

Strategies for the conservation of a pioneer tree species, *Populus nigra* L., in Europe

François Lefèvre^{*a}, Agnès Légionnet^b,
Sven de Vries^c, Jozef Turok^d

^a Unité de recherches forestières méditerranéennes,
Institut national de la recherche agronomique, 84000 Avignon, France

^b Station d'amélioration des arbres forestiers,
Institut national de la recherche agronomique, 45160 Ardon, France

^c IBN/DLO, P.O. Box 23, 6700 AA Wageningen, The Netherlands

^d IPGRI, via delle Sette Chiese 142, 00145 Rome, Italy

Abstract – The European black poplar is a pioneer tree species of the riparian ecosystem. Its natural habitat is exposed to anthropogenic alteration. Overexploitation of the trees, and interaction with a narrow-based cultivated gene pool also contribute to the decline of the species. National programmes for the conservation of *Populus nigra* exist in most European countries, and it was elected as one of the pilot species in the EUFORGEN programme. The strategies developed in 17 European countries over the species range, and 3 years of collaborative efforts within the EUFORGEN *P. nigra* Network are reviewed here. The conservation strategies need to be adapted to the biological characteristics and ecological requirements of black poplar. Applied conservation includes ex situ methods for the conservation of genotypes, and, for long-term gene conservation, in situ management of sites in relation to the preexisting natural reserves, or dynamic conservation in the framework of poplar breeding programmes. The social and cultural impact of poplars also interfere with applied conservation.
© Inra/Elsevier, Paris

Populus / genetic resources / conservation strategy

Résumé – Conservation d'une espèce forestière pionnière, *Populus nigra* L., En Europe. Le peuplier noir européen est une espèce pionnière de l'écosystème ripisylve. Son habitat est soumis à une forte altération d'origine anthropique. La surexploitation de la ressource, et les interactions des compartiments sauvages et cultivés de la peupleraie, participent également au déclin de l'espèce. Des programmes de conservation de *Populus nigra* existent dans la plupart des pays européens;

* Correspondence and reprints

leur coordination est assurée au sein d'un réseau du programme Euforgen pour la conservation des forêts en Europe. Une synthèse des stratégies de conservation adoptées dans 17 pays de l'aire de répartition de l'espèce est présentée, ainsi que les résultats de trois années de collaboration au sein du réseau Euforgen *P. nigra*. Les caractéristiques biologiques du peuplier noir en font un modèle original au sein des espèces forestières, les stratégies de conservation doivent donc être adaptées. Les programmes appliqués de conservation intègrent des méthodes ex situ pour la conservation de génotypes, et, pour la conservation de gènes sur le long terme, des méthodes de gestion in situ en liaison avec le réseau des réserves naturelles, ou une gestion dynamique en liaison avec les programmes d'amélioration génétique. La place socioculturelle du peuplier est également prise en compte dans ces programmes. © Inra/Elsevier, Paris

***Populus* / ressources génétiques / stratégie de conservation**

1. INTRODUCTION

Populus nigra L., the European black poplar, has a wide distribution area ranging from the Mediterranean border in the south to 64° latitude in the north, and from the British Isles to western Asia (Zuffa, 1974). It is a pioneer tree species of the riparian ecosystem, strictly heliophilous, which forms metapopulations by colonising open areas through seeds and propagules (cuttings, root suckers) (Zuffa, 1974; Herpka, 1986). *P. nigra* is characterised by a great diversity of population types, from isolated trees to huge pure or mixed stands. Individual trees may live over 400 years (Popivshchy et al., 1997). The species is dioecious and anemogamous. Seeds have a very short viability (Muller and Teissier du Cros, 1982), they are disseminated through wind and water, and need strict water-soil conditions for germination. Regeneration is generally poor within old established stands; the riparian forest naturally evolves towards hardwood formations.

P. nigra is of economic interest as a pure species. It is widely planted in Turkey for domestic use: 60 000 ha of row plantations, compared to 70 000 ha of hybrid plantations for industry (Tunçtaner, 1995). Due to its plasticity, *P. nigra* is also used as a pure species for soil protection, and afforestation in polluted industrial zones (Popivshchy et al., 1997). *P. nigra* was extensively planted to fix moving sands on the Great Hungarian Plain at the beginning of the eighteenth century, and was then replaced by the so-called euramerican hybrids, *Populus deltoides* × *P. nigra*, in the twentieth century (Toth, 1995).

However, the species is mainly used as a parent pool for poplar breeding programmes. *P. nigra* is hybridised with *P. deltoides* and other exotic *Populus* species, providing adaptability to various soil and climate conditions, rooting ability, high resistance to *Xanthomonas populi*, fair resistance to *Marssonina brunnea* and to poplar mosaic virus (Cagelli and Lefèvre, 1995).

The European black poplar was recognised as a priority species for international collaborative activities on forest genetic resources in Europe. A political framework for strengthening gene conservation activities was created by adopting Resolution 2 at the Strasbourg Ministerial Conference on the Protection of Forests in Europe in 1990. The follow-up committee then organised an international survey on the status and perspectives of forest genetic resources in European countries and suggested collaboration in four 'pilot' networks of

the European Forest Genetic Resources Programme (EUFORGEN), including *P. nigra*. The programme focuses on providing guidance for the development of national policies and encouraging long-term activities in the participating countries.

