

Comment

# Native or Overlooked Translocation? Comment on Antognazza et al. Current and Historical Genetic Variability of Native Brown Trout Populations in a Southern Alpine Ecosystem: Implications for Future Management. *Fishes* 2023, 8, 411

Tommaso Righi <sup>1,2,\*</sup> , Andrea Splendiani <sup>1</sup> , Vincenzo Caputo Barucchi <sup>1</sup>  and Andrea Gandolfi <sup>2</sup> 

<sup>1</sup> Dipartimento di Scienze della Vita e dell'Ambiente, Università Politecnica delle Marche, Via Brecce Bianche, 60100 Ancona, Italy; a.splendiani@staff.univpm.it (A.S.); v.caputo@staff.univpm.it (V.C.B.)

<sup>2</sup> Conservation Genomics Research Unit, Research and Innovation Centre—Fondazione Edmund Mach, 38098 San Michele all'Adige, TN, Italy; andrea.gandolfi@fmach.it

\* Correspondence: t.righi@staff.univpm.it

**Abstract:** The recent revision of Italian legislation on nature conservation has highlighted the pressing necessity of elucidating the native distribution range of managed species. A recent study by Antognazza et al. (Current and Historical Genetic Variability of Native Brown Trout Populations in a Southern Alpine Ecosystem: Implications for Future Management. *Fishes* 2023, 8, 411) provides insights into the native status of brown trout in the Lombardy Prealps, northern Italy, and advocates urgent conservation measures. However, the possible effect of historical and recent anthropogenic impacts was dismissed in the paper. Here, we present how human-mediated activities plausibly might contribute to the observed distribution of population genetic variation, considering both the available literature and ongoing “Mediterranean trout” stocking activities in the region. Implementing management strategies without clear scientific evidence poses significant risks to native biodiversity conservation.

**Keywords:** brown trout; freshwater Italian biodiversity; conservation genetics; invasive species; *Salmo trutta*; conservation biology



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In Italy, local lineages of the brown trout (*Salmo trutta*) complex stand as a vulnerable component of native freshwater biodiversity, threatened by various anthropogenic stressors, including habitat degradation and invasion by non-native salmonids [1]. Trout management traditionally revolves around angling-centric approaches. In the last century, reared Atlantic brown trout and rainbow trout (*Oncorhynchus mykiss*) have been stocked yearly by local Italian administrations to support fisheries, continuing despite the legal ban on introducing non-native fishes into nature [2,3]. In addition, stocking activities made use of strains originating from peninsular populations, branded as “Mediterranean trout” and marketed as a conservation-minded substitute to non-native Atlantic brown trout. This practice, ongoing for at least the last decade, has ignored the widely reported differentiation of native brown trout populations [4–6], leading to the recurrent spread of introgressed individuals and the translocation of reared native trout beyond their natural distribution range, including to protected areas such as Natura 2000 sites and National Parks [4]. In this context, it is imperative to consider the potential past and present human impact when making inferences on the natural distribution of trout populations. The misinterpretation of data—already feared at the beginning of the massive stocking era [7]—and subsequent implementation of management strategies lacking robust scientific foundation pose considerable risks to biodiversity conservation efforts. Hence, we strongly dissent from the conclusions drawn by Antognazza et al. [8] regarding the native status of Adriatic and Mediterranean trout lineages in the Lombardy Prealps region of northern Italy; even more,

we disagree with their conservation and management recommendations. Our critique seeks to elucidate why we diverge from their interpretation of results, highlighting the necessity for considering anthropogenic impacts in the observed population genetic data distribution.

The article by Antognazza et al. titled “Current and Historical Genetic Variability of Native Brown Trout Populations in a Southern Alpine Ecosystem: Implications for Future Management” (<https://doi.org/10.3390/fishes8080411>) [8] investigated the distribution of brown trout within the Lombardy prealpine region. The authors genetically analysed historical *S. trutta* specimens originating from the Po Basin, focusing on the 326 bp partial sequence of the mitochondrial control region (mtDNA CR). Additionally, modern samples were assessed for partial CR (407 bp) sequences and restriction fragment length polymorphism of the LDH-C1\* nuclear locus. Based on the detection of the Adriatic (AD) haplogroup in two historical specimens predating the massive introduction of the Atlantic strain, and the presence of five already described and three new AD, Marmoratus (MA) and Mediterranean (ME) haplotypes in modern samples, the authors concluded that these haplogroups are naturally widespread in the prealpine region. They advocate urgent conservation efforts to protect indigenous trout lineages despite the introgression with hatchery variants.

