Conserving intraspecific biodiversity of forest trees in France and Europe

Using examples drawn from his work on elms and his experience as secretary of the Forest genetic resources commission, Éric Collin illustrates the issues and methods for the conservation of genetic diversity of forest trees in light of climate change.

For the general public, the slogan "Stop biodiversity erosion" means taking measures to protect plants and animals whose survival as a species is threatened. The Eurasian lynx, the common snipe, Hermann's tortoise and the Aveyron ophrys are thus all listed for protection in the 2010 Biodiversity agenda for continental France. Forest trees will probably be excluded, except perhaps elms, the well known victims of a terrible epidemic, and the Spanish black pine, a rare species of which only a few communities still exist in Languedoc and the eastern Pyrenees.

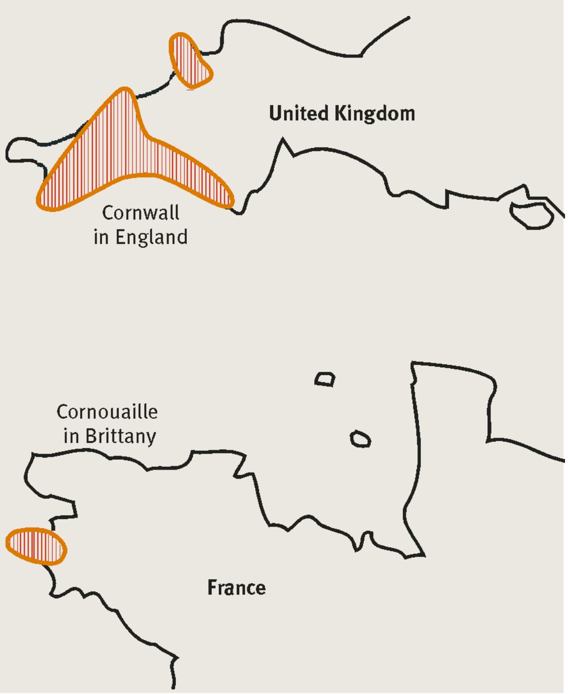
For specialists in forest genetic resources, it is however indispensable and urgent to address the intraspecific biodiversity of forest trees, i.e. the diversity within species. This diversity is not very visible and generally has no name, but it is crucial for species adaptation. It is necessary to stop its erosion if we want to avoid endangering species. This is true even for very common species such as silver fir and beech.

Are species names misleading?

Species and morphological variations

The concept of species is vague and no definition has been found (morphological, biological, ecological, phylogenetic) that is perfectly satisfactory. The effort to create a linguistic discontinuity (a name for a species) in an evolutionary continuum (progressive changes in populations over time and space) may be considered a project that will necessarily remain imperfect. The concept of evolutionary significant units (ESU), used by conservation biologists, takes this double variation better into account, but the criteria or combinations of criteria used to define an ESU (inter-population differentiation measured using molecular markers and/or adaptive characters) are simply guidelines for our work, not absolute truths.

❶ Distribution of the Cornish elm (*Ulmus stricta* Lindley) according to Richens (1983).



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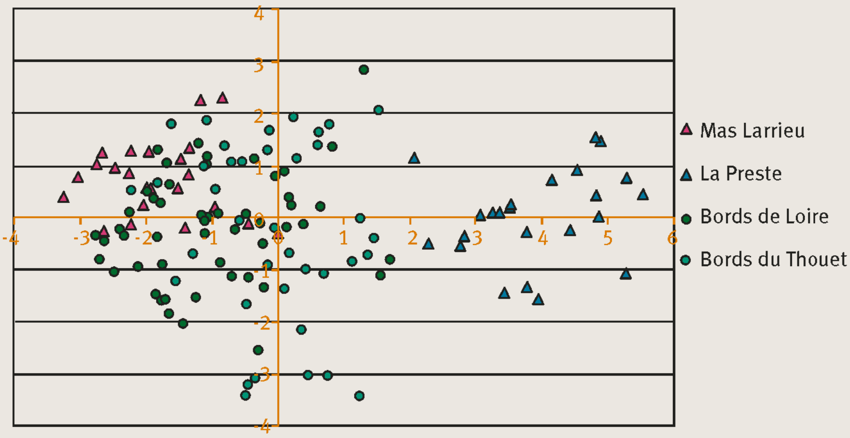
❶ Elm (*Ulmus minor* Mill. *sensu latissimo*), a species devastated by the Dutch elm disease.

