# TreeBreedex Seminar – abstract book



What do large genetic field experimental networks across Europe bring to the scientific community?

Organized by:

Forest Research Institute, Sękocin Stary, Poland Institute of Dendrology, Polish Academy of Sciences, Kórnik, Poland Staatsbetrieb Sachsenforst, Pirna, Germany





Forest Research Institute Instytut Badawczy Leśnictwa, Sękocin Stary 22-24. 06. 2010

## Program

## Monday, June 21, 2010

Arrival of participants

# Tuesday, June 22, 2010

8.00	Leave hotels by minibus or by foot (a twenty minute walk)
8:30 - 9.15	Registration and coffee
9.15 – 9.30	Welcome address of Prof. dr hab. Tomasz Zawiła-
	Niedźwiecki, Director of Forestry Research Institute in
	Sękocin Stary.
9.30 – 9.45	Introduction by Luc. E. Paque, TREEBREEDEX Coordinator
	Technical announcements by Jan Kowalczyk
9.45 – 10.15	1 <sub>st</sub> invited presentation:
	Usefulness of the genetic field experiments for biological
	sciences
	Jacek Oleksyn
10.15 – 10.50	Voluntary paper
10.15 – 10.35	The role of common garden studies in adapting forests to
10.05 10.50	climate change in the Northwestern U.S Daniel Chmura
10.35 – 10.50	Population variability of Fagus sylvatica leaves: a preliminary
10 50 11 00	study- Marzenna Guzicka
10.50 - 11.00	Discussion
11.00 – 11.30	Coffee and group photograph
11.30 - 13.00	Visit to the experimental plot of Scots pine (IUFRO 1982)
13.00 - 14.00	Lunch
14.00 - 14.30	2nd invited presentation:
	What do genetic field trials tell about the future use of
	forest reproductive material?
	Csaba Matyas
14.30 - 15.30	Voluntary papers
14.30 - 14.50	Genetic reactivity of Norway spruce to climate change based
	on experimental results from IPTNS-IUFRO 1964/68 test in
	Poland Janusz Sabor
14.50 - 15.10	Short review of the tree improvement work with birch and
	alder in Ireland – Ellen O'Connor
15.10 - 15.30	International trials concerning forest species in Italy –
	Anna De Rogatis

15.30 – 15.45	Discussion
15.45 – 16.00	Coffee

16.00 - 16.30	3rd invited presentation:
	Provenance experiment networks as a tool for biochemical
	and molecular genetics of forest trees.
	Berthold Heinze
16.30 - 17.10	Voluntary papers
16.30 - 16.50	Characteristics of genetic diversity and differentiation of
	progeny and mother stands of European Beech in Poland
	Małgorzata Sułkowska
16.50 – 17.10	Microsatellites and genetic diversity in seed orchard and
	provenance test - Magdalena Trojankiewicz

17.10 – 17.25 Discussion

17.25 – 18.30	TREEBREEDEX Consortium meeting
	Efficiency of tree breeding strategies in Europe. Report from the
	Questionnaire "Testing strategies in tree breeding"
	Darius Danusevicius
18.30	Free evening or any other busses

## Wednesday, June 23, 2010

9:00 - 9.30	4th invited presentation:
	Impact of the results of large genetic field experimental
	networks to practical forestry supporting industry.
	Dag Lindgren
9.30 - 11.00	Voluntary papers
9.30 – 9.55	Climate-growth-relations of Fagus sylvatica provenances of
	the International Beech Provenances Experiment of 1993/95
	growing in Central Europe - Mirko Liesebach
9.55 – 10.20	Euro-Asiatic transcontinental provenance experiment on
	Scots pine (Pinus sylvestris L.)– Władysław Chałupka
10.20 - 10.40	Larix decidua polonica in Western Europe. First results from a
	joined European network of progeny trials— Luc Pacues
10.40 - 10.55	Last Evaluation of the Provenace Plot Podbanské, Slovakia
	(IUFRO I. Larch Series 1944)– Elena Foffová, Vadimir Foff
10.55 – 11.10	Eurasian provenance experiment trial of Scots Pine at
	Sambor in Ukraine– Roman Gout, Jan Kowalczyk
11.10 - 11.30	Coffee
11.30 - 12.10	Voluntary papers

11.30 - 11.50	Adaptability of oak (Quercus robur L.) ecotypes in conditions of climate change - Igor Neyko
11.50 - 12.10	PLANTACOMP: Genetic experimental network of French
11.50 - 12.10	National Institut for Agricultural Research- Anger Christel
12.10 12.20	
12.10 – 12.30	Discussion
12.30 - 13.30	Lunch
13.30 - 14.00	5th invited presentation:
	Large forest tree provenance experimental networks: their
	advantages, limitations and importance for future
	experiments.
	Mirko Liesebach and Heino Wolf
14.00 - 14.30	Poster session and common discussion
14.30 - 14.45	Final Discussion and conclusions
15.00 - 15.30	Coffee
15.30	Leave for Warsaw to visit the Royal Castle and the Old Town
19.00	Social dinner
22.00	Arrive hotels
Thursday, 24 June	, 2010 - Field trip
7.30	Leave Sękocin
8.45 - 9.30	Visit to the European larch reserve "Modrzewina"
11.30	Arrive Bliżyn

EVOLTREE Intensive Study Site (ISS) Bliżyn

Świnia Góra larch stand

- Open air lunch
- 19.00 Arrive hotels

# Friday, 25 June, 2010

Departures

## Abstracts

# Usefulness of the genetic field experiments for biological sciences Jacek Oleksyn

Institute of Dendrology, Polish Academy of Sciences, ul. Parkowa 5, 62-035 Kórnik, Poland

Field experiments to test the adaption of new and different origins of genetic material (provenances) began when explorers bought new species to Europe from diverse parts of the world in the 16th and 17th century. 'Riga Pine' from the Baltic was the source of pine trees suitable for ship masts in the 18th and 19th centuries and these Baltic seed sources were collected and tested in 18 sites in France by Philip de Vilmorin at around 1810. The first concerted international trials decided on the parameters to record to compare sources, such as dbh and tree height. Scots Pine was the first international scientifically based trial established in 1938-39 by IUFRO and thereafter a series of trials used a broad range of provenances.

