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Silent survivors: how monitoring and genetics reveal the recovery of the Adriatic sturgeon

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ABSTRACT

The Adriatic sturgeon (*Acipenser naccarii*) is a critically endangered species that has been the focus of conservation efforts for the past three decades. Recent monitoring programs, supported by genetic analysis, suggest encouraging recovery signals. Notably, the latest IUCN report identifies *A. naccarii* as the only sturgeon species showing an improved conservation status, moving from "Extinct in the Wild" to "Critically Endangered". This study provides an overview of a monitoring activities conducted over the past ten years, focusing on key findings that support this positive trend, such as evidence of spontaneous reproduction. In this context, genetic studies have been crucial in distinguishing between individuals resulting from past restocking efforts and potentially wild-born specimens. Unexpectedly, a few individuals were unambiguously classified as interspecific hybrids, raising concerns about the potential impact of allochthonous sturgeons on the genetic integrity of the Adriatic sturgeon. These findings underscore the importance of long-term monitoring and genetic assessments in conservation strategies for *A. naccarii*.

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Hybrid identification; introgression; alien species; population monitoring; genetic tagging

Introduction

Sturgeons are an ancient lineage of fish that have survived dramatic environmental changes but now face critical threats due to habitat loss, overfishing and pollution. Among them, the Adriatic sturgeon (*Acipenser naccarii*) is an endemic species historically distributed across the Po River basin and other major river systems draining into the Adriatic Sea. Once abundant, its populations have suffered severe declines over the past century, leading to its near extinction in the wild by the late 20th century (Rossi et al. 1992). These pressures culminated in the species being listed as "*Critically Endangered and possibly extinct in the wild*" on the IUCN Red List.

Many conservation actions, including habitat restoration, captive breeding and restocking programs, have been implemented since the 1990s to support its recovery. Two independent LIFE projects (LIFE03/nat/it/000113 and LIFED04nat/it/000126) funded by the European Community (2003–2004) and coordinated by the Ticino River Park and the Delta Po River Park, contributed significantly to conservation efforts by releasing thousands of individuals in the Po River basin and surrounding areas and conducting systematic monitoring and recaptures. The collected data have been stored in a geo-referenced database managed by the Delta Po River Park and are available upon request through the Institute Delta Ecologia Applicata (www.istitutodelta.it). Structured monitoring efforts concluded in 2010, leaving a gap in official data that has since been partially filled by informal reports.

Beyond these projects, a major contribution to conservation has been provided by a private aquaculture initiative. In 1977, just before the species was on the brink of disappearing from its natural habitat, around 90 young wild individuals (F0) were transferred from the Po River to the aquaculture facility Azienda Agricola V.I.P. (Orzinuovi, Brescia, Italy). Of these, 50 survived and reached reproductive maturity after approximately a decade. The first successful captive breeding event was recorded in 1988, marking the beginning of a continuous propagation program (Arlati et al. 1988; Giovannini et al. 1991). Over the following 35 years, this initiative generated more than 30 successive F1 breeding groups (Boscari et al. 2014b), allowing the release

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of more than 500,000 juveniles into the wild through multiple, largely uncoordinated, restocking programs (Arlati & Poliakova 2009). While these efforts have maintained a presence of *A. naccarii* in its historical range, the self-sustainability of the population through natural reproduction is far from being achieved. The species' long-term survival still depends on sustained reintroduction efforts and careful genetic management of the reintroduced population.

However, these initiatives appear to have yielded a glimpse of hope, as reflected in the latest IUCN assessment released in 2022, where *A. naccarii* was the only sturgeon species to show an improved conservation status (Congiu et al. 2023). Despite this encouraging trend, the species' long-term viability remains uncertain, requiring continued monitoring and research to ensure its persistence. In this context, genetic analyses are crucial to understanding population dynamics, genetic diversity and the effectiveness of conservation interventions.