After carefully reviewing the paper by Antognazza and colleagues, several criticisms have emerged regarding both the data presented and their interpretations.

The first doubt regards the accuracy in interpreting the sequencing data. Two sequences from museum samples, ADSZ3 (GenBank acc. num. OQ676373) and MASZ12 (GenBank acc. num. OQ676374), reported as new, exhibit transitions in sites conserved among haplotypes and haplogroups. These are type-2 transitions (TS2: C → T/G → A), commonly observed in ancient DNA (aDNA) studies and stemming from PCR-induced errors due to postmortem DNA modifications, such as cytosine deamination (and its analogue, 5-methyl cytosine) to uracil (and thymine) [9,10]. Following the correction of such an artefact, ADSZ3 would correspond to at least 100 sequences available in GenBank, including ADcs1, whereas MASZ12 would be reassigned to a haplotype (Ma2c) described for both *S. marmoratus* (GenBank acc. num. JQ582461) and *S. carpio* (GenBank acc. num. KJ834825). The underestimation of potential sequencing errors might result in an overestimation of diversity and its consequent interpretation: great discrepancy can be observed in numbers of haplotypes detected in Zaccara et al. [11] compared to Berrebi et al. [4] and Splendiani et al. [12] for the same locations in Sardinia (see also [13]). Potential misinterpretations could also affect the modern haplotype MA1A, which shows a C-T mutation at position 15690, a conserved site in all previously described MA haplotypes.

The detection of the AD haplogroup in the historical sample is consistent with that in a previous investigation [14]. Splendiani et al. [15] suggested the colonisation and wide distribution of ADcs1 in the Po River before the last glacial periods. Subsequently, glacial expansion would have driven the wide extinction of brown trout in the Alpine area except in refugia at the periphery of glacial coverage, where isolation would have locally promoted the emergence of new haplotypes. For instance, ADporh-1 appeared in the Maritime Alps refugium, while the *S. carpio* AD haplotypes (ScarAD-1 and ScarAD-2) emerged within a refugium close to Lake Garda. Other remnant brown trout populations may have survived south of the Alps, in parapatry with native marble trout, although they have not yet been documented. Antognazza et al. [8] conclude on the natural presence in the Lombardy prealpine region of the Adriatic lineage based on the finding of an AD haplotype in the two historical samples from Torbole (Lake Garda, 1821) and Serio River (1858), predating the beginning of massive stocking activities. However, the occurrence of trout translocations were documented in this area during the first half of the 1800s [7,16] and Lake Garda harbours endemic *S. carpio*, which exhibits both the AD and MA haplogroups [6,15]. The data align well with other historical samples (1884) from the same locality identified as *S. carpio* [8]. From this limited information, Antognazza et al.’s [8] conclusion about the natural presence of the Adriatic lineage in the entire prealpine ecosystem seems speculative and vague.