With regard to *P. nigra*, the objectives assigned to the genetic conservation vary from one country to another, and include:

- ensuring potential for the natural adaptation of the species;
- providing basic material, genes or genotypes for further breeding;
- maintaining the diversity and stability of forest ecosystems;
- preventing loss of unique native stands characterised by high productivity and stability.

The EUFORGEN *Populus nigra* Network started its activities in 1994. Network members from 17 countries meet regularly to exchange information and experiences, coordinate the activities in individual countries, identify common needs and priorities, develop and implement joint tasks of their work plan, prepare collaborative projects and promote awareness at national and international levels.

The aim of this paper is to give an overview of the threats to *P. nigra*, to summarise the current knowledge of the species' genetic diversity and to review critically the static and dynamic approaches to conservation from a European perspective. The role of international collaboration and the outputs provided by the network are mentioned. Particular attention is given to the need to raise public awareness. Perspectives of coordinated research and further practical conservation activities are suggested.

2. THREATS AND THEIR INDICATORS

The situation for *P. nigra*, which is still found over a wide area and locally shows a remarkable colonising potential, may not seem as critical as for highly endangered forest tree species such as *Abies nebrodensis*, for which only 29 individuals are left (Vendramin et al., 1997; Venturella et al., 1997), or *Ulmus* sp., under high parasitic pressure (Collin, 1998). However, three main factors threaten black poplar in Europe. The first is the alteration of the riparian ecosystem, all over the species range. With hydraulic engineering, native poplar stands were displaced by agriculture and other human activities (including poplar plantations). Moreover, in the still wild areas, the regulation of floods has favoured the succession of poplar stands by hardwood forests. Second, the autochthonous black poplar resource has been overexploited. *P. nigra* was deliberately removed from Danube flood plains forest at the beginning of the century because of poor wood quality, then replaced by hybrid poplars (Heinze, 1997). In Bulgaria, very old *P. nigra* trees were overexploited because of specific wood designs (Tzanov, personal communication). Finally, gene introgression from cultivated poplars is a potential threat for *P. nigra*, in the sense that very few clones are extensively cultivated and contribute to a large extent in the pollen and seed pools. Not only exotic hybrids are concerned, but also pure *P. nigra* varieties like the male tree 'italica' distributed all over the continent (Cagelli and Lefèvre, 1995). In addition, the cultivated poplars seem to alter the preexisting pathogen populations (Pinon and Frey, 1997). The decline of *P. nigra* is evidenced by different types of indicators.

The first type of indicator is given by inventories which show scattered distribution patterns of black poplar in many countries, particularly in Western Europe. *P. nigra* is one of the most endangered trees in Belgium: it has totally disappeared in the Meuse Valley (van Slycken, 1995). A similar situation is found in the Netherlands and Greece (de Vries, 1995, Panetsos, personal communication). In the United Kingdom, the total number of remaining black poplars is estimated at no more than 2 000 to 3 000 trees; moreover, the sex ratio is exceptionally reduced to one female for seven male trees, probably due to the removal of cotton-producing trees, and only 150 female trees are known, which represents a drastic reduction of effective population size (White, 1993; Tabbush, 1996a). The decline of the *P. nigra* populations in Austria along the Danube has been noticeable since the 1960s (Heinze, 1997). Large stands of flowering trees are not known in these countries.

Another indication is provided by population dynamics. In the previous countries particularly, natural regeneration is exceptional due to the lack of suitable condition areas for germination. In the United Kingdom, a specific inventory made in the 1970s revealed that present populations consist mainly of planted cuttings, in the flood plains where species are assumed to have been native (Tabbush, 1996a). Additionally, the follow-up of an experimental plot in a reserve area from 1987 to 1994 showed a significant decline of *P. nigra* trees in terms of basal area, which was not observed for other riparian tree species such as *Fraxinus* sp. or *Tilia* sp. (Pont, 1995).

Finally, the third category of indicators results from the analysis of diversity and its predictable evolution. The situation in the United Kingdom illustrates the risk of species extinction in the margins of its range area, and subsequent loss of singular genotypes. In a genetic study based on isozymes, Legionnet and Lefèvre (1996) found a lower diversity (mean expected heterozygosity index H) in French populations than in Central European accessions, which also indicates a lower diversity in the marginal part of the species range. Moreover, Legionnet (1996) found evidence of surprisingly limited gene flow. The mechanisms remain unknown, but the author proposed a pessimistic scenario where, given the genetic load that we can expect for such an allogamous species, the fragmentation of populations resulting from human activity would increase inbreeding and subsequently reduce the mean fitness of individual genotypes. Further study of black poplar population dynamics is deeply needed in order to clarify the main features of the evolution of the species.

