siluriformes) from the Tietê and Paranapanema Rivers (Brazil). Genet Mol Biol. 2003; 26(3):301–05. https://doi.org/10.1590/S1415-47572003000300014

- **Almeida FS, Fungaro MHP, Sodré LMK**. RAPD and isoenzyme analysis of genetic variability in three allied species of catfish (Siluriformes: Pimelodidae) from the Tibagi River, Brazil. J Zool. 2001; 253(1):113–20. https://doi.org/10.1017/S0952836901000103

- **Álvarez S, Jessick AM, Palacio JA, Kolok AS**. Methylmercury concentrations in six fish species from two Colombian rivers. Bull Environ Contam Toxicol. 2012; 88:65–68. https://doi.org/10.1007/s00128-011-0458-x

- **Angarita H, Wickel AJ, Sieber J, Chavarro J, Maldonado-Ocampo JA, Herrera-R GA** *et al*. Basin-scale impacts of hydropower development on the Mompós Depression wetlands, Colombia. Hydrol Earth Syst Sci. 2018; 22(5):2839–65. https://doi.org/10.5194/hess-22-2839-2018

- **Autoridad Nacional de Acuacultura y Pesca (AUNAP)**. SEPEC-Servicio Estadístico Pesquero Colombiano [Internet]. Bogotá; 2020. Available from: http://sepec.aunap.gov.co/

- **Barletta M, Jaureguizar AJ, Baigun C, Fontoura NF, Agostinho AA, Almeida-Val VMF** *et al*. Fish and aquatic habitat conservation in South America: A continental overview with emphasis on Neotropical systems. J Fish Biol. 2010; 76(9):2118–76. https://doi.org/10.1111/j.1095-8649.2010.02684.x

- **Batista JDS, Alves-Gomes JA**. Phylogeography of *Brachyplatystoma rousseauxii* (Siluriformes - Pimelodidae) in the Amazon Basin offers preliminary evidence for the first case of "homing" for an Amazonian migratory catfish. Genet Mol Res. 2006; 5(4):723–40. Available from: http://www.funpecrp.com.br/gmr/year2006/vol4-5/pdf/gmr0231.pdf

- **Betancur T, Mejía O, Palacio C**. Conceptual hydrogeology model to Bajo Cauca antioqueño: a tropical aquifer system. Rev Fac Ing Univ Antioquia. 2009; 48(1):107–18.

- **Carvalho DC, Oliveira DAA, Beheregaray LB, Torres RA**. Hidden genetic diversity and distinct evolutionarily significant units in an commercially important Neotropical apex predator, the catfish *Pseudoplatystoma corruscans*. Conserv Genet. 2012; 13:1671–75. https://doi.org/10.1007/s10592-012-0402-6

- **Contreras-C P, Zapata-B B, Rosado-P R**. Aspectos preliminares del manejo reproductivo en cautiverio de la doncella (*Ageneiosus pardalis* Lütken, 1874). Rev MVZ Cordoba. 2012; 17(3):3147–53. Available from: http://www.scielo.org.co/pdf/mvz/v17n3/v17n3a08.pdf

- **Coronel JS, Maes GE, Claus S, Van Damme PA, Volckaert FAM**. Differential population history in the migratory catfishes *Brachyplatystoma flavicans* and *Pseudoplatystoma fasciatum* (Pimelodidae) from the Bolivian Amazon assessed with nuclear and mitochondrial DNA markers. J Fish Biol. 2004; 65(3):859–68. https://doi.org/10.1111/j.0022-1112.2004.00498.x

- **Dahl G**. Los peces del Norte de Colombia. Bogotá (D.C.): Ministerio de Agricultura, Instituto de desarrollo de los recursos naturales renovables (INDRENA); 1971.

