Genetic Diversity and Management of Black Pine (*Pinus nigra*) habitat of Mount Athos

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Introduction

Black pine (*Pinus nigra* J.F. Arnold subsp. nigra) is a shade-tolerant species, which can grow in dry and arid stations, regardless of the subsoil rock, and has a characteristic adaptability to soils with high metal concentrations.
Due to its great geographical diversity and colonization of extreme sites, the species is classified as a priority habitat and is protected according to the European Directive 92/43, under "Priority habitat type of (Sub)-Mediterranean pine forests with endemic black pines (9530*)".
In Mount Athos (MA) Black pine occurs scattered in the central part of the peninsula, along the ridge of MA. Usually, it is found either as dispersed individual trees or in pure / mixed clusters and groups of trees.
Management actions aim at the conservation of Natura 2000 habitat, favor its regeneration through treatments assisting the establishment of seedlings and the spread of the species in areas where it demonstrates sporadic occurrence.
The assessment of genetic diversity of black pine population had the objectives: to find out the origin of the black pine population in MA, determine differences form other populations in NE Greece, assess if genetic diversity is enough for a viable population, discover indicators of genetic erosion and derive conservation priorities and suggested management measures.

Methodology

Sampling (needles collection)
- Representative for most MA occurrences
- Samples from Sithonia, Thassos and Perama (Evros)

Analysis of 7 genetic loci
- DNA extraction
- Polymerase Chain Reaction (PCR) with nuclear SSR primers
- Fragment Analysis
- Allele binning and Null Allele Adjustment

Data Analysis
- Diversity and Differentiation, PCA, DAPC, spatial PCA, MCMC
- Geospatial Analysis (QGIS)

Results

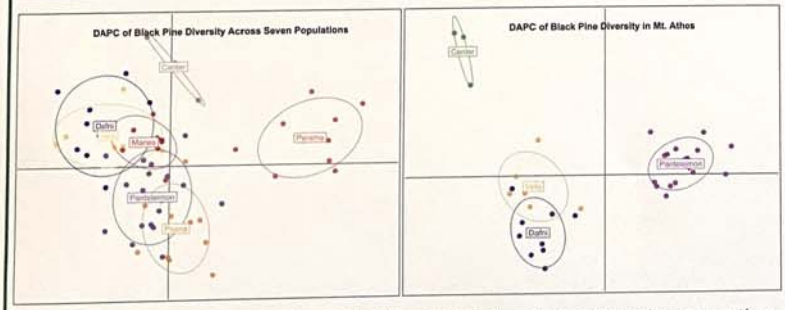
	Population	Mean (SE)		
		Ho	He	F value
Mount Athos	Dafni	0.499 (0.071)	0.662 (0.050)	0.217 (0.126)
	Panteleimon	0.471 (0.095)	0.667 (0.096)	0.373 (0.130)
	Center	0.405 (0.170)	0.446 (0.118)	0.054 (0.292)
	Vela	0.571 (0.093)	0.614 (0.052)	0.093 (0.052)
Sithonia	Psaria	0.436 (0.078)	0.666 (0.075)	0.375 (0.076)
Thrace	Perama	0.388 (0.149)	0.485 (0.128)	0.215 (0.188)
Thasos	Maries	0.502 (0.139)	0.628 (0.077)	0.301 (0.185)
	Total	0.467 (0.043)	0.596 (0.034)	0.241 (0.060)

Genetic diversity (Observed Heterozygosity - Ho, Expected Heterozygosity - He) was relatively low in all populations of black pine in NE Greece, including Mt. Athos subpopulations, indicating limited genetic variability within the local groups of trees.

All populations of NE Greece demonstrated a heterozygote deficit and an increased inbreeding index (F).

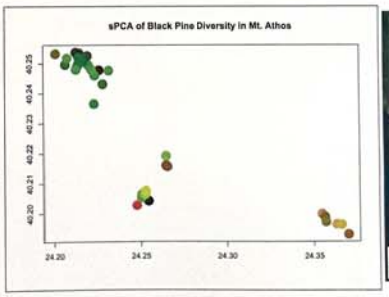
Black pine populations of Mt. Athos demonstrate similar levels of diversity and inbreeding with the populations used in this comparison, possibly because all of them are small, isolated and fragmented.

The results suggest a moderate genetic isolation among the black pine groups in Athos, supporting the hypothesis of natural origin. Planted trees usually exhibit striking genetic differentiation, a trend which was not observed in our data.



The Discriminant Analysis plot shows that the subpopulations of Athos cluster together with the populations in Sithonia and Thasos, while Perama is genetically more distant.

The subpopulations of Mt. Athos are not genetically uniform.



The spatial PCA plot and the relative map demonstrate that the subpopulations of Mt. Athos are not genetically uniform.

Conclusions

- Indication of low genetic diversity and increased inbreeding, possibly due to fragmentation in Mt. Athos.
- Increased risk of genetic erosion and population loss.
- Clear differentiation pattern among subpopulations in Mt. Athos confirm genetic fragmentation.
- Patterns of diversity and differentiation suggest a natural origin of the population.
- Possible fragmentation of an initially larger population and several subsequent founder effects due to the ability of black pine to colonize degraded areas.

Management recommendations

In situ management for the preservation of patches and small populations of black pine
- Silvicultural measures to favor species establishment and regeneration

Restoration activities
- Seed collection and growing of seedlings of local origin in nurseries in Mt. Athos or NE Greece
- Planting of local seedlings in selected areas to increase population density
- Enriching genetic diversity (introduction of other black pine provenances from N. Greece, at a small scale and in small numbers, in locations away from the patches of the local black pine forests)

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