CONSERVATION GENOMICS OF ENDANGERED ALPINE ICHTHYOFAUNA

Barbara Sofia Ilardo [a, b, *], Richard Brown [b], Hazel Nichols [c], Diego Micheletti [a], Andrea Gandolfi [a]

[a] Research and Innovation Centre, Fondazione Edmund Mach, Via E. Mach 1, 38010 San Michele all'Adige, Italy | [b] Liverpool John Moores University, Faculty of Science, School of Natural Sciences and Psychology, Liverpool, UK | [c] Swansea University, Department of Biosciences, Swansea, UK | *Corresponding author: B.S.Ilardo@2018.ljmu.ac.uk

BACKGROUND & AIM

The recent discovery of a pike species, ***Esox flaviae***[1] or ***Esox cisalpinus***[2] ("Italian pike") native to the Italian region has fuelled debate on the conservation status of this prized game fish and apex predator. **Introgressive hybridization** with allochthonous Northern pike (*E. lucius*, here "European pike") is one of the **main threats to the genetic identity of the newly described pike.**

We aim to understand the **underlying genomic adaptions** of this species to its environment, its population structure and evolutionary relationship with European pike, through a **Whole Genome Sequencing** (WGS) approach.

METHODS

Variant calling

- 19x coverage WGS data aligned to *E. lucius* genome[3]
- 28 samples[4]: Italian (16), European (6), Hybrid (6)
- GATK Best Practices[5] yielded 7.3 million SNPs

Population structure

- Principal Component Analysis
- 133k minimally-linked ($r^2 < 0.2$) SNPs
- Identifcation of clusters and exclusion of hybrids

Genome-wide selection scan

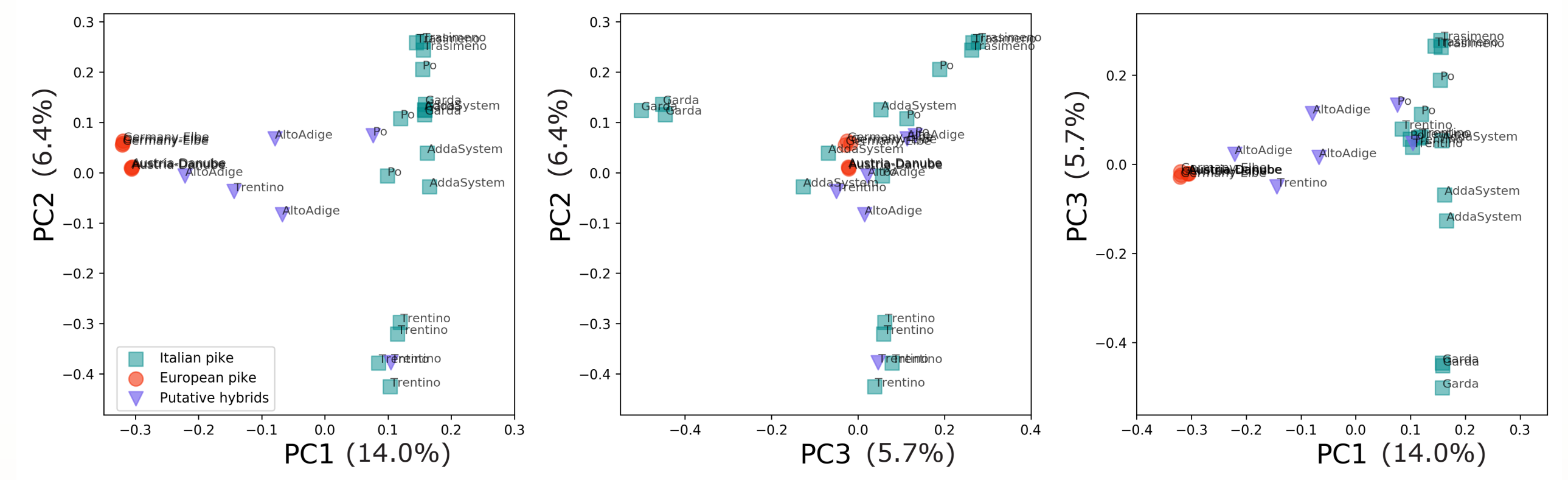
- Interspecies F_{ST} and intraspecies Tajima's D in 50 Kbp non-overlapping sliding windows
- Outliers at 99th and 1st percentiles, respectively

