

Final report

The two year studies of the *Origanum vulgare* (common marjoram) and *Hyssopus officinalis* (hyssop) species, referring to the existence of genetic polymorphism, revealed the following:

- The methods of quantitative and qualitative phytochemical analysis are thin layer chromatography (TLC), spectrophotometry, high performance liquid chromatography (HPLC) and gas-chromatography coupled with mass spectroscopy (GC-MS).
- To determine the genetic polymorphism, we used the RAPD-PCR amplification technique with randomized primers.
- The morphological analysis on marjoram plants harvested near Orheiul Vechi is not determining for the biosynthetic capacity of the active principles (polyphenols, flavons, volatile oils).
- The populational studies of the marjoram plants, harvested in Romania and in the Republic of Moldova, to highlight the chemical variability, showed a restricted intraspecific variability; the quantities of rosmarinic acid are similar in all the studied plants.
- The compounds with antioxidant action from the polyphenols class are the rosmarinic, caffeic and chlorogenic acids, and from the flavonoids class, rutoside, luteoline and apigenine.
- The composition of the volatile oil was determined by gass-chromatographical analysis, which highlighted the existence of the same volatile fractions for *Origanum vulgare*, prelevated from the populations in Romania and those from the Republic of Moldova.
- The genetic analysis, coupled with aphytochemical one, confirmed the existence of a **chemotype** that synthesizes compounds of the type of *sabinen/4(10)-thijuene*, *trans- β -ocimene*, *γ -terpinen*, *β -caryiophyllene*, *germacrene d* and not of the *carvachrol* and *thymol* types in the quantities stipulated by the European Pharmacopeaea.
- The pedoclimatic conditions influence the quantity of the active principles that are synthesized by the marjoram plants and not the spectrum (quality) of the determined compounds.

- The chemotype identified by the studies on the *Origanum vulgare* species may be used as a furnisher of raw material for the units interested to use it in cosmetics, food industry, aromatherapy but not as an adjuvant in phototherapy.
- The clusterization of the volatile oil compounds from 7 *Origanum vulgare* genotypes, showed three groups, dependent on the genotype. Likewise, we proved, by the RAPD method, that three groups performed genetic polymorphism clusterization. As such, the RAPD technique may be used to **identify chemotypes / chemovarieties**, that could be used in the studies of biodiversity preservation, phylogenetic analysis, as well as in the researches of medicinal plant selection and melioration.
- The chromatographic profiles of the volatile oil content and the molecular markers from *Origanum.vulgare*, favoured the intrapopulation, interpopulation and interspecific discrimination.
- The highest molecular polymorphism in *Origanum vulgare ssp. vulgare* was revealed by the RAPD-PCR analysis with the primers OPB10, OPG10 and UBC215.
- We collected and characterized samples of *Hyssopus officinalis* from conventional cultures in the Republic of Moldova and Romania. We studied their intraspecific diversity by the implication of the techniques of molecular analysis (RAPD-PCR), noticing the effectiveness of using RAPD primers in the appreciation of the intra- and interpopulation polymorphism.
- The GC-MS analysis of the hyssop oil confirmed the presence of the two ketones **pinocamphon** and **isopinocamphon** in variable limits depending on the colours of the flowers and on the origin from 42.68% to 57.18%.
- Referring to the safety of the volatile oil administration, the presence of the two ketones in high quantities does not affect the pharmaceutical quality in case the hyssop may be used in associated formulas of teas.
- The investigations on *Hyssopus officinalis* show a great variety of volatile fractions depending on the colour of the flowers, and especially on the pedoclimatic conditions of the harvesting habitat. It seems that for this species one cannot assert for sure that there might exist a **chemotype** but only a **chemovariety** depending on the pedoclimatic conditions.

- We highlighted unique strips present in all the individuals of a hyssop population and their absence in the individuals of other populations – potential markers to be used for the identification of the population.
- We noticed a slight correlation of the genetic similarity degree of the samples with their geographical location. Thus, the average value of the similarities among the genotypes of the Romanian populations was of 0.580 while the similarity degree between the Romanian genotypes and those from the Republic of Moldova was 0.556.
- Further studies on the *Hyssopus officinalis* species will contribute to the elucidation of all the aspects linked to the existence of a chemotype or will confirm the existence of chemovarieties.