

## **The utilization of genetic approaches for effective conservation of endangered species ConGRESS project, regional workshop, Zvolen, January 25–27, 2012**

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The ConGRESS (Conservation Genetic Resources for Effective Species Survival, within 7<sup>th</sup> EU Framework Program) is an EU consortium dedicated to transferring current knowledge in conservation genetics and in the analysis of genetic variation data to management professionals and policy makers (<http://www.congressgenetics.eu>). The project ConGRESS is aimed at the activities at the regional level, with particular emphasis on improving skills and knowledge regarding biological diversity and methods of protection in the regions of Europe, where it can be particularly lacking today. By building a network based on the scientists, specialists, practitioners involved in the protection of plant and animal species management and protection policies will be possible to provide a forum for communicating ideas, experiences and support that will strengthen the protection of European biodiversity and the future will improve coverage of its activities (<http://review.j4b.com/Congress/default.aspx>).

The meeting initiated by the ConGRESS project was contributed to educate a new generation of conservation managers and politicians who could be able to take into account and apply genetic research in biodiversity conservation projects in the future. The meeting was attended by about 70 scientists from four European Union member states: Czech Republic, Slovakia, Italy, Poland and Croatia. The initiator of the organisation of scientific workshop were Ladislav Paule, Professor (Slovakia) and Josef Bryja, Associate Professor (Czech Republic). The main topics were related to the following issues:

Presentation of the project's ConGRESS and decision support tools for species conservation:

- Description and utilization of modeling package available on the website of the project
- Testing of different scenarios of decision in modeling tools of project work package
- Testing of different scenarios in simulation tools of project work

The summary discussion of the symposium with the main topics:

- What are the main limitations of research results on conservation of genetic resources?
- What should be done to improve the co-operation between scientists and geneticists to make it more efficient in practice?
- Where could be find funding to build on their achievements in genetic decision-making processes regarding environmental protection?

The Regional Workshop – “Utilization of genetic approaches for effective conservation of endangered species,” provides a platform linking the studies molecular biologists working in the assessment of genetic variability of endangered populations of plants and animals on one side and biologists practitioners actively involved in the protection of species.

Long-term stability of gene pool in forest ecosystem is crucial to protect their ability to adapt to the impact of environmental changes, so conservation of adaptive genetic abilities of forest trees is fundamental in genetic management of forest stands. One of the fundamental fac-

tor in this field is understanding of biological factors that are basis of genetic structure of forest tree populations.

Human activities are the source of changes and destructions of the forest ecosystems and their genetic resources. The needs of minimising this influence and efficient protection of forest are very important to save their value for future. First of all it is urgent to understand natural ecological process of ecosystems and their genetic resources to understand their evolutionary forces to be able to protect their genetic variation. So, the crucial factor is the utilization of genetic makers, which enables to characterise an object of our investigation – a species, a population, an individual, a tissue etc. For this particular reasons the marker should be characterised with some traits (Szmidi and Wang 1991):

- Detect qualitative and/or quantitative variation
- Do not show environmental and developmental interactions
- Possess simple codominant inheritance
- Detect silent nucleotide substitutions without phenotypic effect
- Show changes within coding and non-coding parts of genome.

There is no genetic markers to be recommend to fulfill all traits to be desire. It should be taken into account to choose the best marker among others: marker system stability, simplicity of the techniques and their costs, the size of studied population, availability of adequate skills and equipment and so on.

Up to last 40 years, the history of our knowledge as far as genetic variation of forest trees population is concerned was based on investigation of allozyme diversity (isoenzyme studies). This markers are narrow group of proteins still very useful to study genetic variation of forest populations, but not representative for whole complicated genome and its adaptive ability under environmental changes. The important group of genetic markers are molecular markers developed last 15 years. The molecular markers used more common within last twenty years can be divided as follows:

### **HYBRIDIZATION-BASED MOLECULAR MARKERS**

RFLP (Restriction Fragment Length Polymorphism) methods were implemented by Rivin et al. (1983). This techniques are still used to describe the variation of or-

ganisms especially in pilot studies. Such fragments of DNA are detectable by restriction analysis or fragment hybridization. Fragments of DNA are detecting by cutting with specific enzymes, which are able recognise specific nucleotides (Botstein et al. 1980). Application of DNA in genetics of forest tree populations is very wide e.g.: phylogenetic relationship studies reconstructions, analysis of interspecies hybrids, estimation of genetic structure.