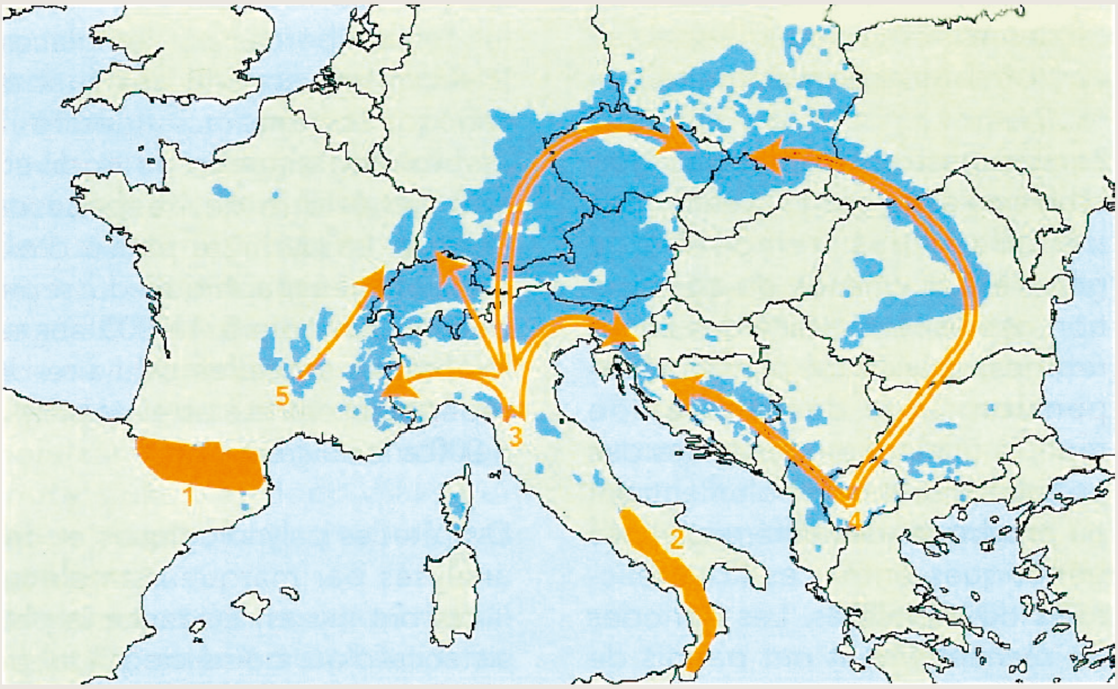
The case of elms in Europe is a good example of the difficulty in distinguishing between species. According to the eminent specialists, all British, who hotly disputed the topic during the second half of the last century, there are three, four or six species of elm in Europe, or many more with up to 61 micro-species. The problem lies mainly in the taxonomic status that should be assigned to the Field elms endemic to different parts of the British Isles, the Channel islands or even Brittany. According to R. Melville (Kew Arboretum), they are endemic species, but according to R. Richens (University of Cambridge), they are simply varieties of the large Field elm species *sensu latissimo* (see photo ❶). These disputes would appear trivial if they did not have consequences for the symbolic presentation of the patrimonial value. For example, owing to its rarity and value as an endemic species, Plot's elm (*Ulmus plotii* Druce) was granted a high degree of protection until genetic studies using molecular markers showed that the taxon was not a true species, but probably a clone spread by the inhabitants of the English Midlands. Preliminary research conducted in 1996 in Finistère (Hollingsworth, pers. com.) led to the belief that the Cornish elm (*U. tricta Lindley*), distinguishable owing to its slender form and late bud break, is also a variety with a very narrow genetic base. According to R. Richens, its current distribution (see figure ❶) is probably due to population movements between Cornouaille in Brittany and Cornwall in England prior to the 800s.

