A Programme for the Management of Forest Tree Genetic Resources in the Azores Islands

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Abstract. Since few forest tree gene conservation activities have been carried out in the Azores islands with its unique flora, it is urgent to develop a strategy for the management of the Azorean forest tree genetic resources. Safeguarding the potential for adaptation was identified as the prime objective for gene conservation of tree species in the Azores. Three species were identified of potential interest for tree genetic improvement. *Prunus lusitanica spp azorica* was selected due to its rarity and is close to extinction. *Piconia azorica* and *Juniperus brevifolia spp azorica* were selected based on their high wood quality and the interest in these trees by the Azorean Forest Service. Combined gene conservation and breeding was suggested for these species. The breeding of all of these species will follow the Multiple Population Breeding System concept. Open-pollinated seed will be collected for establishment of seedling seed orchards for each ecogeographic zone (cf Figure 4). Culling of trees with undesired characteristics will be carried out before seed collection. Seeds will be used for establishment of new progeny plantations. For *Prunus lusitanica spp azorica* only 1-2 seedling seed orchards are suggested. For the two other species several seedling seed orchards are suggested. For tree species not included in the breeding programme, *in situ* subpopulations are recommended. They should cover the entire range of distribution of these tree species. Whenever two or more species coexist, combined gene resource populations could be used to reduce the cost for conservation. Promotion of flowering by cutting competing tree species should be carried out to guarantee regeneration of the gene resource populations. Supportive research for the management program is urgently needed.
Key words: *Prunus lusitanica spp azorica*; *Piconia azorica*; *Juniperos brevifolia spp azorica*

Um Programa para a Gestão dos Recursos Genéticos de Árvores Florestais nas Ilhas dos Açores

Sumário. A flora única do arquipélago açoriano e os esforços diminutos para a sua conservação justificam que se desenvolva urgentemente uma estratégia para a gestão dos recursos genéticos florestais. Como objectivo primordial na conservação genética das espécies florestais dos Açores foi considerado a salvaguarda do seu potencial de adaptação. Foram eleitas três espécies endémicas para melhoramento florestal: o *Prunus lusitanica spp azorica* foi seleccionado por ser

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uma espécie de ocorrência muito rara e perto da extinção; o *Piconia azorica* e o *Juniperos brevifolia spp azorica* foram seleccionados com base na excelente qualidade da sua madeira e no interesse demonstrado pela Direcção Regional dos Recursos Florestais dos Açores. Para estas espécies foi sugerido um projecto combinado de conservação genética e melhoramento, o qual seguiria o conceito de "Multiple Population Breeding System". Por cada zona ecogeográfica deveriam ser colhidas sementes de polinização livre para a instalação de pomares produtores de sementes (Figura 4). O desbaste de árvores com características indesejáveis seria efectuado antes da colheita de sementes. Estas seriam usadas para o estabelecimento de novas plantações. No caso do *Prunus lusitanica spp azorica* sugere-se o estabelecimento de apenas um ou dois pomares produtores de sementes. Para as outras duas espécies sugere-se a instalação de vários pomares produtores de sementes e para espécies florestais não incluídas no melhoramento, recomenda-se a selecção de subpopulações *in situ*. Estas devem cobrir totalmente o habitat ocupado por estas espécies florestais. Quando coexistirem duas ou mais espécies podem ser combinadas subpopulações de recursos genéticos, para reduzir os custos de conservação. Para favorecer a floração, garantindo a regeneração, sugere-se o desbaste das espécies florestais concorrentes. A gestão deste projecto requer que seja desenvolvida urgentemente investigação de apoio.
Palavras chave: *Prunus lusitanica spp azorica*; *Piconia azorica*; *Juniperos brevifolia spp azorica*

