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First Record of *Silurus aristotelis* Garman, 1890 in Schinias Olympic Rowing and Canoeing Center (Schinias National Park, Attica, Greece)

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Abstract: This study reports the presence of the endangered endemic Aristotle's catfish (*Silurus aristotelis* Garman, 1890) outside of its natural distribution and within the Schinias National Park (Attica Region, Greece), based on morphometric and molecular analyses. The capture of three adult specimens within a small surveyed area suggests that the species is widespread within the National Park and possibly has established a viable population. Given its ecological traits – high environmental adaptability, top predator status, habitat modification tendencies, and territorial behavior – *S. aristotelis* is expected to exert significant competitive pressure on the park's endemic and native species, particularly the Marathon minnow (*Pelasgus marathonicus* Vinciguerra, 1921) and the European eel (*Anguilla anguilla* L.). Additionally, its invasion risk is amplified under projected climate change scenarios, raising concerns about its long-term ecological impact. The detection of *S. aristotelis* within this protected area highlights the urgent need for immediate management actions, including eradication or population control measures, particularly since angling is strictly prohibited in this area.

Key words: Aristotle's catfish, translocation, molecular identification, invasiveness risk, Attica

Introduction

The translocation of native freshwater fish species beyond their historical ranges, yet within the same country, represents a complex and often understudied ecological phenomenon with significant implications for aquatic ecosystems (Koutsikos et al. 2019, Vitule et al. 2019, Tarkan et al. 2024). While some translocations are conducted under scientific supervision for conservation purposes (Kalogianni

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et al. 2024) or stock enhancement, primarily in recreational fisheries (Lintermans et al. 2015), the majority result from informal, poorly regulated, or even unintentional stocking practices (Economidis et al. 2000, Vardakas et al. 2022). These actions, often undertaken without rigorous ecological assessment, can negatively affect local biodiversity, compromise ecosystem functioning, and lead to unpredicted ecological consequences. Freshwater fish translocated beyond their native ranges can disrupt local ecosystems by competing with the local fish fauna for food and habitat, preying on native fauna, altering nutrient cycling, and serving as vectors for diseases or parasites, potentially leading to declines in native populations (Copp et al. 2005, Savini et al. 2010, Ribeiro & Leunda 2012). Moreover, translocations can have profound effects on genetic diversity, often surpassing the impacts of non-native species introductions. This is primarily due to the increased likelihood of introgressive hybridization between translocated populations or closely related species, which can lead to the erosion of unique genetic lineages and the loss of local adaptations (Apostolidis et al. 2008). Furthermore, potential adverse impacts on the gene pool and the overall genetic integrity of the donor population(s) are an overlooked aspect particularly in poorly designed translocations of endangered species. As freshwater species are among the most threatened globally (Sayer et al. 2025), understanding the impacts of these translocations is critical for informing sustainable management practices and mitigating unintended consequences.

The translocation of fish species beyond their native ranges remains a common pattern in Greece, often driven by economic interests and recreational fishing. Recurring stocking efforts by local fisheries and forest authorities, professional fishermen associations, and recreational anglers have contributed to the widespread introduction of species into new localities (Koutsikos et al. 2019, Vardakas et al., 2022). In many cases, these stockings are undocumented, involve domesticated or cultured varieties of foreign origin, and, more concerningly, take place without scientific oversight or clear justification for population enhancement. Specifically, over the past decade, more than 29 native fish species have been translocated to various lotic and lentic ecosystems of Greece (Koutsikos et al. 2019, Vardakas et al. 2022). The most widely translocated species in lentic ecosystems of Greece are those of high commercial value, including common carp (*Cyprinus carpio* L.), tench (*Tinca tinca* L.), European perch (*Perca fluviatilis* L.), Wels catfish (*Silurus glanis* L.) and Aristotle's catfish (*Silurus aristotelis* Garman, 1890) (Vardakas et al. 2022).

