| 1 | Rediscovery and Habitat Assessment of the Endemic Philippine Medaka (Oryzias luzonensis) | | | | | |
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| 2 | in Northern Luzon | | | | | |
| 3 | Faith S. Tadeo ^{1*} , Andie John D. Tadeo ^{1,2} , Marvin M. Aquino ¹ , Shara Mae P. Puh-hay ¹ , Marycris | | | | | |
| 4 | Fiatacag ¹ , Bryan Baldemor ¹ , Christian C. Molina ³ , Nico Jose Leander ⁴ , Rodolph Charles N. | | | | | |
| 5 | Rosel ¹ , Brian Wade Jamandre ⁵ | | | | | |
| 6 | ¹ College of Fisheries, Don Mariano Marcos Memorial State University, La Union, Philippines | | | | | |
| 7 | ² Fisheries Training Research Institute, Don Mariano Marcos Memorial State University, L | | | | | |
| 8 | Union, Philippines | | | | | |
| 9 | ³ Ilocos Sur Polytechnic College, Ilocos Sur, Philippines | | | | | |
| 10 | ⁴ Bureau of Fisheries and Aquatic Resources 3, Pampanga, Philippines | | | | | |
| 11 | ⁵ National Fish and Wildlife Foundation, Washington, D.C., USA | | | | | |
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| 13 | *Corresponding Author: ftadeo@dmmmsu.edu.ph | | | | | |
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1 Abstract

| The endemic Philippine medaka Oryzias luzonensis, confined to the Laoag (Padsan) | | | | | |
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| watershed in Ilocos Norte, Northern Luzon, had not been recorded in the wild for over 40 years, | | | | | |
| prompting its classification as Endangered by the IUCN in 2021. Field surveys conducted from | | | | | |
| January to May 2024 across six sites using traditional push nets successfully rediscovered the | | | | | |
| species in three Solsona municipality sites, yielding 54 specimens (32 at Maananteng, 12 at | | | | | |
| Aguitap, 10 at San Juan; mean length 2.52 ± 0.21 cm). No individuals were found in three Dingras | | | | | |
| sites, which exhibited moderate to severe habitat degradation from agricultural intensification, | | | | | |
| sand and gravel mining, and channelization. Habitat survey showed significant environmental | | | | | |
| differences among sites for temperature and turbidity (F _{5,12} = 28.7, p < 0.001), with O. luzonensis | | | | | |
| presence associated with cooler, clearer waters. Only juvenile and subadult specimens were | | | | | |
| captured, suggesting potential recruitment limitations. This rediscovery confirms species | | | | | |
| persistence but highlights extreme vulnerability, necessitating immediate habitat protection and | | | | | |
| invasive species control to prevent extinction of the Philippines' sole medaka species. | | | | | |
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Keywords: Endemic fish, conservation, freshwater ecosystems, Philippines, habitat degradation,

Oryzias

Introduction

Medakas or ricefishes of the genus *Oryzias* are small, oviparous freshwater fishes in the family Adrianichthyidae, distributed throughout East, South, and Southeast Asia (Parenti 2008; Magtoon and Termvidchakorn 2009). These fishes serve as valuable model organisms in scientific research due to their ease of maintenance, short life cycles, transparent embryos, and relatively compact genome (Wittbrodt et al. 2002; Furutani-Seiki and Wittbrodt 2004; Takehana et al. 2005; Naruse et al. 2011; Hilgers and Schwarzer 2019). Beyond laboratory applications, medakas are increasingly utilized in environmental monitoring and toxicological assessments (Padilla et al. 2009).

The endemic Philippine species *Oryzias luzonensis*, locally known as "coscosleng," represents the sole medaka species in the Philippines and is confined to the Laoag or Padsan watershed in Ilocos Norte, Northern Luzon. As the only member of its genus in the archipelago, this species represents a unique component of Philippine freshwater biodiversity with unique evolutionary significance. Originally described as *Aplocheilus luzonensis* by Herre and Ablan (1934) based on specimens collected from rice paddies and creeks in Solsona, the species was later reclassified under *Oryzias*. Historical records extended its range to the municipalities of Bacarra, Batac, Dingras, and Laoag (Blanco 1947). The last documented field collection occurred in 1982 in Maananteng, Solsona, where individuals were used to establish a captive population maintained in Japan (Formacion and Uwa 1985; Kinoshita et al. 2009).