Un Programme de Gestion des Ressources Génétiques de la Forêt des Açores

Résumé. Peu d'études sur la conservation des gènes des arbres de forêt ont été menées dans les îles des Açores, où la flore est unique; il est donc urgent d'y développer une stratégie de gestion des ressources génétiques des arbres forestiers. La sauvegarde du potentiel d'adaptation est considérée comme premier objectif de la conservation des gènes des arbres forestiers. Trois espèces ont été retenues pour l'intérêt de leur potentiel d'amélioration. *Prunus lusitanica* a été choisi car il s'agit d'une espèce rare et proche de l'extinction. *Piconia azorica* et *Juniperus brevifolia* ont été sélectionnées pour la bonne qualité de leur bois et pour l'intérêt que leur porte le service forestier des Açores. Une culture associée à la conservation des gènes a été proposée pour ces espèces. La culture de chacune d'elles suivra le concept de Multiple Population Breeding System. Dans chaque zone éco géographique, des graines issues de pollinisation ouverte seront collectées afin de planter de nouveaux vergers producteurs de semences (cf. Fig. 4). L'abattage des arbres aux caractéristiques indésirables sera réalisé avant la collecte des graines. Les graines seront utilisées pour les plantations de la génération suivante. Pour *Prunus lusitanica* seul un à deux vergers producteurs de semences sont suggérés. Pour les deux autres espèces deux sont proposés. Pour les essences d'arbres non inclues à la culture, des sous populations *in situ* sont recommandées. Elles devraient couvrir toute la gamme de distribution de ces espèces. Si deux espèces ou plus coexistent, des populations de ressources génétiques combinées pourraient être utilisées pour réduire les coûts de conservation. La floraison devrait être accélérée en abattant les arbres compétiteurs afin de garantir la régénération des populations de ressource génétique. Une recherche approfondie sur le programme de gestion se révèle nécessaire.
Mots clés: *Prunus lusitanica spp azorica*; *Piconia azorica*; *Juniperos brevifolia spp azorica*

Introduction

The essence of forest tree gene conservation can be phrased in the following way

The methods of gene conservation should ensure that the objectives in gene conservation are fulfilled while taking the structure and dynamics of the species into account. Since long-term breeding is one option for gene conservation it may be more accurate to describe gene conservation in broad sense as management of forest tree gene resources. As an

example, GRAUDAL *et al.* (1997) stated that a breeding program, which covers a large area of the distribution of that species, is of great significance for gene conservation.

Dynamics of any species is dependent on the interaction of the evolutionary factors. Similarly, understanding of the dynamics is of significance for identifying the objectives of gene conservation and for development of gene conservation methods. Before a discussion of evolutionary factors, gene conservation objectives, and gene conservation methods is carried out: a brief introduction to the forest of the Azores islands will be given.

Geography and climate

The Azores is an archipelago constituted by 9 islands in the North Atlantic Ocean with an area of 2.333 km². There are three groups of islands, Flores and Corvo compose the western group, in the central group there is Faial, Pico, São Jorge, Graciosa and Terceira, in the closest group to the mainland there are two islands, São Miguel and Santa Maria. The latitudinal range is 36º55' - 39º42'N, and the longitudinal range is 25º - 31º30'W. All islands are of volcanic origin, resulting in a large variation of hills and valleys.

The islands have an oceanic climate although with a considerable increase of precipitation as we move from São Miguel (751 mm) to Flores (1592 mm). These figures are valid for sea level. The precipitation increases approximately 25% for each 100 m of altitude (SJÖGREN, 2001). The temperature varies from an annual minimum of 14ºC to a maximum of 24.8ºC. There is a high level of humidity with an annual average of 77%.

Flora

SJÖGREN (2001) gave an introduction to the flora of the Azores islands, in which he stressed the unique character of this flora with many endemic vascular plants. The autochtonous forest of the Azores islands is frequently referred to as *Laurisilva* and it contains several tree species such as Cedar (*Juniperus brevifolia*), English holly (*Ilex perado ssp.azorica*), Sheepberry (*Viburnum tinus, ssp.subcordatum*), Scotts heather (*Daboecia azorica*), Tree heath (*Erica scoparia, ssp.azorica*), Wildberry (*Vaccinium cylindraceum*), Spurge-flax Daphne (*Euphorbia stygiana*), "Pau Branco" (*Picconia azorica*), "Faia" (*Myrica faia*), Morello wild cherry (*Prunus lusitanica ssp.azorica*), several of them facing serious threats.

Most of the autochthonous species exist in the four archipelagos Azores, Cape Verde, Canarias and Madeira; this group of archipelagos is called Macaronesia. However, there are several species endemic to Azores islands only.

The main economical activity is farming with the raising of dairy cows and high quality beef, even some of the steepest slopes are used for grazing year round. This lead to a destruction of the native forest converting it to pastures in all the nine islands, with a stronger impact in some of them. The construction of roads enabled and promoted the planting of large areas with the imported species, *Cryptomeria japonica*, which is now the most abundant species in the Azores. There is also some production of *Eucalyptus globulus*, but this species has probably a gloomy future since the cellulose companies gave up their forestry areas in Azores. There are species that were introduced as garden

trees and now are invading and competing with the natural forest, like the *Acacia melanoxylon* and the *Pittosporum undulatum*. They are probably the most devastating ones with a very fast growth and regeneration combined with their ability to grow in very poor soils.