Gene Ontology Enrichment Analysis

- Detection of genes within 50 Kb of outlier F_{ST} regions
- GOEA of this gene set using g:Profiler[6]

RESULTS

POPULATION STRUCTURE

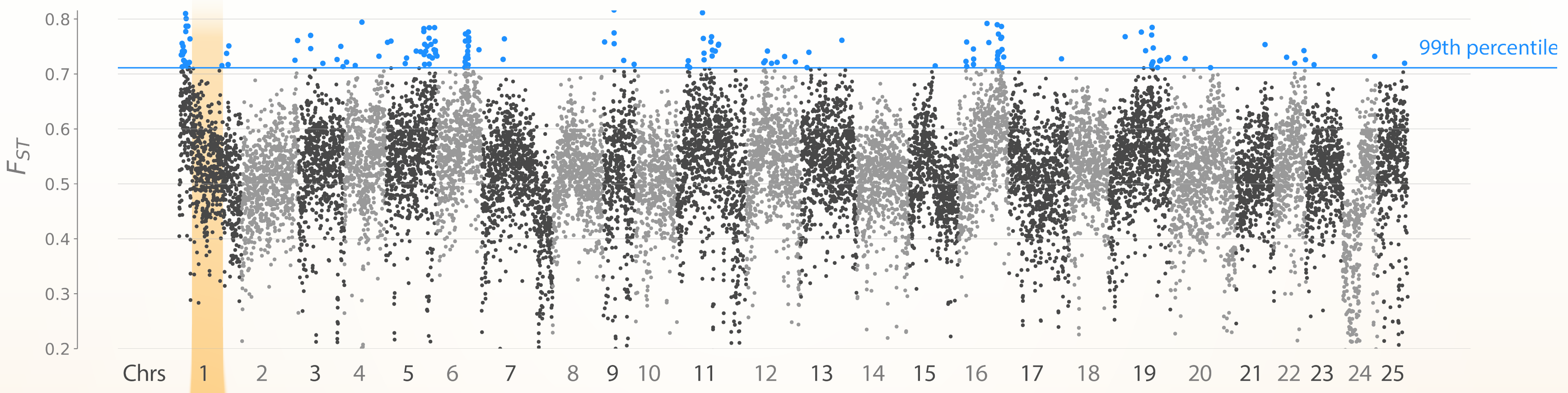


The two species are clearly differentiated in PC1 (14% of variance explained). Also, some differentiation along PC2 (6.4%) and PC3 (5.7%) is present within Italian pike subpopulations, corresponding to the different localities of the samples.

GENOME-WIDE SELECTION SCAN

Candidate chromosomal regions (blue points) under positive selection were detected between species by estimating F_{ST} values in non-overlapping 50 Kb windows and selecting outliers above the 99th percentile of the empirical distribution.

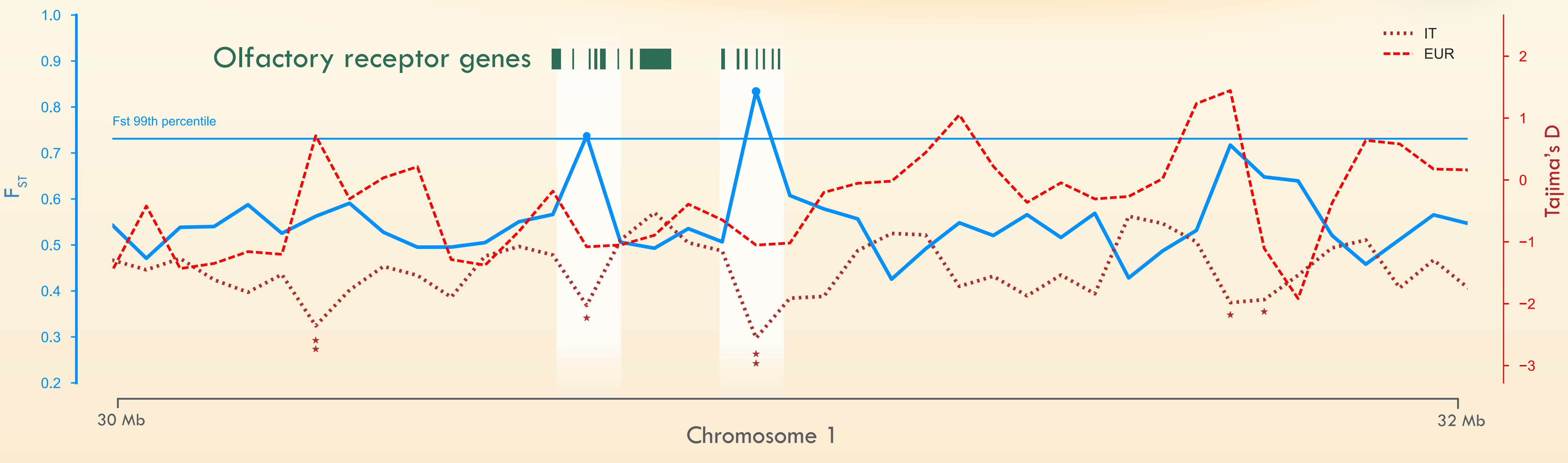
Within 50 Kb of these outlier regions, **746 candidate genes were identified.**