Unfortunately, at present, there is no structured nationwide monitoring network capable of systematically collecting and integrating data into a unified database. Consequently, data on the sturgeon presence in the wild largely relies on informal sightings, gathered through a network of voluntary contacts. Microchip reading from recaptured individuals also presents logistical challenges, as personnel equipped with scanners are not always available to reach the site promptly. Nevertheless, in some instances – thanks in particular to the commitment of volunteers from the Italian Federation of Sport Fishing and Underwater Activities (FIPSAS) in Venice – it has been possible to obtain tissue samples, which were subsequently subjected to genetic analysis.

Over the past 20 years, advances in genetic tools have enabled researchers to assess the impact of restocking programs and distinguish between released individuals and their offspring versus potential wild specimens (Boscari et al. 2014b; Barca et al. 2022). This is possible because all animals released in the past 35 years descend from the original F0 broodstock. Genetic compatibility testing with the F0 stock allows us to advance a hypothesis about the origin of captured individuals. Additionally, genetic markers originally developed for species and hybrid identification to monitor illegal caviar trade (Boscari et al. 2014a) are useful to inform conservation programs. These tools, in fact, are essential to assess genetic purity of individuals used in conservation programs and detect the presence of exotic or hybrid sturgeons in our waters.

In this context, the present study provides a comprehensive overview of monitoring activities conducted over the last few years in collaboration with FIPSAS in the Veneto Region. Monitoring was focused on the Province of Venice, particularly in the Livenza River. Local fishermen notified authorities upon sturgeon sightings, enabling microchip scans and biometric data collection. If no microchip was found and the specimen exceeded a certain size threshold, one was implanted and tissue samples were collected whenever possible for genetic analyses to determine the origin of these individuals. These analyses were conducted to assess whether the individuals originated from controlled breeding programs – possibly released without a microchip – or whether they could instead be of wild origin. This information is essential to evaluate the species' natural reproductive success and to detect the potential presence of remnant wild breeders in the population.

By integrating these data, we aim to evaluate the success of conservation strategies, highlight key findings supporting the recovery of *A. naccarii*, and identify future priorities for its sustainable management. Ultimately, this study contributes to a deeper understanding of the current status of the species in the wild and underscores the critical role of integrated genetic and ecological monitoring in guiding effective long-term conservation efforts. Moreover, it is intended as a starting point to promote a more coordinated national monitoring framework and the establishment of a centralized and standardized database of sturgeon recaptures, in line with the objectives of the recently approved LIFE RESTORE project (101216004 - LIFE24-NAT-IT-LIFE-RESTORE).

It is important to clarify that this study does not aim to provide a comprehensive assessment of the conservation status of *A. naccarii* across its entire historical range. Rather, it reports on data collected through a regional, multi-year monitoring program carried out by the Veneto Region with the support of FIPSAS, focusing mainly on the Livenza River. Most of the individuals considered in this study were captured in this river in recent years, although a few additional sightings were recorded in adjacent systems such as the Piave and Tagliamento Rivers, and in the Adriatic Sea. For untagged individuals, tissue samples were collected for genetic analysis whenever possible. The data presented here do not result from a systematically designed national monitoring effort; instead, this work attempts to bring clarity and structure to a set of opportunistically collected observations, integrating genetic analyses where feasible to provide a better understanding of the current situation. The findings discussed in this manuscript should therefore be interpreted as partial but valuable insights into the status of A. naccarii within a specific region, highlighting both positive signals and emerging risks for the species.

Materials and methods

Monitoring area and sampling

The monitoring program conducted by FIPSAS from 2019 to 2024 focused on the Province of Venice, particularly the Livenza River. Collaboration with local fishermen enabled the reporting of sturgeon sightings. Captured individuals were scanned for microchips, and biometric data (length and weight) were collected. If no microchip was detected and the specimen exceeded a certain size threshold (30 cm), one was implanted. Tissue samples for genetic analysis were taken when authorized personnel were available on-site. All fish were released after examination.

Genetic analysis

DNA was extracted from all collected tissue samples using the Qiagen Blood and Tissue Kit following the manufacturer's protocol.

Species identification was conducted using both mitochondrial and nuclear markers. The maternal lineage was determined by sequencing the mitochondrial control region and comparing haplotypes with reference databases. The amplification process was carried out using sturgeon-specific primers (PRO_1F and PHE_1R) following the protocol detailed in Boscari et al. (2014a). Species-specific nuclear markers were amplified to confirm both parental contribution and exclude the presence of interspecific hybrids.