There are many reasons why investments should be focused on autochthonous species. One important reason being the risk with introduction of species like those mentioned above. The wood of some of the autochthonous species is quite valuable such as *Juniperus brevifolia ssp. azorica, Prunus lusitanica L. ssp. azorica, Persea indica, Piconia azorica*. These species are already adapted to the Azores climate, so problems with maladapted species will be reduced or eliminated.

One of the questions that may be raised is "For which market will the wood be produced"? The answer is that it will be for the local market in Azores as well as for export to the mainland. Contrary to the *Cryptomeria* or *Eucalyptus* characterized by low-value wood, some selected autochthonous tree species produce quality timber much appreciated for furniture. Generally it is always useful to export refined products with a higher economic value than raw material.

The autochthonous species will probably not grow as fast as some introduced species, but still the income is expected to be larger than compared with fast growing species. There is also another economical factor favouring the autochthonous species, they have priority to be supported by funding through EU. This is of particular relevance now with the new CAP (Common Agricultural Policy) measures

that subsidise by area and not occupation, which previously was favouring an intensive farming. This will be a help to attract forest owners to invest, but with the kind of forest use that is going on now it will not be enough. Therefore, it is urgent that the State increases the awareness of the value of the existing forest resource of the Azores islands. Without such awareness no responsibility for regeneration may be taken.

We are preserving our forest patrimony, that will attract more tourists interested in ecology and natural flora to come and see our endemic forest, something that they will not do with an island covered with a tree from Japan or Australia. Tourism will be one more source of income to the islands and will increase the importance of a project concerning the autochthonous species.

In conclusion, the main problem in Azores, which is shared by many places in the world, is that abusive use by man of our forests for over 5 centuries has led to a substantial reduction of the native forests.

Evolution

The mode of inheritance of adaptive traits is of importance for the speed of evolution. Traits regulated by major genes can evolve much faster than traits regulated by minor genes. The latter have each a small effect on the trait. Most traits of significance for adaptation in forest trees are quantitative, and as such regulated by many genes. These traits mostly show normal distribution with a bell-shaped curve (Figure 1).

Different authors interpret the terms adaptability, adaptation, and adaptedness differently. Therefore, the defini-

tions used in this paper are given in Box 1, taken from ERIKSSON and EKBERG (2001).

Evolution is influenced by four main factors: natural selection, genetic drift, gene flow, and mutations. The definitions of these four factors are given in Box 2, taken from ERIKSSON and EKBERG (2001).

As stated in the definition of natural selection this factor is a result of differential transfer of alleles and natural selection should not be regarded as an active force. Natural selection requires that different phenotypes exist and that these differences are genetically conditioned. After an extensive review of several papers on natural selection in the wild ENDLER (1986) concluded that natural selection varies from strong to weak.

Three main types of natural selection can be distinguished (Figure 1): stabilizing selection, in which individuals in both tails of the distribution are selected against, directional selection, in which individuals in one of the tails of the curve are favoured by the existing conditions, and disruptive selection when individuals in both tails of the curve have the highest adaptedness for the existing conditions.

Genetic drift is of greatest significance in small populations. This will consequently lead to inbreeding and homozygozity by loss or fixation of alleles. This factor is of great relevance and has to be controlled in breeding or gene conservation projects, but is probably of minor significance in wind-pollinated species with large populations.

Box 1 - Definitions of adaptability, adaptation, adaptedness, and fitness
Adaptability = the ability of a population to respond genetically or phenotypically to changed environmental conditions
Adaptation = the process that leads to a better adaptedness in a specific environment
Adaptedness = is the degree to which an organism is able to live and reproduce in a given set of environments
Fitness = is an expression for one individual's contribution to the next generation in relation to the other individuals in the same population

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Box 2 - Definitions of evolutionary factors
Mutations = change of genes
Natural selection = differential transfer of alleles to the next generation resulting in increased fitness
Random genetic drift = random loss of alleles in small populations
Gene flow = migration to a recipient population from another population with a different allele frequency
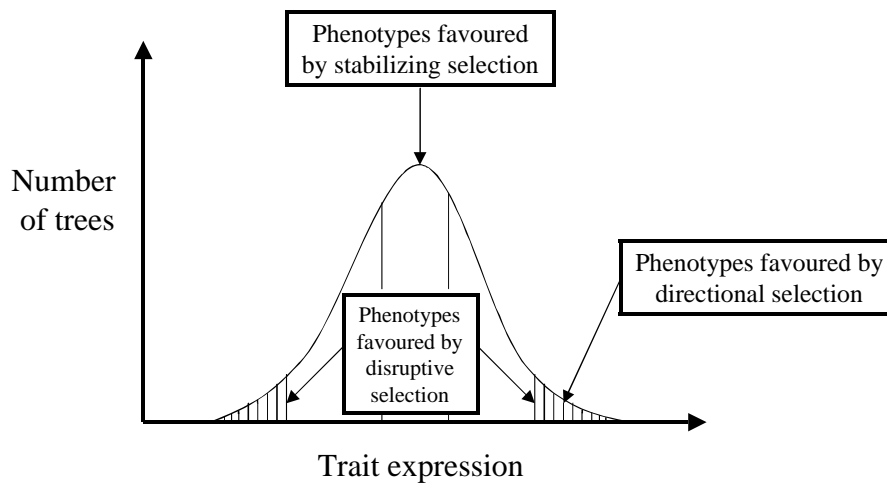
Figure 1 - Bell shaped curve representing the normal distribution of traits. The arrows indicate which individuals in the curve will be favoured in each of the three most relevant kinds of natural selection

