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Cover: Pseudo-flying animals and wind-dependent seed & spore dispersers – made with digital painting in Krita. © Melito Prinson Pinto



Range extension of *Isthmoheros tuyrensis*, a threatened species of fish (Cichlidae) in Panama: including new ecological and morphological data

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INTRODUCTION

Cichlids dispersed to northern Central America (from South America) early in the Cenozoic, long before the Plio-Pleistocene rise of the Isthmus of Panama (IOP; Matamoros et al. 2015). Currently, there is a relatively low species diversity and a limited number of cases of endemism within the Cichlidae in southern Central America. This is especially evident in the Pacific Slope of Eastern Panama (PSEP), as compared to the western side of Panama (which borders Costa Rica) and the rest of Central America, including Mexico (Matamoros et al. 2015). The PSEP includes 13 major river drainages starting from the west side of the Panama Canal (Comité de Alto Nivel de Seguridad Hídrica 2016) and is recognized within the Chocó Biogeographical Region (Matamoros et al. 2015).

Only two endemic species of cichlids have been documented in the PSEP, particularly in the Darien and Bayano River tributaries (Comité de Alto Nivel de Seguridad Hídrica, 2016). One of these is *Darienheros calobrensis* and the other is *Isthmoheros tuyrensis*, both recognized in monotypic genera (Rican et al. 2016). *Isthmoheros tuyrensis*, commonly known as “Aviente” in Spanish (González-Gutiérrez 2021), has been reported in both the Tuíra & Bayano river basins, in the Balsas & Urugantíto rivers within Darien National Park, as well as in the Mamatí river (Lyons 2020). It has been described as a detritivore with a lentic postcranial morphology (Rican et al. 2016).

Isthmoheros tuyrensis was previously classified in the genus *Vieja* (Kullander 2003; Garcés & García 2007; McMahan 2010; McMahan et al. 2015), however, Rican et al. (2016) concluded that *Vieja* is actually part of the herichthyine clade, while *Isthmoheros* is an amphilophine, more closely related to other middle American genera such as *Amatitlania*, *Amphilophus*, and *Parachromis*, among others. Moreover, Rican et al. (2016) stated that *Isthmoheros* has its sister genus on the opposite side of the Isthmus in western Panamá and southeastern Costa Rica (i.e., *Talamancaheros*), which also present a detritivore cranial morphology and a lentic postcranial morphology with an obscure breeding coloration (vs. a scraping cranial and a lotic postcranial morphology with a white and black breeding coloration). Moreover, despite some ecomorphological differences, both *Isthmoheros* and *Talamancaheros* share a similar semi-herbivorous diet, based in tooth morphology (Conkel 1993), and they are separated, according to Rican et al. (2016), by a long-isolated monophyletic lineage within the amphilophines, being the sister clade

of the aforementioned *Darienheros* plus *Panamius* (Matamoros et al. 2015; Rican et al. 2016).

Isthmoheros tuyrensis faces several threats due to the increased spread of human activities in the eastern region of Panama; mainly due to the expansion of the urban footprint of the Panama City, originating from the Pacific entrance of the Panama Canal. Information on distribution and ecology of *I. tuyrensis* in the Eastern side of the IOP is relatively scarce and only a few comparative studies on ecomorphology have been done from collections in the Bayano River tributaries and the Darien region (Rican et al. 2016). Moreover, river basins towards the west of Panama (i.e., in the Panama District), have been relatively under sampled for freshwater fish species in general, including a lack of information on the distribution of endemic cichlids such as *I. tuyrensis*.

In this paper we report a new distribution range extension for *I. tuyrensis* in the Panama City area. This record is presented after conducting seasonal surveys in three river basins of the region. A morphological description of specimens is included as taxonomical validation for this new range extension. Moreover, our survey locations give us an idea of the potential barriers and distribution limits leading to the possible threat of extirpation of the species in this area, in particular from heavy pollution towards the west of Panama. Finally, we also provide and discuss data on several environmental parameters as a reference for the species' habitat condition in this region. This information will be relevant for future taxonomic and conservation studies, contributing to a better understanding on the biology of the species.

MATERIALS AND METHODS

Study Area

Although this paper is focused on specimens collected only in the Pacora river basin, the sampling effort was part of a broader study between September 2020–May 2021 in three rivers in the District of Panama: Matasnillo, Juan Díaz, and Pacora (Figure 1). All these rivers drain to the Pacific Ocean via the Bay of Panama and experience different levels of degradation due to human activities. These rivers are surrounded by commercial, industrial, and residential land, with an estimated of 1,098,068 people residing in an area of 191 km² (i.e., 540 inhabitants/km²) (Municipio de Panamá 2019). Pressures such as water diversion, extraction of sand & gravel, polluted runoff from nearby agricultural & livestock production, improper use of soils, sanitary