3. DESCRIPTION OF THE GENETIC DIVERSITY

As a preliminary step for conservation, the description of existing diversity is needed:

- to describe the resource, and evaluate the efficiency of the conservation activities;
- to identify the areas of interest for conservation: where the species has rarefied, where it can be maintained, where it could be re-introduced;
- to define a sampling strategy.

3.1. Demographic and ecological survey

Two difficulties are encountered. First, the many isolated individuals or small linear groups of trees are generally not considered in the forest inventories. Second, the occurrence of *P. nigra* in mixed stands of secondary economic interest does not allow a detailed identification of the species ('poplars' often group *P. nigra* with *Populus alba*, aspens and cultivated hybrids if any). Therefore, rough data are available for most of the countries, but the actual situation is hardly estimated. In Hungary, experts consider that only 10 % of the area officially registered as autochthonous poplar really corresponds to the native black poplar (Toth, personal communication). Nevertheless, we can imagine that information such as 100 or 200 ha for native black poplar in Bulgaria and Moldova (Tzanov, 1995; Postolache, 1997) is clear evidence of a reduced area.

Inventories specifically dedicated to black poplar are still exceptional. In the United Kingdom, the objective is to record and precisely map 90 % of the remaining trees within 5 years (Tabbush, 1996a). This work is done in Belgium, the Netherlands, Slovakia and the Czech Republic on a national basis (van Slycken, 1995; de Vries, 1995; Mottl et al., 1997; Varga, 1997), and in Italy on a regional basis (Cagelli, 1995). This work was done in Germany in 1963, but data could not be updated since further inventories grouped *P. nigra* with other deciduous trees (Schulzke, 1995). In Spain, inventory of both individual trees and stands started recently (Alba, 1995). A similar approach is planned in France (Lefèvre et al., 1996).

Successive inventories would provide a precise evaluation of the situation. One example is given in Hungary where *P. nigra* decreased from 20 % of total poplar area in 1953, to 13 % in 1957 and 3 % in 1973 due to the extension of hybrid poplar, although the '*P. nigra*' area remained quite stable, at 4 452 ha in 1957 and 4 477 ha in 1973 (Toth, 1995). This measures the development of the cultivated poplar pool (remembering that probably 10 % only of the so-called native '*P. nigra*' is the true species). Five years' delay between two surveys should provide preliminary information on the short-term evolution of the resource (Lefèvre et al., 1996).

Ecological maps of soil and climate conditions revealed that most of the sites suitable for poplars along the Danube have been dedicated to hybrid plantations (Heinze, 1997).

3.2. Assessment of genetic diversity

Advanced generations of introgression, if they do occur, would hardly be detected in the field on the basis of morphological traits. Species identification can be completed in the laboratory by isozymes (Legionnet, 1996; Mottl et al., 1997) and deoxyribonucleic acid (DNA) markers (D'Ovidio et al., 1991; Faivre Rampant et al., 1992a, b). Preliminary results suggest that introgression might be counterselected in the young seedling stage (Legionnet, 1996), but further investigation is needed. This identification is complicated since the taxonomy of the genus *Populus* is currently being re-examined using DNA markers (Smith and Sytsma, 1990; Eckenwalder, 1996).

Mean diversity for isozyme data in *P. nigra* corresponds to other Angiosperm tree species ($H_e = 0.17$; Legionnet and Lefèvre, 1996). Like other forest tree

species, very little differentiation is found with isozymes between stands ($Gst = 0.06$) or between rivers ($Gst = 0.03$). However, the stands do differentiate somewhat: contrasted diversity values are found in neighbouring populations in France, and alleles can be undetected in a stand while they are regularly detected some kilometres away (Legionnet and Lefèvre, 1996). Significant differentiation may also be found with adaptive traits: for various components of the resistance to rusts, 0 to 20 % of the genetic variance was found between stands (Legionnet, 1996). Thus, most of the variation, neutral or not, is found within stands, but the level of differentiation may vary from one trait to another. Therefore, sampling for undetermined genotypes requires a balanced within/between-population strategy, whereas sampling for a particular trait of interest would benefit from a preliminary study of the differentiation for that trait.

Vegetative propagation is regularly observed at a juvenile stage, but Legionnet et al. (1997) found almost no genetic redundancy in adult trees. At least two hypotheses could explain this result: (1) vegetative propagation is often found in disturbed areas where the plants are regularly coppiced and remain in a juvenile stage; (2) when conditions allow the trees to grow up, the high competition could result in a single survival shoot per bunch.

3.3. International coordination

Inventories and collection of genetic material pose a particular problem because of the difficult detection of introgression in the field. The main objective is always to preserve diversity, not restrict it arbitrarily, and therefore only indications can be given with regard to 'pure' species. After having exchanged their long-term experiences from individual countries, the EUFORGEN *P. nigra* Network members therefore decided to produce an identification sheet for *P. nigra*. It was prepared on the basis of the most characteristic morphological traits in the wild and at the nursery stage. The practical identification sheet was printed in six languages and is used widely in European countries for inventories.

The network is currently working on a practical list of stand descriptors specifically dedicated to *P. nigra*. This will help a survey of the situation at the species range level.