Mutations are errors that occur during the process of DNA replication or induced physically or chemically by mutagens at a very low rate, 1:10 thousand to 1:1 million, so this is not a main factor in population differentiation. Most of these errors are largely with reduced fitness leading to poor survival of the individual carrying the mutation, but if the mutation is a minor defect, individuals may survive to the next generation.

Gene flow is mediated by pollen, transfers of seeds, fruits, nuts or vegetative propagules and can be a strong evolutionary factor. One migrant per generation is enough to prevent fixation of neutral alleles (SLATKIN, 1987).

Besides these four evolutionary factors, recombination ought to be touched upon since it leads to creation of new genotypes. Recombination does not change the gene frequency directly, only indirectly, creating new options for change by natural selection or genetic drift.

In Figure 2 it is visualised that disruptive natural selection, genetic drift and to some minor extent mutations, will raise the level of differentiation among populations. Stabilising selection within populations is the most common type of selection in nature. At the species level this kind of selection will be experienced as disruptive selection between populations (cf ERIKSSON, 2001). Since alleles are fixed in a random way by genetic drift, two populations will not have the same set of alleles fixed. Genetic drift thus promotes differentiation among populations. Mutations occur at a very low rate, and the chance of the same mutation occurring in two different populations is practically nil, so the populations will differ from each other in relation to mutations, this is the reason why we can see mutations as a promoter, but a very weak one, in Figure 2. Gene flow causes a reduction of the variation between populations and thus constitutes a constraint to population differentiation.
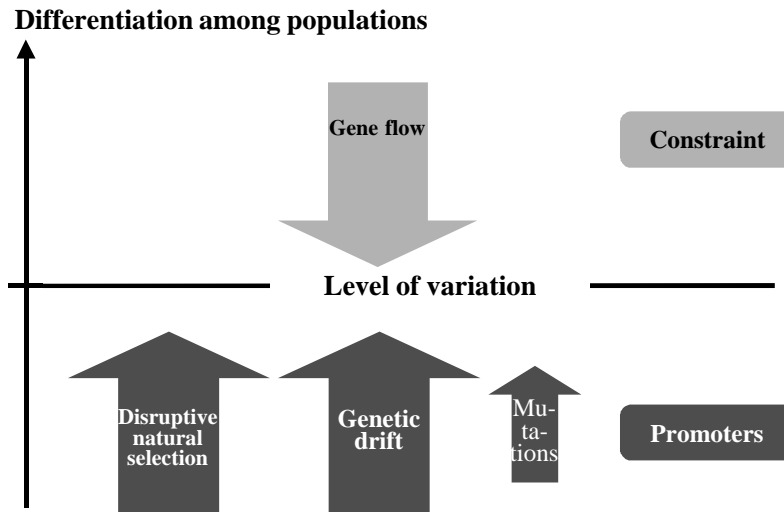
Differentiation among populations

Figure 2 - The influence of the four evolutionary factors on differentiation among populations. Arrows pointing upwards increase the differentiation and thus are promoters of differentiation, while the downward pointing arrow reduces the differentiation among populations and thus it is a constraint

The four evolutionary factors interact in a complex way. Therefore, it is impossible to know which factor is most important in one specific situation. It is of greatest significance to stress that the present genetic constitution is never or very rarely dependent on natural selection alone. The hypothesis that many genotypes may give rise to the same quantitative trait phenotype means that the belief that the present genetic constitution is the one and only good constitution is false.

As can be seen from Figure 3 the within-population variation constitutes a contrast to Figure 2, in this case gene flow leads to a higher genetic variation, while stabilizing natural selection, genetic drift and inbreeding will repress it. Gene flow will bring new alleles from other populations, which increases the variation, the same happens with mutations that will generate new alleles. Stabilizing selection will reduce variation through the favouring of the existing genotypes at the centre of the distribution curve, genetic drift and inbreeding are known to reduce variability and lead to homozygozity.

