



PATTERNS OF GENETIC DIVERSITY IN WILD AND CULTIVATED POPULATIONS OF DALMATIAN SAGE (*SALVIA OFFICINALIS* L., LAMIACEAE)

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Dalmatian sage

Distribution:

- coastal region of the western Balkan and central and southern Apennine Peninsulas

Cultivation:

- throughout the Mediterranean region, in Australia, Germany, USA etc.

Naturalized populations:

- plants that have escaped from earlier cultivation
(throughout the Mediterranean region)

Gathering from the wild

- negative impact on biodiversity



Aims of research:

- to assess levels of genetic diversity of natural populations compared to planted populations
- to discuss the extent of genetic diversity reduction occurring in naturalized and cultivated populations
- to provide insight into the phylogeographic history of wild populations



SAMPLING

30 populations

Slovenia (2)

Croatia (8)

BiH (4)

Montenegro (2)

Albania (2)

Macedonia (2)

Greece (2)

Kosovo (2)

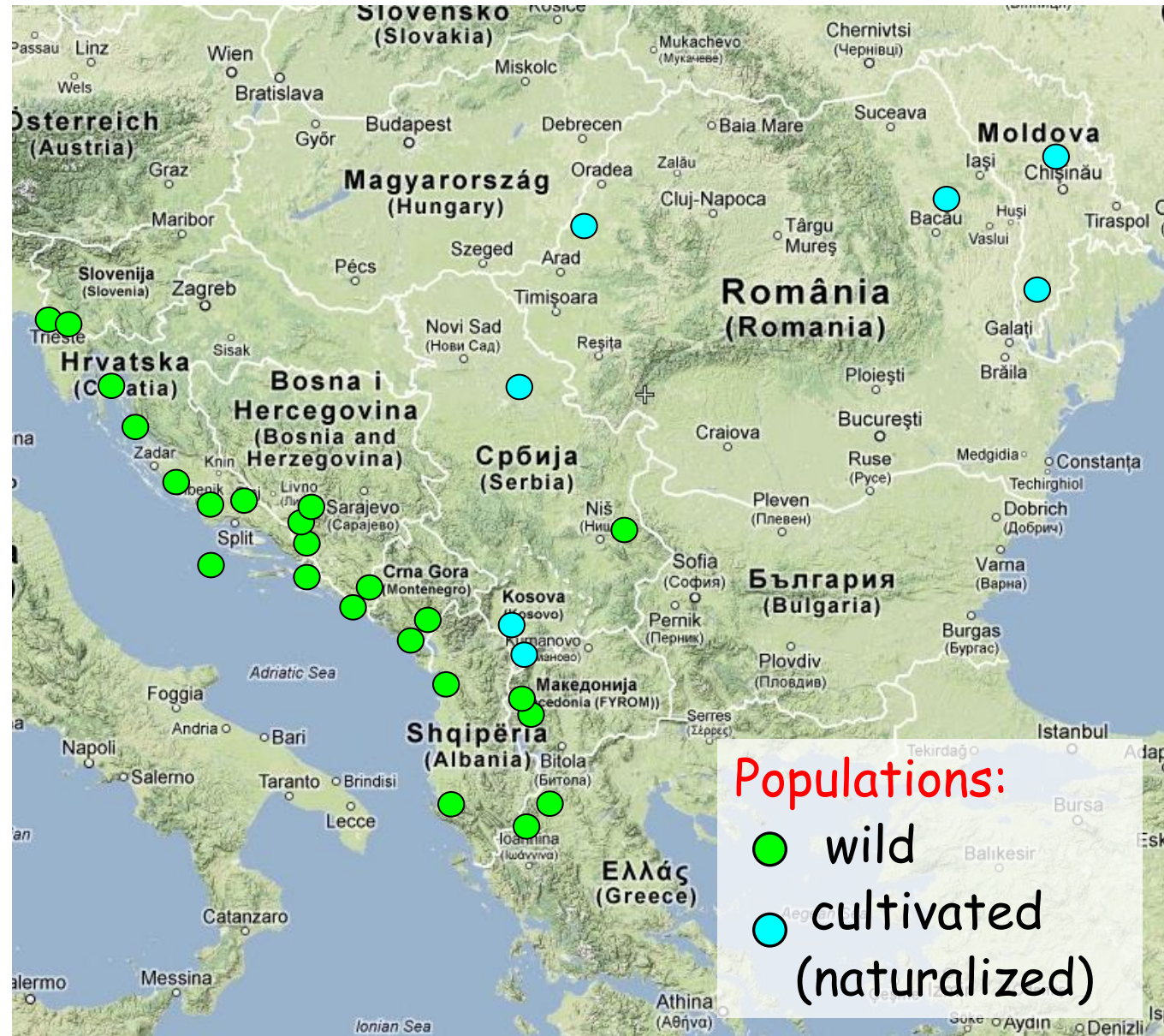
Serbia (2)