Relatedness analysis was carried out using multiple microsatellite loci (Barca et al. 2022) to identify, when possible, groups of full sibs. For microsatellite analyses, a panel of 15 loci was amplified using two multiplex PCR reactions (M1 and M2), with the Qiagen Multiplex PCR Master Mix. The loci included in the analyses were: Anac_B11, Spl120, AfuG132, Spl163, AfuG113, Anac_c3133 and Anac_c2588 for M1, AoxD161, AfuG41, Anac_c6784, AoxD241, AfuG112, Anac_B7, Anac_c14336 and Anca_c12159 for M2. The thermal cycling conditions were: 15 minutes at 95°C, followed by 35 cycles of 30 seconds at 94°C, 90 seconds at 60°C and 60 seconds at 72°C, with a final extension of 30 minutes at 60°C. Locus Afug113 was initially amplified via a single touchdown PCR and later included in the M1 multiplex. Loci AoxD241, Afug112 and AnacB7 were amplified separately and then integrated into the M2 multiplex. Detailed information on each locus, including primer sequences, expected size ranges, multiplex groupings, thermal profiles and original references, is provided in Barca et al. (2022). PCR products were genotyped by BMR Genomics (Padua, Italy; https://www.bmr-genomics.it/), and allele scoring was performed using GeneMarker software version 1.95 (SoftGenetics LLC, State College, PA, USA).

To visualize the genetic similarity among sampled individuals, we performed a multidimensional scaling (MDS) analysis based on pairwise genetic distances. Given the tetraploid condition of sturgeon, which prevents accurate determination of allele dosage, genetic distances were estimated using presence/absence data of alleles across the 15 microsatellite loci. Sørensen's distance metric was applied to quantify genetic dissimilarity among individuals, under the assumption that more closely related genomes exhibit lower genetic distances (Sørensen 1948). The distance matrix was computed using the R package "ade4" (Thioulouse et al. 2018). The final plot was generated using the "ggplot2" package in R (Wickham 2016).

Results

Sightings

Between 2019 and 2024, a total of 104 sightings of putative *A. naccarii* specimens were recorded by FIPSAS in the north-eastern Italian river basins and coastal waters (Table 1). Among these, five individuals were smaller than 25 cm and were therefore not microchipped, as per handling guidelines. Since this size is below the minimum threshold used for restocking, these individuals are unlikely to be the result of artificial releases

Table 1. Summary of *A. naccarii* sightings reported by FIPSAS from 2019 to 2024. For each year and location, the total number of reported individuals is shown, along with the number of individuals smaller than 25 cm (in parenthesis), the number of tissue samples collected and the number of untagged individuals in which a microchip was implanted. Individuals smaller than 25 cm were not microchipped due to handling protocols.

Year	River/Sea	N. sightings ($N_{<25\ cm}$)	Tissue samples collected	Microchips added
2019	Livenza River	3 (3)	0	0
2020	Livenza River	3 (1)	0	0
2021	Livenza River	18 (0)	4	4
2022	Livenza River	4 (0)	0	1
2023	Livenza River	39 (0)	13	21
	Tagliamento River	5 (0)	0	0
	Piave River	3 (1)	0	0
2024	Livenza River	29 (0)	10	11
	Adriatic Sea	2 (0)	0	0
Total		**104 (5)**	**27**	**37**

and are instead possibly attributable to natural reproduction events. A microchip was implanted in 37 individuals that were found to be untagged at the time of capture, and tissue samples were successfully collected from 27 of them. Most of the sightings and sampling activities occurred in the Livenza River, which represents a key area for monitoring, with occasional records also reported from the Tagliamento and Piave rivers, and from the Adriatic Sea.