Phenotypic plasticity is sometimes seen as an evolutionary factor (BRADSHAW, 1965). It allows plants with the same genotype to express different phenotypes in different environments. Phenotypic plasticity may contribute to fitness in long-lived tree species, which are experiencing large changes in their environment during their lifetime (ENDLER and MAC LELLAND, 1988). However, phenotypic plasticity may be regarded as a disguise of the genotype and in this way it is a constraint to natural selection.
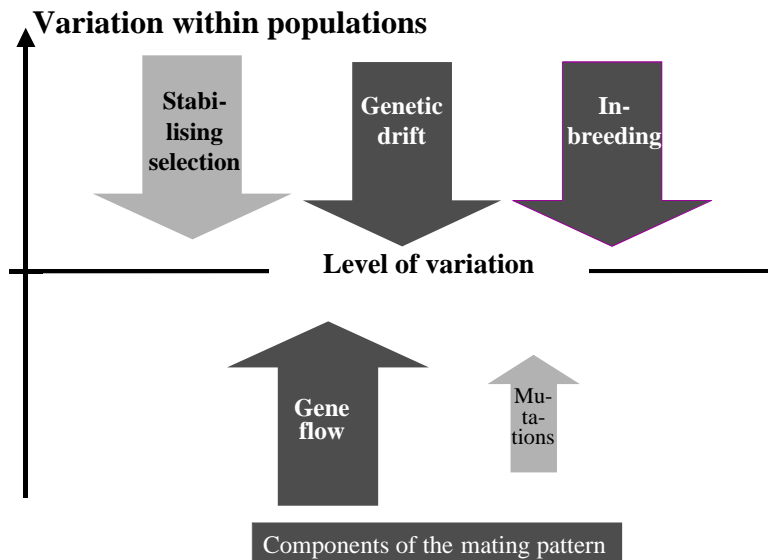
Variation within populations

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Stabi-        Genetic        In-
lising         drift        breeding
selection

                Level of variation


          Gene              Mu-
          flow              ta-
                            tions

          Components of the mating pattern
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Figure 3 - The influence of the four evolutionary factors on within-population variation. Arrows pointing upwards increase the variability and thus are promoters of within-population variation while the downward pointing arrows reduce the variability within the population and thus are constraints

Management of gene resources

The essence of management of forest tree gene resources was outlined in Introduction. Independent of the phrasing of the activity as gene conservation or management of gene resources, it is important to define the meaning of gene resource populations and to identify and rank objectives in management of gene resources. Gene resource populations were defined as *the seeds, acorns, nuts, plants, or trees that are included in the gene conservation* by ERIKSSON and EKBERG (2001).

Objectives

SOULÉ and MILLS (1992) stated that *Conservation genetics exists for one reason only: To promote the fitness of targeted populations.* This is in good agreement with the statement by ERIKSSON *et al.* (1993) that the prime objective of forest tree gene conservation is *creation of good conditions for future evolution.* Since genetic drift may lead to evolution this objective was later reformulated as safeguarding the potential for adaptation (ERIKSSON, 2001). The reason for identifying this objective as the prime objective in gene conservation is that long-lived forest trees are exposed to continuous changes during their lifetime and their long-term survival and thriving depends on their genetic ability to cope with these changes, *ie* that they have the potential for adaptation. It ought to be observed that phenotypic plasticity may be useful for survival during a single generation but may be a constraint to adaptation in a multi-generation

perspective. In many instances the breeding objectives of forest trees are to develop well-adapted material to the prevailing growth conditions and therefore breeding and gene conservation share the same objective.

ERIKSSON *et al.* (1993) reported that the objectives of many forest tree gene conservation programs were not explicitly expressed. However, it seems, as the present genetic constitution was the target in many conservation programs. In the paper by ERIKSSON *et al.* (1993) it was argued that the present genetic constitution was transient and one of several possible and for this reason it should not be the objective in gene conservation. Rather it should be the starting material for gene conservation. Simulations by HOLSINGER (1993) lend support to the arguments by ERIKSSON *et al.* (1993).

To estimate breeding gains plant breeders have benefited from comparisons of modern varieties with old cultivars. Even if such an objective seems less likely in tree breeding it cannot be excluded that tree breeders would find it useful to have a reference material for comparisons in the future. Thus preservation of the present genetic constitution becomes one objective.