Romania (2)

Moldova (2)

cca. 25 plants per
population

709 samples



Microsatellite analysis

Molecular markers:

8 microsatellite loci

165 alleles (average: 20.63 alleles per locus)

Data analyses:

Genetic distance / Cluster analysis

Allelic richness

Hardy-Weinberg equilibrium: H_O , H_E , F_{IS}

Bottleneck

Isolation by distance

Structure

DALMATIAN SAGE

	Wild	Cultivated
No.	23	7
N_{av}	8.696	3.821
N_{ar}	7.920	3.672
Range	5.13-10.30	2.71-4.19
$P(N_{ar})$	P < 0.001	
N_{pr} (total per population)	20	0
N_{pr} (wild vs. cultivated)	115	0

No.	Population	n	N_{av}	N_{ar}	N_{pr}
P01	SVN-1	23	8.750	7.930	0
P02	SVN-2	24	9.125	8.258	0
P03	HRV-1	25	8.500	7.708	0
P04	HRV-2	25	8.750	7.948	1
P05	HRV-3	24	8.000	7.107	0
P06	HRV-4	24	9.875	9.071	1
P07	HRV-5	25	10.000	8.990	1
P08	HRV-6	24	7.125	6.655	0
P09	HRV-7	25	11.250	9.949	2
P10	HRV-8	25	11.375	10.300	0
P11	BIH-1	24	8.000	7.372	2
P12	BIH-2	25	10.625	9.709	1
P13	BIH-3	25	10.500	9.471	1
P14	BIH-4	22	10.625	9.847	2
P15	MNE-1	24	7.375	6.721	0
P16	MNE-2	22	8.375	7.651	1
P17	ALB-1	24	9.375	8.633	4
P18	ALB-2	23	9.625	8.694	0
P19	MKD-1	24	7.250	6.458	2
P20	MKD-2	22	6.625	6.210	1
P21	GRC-1	24	6.375	5.752	0
P22	GRC-2	22	7.000	6.600	1
P23	KOS-1	23	4.250	4.089	0
P24	KOS-2	24	4.375	4.076	0
P25	SRB-1	20	2.750	2.707	0
P26	SRB-2	24	5.500	5.134	0
P27	ROU-1	24	4.250	4.189	0
P28	ROU-2	22	3.750	3.639	0
P29	MDA-1	23	4.125	3.886	0
P30	MDA-2	24	3.250	3.121	0
Mean Total		23.63 709	7.558	6.929	20

DALMATIAN SAGE: ALLELIC RICHNESS

$N_{ar} > 9$
Southern
Dalmatia
(HRV)
Herzegovina
(BiH)

$N_{ar} < 5$
cultivated
populations
only



DALMATIAN SAGE

	Wild	Cultivated
No.	23	7
H_O	0.733	0.590
Range	0.54-0.85	0.31-0.79
$P(H_O)$	$P < 0.001$	
H_E	0.751	0.545
Range	0.61-0.85	0.38-0.61
$P(H_E)$	$P < 0.001$	

- two populations exhibited significant deviations from HWE
- cultivated populations from Romania (ROU-1, ROU-2) show heterozygote excess
- artificial selection favouring heterozygotes?