### **PCR-BASED MARKERS**

The possibilities of using the polymerase chain reaction (PCR) as a scientific technique in molecular biology, which enables to amplify a fragments of DNA and to produce copies of a particular DNA sequence was the basis to elaborate the powerful tools of analysing of the genetic variation of tested organisms. The DNA analysis offers much more markers with specific advantages useful in particular needs of the studies.

One of the first methods used to study DNA variation was RAPD (Random Amplification of polymorphic DNA). The RAPD method (Nakamura et. al. 1987) could characterised migration path after glaciation period and in the forestry was used to describe phylogeny of tree species. Short fragments of DNA (8–12 nucleotides) were used as primers to amplified the total DNA of sampled organism. Practically all of RAPD markers are dominant so, it is not possible to distinguish if a DNA fragment is amplified from a heterozygous locus (1 copy) or homozygous (2 copies). At the present, the experiment errors in the reproducibility of the analysis results of this method is not accepted by scientific journals to publish.

AFLP-PCR method (Vos et al. 1995) is utilised in genetics research, e.g. DNA fingerprinting, or genetic engineering. The restriction enzymes are used to digest genomic DNA, and next ligation of adaptors to the sticky ends of the restriction fragments. The restriction fragments are selected to be amplified. AFLP is not an acronym – the idea is to score the presence-absence of polymorphisms. AFLP-PCR is a highly sensitive method for detecting polymorphisms in DNA.

SSR (Simple Sequence Repeats) – short tandem repeats of 2–6 base pairs of DNA (STRs), among other there were developed: the microsatellites markers, expressed

sequence tags (EST), sequence characterised amplified region (SCAR), sequence tagged regions (STS), single nucleotide polymorphism (SNPs), diversity arrays technology (DArT) – Turnpenny, Ellard (2005), Semagn et al. (2006). The co-dominant character of the tools is very useful in genetics, for species and population genetic diversity evaluation, assessment of genetic distances as well as to study gene duplication or deletion.

The website of the ConGRESS project is still developed and it can found a lot of very useful links to get information concerning the wide set of molecular markers, techniques of their utilization, evolution of organisms, population genetic structure dynamics e.g. selection, gene flow methods of elaboration and many, many other problems.

The platform of The ConGRESS Project consortium helps to find information about the people and institutions dealing with forest conservation genetics:

- Albert Ludwigs Universität, Freiburg, Germany
- Cardiff University, Cardiff, United Kingdom
- Centre National de la Recherche Scientifique, Grenoble, France
- Durham University, Durham, United Kingdom
- Estación Biológica de Doñana, Sevilla, Spain
- Universiti Degli Studi di Ferrara, Ferrara, Italy
- Fondazione Edmund Mach, Trentino, Italy
- Institute of Vertebrate Zoology, Brno, Czech Republic
- Kobenhavns Universitet, Copenhagen, Denmark
- Koninklijke Maatschappij voor Dierkunde van Antwerpen, Antwerp, Belgium
- Queen Mary College, University of London, London, United Kingdom
- Stichting Nationaal Natuurhistorisch Museum Naturalis, Leiden, The Netherlands
- Turun Yliopisto, Turku, Finland

Conservation of biodiversity for future seems to be the most important activity, when the changes of environment are not possible to predict. Especially it is for gene-pool reserves it important to protect their genes without changes.

The mating system and selection of trees are usually unknown and not possible to assess under the influence of silvicultural treatments.

The importance of preservation of genetic variation is poorly recognised in practice compared to the other biodiversity components – species and ecosystems. ConGRESS project idea is to provide a tool to assess the relevance of genetic data and to show the processes that may result in genetic depletion, including a decision-support module and to allow end-users to establish optimal policy and management options given the genetic information.

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