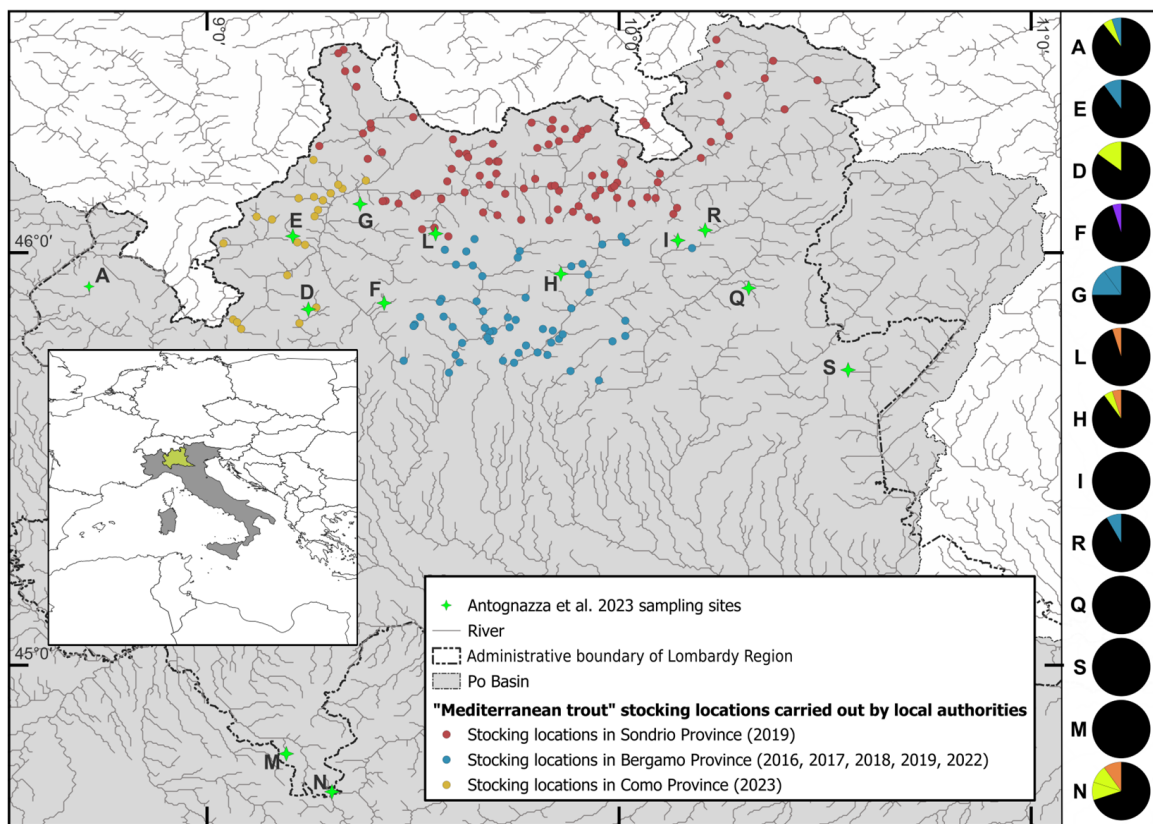
The most significant concerns pertain to the interpretation of the results obtained from modern samples. Firstly, the authors juxtapose the observed mtDNA variability within the non-native Atlantic (AT) and native AD, MA and ME haplogroups to formulate a hypothesis of prolonged native brown trout presence in the area, affording them sufficient time to accrue genetic variability. In contrast, the presence of solely the exotic ATcs1 haplotype is interpreted as indicative of a single stock origin. However, as reported in Table S3 and Figure 2 by the authors [8], nine of the twelve AT haplotypes used in the analysis collapsed to the ATcs1 due to the limited sequence length considered. Among these, the four most common haplotypes found in European and Italian hatchery strains (ATcs1, ATcs2, ATcs3 and ATcs4) are listed. Therefore, the observed genetic uniformity of the Atlantic haplogroup must be attributed to the sequence length analysed in this study rather than to a signal of single stock origin. This is further underscored by the presence of the DA haplotype in a sample. Likewise, it cannot be ruled out that the presence of highly differentiated AD, ME and MA haplotypes detected in modern samples could be the result of fish introduction from multiple or mixed stocks (see below).