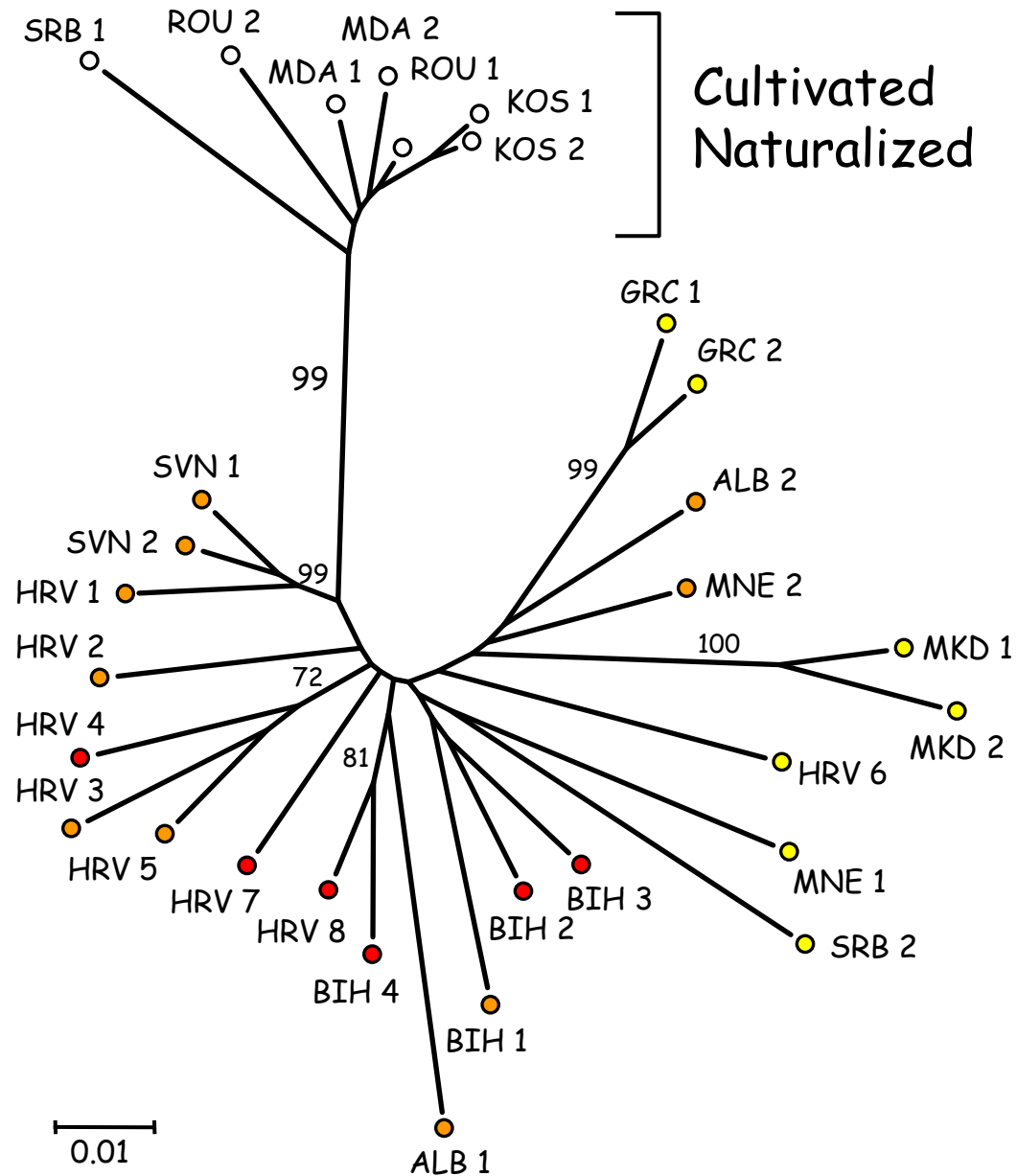
No.	Population	H_O	H_E	F_{IS}	
P01	SVN-1	0.745	0.795	0.063	ns
P02	SVN-2	0.854	0.795	-0.075	ns
P03	HRV-1	0.717	0.734	0.024	ns
P04	HRV-2	0.754	0.759	0.008	ns
P05	HRV-3	0.707	0.708	0.002	ns
P06	HRV-4	0.698	0.743	0.061	ns
P07	HRV-5	0.736	0.771	0.045	ns
P08	HRV-6	0.717	0.712	-0.007	ns
P09	HRV-7	0.754	0.769	0.019	ns
P10	HRV-8	0.825	0.847	0.026	ns
P11	BIH-1	0.817	0.758	-0.077	ns
P12	BIH-2	0.762	0.796	0.043	ns
P13	BIH-3	0.818	0.815	-0.004	ns
P14	BIH-4	0.777	0.833	0.067	ns
P15	MNE-1	0.693	0.745	0.070	ns
P16	MNE-2	0.699	0.718	0.026	ns
P17	ALB-1	0.744	0.766	0.029	ns
P18	ALB-2	0.743	0.802	0.073	ns
P19	MKD-1	0.683	0.699	0.024	ns
P20	MKD-2	0.720	0.725	0.007	ns
P21	GRC-1	0.679	0.673	-0.009	ns
P22	GRC-2	0.672	0.702	0.042	ns
P23	KOS-1	0.598	0.561	-0.067	ns
P24	KOS-2	0.505	0.561	0.100	ns
P25	SRB-1	0.313	0.377	0.171	ns
P26	SRB-2	0.537	0.612	0.123	ns
P27	ROU-1	0.783	0.610	-0.283	***
P28	ROU-2	0.793	0.612	-0.296	***
P29	MDA-1	0.592	0.595	0.004	ns
P30	MDA-2	0.545	0.497	-0.096	ns
Mean		0.699	0.703	0.004	