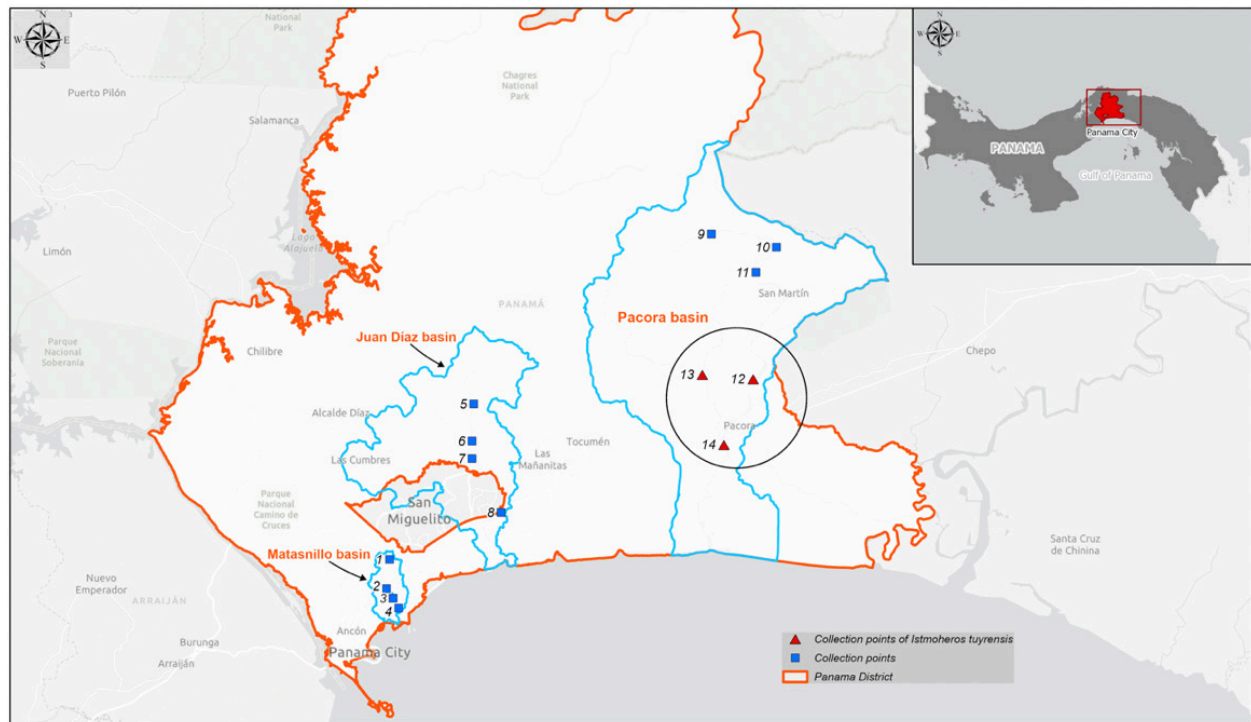


Figure 1. The three sampled basins and localities in the Panama District area (Panama City), specific locations where *I. tuyrensis* was collected are pointed out.

landfills, urban development near drainage areas, sedimentation resulting from deforestation, and untreated sewage affect these three rivers. Moreover, previous reports suggest that these impacts are higher in the city's western side (ANAM 2009).

The Matasnillo River is the main tributary of river basin No. 142 (between Caimito and Juan Díaz River) and is located in the center of Panama City. It is 6 km long, with an annual precipitation of 1,500 l/m² and a 33 m³/s flow. The whole basin, divided by the Panama Canal to the west, has an area of 137 km² and according to the last Panama census of population in 2010 has an estimated of 1,013,714 inhabitants. Both the Arraiján District at the west side of the Panama Canal and the main river at the city center are extremely channelized with little vegetation (Comité de Alto Nivel de Seguridad Hídrica, 2016). Compounding these threats are several sites where sewage tanks occupy river and stream easements, in parallel, the uncontrolled urban development hinders sewage infrastructure maintenance and repairs; this is a critical problem for many urban rivers in the Republic of Panama (MINSa 2019).

The Juan Díaz basin (basin No. 144) includes some of the largest rivers in the east side of the city. The basin is 351 km² & 22.5 km long, with an annual precipitation of 3,000 l/m² & flow of 5.1 m³/s, and an estimated of

868,401 inhabitants (Comité de Alto Nivel de Seguridad Hídrica 2016). To date, the biodiversity of this basin has not been well studied and there is a lack of awareness regarding the area's natural capital, although recent studies have highlighted its importance and relevance at the ecosystem level (Charris-Palacios 2020). Moreover, there are several high-income housing projects currently planned in the basin, which threaten these natural areas and are faced with opposition from local communities, which depend on drinking water from the river (Ruiz 2018). The upper basin is also used by some local communities as a tourist attraction.