The first trials with Scots pine showed that 'local' sources were best however modest increases in height and survival rates can be obtained using different sources to local. Recent experiments have shown that Northern provenances have more root production biomass per year and that the populations from North or high elevation had a higher proportion of live roots. Clear differences in the start and end of the growth cycle were detected with an earlier flushing (by 10 days), a longer period of needle growth (by 45 days) and a larger growing season (+60 days) was found for Polish provenances compared to Swedish Provenances. Ecophysiological studies have shown that Northern Provenances of Norway spruce and those from higher altitudes have a higher concentration of nitrogen in their leaves and dark respirations and photosynthesis was also higher.

# The role of common garden studies in adapting forests to climate change in the Northwestern U.S.

### Daniel J. Chmura<sup>1</sup>, Glenn T. Howe<sup>2</sup>, J. Brad St. Clair<sup>3</sup>, Paul D. Anderson<sup>3</sup>

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Population variation revealed in common garden studies – long term provenance tests and short-term nursery experiments – indicates that populations of trees are adapted to climates of their origin. Therefore, populations, rather than whole species, should be regarded the units of interest with respect to forest adaptation to climate change. In the Nortwestern U.S., projected climatic changes will likely include atmospheric warming and related changes in hydrology that may lead to increases in drought, wildfires, and insects and disease epidemics. Contemporary populations may become maladapted to their locations if these projected changes occur. Adaptation to novel climates may require active management of genetic resources at the population level, which includes assisted migration and conservation or enhancement of genetic diversity.

The Seedlot Selection Tool – an on-line GIS-based program – will help forest managers to identify seedlots expected to be adapted to future climates at their sites based on selected climate change scenarios (http://sst.forestry.oregonstate.edu/). The Center for Forest Provenance Data (http://cenforgen.forestry.oregonstate.edu/) will gather information from existing networks of provenance and other genecological studies in one database. Data will be available to researchers seeking to understand plant adaptation, population's responses to climate change and to support management decisions for adapting forests to new climates in the Northwestern U.S. and elsewhere.

#### Population variability of Fagus sylvatica leaves: a preliminary study

#### Marzenna Guzicka & Roman Rożkowski

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The aim of the present study was to examine of population variability in *Fagus sylvatica* leaves. The leaves were collected in August 2009 in the experimental site in Choczewo. The site is a part of a project testing diversity of beech in Poland. 12 populations in 2 block were selected for the study, and the block set was rearranged. The leaves were collected only from the upper part of the tree crown, and each population was represented by six individuals (3 trees × 2 blocks). The following characters were analysed: perimeter, area, total holes area, leaf length, blade length, blade width, mass of 10 leaves, and LSA. The variance analysis showed that the block impact was not important, and the population had a significant impact. We found a positive correlation between almost all examined leaf characters and the tree productivity.

#### What do genetic field trials tell about the future use of forest reproductive material?

### Csaba Mátyás

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#### 1. Changing forest communities in changing environments

Adjusting the policy of forest reproductive material use to the challenges of climate change, requires genetic information on the tolerance and adaptability of tree species. In practice, the only possibility to study adaptive responses under experimental conditions is the analysis of common-garden tests (i.e. provenance trials). The transfer analysis of forest reproductive material, i.e. modelling and forecasting responses based on field trials, is a useful tool to investigate the effects of environmental changes on growth and health. As part of this analysis, the phenotypic response of a provenance across test sites is interpreted as norm of response to changing climatic conditions. The response functions can be used to validate models describing adaptive reactions to changed climate.

### 2. Quantitative response to changes: results of field trials

Results of field trials show a remarkable range of adaptability even to dramatic changes in thermal and moisture conditions. The effects of environmental changes on tree populations in different parts of the distribution range are, however, divergent, as different climatic factors exert their selection pressure.

In the boreal zone, expected rise of temperature will lead to accelerated growth without any significant genetic changes of tree populations. In the dry, warm-temperate zone and in particular in the Mediterranean region even relatively small temperature increases, coupled with growing drought stress, will trigger loss of compatibility, growth and productivity and increase susceptibility to pests and diseases. Subsequently, this will increase mortality in tree populations and challenge the economy of forestry operations.

The superiority of locally adapted, autochthonous populations shows also a differentiated picture. It seems that the fitness of local seed sources is the most sensitive to changes at locations with extreme conditions. The more favourable the local conditions are, the more environmental change locally adapted provenances can tolerate.

A fundamental question is how much the natural genetic processes such as migration, gene flow and natural selection, can compensate the effects of climate change. It seems that the simultaneous action of these processes creates an adaptive genetic disequilibrium, instead of adaptive optimisation, a preconception implicitly regarded as basis of forest reproductive material use in the past.

In view of expected changes, these facts and responses will have to be taken in consideration when shaping the future strategy of reproductive material deployment.

### 3. Recommendations

Adaptation strategy to climate change cannot rely on natural self-regulation and acclimation of ecosystems. Therefore, human interference is essential to ensure adaptability of forests and reduce environmental and social pressures on sustainability.

### 3.1 Adaptation through appropriate use of forest reproductive material

The artificial support to migration and gene flow, i.e. the controlled transfer of well documented forest reproductive material and the preference of populations exhibiting plasticity, offers the best possibilities for human-facilitated adaptation. This is certainly more efficient than reducing landscape fragmentation with green corridors. Silvicultural measures, such as selection of appropriate natural regeneration techniques and regulation of species mixture, can also facilitate this mitigation.