- **Dantas HL, Santos Neto MA, Oliveira KKC, Severi W, Diniz FM, Coimbra MRM**. Genetic diversity of captive and wild threatened catfish *Pseudoplatystoma corruscans* in the São Francisco River. Rev Fish Sci. 2013; 21(3-4):237–46. https://doi.org/10.1080/10641262.2013.800787

- **Evanno G, Regnaut S, Goudet J**. Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. Mol Ecol. 2005; 14(8):2611–20. https://doi.org/10.1111/j.1365-294X.2005.02553.x

- **Excoffier L, Lischer HEL**. Arlequin suite ver 3.5: A new series of programs to perform population genetics analyses under Linux and Windows. Mol Ecol Resour. 2010; 10(3):564–67. https://doi.org/10.1111/j.1755-0998.2010.02847.x

- **FAO**. Colombia: pesca en cifras/2014. Bogotá (D.C.): Ministerio de Agricultura y desarrollo rural, Organización de las naciones unidas para la alimentación y la agricultura (FAO); 2015. Available from: https://www.aunap.gov.co/wp-content/uploads/2016/05/Pesca_en_cifras.pdf

- **Ferraris CJ Jr**. Checklist of catfishes, recent and fossil (Osteichthyes, Siluriformes), and catalogue of siluriform primary types. Zootaxa. 2007; 1418(1):1–128. https://doi.org/10.11646/zootaxa.1418.1.1

- **Fonseca FS, Domingues RR, Hallerman EM, Hilsdorf AWS**. Genetic diversity of an imperiled Neotropical catfish and recommendations for its restoration. Front Genet. 2017; 8:196. https://doi.org/10.3389/fgene.2017.00196

- **Frankham R, Bradshaw CJA, Brook BW**. Genetics in conservation management: Revised recommendations for the 50/500 rules, Red List criteria and population viability analyses. Biol Conserv. 2014; 170:56–63. https://doi.org/10.1016/j.biocon.2013.12.036

- **Fricke R, Eschmeyer WN, Van der Laan R**. Eschmeyer's catalog of fishes: genera, species, references [Internet]. San Francisco: California Academy of Science; 2020. Available from: http://researcharchive.calacademy.org/research/ichthyology/catalog/fishcatmain.asp

- **Gallo H, Díaz-Sarmiento J**. Variabilidad genética del bagre rayado *Pseudoplatystoma fasciatum* (Pisces: Pimelodidae) en el río Magdalena (Colombia). Rev Acad Colomb Cien Exactas, Fis Nat. 2003; 27(105):559–606.

- **Galvis G, Mojica JI**. The Magdalena River fresh water fishes and fisheries. Aquat Ecosyst Health Manag. 2007; 10(2):127–39. https://doi.org/10.1080/14634980701357640

- **Galvis G, Mojica JI, Camargo M**. Peces del Catatumbo. Bogotá (D.C.): ECOPETROL; 1997.

- **Garza JC, Williamson EG**. Detection of reduction in population size using data from microsatellite loci. Mol Ecol. 2001; 10(2):305–18. https://doi.org/10.1046/j.1365-294x.2001.01190.x

- **Hilsdorf AWS, Hallerman EM**. Genetic resources of Neotropical fishes. New York: Springer International Publishing; 2017. https://doi.org/10.1007/978-3-319-55838-7

- **Hoyos N, Escobar J, Restrepo JC, Arango AM, Ortiz JC**. Impact of the 2010-2011 La Niña phenomenon in Colombia, South America: the human toll of an extreme weather event. Appl Geogr. 2013; 39:16–25. https://doi.org/10.1016/j.apgeog.2012.11.018

- **Jiménez-Segura LF, Galvis-Vergara G, Cala-Cala P, García-Alzate CA, López-Casas S, Ríos-Pulgarín MI *et al***. Freshwater fish faunas, habitats and conservation challenges in the Caribbean river basins of north-western South America. J Fish Biol. 2016; 89(1):65–101. https://doi.org/10.1111/jfb.13018

- **Jiménez-Segura LF, Palacio J, Leite R**. River flooding and reproduction of migratory fish species in the Magdalena River basin, Colombia. Ecol Freshw Fish. 2010; 19(2):178–86. https://doi.org/10.1111/j.1600-0633.2009.00402.x

- **Jiménez-Segura LF, Villa-Navarro FA**. *Pimelodus grosskopfii* Steindachner 1879. In: Lasso CA, Agudelo Córdoba E, Jiménez-Segura LF, Ramírez-Gil H, Morales-Betancourt M, Ajiaco-Martínez RE *et al*., editors. I. Catálogo de recursos pesqueros continentales de Colombia. Serie Editorial Recursos Hidrobiológicos y Pesqueros Continentales de Colombia. Bogotá (D.C.): Instituto de investigaciones de Recursos Biológicos Alexander von Humboldt; 2011, p.466–71.

