Development of SNP markers to monitor genetic relationship and hybridisation in natural population of *Abies nebrodensis*

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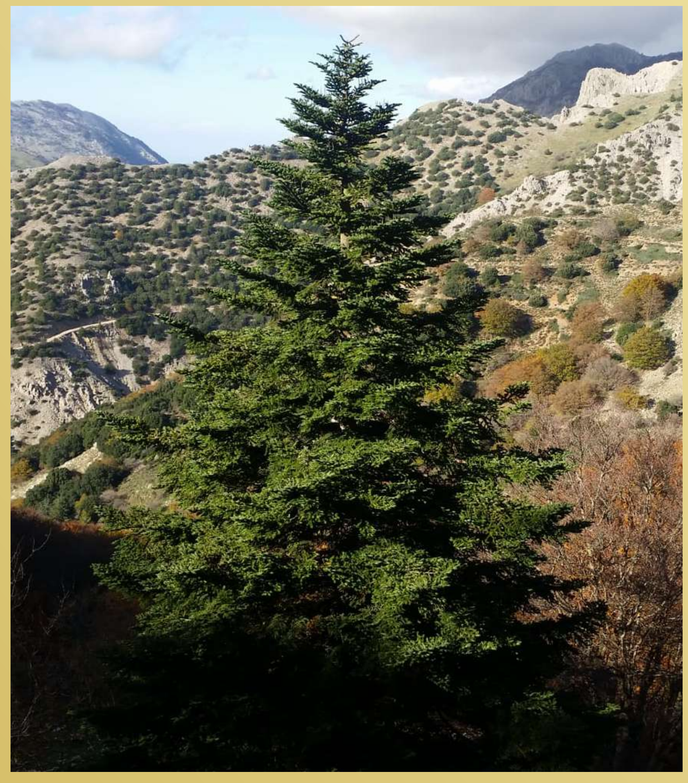

Fig. 1. Adult tree of *Abies nebrodensis* from the Madonie Park (Sicily).

Fig. 2. Young A. *nebrodensis* tree of the natural regeneration, growing in the Madonie Park.

Fig. 3. Seedling coming from different mothers, from the local nursery “Vivaio Piano Noce” (Madonie Park).

INTRODUCTION

Abies nebrodensis is an endemic species to the north–central part of Sicily (Fig.1). The Sicilian/Nebrodi fir is classified as critically endangered by the IUCN Red List. According to recent estimates, this species is limited to a unique relict population that harbors 30 adult trees and a fluctuating number of juveniles derived from natural regeneration (170 according to the last census) (Fig. 2); besides, some thousands of cultivated seedling are preserved as *ex situ* collection (Fig. 3). Hybridization between *A. nebrodensis* and the closely related *A. alba* and *A. cephalonica* is one of the most important concerns in the conservation of this endangered fir, and conservation authorities suspect the hybrid origin of some seedlings in the natural population.

We used restriction site associated DNA sequencing (RAD-seq) to identify high-quality and information-rich SNPs in samples of *A. nebrodensis*, *A. alba* and *A. cephalonica*. We developed a set of SNP-array for genotyping of *A. nebrodensis* adults and juveniles. This SNP panel will be tested to, (1) evaluate the variability and degree of genetic relationship among the adult mature plants of the original population, (2) determine the rate of outcrossing, inbreeding and self-fertilization and (3) assess the eventual hybridization due to pollen coming from non-native *Abies* species planted in the park (*A. alba* and *A. cephalonica*).

MATERIAL & METHODS

We built a bioinformatic pipeline (Fig. 4) using:

Stacks PLINK2 adegenet 2.1.0 SnpEff

RESULTS

We finally selected:

- 20 high quality SNPs for the **hybridization test** between *A. nebrodensis* and the other two species (Fig. 5).
- 100 high quality SNPs were developed for the **paternity analysis** using just **intergenic regions** (Fig. 6) to reduce selection bias on subsequent analyzes.

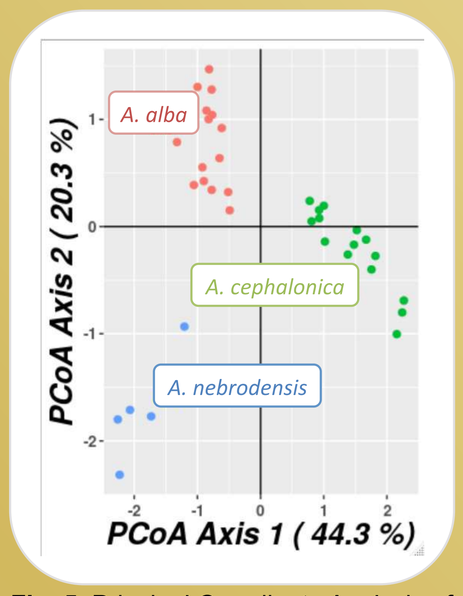

Fig. 5. Principal Coordinate Analysis of *Abies* samples using Euclidean distance from the 20 selected SNPs.