Secondly, although the authors selected sites in Lombardy presumed to be free from stocking practices, the influence of trout introduction is evident through the high percentage of non-native Atlantic haplotypes (90%) and the PCR-RFLP allele LDH-C1\* 90 (98%). Consequently, the authors' decision to dismiss a priori the artificial presence of "Mediterranean trout" in these sites is at least questionable. This is even more true since the authors did not consider official stocking of "Mediterranean trout" records as a criterion to exclude sampling sites. The investigated Alpine region, in fact, is subject to extensive yearly stocking activities, involving the utilisation of both Atlantic and "Mediterranean trout" hatchery stocks. Already introduced as early as 2009 in the Alpine lake in Valtellina [17], the "Mediterranean trout" represents a substantial proportion of fish stocked in the Como, Sondrio, Bergamo and Brescia provinces by the local angling associations [18–29] (Figure 1). Moreover, the study by Antognazza and colleagues [8] lacks a phylogeographic rationale to elucidate the presence of putative native haplotypes, considering their geographic distribution as described in the literature. The AD-Tyrrh1 haplotype has been described in the Peri-Tyrrhenian area, being widespread in Liguria, Corsica and Sardinia and sporadically present in the Tyrrhenian slope of Central Italy [4,12,30,31]. Its presence in Tuscany–Emilian Apennine populations is the result of stocking activities [31], while it has been never documented along the Italian Adriatic slope [4,15]. The new haplotype AD-A1, found in the Apennine river Trebbia, stocked with "Mediterranean trout" [32], differs by a single-step mutation (C-T at position 15690) from the haplotype AD-PostaFibreno (GenBank acc. num. JQ314219), surprisingly not included in the lists of haplotypes used as reference in the paper, which is an endemic and fixed haplotype in Posta Fibreno Lake and Fibreno Basin in the Tyrrhenian slope of Central Italy [5,6]. The haplotype ADK1 has been reported in the Adriatic Basin in Croatia (Krka) [33] and the western Balkan Aegean Basin (Struma; haplotype AdAE1, GenBank acc. num. GQ357908) [34], and it completely overlaps with 561 bp of haplotype ADBoz (GenBank acc. num. DQ318128) found in the Aegean drainage in Serbia [33,35]. Finally, the haplotype misinterpreted as newly described, MeA1, has been previously identified in the Panigale Hatchery, Emilia Romagna [36] (GenBank acc. nums. KY661248, KY661249 and KY661254).

Notably, the AD-Tyrrh1 and AD-PostaFibreno haplotypes are commonly present in reared Mediterranean brown trout from northern and central Italian hatcheries [31,36,37] (GenBank Acc. nums. KY661208-KY661211 and KY661213-KY661225) and their dissemination beyond their natural range has been reported [31,38].

Stocking material used in the Lombardy Prealps came from—and was exchanged among—several hatcheries [18–20]. Although the precise origins of the reared strains are not explicitly documented, the detected similarity in genetic make-up may suggest a shared mixed ancestry with the Panigale hatchery. The human-mediated introduction of "Mediterranean trout" may also explain the limited percentage of both AD and ME mtDNA

haplotypes and the non-Atlantic allele LDH-C1\*100—if not from Danubian or marble trout [39]—in prealpine sites compared to the reference southwestern native populations.



**Figure 1.** Recorded “Mediterranean trout” stocking locations in the prealpine area of the Lombardy region (Italy) authorised by three local authorities (provinces), overlapping most sampling sites from Antognazza et al. [8]. Data were obtained from fishing associations’ web sources [20,21,28] and requested from the relevant regional office [22–25]. Pie charts report mtDNA haplogroup distributions detected by Antognazza et al. [8]; yellow = Adriatic (AD), orange = Mediterranean (ME), blue = Marmoratus (MA), purple = Danubian (DA) and black = Atlantic (AT). The ID letters have been kept identical to the original work.

Considering the aforementioned observations, the presence of AD and ME haplogroups cannot represent—alone—evidence of natural brown trout presence in this area, excluding a priori their more plausible origin from stocking activities.

Urgent management actions proposed solely on the sporadic presence of mtDNA haplogroups should be approached with caution [40]. In fact, if the presence of Atlantic haplotypes in Mediterranean populations identified anthropogenic manipulation, relying solely on the mtDNA haplogroup to inform management strategies has significant limitations in cases with complex evolutionary histories characterised by extinction/colonisation and isolation/contact events, such as for brown trout inhabiting the Mediterranean Basin [6]. The AD, ME and MA haplotypes are widespread throughout the entire Mediterranean Basin [41] and are shared among ecologically different species [4,6,15,42,43]. In these complex scenarios, including human manipulation, the analysis of complete mtDNA CR sequences emerges as imperative for biogeographic reconstruction, surpassing the mere examination of mtDNA haplogroups [12,15]. In any case, mtDNA CR alone is not sufficient for a Management Unit definition [44]. This aspect cannot be neglected if the conservation of the species is called into question, especially if well-structured populations exist. The conservation of biodiversity, recently enshrined in the Italian Constitution [45], must not be

jeopardised by the implementation of management practices based on erroneous data and simplistic or speculative interpretations.