4. STATIC CONSERVATION

Static conservation is probably the easiest strategy in the short term. This strategy is oriented towards the conservation of genotypes rather than conservation of genes. The objectives are the following:

- maintain through vegetative propagation a large proportion of the remaining resource when locally endangered (e.g. United Kingdom);
- maintain locally adapted genotypes when in situ conservation is not relevant (e.g., isolated trees or small linear stands near agricultural land);
- provide identified material for planting, breeding and experimental studies.

4.1. Sampling strategy

Poplar trees can be collected through seeds or cuttings. Various arguments are in favour of cuttings. First, cuttings are easier to handle than seeds: they can be collected all winter long, and conserve and propagate easily. The sampling strategy can be adapted to avoid genetic duplication among clone accessions, whereas a screening procedure within half-sib progenies is required to obtain the same result from seeds. As for other forest tree species, individual heterozygosity was found to increase with age, and particularly during the first years of the seedlings when demographic competition is at a maximum (Legionnet, 1996): thus sampling diversity in adult trees is expected to be more efficient than in the seeds. Finally, isolated male trees can be sampled; and screening against hybrid types is readily done during the collection.

The sampling strategy should provide the maximum diversity at a low cost. The Italian collection was based on the following criteria: distance from large-scale poplar cultivation, old age of the trees, distance between sampled trees and ratio between sexes (Bisoffi et al., 1987). Therefore, the risks of introgression and genotype duplication are reduced, and the potential local adaptations are extensively sampled all over the country. In France, the sampling strategy takes into account the distribution of the species (many isolated trees or small groups, fewer large populations) and the structure of diversity: collecting sites should be regularly identified all over the country, each one being represented in the collection by one to five genotypes according to the logarithm of the number of flowering trees on site (*table I*).

Table I. Number of accessions per collecting site in the French clone collection.

Number of adult trees in the population	Number of clones per population maintained in the collection
< 10	1
10–20	2
20–50	3
50–150	4
> 150	5

The objective size of the national collections is generally around 400 to 500 clones (Alba, 1995; Cagelli, 1995; Krstinic and Kajba, 1995). In France, the active gene bank will consist of a collection of 500 clones representing the whole territory as previously described, and submitted to evaluation. The set of 500 genotypes will be regularly updated in a limited proportion, first to allow the inclusion of particular phenotypes of interest, and then to adjust the representation of the various origins when new information on the structure of genetic diversity is available (Lefèvre et al., 1996).

4.2. Conservation methods

The simple record of individual trees, precisely mapped, is often considered as a form of collection, even in the absence of any conservation rule or *ex situ*

propagation. These trees are generally 'plus trees', selected by foresters for their vigour, health and shape: several hundreds of trees may be concerned (Mottl et al., 1997; Varga, 1997; Toth, personal communication). Specific regulations are sometimes defined by local authorities for the preservation of individual trees in situ, generally because of particular aesthetic or historical value: a status of 'Natural Monuments' was declared for black poplars near Vienna (Heinze, 1997), a 'Tree Preservation Order' has been issued in the United Kingdom (Tabbush, 1996a).

Nevertheless, the clone gene banks are generally maintained as ex situ collections in the field. In France, the conservation consists of a nursery stool-bed, adult tree plots, as well as any plantation linked to the poplar breeding programme. Stool-beds consist of three stools per genotype, annually coppiced, and renewed every 7 years. Adult conservation plots result in all in three trees per clone, at approximately 300 trees per hectare, to be renewed every 20 years. These figures give an idea of the cost of such a conservation strategy. The active collection is currently evaluated in the nursery for the following criteria: adaptive traits of high heritability (phenology, disease resistance), morpho-physiological descriptors and simple gene markers (isozymes, DNA).

P. nigra is also a good biological model for long-term storage based on tissue culture techniques or cryoconservation (Maestro, 1995). These methods could reduce the cost of ex situ conservation, especially when the term of in vitro conservation exceeds the rotation in stool-beds or adult tree plots. This is not used yet.

4.3. International coordination

A European database of the clone collections has been set up, including passport data and conservation status. National collections are progressively included (*table II*), and updating is planned several times a year. The database is freely available in computerised form or on paper.

Table II. Content of the EUFORGEN database of *P. nigra* clone collections (March 1997).

National collection	Number of accessions
Belgium	61
Croatia	68
France	383
Italy	534
The Netherlands	228
Spain	131
Turkey	298
UK	106
Total	1 809

Precise guidelines for maintenance and duplication of ex situ collections have been published (de Vries, 1996), including technical aspects for the estimation of conservation cost. Guidelines for seed and pollen conservation based on the most recent results are also available (Cagelli, 1997).

A series of adult trees and nursery plant descriptors has been determined (van Slycken, 1996), and a list of reference clones for each trait was identified: cuttings from these clones, coming from a single nursery, are made available.

A restricted collection of *P. nigra* clones from all origins is currently being prepared and propagated (two clones per country): cuttings will be available and used as a common set of control clones for comparison of national collections.