Only few countries have already adopted the above principles in their forest strategy. There is a need to develop *flexible and genetically well-founded pan-European guidelines* for using forest reproductive material.

### 3.2 Priorities

Tree populations and rare tree species at the southern or continental distribution limits are specially threatened. In case of common, widely distributed tree species certain populations will also need special consideration, such as populations with high phenotypic plasticity as well as valuable and isolated populations on the southern or low-elevation fringes of the distribution area.

### 3.3 Need for a common plan of action

The mitigation of the effects of climate change presents tasks of a magnitude not encountered before in European forestry. The nature of the problems requires cross-border collaboration, strengthening of international cooperation and sharing of responsibilities. At country level, national forest programmes have to incorporate the above-described tasks and principles into their agenda.

# Genetic reactivity of Norway spruce to climate change based on experimental results from IPTNS-IUFRO 1964/68 test in Poland.

#### Janusz Sabor

University of Agriculture in Krakow, Faculty of Forestry, Department of Forest Tree Breeding, e-mail: rlsabor@cyf-kr.edu.pl

Genetic diversity of spruce provenances tested in Krynica (Poland)n was presented. The trial has a full set of 1096 provenances. It is the most elevated planting site (750 m) for the whole experiment. The experiment covers provenances from the natural range of the species and from the area where spruce was introduced by man. Poland is represented by 92 provenances. Among all the provenances considered, 528 have a strictly defined (accurate to a stand) location, so they can be reproduced and used in practice. The material is thus representative of the whole Picea abies species to the degree that has no parallel in any previous research.. Statistical analysis was performed on all data and an analysis of variance showed no effect of blocking on all characters except blade length. Significant provenance effects were found for all of the characters which were measured. As suggested by the height of trees aged 25 years and the frost resistance (late flushing) of spruces, the provenances from regions 67 East Pomeranian Lakeland, Masuria Poland, 69 Augostów, Lakeland Poland, 50 Slovenskie Rudohorje , 75 Belarus, 96 Canada (Hudson, Ontario) and 58 Bihor Mts., Transylwania, Romania have the greatest genetic and breeding value.

### A Review of the Irish Birch and Alder Improvement Programmes.

# <u>Ellen O' Connor<sup>1</sup></u>, Niamh O' Dowd<sup>2</sup>, Martin Steer<sup>3</sup>, Michael Bulfin<sup>4</sup>, Nuala Ni Fhlatharta<sup>4</sup>, and Barbara Doyle<sup>1</sup>.

1) University College Cork, 2) Dublin City University, 3) University College Dublin, 4) Teagasc.

Two species of birch are native to Ireland; *Betula pubescens*, downy birch, and *B. pendula*, silver birch. Poor stem quality of naturally regenerated birch has prevented the listing of birch as a recommended species for afforestation programmes. Previous field tests of seed obtained from Sweden and Finland in the 1960's and 1980's have resulted in low survival and slow growth rates in the imported material. This poor performance was attributed to mal-adaptation to environmental cues, especially photoperiod, in these species. Based on this experience, a programme of birch improvement founded on native plus-tree selection was started in 1998; supported and funded by COFORD, the council for forestry research and development. The improvement of birch has progressed with 10 year-old progeny trials and an indoor seed orchard established at this stage. However, it has become apparent that an insufficient number of plus-trees of *B. pendula* have been identified to date.

Seven Scottish provenances, two German breeding populations and one French family of *B. pendula* were included in the progeny trials established in 2001. Many of these overseas families have shown good survival rates, growth rates within the range of native material and quality that is comparable to many of the Irish *B. pendula* families. It is, perhaps, time to consider the use of overseas material again. The best trees from the best of the Scottish, French and German families could provide a fast route to expand the base of *B. pendula* used in the improvement programme. In addition, the modern approach of matching 'environmental envelopes' could direct new movements of *B. pendula* from Europe. It is envisioned that new introductions would be incorporated into the improvement programme rather than used directly.

Alder, *Alnus glutinosa*, is on the recommended list for afforestation and has been in such demand in recent years that the development of an improved sustainable, healthy native seed source was warranted. The improvement of alder was initiated in 2005. Grafts of eighty-five plus-trees have been collected for use in an untested seed orchard.

#### International trials concerning forest species in Italy.

#### Anna De Rogatis, Fulvio Ducci & Lorenzo Vietto

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Italy specially CRA SEL, and CRA PLF, respectively the former Istituto Sperimentale per la Selvicoltura of Arezzo and the Istituto di Sperimentazione per il Pioppo of Casale Monferrato always had shared efforts for establishing international experiments on forest species.

Since the first years of the XX century the meaning and the need of establishing wide national or international multisite experimental tests was clear, due to the high environmental variation of the national territory. Indeed, only large experiments can allow the understanding of productive and adaptive potential of species, provenances and taxa produced with breeding and improvement programmes.

Initially, most of international tests were initially focused on conifers, mostly exotics, but then also hardwood species were considered.

Materials from species of wide economic interest were tested in the framework of wide international trials as Larch, *P. sylvestris, Pinus* sp. sect. *Halepensis, Pseudotsuga menziesii,* Mediterranean firs, *Prunus avium, Juglans regia, Populus* sp., and many others not included within the range of Treebreedex model species *Cedrus* sp., *Cupressus* sp., *Fagus sylvatica, Pinus pinaster, Robinia pseudoacacia, Ecalyptus* sp. studied by CRA or universities or CNR institutes.

Most of these trials were planted thanks to IUFRO, FAO Silva Mediterranea, AIR, FAIR or COST European programmes or even within bilateral collaborations.

Materials are in general subject to genetic, breeding and monitoring studies on phenology or physiology for resistance/tolerance to biotic or abiotic stress, as well as productive and economic value traits were investigated.