- **Jombart T**. *adegenet*: a R package for the multivariate analysis of genetic markers. Bioinformatics. 2008; 24(11):1403–05. https://doi.org/10.1093/bioinformatics/btn129

- **Kopelman NM, Mayzel J, Jakobsson M, Rosenberg NA, Mayrose I**. Clumpak: a program for identifying clustering modes and packaging population structure inferences across K. Mol Ecol Resour. 2015; 15(5):1179–91. https://doi.org/10.1111/1755-0998.12387

- **Landínez-García RM, Márquez EJ**. Microsatellite loci development and population genetics in Neotropical fish *Curimata mivartii* (Characiformes: Curimatidae). PeerJ. 2018; 6:e5959. https://doi.org/10.7717/peerj.5959

- **Landínez-García RM, Márquez EJ**. Development and characterization of 24 polymorphic microsatellite loci for the freshwater fish *Ichthyoelephas longirostris* (Characiformes: Prochilodontidae). PeerJ. 2016; 4:e2419. https://doi.org/10.7717/peerj.2419

- **Li YL, Liu JX**. StructureSelector: a web-based software to select and visualize the optimal number of clusters using multiple methods. Mol Ecol Resour. 2018; 18(1):176–77. https://doi.org/10.1111/1755-0998.12719

- **Littmann MW, Burr BM, Nass P**. *Sorubim cuspicaudus*, a new long-whiskered catfish from northwestern South America (Siluriformes: Pimelodidae). Proc Biol Soc Wash. 2000; 113(4):900–17.

- **Loir M, Cauty C, Planquette P, Le Bail PY**. Comparative study of the male reproductive tract in seven families of South-Arnerican catfishes. Aquat Living Resour. 1989; 2(1):45–56. https://doi.org/10.1051/alr:1989005

- **López-Casas S, Jiménez-Segura LF, Agostinho AA, Pérez CM**. Potamodromous migrations in the Magdalena River basin: Bimodal reproductive patterns in Neotropical rivers. J Fish Biol. 2016; 89(1):157–71. https://doi.org/10.1111/jfb.12941

- **Luikart G, Cornuet J**. Empirical evaluation of a test for identifying allele recently data bottlenecked populations from frequency. Conserv Biol. 1998; 12(1):228–37. https://doi.org/10.1111/j.1523-1739.1998.96388.x

- **Mancera-Rodríguez NJ, Márquez EJ, Hurtado-Alarcón JC**. Uso de citogenética y técnicas moleculares en estudios de diversidad genética en peces colombianos. In: López-H A, editor. Biología Molecular aplicada a la producción animal y la conservación de especies silvestres., Medellín: Universidad Nacional de Colombia; 2013. p.237–312.

- **Márquez E., Restrepo-Escobar N, Yepes-Acevedo AJ, Narváez JC.** Diversidad y estructura genética de los peces de la cuenca del Magdalena, Colombia. In: Jiménez-Segura LF, Lasso CA, editors. XIX. Peces de la cuenca del río Magdalena, Colombia: diversidad, conservación y uso sostenible. Serie Editorial Recursos Hidrobiológicos y Pesqueros Continentales de Colombia. Bogotá, (D.C): Instituto de Investigación de Recursos Biológicos Alexander von Humboldt; 2020. p.115–57.

- **Marrugo-Negrete J, Verbel JO, Ceballos EL, Benítez LN**. Total mercury and methylmercury concentrations in fish from the Mojana region of Colombia. Environ Geochem Health. 2008; 30:21–30. https://doi.org/10.1007/s10653-007-9104-2

- **Meirmans PG**. Using the AMOVA framework to estimate a standardized genetic differentiation measure. Evolution (N Y). 2006; 60(11):2399–402. https://doi.org/10.1111/j.0014-3820.2006.tb01874.x

- **Meirmans PG, Hedrick PW**. Assessing population structure: FST and related measures. Mol Ecol Resour. 2011; 11(1):5–18. https://doi.org/10.1111/j.1755-0998.2010.02927.x