5. DYNAMIC CONSERVATION

When the objective is to preserve gene diversity, and not existing genotypes, dynamic conservation is preferable. This can be achieved through in situ conservation of native stands, long-term breeding programmes or both: a combined strategy is planned for *P. nigra*, as for other tree species like cork oak (Varela and Eriksson, 1995). Stand management rules for in situ conservation have been defined for different social broadleaves (Teissier du Cros and Bilger, 1995) and conifers (Koski et al., 1997); 'pseudo in situ' strategies have also been described for species showing scattered distribution, based on artificial seed orchards (Collin and Bilger, 1995). However, none of these in situ methods can be directly applied to black poplar, due to its pioneer habit, and original approaches to the management of disturbed riparian zones are needed: both the management of the forest itself and the management of the river have an effect on the population dynamics of poplars (Peterken and Hughes, 1995). In addition, principles of long-term breeding aimed at gene conservation are successfully applied to forest trees (Namkoong, 1976, 1997; White et al., 1993), and can be readily extended to the case of black poplar.

When *P. nigra* is not a protected species itself, its preservation relies on the protection of its biotope (Gencsi and Bordacs, 1997). The best strategy depends on the funds available for the species. In situ conservation is obviously cheaper than long-term breeding programmes when based on preexisting reserve areas, when a specific objective for black poplar preservation can be assigned (however, active management is required, which means specific costs). In the areas where the species has been threatened seriously, or where riverbank reserve areas are unavailable, specific re-introduction of the species is planned.

5.1. In situ conservation units

In France, in situ conservation of *P. nigra* relies indirectly on reserve areas: mainly the natural reserves are perennial and 'managed'. More than 20 reserve areas are grouped in a Network of Riverside Reserves (Michelot, 1995). Similar reserves exist all over Europe. Some countries, like Croatia, Hungary, Russia and Slovakia, have specifically dedicated some wild populations to the objective of *P. nigra* conservation: they consist of sustained seed stands, also used for the breeding programmes (Krstinic and Kajba, 1995; Popivshchy et al., 1997; Varga, 1997). The objective is to maintain a mixed forest as similar to the natural composition as possible for stability (Krstinic and Kajba, 1995).

The re-introduction of *P. nigra* on riverbanks by plantation has been achieved in the Czech Republic, Hungary and the Netherlands (de Vries, 1995; Mottl et al., 1997; Toth, personal communication). Belgium and the United

Kingdom plan to reestablish a complex flood-plain ecosystem where it has disappeared (Peterken and Hughes, 1995; Tabbush, 1996b). In Belgium, it was decided to expropriate interdiike land along the River Scheldt for this purpose (van Slycken, personal communication). Private or public areas are involved in these projects, and are protected by law. The production of planting material for these projects is regulated: the objective of conservation excluding commercial production has to be officially recognised in order to be able to grow unregistered material in the nurseries.

5.2. Specific management and regulations

Integral reserves generally do not have any technical or silvicultural management, just a regulation on exploitation. In this case, the preservation of *P. nigra* is totally controlled by the natural disturbance of the ecosystem. In some Italian reserves, the only regulation concerns hybrid plantations within the park (Cagelli, 1995). Within Austrian reserves along the Danube, the situation has degraded so much that eradication of hybrid poplars is only planned gradually to avoid dangerously large clear-cuts (Heinze, 1997). So far, no specific laws exist in any country, regulating the type of material to be (or not to be) planted in the vicinity of protected areas; thus, protection against 'genetic pollution' from exotic hybrids or *P. nigra* clone cultivars is directly related to the size of the reserve area.

Specific in situ management of selected *P. nigra* stands is expensive. Special care is needed to obtain regeneration: removal of weeds three times a year and pesticides result in 20 % of the germination rate normally obtained in the nursery, and cleaning is required for several years due to the poor growth in natural conditions (Varga, 1997). Thinnings start at 10 years of age (Krstinic and Kajba, 1995). An interesting example of the positive effect of hydraulic engineering for black poplar is found along the Volga: distribution of poplar was previously limited by intense flooding. Artificial regulation has drastically reduced the inundation period by a factor of one to three, which is more suitable for black poplar (Popivshchy et al., 1997).

The re-introduction of *P. nigra* on the banks of river systems is generally based on material of local origin, either clonal material (Toth, personal communication) or 2-year-old seedlings (Mottl et al., 1997). A more advanced procedure is planned for artificial re-establishment of flood-plain forest, although it is not yet finalised. One strategy would be to plant a range of species, including at least 30 to 50 male and 30 to 50 female black poplar trees from the gene bank in each location, and let nature do its work (de Vries, 1995; Tabbush, personal communication). Large spacing, 10 m × 10 m or more, would favour flowering, and minimum care during the 2 years after plantation would help initial establishment (Lefèvre et al., 1996).

Either for native or artificial in situ conservation plots, natural regeneration is certainly needed, but is probably not enough to ensure diversity. Legionnet (1996) studied the occurrence of seedlings. Regeneration in a single location consists of a mosaic of seedling patches from various origins. Different factors contribute to the diversity found in the recruitment: the variation for flower phenology among neighbouring trees, the huge variation in space and time of site conditions suitable for seed germination in such an ecosystem, the

dissemination potential of the species and the poor viability of its seeds. In situ management should be compatible with diverse recruitment.