The Pacora River basin (basin No. 146) is 368 km² & 48 km long, with 2,750 l/m² in annual precipitation, an average flow rate of 11.1 m³/s, and about 253,131 inhabitants (Comité de Alto Nivel de Seguridad Hídrica 2016). Although the Pacora River faces many of the same threats from expanding urbanization from the east, it also hosts some of the most important natural features (including beaches, pools, waterfalls, and forested areas) in the city. These attractions are connected to the city by a relatively good road network and are enjoyed predominantly by the local communities in addition to in-country tourists. The ecotourism potential of this area is recognized by the local government in the Panama City Resilience Strategy (Municipio de Panamá

2019), which recommends promoting the river's natural ecosystems and biodiversity, along with training tour guides, as a potential income generation activity for local communities. In addition, the Pacora River is a source of drinking water (after treatment in nearby plants), although it's not the sole water source for the urbanized area (García-Armuelles 2020).

Sampling effort at the three explored rivers

Fish sampling was conducted in the three river basins in a one-week period per season, including August–September (rainy season) 2020, February–March (dry season), and April (transition season) 2021. A total of 13 sampling sites were selected from the upper, middle, and lower river basins (Figure 1). At each site, depending on the riparian river structure, physiography, and river length & width, we selected a representative 100-m long transect. Fish were sampled in each transect using an Electro fisher (Halltech, HT-2000, 2020) for 45 minutes, according to the methodology described by Barvour et al. (1999), with voltage limits to 250 volts for areas with high conductivity ($>300 \mu\text{S}/\text{cm}$) and 750 volts for water with moderate to low conductivity ($100\text{--}300 \mu\text{S}/\text{cm}$). Since saltwater intrusion limits the use of electrical devices, we also employed a 213 cm long cast net with 1 cm mesh width, utilizing random throws for 30 minutes at each 100 m long transect.

Testing of water quality and physical parameters were carried out mostly using a multi parameter device (YSI Professional Plus 2015). The basic parameter data presented here include water temperature, pH, dissolved oxygen, and conductivity. Water samples were also collected in each locality to test for nitrate and fecal coliform levels. Samples were analyzed by a local laboratory (Ambitek Service Inc). River width was measured using a metric tape, depth with a limnometric rod, and flow with a current meter (Global Water BA1100 Model Fp111 Probe 3.7–6', 2017). Forest condition was assessed according to Munné et al. (2003) including qualitative in situ observations of gallery forest (as coverage percentages) within the upper, middle and lower sections on each river with 50 m long transects measuring on each side of the main riverbed.

Matasnillo River

Four fixed monitoring stations were established for repeated sampling in this basin during all seasons, one in the upper basin, one in the middle basin, and two in the lower basin. A single 100 m long section, due to the narrow river width, was sampled at each locality, totaling 400 m of sampling per season. This means that a

total of 1,200 m were sampled during a total of 540 min (9 h), with heavy limitations due to saltwater intrusion, mainly in the lower basin. The river condition's regarding obstructive garbage, such as metal wires and cement structures, in addition to heavy pollution from sewage disposal, made it impossible to use a cast net in this river basin.

Juan Díaz River

Three fixed monitoring stations were established for repeated sampling in this basin during the rainy season and in-between seasons. During the dry season, an additional site located on the upper basin, inaccessible during the rainy season, was sampled. At all localities (one in the upper basin, one in the middle basin, and one in the lower basin), two 100 m long sections were sampled, totaling 600 m sampled during the rainy season, 600 m between seasons, and 800 m during the dry season. Grouping all the sections monitored during the three seasons using electrofishing, a total of 2,000 m were sampled for 900 min (15 h). For cast net sampling, we spent a total of 600 min (10 h) in this basin across all seasons.

Pacora River

Surveys were conducted in this basin in five fixed monitoring stations during the rainy season and the transition season. Due to the inclusion of one site that was unreachable in other seasons, six stations were monitored during the dry season. At all localities (one in the upper basin, one in the middle basin, and one in the lower basin), two 100 m long sections were sampled. For the rainy and transition seasons we completed a total of 1,000 m sampled; during the dry season a total of 1,200 m were sampled with electrofishing. Considering all the sections monitored during the three seasons, we had a total of 3,200 m sampled for 1,440 min (24 h). For cast net sampling we spent a total of 960 min (16 h).

For practical purposes, this paper is focused on the localities where specimens of *I. tuyrensis* were found (Tables 1 & 2).

Species identification and morphological assessment

For the identification of the species collected we consulted the specialized literature (e.g., Bussing 1998; Gonzalez 2021), including revisionary works and the original description of the species known to occur in the sampled area after Matamoros et al. (2015) and Rican et al. (2016). Specimens identified as *I. tuyrensis* were retained (both preserved and alive), photographed, and measured (see Table 3) according to McMahan et al.

Table 1. Collection localities, season, collecting methods and number of individuals of *Isthmoheros tuiyensis* found in the Pacora River.