Silurus aristotelis, is a species with a natural distribution restricted to the Ionian ichthyogeographic ecoregion in western Greece (Barbieri et al. 2015), and primarily found in lentic ecosystems associated with the lower Acheloos River drainage, including the natural Lakes Trichonis, Lysimachia, Ozeros, and Amvrakia (Economou et al. 2007, Barbieri et al. 2015, Vardakas et al. 2022). Despite being classified as Least Concern (LC) in the Greek Red Book, *S. aristotelis* is listed as Endangered (EN B1ab(iii)+2ab(iii)) under the IUCN Red List and Directive 92/43/EEC due to its restricted distribution and habitat degradation (Ford 2024a).

Beyond its native range, *S. aristotelis* has been translocated to various other lentic ecosystems, such as Lake Volvi in the Macedonia–Thessaly ichthyogeographic ecoregion, where it was translocated in 1986 but became extirpated by the 1990s (Economidis 1991, Economidis et al. 2000, Ford 2024a). While this translocation, reinforced the local fisheries in Lake Volvi, it possibly also led to the extinction through competition of the native population of *S. glanis* (Economidis et al. 2000). Additional translocations include Lake Pamvotida, where the transfer occurred during the 1960s or earlier (Economidis et al. 2000), and Kalodiki marsh, which may have been a primary translocation from its native habitat over 25 years ago or a secondary translocation from Lake Pamvotida (Konstantinidis et al. 2018). Other translocations include the lower Boeotian Kifissos River, encompassing Lakes Yliki and Paralimni, where they likely occurred in the early 2000s, possibly as accidental consequences of carp stockings (Koutsikos et al. 2012, Zogaris 2013). Non-native populations have also been recorded outside Greece, with a small artificial lake in northwestern Bulgaria stocked with juveniles from southwestern Bulgaria (Pehlivanov & Atanasov 2007), reflecting successive translocations originating from Greece.

This study documents the first recorded instance of *S. aristotelis* in Schinias National Park, (Attica, Greece), providing insights into the ecological dynamics and potential risks associated with intra – country translocations of freshwater fish in an era of increasing anthropogenic pressure on aquatic environments.

Materials and Methods

Study area and sampling methods

During ad hoc surveys within the Schinias Olympic Rowing and Canoeing Center's (SORCC), one site was sampled on July 24, 2024, using a backpack electrofishing device (IG200/2, 250W, Hans

Fig. 1. Location of the Schinias Olympic Rowing and Canoeing Center (SORCC) within the Schinias-Marathon National Park (area within the red boundaries). The red arrow indicates the specific area where *Silurus aristotelis* was discovered. (Map data: Google© 2025).

Fig. 2. The warming–up lake of the Schinias Olympic Rowing and Canoeing Center (SORCC), where specimens of *Silurus aristotelis* were recorded.

Grassl GmbH) in the SORCC warming-up lake (38.144439°N, 24.014835°E, Fig. 1, Fig. 2). A modified fish sampling method was followed (IMBRIW 2013), by point sampling along the shoreline. Physicochemical parameters (i.e., water temperature (°C), dissolved oxygen (mg/l), conductivity (mS/cm) and salinity (psu)) were measured with a HACH HQ40D polymeter, while habitat characteristics (depth (m), width (m), velocity (m/s) and substrate (%)) were recorded. Three catfish specimens were collected, photographed, and one was preserved in formalin for further analysis, while a fin clip was obtained for DNA analysis.

The SORCC was constructed for the 2004 Athens Olympics (Fig. 1). The facility consists of two interconnected semi-natural lakes, one for racing (2,240 m length x 160 m width), one for warming up (1,350 m. length x 135 m width), and an artificial island with a total of 0.57 km² of water surface and 0.21 km² of constructed environments. The water supply to the rowing arena and to the Schinias wetland is derived from the Makaria spring. The area belongs to the Natura 2000 European Network (GR3000016-Schinias Wetland and GR 3000003-Schinias-Marathon National Park; BISE 2025a,b).