5.3. Breeding populations

Advanced strategies have been applied to poplar breeding (Bisoffi and Gullberg, 1996). In Italy, *P. nigra* is involved in a semi-reciprocal recurrent selection scheme, in order to produce interspecific varieties with *P. deltoides*. The objectives of drastic clone selection, and continuous smooth population improvement, are clearly disconnected, allowing for the long-term preservation of the potential for improvement, which is precisely gene conservation. This kind of breeding programme is optimal for gene conservation, but is reserved for species of great economic interest. In this programme, 139 *P. nigra* males were crossed with four to six *P. deltoides* females, and 41 *P. nigra* females were involved in intraspecific polycrosses. From the study of correlations between intra- and interspecific breeding values, 39 males and 15 females were selected, plus 71 clones within female progenies, to provide progenitors for the next generation (Cagelli, 1995).

A long-term poplar breeding strategy was recently defined in France, involving three *Populus* species including *P. nigra*. For this species, population management aims at preserving a large genetic base through generations (no family selection, but only clone within-family), with control of inbreeding. This programme should start on a 50 to 60 genitors basis. For poplar breeding populations (but not for final selection), improvement relies on nursery evaluation. In this case, the generation length would be determined by the age at flowering of the trees, which is around 8 to 10 years in controlled conditions, probably earlier than in nature (Zsuffa, 1974).

5.4. International coordination

The collaborative efforts of the network have so far concentrated on the identification of common research needs. Based on the discussions, a joint research project proposal, coordinated by the Netherlands, was submitted for funding. The aim is to carry out a broad study of genetic diversity within and among European river systems, and of the history traits of the species on which dynamic management should be based.

6. PUBLIC AWARENESS

Public awareness was considered an important area where members of the EUFORGEN *P. nigra* Network could have an impact and contribute to its improvement. Poplar has an important social and cultural meaning, mainly in its domesticated form, either planted or ornamental (Le Floch, 1996). Poplar is often associated with clonal forestry and exotic hybrids, but the question of the native *P. nigra* itself is neglected. Concerning flood-plain conservation efforts, emphasis is first put on water, then on forest ecosystems, and, within them, on animal species (Heinze, 1997). These features are well illustrated during inventories: recording of the remaining trees in the United Kingdom was based on information received from "enthusiastic citizens" after several

campaigns in the national newspapers (Tabbush, 1996a). However, *P. nigra* is rarely identified in the regular inventories.

The objective then is to inform the public about the needs, objectives and strategies of black poplar conservation. Various communication media have already been used in the different countries: newspapers, reports, leaflets, conferences, videos, excursions and meetings. Collaborative efforts are made within the network, starting with a collection of European illustrations. All the outputs from the network (identification sheet, list of references, publications, guidelines and database) can be obtained through the Internet. One future question will be to conciliate the 'native status' of the species and 'active management' when it is needed (Tabbush, personal communication).

7. OUTLOOK

The *P. nigra* Network brings together a multitude of partners from different countries. Their coordinated work, with shared responsibilities, aims to benefit national gene conservation activities in all participating countries. The network members accomplish common tasks which result in an 'added value' of networking over individual efforts. Development of conservation strategies and methodologies, as well as application of common standards, are essential tasks. While research collaboration is a characteristic part of the activities, the network has thus far been primarily concerned with the practical application of research results to forestry practice.

A comprehensive approach integrating the in situ protection and management with ex situ conservation, and including effective methods for the re-establishment of riparian ecosystems, is needed in the long term. This concerns not only *P. nigra*, but other species like *Ulmus laevis*. From a European perspective, activities will focus on the improvement of methodologies for in situ conservation in the coming years. Various concepts ranging from the 'natural monument preservation' to the active management of in situ gene conservation areas, should be taken into consideration. A unique representative network of in situ gene conservation areas could be established in the future, covering the most important genetic resources not only in the participating, European countries, but throughout the whole distribution area of the species.

Forest gene conservation is a long-term objective, and criteria are needed to follow up the sustainability of forest management (Namkoong et al., 1996). More information is needed with regard to the genetic architecture and processes in *P. nigra* in its entire range. Gene flow efficiency should be further investigated, in order to optimise the sampling efforts within and between populations. Beyond this immediate question of sampling, a dynamic approach including population genetics, demography and ecology is needed to predict the evolution of the species under anthropic influence. Another major study area is the effects of introgression. Beyond merely theoretical discussions, the extent to which the network members, and all involved scientists and responsible authorities in general, will be concerned about interaction between wild and cultivated poplar gene pools may have important economic consequences. A common strategy should thus be pursued to address this particular aspect of *P. nigra* conservation biology. Metapopulation dynamics in relation to the ecosystem and its regulatory processes will be an important area linking the

pure scientific interest with practical considerations about the foreseen network of gene conservation areas in the whole distribution range.