Site number	Elevation (m)	Basin / Season*	Method	Ind	Size (cm)	Weight (g)
12	40	M/T	Cast net	1	16.0	63
13	52	M/R	Electrofishing	1	8.1	10
13	52	M/T	Electrofishing	1	8.1	10
14	20	L/ T	Cast net	2	8.1	10

*M—Middle Basin | L—Lower Basin | R—Rainy Season | T—Transition Season | Ind—Number of individuals collected

Table 2. Average Physical Parameters in the Pacora River, Panama City.

Parameter	Units	Average	Minimum	Maximum	SD
Temperature	°C	27.9	23.0	31.6*	2.14
PH	-	7.73	7.01	8.5	0.28
Conductivity	μS/cm	166.5	124.4	207.7**	21.83
DO	%	7.90	6.11	9.93	1.03
TDS	mg/L	87.04	2.8***	206.2	44.83
Nitrate	Mg/L N-NO ₃	1.87	0.5	5.2****	1.53
Fecal Coliform	MPM/100 mL	1897	63	7701****	3126
Flow	Meters/ second	0.29	0	2.44	0.40
Width	M	22.47	6	53.6	9.30
Depth	Cm	39.08	0.8	100	27.76
Forest Condition	%	53	20	90*****	28.31

*Dry and transitional season, middle and lower basin | ** Transitional season, lower basin | *** Rainy season, middle basin | **** Rainy season, lower basin | ***** Upper basin.

(2015) and Rican et al. (2016). Counts (see Table 3) were done on preserved specimens according to Rican et al. (2016). Comparative morphometric and meristic data was obtained from the literature (McMahan et al. 2015; Rican et al. 2016).

RESULTS

Fish diversity

From a total of 9,259 fish specimens found in the sampled period, including 21 families, 40 genera, and 43 species (data under analysis for further publication), there were only two species of native cichlids. The most abundant species, with a total of 374 individuals, was the Chogorro (*Andinoacara coeruleopunctatus*). Of these 374 specimens, 134 were collected in Juan Díaz and 240 in Pacora, with zero individuals in Matasnillo.

On the other side, only five specimens of *I. tuiyensis* were collected (see morphological details/data below), all in the Pacora River (Table 1). Of these five specimens, two were found in the middle basin and captured with

electrofishing (Figure 1, Site 13, Table 1), one was collected during the rainy and the other in the transition season; a third specimen was found at a middle basin (Figure 1, Site 12, Table 1), during the transition season; and the last two specimens were captured in the lower basin (Figure 1, Site 14, Table 1), during the transition season. These last three specimens (Sites 12 and 14) were captured using cast net.

At the time of this writing, three specimens of *I. tuiyensis* are preserved and housed at the “Dr. Luis Howell Rivero” Museum at the Center of Marine Biology and Limnology (CCML) in the University of Panama, Catalogue number MBML No 2151; while the two other specimens are maintained alive in an aquarium located in the International Maritime University of Panama’s laboratory, in the Faculty of Marine Sciences. Pictures of one preserved specimen are provided in Images 1–2. Live specimens of *I. tuiyensis* are illustrated in Images 3–5.

Two exotic species of cichlids were also collected during our study. A total of 426 individuals of the Nile Tilapia (*Oreochromis niloticus*) were captured, 423

in Juan Díaz, 3 in Pacora, and zero in Matasnillo. A single specimen of the Jaguar Guapote (*Parachromis managuensis*), which is native from Honduras, Nicaragua, and Costa Rica, was found in the Juan Díaz river.

Species identification (*I. tuyrensis*) and morphological assessment

Identification of fishes as *I. tuyrensis*, comprising a new record for the Pacora river, was based on the following combination of distinctive characteristics which separate it from the other cichlid species occurring in the southern portion of Central America: body relatively robust and wide; second lower lip missing; teeth conical, without second cusp, but with tip labiolingually flattened; lateral line scales 31–32; anal fin spines 6–7; and coloration pattern (body grayish-green to greenish-brown, with 8–9 lateral black blotches, and several longitudinal series of small dark spots on the sides and fins). Complementary morphometric and meristic data for the species, based on three specimens measured, and analyzed, is provided in Table 3.

Environmental parameters at the sampled localities

The results for the environmental analysis are restricted to the Pacora River, since it is the only river where *I. tuyrensis* was found. The physiochemical and physical parameters are detailed in Table 2.

Physicochemical parameters

Temperature averaged 27.9 °C, with maximum values in the dry season. The average pH was 7.73, with a maximum of 8.5 and similar values across all seasons. Conductivity showed high variation with higher values in the lower basin. Dissolved oxygen averaged 7.90%, with relatively low variation and maximum values in the upper basin. Nitrate levels showed an average of 1.87, increasing in the middle and lower basins during the rainy season. Fecal Coliform concentration showed higher values in the localities at the middle and lower portion of the basin, increasing during the rainy season (Table 2).