VARELA and ERIKSSON (1995) argued for inclusion of gene conservation of associated species to tree species as one important conservation objective. The meaning of associated species is a species dependent on another species for its existence. The tree species included in conservation are called target species.

Saving of endangered populations is frequently referred to as an objective in forest tree gene conservation (*e.g.* MELCHIOR *et al.*, 1986)

Methods

Of great significance in forest tree gene conservation is that the method used or suggested allows for a continued adaptation of the species. Therefore, gene conservation must be dynamic so that species can benefit from natural or artificial selection and thereby reach higher adaptedness to its environmental conditions.

Gene Namkoong, a leading forest geneticist, introduced the MPBS concept during the 1970s (NAMKOONG, 1976), to prepare the breeding population for changed breeding goals as well as changing environmental conditions. It was later elaborated to encompass gene conservation objectives (NAMKOONG, 1984). The *Multiple Population Breeding System* (MPBS) is a method that not only allows for adaptation under changes in environment but also permits breeders to respond swiftly on possible changes in value of characters. It thus fulfils the condition given above for dynamic gene conservation with inclusion of maximum existing variation among populations.

MPBS means that the gene resource population is split into approximately 20 subpopulations each with an effective population size, N_e, of approximately 50. If the entire breeding population has 20 subpopulations the census number of trees would be 1000. With such a population size few alleles would be lost for random reasons unless they are extremely rare. Each subpopulation has a separate breeding goal, which could be the improvement of the same trait under a number of different site conditions or the improvement of a number of different traits. In gene conservation the meaning is rather adaptation to different environmental conditions of the different sublines. The split of the gene

conservation populations into several subpopulations allows for an increase of the total additive variance of the gene conservation population with marginal loss of within-subpopulation variance (ERIKSSON *et al.*, 1993). Thus, inbreeding will be on an acceptable rate because for an effective population size (N_e) = 50, the coefficient of inbreeding $F=1/(2N_e) = 0.01$ (VARELA and ERIKSSON, 1995).

There are different degrees of sophistication of MPBS, from the most intensive breeding to *in situ* gene conservation without any human interference. If the subpopulations originate from different site conditions it increases the possibility to include alleles, which are rare at the species level but are common in some subpopulations.

In summary, the main advantage of the MPBS is that it combines the capture of the total existing genetic variability with a satisfactory variability within each subpopulation and that it allows the gene resource populations to adapt to the prevailing environmental conditions. Another advantage is that the speed of evolution might be faster in a population of 50 trees than in one large population containing thousands of trees.

The most sophisticated MPBS involves sampling, plantation, management and regeneration of a net of gene resource populations located over a wide span of site conditions. There may be different evolution in each of the several small subpopulations since each subpopulation can be exposed to various types of climatic and edaphic conditions.

The Gene Resource Populations (GRPs) can be *in situ* or *ex situ*; the definition of these concepts differs dependent on the author. The definitions used in this paper are, *in situ* = already existing self-regenerated natural forests and *ex situ* = man made forests even if it is the same natural species in the same normal habitat.

Conservation *in situ* has its benefits because it keeps the target species in their natural habitat what will also benefit other species living in that habitat. With the conservation *ex situ* we are focusing on the target species, so other species will not benefit much from that, but a bigger variability for the target species will or should be present in this kind of conservation.

Any method must be classified according to how well it represents the existing genetic constitution and capacity to promote adaptation. In their analysis of different methods for gene conservation VARELA and ERIKSSON (1995) stated that the *ex situ* version of MPBS is one of the most elaborate methods. In the methods that use large subpopulations, 200-300 hectares, there are possibilities to include the conservation of associated species. Provenance and progeny trials can also be a good part of gene conservation if they represent the existing variation in a species. However, evolution is not promoted. Some static methods will conserve pollen, seed and tissue banks. The limited space and the low cost of these methods are advantageous. Unmanaged natural reserves are not fulfilling our aims unless preserving a structure of endemic species. Botanical gardens have just few individuals, so there is almost no value at all from a gene conservation perspective (VARELA and ERIKSSON, 1995).

Suggestion for management of the Azorean forest tree gene resources

Objectives and funding. The role of gene conservation in Azores islands is to

assure that the existing species will not become extinct in the near future. The objective of this project is to develop a strategy for conservation of the genetic material of the tree species in Azores, at the same time as we have some income from the breeding of the same species. Therefore, evolution will be favored in this project by allowing the species to cope with future changes in climate via adaptation. Some species are facing a great risk of extinction, like *Prunus lusitanica L. ssp. azorica*, to save this species will be a top priority.