For over four decades, no verified records of wild *O. luzonensis* populations existed, prompting conservation concerns. In 2021, the IUCN Red List assessed the species as Endangered due to threats including agricultural pollution, habitat modification, land use change, and invasive alien species (Gonzalez et al. 2021). The assessment emphasized the urgent need to verify the

1 species' continued existence and reassess population status in the wild. This study addresses these

2 critical knowledge gaps through comprehensive field surveys across the Laoag or Padsan River

watershed. Our objectives were to: (1) verify the current distribution of O. luzonensis, (2) assess

habitat conditions and document other fish communities, and (3) develop conservation

recommendations.

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Materials and Methods

Study Area and Site Selection

9 Field surveys were conducted from January to May 2024 within the Laoag or Padsan River

watershed in Ilocos Norte, Northern Luzon, Philippines (Fig. 1). Based on historical records of O.

luzonensis, six sampling sites were selected: three in Solsona municipality (Sites 1–3: Maananteng,

12 Aguitap, San Juan) and three in Dingras (Sites 4–6).

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Fish Collection and Identification

Sampling employed traditional push nets, locally known as "Kapot," a common fishing gear used by local communities to capture small fish in shallow streams (Fig. 2). The push net functions as a trap net strategically positioned in areas where schools of Medakas and similar small fish (such as guppies and mosquitofish) are abundant. The sampling technique was modified from traditional fishing methods: fishermen scared and lured target fish into the Kapot by waving a stick with plastic strips attached to the end (similar to a fly swatter or "pambugaw ng langaw" in the local language). Sampling was conducted along transects in accessible stream segments during

1 both dry and early wet seasons, when water levels allowed optimal access to target habitats.

2 Captured specimens were temporarily held in aerated containers, manually sorted, and

photographed. Morphological identification was confirmed through comparison to identification

key (Herre and Ablan 1934) expert consultation, with specimens preserved in 70% ethanol for

voucher purposes. All other fish species encountered were recorded and identified to species level

when possible.

Habitat and Water Quality Assessment

In situ measurements of water temperature (°C), pH, and turbidity (NTU) were taken using calibrated portable meters (YSI Pro1020, USA). Physical habitat assessments included water depth, flow velocity, canopy cover, and dominant substrate classification. Visual inspections documented anthropogenic impacts including natural or man-made erosion, agricultural runoff, and waste accumulation. To assess differences in water quality parameters within sites, statistical comparisons were conducted using Analysis of Variance (ANOVA). ANOVA was applied to rigorously test spatial variability and identify statistically significant differences, with post-hoc Tukey HSD tests performed for pairwise group comparisons when appropriate. All statistical analyses were conducted using the R programming language (R Core Team 2024).

Results

Rediscovery of Oryzias luzonensis

Live *O. luzonensis* specimens were confirmed exclusively in Solsona sites, representing the first verified wild collection in over 40 years (Fig. 3-4). A total of 54 individuals were captured: 32 at Site 1 (Maananteng), 12 at Site 2 (Aguitap), and 10 at Site 3 (San Juan) (Table 1). All

specimens were juveniles or subadults (mean standard length: 2.52 ± 0.21 cm), with no mature
 adults observed. The species was completely absent from all Dingras sites (Sites 4–6).

Collected specimens exhibited typical *O. luzonensis* characteristics, consistent with the original species description and historical collections from the same locality. The 54 specimens displayed small, slender bodies with an average standard length of 2.52 ± 0.21 cm (Fig. 4). Bodies were laterally compressed and semi-transparent, exhibiting pale yellow to grayish ground coloration with a silvery sheen along the sides. A distinct dusky stripe extended from the upper angle of the opercle to the caudal base in all specimens. Meristic counts revealed 5-7 soft rays in the dorsal fin and 15-18 soft rays in the anal fin, consistent with the species' diagnostic characters. Notably, all specimens showed characteristic melanophores on the dorsal to caudal region, a diagnostic feature relatively unique to *O. luzonensis* among *Oryzias* species (Fig. 3-4) (L. Parenti and K. Naruse, personal communication). These morphological features were identical to those described by Herre and Ablan (1934) from specimens collected at the same locality in December 1933, confirming species identity and morphological consistency across nearly nine decades.