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## References

- Rondinini, C.; Battistoni, A.; Teofili, C. (Eds.) *Lista Rossa IUCN dei Vertebrati Italiani 2022*; Comitato Italiano IUCN e Ministero dell’Ambiente e della Sicurezza Energetica: Roma, Italy, 2022.
- D.P.R. 12 Marzo 2003, n. 120. Regolamento Recante Modifiche ed Integrazioni al Decreto del Presidente Della Repubblica 8 Settembre 1997, n. 357, Concernente Attuazione della Direttiva 92/43/CEE Relativa Alla Conservazione Degli Habitat Naturali e Seminaturali, Nonché Della Flora e Della Fauna Selvatiche. (G.U. 30 Maggio 2003, n. 124). 2003. Available online: [https://www.gazzettaufficiale.it/atto/serie\\_generale/caricaDettaglioAtto/originario?atto.dataPubblicazioneGazzetta=2003-05-30&atto.codiceRedazionale=003G0142](https://www.gazzettaufficiale.it/atto/serie_generale/caricaDettaglioAtto/originario?atto.dataPubblicazioneGazzetta=2003-05-30&atto.codiceRedazionale=003G0142) (accessed on 5 May 2024).
- D.P.R. 5 Luglio 2019, n. 102. Regolamento Recante Ulteriori Modifiche dell’Articolo 12 del Decreto Del Presidente Della Repubblica 8 Settembre 1997, n. 357, Concernente Attuazione della Direttiva 92/43/CEE Relativa Alla Conservazione Degli Habitat Naturali e Seminaturali, Nonché Della Flora e Della Fauna Selvatiche. (19G00108) (GU Serie Generale n.208 Del 05-09-2019). 2019. Available online: <https://www.gazzettaufficiale.it/eli/id/2019/09/05/19G00108/sg> (accessed on 5 May 2024).
- Berrebi, P.; Caputo Barucchi, V.; Splendiani, A.; Muracciole, S.; Sabatini, A.; Palmas, F.; Tougard, C.; Arculeo, M.; Marić, S. Brown trout (*Salmo trutta* L.) high genetic diversity around the Tyrrhenian Sea as revealed by nuclear and mitochondrial markers. *Hydrobiologia* **2019**, *826*, 209–231. [CrossRef]
- Rossi, A.R.; Talarico, L.; Petrosino, G.; Crescenzo, S.; Tancioni, L. Conservation Genetics of Mediterranean Brown Trout in Central Italy (Latium): A Multi-Marker Approach. *Water* **2022**, *14*, 937. [CrossRef]
- Gratton, P.; Allegrucci, G.; Sbordoni, V.; Gandolfi, A. The Evolutionary jigsaw puzzle of the surviving trout (*Salmo trutta* L. Complex) diversity in the Italian region. A Multilocus Bayesian Approach. *Mol. Phylogenetics Evol.* **2014**, *79*, 292–304. [CrossRef] [PubMed]
- Pavesi, P. L’ultima Sementa di Pesci nei Nostri Laghi. Reale Istituto Lombardo di Scienze e Lettere. Rendiconti. Serie II, 14, p. 227. 1881. Available online: [https://books.google.it/books?id=np89AAAAYAAJ&source=gbs\\_navlinks\\_s&redir\\_esc=y](https://books.google.it/books?id=np89AAAAYAAJ&source=gbs_navlinks_s&redir_esc=y) (accessed on 5 May 2024).
- Antognazza, C.M.; Palandačić, A.; Delmastro, G.B.; Crosa, G.; Zaccara, S. Current and Historical Genetic Variability of Native Brown Trout Populations in a Southern Alpine Ecosystem: Implications for Future Management. *Fishes* **2023**, *8*, 411. [CrossRef]