International trials allow the evaluation of materials based on large environmental range, either for interaction *genotype x environment* for <u>multiple productive</u> characters and parameters either for <u>adaptive</u> traits.

Nowadays, in view of the global change effects, international trials are open air laboratories for studying deeply <u>adaptation</u> and <u>genetics of adaptation</u> and supply information on FGR reactions strategic for <u>mitigation</u> activities and <u>preserving</u> resources *in situ* and *ex situ*.

Many are problems for long term managing, for maintainance, conserving continuity in the time, problems due to changes in managers and to social aspects, but now also to the increased ferquence of extreme events is becaming significant.

# Provenance trial networks as a tool for biochemical and molecular genetics of forest trees

#### **Berthold Heinze**

#### BFW - TBX P02 - Vienna, Austria, e-mail: berthold.heinze@GMX.AT

Field trials are often a "quick and easy" way to collect material for genetic marker studies. Lagercrantz and Ryman (1988, 1990) have provided one of the first examples in their efforts to assess range-wide variation in a forest tree, with isoenzyme (allozyme) markers, utilizing material in the Norway spruce IUFRO 1964/68 trial in Sweden. Other types of markers have also been studied in field trials - actually any kind of marker can be studied. Nuclear DNA markers include sequencetagged sites, chloroplast microsatellites, mitochondrial minisatellites, or a combination of several types of markers. Disadvantages of using field trials for this purpose include the uncertainty of identification of the exact source, especially for older trials; the exact descent of material (how many mothers, and the identification of half-sibs); the possible disappearance of the original seed stands; possible natural genetic selection in the nursery or at the trial site; and the poor representativeness of the collection for the whole species range. Alternatives for obtaining diverse material are to request seeds or collect on one's own, or to ask correspondents for postal sending of material. The important question for this seminar is, however, what is the "added value" of large trial networks? Up to now, this is not really evident - multiple-site studies are rare, and multiple-lab studies are rare as well. There are a number of ways were several sites could be utilized for investigating, e.g. selection effects at different sites, pedigree reconstruction, genetic diversity and plasticity, selection, adaptation and epigenetic effects, etc. One concern was often that such studies have little "marking" capacity for really interesting growth traits (incongruence between observable growth and marker patterns in many examples).

At the start of the age of genomics, genetic mapping required family pedigrees, not provenances. Alternatives from human genetics, were building large pedigrees is also not feasible, include admixture mapping and association studies. In the latter, simple correlations between markers and traits are studied. Technological advances make it possible to analyse many genes in many individuals now, at least for some organisms. In a typical association study, the steps are to collect material, measure phenotypes (height, diameter, disease prevalence, ...), and then to analyse as many markers as possible. Alternatively, even whole genome sequencing for individual genomes is possible in Arabidopsis. Single nucleotide polymorphisms (SNPs) can be analysed on several platforms. Next, it is necessary to assess any population or family structure in the sample. To calculate the statistical associations, dedicated software is available. It is desirable to verify any associations in an independent sample. Advantages of association studies include the ease of the approach for sampling, and its inherent simplicity. Although no building of pedigrees is necessary, family pedigrees can enhance the study. Current examples of association studies in trees will be presented in an overview, including examples for studying range-wide demography, local adaptation, and wood traits. This first generation of studies, using traditional Sanger sequencing of some candidate genes and / or SNP assays on many individuals, included testing for deviation from evolutionary neutrality, for genes or alleles that show reduced or enhanced diversity and thereby "footprints of selection" or "selective sweeps", and testing for association with "geography" or measured wood traits. Issues with association studies will be addressed; most notably, association (statistical correlation) does not always mean causal explanation. Nevertheless, genome-wide ("genomic") association studies will hopefully reveal genetic control of traits in many species soon, because technology advances make it possible to study many genes and whole genomes. Experimental networks are an ideal basis for such studies - both provenance and progeny trials can be used. Basic research into gene function is necessary before gene markers can be used for selection.

# Characteristics of genetic diversity and differentiation of progeny and mother stands of European Beech in Poland.

#### Malgorzata Sulkowska, Justyna Nowakowska

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European Beech (Fagus sylvatica L.) is one of the most important forest tree species, which covers 5.1 % of forest area in Poland. Present genetic structure of beech populations was formed by many different factors, not only environmental and genetic ones but also anthropogenic. It attains its north-eastern limit of natural range in Poland. There were investigated 6 beech populations classified according to phytosociological characteristics to the following plant associations: Galio-odorati-Fagetum (Gryfino and Kwidzyn), Dentario glandulosae-Fagetum (Bieszczadzki National Park), Luzulo-luzuloides-Fagetum (Suchedniów, Tomaszów), Dentario enneaphyllidis-Fagetum (Zdroje). The genetic structure of these populations was analysed. Thirty individuals per one generation (mother, progeny stands) in every provenance were investigated. The genetic variation and differentiation of mother stands and their open-pollinated progeny were characterised on the basis of following isoenzyme: glutamate-oxaloacetate transaminase (GOT – EC 2.6.1.1 – Got-2), leucine aminopeptidase (LAP - EC 3.4.11.1 - Lap-1), malate dehydrogenase (MDH - EC 1.1.1.37 -Mdh-1, Mdh-2, Mdh-3), menadione reductase (MNR – EC 1.6.99.2), phosphoglucomutase (PGM – EC 2.7.5.1), phosphoglucose isomerase (PGI - EC 5.3.1.9 – Pgi-2), shikimate dehydrogenase (SKDH - EC 1.1.1.25) and DNA chloroplast markers: ccmp4, ccmp7 and ccmp10. There were calculated following genetic parameters: average number of alleles per locus, percentage of polymorphic loci and heterozygosity observed and expected on the basis of isoenzyme analysis. The frequency of obtained DNA haplotypes as well as average number of alleles per locus were characterized. The parameters of genetic diversity (Hs and Ht) and differentiation (Gst and Nst) of DNA markers were counted and compared between mother and progeny generation. The results pointed at the very high inter-population diversity of mother stands as well as their progeny. The investigations revealed the importance of using local European beech ecotypes, taking into account its plasticity, which is the best advice to obtain success in forest management and for protection of genetic resources of the species.