The SORCC is situated within the Schinias–Marathon National Park, historically known as the "Marathon Marsh,", or the "Great Marsh", the largest wetland in the Attika Region. It represents a coastal biotope located within the Marathon plain, approximately 45 km to the northeast of the metropolitan city of Athens. Established as a National Park in 2003, in a land area of 9 km² it showcases an array of natural habitats: freshwater springs, coastal wetlands, coastal sand dunes covered by pine forest and a rocky substrate covered by Mediterranean maquis. The ecological significance of Schinias is noteworthy, as it harbors over 320 species of flora and a considerable diversity of fauna, which includes numerous threatened resident and migratory avian species, freshwater ichthyofauna, as well as amphibians and reptiles (Hadjibiros 2010). Based on the most recent inventory of lentic ecosystems (Vardakas et al. 2022), four fish species have been documented in the Schinias–Marathon National Park, comprising two native and two non – native species. The native species include the European eel (*Anguilla anguilla* L.), which is classified as Critically Endangered by the IUCN, and the Marathon minnow (*Pelasgus marathonicus* Vinciguerra, 1921), listed as Near Threatened (Ford 2024b). The two non-native species recorded in the park are the eastern mosquitofish (*Gambusia holbrooki* Girard, 1859) and the goldfish (*Carassius auratus* L.).

Morphometric analyses

Morphometric study was performed following the key to species of *Silurus* in Europe and the morphological/morphometric/meristic characters described by Kottelat & Freyhof (2007).

DNA extraction, PCR amplification and DNA sequencing

For the DNA extraction, a modified CTAB protocol (Doyle & Doyle 1987) was used. To assess the quality and quantity of the extracted DNA, semi-quantification was carried out using 1.5% agarose gel electrophoresis. Finally, 1 μl of the extracted DNA was used in PCR to amplify the COI gene (Cytochrome c oxidase subunit I), using the universal primers of Folmer et al. (1994). The PCR protocol began with an initial DNA denaturation at 95°C for 2 min, followed by 37 cycles of: 94°C for 30 sec, 48°C for 30 sec, and 72°C for 1 min, concluding with a final extension at 72°C for 10 min. PCR product purification was conducted using ExoSAP–IT™ PCR Product Cleanup (Thermo Scientific, Santa Clara, CA 95051, USA) yielding a final product of approximately 700 base pairs. Purified PCR products were sent for bi-directional sequencing to CEMIA A.S (Larissa, Greece). Chromatograms were visually examined with FinchTV 1.3.1 (Geospiza, Inc.; Seattle, WA, USA; http://www.geospiza.com). The sequences, after visual inspection, were subjected to a MEGABLAST search (National Center for Biotechnology Information Basic Local Alignment Search Tool, NCBI BLAST; http://www.ncbi.nlm.nih.gov/).

Invasiveness risk assessment

To assess the invasiveness risk of *S. aristotelis* in the risk assessment (RA) area (i.e., the SORCC), a risk screening was conducted by a single assessor (CP) using the AS-ISK tool (AS-ISK: www.cefas.co.uk/nns/tools/), which is described in detail in Copp et al. (2021). Upon completion of the screening, the species received both a Basic Risk Assessment (BRA) score and a BRA+ Climate Change Assessment (CCA) composite score (ranging from −20 to 68 and from −32 to 80, respectively). Threshold values for freshwater fish species to distinguish medium – and high–risk levels (of becoming invasive) were applied for BRA (i.e. 25.5) and BRA+CCA (i.e. 28.75) scores, following Tarkan et al. (2021).

Results

Environmental setting and habitat attributes

The study site was located at an altitude of 2 meters above sea level and 1,000 meters inland from

the coastline, with a flat topography (0% slope). Water quality parameters indicated that the water was slightly brackish with a conductivity of 6.09 mS/cm, a pH of 8.02, and a salinity of 3.12 psu. The water temperature averaged 27.5°C. Dissolved oxygen levels were measured at 8.46 mg/l, suggesting adequate oxygen availability to support aquatic life. The water exhibited moderate turbidity with suspended particulate matter, while some foam was observed, possibly resulting from our sampling efforts, which may have mixed the bottom substrate. The overall fished area was 140 m^2. The maximum depth was recorded at 2.0 meters, with a mean depth of 1.2 meters. Water velocity was low (<0.1 m/sec), while the substrate of the study area was dominated, based on a modified Wentworth scale (Cummins 1962), by rocky materials, with 40% boulders (>256 mm) and 40% cobbles (64-256 mm), artificially placed in the site area to stabilize the banks. Silt constituted 20% of the substrate composition. The site also exhibited dense riparian vegetation, primarily reeds, and abundant bottom vegetation, specifically *Ruppia* sp.