DALMATIAN SAGE: PAIRWISE F_{ST} VALUES

- average pairwise F_{ST} : 0.153
- minimum F_{ST} : 0.009
 - between two adjacent wild populations from Slovenia (P01/P02)
- maximum F_{ST} : 0.414
 - between the cultivated (P25) and the wild (P26) populations from Serbia
- all but three pairwise F_{ST} values are significant ($P < 0.05$)
 - significant population differentiation
- non-significant population pairs
 - P01/P02 two adjacent wild populations from Slovenia
 - P10/P14 two adjacent wild populations: one from Croatia and the other from Bosnia and Herzegovina
 - P23/P24 two adjacent naturalized populations from Kosovo

DALMATIAN SAGE

- wild populations grouped together in accordance with geographical position of the collecting sites, from Slovenia in the North-West of the region to Greece in the South-East
- seven cultivated populations grouped separately from the rest and formed a well-supported clade (bootstrap support 99%) suggesting the common origin of the cultivated plant material
- population SRB-2 from Sićevo gorge is undoubtedly indigenous

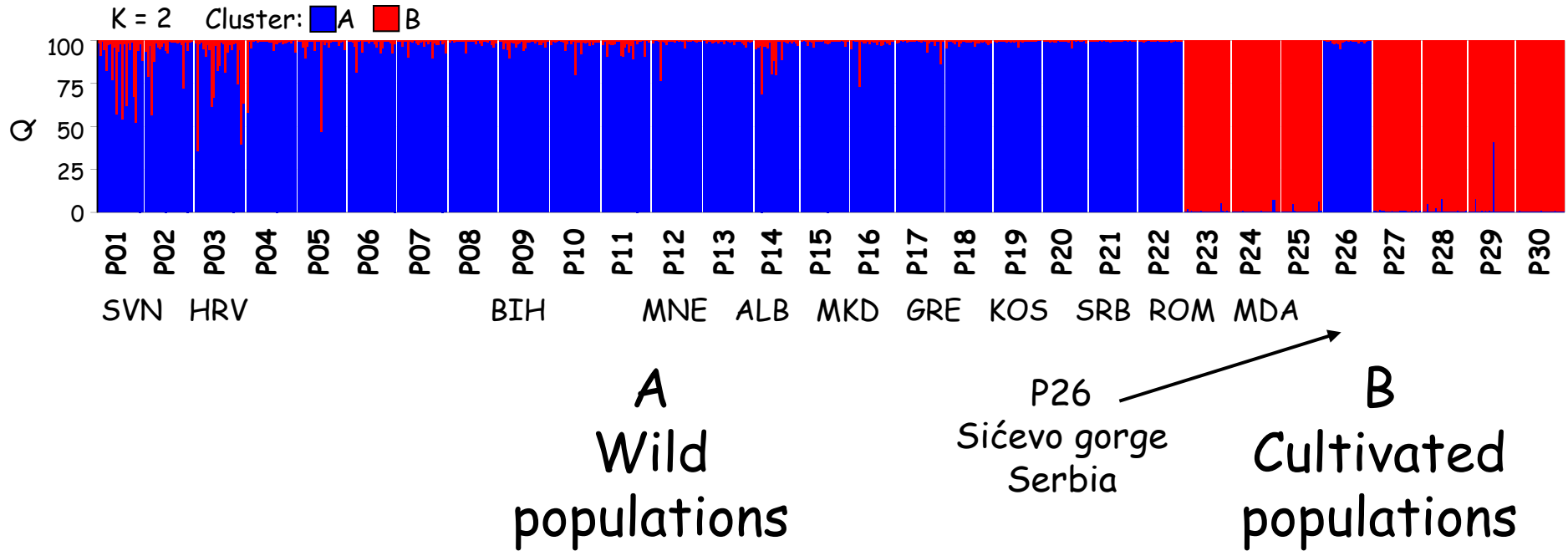


DALMATIAN SAGE: AMOVA

Source of variation	df	Variance components	% Total variance	ϕ -statistics	P(ϕ)
Between groups (wild vs. cultivated)	1	σ_a^2 : 0.419	11.94	ϕ_{CT} : 0.119	< 0.001
Among populations within groups	28	σ_b^2 : 0.342	9.77	ϕ_{SC} : 0.111	< 0.001
Among individuals within populations	1388	σ_c^2 : 2.744	78.29	ϕ_{ST} : 0.217	< 0.001

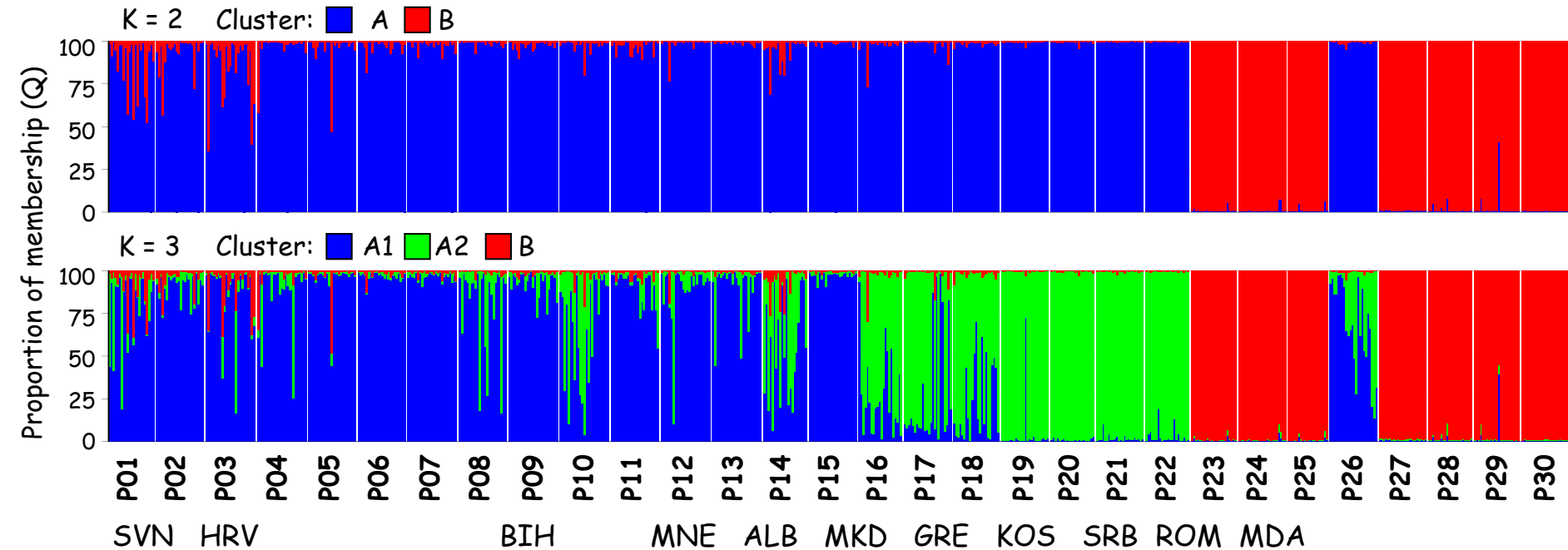
- the most of total genetic diversity was attributable to differences among individuals within populations
- overall structure is highly significant
- both sources of variations are highly significant
 - populations differ within groups
 - groups (wild vs. cultivated) are clearly distinct

DALMATIAN SAGE: STRUCTURE AT K = 2



- proportions of membership (Q) of each individual plant in each of the two clusters:
 - each individual plant is represented by a single vertical line divided into colours representing different clusters
 - the length of the coloured segment shows the individual's estimated proportion of membership in that cluster