# Efficiency of tree breeding strategies in Europe. Report from the Questionnaire "Testing strategies in tree breeding

Darius Danusevicius<sup>1</sup>, Alfas Pliura<sup>1</sup>, Gunnar Jansson<sup>2</sup>, Dag Lindgren<sup>3</sup>

1- Lithuanian Forest Research Institute, Lithuania (LFRI, P15),

2- Skogforsk, Sweden (P21),

3-SLU, Sweden (P25).

Efficient breeding implies optimum allocation recourses between high and low input breeding and optimal combination of genetic gain, gene diversity, costs and time. This combination strongly depends on the long-term breeding plans and the input in breeding. The experience is gained, but not equally among the European countries, where breeding is driven by variable ownership types and interests. To maximise the efficiency of breeding at the pan-European perspective, it is beneficial to gain from experience of scientifically-sound strategies. The objective of this questionnaire is to prepare a review on how breeding programs of forest trees are designed and what testing strategies are used in European countries. The ultimate goal is to improve efficiency of breeding by taking advantage of the efficient practice. The questionnaire consists of 3 parts: (I) breeding strategies and testing/selection methods used for each species,(II) tools available to optimise the testing strategies and (III) literature list on optimization of breeding strategies of forest trees. In total, answers on 115 breeding programs from 28 forest tree species were obtained from 19 Treebreedex institutions (representing 19 countries). The main forest countries responded. No breeding programmes were reported for such wide-spread conifers as Juniperus and Taxus bocata. Most breeding efforts are focused 3 coniferous species (Pinus sylvestris, Picea abies and Larix sp.) and on 4 broadleaved species (Populus sp., Betula sp., Fraximus sp. and Prunus avium). The general statistics on breeding is as follows: 60% of all are long-term programmes; 52% high input; 30% do not subdivide the breeding stock into breeding populations and as much as 40% use the site type and natural species distribution as the main criterion for subdividing into breeding populations (meaning not eco-climatic zones or adaptive environments); only 10% maintain nucleolus breeding population for generating high gain; 47 % uses closed breeding populations with no infusion of genetic material from outside; only 33% use controlled mating among breeding populations members; 87% use the same testing strategy for different traits; 48% breeding and multiplication populations are not separated; 69 % use among and within family selection; 50% uses two-stage phenotypeprogeny testing strategy; 8 % use molecular markers in breeding and 5% use simulations to optimise breeding (most were willing to use simulations). In the analyses of the answers, the breeding strategies were subdivided into 4 categories based on terms and input: "long-term high-input"; "long-term-low-input"; "short-term high-input" and "short-term low-input" and methods of breeding with each of these 4 strategies were analysed.

#### Microsatellites and genetic diversity in seed orchard and provenance test.

#### Magdalena Trojankiewicz

Kazimierz Wielki University in Bydgoszcz, e-mail: trojankiewicz@ukw.edu.pl

Microsatellites are a very efficient tool for many population genetics application. The effective population size is one of the most important parameters of a mating system and it defines the number of individuals participating to an equal degree in reproductive processes. The aim of this study was to investigate reproductive processes in seed orchard. The survey was applied to a Scots pine (Pinus sylvestris L.) located in Gniewkowo Forestry. Genetic surveys were carried using nuclear microsatellites DNA as genetic markers. Genotype of parents population individuals were defined for set of 187 seedling growing in the central part of plantation. Genetic structure of parental and progeny population was surveyed as well as associating system and so called pollen contribution. Comparative analysis of effective size of male parents were carried rated by many different survey methods. A high level of genetic variability characterize parents population. In a single locus the number of alleles varied from 8 to 26, with the average 14.33. High values of expected hetoerozygosity  $H_e = 0,803$  and observed  $H_o = 0,785$  additionally bear evidence of high polyforphizm of markers. We observed differential participation of individual seedling in creating the genetic pool of analyzed progeny population. In every observed loci new alleles were noted, which could be caused by pollen immigration. Variance effective population size estimated for an individual loci varied form 8 to 77. Variance effective population size estimated on a base of all loci equaled  $N_{e(v)}$  = 24,80. The analyze of genetic structure of pollen pool based on the TWOGENER method allowed to estimate the genetic differentiation degree of pollen gametes that took part in creating progeny of analyzed maternal trees. The heterogeneous pollen pool factor was being  $\Phi_{FT}$  = 0,0095 and tha calculated effective population size of male parents based on the previous calculation gave as  $N_{e(p)}$  = 52,57. Mating system analyzes showed that in the majority of analyzed loci the value of cross- fertilization ( $t_s$ ) was higher than 1 given the average  $t_s = 1,029$ . The value of cross- fertilization estimated with *multilocus* method equaled  $t_m$ =1,014. The average male parents correlation factor equaled  $r_p$  = 0,046. Effective male parents population size estimated the base counted the bases on significant differentiation from 6,7 to 1000, given the harmonic-average  $N_{e(r)} = 21,74$ . Fatherhood analyzes showed that ten the most productive clones were responsible for up to 63% of pollinations happening in local population. Differential success of linking leads to the reduction of effective population size, which in that case equaled amounting to  $N_{e(f)} = 17,14$ with account for 40% of total number of clones. The neighbourhood focusing on such personal characteristic as: the distance from female parents trees, fertility, the amount of pollen produced, allowed to rate an effective number of seedling that equaled  $N_{e(s)} = 92,7$ , which account to 49,6% all seedlings present in the second seed orchard. Effective population size of male parents on the analyzed plantation is big, considering the fact that the level of contamination is 60%.