REFERENCES

Alba N., Conservation of forest genetic resources of *Populus* in Spain, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 25–26.

Bisoffi S., Gemignani G., Gras M.A., May S., Mughini G., Establishment of *Populus nigra* genetic reserves in Italy, *Genet. Agr.* 41 (1987) 105–114.

Bisoffi S., Gullberg U., Poplar breeding and selection strategies, in: Sttetter R.F., Bradshaw H.D., Heilman P.E., Hinckley T.M. (Eds.), *Biology of Populus and Its Implications for Management and Conservation*, NRC Res. Press, Ottawa, 1996, pp. 139–158.

Cagelli L., *Populus nigra* genetic resources in Italy, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 19–24.

Cagelli L., Guidelines for seed and pollen storage, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 12–13.

Cagelli L., Lefèvre F., The conservation of *Populus nigra* and gene flow with cultivated poplars in Europe, *For. Genet.* 2 (1995) 135–144.

Collin E., European gene conservation strategy for Noble Hardwoods in the long term – Elm (*Ulmus* spp.), in: Turok J., Collin E., Demesme B., Eriksson G., Kleinschmidt J., Musanen M., Stephan R. (Eds.), *Noble Hardwoods Network Report of the second meeting*, Lourizan, Spain, 22–25 March 1997, IPGRI, Rome, 1998, pp. 44–47.

Collin E., Bilger I., Programme de conservation des ressources génétiques du merisier. Document de la Commission technique nationale de conservation des ressources génétiques forestières, Cemagref, Nogent-sur-Vernisson, 1995.

de Vries S.M.G., *Populus nigra* in the Netherlands, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, p. 46.

de Vries S.M.G., Guidelines for maintenance and duplication of ex situ field collections of *Populus nigra*, in: Turok J., Lefèvre F., Cagelli L., de Vries S. (Eds.), *Populus nigra* Network. Report of the second meeting, Casale Monferrato, Italy, 10–12 September 1995, IPGRI, Rome, 1996, pp. 11–12.

D'Ovidio R., Mugnozza G.S., Tanzarella A., rDNA cloning and rapid hybrid identification in *Populus* spp. (*Salicaceae*), *Plant Syst. Evol.* 177 (1991) 165–174.

Eckenwalder J.E., Systematics and evolution of *Populus*, in: Sttetter R.F., Bradshaw H.D., Heilman P.E., Hinckley T.M. (Eds.), *Biology of Populus and Its Implications for Management and Conservation*, NRC Res. Press, Ottawa, 1996, pp. 7–32.

Faivre-Rampant P., Bodergat R., Bervillé A., Une méthode moléculaire de classement des clones de peupliers (*Populus*) dans les sections *Tacamahaca*, *Aigeiros*, *Leuce* et *Leucoides* par des fragments de restriction des unités ribosomiques, *C.R. Acad. Sci.* 3 (t315) (1992a) 133–138.

Faivre-Rampant P., Jeandroz S., Lefèvre F., Lemoine M., Villar M., Bervillé A., Ribosomal DNA studies in poplars: *Populus deltoides*, *P. nigra*, *P. trichocarpa*, *P. maximowiczii* and *Populus alba*, *Genome* 35 (1992b) 733–740.

Gencsi Z., Bordacs S., Conditions of in situ conservation of *Populus nigra* in Hungary on the bases of the new Nature Protection Law, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 18–20.

Heinze B., *Populus nigra* in Austria – rare, endangered, not recognised? in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 34–40.

Herpka I., A survey of development and possibilities of growing: natural forests of poplars and willows, in: Poplars and Willows in Yugoslavia, Poplar Research Institute, Novi Sad, 1986, pp. 21–36.

Koski V., Skroppa T., Paule L., Wolf H., Turok J., Technical guidelines for genetic conservation of Norway spruce (*Picea abies* (L.) Karst.), IPGRI, Rome, Italy, 1997.

Krstinic A., Kajba D., Conservation of poplar and arborescent willow genetic resources in Croatia, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 29–36.

Le Floch S., Regards sur le peuplier, un arbre entre champs et forêts – du rationnel au sensible, thèse, Ecole nationale du génie rural, des eaux et des forêts, 1996.

Lefèvre F., Legionnet A., Valadon A., Villar M., Programme national de conservation de *Populus nigra*. Document de la Commission technique nationale de conservation des ressources génétiques forestières, Inra, Avignon, 1996.

Legionnet A., Diversité et fonctionnement génétique des populations naturelles de *Populus nigra* L., espèce pionnière des ripisylves européennes, thesis, Université Montpellier, France, 1996.

Legionnet A., Lefèvre F., Genetic variation of the riparian pioneer tree species *Populus nigra* L. I. Study of population structure based on isozymes, *Heredity* 77 (1996) 629–637.

Legionnet A., Faivre-Rampant P., Villar M., Lefèvre F., Sexual and asexual reproduction in natural stands of *Populus nigra*, *Bot. Acta* 110 (1997) 257–263.

Maestro C., Poplar germplasm conservation: ex situ conservation methods under controlled conditions, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 11–14.