Natural hybridisation and species complexes

Officially assigning a species name is not only a regulatory requirement for the sale of forest-tree seeds and planting stock, but also indispensable when drawing up the floristic inventory of a natural site that must be protected or restored. Legal proceedings concerning a batch of ash seeds from the Saône valley and a recent study in the Loire valley (Henry, 2009, see figure ❷) show that for native ash trees in France, assigning a species name is not always easy. The morphological distinction between the common ash (**Fraxinus excelsior**) and narrow-leafed ash (*F. angustifolia*) is particularly difficult owing to hybridisation in the mentioned valleys. Generally speaking, the opinion that the narrow-leafed ash is a more Mediterranean species, totally distinct from the common ash, is a great oversimplification. For forest geneticists, gene exchanges between these two types of ash have resulted in a currently evolving species complex rather than the two clearly defined species that botanic or commercial names would have us believe. The same is true for the Field elm (*Ulmus minor*

❷ Discriminant analysis of the foliar morphology of ash trees in the Saumur region (green) compared to a population of common ash (blue) and narrow-leafed ash (red), according to Henry (2009).





❸ **Glacial refuges of the silver fir (*Abies alba L.*) and the main colonisation routes (adapted by Plas *et al.*, 2008, based on Konnert et Bergmann, 1995).**

Mill. *sensu latissimo*) and the Wych elm (*U. glabra* Huds.) which together form a species complex that makes specific determination of some elms very difficult. A study using molecular markers on 535 clones of native European elms (Goodall-Copestake *et al.*, 2005) clearly shows a continuum of intermediate forms between the Field elm and the Wych elm. The European white elm (*U. laevis* Pall.), however, does not crossbreed with the above two species and is phylogenetically speaking quite different. The list of forest species that in fact constitute species complexes is long and includes the Aleppo and Turkish pines, the common, sessile and downy oaks, the silver and Greek firs, etc.

Diversity without a name because it is not very visible

The above shows that our linguistic categories do not always coincide perfectly with the relevant biological groups that should be included in biodiversity-conservation and monitoring policies. For Plot's elm, the attention paid to a few morphological specificities led to a proposal for a major conservation effort in favour of a variety with little genetic diversity and belonging to the vast species complex of the Field elm. Conversely, the absence of a name or of an easily identifiable morphological characteristic may result in overseeing entire aspects of variability in living beings and their populations. In the case of forest trees, over a century of experimentation in plantations comparing different provenances has made it possible to understand some of this unnamed variability. More recently, DNA studies using molecular markers have been the means not only to better assess the degree of diversity within populations and genetic differentiation between populations, but to obtain information on the post-glacial history of these populations (refuges during the last glacial maximum and recolonisation routes during the Holocene). These various elements are most useful in determining relevant ESUs for conservation purposes.

Regarding the silver fir, for example, we now know that the Aude and Alpine populations come from different glacial refuges (see figure ❸). For the European white elm, a recent study using several different types of molecular markers (Fuentes Utrilla, 2008) revealed the singular characteristics of the small populations in Spain and southwestern France compared to those in other parts of the distribution range of the species. This study contradicts the generally accepted opinion that the rare populations of European white elm in Spain are not autochthonous and thus do not merit conservation efforts. It also reinforces our hypothesis that the species may be native to southern France (Timbal et Collin, 1999).

In conclusion, it is worthwhile to keep in mind that species names may be misleading and that biological rareness does not depend exclusively on morphological particularities, but it is above all a question of genetic originality and diversity.

Dangers of genetic erosion

Loss of species and of diversity

For the general public, erosion of biodiversity is more or less synonymous with the risk of species extinction. In the case of European forest trees, we have thankfully not reached that point, though there are a few exceptions such as the Sicilian fir and the Serbian spruce. Even Field elms, that were spectacularly decimated by Dutch elm disease in the 1970s, are not threatened with extinction because they continue as shrubs in the form of resprouts regularly cut back by the disease. In certain countries or regions of

Europe, the species is nonetheless listed as "near threatened", "vulnerable" or even "endangered" as per the IUCN (International union for the conservation of nature) criteria. But for our common forest trees, the issue of biodiversity erosion should be addressed in terms of quality (loss of diversity and capacity to adapt) rather than in the overly simple terms of quantity (rarity and/or regression).

Obviously, the size and dynamics of populations play a crucial role in the stability and evolution of the genetic diversity of species and their populations, but it may not be concluded that a common species is automatically immune to diversity loss. In the case of the silver fir, the species is widely spread throughout Europe, including in France, but a number of factors are nonetheless likely to result in a reduction of the genetic diversity of the species as a whole, as well as within individual populations. On the local level, certain sylvicultural techniques may increase the risks of self-fertilisation or, on the contrary, facilitate the penetration of non-native pollen and seeds, at the expense of autochthonous flora (Plas *et al.*, 2008). Overall, there is the risk that climate change will result in a rapid reduction in the distribution range of species and in the loss of ecotypes in marginal geographic situations. The drought in 2003, which caused high losses of fir trees in south-eastern France, was a serious warning.