# Impact of the results of large genetic field experimental networks to practical forestry supporting industry.

### Dag Lindgren

Department of Forest Genetics and Plant Physiology, SLU, SE-901 83 Umeå SWEDEN e-mail: Dag.Lindgren@genfys.slu.se

Some expected impacts for Industry:

- More reliable and applicable breeding values
- Better forest regeneration materials now and in the future
- Better known and documented forest regeneration materials
- Reduced risk of failures with FRM
- Better forecasts of forest growth
- More discussion and attention focusing on the forest in the field
- Better contacts among those dealing with similar forests in different organizations (countries)
- More focus of scientists (like forest geneticists), education and administrators of what happens with industrial plantations
- Easier to claim that Industry knows something about what they are doing and tries to get it better known (e.g. diversity)!

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Large genetic field experiments are one of the keys to survival of the human race and civilization!

- Without them we do not know what we should do or have done when managing forest land.
- Gives a sustainable support for an increasing world population with a reasonable standard of living!
- Emphasize on sustainability and basic environment friendliness. The forest creates raw material from air, water and sun-shine.
- Demonstration that we care for the future and plan long term.
- Basis for predicting the impact of the present and future forest.

#### Climate-growth-relations of *Fagus sylvatica* provenances of the International Beech Provenances Experiment of 1993/95 growing in Central Europe.

### Mirko Liesebach<sup>1</sup>, Silvio Schüler, Heino Wolf

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European beech covers a large geographic area of Europe and is a strong competitor with other tree species in Central Europe. At the Institute of Forest Genetics in Grosshansdorf in 1993 seed lots were sown for the establishment of an International Beech Provenance Experiment, and subsequently 23 trials were established in spring 1995. The presentation will concentrate on three sites with 47 common provenances, where survival and height growth were monitored: Schaedtbek (Germany, Schleswig-Holstein), Malter (Germany, Saxony) and Gablitz (Austria). Climate data are available for these trial sites and the sites of provenance origin.

The seed lots where many of the tested provenances originated lay to the west or southwest of the trial sites, in a cooler damper habitat than the trials. Hence for practical purposes this east- / northeast-wards translocation of provenances simulates the predicted climate change for this region.

In addition to climate, growth rate could be influenced by the previous use of the sites. Beech is growing much better on the two sites established on agricultural soils than on forest land. Overall survival is similar among the sites or likewise affected by management.

Height growth of provenances from the centre of the distribution area was rather unresponsive to climate, but provenances from the eastern part did response to climate. On the three trial sites some provenances from the eastern distribution area grow consistently under different climate conditions. Other provenances were sensitive to transfer to different sites: i.e. they have the tendency to grow better on some sites than others.

# Euro-Asiatic transcontinental provenance experiment on Scots pine (Pinus sylvestris L.)

#### Władysław Chałupka

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In 1976 on the initiative of All-Union Forest Research Institute in Pushkino, near Moscow, a major Scots pine provenance experiment was established on the territory of former Soviet Union. Seed were collected at 113 stands, and 33 experimental trials were established on various sites. A joint evaluation of five quantitative and qualitative characters was conducted at age 10-15, and results were reported by A. M. Shutyaev and M. Giertych.

The presentation included both basic data on the lay out of experiment and some results of provenance performance at experimental trials. A division of Scots pine range proposed by Shutyaev and Giertych was also demonstrated.

# Larix decidua polonica in Western Europe. First results from a joined European network of progeny trials

Luc Pacues

INRA, Orleans, France

Last Evaluation of the Provenace Plot Podbanské, Slovakia (IUFRO I. Larch Series 1944)

Elena Foffová, Vadimir Foff

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#### Eurasian provenance experiment trial of Scots Pine at Sambor in Ukraine

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In the years 1974 to 1976, on the inspiration of the Forest Research Institute in Pushkino, a major Scots pine (*Pinus sylvestris* L.) provenance experiment was established on 33 planting sites scattered over the former territory of USSR. One of the testing sites was located in the west Ukraine in East Roztochia, locality Sambor. The trail was planted in 1975 year using one year bare rot seedlings. The spacing was  $2.0 \times 0.75$ . In total 34 provenances were distributed in 3 randomized block design. The trail was measured last time in autumn 2006 after 33 years of growth. The present brief report gives results of this trail. The provenance from Ukraine have good growth and survival among them the local one no 34a is the best on volume. The relationship can be observed between volume and longitude of origin of the respective provenance(r2=0.5). The results pointed at the very high inter-population diversity. Provenance heritability value for volume is 0.9 +/- 0.2. The main conclusion is that for the Ukrainian conditions the most suitable from studied populations are local one and population Gomel from Belarus.

Adaptability of oak (*Quercus robur L.*) ecotypes in conditions of climate change.

#### Ihor Neyko

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Adaptability of oak population was studied on the provenance tests located in the central forest-steppe zone of Ukraine (Vinnitsa region). Oak provenance tests were created in spring 1964 by acorn sowing. The seed material was represented from 32 regions of Ukraine, 20 – of Russia, 6 – of Belarus, 2 – of Moldova, Latvia, Lithuania and Estonia. Local oak population was sown as a control. Each geographical location was represented by one variant. Necessary measures on the removal of died examples were regularly taken.

The data analysis of 1964-2010 specifies essential ecological and geographical influence of seeds origin, phenological forms on the growth and productivity of climatic ecotypes as well as on selection and quality indicators. The worst seed germination intensity was characteristic for the most remote northern and north-east ecotypes: Moscow, Volgograd, St.-Petersburg, Chuvash, Estonian, Bashkir, Latvian, and Pskov. But it is necessary to note that some remote ecotypes had tendencies for the improvement of adaptability and decrease of tree dying intensity (some populations from Estonian, Bryansk, Brest, Latvian and Minsk ecotypes).