The method to be used in this project must insure that a wide genetic variability is included and that the most important objective, to safeguard the potential for adaptation, is granted. The tree species in the Azores islands are suffering a reduction in their natural populations. Therefore, it is the responsibility of politicians to provide them with such a protection that future evolution is granted.

Support for the funding of this gene conservation ought to come from EU programs, the regional government, and also from breeding of the selected species.

Grouping of species. A separate gene management program for each species is unrealistic for economic reasons. This means that a grouping of species has to be carried out. One important factor for grouping is whether a species is commonly occurring or rarely occurring (cf ERIKSSON, 2001). Demand with respect to site conditions is another factor to consider in the grouping. The tree species to be included in the gene management program for the Azores islands are listed in Table 1. Besides the factors discussed above some other relevant biological and economic factors are given in this table.

Methods. For the commonly occurring species we suggest the use of *in situ* MPBS with low-intensity management. We will opt for the gene resource population *ex situ* if it does not require much higher cost, or if the species is included in breeding. The principle for selection of the subpopulations is shown in the schematic illustration in Figure 4. This suggestion is based on the climatic variation in east-west direction as well as in elevation. In addition to this, edaphic conditions should also be considered in the selection of subpopulations. Since farming dominates below 300 meters no subpopulations are suggested for lower elevation, where farming is dominating. Figure 4 also indicates that joint conservation of two species is recommended whenever there is a common distribution of the species. This suggestion may be extended to any number of species with common distribution.

Before a subpopulation is identified as a gene resource population we need to know the abundance of the selected species. Just the census number is not enough, since it will only give the total number of existing trees but for this project it is particularly important to know the number of trees that can reproduce. As a rule of thumb the census number must be at least three times larger than the required effective population size. Even if the requirement for a subpopulation is 150 trees the area of a subpopulation will in many cases not be large. For the time being the most probable problem for gene conservation in the Azores islands is the small size of the scattered populations/individuals that may lead to genetic drift. Terceira, Pico and Faial islands support the wider areas of still natural forest.
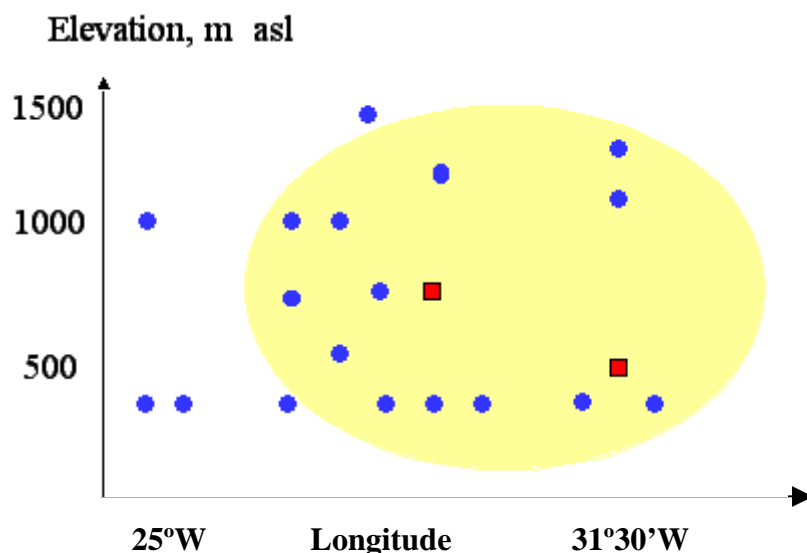
Figure 4 - Principle of how the gene conservation of tree species may be carried out in the Azores islands. Each dot represents a subpopulation in a gradient of longitude and altitude, the shaded area indicates that two species coexist in this area and joint gene conservation of the two species may occur. The potential location for two progeny trials for *Prunus lusitanica* is indicated by squares

If the requirement for an effective population size of 50 in each subpopulation is not met it is necessary to increase the population size to avoid genetic drift. Preferably, this should be done by planting material from similar site conditions in the subpopulations with too low N_e. If this is not possible, material from Madeira or Canary islands may be used.

For the endemic and rarely occurring species, *Prunus lusitanica*, *ex situ* measures are required to safeguard the potential for adaptation of this species in the five islands, in which it occurs. In this case material must be brought together in one or two clone archives or seedling plantations to increase the population size. Seeds produced in the clone archives or seedling plantations should

be used to generate a new generation of this species in the way suggested by KLEINSCHMIT (1994). It is important to stress that the trees in each subpopulation should as far as possible originate from similar site conditions.