DALMATIAN SAGE: STRUCTURE AT K = 3



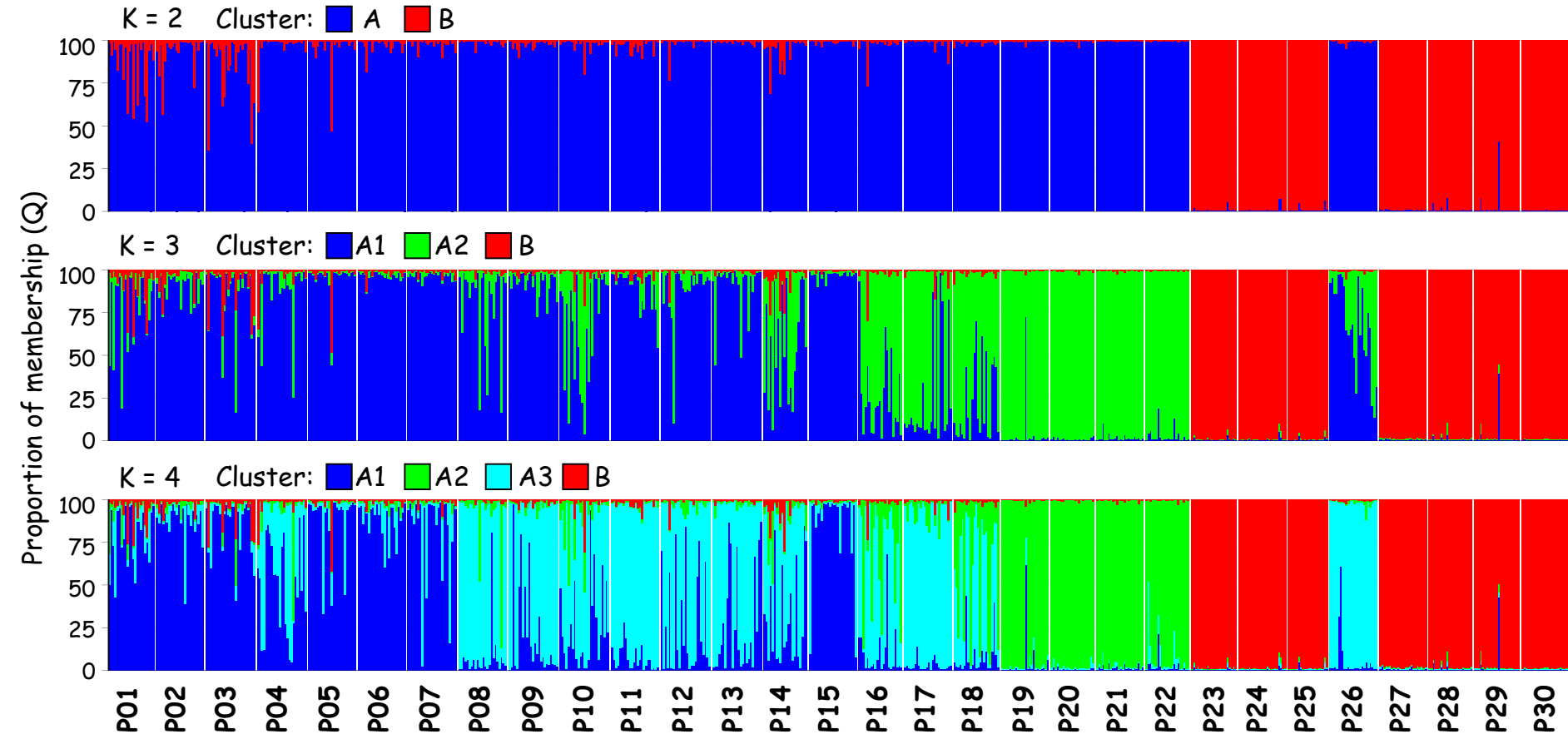
A1
North-West
populations

A2
South-East
populations

B
Cultivated
populations

- the cluster A (Wild populations) is at K = 3 split into two clusters according to geographical locations
- A1 North-West: Adriatic populations**
- A2 South-East: Montenegro-Albania-Macedonia-Greece**