Progeny of the ecotypes of the most remote northern, northeast and east regions (Moscow, Tambov, St.-Petersburg, Bashkiria, Estonian, Chuvashia ecotypes) are marked by the slowest growth in height and diameter. Analysis of the results on the growth dynamics of oak ecotypes testifies that the greatest differentiation in height was marked at the initial stages of growth. Up to 10-year age the difference of growth intensity in height was more than 60 %. At the age of 25 - 40 there was a tendency towards activization of growth intensity of the northern and north-east ecotypes (Estonian, Tula, Tatarstan ecotypes). Intensity increase of the growth processes specifies the increase of adaptability of the remote ecotypes. Acclimatization of the remote geographical oak ecotypes makes up about 20-30 years.

#### PLANTACOMP: Genetic experimental network of French National Institut for Agricultural Research

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Since several dozens of years, numerous genetic experiments of forestry species were installed through France. Genetic breeders of French national Institut for Agricultural Research actively participated in the implementation of this network, recently baptized PlantaComp. The initial objectives of these tests were to analyze the expression of genetic variability and to study genetic parameters in order to create improved materials of reforestation. This network aggregates tests comparing species, provenances, progenies and clones.

PlantaComp is a remarkable storage of genetic diversity and thus presents a strong stake in preservation of the biodiversity. Furthermore, the setting-up of the same genetic units in varied environments, and the follow-up over long periods of these trials, allow to have access to precious information on the plasticity and the adaptation of this material in front of a variable environment, which mimes the impact of the announced climate change. Finally, these genetic experiments, installed in a rigorous way in contrasted environments and followed regularly for several years can also serve as a support for studies concerning other themes: following-up of invasions of insects, efficiency of the use of the water, the detection of QTL ...

To make the most of the potential of this network, and so face the new stakes in the forestry sector, French National Institut for Agricultural Research showed one's willingness to organize this network. So, PlantaComp could be valued through national or international projects, included several partners, linked with emergent issues of forestry research (climate change but also production of biomass, biomolecule) while staying most closely with preoccupations of the profession (selection of improved material).

# Large forest tree provenance experimental networks: their advantages, limitations and importance for future experiments

### Mirko Liesebach<sup>1</sup>, Heino Wolf<sup>2</sup>

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Provenance research in large experimental networks is done since more than one hundred years in Europe forced by the serious and frequent failure of cultivation of seed sources without consideration of the origin of seed. For the main conifer species and broadleaved species several international provenance experimental networks were establishment and maintained over a long period since the beginning of the 20<sup>th</sup> century. In addition, first attempts for the screening of material approved as tested on European level were done with Hybrid-larch as an example in the end of the 20<sup>th</sup> century.

The research work done resulted in a more or less sound knowledge on the variation of provenances of species investigated under existing climate conditions. However, climate change will affect the cultivation conditions more or less for all forestry species. Therefore, the experimental networks already existing could be used as an excellent base for on-going research on the adaptability of species and their provenances to changing climate conditions. Above that, large experimental networks newly to be established can be used as a tool to evaluate and to assess the cultivation value of emerging species and their provenance.

Despite of many problems involved in the management and the financing of these long term projects, large experimental networks are the only and practical way to explore the possibilities and limitations of genetic resources until something better is developed.

#### Poster session

### Variation of pedunculate oak (Quercus robur L.) tested on "Chrostowa II" experimental plot located in Carpathian Foothills (southern Poland)

#### J. Banach

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Between 1996 and 2000 there were 17 provenance-family experimental plots with pedunculate oak established in Poland, including three plots in southern part of the country, in Brzesko Forest District (two in Chrostowa Forest Range, and one in Jodłówka Forest Range). The paper presents the results of analyses on second plot situated in Chrostowa, which was established in 1999 by planting 3-year-old oaks.

The material tested on the "Chrostowa II" experimental plot consisted of progeny of 5 Polish provenances of pedunculate oak. They represented the northern (Młynary-1 and Młynary-2) and southwestern (Krotoszyn, Milicz, and Opole) parts of Poland.

Oaks' average survival (at the age 13) on experimental plot was high (93.2%). The influence of genotype (provenance) on changeability was not significant. This shows on good adaptation of tested progenies to conditions of Carpathian Foothills. The oaks of Krotoszyn and Opole provenance were characterized by good growth (respectively 4.80 and 4.75 m). Differentiation between families was higher in comparison to provenances level, with high value of family heritability (0.74). This suggests larger role of individual selection.

In assessment of shoot morphology the classification proposed by Krahl-Urban (1959) for young oaks was used. It includes three stem forms: axial, forked, and shaggy. In population of pedunculate oak tested on experimental plot 34.2% of trees had stems of an axial form, 45.2% of a shaggy form, and 20.6% of a forked form. The highest percentage of an axial form was found in the provenance of Opole (38.9%), and the lowest in the Milicz provenance (23.6%). A significant variation in proportion of respective forms on the "Chrostowa II" plot indicated the possibility to choice the provenances characterized by a grater percentage of good stem forms.

The observation of retention of leaves during the winter rest period was made using threedegrees' classification based on a visual estimation of the number of dry leaves remaining on a single tree. Percentages of trees in respective leaf retention classes varied. Taking the entire population into account the leafless oaks prevailed (62.5%), while the remaining trees were characterized by a low (19.3%) or a high (18.2%) occurrence of dry leaves. Among provenances the highest percentage of leafless oaks occurred in provenances Krotoszyn (78.6%) and Młynary-2 (74.5%), while the lowest in progeny of Opole (48.4%). The highest percentage of trees with a high occurrence of dry leaves was recorded for Opole (28.4%), the lowest for Krotoszyn (5.6%). On the provenance level a high tendency to a full shedding of leaves in winter was exhibited by the northern provenance (Młynary-2), and the provenance from western Poland (Krotoszyn).

