Microsatellite evidence for high clonality in narrow endemic and endangered *Salvia brachyodon* (Lamiaceae): implications for management and conservation

I. Radosavljević, Z. Šatović, Z. Liber
Salvia brachyodon Vandas – short-toothed sage

- Family: Lamiaceae
- Subfamily: Nepetoideae
- Tribus: Mentheae
- Genus: Salvia
- Section: Salvia
  
  (30-40 species – Mediterranean, Irano-Turanian region)
Distribution range
- eastern Adriatic coastal region
- three known populations: Pelješac peninsula (1), Konavle (2) and Mt. Orjen (3)
Earlier research carried out in 2010 suggested the possibility of clonal reproduction in *S. brachyodon*. Due to lack of any prior knowledge concerning clonality in the species, the sample size was too small and the sampling strategy was not appropriate for genetic analysis of clonal organisms.

Aims of the research:
- to assess the levels of genetic diversity and clonality within the population od *S. brachyodon* from Pelješac peninsula.
- to assess the levels of vulnerability of *S. brachyodon* population and to discuss possible conservation activities.
Pelješac peninsula
- ~ 900 m.a.s.l.
- the largest population
  - few thousands individuals
  - ~ 5000-6000 m²
Sampling
- 5 plots, each 5 x 5 m
- the exact position for each plot was determined based on a number of available plants
- 120 specimens (22-26/plot)

Genetic analysis

Eight microsatellite markers:

<table>
<thead>
<tr>
<th>Locus</th>
<th>GeneBank No.</th>
<th>Repeat Motif</th>
</tr>
</thead>
<tbody>
<tr>
<td>SoUZ001</td>
<td>GQ376512</td>
<td>(AG)$_{15}$</td>
</tr>
<tr>
<td>SoUZ002</td>
<td>GQ376513</td>
<td>(TG)$_{11}$</td>
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<td>(AGA)$_{10}$</td>
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</tbody>
</table>
Individual plants are grouped into patches!
The exact location of each plant was geocoded.
PLOTS 4 AND 5
RESULTS

An unrooted Fitch-Margoliash tree based on the proportion-of-shared-allele distances between *Salvia brachyodon* samples. Bootstrap support values higher than 50% base on 1,000 replicates are indicated on the branches.

Ramets/genets:

[H$_O$ = 0.708, H$_E$ = 0.699]
Total number of genets: 36

Percentage of the samples belonging to the most frequent clone: 18.5 %

Clonal subrange: 11.63 m
PLOTS 4 AND 5

Ramets/genets:
- Red = 22
- Green = 8
- Blue = 4
- Pink = 3
- Cyan = 2
- Green = 2
- Brown = 2
- Orange = 2

Scale: 1 m
DISCUSSION

- clonal propagation is achieved by underground stolons
- high no. of alleles / high gene diversity: unusual but not unprecedented in narrow endemic species
- Hardy-Weinberg equilibrium: sexual reproduction is also present
- clonality helps in maintaining high levels of observed heterozygosity: new mutations can not be lost through genetic drift.
- **S. brachyodon** population is not in immediate threat.

- being a typical heliophyte, **S. brachyodon** requires clean, open space to thrive.

- the abandonment of the traditional pasture management has a loss of a suitable open habitats (i.e. Mediterranean grassland) for a consequence.

- ecological succession towards the indigenous black pine (**Pinus nigra** Arnold) forest throughout the region is inevitable!

- to protect **S. brachyodon**, habitat management is needed - open habitat must be maintained!

- agro-pastoral system that was once present in this location, should be encouraged and re-established.

- currently, **S. brachyodon** thrives in Pelješac peninsula. Why?........
…thanks to ‘devastating’ wildfire that overran St. Ilija in 1998 and cleared the entire area!
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