- Binladen, J.; Wiuf, C.; Gilbert, M.T.P.; Bunce, M.; Barnett, R.; Larson, G.; Greenwood, A.D.; Haile, J.; Ho, S.Y.; Hansen, A.J. Assessing the fidelity of ancient DNA sequences amplified from nuclear genes. *Genetics* **2006**, *172*, 733–741. [CrossRef]
- Orlando, L.; Allaby, R.; Skoglund, P.; Der Sarkissian, C.; Stockhammer, P.W.; Ávila-Arcos, M.C.; Fu, Q.; Krause, J.; Willerslev, E.; Stone, A.C. Ancient DNA analysis. *Nat. Rev. Methods Primers* **2021**, *1*, 14. [CrossRef]
- Zaccara, S.; Trasforini, S.; Antognazza, C.M.; Puzzi, C.; Britton, J.R.; Crosa, G. Morphological and genetic characterization of Sardinian trout *Salmo Cettii* Rafinesque, 1810 and their conservation implications. *Hydrobiologia* **2015**, *760*, 205–223. [CrossRef]
- Splendiani, A.; Righi, T.; Fioravanti, T.; Sabatini, A.; Palmas, F.; Tougard, C.; Berrebi, P.; Talarico, L.; Caputo Barucchi, V. Population genetics, demography and conservation of mediterranean brown trout from Sardinia. *Aquat. Conserv. Mar. Freshw. Ecosyst.* **2024**, *34*, e4099. [CrossRef]
- Sanz, N.; Araguas, R.-M.; Giampiccolo, M.; Duchi, A. Native population structure beyond hatchery introgression in the endemic Sicilian trout. *Diversity* **2023**, *15*, 274. [CrossRef]
- Splendiani, A.; Fioravanti, T.; Giovannotti, M.; Olivieri, L.; Ruggeri, P.; Nisi Cerioni, P.; Vanni, S.; Enrichetti, F.; Caputo Barucchi, V. Museum samples could help to reconstruct the original distribution of *Salmo trutta* Complex in Italy. *J. Fish Biol.* **2017**, *90*, 2443–2451. [CrossRef]
- Splendiani, A.; Berrebi, P.; Tougard, C.; Righi, T.; Reynaud, N.; Fioravanti, T.; Lo Conte, P.; Delmastro, G.B.; Baltieri, M.; Ciuffardi, L. The role of the south-western alps as a unidirectional corridor for mediterranean brown trout (*Salmo trutta* Complex) lineages. *Biol. J. Linn. Soc.* **2020**, *131*, 909–926. [CrossRef]
- Monti, M. Notizie dei Pesci Delle Provincie di Como e Sondrio e del Cantone Ticino; Dalla tip. di Carlo Franchi, 1864. Available online: [https://books.google.it/books?id=xvg-AAAAYAAJ&printsec=frontcover&hl=it&source=gbs\\_ge\\_summary\\_r&cad=0#v=onepage&q&f=false](https://books.google.it/books?id=xvg-AAAAYAAJ&printsec=frontcover&hl=it&source=gbs_ge_summary_r&cad=0#v=onepage&q&f=false) (accessed on 5 May 2024).
- Torri, M. La Reintroduzione della Trota Mediterranea. Cronache di una Stagione di Ripopolamenti. *Pescare in Valtellina*. 2009. Available online: <https://issuu.com/upssondrio/docs/piv-2-2009> (accessed on 5 May 2024).
- APS COMO. Relazione Attività 2018 Incubatoio Ittico Di VALMOREA. *Relazione Sulla Gestione Relativa All’esercizio Sociale 2018—Bilancio Preventivo Esercizio Sociale Anno 2019*. 2018. Available online: <https://www.aps-como.it/wp-content/uploads/relazione-assemblea-2019.pdf> (accessed on 5 May 2024).