Physical parameters

Water flow averaged 0.29 m/s; with a range from 0 (no current) –2.44 m/s. River depth and width showed strong variation across sampled localities, both increasing during the rainy season. Forest coverage tended to increase towards the middle and upper basins, with a maximum of 90% coverage in the upper basin and an average of 53% along the entire basin (Table 2).

Table 3. Morphometric and meristic data of individuals of *I. tuyrensis* found in the Pacora River. Head measurements are expressed as percentages of the head length; body measurements are expressed as percentages of the standard length.

Measurement/Count	N1	N2	N3
Total length (cm)	10.51	10.12	19.47
Standard length (cm)	8.10	8.10	16.00
Head length (cm)	2.69	2.40	4.69
Snout length	27.43	25.54	26.08
Mouth length	16.43	19.85	19.09
Eye diameter	29.33	27.23	25.00
Post-ocular length	42.35	46.15	42.74
Head depth	106.57	120.00	130.11
Predorsal length	31.51	32.28	29.18
Prepectoral length	33.70	30.78	31.07
Pectoral length	28.24	27.95	30.60
Prepelvic length	35.67	30.59	35.96
Pelvic length	25.35	29.63	28.23
Preanal length	64.71	67.40	64.12
Dorsal fin base	55.31	56.48	59.07
Dorsal fin height	11.29	13.29	12.93
Anal fin base	24.26	25.02	24.05
Body depth	47.25	50.59	51.03
Caudal peduncle length	11.41	12.10	12.22
Caudal peduncle depth	14.06	15.34	14.91
Dorsal fin elements	XVI, 10	XVII, 10	XVII, 11
Pectoral fin elements	14	13	14
Anal fin elements	VII, 7	VI, 8	VI, 7
Lateral line scales	32	31	32

DISCUSSION

Species distribution and environmental limitations

Toward the western-most range limit for *I. tuyrensis*, Loftin (1965) reported its presence almost 6 km east from the town of Pacora (not the river basin), near the Señora River, which drains to the Bayano River. Our findings report, for the first time, the presence of this species in the Pacora River basin, about 20 km in a straight line from previously known localities in Central Panama. This finding raises questions about the possible past distribution of the species in other rivers in Central/Western Panama, given there is no previous records of the species in the rivers of Panama City.

In the neighboring Juan Díaz river, not a single specimen of *I. tuyrensis* was found during the three sampled seasons, although another native and even two others exotic (more generalist and less sensitive to



Image 1. Preserved specimen of *I. tuyrensis*, right side, 16 cm, collected at the Prison La Joya (Site #12) on transition season in May 2021, picture from 3 July 2021. © Javier Pardo, UP.



Image 2. Preserved specimen of *I. tuyrensis*, left side, 16 cm, collected at the Prison La Joya (Site #12) on transition season in May 2021, picture from 3 July 2021. © Javier Pardo, UP.



Image 3. Live individual of *I. tuyrensis*, 15 cm, kept at the UMIP Aquarium, collected at the Restaurante Cabobre (Site #13) in May 2021, picture from 13 December 2021. © Jafet Santos, UMIP.



Image 4. Same live individual of Image 3 kept at the UMIP Aquarium, 18 cm, picture from 21 March 2022. © Javier Pardo, UP.



Image 5. The two live individuals collected at the Restaurante Cabobre (Site #13) kept at the UMIP Aquarium, 18 cm and 10 cm, picture from 21 March 2022. © Javier Pardo, UP.

environmental changes/disturbances) cichlid species were found. This river (Juan Díaz) is surrounded by densely populated areas at its middle and lower portions and is currently undergoing a rapid urbanization process. It is most likely an unsuitable habitat for *I. tuyrensis*, although that does not mean that this species was not present in this river before 1970 when the uncontrolled urbanization started (Municipio de Panamá 2019). Both Juan Díaz and Pacora Rivers drain to the Panama Bay Ramsar Site (Kaufmann 2012; Suman 2014), however *I. tuyrensis* is not reported in any study in the area; moreover, in the past, only 9 individuals were found in surveys on the Darien Province in rivers such as Balsas, near the Colombian border (Garcés & García 2007). Unfortunately, this Ramsar Site does not include the middle river basin and covers little freshwater habitat, even though watersheds can be considered wetlands according to the Ramsar technical classification (Ramsar Convention Secretariat 2016).

Conservation issues

Some conservation issues, mainly related to the agricultural-urban expansion, habitat loss, and pollution, that are affecting negatively the current conservation and populational status and distribution of *I. tuyrensis* are discussed below.