Fish fauna, morphometrics and molecular verification

Three adult specimens of *S. aristotelis* were caught near the shoreline, at a depth of approximately 1.5 m. Along with *S. aristotelis*, the following species were also recorded: *A. anguilla*, *G. holbrooki* and the Marathon minnow *P. marathonicus*, the latter of which was found in the "Makaria" spring that supplies water to the SORCC (about 1,800 m from the main sampling area). In addition, five adult individuals belonging to the *Carassius* genus were collected. Since distinguishing *C. auratus* and *Carassius gibelio* (Bloch, 1782) in the field is challenging, we tentatively based our identification on their color and previous published work (Vardakas et al. 2022), with *C. auratus* identified by its golden-brown bronze color, as opposed to the silvery-brown color of *C. gibelio*, a key distinguishing feature of *Carassius* species in Europe (Kottelat & Freyhof 2007).

Silurus aristotelis was distinguished from the other native silurid species (i.e., *S. glanis*) by the following unique characters: one pair of mental barbels (Fig. 3) and anal fin with 66-75½ rays (Kottelat & Freyhof 2007) (Table 1).

Table 1. Morphological/morphometric/meristic measurements of *Silurus aristotelis* specimen caught in SORCC.

Total weight (TW, g):	125.7
Total length (TL, mm):	266
Standard length (SL, mm):	238
Head length (HL, mm):	56.75
Body depth (BD, mm):	46.75
Caudal peduncle length (CPL, mm):	11.40
Total anal rays:	73½
Mental barbels:	1 pair