- **MejíaRivera O, Betancur-Vargas T, Londoño-Ciro L**. Aplicación de técnicas geoestadísticas en la hidrogeología del bajo Cauca antioqueño. Dyna. 2007; 74(152):137–49. Available from: http://www.scielo.org.co/pdf/dyna/v74n152/a12v74n152.pdf

- **Mojica JI, Castellanos C, Álvarez -León R, Villa-Navarro FA**. *Ageneiosus pardalis* Lütken 1874. In: Mojica JI, Usma-Oviedo JS, Álvarez-León R, Lasso CA, editors. Libro rojo peces dulceacuícolas de Colombia 2012. Bogotá (D.C.): Investigación de Recursos Biológicos Alexander von Humboldt, Instituto de Ciencias Naturales de la Universidad Nacional de Colombia, WWF Colombia y Universidad de Manizales; 2012a. p.71–73.

- **Mojica JI, Galvis G, Sánchez-Duarte P, Castellanos C, Villa-Navarro FA**. Peces del valle medio del río Magdalena, Colombia. Biota Colomb. 2006; 7(1):23–37.

- **Mojica JI, Usma-Oviedo JS, Álvarez-León R, Lasso CA**. Libro Rojo de peces dulceacuicolas de Colombia 2012. Bogotá (D.C.): Instituto de Investigación de los Recursos Biológicos Alexander von Humboldt, Instituto de Ciencias Naturales de la Universidad Nacional de Colombia, WWF Colombia, Universidad de Manizales; 2012b. https://doi.org/10.1007/s13398-014-0173-7.2

- **Nelson JS, Grande TC, Wilson MVH**. Fishes of the world. Hoboken, New Jersey: John Wiley & Sons; 2016. https://doi.org/10.1007/bf00005935

- **Oosterhout C Van, Hutchinson WF, Wills DPM, Shipley P**. MICRO-CHECKER: Software for identifying and correcting genotyping errors in microsatellite data. Mol Ecol Notes. 2004; 4(3):535–38. https://doi.org/10.1111/j.1471-8286.2004.00684.x

- **Ortega-Lara A, Lasso-Alcalá OM, Lasso CA, Pasquier GA, Bogotá-Gregory JD**. Peces de la subcuenca del río Catatumbo, cuenca del Lago de Maracaibo, Colombia y Venezuela. Biota Colomb. 2012; 13(1):71–99. Available from: http://revistas.humboldt.org.co/index.php/biota/article/view/258

- **Ortega-Lara A, Usma JS, Bonilla PA, Santos NL**. Peces de la cuenca alta del río Cauca, Colombia. Biota Colomb. 2006; 7(1):39–54.

- **Peakall R, Smouse PE**. GenALEx 6.5: Genetic analysis in Excel. Population genetic software for teaching and research-an update. Bioinformatics. 2012; 28(19):2537–39. https://doi.org/10.1093/bioinformatics/bts460

- **Pereira LHG, Foresti F, Oliveira C**. Genetic structure of the migratory catfish *Pseudoplatystoma corruscans* (Siluriformes: Pimelodidae) suggests homing behaviour. Ecol Freshw Fish. 2009; 18(2):215–25. https://doi.org/10.1111/j.1600-0633.2008.00338.x

- **Piry S, Luikart G, Cornuet JM**. BOTTLENECK: a computer program for detecting recent reduction in the effective population size using allele frequency data. J Hered. 1999; 90(4):502–03. https://doi.org/10.1093/jhered/90.4.502

- **Prado FD, Fernández-Cebrián R, Foresti F, Oliveira C, Martínez P, Porto-Foresti F**. Genetic structure and evidence of anthropogenic effects on wild populations of two Neotropical catfishes: baselines for conservation. J Fish Biol. 2018; 92(1):55–72. https://doi.org/10.1111/jfb.13486

- **Prado FD, Fernández-Cebrián R, Hashimoto DT, Senhorini JA, Foresti F, Martínez P, Porto-Foresti F**. Hybridization and genetic introgression patterns between two South American catfish along their sympatric distribution range. Hydrobiologia. 2017; 788:319–43. https://doi.org/10.1007/s10750-016-3010-5