19. APS COMO. Relazione Attività 2019 Incubatoio Ittico Di VALMOREA. *Relazione Sulla Gestione Relativa All'esercizio Sociale 2018-Bilancio Preventivo Esercizio Sociale Anno 2020*. 2019. Available online: <https://www.aps-como.it/wp-content/uploads/relazione-Presidente-Assemblea-2019-20-web.pdf> (accessed on 5 May 2024).
20. APS COMO. *Ripopolamenti 2023*; APS COMO: Lora, Italy, 2023.
21. FIPSAS Bergamo. Campagna Ittiogenica 2022: Nel Mese di Maggio Saranno Seminati 260mila Avannotti di Marmorata e 65mila di Trota Fario Mediterranea. Available online: <https://www.fipsasbergamo.it/campagna-ittiogenica-2022-nel-mese-di-maggio-saranno-seminati-260mila-avannotti-di-marmorata-e-65mila-di-trota-fario-mediterranea/> (accessed on 5 May 2024).
22. Regione Lombardia. Piano Annuale di Ripopolamento Ittico Delle Acque Bergamasche per l'anno 2016. 2016.
23. Regione Lombardia. Piano di Riparto del Novellame Di Trota Marmorata, Trota Fario e Trota Lacustre Prodotta Negli Incubatoi Ittici di Valle Finalizzato al Ripopolamento delle Acque Bergamasche per l'anno 2017. 2017.
24. Regione Lombardia. Piano di Riparto del Novellame di Trota Marmorata, Trota Fario e Trota Lacustre Prodotta Negli Incubatoi Ittici di Valle Finalizzato al Ripopolamento delle Acque di Competenza dell'UTR Bergamo per L'anno 2018. 2018.
25. Regione Lombardia. Piano di Riparto del Novellame di Trota Marmorata, Trota Fario e Trota Lacustre Prodotta Negli Incubatoi Ittici di valle Finalizzato al Ripopolamento delle Acque di Competenza Dell'UTR Bergamo per L'anno 2019. 2019.
26. Vezza d'Oglio: Oltre 80mila Trotelle Trovano Casa nell'Oglio in Alta Valle Camonica. *Gazz. Delle Valli*. 2015. Available online: <https://www.gazzettadellevalli.it/valle-camonica/edolo/vezza-doglio-oltre-80mila-trotelle-trovano-casa-nelloglio-in-alta-valle-camonica-52929/> (accessed on 5 May 2024).
27. Vezza d'Oglio: Oltre 110mila Trotelle Introdotte nel Fiume Oglio in Alta e Media Valle Camonica. *Gazz. Delle Valli*. 2016. Available online: <https://www.gazzettadellevalli.it/edolo/vezza-doglio-oltre-110mila-trotelle-introdotte-nel-fiume-oglio-in-alta-e-media-valle-camonica-97428/> (accessed on 5 May 2024).
28. UP SONDRIO. Semine Novellame 2019. 2019. Available online: <https://www.unionepecasondrio.it/wp-content/uploads/2020/02/PIANO-SEMINE-NOVELLAME-2019.pdf> (accessed on 5 May 2024).
29. Regione Lombardia. Piano Ittico Regionale. 2022. Available online: <https://www.regione.lombardia.it/wps/portal/istituzionale/HP/DettaglioRedazionale/servizi-e-informazioni/cittadini/agricoltura/pesca/piano-ittico-regionale-vas-2022/piano-ittico-regionale-vas-2022> (accessed on 5 May 2024).
30. Talarico, L.; Mucci, N.; Greco, C.; Caniglia, R.; Mengoni, C. D5 Report on Population and Habitat in the Six Parks, Parte 2, Analisi Genetiche. 2021. Available online: <https://www.lifestreams.eu/download/d5-report-on-population-and-habitat-in-the-six-parks/> (accessed on 5 May 2024).
31. Talarico, L.; Caniglia, R.; Carosi, A.; Lorenzoni, M.; Greco, C.; Padula, A.; D'Antoni, S.; Alberti, D.; de Paoli, A.; Casali, L. Population structure, genetic diversity and demographic patterns unveil massive mediterranean brown trout manipulations in a protected area of the northern apennines (Italy). *Eur. Zool. J.* **2023**, *90*, 470–486. [CrossRef]
32. FIPSAS GENOVA. FIPSAS GENOVA: Prosegue con Successo il Progetto Aveto/Trebbia. Available online: <https://www.fipsas.it/news/2847-fipsas-genova-prosegue-con-successo-il-progetto-aveto-trebbia> (accessed on 5 May 2024).