Genetic analyses for species identification and estimation of genetic diversity

A total of 27 tissue samples were analysed. Mitochondrial DNA analysis revealed that 24 individuals carried haplotypes consistent with *A. naccarii* and compatible with those of the original F0 broodstock. In contrast, three individuals displayed mitochondrial haplotypes belonging to other sturgeon species: two to *A. gueldenstaedtii* and one to *A. transmontanus*, indicating non-naccarii maternal origin. Species identification based on nuclear markers confirmed the presence of *A. naccarii* genetic material in all 27 individuals, thus demonstrating that each specimen – including those with non-naccarii mitochondrial haplotypes – had at least one *A. naccarii* parent. In the three discordant cases, nuclear amplification also revealed the presence of diagnostic alleles from *A. gueldenstaedtii* or *A. transmontanus*, confirming the hybrid nature of these individuals.

In summary, of the 27 individuals analysed, 24 were identified as pure *A. naccarii*, while three were interspecific hybrids: two resulting from crosses between *A. gueldenstaedtii* and *A. naccarii* – species that are phylogenetically closely related – and one individual from a cross between *A. transmontanus*, a North American species, and *A. naccarii*.

The genetic distances among sampled individuals were visualized through a multidimensional scaling (MDS) analysis based on microsatellite data (Figure 1). The plot shows that individuals genetically identified as pure *A. naccarii* form a compact and homogeneous cluster, indicating low genetic variability within this group and confirming their common origin from the same F0 broodstock. In contrast, the three interspecific hybrids – two with *A. gueldenstaedtii* (hyb_GxN) and one with *A. transmontanus* (hyb_TxN) – are clearly separated from the main cluster, reflecting substantial genetic divergence at the nuclear level. This separation supports the molecular evidence of hybridization and highlights the genetic distinctiveness of these individuals. The fact that all three individuals showed at many loci, two alleles compatible with *A. naccarii* and two with the alien species, supports the hypothesis that these animals are first-generation hybrids.

Discussion

Genetic evidence supports the success of restocking efforts

The results of this study highlight the significant contribution of restocking programs to the current population of Adriatic sturgeon (*Acipenser naccarii*) in the Livenza River. The genetic analyses confirmed that the majority of recaptured individuals for which a tissue sample was collected are genetically compatible with the F0 breeding stock originally used for propagation. However, this does not allow us to distinguish between animals that were released directly and those that may have been naturally born from previously released individuals that have reached sexual maturity. In