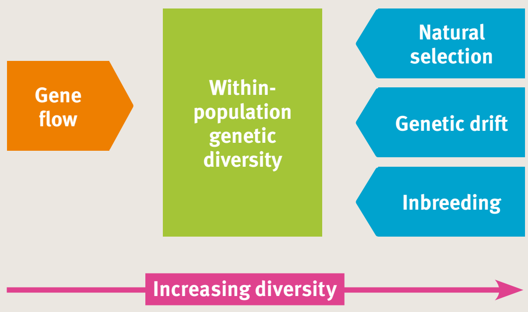
Gene transfers, ma no troppo!

The issues of demographics and the introduction of non-native flora rapidly discussed above require a comment on the essential role of gene transfers between populations and even between species. Transfers occur naturally owing to wind or animals that disseminate pollen and seeds, sometimes over long distances, but are also caused artificially by humans who introduce seeds or planting stock from other regions or even other continents. As shown in figure ❹, transfers are essential to maintain and increase genetic diversity within a population and counteract the effects of genetic drift, inbreeding and natural selection, which tend to reduce diversity.

Transfers are particularly necessary for small populations whose habitat has been reduced or fragmented. That is notably the case for forests along rivers, the habitat of the European white elm, of which there remain only fragments following clearances for agriculture, poplar stands or urbanisation. For these small populations separated from each other, major, random losses of genetic diversity may occur through genetic drift. Even between species, gene transfers have contributed to forming the forest-tree species as we know them today. The examples of ash and elm trees discussed above show that interspecific transfers continue unabated and could play a role in the adaptation of forest-tree populations to climate change.

Should we consider that gene transfers are a normal process when they occur spontaneously among autochthonous populations or species, and that they are an intolerable source of genetic pollution when human intervention is involved? A pragmatic means to avoid this rather caricatured ideological debate would be to examine the possible consequences of such transfers from non-native provenances or species. The consequences could be positive, negative or negligible depending on the respective size, diversity and adaptation of the receiving population and the non-native genes. The worst case scenario is a small population receiving a large quantity of ill-adapted plant material offering little genetic diversity. The best case scenario is a small population receiving a large quantity of well adapted and genetically diverse plant material.

❹ Factors influencing diversity within populations (based on Eriksson, 1998).



Maintain processes rather than situations

Adaptation and adaptability (see box ❶)

The conservation goals set on both the French (CRGF) and European (European forest genetic resources programme, EUFORGEN) levels are to maintain the adaptive capacity of forests in a changing environment (Balsemin et Collin, 2004). This approach, going beyond efforts to simply maintain the current state of adaptation, is based on the concept of "dynamic conservation" of genetic resources. Contrary to static conservation, e.g. collections of elm clones preserved in nurseries or as buds in liquid nitrogen, dynamic conservation attempts to facilitate sexual reproduction as a source of new genetic combinations and natural selection in populations that regenerate themselves *in situ* (see figure ❺). If necessary, dynamic conservation can also be carried out in populations reconstituted *ex situ*, e.g. conservation plantations or conservation seed-orchards. Habitat protection, dynamic conservation and static conservation are not approaches exclusive of each other, on the contrary, they are complementary methods that can be combined depending on the specific biological, technical and land constraints weighing on the resource to be conserved.

For the European white elm in the Loire and Garonne river basins, our work was conducted in close partnerships with the naturalists managing the sites and dealt essentially with habitat protection and dynamic conservation *in situ*. In Finland, the rare populations of European white elm are ▶