Michelot J.L., Gestion patrimoniale des milieux naturels fluviaux. Guide technique, Réserves naturelles de France, atelier technique des espaces naturels, Montpellier, 1995.

Mottl J., Benetka V., Dubsky M., Vackova K., Cikova L., Conservation of genetic resources of *Populus nigra* in the Czech Republic, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 30–33.

Muller C., Teissier du Cros E., Conservation pendant cinq ans de graines de peupliers noirs (*Populus nigra* L.), *Ann. Sci. For.* 39 (1982) 179–185.

Namkoong G., A multiple index selection strategy, *Silvae Genet.* 25 (1976) 199–201.

Namkoong G., A gene conservation plan for loblolly pine, *Can. J. For. Res.* 27 (1997) 433–437.

Namkoong G., Boyle T., Gregorius H.R., Joly H., Savolainen O., Ratnam W., Young A., Testing criteria and indicators for assessing the sustainability of forest management: genetic criteria and indicators, CIFOR Working Paper no. 10, Bogor, Indonesia, 1996.

Peterken G.F., Hughes F.M.R., Restoration of floodplain forests in Britain, *Forestry* 68 (1995) 187–202.

Pinon J., Frey P., Structure of *Melampsora larici-populina* populations on wild and cultivated poplar, *Eur. J. Plant Pathol.* 103 (1997) 159–173.

Pont B., Suivi à long terme de la dynamique forestière spontanée des ripisylves – 1e phase : mise au point de la méthode et test sur six réserves naturelles – Rapport final, Internal document, Réserves naturelles de France, 1995.

Popivshchy I.I., Prokazin A.E., Routkovsky I.V., Black poplar in the Russian Federation, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 46–52.

Postolache G., The status of black poplar (*Populus nigra*) in Moldova, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 44–45.

Schulzke R., *Populus nigra* in Germany: a case study, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 37–39.

Smith R.L., Sytsma K.J., Evolution of *Populus nigra* (sect. *Aigeiros*): introgressive hybridization and the chloroplast contribution of *Populus alba* (sect. *Populus*), *Am. J. Bot.* 77 (1990) 1176–1187.

Tabbush P., The status of black poplar conservation in Britain, in: Turok J., Lefèvre F., Cagelli L., de Vries S. (Eds.), *Populus nigra* Network. Report of the second meeting, Casale Monferrato, Italy, 10–12 September 1995, IPGRI, Rome, 1996a, pp. 7–10.

Tabbush P.M., Native poplars and the restoration of floodplain forests, *Q. J. For.* 90 (1996b) 128–134.

Teissier du Cros E., Bilger I., Conservation of beech genetic resources in France, in: Genetics and Sylviculture of Beech, Proc. IUFRO Project Group P1.10-00, Mogenstrup, Denmark, 1995, pp. 196–209.

Toth B., Black poplar (*Populus nigra*): the situation in Hungary, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 27–28.

Tunçtaner K., Conservation of genetic resources of *Populus nigra* in Turkey, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, pp. 41–44.

Tzanov T., Bulgarian national programme project for the conservation of *Populus nigra*, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, p. 45.

van Slycken J., Short note about *Populus nigra* in Belgium, in: Frison E., Lefèvre F., de Vries S., Turok J. (Eds.), *Populus nigra* Network. Report of the first meeting, Izmit, Turkey, 3–5 October 1994, IPGRI, Rome, 1995, p. 40.

van Slycken J., Plant descriptors for *Populus nigra*, in: Turok J., Lefèvre F., Cagelli L., de Vries S. (Eds.), *Populus nigra* Network. Report of the second meeting, Casale Monferrato, Italy, 10–12 September 1995, IPGRI, Rome, 1996, pp. 13–24.

Varela M.C., Eriksson G., Multipurpose gene conservation in *Quercus suber* – a Portuguese example, *Silvae Genet.* 44 (1995) 28–37.

Varga L., Preservation and reproduction of black poplars in Slovakia, in: Turok J., Lefèvre F., de Vries S., Toth B. (Eds.), *Populus nigra* Network. Report of the third meeting, Sarvar, Hungary, 5–7 October 1996, IPGRI, Rome, 1997, pp. 41–43.

Vendramin G., Michelozzi M., Tognetti R., Vicario F., *Abies nebrodensis* (Lojac.) Mattei, a relevant example of a relic and highly endangered species, *Bocconea* 7 (1997) 383–388.

Venturella G., Mazzola P., Raimondo F.M., Strategies for the conservation and restoration of the relict population of *Abies nebrodensis* (Lojac.) Mattei, *Bocconea* 7 (1997) 417–426.

White J., Black poplar: the most endangered native timber tree in Britain, Research Information Note – Forestry Authority Research Division (United Kingdom), no. 239, 1993.

White T.L., Hodge G.R., Powell G.L., An advanced-generation tree improvement plan for slash pine in the Southeastern United States, *Silvae Genet.* 42 (1993) 359–371.

Zsuffa L., The genetics of *Populus nigra* L., *Annales Forestales* (Zagreb) 6/2 (1974) 29–53.