- **Pritchard JK, Stephens M, Donnelly P**. Inference of population structure using multilocus genotype data. Genetics. 2000; 155(2):945–59. Available from: https://www.genetics.org/content/genetics/155/2/945.full.pdf

- **Puechmaille SJ**. The program structure does not reliably recover the correct population structure when sampling is uneven: subsampling and new estimators alleviate the problem. Mol Ecol Resour. 2016; 16(3):608–27. https://doi.org/10.1111/1755-0998.12512

- **Ramella MS, Kroth MA, Meurer S, Nuñer APDO, Zaniboni Filho E, Arisi ACM**. Genetic variability in four fish species (*Pimelodus maculatus*, *Prochilodus lineatus*, *Salminus brasiliensis* and *Steindachneridion scripta*) from Uruguay River Basin. Braz Arch Biol Technol. 2006; 49(4):589–98. https://doi.org/10.1590/S1516-89132006000500008

- **R Development Core Team**. R: A Language and Environment for Statistical Computing; 2017. Available from: https://www.R-project.org/

- **Rangel-Medrano JD, Ortega-Lara A, Márquez EJ**. Ancient genetic divergence in bumblebee catfish of the genus *Pseudopimelodus* (Pseudopimelodidae: Siluriformes) from northwestern South America. PeerJ. 2020; 8:e9028. https://doi.org/10.7717/peerj.9028

- **Restrepo-Escobar N, Márquez EJ**. Microsatellite loci development for three catfish species from northwestern South America. Neotrop Ichthyol. 2020; 18(1):e190079. https://doi.org/10.1590/1982-0224-2019-0079

- **Restrepo-Gómez AM, Rangel-Medrano JD, Márquez EJ, Ortega-Lara A**. Two new species of *Pseudopimelodus* Bleeker, 1858 (Siluriformes: Pseudopimelodidae) from the Magdalena Basin, Colombia. PeerJ. 2020; 8:e9723. https://doi.org/10.7717/peerj.9723

- **Restrepo-Moreno SA, Foster DA, Stockli DF, Parra-Sánchez LN**. Long-term erosion and exhumation of the Altiplano Antioqueño, Northern Andes (Colombia) from apatite (U-Th)/He thermochronology. Earth Planet Sci Lett. 2009; 278(1-2):1–12. https://doi.org/10.1016/j.epsl.2008.09.037

- **Ribeiro FRV, Rapp Py-Daniel LH, Walsh SJ**. Taxonomic revision of the South American catfish genus *Ageneiosus* (Siluriformes: Auchenipteridae) with the description of four new species. J Fish Biol. 2017; 90(4):1388–478. https://doi.org/10.1111/jfb.13246

- **Ribolli J, Melo CMR, Zaniboni-Filho E**. Genetic characterization of the Neotropical catfish *Pimelodus maculatus* (Pimelodidae, Siluriformes) in the Upper Uruguay River. Genet Mol Biol. 2012; 35(4):761–69. https://doi.org/10.1590/S1415-47572012005000060

- **Rice WR**. Analyzing Tables of Statistical Tests. Evolution (N Y). 1989; 43(1):223–25. https://doi.org/10.2307/2409177

- **Ríos N, Bouza C, García G**. Past hybridisation and introgression erased traces of mitochondrial lineages evolution in the Neotropical silver catfish *Rhamdia quelen* (Siluriformes: Heptapteridae). Hydrobiologia. 2019; 830:161–77. https://doi.org/10.1007/s10750-018-3861-z

- **Ríos N, Bouza C, Gutiérrez V, García G**. Species complex delimitation and patterns of population structure at different geographic scales in Neotropical silver catfish (*Rhamdia*: Heptapteridae). Environ Biol Fishes. 2017; 100:1047–67. https://doi.org/10.1007/s10641-017-0622-1

- **Sekine ES, Prioli AJ, Prioli SMAP, Júlio Júnior HF**. Genetic differentiation among populations of *Pseudoplatystoma corruscans* (Agassiz, 1829) (Osteichthyes, Pimelodidae) isolated by the Guaíra Falls in the Paraná River. Acta Sci. 2002; 24(2):507–12. Available from: http://repositorio.uem.br:8080/jspui/bitstream/1/5222/1/375.pdf

- **Soulé ME**. Thresholds for survival: maintaining fitness and evolutionary potential. In: Soulé ME, Wilcox B, editors. Conservation biology: an evolutionary-ecological Perspective. Sunderland: Sinauer Associates Inc; 1980. p.151–69.