DALMATIAN SAGE: STRUCTURE AT K = 4



A1
Nothorn
Adriatic

A3
Southern
Adriatic

A2
Macedonia
Greece

B
Cultivated
populations

- wild populations split further into three subclusters

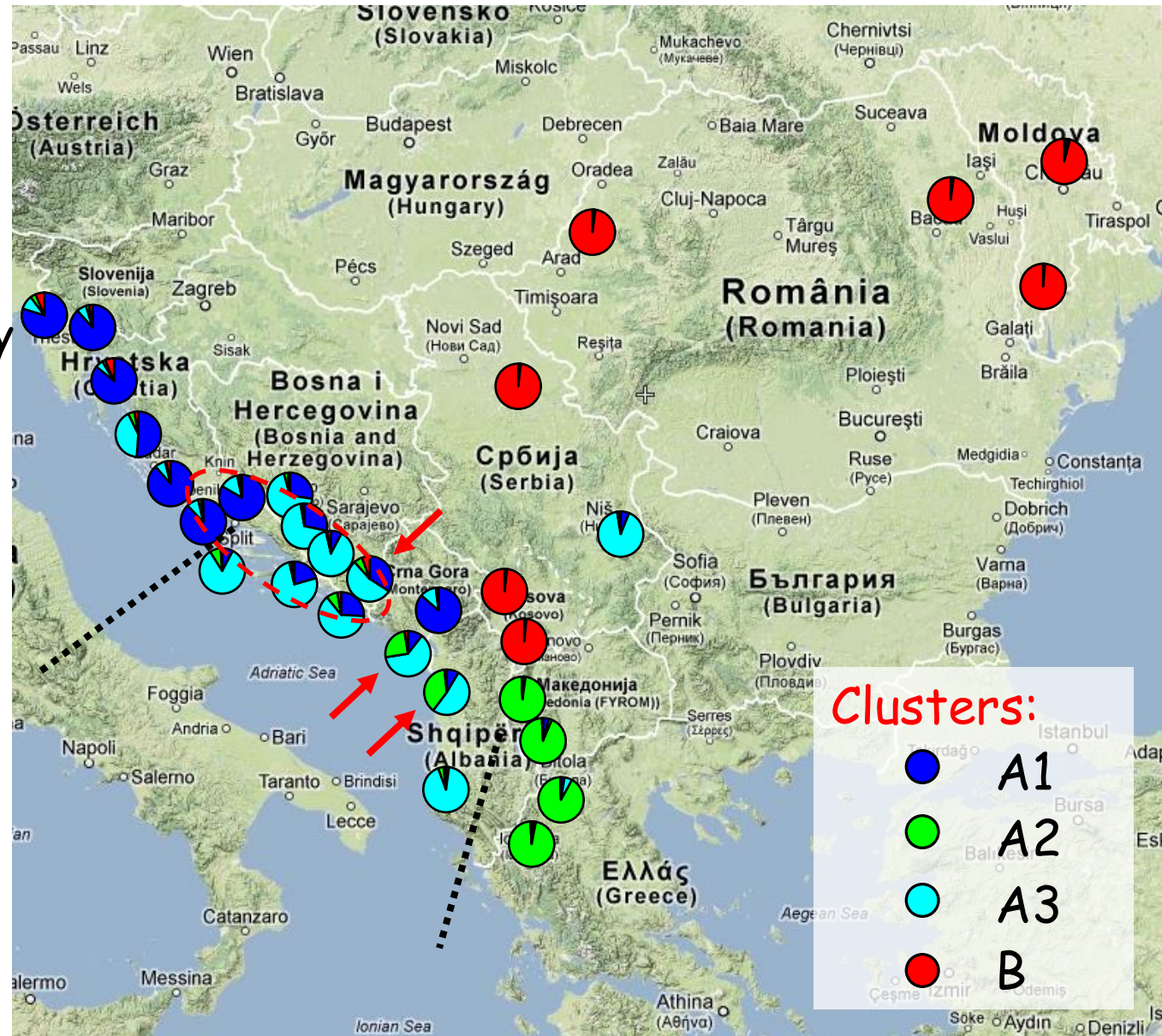
DALMATIAN SAGE: AVERAGE Q AT K = 4

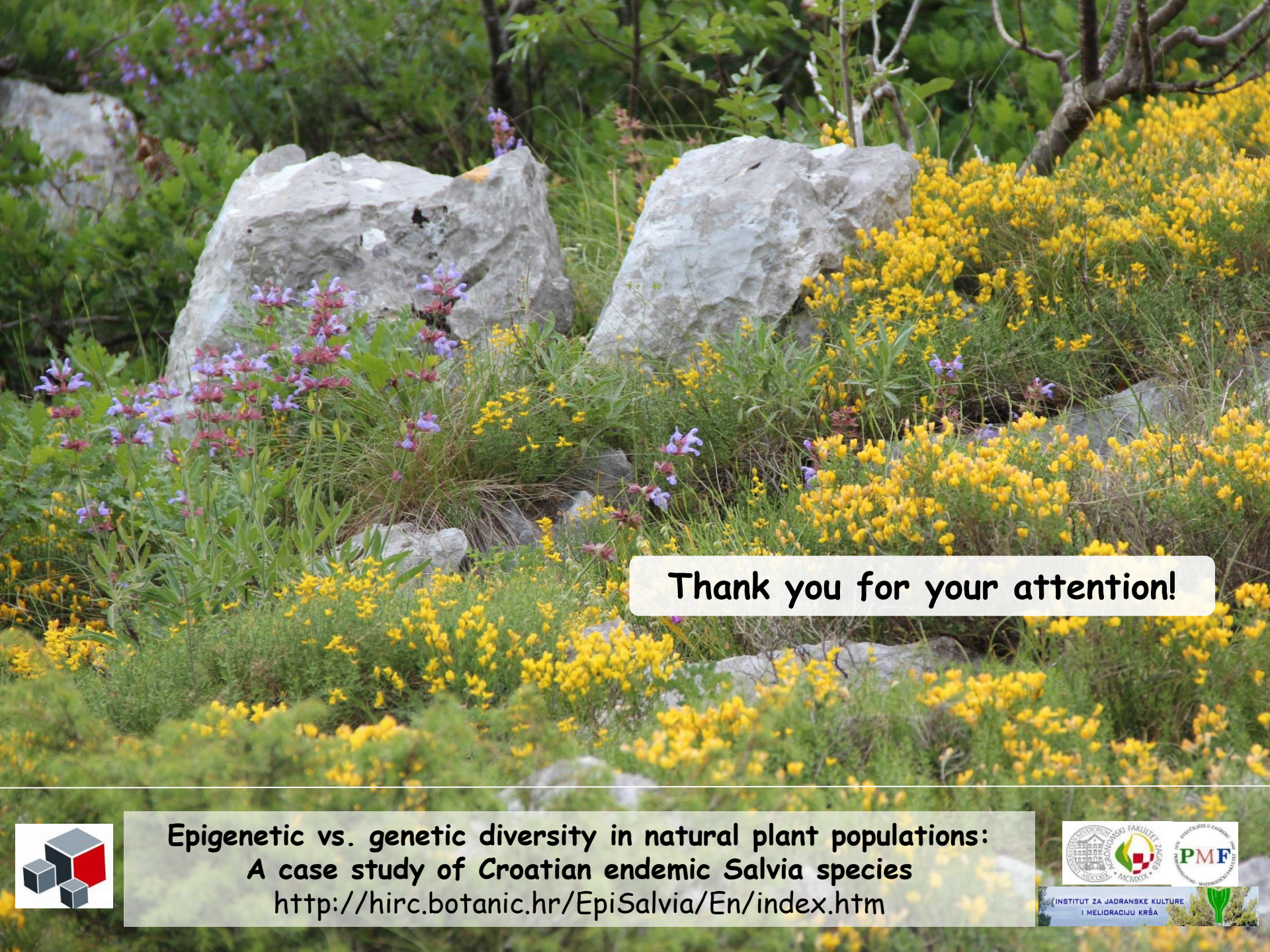
The most admixed populations:

→ BiH-4
ALB-2
MNE-2

The highest diversity (N_{ar} , H_E):
the populations belonging to cluster A3

Possible origin of cultivated populations:
Slovenia, N. Croatia (Istrian peninsula)





Thank you for your attention!



**Epigenetic vs. genetic diversity in natural plant populations:
A case study of Croatian endemic *Salvia* species**
<http://hirc.botanic.hr/EpiSalvia/En/index.htm>