# Some forest genomics applied to study of adaptation in Polish *Pinus sylvestris* L.

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Genetic diversity is the basis of the ability of organisms to adapt to changes in their environment through natural selection. Most of adaptation and growth traits are characterized by polygenic inheritance and they are an effect of interactions between two or more genes and their environment. Selection of forest trees should be directed on improvement of important economic traits and should be based on genetic knowledge.

The study presents an estimation of diversity of selection important traits such as survival and diameter of *Pinus sylvestris* L. populations tested in stand conditions of Beskid Sądecki. Also paper provides an overview on the use of genomics to research pine adaptation to environmental conditions of Polish Mountains.

#### Conservation of silver fir (Abies alba Mill.) gene resources in CarpathianGene Bank

#### Marta Kempf

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The Carpathian Mountains belong to the most interesting natural regions in Poland. Silver fir (*Abies alba* Mill.) is very important species in this area. Its share in the forest stand is assessed at the level of 25.0%. To protect and preserve of valuable stand of silver fir in 1999 created a security program to conservation of its gene resources called Carpathian Gene Bank. In this program 41 provenances are tested on nine experimental plots localizeted in Carpathian.

The study investigated the assessment of the adaptive traits (survival rate and height growth) of 10-year-old silver fir on conservation plots of the Carpathian Gene Bank located in the territory of Wisla, Kroscienko oraz Bircza forest districts. Present studies revealed a significant variation of tested characteristics in the juvenile growth age of protected provenances. A significant effect of genotype on the majority of analyzed characteristics was observed. Calculated values of inheritance of tested traits were high indicating a possibility of enhancing the genetic gain of forest propagation material through juvenile selection.

### Isoenzyme polymorphism in progenies of Norway spruce from some Krutzsch regions of

#### IPTNS-IUFRO 1964/68 provenance test in Krynica

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The genetic structure of Norway spruce [*Picea abies* (L.) Karst.] provenances on the IPTNS-IUFRO 1964/68 experimental plot in Krynica were studied. Two hundred and fifty trees from fourteen Krutzsch regions were analysed by using seven isoenzyme systems. No geographical grouping was observed using these types of markers. Parameters of genetic diversity including mean number of alleles per locus and observed heterozygosity were similar to values obtained for whole range of Norway spruce occurrence in Europe and were equal to 1.4675 and 0.1162, respectively. The spruces from Belarus were characterized by the highest genetic polymorphism while trees from south – eastern Styria - the lowest. Values of Wright's inbreeding coefficient varied from -0,4167 for Romanian provenances to +0,2233 for spruces from 28 Krutzsch region (Tyrol – Salzburg, Austria).

#### DNA variation of Scots pine (Pinus silvestris L.) populations from IUFRO 1982 experiment

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Random Amplified Polymorphic DNAs (RAPD) technique with single primers was tested for its usefulness in genetic distance estimation and population studies in the coniferous species of *Pinus silvestris*. DNA markers allow for direct analysis both coding and noncoding regions of the genome. The technique of detection DNA variations using RAPD markers has become a popular tool in genetic studies.

Needles for DNA analysis were collected from 60 individuals from 15 provenances (four individuals per provenance). An initial screening of four trees for each population was undertaken to ascertain which primers (from a set of decamer oligonucleotide) would produce polymorphic PCR products. Different reaction conditions were tested in order to get the optimal resolution of fragments and specificity and reproducibility of patterns.

In this preliminary investigation, a high  $MgCl_2$  concentration (5,5 mM) together with a low primer concentration (0,2  $\mu$ M) in the polymerase chain reaction (PCR) mixture yielded the best amplification products. Amplified fragments were scored as the presence or absence of a fragment. The analyses were performed to express (in the form of dendrograms) the relationships among the populations and genetic variability detected.

We have also examined quantitative and qualitative traits in the provenance experiment to test genetic correlation among traits in population from several distinct areas.

#### Conservation of forest genetic resources in western Ukraine

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The system work concerning the conservation of forest genetic resources has been started in the early 80th years last century in Ukraine. Most of the objects in situ and ex situ measures were selected just at that time and they are concluded in national register.

The area of 497 genetic reserves was nearly 26 000 ha and 3079 ha of plus stand as well as 4065 plus trees were selected, 1569 ha of archive, clone seed orchards and the family plantation were established, 406 ha of provenance and progeny trial were planted at the middle 90-th. A large part of them is located in western Ukraine.

An inventory, survey and research of in situ and ex situ conservation units has been performed, especially in the western region, as well as in Ukraine in general. These elaborations were supported by EUFORGEN and Ministry of Finance of Luxembourg for the last decade. It allowed to make the following generalizations: (1) the network of genetic reserve forest tree species isn't optimal: regarding the number, area and list endangered, vulnerable and rare species; (2) extension of such network is appropriate by selection of new reserves of social broadleaves, in particular *Quercus robur* and *Quercus petraea* and noble hardwoods (*Prunus avium, Sorbus torminalis, Ulmus* sp.); (3) it would be useful to optimize structural and spatial organization of conservation units in situ, by zoning of the area unit into core and buffer zones; (4) it is necessary to increase the minimum area of core genetic reserve of main forest species up to 10 - 12 ha; (5) would be needed to improve national legislation of the conservation of forest genetic resources; (6) the effective use of forest genetic resources could be increased by more fully involving them in seed and genetic-breeding programs; (7) it would be useful to come from passive principles to active management methods of conservation units; (8) the conservation of genetic diversity has to be associated with sustainable forest management.

Draft of "Concept for the conservation and sustainable utilisation of forest genetic resources in Ukraine" and "Regulations for establishment, conservation and sustainable use of forest genetic resources in Ukraine" can be considered as the practical steps of these proposals implementation.