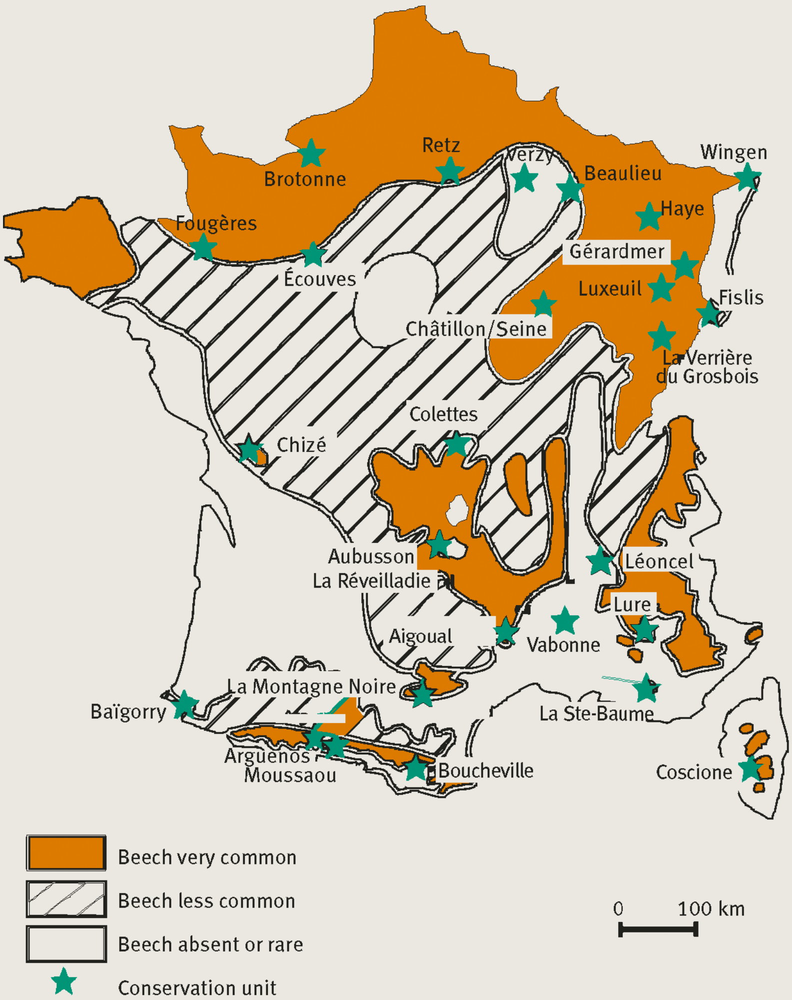
❶ Adaptation and adaptability

The term **adaptation** defines the quality of survival, growth and reproduction of a population under constant environmental conditions.
The term **adaptability** defines the capacity of a population to evolve in a changing environment. That capacity depends heavily on the genetic diversity which supplies the "fuel" for evolution, where natural selection is the "motor".

too small and too depleted, owing to genetic drift, to be very suitably conserved in situ. Finnish colleagues proceeded as in France, but also created an *ex situ* collection of clones reflecting the genetic diversity of the Finnish populations. They plan to create a conservation seed-orchard that would make it possible to reintroduce the species to its original environment or a suitable one. The composition of the seed batches would be diverse enough and not excessively different than that of the population gene pool prior to its fragmentation.

Static conservation *ex situ* was judged the most appropriate method to maintain the diversity of the Field elm in France. This decision was due to technical reasons (easy cloning) and to the hope that the pathological tests (initially run by INRA, the National institute for agronomic research) carried out on the collected material would enable selection of genotypes resistant to Dutch elm disease (DED). This decision appears to have been the right one for other reasons as well, based on the extreme taxonomic complexity of *Ulmus minor* Mill. *sensu latissimo* and the role played by humans over millennia in propagating clones of the species far beyond the original habitat.

❺ **Network for *in situ* conservation of the genetic resources of the common beech (*Fagus sylvatica*).**



Sampling efforts carried out over large parts of France (over 250 clones with the Field elm phenotype or that of a close hybrid) probably succeeded in acquiring a major part of the species' variability in France, including the endemic "species" such as the Cornish elm. None of the clones resists DED well, but the artificial inoculation tests carried out in the partnership with INRA Nancy, CNR (*Consiglio nazionale delle ricerche*) in Florence and the French National conservation nursery of Guéméné-Penfao enabled the selection of plant material less susceptible to the disease and suitable for cautious use in the reconstruction of field hedges.

If the introduced trees are resistant enough to survive until maturity and regularly blossom, they may contribute to dynamic conservation of the local genetic resources. To avoid the excessive parental contribution of certain genotypes, clonal composition of the elm batches planted in the hedgerows must remain diverse and, if possible, regionally structured and gradually renewed over time. A conservation seed-orchard with a large genetic base may also be established.