GO ENRICHMENT ANALYSIS

Allochthonous European pike | Endemic Italian pike

GO ID	GO TERM NAME	ADJUSTED P-VALUE
GO:0050911	Detection of chemical stimulus involved in sensory perception of smell	6.0×10^{-5}
GO:0050907	Detection of chemical stimulus involved in sensory perception	7.0×10^{-5}
GO:0007608	Sensory perception of smell	8.2×10^{-5}
GO:0009593	Detection of chemical stimulus	8.2×10^{-5}



* and ** respectively indicate Tajima's D negative outliers at the 0.5th and 1st percentiles of the genome-wide empirical distributions. Negative D values suggest positive selection.

Several functional categories are significantly enriched for **olfactory perception, all attributed to a cluster of 16 G protein-coupled odorant receptor genes on chromosome 1.** Within these regions, negative outlier Tajima's D values in Italian but not in European pike suggest positive selection acting in the endemic cisalpine species.

CONCLUSIONS

Pike, being apex ambush predators, rely greatly on their olfactory perception, perhaps more so than previously believed[7]. Our results suggest **olfactory receptors are undergoing positive selection in Italian pike,** possibly due to adaptation to local ecosystems. Because this species is threatened by introgressive hybridization with non-native European pike, used in stocking practices for angling purposes, this study informs the **need to preserve endemic biodiversity and its genomic adaptations.**

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the financial, technical and material support provided by a matched-funding scheme between Liverpool John Moores University and Fondazione Edmund Mach.

REFERENCES

1. Lucentini, Livia, et al. "Molecular and phenotypic evidence of a new species of genus Esox (Esocidae, Esociformes, Actinopterygii): the southern pike, Esox flaviae." PLoS One 6.12 (2011): e25218.

2. P.G. Bianco, G.B. Delmastro, Recenti novità tassonomiche riguardanti i pesci d'acqua dolce autoctoni in Italia e descrizione di una nuova specie di luccio, Res., Wildl. Conserv. 2 (Suppl.) (2011) (14 p.).

3. Rondeau, Eric B., et al. "The genome and linkage map of the northern pike (Esox lucius): conserved synteny revealed between the salmonid sister group and the Neoteleostei." PLoS One 9.7 (2014): e102089.

4. Gandolfi, Andrea, et al. "Population genetics of pike, genus Esox (Actinopterygii, Esocidae), in Northern Italy: evidence for mosaic distribution of native, exotic and introgressed populations." Hydrobiologia 794.1 (2017): 73-92.

5. Van der Auwera, Geraldine A., et al. "From FastQ data to high-confidence variant calls: the genome analysis toolkit best practices pipeline." Current protocols in bioinformatics 43.1 (2013): 11-10.

6. Reimand, Jüri et al. "g:Profiler--a web-based toolset for functional profiling of gene lists from large-scale experiments." Nucleic acids research vol. 35,Web Server issue (2007): W193-200. doi:10.1093/nar/gkm226

7. Raat, Alexander JP. Synopsis of biological data on the northern pike: Esox lucius Linnaeus, 1758. Food & Agriculture Org., 1988.

Caldonazzo Lake, Italy. Ph. credit www.visitvalsugana.it