Agricultural-urban expansion vs. habitat loss

A study of land uses in the Pacora River basin conducted by Rodríguez-Martínez (2019) discusses transitions of land type measured through Geographic Information Systems (GIS) via three classifications—deforestation, gain or loss of agricultural land, and expansion of the urban footprint; with a variation trend between the periods corresponding to the years 1992, 2000, 2009, and 2019. The transition that presented the greatest magnitude of variation was deforestation, with the greatest loss between 2009 and 2019 (4,996.4 ha). These results indicate that the most significant transition that occurred was the transformation of forest land into pasture for livestock, with losses of 3,031.7 ha, 1,991.6 ha, and 3,466.9 ha, respectively, in the three periods assessed. Another significant change presented in the study is the growth of the urban footprint, which went from 259.6 ha in 1992, to 642.2 ha in 2000, to 2,412.0 ha in 2019; a tenfold increase in less than 30 years. A management plan proposal for the Pacora River basin developed by PREVDA (2008) states that the basin is (in addition) exposed to a series of climate risks and extreme events ranging from heavy rainfall and floods to some periods of drought. Moreover, we observed

that the river was blocked in multiple areas by local communities, especially during dry season, in order to create swimming areas. Currently there are no studies at this basin addressing the impact of the aforementioned factors, as well as of the deforestation due to livestock increasing, on the water availability, hydrological capacity, and biodiversity.

The impact from pollution

Regardless of the habitat connectivity and of their ability to survive in estuarine areas (as we found some specimens in the lower Pacora basin). Our surveys indicated that the Pacora River has an average pH of 7.7, with maximum values of 8.5, pointing to relatively alkaline waters with significant mineral input and less accumulation of organic material (Nilsson & Renöfält 2008). The presence of anthropogenic impacts from agricultural activity and urban sewage in some areas can contribute to denitrification, which may cause an increase of pH levels (He et al. 2017). On the other hand, our nitrate values showed an average value of 1.87 mg NO₃-N/l, with a maximum of 5.2 mg NO₃-N/l; little surprising was the fact that we did not find individuals of *I. tuyrensis* in localities with the highest nitrate values (Table 1 & Table 2). Although no information is available on nitrate toxicity for *I. tuyrensis* or for any of its close relatives, some studies have found that many freshwater fishes can exhibit increased mortality with nitrate levels between 1.1 and 4.5 mg NO₃-N/l (Camargo et al. 2005).

Many studies, on the other hand, have considered the agricultural leaching as the major factor driving the increase of conductivity and dissolved solids. For instance, in the geographically proximate region of the Costa Rican Pacific, Pérez-Castillo & Rodríguez (2008) incorporated the conductivity variable in their analyses of water quality in lagoons of the Palo Verde National Park, considering it an indicator for inorganic fertilizer presence and poor water quality. They established a 250 µS/cm maximum value for uncontaminated waters and a value of 1,500 µS/cm for heavily polluted waters. In other studies, specifically the Rincón River basin, also in the Costa Rican Pacific region, Beita-Sandí & Barahona-Palomo (2010) determined that average conductivity was 161.8 µS/cm, with a range from 92.7 µS/cm up to 249.6 µS/cm, thus, suggesting the area to be free of marine influence since none of the records exceeded 45.2 µS/cm (Villegas-Arguedas 2011). For the Pacora River, our minimum conductivity values were 124.4 µS/cm, with an average of 166.5 µS/cm, and tended to be higher in the middle and lower basin. This may be a product of the cumulative impact of agricultural activities from the

upper to the lower sections of the basin.

Moreover, Beita-Sandí & Barahona-Palomo (2010) found that the conductivity range in natural fresh waters in some Pacific Rivers in Costa Rica varied between 10 $\mu\text{S}/\text{cm}$ & 350 $\mu\text{S}/\text{cm}$, while in areas with marine influence the values varied between 125 $\mu\text{S}/\text{cm}$ & 2,200 $\mu\text{S}/\text{cm}$. We consider our conductivity and other pollution indicators discussed here to be high in the lower section of Pacora River, leading us to infer that the river, in addition to those discussed above, also has a marked marine influence. However, since higher conductivity values were obtained from collection localities near urbanized areas, we cannot conclude that these values are a natural characteristic of this river.

Although we used different voltage settings, electrofishing was probably affected in some cases by the high conductivity; on the other hand, in the lower & middle basin most collections were done using cast nets. More studies are necessary to confirm if *I. tuyrensis* prefer particular conditions at proximity with estuaries as occur in other cichlid genera (e.g., *Vieja*; Bussing 1998). Moreover, several studies mention that some fish can prefer aquatic habitats with specific requirements such as elevated values of water conductivity, but this can vary among species (Vieira & Tejerina; Garro 2020). The few individuals of *I. tuyrensis* that we found in our study were collected in sites in the middle and lower river sections with intermediate to relatively high conductivity values. Thus, there is a possibility that these conductivity values are negatively affecting the population status of the species since they may be due to anthropogenic activities.