Working on different scales

The major role played by France in establishing the European programme EUFORGEN following the Ministerial conference on the protection of forests in Europe in (Strasbourg, 1990) explains the consistent methods employed and the close ties between the French programme set up by CRGF and the EUFORGEN strategy, formulated by representatives from the 30 countries participating in EUFORGEN. CRGF and EUFORGEN work in two ways to conserve the intraspecific biodiversity of forest trees by 1) promoting sylvicultural techniques that take genetic diversity better into account, and 2) establishing conservation networks for the species.

In France, the networks are set up in public forests in various pedoclimates and using different models depending on the types of species in question, i.e. species with large populations (sessile oak, beech, silver fir, etc.) and rarer species that are threatened or scarcely spread (elms, black poplar, wild cherry, etc.). For a species in the CRGF programme, if conservation efforts cannot be implemented simultaneously in different parts of France, priorities are determined taking into account the Pan-European range of the species. For the European white elm, for example, given that German and Polish foresters can conserve it in the mid-European section of its range, efforts are focussed on populations in the Loire and Garonne river basins rather than in the east of France.

Through the EUFGIS EU project (EUFGIS, Establishment of a European information system on forest genetic resources), EUFORGEN will soon put on-line a spatialised database, including maps, of the *in situ* conservation units for European forest-tree species in all the participating countries. This project is the result of long discussions on criteria and management rules for these units, which must not indicate simply that the given species is present, but also certify that the local population meets the conditions required for truly dynamic conservation, i.e. sufficient number of trees capable of blossoming and contributing to regeneration, installation and sustainable protection of regeneration.

Projects on the regional level can, of course, fit perfectly in national and European projects. For the European white elm, funding from the *Plan Loire-Grandeur Nature* facilitated better understanding and conservation of two local populations. Partnerships targeting the experimental reintroduction of Field elm clones in hedgerows in western France will be an opportunity to acquire data on natural contamination by the Dutch elm disease and the healing capacity of clones where current testing is limited to artificial inoculations in tree nurseries.

Confronting climate change

Forecasts based on the various scenarios published by IPCC (Intergovernmental panel on climate change) indicate a very strong and fast contraction in the distribution ranges of many forest-tree species found in France. We can hope that the individual phenotypic plasticity of trees (i.e. the capacity of a genotype to generate different phenotypes depending on the environment and thus adapt), the high variability in adaptive characteristics (vigour, time of bud break, resistance to drought) naturally present in all tree populations and the existence of small, favourable spots (shade, soil depth and water) will support pockets of resistance. Genetic modifications can occur in a few generations through natural selection of the most resistant trees. On the other hand, migration of populations will probably be too slow compared to the speed of the expected changes, unless the migration is "assisted" and accelerated by humans. To that end, the CRGF has made the following main recommendations to forest managers.

- Avoid inconsiderate use of a supposedly "providential" species or provenance.

- Adopt sylvicultural techniques that maintain genetic diversity over the long term (ensure the quantity and diversity of natural regeneration or acquire guarantees concerning the genetic diversity of the seed and seedling batches used).

- Encourage natural selection and, if losses occur, regeneration of the surviving trees likely to have made special, very useful genetic adaptations.

- In failing communities, assist regeneration using material from a neighbouring provenance with a hotter and drier climate (assisted migration). If a species or provenance is completely replaced, keep all documents concerning the origin of the new material.

In addition to these general recommendations for forest management, CRGF is planning specific conservation efforts and the experimental transfer of genetic resources.

Brief discussion on the formulation of public policies

Forests as natural and cultural phenomena

At a time when the threat of climate change imposes new constraints, it is worth looking at the forces that determined the approach to conservation of the intraspecific biodiversity of forest trees in France. That approach is particularly impacted by the special position of forest trees at the interface between wild plants, whose conservation falls naturally to the Ecology ministry, and the genetic resources of cultivated plants, whose conservation is the due responsibility of the Agriculture ministry. This difficult position has raised a number of issues, positive and negative.

It may be regarded as positive and productive that forest-tree species were not considered a part of nature that had to be completely protected from the effects of human activities, but rather as a group of genetic pools being constantly modified under the pressure of both environmental and anthropogenic change, and whose conservation should be managed rationally and sustainably. That is not to say that forest reserves completely cut off from all human activities are not useful for intraspecific conservation. However, it is certainly worthwhile to consider conservation of intraspecific forest biodiversity not only in biological terms, but also in light of the complex relationship between a resource and its manager.