In total, four other fish species and one invertebrate were documented across all sites: *Gambusia affinis*, *Poecilia reticulata*, *Oreochromis niloticus*, and freshwater shrimp (*Macrobrachium* spp.). All sites were dominated by invasive taxa, particularly *G. affinis* and *P. reticulata*, especially in Dingras sites where only non-natives were collected.

Habitat Conditions and Water Quality

Qualitative assessments revealed marked differences between Solsona and Dingras habitats. Solsona streams were characterized by shallow depths (15–36 cm), slow flow, and partial canopy cover from native vegetation, particularly taro (*Colocasia esculenta*) and grass (Fig. 5).

Conversely, Dingras sites exhibited moderate to severe degradation: channelized streams, exposed eroding banks, minimal vegetation, and substrates dominated by fine sediments. Agricultural impacts were evident through irrigation modifications, and livestock disturbance. Additionally, extensive sand, gravel, and stone mining operations for construction materials were observed throughout the Dingras watershed, contributing to habitat destruction, increased sedimentation, and altered stream morphology. These combined anthropogenic pressures likely rendered habitats unsuitable for *O. luzonensis*.

Water quality measurements revealed differences between Solsona and Dingras sites, though these differences were not statistically significant (Table 1). Solsona sites exhibited slightly cooler temperatures (27.67 \pm 1.53°C), similar pH values (6.90 \pm 0.10), and lower turbidity (0.67 \pm 0.58 NTU). Dingras sites were characterized by slightly elevated temperatures (29.50 \pm 0.50°C), comparable pH (6.83 \pm 0.29), and higher turbidity (2.67 \pm 2.08 NTU). Notably, all sites maintained relatively clear water conditions with low turbidity values overall.

One-way ANOVA revealed significant differences among sites for temperature (p < 0.001) and turbidity (p < 0.001), but not for pH (p = 0.13). Post-hoc Tukey tests showed that Site 1 (26.0°C) had significantly lower temperature than Sites 4-6 (29.0-30.0°C), while Sites 2-3 were intermediate. For turbidity, Site 5 (5.0 NTU) was significantly higher than Sites 1-3 (0.0-1.0 NTU).

Discussion

The rediscovery of *Oryzias luzonensis* in Solsona after more than four decades confirms the species' persistence in the wild, albeit within severely restricted and degraded habitats. Critically, our findings reveal a dramatic range contraction from the species' historical distribution.

While historical records documented *O. luzonensis* across multiple municipalities within the Laoag (Padsan) watershed including Bacarra, Batac, Dingras, Laoag, and Solsona (Blanco 1947), current ground-truthing confirms the species' presence only within Solsona municipality. The complete absence from Dingras, despite historical occurrence and intensive sampling efforts, suggests local extinction and represents a significant reduction in the species' geographic range. This range contraction pattern, from a watershed-wide distribution to a single municipality, indicates severe population decline and habitat loss that has compressed the species into its last remaining refugia. The failure to detect *O. luzonensis* beyond this small area in Solsona underscores the species' precarious conservation status and suggests that previous distribution records may now represent extirpated populations rather than current habitat.

Our findings demonstrate subtle differences in some environmental parameters between sites with and without *O. luzonensis*. The slightly higher temperatures and turbidity in Dingras sites, combined with the more pronounced habitat degradation observed in the field, may contribute to the species' absence. The association of this endemic species with the cooler, clearer waters of Solsona aligns with habitat requirements of other *Oryzias* species (Kinoshita et al. 2009). The preference for shallow, slow-flowing waters with riparian vegetation, particularly *Colocasia esculenta*, underscores the importance of maintaining vegetated buffer zones that stabilize water quality and provide essential habitat structure. The presence of invasive species in degraded Dingras sites may contribute to decline of native fish. *Gambusia affinis* and *Oreochromis* species are aggressive competitors known to displace native fauna through predation, resource competition, and habitat modification (Pyšek et al. 2020).