Authors including Mondal & Bath (2020) have found that conductivity and total dissolved solids affect negatively the water quality conditions; thus, tolerant species, particularly those peripheral and with broader distributions on the whole basin (which does not seem to be the case of *I. tuyrensis*), are able to survive on high conductivity values. The same authors also stated that increased total dissolved solids and conductivity is related to reduced species richness and diversity of freshwater fish in tropical river basins. The same review concludes that an increase of nutrient contents in the water leads to an increase in primary productivity and persistence of periphyton feeding fishes, producing excessive algal growth, increased sediments, and an imbalanced food chain, which, again, seems not to be the case for *I. tuyrensis* a detritivorous species (Rican et al. 2016).

Since the bacteria *Escherichia coli* is predominant in sewage; we consider that fecal pollution may represents

a potential threat to *I. tuyrensis*. A study by Guzmán et al. (2004) where *E. coli* concentration was determined in digestive tracts and muscles of two species of fishes (*Jenynsia multidentata* and *Bryconamericus iheringi*) sampled at the same sites, showed higher concentrations of the bacteria in *J. multidentata* than in *B. iheringi*, thus indicating that the former species is more sensitive to the accumulation of the bacteria. Moreover, these authors concluded that increased bacteria concentration compromises the immunological system of these fish. Although we found no specific information for cichlids and considering that *E. coli* (measured by us as fecal coliform) is present in variable concentrations in all the sampled localities, we can conclude that *I. tuyrensis* is a sensitive species, since it was not found in the most polluted rivers (e.g., Juan Díaz;). On the other hand, no other cichlids were found in the nearby and heavily polluted Matasnillo river, which can provide us information on the tolerance levels of the species of this family to the fecal pollution.

Conservation measures

Habitat conditions and the permanence of this endemic species in the PSEP is not guaranteed if measures are not taken to control agricultural and urban footprint expansions. The Pacora River has a population of one of the only two endemic species of cichlids from this region, which is struggling to survive after its possible disappearance from nearby rivers toward the west. In terms of planning and environmental policies, recent management plans are non-existent except for an expired initiative that proposed integral management for the basin more than 10 years ago (PREVDA 2008). There are multiple threats to the Pacora river, and this species, posed by increased water demand for livestock, crops and industry, including the extraction of gravel, sand, and of non-metallic minerals directly from the river. These factors affect the biophysical and social components of the river basin where local communities, mostly living in poverty, are fighting for the right to healthy rivers, and ecosystems (Espinoza 2021).

Freshwater fishes are among the most threatened groups of species on the planet (Lacy et al. 2017). They have persisted for decades in tropical river basins and their ecological/environmental and socio-economical value is probably not fully understood by human communities, particularly in urban cities. As in other countries (see Lacy et al. 2017), neither local Panamanian stakeholders nor governments consider freshwater fishes to be a priority group in their Environmental Impact Assessment processes. We hope that this study can begin raising

awareness for riverine fishes and particularly for members of the Cichlidae. Panama's central and local governments should monitor biological indicators in its rivers and set priorities such as connecting the sewage system to the Juan Díaz Treatment Plant and increase sewage treatment capacity, instead of depositing sewage directly to the river (MINSA 2019; Municipality of Panama 2019). We also recommend training local tourist guides for eastern rivers such as Pacora, including the recognition of their unique and local biodiversity. Finally, actions outlined in the Panama City Resilience Strategy should be implemented within the next 10 years, according to existing regulations (Municipio de Panamá 2019).

Other functional and taxonomical aspects

Previous studies on *I. tuyrensis* noted the preference of this species for slow-moving waters. However, for the Pacora River, particularly at the lower basin, current velocity is relatively high. This is typical from rivers in this region of Panama, which is characterized by steep profiles and a shorter distance to the coast. This contrast, for example with the Bayano & Darien Rivers, in particular the Tuyra & Balsas rivers, where freshwater wetland ecosystems include lagoons with aquatic plants adapted to intermittent flooding (Ibáñez & Flores 2021).

Regarding the morphological data, despite our specimen count is scarce due to the low population densities of the species, the information provided here agree with the morphometric and meristic information published by previous authors (e.g., Kullander 2003; Rican et al. 2016); moreover, this study adds new and relevant information on the morphology of *I. tuyrensis*, contributing to its further diagnosis and characterization. This information could be relevant for taxonomic and descriptive studies, as well as in applied ecology research. Finally, our data provided limited information in terms of size classes, since four specimens measured about 8 cm and the maximum size reported was 16 cm. In this regard, previous authors (Kullander 2003; Rican et al. 2016) reported a maximum size of 23.5 cm for the species.