- **Telles MP, Collevatti RG, Braga RS, Guedes LBS, Castro TG, Costa MC *et al***. Geographical genetics of *Pseudoplatystoma punctifer* (Castelnau, 1855) (Siluriformes, Pimelodidae) in the Amazon basin. Genet Mol Res. 2014; 13(2):3656–66. https://doi.org/10.4238/2014.May.9.8

- **Unidad de planeación minero energética (UPME)**. Producción, regalías y comercio exterior [Internet]. Bogotá (D.C.): Sistema de información minero colombiano (SIMCO).; 2019. Available from: http://www1.upme.gov.co/simco/Cifras-Sectoriales/Paginas/Informacion-estadistica-minera.aspx

- **Usma-Oviedo JS, Valderrama M, Escobar MD, Ajiaco-Martínez RE, Villa-Navarro FA, Castro F *et al***. Peces dulceacuícolas migratorios en Colombia. In: Naranjo LG, Amaya Espinel JD, editors. Plan nacional de las especies migratorias. Ministro de ambiente, Vivienda y Desarrollo Territorial, WWF Colombia; 2009. p.103–32. Available from: http://d2ouvy59p0dg6k.cloudfront.net/downloads/plan_migratorias_version_web.pdf

- **Vaini JO, Crispim BA, Silva DBS, Benites C, Russo MR, Grisolia AB**. Genetic variability of pure *Pseudoplatystoma corruscans* and *Pseudoplatystoma reticulatum* individuals in the Paraná and Paraguay River basins. Fish Sci. 2016; 82:605–11. https://doi.org/10.1007/s12562-016-0999-3

- **Villa-Navarro FA, Acero P A, Cala-Cala P**. Taxonmic review of Trans-Andean species of *Pimelodus* (Siluriformes: Pimelodidae), with the description of two new species. Zootaxa. 2017; 4299(3):337–60. https://doi.org/10.11646/zootaxa.4299.3.2

- **Villa-Navarro FA, Usma-Oviedo JS, Mesa-Salazar L, Sanchez-Duarte P**. *Pimelodus grosskopfii*. The IUCN red list of threatened species 2016:eT49829828A61473588 [Internet]. http://dx.doi.org/10.2305/IUCN.UK.2016-1.RLTS.T49829828A61473588.en

- **Villa-Navarro FA, Zúñiga-Upegui PT, Castro-Roa D, García-Melo JE, García-Melo LJ, Herrada-Yara ME**. Peces del alto Magdalena, cuenca del río Magdalena, Colombia. Biota Colomb. 2006; 7(1):3–22.

- **Zamudio KR, Robertson JM, Chan LM, Sazima I**. Population structure in the catfish *Trichogenes longipinnis*: Drift offset by asymmetrical migration in a tiny geographic range. Biol J Linn Soc. 2009; 97(2):259–74. https://doi.org/10.1111/j.1095-8312.2009.01209.x

AUTHOR'S CONTRIBUTION ⓒRT

Natalia Restrepo–Escobar: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing–original draft, Writing–review and editing.

Anny Johanna Yepes–Acevedo: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing–original draft, Writing–review and editing.

Edna Judith Márquez: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Visualization, Writing–original draft, Writing–review and editing.

ETHICAL STATEMENT

Environmental license of Ministerio de Ambiente, Vivienda y Desarrollo Territorial # 0155 of January 30th, 2009.

COMPETING INTERESTS

The authors declare no competing interests.

HOW TO CITE THIS ARTICLE

• **Restrepo-Escobar N, Yepes-Acevedo AJ, Márquez EJ**. Population genetics of three threatened catfish species in heterogeneous environments of the Cauca River, Colombia. Neotrop Ichthyol. 2021; 19(1):e200040. https://doi.org/10.1590/1982-0224-2020-0040