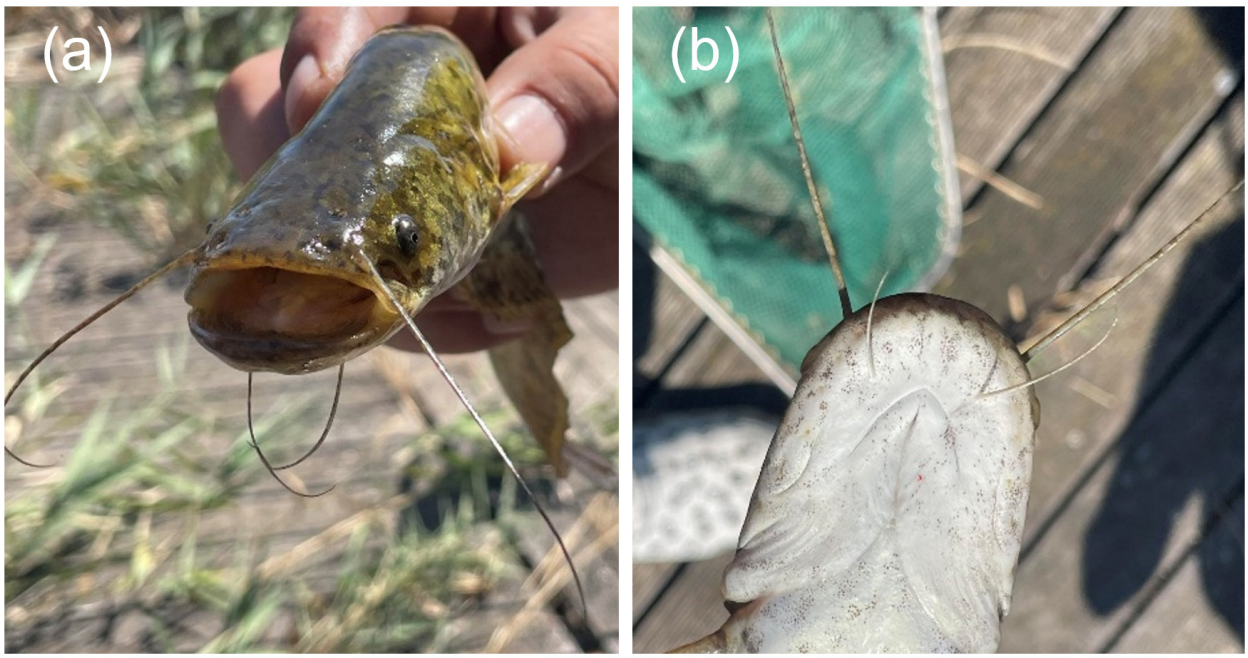

Fig. 3. Basic characteristics of the *Silurus aristotelis* specimen; (a) front (head) view and (b) jawbone view with the mental barbels.

The specimen was visually identified as *S. aristotelis* and verified by molecular methods. BLAST results from Genbank exhibited 100% similarity with the accession number OX327782.1, confirming that the analysed sample herein was *S. aristotelis*. The sequences were translated into amino acids using the Sequence Manipulation Suite platform, following the Vertebrate Mitochondrial Code (translation table 2) from the second codon position on the direct strand. No stop codons were observed within the reading frame. The sequence identified in this study has been assigned the GenBank accession number PV012548.

Invasiveness risk scoring

The application of the AS–ISK tool for *S. aristotelis* resulted in BRA score of 25 and BRA+CCA score of 33, suggesting a species with marginal medium risk of becoming invasive in the RA area based on the BRA score and with high risk based on the combined BRA+CCA score.

Discussion

This study reports on the first occurrence of *S. aristotelis* within the Schinias National Park in the Attica region. The presence of this species in the region shows the potential risks associated with intra–country translocations of native/endemic species while it also highlights the vulnerabilities of artificial and semi–artificial aquatic environments near urban areas to the introduction of translocated species. Artificial and semi–artificial lentic systems, such as the one present in SORCC, are particularly vulnerable to the establishment of non–native species (Vardakas et al. 2022). The current study verifies this trend, as evidenced by the presence of two top invasive species and one translocated in the area.

Intentional and unintentional translocations of freshwater species into freshwater ecosystems near major urban centres, such as Athens, Thessaloniki, Ioannina and Kastoria, has been a common practice in Greece (Leonardos et al. 2008, Koutsikos et al. 2021a, Petriki & Bobori 2024). The Attica region has experienced the translocation of several native fish species, including the following *Barbus sperchiensis* Stephanidis, 1950, *C. carpio*, *Economidichthys pygmaeus* (Holly, 1929), *Ladigesocypris ghigii* (Gianferrari, 1927), *Luciobarbus graecus* (Steindachner 1895), *Leucos ylikiensis* (Economidis, 1991), *Scardinius graecus* Stephanidis, 1937 and *Squalius vardarensis* Karaman, 1928 (Barbieri et al. 2015, Vardakas et al. 2022, Koutsikos et al. 2023, Zogaris et al. 2023). Similar to the case of *S. aristotelis*, these introductions contribute to ecosystem alterations and may lead to the displacement of endemic species, such as the Marathon minnow. This growing trend of translocation raises significant concerns about the ecological integrity of freshwater systems in the region, particularly in the face of increasing anthropogenic pressures.

The pathway and vector responsible, as well as the exact date for the translocation of *S. aristotelis* within Schinias National Park remain unclear. However, given (a) the proximity of SORCC to other lentic ecosystems where recreational fishing is actively practiced and where the species is present (e.g., Lakes Yliki and Paralimni), and (b) the documented translocation of three co–occurring species (*L. graecus*, *L. ylikiensis*, and *S. graecus*) from these lakes to Marathon Lake − situated in near proximity with SORCC − it is highly plausible that *S. aristotelis* was introduced as a secondary introduction, likely facilitated by anglers.