As discussed above many measures need to be taken for gene conservation in Azores. The most important part is the regeneration of the gene resource subpopulations once the gene resource populations are identified. Promotion of flowering in the *in situ* subpopulations should be carried out by cutting trees competing with the trees of the target species. Another important issue is that strong protection measures must be applied to protect the still existing stands/individuals of native species and their habitats. This means that in some

cases the foreign species will have to be controlled to reduce the threat they constitute. A study must be done to determine if any other threats are endangering these species, for example it has been mentioned that *Armilaria* disease is attacking some stands of *Persea indica* (JORGE BELERIQUE pers. comm.). For the time being no studies have been made, that we are aware of.

Breeding project

The following tree species were selected for breeding:

Prunus lusitanica L. ssp. Azorica,

Piconia azorica spp azorica,

Juniperus brevifolia.

Prunus lusitanica was selected because it is one of the most endangered species in the Azorean flora. It has a valuable wood with a fast growth. *Piconia azorica* and *Juniperus brevifolia* were selected based on their good wood quality and the interest in these trees by the Azorean Forest Service. Since *Persea indica* may not be autochtonous it was not selected for breeding. The main breeding objective of all three species is to have fast growth combined with good wood quality. Furniture production is the expected use of the wood, which should be free of all kinds of defects such as spike knots, twisted wood, and without fungus attacks.

The principle for breeding is the MPBS concept but the number of subpopulations will be less than for gene conservation, which encompasses the entire distribution of the three species. No high elevation subpopulations will be included in the suggested breeding program.

For the extremely rare *Prunus lusitanica* only one or two progeny trials will be established on Terceira island. Seeds from open-pollination must be collected from all fruit-bearing trees in the five islands, in which this species occur. The seedlings produced should be planted in progeny trials that will be converted to seedling seed orchards after assessments have been taken.

For *Piconia azorica*, and *Juniperus brevifolia* seeds will be collected from plus trees with the desired traits. One progeny trial ought to be established in each of the eight islands, in which these species occur.

When the progeny trials of the three species have reached such an age that assessments are possible to carry out estimates of heritability and the coefficient of additive variance for traits selected for should be calculated. Based on the results from this evaluation, selection of superior trees in superior families will be carried out. Inferior trees will be culled. After culling the progeny trial is converted into a seedling seed orchard. Once seeds are available from the seedling seed orchards the seeds should be used in nurseries for propagation or for direct seeding by forest owners.

Tree breeding has to start without all genetic and biological knowledge required. A research support program ought to be started. First of all it is of significance to get estimates of genetic parameters for the traits selected for. This can be done by the aid of the progeny trials suggested above. Reproductive biology - including seed crop per tree, flower initiation, flowering phenology, seed ripening, pollen vector(s), pollen dispersal, and self incompatibility - is important for future design of the breeding.

Conclusions

For centuries the landscape and flora composition of Azores islands was affected by human activities. This led to endangering of some of the existing autochthonous tree species. Presently it has become more important to save those species for ecological reasons as well as moral reasons.

Like everything else there is a limit of investment that can be done without revenue, so conservation will be achieved in a way that produces income through a breeding program, and so increasing the possibilities of success in this project. It is of great importance that the selected species will have conditions matching the prime objective in gene conservation *to safeguard the potential for adaptation*.

Only three species were selected for combined breeding and gene conservation not because they were the only ones that could be used but because more than that would be unrealistic. One of the main factors in the selected species is that they must produce valuable wood in order to have the refunding that will support part of the project.

The species *Prunus lusitanica L. ssp. azorica* was selected for that reason and because it is one of the most threatened species in Azores. For this reason a more intense conservation project was designed for it to try to reduce the effect of genetic drift may have in this species. One or two seedling seed orchards are recommended for this species owing to its rarity.

As regards the other two species there is also a need for conservation, but not as strong as for *Prunus lusitanica L. ssp. azorica*. The conservation of *Piconia azorica and Juniperus brevifolia ssp. azorica* will be taken care of in the breeding part of the project, which will be carried out according to the *ex situ* version of MPBS. There is no need for an intense project for their conservation, in the meantime the breeding project must be careful enough to include all alleles in frequencies over 1%.

For the rest of the autochthonous tree species *in situ* MPBS gene conservation is suggested with subpopulations distributed over the entire range of distribution. Whenever species coexist combined gene conservation can be carried out. The associated species will benefit from the conservation of our autochthonous tree species. Reduction of exotic species will also be very important for the conservation program, presently exotic species constitute a threat to our natural flora.

The designed project needs to be flexible to be able to adapt to new conditions that may arise, and to new knowledge about these species that may come further on in the project.

Supportive research must be carried out to improve biological and genetic knowledge of the selected species. Not that many such studies were carried out so far.

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