MOLECULAR MARKERS APPLICATION FOR GENETIC RESOURCES
CHARACTERIZATION OF DIFFERENT PLANT SPECIES

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Summary
Results on different genetic markers application, including proteins and DNA, to characterize genome structure and diversity of maize, beans, soybean red pepper, pine and pine tree will be presented. Dendograms for different species genotypes, based on cluster analysis (UPGMA) of genetic distances from both protein and DNA markers indicated that molecular markers are suited to measure the average level of relatedness and to identify closely related genotypes.

Introduction
Plant genetic resources include the reproductive or vegetative propagated material of (i) cultivars in current use and newly developed varieties, (ii) traditional cultivars and landraces (iii) wild relatives of cultivated species and (iv) elite breeding materials, aneuploids and mutants. Possible application of molecular markers in germplasm collections are: identification and verification of old and new collected genotypes; detection of duplicates; genetic purity analysis; genetic diversity analysis; construction of „core collection“ and selection of interesting, gene resources; monitoring of viability and health and genetic changes due to long-term storage at low temperature. In this study the particular goal was to confirm that genetic markers, as proteins or DNA, are useful and are reliable indicators of genome structure and function in evolutionary different plant species including maize, beans, red pepper and soybeans.

Material and methods
Embryo tissue from mature seeds has been used for genetic divergence characterization of different maize populations, inbred lines and hybrids, different accession from the species Capsicum annuum L., several soybean genotypes, parental individuals and several hybrid combinations of Serbian spruce (Picea omorika/Panč./Purkyine) and Pinus nigra arn as well. Divergence of Pinus nigra arn has been analyzed also at the salt soluble protein level extracted from young seedlings. Characterization has been done either by application of PCR based DNA molecular markers including SSR, RAPD, AFLP and RFLP or salt soluble seed proteins or both. Salt soluble proteins have been separated by PAGE according to the molecular weight. Dendograms have been constructed using unweighted pair – group method with arithmetical averages (UPGMA) and computer program NTSYS – pc.

Results and discussion
Soybean. Soybean (Glycine max.) is a self-pollinating crop of limited genetic variability, thus impairing genetic identification exclusively on the basis of morphological traits. Using molecular markers we are expecting to assess degree of genetic variability between different genotypes, genetic purity control, monitoring viability and genetic changes due to long-term storage at low temperature. More important goal was identification and verification an old and
new genotype in collections of investigated plant species. Seed soluble protein electrophoregrams enabled us, by transformation of presence and absence of protein band to binary data, to calculate genetic diversity and index of similarity. Obtained results generally agreed with pedigree and morphological characteristics of investigated genotypes. Results obtained after use SSR and RAPD markers are more accurate and its application is recommended. **Red pepper.** Grouping of red pepper accessions belonging to different taxonomic units, revealed by the analysis of seed proteins, does not completely agree with classification based on morphological traits. Protein pattern of accessions belonging to the species *Capsicum annuum* L., could not give clear answer concerning connection between varieties and types of peppers. More specific genetic markers are suggested as better solution for understanding phylogenetic relations in *Capsicum annuum* L. **Pine - tree.** Seed salt soluble proteins, originated both from free pollination and half-sib lines, which were functionally female i.e., male, and the seed of parental genotypes and their hybrid combinations have been analyzed. Similarity coefficient has been calculated at the level of female - hybrid, male – hybrid and female – male comparison. Besides polypeptides common for both parents, polypeptides originated either from female or male (co dominance of parental gene expression in the hybrid), polypeptides originated only from female or male, and specific hybrid polypeptides have been identified. **Pine.** Specificity of investigated populations and open pollinated lines, obtained by seed morphology, germination dynamics and water imbibitions as well as behaviors of young seedlings has been confirmed by the analysis of seed salt soluble proteins. High level of individual variability of open pollinated lines was determined at the level of seed proteins, probably as consequence of pollen effect and high level of heterozigity of the female trees. Drought effect on different genes expression has been determined through different polypeptides synthesis in young seedlings. **Maize.** Characterization, classification and evaluation of local and introduced germplazme at Maize Research Institute gene bank have been done by protein markers application. Both quantitative and qualitative differences have been obtained between investigated populations. Discrimination between populations was based on qualitative difference of the protein pattern. Parallel results after protein complex and group analysis indicated that genetic constitution of investigated genotypes is different and this approach is not accurate to identify duplicates in gene bank. Another study attempted to analyze maize inbred lines of different origin by salt soluble protein, SSR, RAPD and AFLP markers. Maize genome polymorphism, identified by AFLP and SSR markers are presented in Fig. 1. Obtained results corroborate the usefulness of protein, SSR, RAPD and AFLP markers for characterization of inbred lines and for assigning inbreeds into heterotic groups.