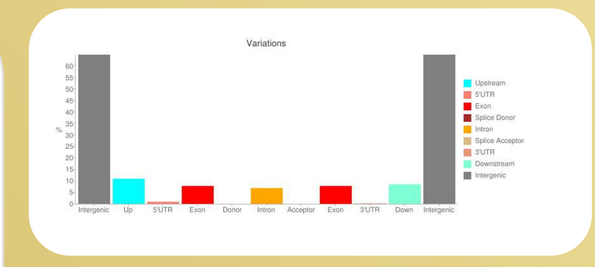

Fig. 6. Frequency of SNPs by genomic regions.

- OpenArray primers will be developed for the selected SNPs.
- We will perform a paternity test using 30 adult mature plants, 118 juveniles plants from natural regeneration and 2060 seedlings at a nursery.

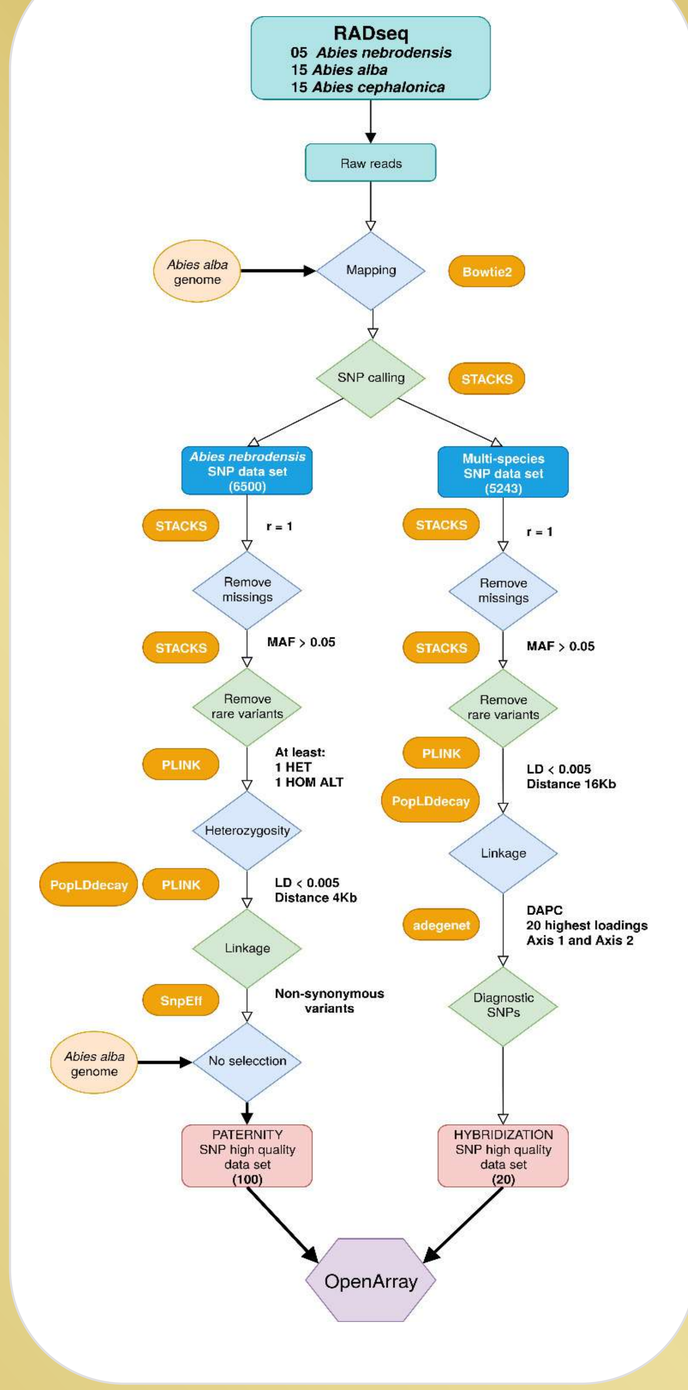

Fig. 4. Flowchart of SNP selection pipeline.

ACKNOWLEDGMENTS

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