This approach is promising in as much as it facilitates dynamic conservation of a given species, whereas an overly protective approach could tend toward elimination of the species owing to natural competitive processes in ecological dynamics. The consent of owners or managers and funding for the additional cost of conservation management are, however, a problem, even in public forests, and CRGF has found it difficult to set up contracts for private forests.

On a more negative note, it is unfortunate that the "genetic resources" approach has produced a utilitarian outlook, in terms of both methods (insufficient sampling in forests that are not or only slightly productive) and the priorities set for species with high economic value, at the expense of non-cultivated species that are potentially under threat (e.g. yew). This is in fact a congenital problem at CRGF because it was assumed at the outset that conservation of species not planted by forest managers would be ensured by protected forest zones (e.g. the biological reserves) and thus the priority for the Agriculture ministry would be conservation of commonly planted tree species. ▶

More informations about...

▶ The Forest genetic resources commission (CRGF)

CRGF was created in 1992 by the French Agriculture ministry. It comprises scientists, public and private forest managers and a representative of the association France Nature Environnement. It defines and implements a strategy to assess and conserve the genetic diversity of forest-tree species in France.
http://agriculture.gouv.fr/sections/thematiques/foret-bois/conservation-ressources

▶ Useful internet sites

http://agriculture.gouv.fr/sections/thematiques/foret-bois/graines-et-plants-forestiers/
http://www.euforgen.org/
(See also the downloadable documents for France on the "Countries" page).

This "division of labour" led CRGF and EUFORGEN to set up specific networks. The advantage of this method is that it more effectively avails the expertise of researchers specialised in the genetic variability of a species or genus. However, it has now met its limits, because the human and financial means are insufficient to create new, specific networks. It would also now be better to improve convergence of the goals for conservation of habitats, species and intraspecific biodiversity. In the field, the people running the Black poplar and Elm networks at CRGF had no difficulties in finding common methods with the managers of river nature reserves. This type of fruitful collaboration could be gradually expanded to other species of riparian forest trees that are currently not or hardly addressed by CRGF (ash, willow, alder, etc.) and to other types of forest environments. Indeed, the contacts made between CRGF and managers for protected zones would indicate that collaborative efforts could renew and enhance the conservation practices of all concerned.

From conservation to ecological engineering

Through the regulations governing the trade of forest seeds and planting stock (called forest reproductive material, FRM), the Agriculture ministry has a means to stimulate forest production through genetic improvements and to avoid unwanted transfers of FRM that could harm conservation of the genetic resources of forest trees. It should also be noted that wild resources constitute a gene reservoir for genetic improvements. That is the case, for example, of the black poplar, deemed "unproductive" in its natural environment, but which paradoxically supplies valuable resistance genes for the cultivated varieties of high-yield poplars.

Clearly, even if the regulations are satisfactory on the whole, there are internal conflicts that can result from the divergent goals, i.e. production (spread genetic progress, rationalise the business of producing seeds and planting stock, comply with European directives on free trade in Europe) and conservation (avoid degrading the autochthonous resources). These contradictions manifest themselves in their application because the regulations, initially designed for forest species and areas intended primarily for production purposes, are poorly suited to other areas (hedgerows, riparian vegetation, etc.) and to new purposes, notably those for ecological engineering (river-bank maintenance, support for biodiversity, ecological restoration, phytoremediation, etc.). What is more, the eligibility criteria for the populations from which FRM harvests are authorised may appear to be overly concerned with the commercial quality of the phenotype (straight trunks, fineness of branching). And of course, the size of the regions of provenance used for product tracing and trade of seed and seedling batches is a source of contention between certain end users, who would like to see a finer mesh, and the nursery owners and seed merchants, who would like a simpler system.

In spite of their imperfections and because they are now an integral part of forestry practices, the regulations will necessarily serve as a basis for measures targeting the preservation and use of forest genetic resources to reinforce the adaptive capacity of forests to climate change. Far reaching decisions must be made to modify the selection criteria for the populations supplying seeds, redefine the regions of provenance and produce new recommendations on how to use plant materials.

These decisions must create a better position to meet the needs of ecological engineering and to determine the degree to which assisted migration from certain provenances should, where applicable, be encouraged. The scope and importance of these tasks would justify the funding of in-depth multi-disciplinary work on the subject. ■

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