The capture of only juvenile and subadult specimens raises concerns about population structure and reproductive success. This pattern could indicate: (1) seasonal breeding with adults

- 1 in refugia, (2) recent recruitment failure, or (3) population bottlenecks affecting size structure. The
- 2 small population size observed (54 individuals across three sites) may suggests extreme
- 3 vulnerability to stochastic events.
- 4 Habitat fragmentation and degradation observed in Dingras represent primary threats to *O*.
- 5 luzonensis persistence. Mining activities for construction materials have particularly severe
- 6 impacts, causing direct habitat destruction, increased sediment loads, altered hydrology, and
- 7 disruption of natural substrate composition essential for medaka survival. These findings align
- 8 with global patterns of freshwater fish endangerment linked to anthropogenic habitat modification
- 9 and extractive industries (Dudgeon et al. 2006).

Conservation Implications and Recommendations

The restricted distribution and small population size of O. luzonensis necessitate immediate

conservation intervention. Priority actions should include: (1) protection of remaining Solsona

habitats through local ordinances, (2) riparian restoration with native vegetation, (3) invasive

species control programs, (4) water quality monitoring, (5) establishment of buffer zones around

critical habitats, and (6) regulation of mining activities within the watershed to prevent further

habitat destruction and sedimentation.

Long-term conservation success requires community engagement and sustainable

livelihood alternatives that reduce pressure on freshwater ecosystems. The species' potential value

for aquaculture and scientific research could provide economic incentives for conservation while

supporting local communities.

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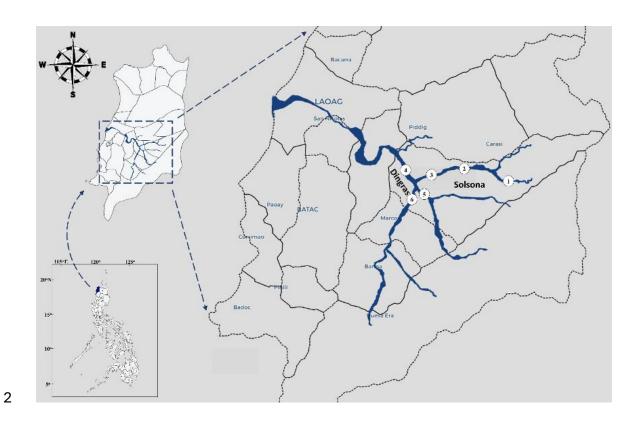
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2 Table 1. Water quality parameters and species occurrence across study sites.

| Site | Temp. (°C) | рН | Turbidity (NTU) | Substrate | O. luzonensis |
|------|----------------|-----|-----------------|-------------|----------------|
| 1 | 26.0 ± 0.2 | 6.9 | 0.0 | Gravel/Silt | Present (n=32) |
| 2 | 28 ± 0.3 | 7.0 | 1.0 | Gravel/Sand | Present (n=12) |
| 3 | 29.0 ± 0.2 | 6.8 | 1.0 | Gravel/Silt | Present (n=10) |
| 4 | 30.0 ± 0.2 | 7.0 | 2.0 | Sand/Silt | Absent |
| 5 | 29.0 ± 0.3 | 6.6 | 5.0 | Gravel/Clay | Absent |
| 6 | 29.5 ± 0.2 | 7.0 | 1.0 | Gravel/Silt | Absent |

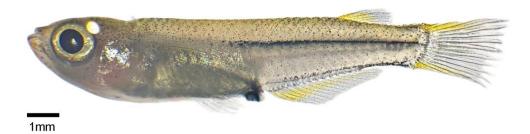


3 Fig. 1. Sampling sites of Sampling sites of O. luzonensis in Solsona (sites 1-3) and Dingras (sites

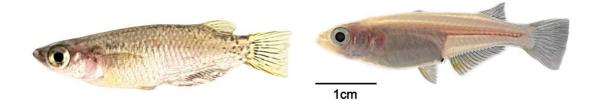
4 4-6), Ilocos Norte.



3 Fig. 2. Collection of *O. luzonensis* using a push net (*Kapot*).



- 3 Fig. 3. O. luzonensis juvenile with its distinct melanophores collected in sampling sites in
- 4 Solsona, Ilocos Norte.



- 9 Fig. 4. Adult O. luzonensis (a. dead and b. live samples) collected in sampling sites in Solsona,
- 10 Ilocos Norte.



3 Fig. 5. Sampling sites where *O. luzonensis* was collected: Site 1 (a,b) and Site 2 (c,d)