33. Jadan, M.; Strunjak-Perović, I.; Topić Popović, N.; Čož-Rakovac, R. Three major phylogenetic lineages of brown trout (*Salmo trutta* Linnaeus, 1758) in the Krka River system (Croatia) revealed by complete mitochondrial DNA control region sequencing. *J. Appl. Ichthyol.* **2015**, *31*, 192–196. [CrossRef]
34. Kohout, J.; Šedivá, A.; Apostolou, A.; Stefanov, T.; Marić, S.; Gaffaroglu, M.; Šlecht, V. Genetic diversity and phylogenetic origin of brown trout *Salmo trutta* populations in eastern Balkans. *Biologia* **2013**, *68*, 1229–1237. [CrossRef]
35. Marić, S.; Sušnik, S.; Simonović, P.; Snoj, A. Phylogeographic study of brown trout from Serbia, based on mitochondrial DNA control region analysis. *Genet. Sel. Evol.* **2006**, *38*, 411. [CrossRef]
36. Zuffi, G.; Papa, G.; Plazzi, F. *Morphological and Genetic Characterization of Salmo trutta Species Complex Specimens from the Panigale Fish Farm (Lizzano in Belvedere, BO)*; University of Bologna: Bologna, Italy, 2017.
37. Righi, T.; Fasola, E.; Iaia, M.; Stefani, F.; Volta, P. Limited contribution of hatchery-produced individuals to the sustainment of wild marble trout (*Salmo marmoratus* Cuvier, 1829) in an Alpine Basin. *Sci. Total Environ.* **2023**, *892*, 164555. [CrossRef]
38. Fabiani, A.; Gratton, P.; Zappes, I.A.; Seminara, M.; D'Orsi, A.; Sbordoni, V.; Allegrucci, G. Investigating the genetic structure of trout from the Garden of Ninfa (Central Italy): Suggestions for conservation and management. *Fish. Manag. Ecol.* **2018**, *25*, 1–11. [CrossRef]
39. Snoj, A.; Marčeta, B.; Sušnik, S.; Melkič, E.; Meglič, V.; Dovč, P. The taxonomic status of the sea trout from the north Adriatic Sea, as revealed by mitochondrial and nuclear DNA analysis. *J. Biogeogr.* **2002**, *29*, 1179–1185. [CrossRef]
40. Meraner, A.; Gratton, P.; Baraldi, F.; Gandolfi, A. Nothing but a trace left? Autochthony and conservation status of northern adriatic *Salmo trutta* inferred from PCR multiplexing, mtDNA control region sequencing and microsatellite analysis. *Hydrobiologia* **2013**, *702*, 201–213. [CrossRef]
41. Bernatchez, L. The evolutionary history of brown trout (*Salmo trutta* L.) inferred from phylogeographic, nested clade, and mismatch analyses of mitochondrial DNA variation. *Evolution* **2001**, *55*, 351–379. [PubMed]
42. Pustovrh, G.; Susnik Bajec, S.; Snoj, A. Evolutionary relationship between marble trout of the northern and the southern Adriatic Basin. *Mol. Phylogenetics Evol.* **2011**, *59*, 761–766. [CrossRef] [PubMed]
43. Splendiani, A.; Giovannotti, M.; Cerioni, P.N.; Caniglia, M.L.; Caputo, V. Phylogeographic inferences on the native brown trout mtDNA variation in central Italy. *Ital. J. Zool.* **2006**, *73*, 179–189. [CrossRef]

44. Meraner, A.; Gandolfi, A. Genetics of the genus *Salmo* in Italy: Evolutionary history, population structure, molecular ecology and conservation. In *Brown Trout: Biology, Ecology and Management*; Lobón-Cerviá, J., Sanz, N., Eds.; Wiley: Hoboken, NJ, USA, 2017.
45. L.C., n. 1. Modifiche Agli Articoli 9 e 41 della Costituzione in Materia di Tutela Dell'ambiente; 2022; Vol. G.U. Serie Generale n.44 del 22-02-2022. Available online: <https://www.gazzettaufficiale.it/eli/id/2022/02/22/22G00019/sg> (accessed on 5 May 2024).

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