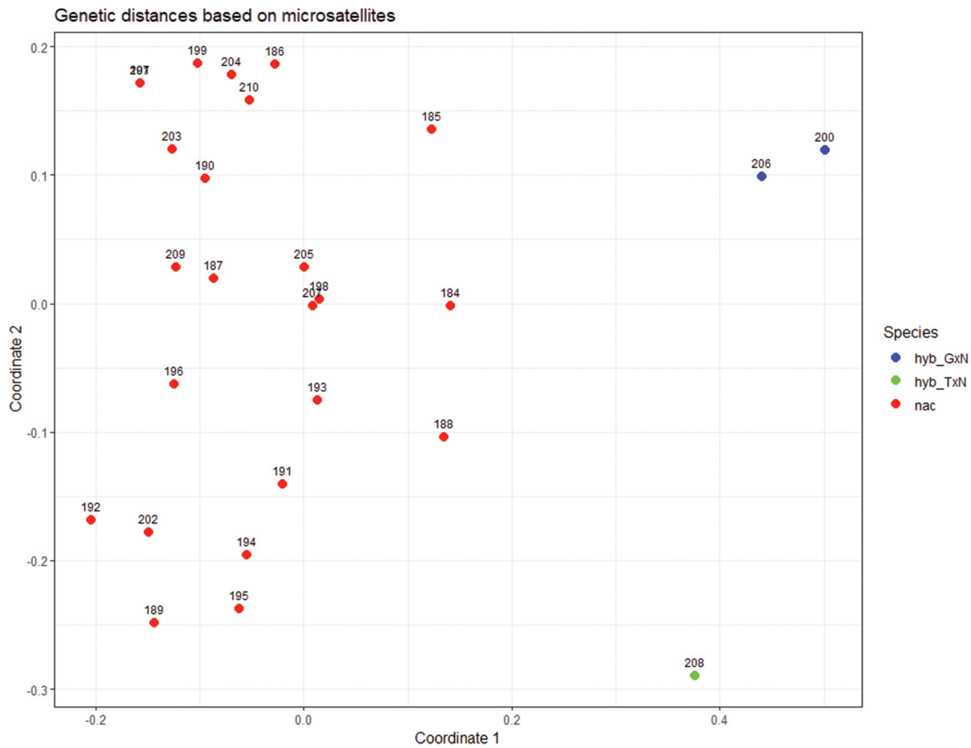

Figure 1. Multidimensional scaling (MDS) plot showing the genetic distances among sampled individuals based on microsatellite data from 15 loci. Each point represents a single individual, identified by the last three digits of the Sample ID reported in Table 2. Colors indicate the maternal species as determined by mitochondrial haplotypes: red for *A. naccarii*, blue for *A. gueldenstaedtii* and green for *A. transmontanus*. The legend reflects the final genetic classification of each specimen, distinguishing pure *A. naccarii* (nac) from interspecific hybrids with *A. gueldenstaedtii* (hyb_gxn) or *A. transmontanus* (hyb_txn).

fact, the Livenza River is one of the rivers where spontaneous reproductive activity of *A. naccarii* has recently been observed. Despite this, the presence of small untagged juveniles strongly suggests natural reproduction events.

This finding highlights the effectiveness of past restocking efforts in maintaining the presence of *A. naccarii* in the wild. However, it also underscores a key limitation of genetic analyses: the inability to distinguish between individuals directly released from hatcheries and those born in the wild from released parents. Since all reintroduced fish originate from the same genetic pool, their offspring remain genetically indistinguishable from newly released individuals, making it impossible to differentiate between captive-bred and naturally born specimens based solely on genetic data.

The only reliable criterion for identifying probably wild-born individuals is their size at recapture, particularly when it falls outside the expected size range for individuals released in the same year. In this regard, field observations of small, untagged juveniles – below the minimum size threshold established for release – provide strong circumstantial evidence that natural reproduction may have resumed in the wild. These individuals, although not sampled for genetic analyses, due to their small size, represent a tangible indication that successful spawning occurs in natural environments. Regardless of their origin, the fact that reproductive activity has been definitively confirmed in at least three of the six years for which data are available underscores the importance of continued conservation efforts.

This evidence – together with the discovery of a stranded female in the Po River carrying mature eggs and the capture of a juvenile *A. naccarii* in the same river (Congiu et al. 2021) – led to the reclassification of the Adriatic sturgeon in the latest IUCN assessment (2022), from "*Critically Endangered and possibly extinct in the wild*" to "*Critically Endangered*". This represents a major achievement, as *A. naccarii* is the only sturgeon species worldwide to have shown an improvement in conservation status compared to the previous assessment in 2009.

Table 2. Biometric data and genetic classification of the 27 individuals sampled between 2021 and 2024. For each specimen, weight (kg), total length (cm), mitochondrial haplotype and results of species-specific nuclear DNA markers are reported. Mitochondrial and nuclear markers were used to detect, respectively, the maternal species and presence of diagnostic alleles for *A. naccarii* (NAC), *A. gueldenstaedtii* (GUE) and *A. transmontanus* (TRA). Final classification indicates whether the individual was identified as a pure *A. naccarii* or as an interspecific hybrid. The individual marked with an asterisk (*), first captured and microchipped in December 2021, was subsequently recaptured in June 2023 in the Tagliamento River, where it was identified via microchip.