Silurus aristotelis has established successful populations in various lentic and lotic ecosystems across Greece (Koutsikos et al. 2019, Vardakas et al. 2022). It inhabits slow–flowing lowland river reaches, often in close proximity with wetlands and lakes, favoring well–vegetated environments with muddy or silty substrates (Koutsikos et al. 2021b). This species is well adapted to turbid, nutrient–rich waters, particularly in lakes with dense aquatic vegetation that provide both shelter and suitable breeding sites (Barbieri et al. 2015). It is a nocturnal predator with its diet primarily consisting of fish, both live and dead, while it can also opportunistically feed on crustaceans, gastropods, aquatic insects, amphibians, and small aquatic snakes (Iliadou & Ondrias 1986) due to the highly developed sensory barbels, which can efficiently detect prey in low–visibility conditions. Their distinctive three pairs of barbels reduce to two as they grow (Economou et al. 1993). Juveniles initially develop three pairs of barbels, but one pair is lost at approximately 95 mm total length (TL). The species reaches sexual maturity at 2–3 years of age. The breeding season extends from April to July, during which males establish and defend small territories at spawning sites. Nest-building occurs in shallow depressions or on cleared substrates at depths of 2–3 meters, utilizing plant materials and woody debris (Kottelat & Freyhof 2007). Spawning occurs in pairs, with the male guarding the nest until the larvae emerge. The conditions at SORCC closely resemble those found in the species' type locality, providing a favorable environment for its establishment. This is supported by the presence of suitable spawning grounds, abundant prey resources, and

optimal environmental factors. The stable hydrological environment, depth, and water volume of SORCC create an ideal habitat for reproduction and fry development for the species. These factors, combined with the species' capacity for environmental modification, make SORCC an optimal location for the continued growth of *S. aristotelis*.

As a top carnivorous predator, *S. aristotelis* plays a crucial role in freshwater food webs. Similar to eels, it exploits a broad spectrum of food sources, with its feeding behavior influencing all trophic levels within an ecosystem. Additionally, *S. aristotelis* exhibits territorial tendencies and actively modifies its habitat by creating shallow depressions or cleaning spawning substrates (Kottelat & Freyhof 2007), leading to structural microhabitat alterations. These traits contribute to its high adaptability, allowing it to dominate its environment and potentially displace native fauna, including fish species that rely on similar habitats. Given these ecological characteristics, the invasiveness risk of *S. aristotelis* was assessed using the Fish Invasiveness Screening Kit (FISK) on country – wide scale at a moderately high level, with a mean score of 18.5 and a calibration threshold of 15.25 between medium – and high-–risk species (Perdikaris et al. 2016). When the newer tool (i.e., AS–ISK) was applied to the SORCC as risk assessment (RA) area, the risk score increased to 25 (BRA), and under climate change scenarios (BRA+CCA), it rose further to 33. These findings suggest that while *S. aristotelis* currently poses a medium risk of becoming invasive, this risk escalates significantly under changing environmental conditions. If favourable conditions persist, *S. aristotelis* could thrive, exerting substantial ecological pressure on the endemic species found in the Shinias National Park and potentially driving shifts in species composition and food web dynamics. The spatial overlap between *S. aristotelis* and the endemic/native species, the Marathon minnow and eel, require further investigation, as they could lead to significant ecological shifts in the local food web and threaten the persistence of these threatened species (Koutsikos et al. 2021a).

The introduction of *S. aristotelis* into Schinias National Park highlights the complex challenges posed by intra–country translocations of native species in freshwater ecosystems. While the species may offer certain ecological functions, such as predator control, its potential to disrupt local biodiversity, particularly through competition with, and predation on native species, warrants careful consideration. The findings highlight the need for continued monitoring and the development of effective management strategies to mitigate the risks posed by invasive species in freshwater environments, particularly in artificially created aquatic systems such as SORCC. Within this context, eradication/population control in a relatively small and confined waterbody could be an achievable target and therefore such measures are especially urged for this species (e.g., De Santis et al. 2024 for *S. glanis*) despite its endangered conservation status.

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