GENERAL CONCLUSIONS

The distribution limit for *I. tuyrensis* towards the west side of the PSEP is extended with our findings as the previous westernmost reports are limited to the Chichebre and Señora Rivers in the Bayano River basin (Lyons 2020). Most rivers of Panama City, such as the

Juan Díaz, are heavily polluted and this could prevent or limit the presence of this species, which, based on our data, can be considered as sensitive to pollution. For the Matasnillo River, pollution and deforestation are even higher; moreover, this river shows high marine influence and conductivity values that exceeds 400 $\mu\text{S}/\text{cm}$ in most sites. These issues (pollution, deforestation, river salinization, among others) call for the urgent implementation of restoration, conservation, and sanitation programs for all these rivers. This includes updating and implementing 1—the Pacora River Management Plan, buffering the spread of new urbanizations in the basin and 2—the Territorial Ordination Plan for the Panama City, which has already been developed, but is pending approval (IDOM SUMA CONTRANS 2017).

Although our study expands the geographic range of *I. tuyrensis* and furthers biological understanding of the species, it does not alter the fact that this species is listed as Vulnerable and likely to become endangered based on the criteria of the IUCN Endangered Species Red List. The relatively few known populations of this species (less than 10, based on Lyons 2020, including the new reported in this study) as herein discussed, are exposed to several threats including deforestation, agricultural expansion, mining activities, and road infrastructure development among others, not only on the central and western portion of the country but also within the Darien Region (Lyons 2020; Arcia-Jaramillo 2022). As we pointed out, this species is virtually lacking any effective protection along their distribution range, even in the eastern portion of the country (Arcia-Jaramillo 2022).

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Abstract: There are two endemic species of Cichlidae in southern Central America, both found in the Pacific Slope of Eastern Panama (PSEP). One is *Isthmoheros tuiyensis*, which until now was presumed to be distributed in the Darien Province and the Bayano River basin. Information on distribution and ecology of *I. tuiyensis* is relatively scarce. In this investigation we report a new range extension for the species and provide additional morphological and ecological data. Fish were sampled using electrofishing and cast nets, in three river basins of the Panama District (Matasnillo, Juan Díaz, and Pacora) from August–September (rainy season) of 2020, February–March (dry season), and April (transition season) of 2021. Fish diversity, water quality, and physical parameters were gathered within the upper, middle, and lower portions of the three basins. This study focused on the localities where specimens of *I. tuiyensis* were found (i.e., Pacora river basin). The presence of the species in localities with significant anthropogenic threats results in a potential barrier for distribution, along with the possibility of extirpation due to heavy pollution – in particular from the rivers on the western side of Panama City. In addition, we note an increase in urban threat from the east of the city due to expanded development and agricultural activities. *I. tuiyensis*, the virtually unknown “Aveinte” in Spanish or the “Isthmian Hero”, is listed as Vulnerable by the International Union for Conservation of Nature (IUCN)’s Red List and inhabits some river basins lacking effective protection, being the only endemic fish species located in an urban basin in the Pacific of Mesoamerica. The information on distribution, morphology, and ecology provided here will contribute to a better understanding of the species’ biology and will aid the creation and implementation of management and conservation measures.

Keywords: Agriculture, Central American ichthyofauna, conservation actions, Eastern Panama, endemism, pollution, urban expansion.

Resumen: Existen dos casos de endemismo de cíclidos en el Sur de Centroamérica, i.e., en la Vertiente Pacífico del Este de Panamá. Una de estas especies es *I. tuiyensis*, cuya distribución conocida incluye los ríos de la Provincia de Darién y el Río Bayano. La información sobre distribución y ecología de *I. tuiyensis* es, no obstante, relativamente escasa. En esta contribución reportamos una extensión en el rango de distribución conocida para la especie, así como datos morfológicos y ecológicos adicionales. Se realizaron muestreos ictiológicos utilizando electropesca y atarrayas, en tres ríos del Distrito de Panamá (Ciudad de Panamá: Matasnillo, Juan Díaz y Pacora) entre agosto y septiembre (estación lluviosa) de 2020, febrero y marzo (estación seca) y abril (transición entre estación seca y lluviosa) de 2021. Se recopilaban datos sobre diversidad de peces y parámetros físicos y de calidad del agua en las zonas alta, media y baja de las tres cuencas. Este reporte se enfoca, no obstante, en la única zona en donde se encontraron ejemplares de la especie endémica mencionada (i.e., Pacora). La frecuencia de los censos frente a las amenazas muestra una posible barrera de distribución con posibilidad de extirpación, debido a la fuerte contaminación en los ríos, hacia el Oeste de la Ciudad de Panamá, acompañada de amenazas producto de la expansión urbana y las actividades agrícolas en el Distrito de Panamá. *I. tuiyensis*, el desconocido “Aveinte” o el “Héroe del Istmo”, es una especie catalogada como vulnerable en la Lista Roja de Especies en Peligro de la UICN que habita en una zona que carece de protección efectiva; siendo la única especie de pez endémica ubicada en una cuenca urbana en el Pacífico de Mesoamérica. La información sobre distribución, morfología y ecología aquí provista se espera que contribuya a un mejor conocimiento y entendimiento de la biología de la especie, así como a la creación y promoción de medidas de manejo y conservación.

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