				Genetic analyses for species identification				
		Biometric data		mtDNA	nDNA			
Data	Sample ID	Kg	cm	Haplotype	NAC	TRA	GUE	Final classification
8 November 2021	**NAC/24/184**	1.20	61	NAC_Hap2	+	–	–	**Pure** *A. naccarii*
11 November 2021	**NAC/24/187**	0.40	46	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
11 December 2021	**NAC/24/185**	27.60	162	NAC_Hap3	+	–	–	**Pure** *A. naccarii*
13 December 2021	**NAC/24/186***	14.00	135	NAC_Hap7	+	–	–	**Pure** *A. naccarii*
8 April 2023	**NAC/24/188**	0.85	55	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
21 April 2023	**NAC/24/189**	1.06	61	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
25 April 2023	**NAC/24/190**	1.68	66	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
10 May 2023	**NAC/24/191**	1.40	64	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
11 May 2023	**NAC/24/192**	4.42	89	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
25 May 2023	**NAC/24/193**	1.22	59	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
2 June 2023	**NAC/24/194**	5.78	101	NAC_Hap2	+	–	–	**Pure** *A. naccarii*
5 June 2023	**NAC/24/195**	2.22	72	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
3 July 2023	**NAC/24/196**	1.82	67	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
10 September 2023	**NAC/24/197**	1.42	61	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
23 October 2023	**NAC/24/202**	1.28	63	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
24 October 2023	**NAC/24/203**	3.61	87	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
24 October 2023	**NAC/24/204**	0.62	52	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
12 April 2023	**NAC/24/198**	0.69	50	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
20 April 2024	**NAC/24/199**	2.40	74	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
22 April 2024	**NAC/24/200**	2.08	72	GUE	+	–	+	**Hybrid GUExNAC**
6 July 2024	**NAC/24/201**	2.26	75	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
16 July 2024	**NAC/24/205**	1.88	67	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
2 Semptember 2024	**NAC/24/206**	6.30	98	GUE	+	–	+	**Hybrid GUExNAC**
11 Semptember 2024	**NAC/24/207**	2.44	76	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
17 Semptember 2024	**NAC/24/208**	7.20	106	TRA	+	+	–	**Hybrid TRAxNAC**
25 Semptember 2024	**NAC/24/209**	1.44	67	NAC_Hap5	+	–	–	**Pure** *A. naccarii*
1 October 2024	**NAC/24/210**	4.40	96	NAC_Hap5	+	–	–	**Pure** *A. naccarii*

Detection of interspecific hybrids and associated conservation risks

Another critical finding of this study is the presence of interspecific hybrids among the recaptured individuals, including two hybrids between *A. naccarii* and the Russian sturgeon (*A. gueldenstaedtii*), and one hybrid between *A. naccarii* and the white sturgeon *(A. transmontanus*). The occurrence of these individuals in the Livenza River highlights the potential risks associated with uncontrolled releases and escapes from aquaculture facilities. Several explanations may account for this finding. Firstly, the production of interspecific hybrids is a common practice in sturgeon aquaculture due to their favourable growth and commercial traits. Such practices have been ongoing for at least two decades, raising the possibility that hybrid individuals were inadvertently included in some *A. naccarii* broodstocks not genetically certified. Compared to the other possible scenarios that will be presented later, this is arguably the most concerning, as it implies that hybrids may have been inadvertently used for reproduction in restocking programs. This possibility is particularly alarming because it would suggest that potentially hundreds of hybrid individuals may have entered natural ecosystems through past restocking events. Fortunately, this scenario is also the easiest to verify: a comprehensive genetic analysis of all current broodstock individuals could readily detect any hybrids present and thus either confirm or exclude this route of introduction. If this hypothesis is excluded, however, distinguishing between the remaining two scenarios becomes more challenging. The presence of hybrids might result from direct escapes of farmed individuals or from releases by private individuals keeping ornamental sturgeons. An additional, not mutually exclusive possibility is that exotic sturgeons or interspecific hybrids that are known to be fertile (Ludwig et al. 2009; Beridze et al. 2022), once introduced into the river system, have participated in natural spawning events, and that the hybrids detected in this study were consequently born in the wild.

Regardless of the origin, the presence of non-native sturgeons poses a serious conservation threat due to the risks of hybridization (with potential long-term introgression) and competition for resources. Hybridization may lead to the progressive loss of the genetic identity and local adaptations of *A. naccarii*, especially if fertile hybrids or their backcrosses reproduce with native individuals over multiple generations. This genetic introgression could compromise both the evolutionary potential and the ecological fitness of the species, making recovery efforts based on genetically pure individuals less effective or even futile in the long-term. Moreover, the cryptic nature of introgression, especially in later generations, may make detection extremely difficult with standard genetic tools, allowing the silent spread of non-native genetic material within the population. It is also important to note that the genetic markers used in this study are single-locus markers, and due to Mendelian segregation, they may be lost from the second generation onwards. This limitation means that additional hybrids – particularly later-generation backcrosses – may not have been detectable using the current marker set. Therefore, the number of hybrid individuals in the wild may be underestimated. These findings underline the urgent need for rigorous genetic screening of all captive sturgeon individuals intended for conservation breeding, along with the implementation of strict legislation and biosecurity measures to prevent further introductions of alien species into natural river systems. In this regard, banning the sale of non-native species for ornamental use could be a useful measure, as such fish are often released into the wild once they outgrow private facilities. Furthermore, the presence of exotic sturgeons in aquaculture plants or recreational fishing lakes significantly increases the risk of accidental escapes. A recent example occurred in the province of Brescia, where more than 100 *A. gueldenstaedtii* individuals escaped from a fish farm. Preventive measures must be adopted to minimize such events and safeguard the genetic integrity of native populations.

Toward a coordinated framework for sturgeon conservation

A broader issue emerging from this study is the fragmentation and lack of coordination in the collection and management of monitoring data. Currently, recapture records are dispersed across multiple entities, making it difficult to construct a comprehensive and coherent picture of both past and present restocking outcomes. Establishing a centralized database that integrates all available information – such as genetic profiles, biometric data and microchip records – is crucial to enhancing the effectiveness of conservation strategies. This critical need is being addressed by the newly approved LIFE RESTORE project, which aims to unify monitoring efforts across the historical distribution range of *A. naccarii*. With the support of five Italian regions, the coordination of the University of Padua, and the involvement of leading research institutions and environmental authorities, LIFE RESTORE represents a significant step forward in the structured and coordinated conservation of the species.

The data presented in this study are highly partial and reflect only a limited subset of the monitoring activities carried out across various past projects. This fragmentation further underscores the urgency of adopting a more unified and scientifically guided approach to sturgeon conservation in Italy.

In 2019, the Pan-European Action Plan for sturgeon conservation was adopted under the Bern Convention, setting forth clear guidelines for the restoration of sturgeon populations throughout Europe. These include not only the coordination of conservation actions, but also habitat restoration, regulation of fisheries and aquaculture, reinforcement of legal protections and the implementation of genetic conservation protocols. Through the LIFE RESTORE project, Italy is beginning to align its conservation efforts with these international directives. The launch of the LIFE RESTORE project could provide a clearer definition of long-term strategies for *A. naccarii* and contribute to the development of a structured, accessible and centralized system for monitoring and managing available data. In addition to data harmonization, LIFE RESTORE will address other critical gaps highlighted by this study. First, the project will carry out a systematic screening of the genetic diversity of broodstock in the different breeding facilities, combined with the adoption of breeding plans aimed at minimizing genetic erosion and enhancing the adaptive potential of the reintroduced population. In addition, the project will include specific actions aimed at evaluating the quality and suitability of riverine habitats for spawning and juvenile development, through the collection of hydromorphological and physicochemical data. Furthermore, a shared tagging protocol and a national digital platform for recording genetic profiles, tagging information and recapture events will be developed to support coordinated monitoring and improve conservation efficiency across the species' range. Combined with strengthened legislation and continued scientific research, these efforts offer renewed hope for the long-term survival of this critically endangered species.

Conclusion

The conservation of *Acipenser naccarii* is at a crucial crossroads. While recent monitoring efforts and genetic analyses reveal encouraging signs of population persistence and possible natural reproduction, the species remains critically endangered and its future is far from secure. This study highlights the value of integrating genetic tools with field monitoring to assess the outcomes of past restocking programs and to detect emerging threats such as hybridization with non-native species. The presence of small untagged juveniles and the detection of interspecific hybrids underscore both the potential for recovery and the fragility of this process. Moving forward, conservation strategies must prioritize the creation of a coordinated national monitoring network, the establishment of a centralized recapture database, the enforcement of stricter controls on the use and movement of non-native sturgeon species and the training of the personnel involved in monitoring activities, so that they are able to identify potentially hybrid animals and temporarily hold them until the results of the genetic analyses are available. These measures, along with habitat restoration and alignment with international conservation frameworks, are essential to ensure the long-term viability of *A. naccarii* in the wild.

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Disclosure statement

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