

11-20-95

Biodiversity : Scientific stakes and inferences

(The case of plants and plant-breeding)

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Summary

Biodiversity perceptions are many and eventually contradictory. However the rising concern on its short-term evolution urges a significant investment in research and in the integration of new information into the decision making process. From a scientific point of view Biodiversity appears to be a loose concept and one needs to associate it with the impact of human activities on nature. This leads to several questions, two of which will be discussed : Is biotechnology a salvage or a scourge to the general biodiversity concern ? Should we promote more stringent conservations or should we rely on an evolution guided by the caution principle ?

For millennia people have taken benefit from biological resources to eat, resist cold, cure diseases,... The spectacle of biodiversity stimulates human imagination and skills. Networks of symbolic representations as well as practical relationships have developed between societies and nature. This complexity slowly evolved through centuries with gradual changes in the environment. The economic expansion and the demographic outbreak of human kind in the last decades have created a fracture in these processes and massively endangered future biological resources. Concomitantly growing urban populations have lost their traditional roots and links with nature. Bleak prospects and present social disfunctionments lead to a growing concern and our societies get more and more aware of their economic, ecological and cultural need of a rich and diverse living environment (review in 1).

As far back as we recall classifications have been elaborated to describe and organize the living world. A decisive step was achieved when, on following Linné (1707-1778), the concept of species was brought in. More than a million of them are recognized today among existing creatures (Table I). We probably know most of the mammals, we are still far from the exhaustive recollection of insects and even farther for micro-organisms and viruses (2). The number of species on our planet is certainly of the order of several tens of millions and this covers only the present repertoire of living forms. Life is indeed also characterized by its active renewal. When some species seem to have come unchanged from the night of geological times, most of them emerged and disappeared and paleontologists show that the mean half-life of one species is around several million years (3). This means that since the origin of life on earth some 3,500 Myr ago a huge number of species were born and vanished, a number much higher than the presently living sample (Table II). This evolution has paralleled major climate changes and geological upsettings, testifying for a considerable adaptive potential of living forms. In the neo-Darwinian view genetic diversity at the individual level is the reservoir from which selection sieves out the winning forms, in the neutralists vision random sampling processes pick out the various organisms that will bring about the new generations in every place : Nature accommodates environmental changes in the long run by a functional and genetic evolution and by reorganizations involving extinctions and emergencies of species and of biological associations.

Diversity is observed from molecules and genes to cells and organisms, from populations and communities to ecosystems, landscapes and biosphere as a whole (4, 5). The abundance and variety of species as well as their activities and breeding regularity are the traditional criteria of biologists to evaluate how rich a given site can be. In doing so they obtain an integrative picture of the consequences of the biological changes that have occurred locally in the past. The evolutionary processes are extremely slow : Judged at the pace of a human life, nature appears to be at equilibrium even if this is a treacherous impression. We know now that human activities modify the ecosystems at a much higher speed and often impose considerable transformations on biological diversity at a pace the normal renewal process cannot keep (6, 7). The way La Réunion Island evolved illustrates these complex situations and the relationships woven between nature and humans. This island has a rich biological diversity and human settlements are recent, well dated and recorded. The volcanic substrate

emerged from the Indian Ocean some 2.5 Myr ago and from that time up to now a rich biological colonization occurred. When human presence became significant on the eighteenth century a complete survey of the fauna and the flora was carried out, the island being small enough to have been covered by thorough studies : 750 plant species were described, one third of them being endemic. As a whole they represent the results of the arrival of new-comers on the volcanic island and of the many years of local evolution. They are distributed into five general ecosystems : the rain forest, the dry forest, the Acacia forest, the mountain forest, and the upper lands. This complex landscape is under constant reelaboration due to the intimate renewal of its biological components and of natural perturbations (volcanic eruptions and cyclones). A dynamic equilibrium seems to have been reached in some 2 Myr. It operates through 'cyclic recolonizations involving the various scattering potentials of species that settle in cascades one after the other when a site has been "cleaned" by external events.

By the end of the seventeenth and especially during the eighteenth century human populations came in. The original hunting and harvesting activities were rapidly replaced by lumber exploitations, land clearings for ploughing and planting. A large part of the country is turned to agriculture and plantations. The spaces occupied by the original ecosystems are reduced and fragmented. Although its maintenance is not threatened the rain forest withdrew to three hundredths of its original size. A systematic survey of the flora of the island carried on in recent years reveals the consequences of two hundred years of man activities on the original biological diversity (8, 9). A dozen of plant species have most probably disappeared and a group of fifty is reduced respectively to a few isolated individuals or to tiny populations where reproduction may not be maintained. In parallel new "interesting" species have been introduced deliberately, others have snaked in. Three hundred years of man-managed introductions are very impressive : some eleven hundred new plants have entered the island and sixty of them are so successful that they have modified the original ecosystems. Although the five original ecosystems are still recognizable their compositions have been modified and more importantly their renewal potential has been altered. When a cyclone cleans some space the new-comers interfere in the recolonization cascade and the original cycle is no longer reproduced. The former ecosystem disappears and is replaced by a new one the sustainability of which is not guaranteed. This phenomenon is amplified by the disappearance of fruit-eating animals that used to contribute to seed-scattering (twenty for out of the thirty three vertebrate species of the island have disappeared since the eighteenth century).

The situation observed in the region overlapping the French-German border (Pfälzerwald Naturpark and Parc des Vosges du Nord) illustrates a different aspect : the recent and drastic changes brought under the pressure of economic and social changes on ecosystems that men had shaped for centuries. This geographical area was largely a forest with scattered peat bogs in the early Middle Ages and Charlemagne used to go there occasionally for hunting. In the following ten hundred years farmers settled and created a diversified landscape at the same time that they made their living out of crop and cattle breeding : the forest withdrew, peat bogs were often dried and partly exploited, space was used as wet and dry grazing lands, upland pastures, farming parcels. This created a complexity which favored the settlement of many wild species (plants, insects and

birds) that could move about, forage and reproduce. By the turn of this century the type of agriculture carried on in this region was no longer profitable, the farming populations and the biological diversity decreased progressively. When recently National Park authorities were given responsibilities on this region, conservation of the natural diversity became a priority. Experiments were first carried on with the idea of relieving some parcels from any human interference and led to the conclusion that in absence of man intervention a plane forest rapidly goes back and speeds up the rate of decrease of the initial variety of living species. Consequently decision makers changed their mind and privileged with some success a policy that keeps some farming activities and maintains the diversity of the ecosystems (10). The Pfälzerwald and Vosges du Nord example tells us that the conservation of biological diversity and its changes are basically the expression of the wills and the deeds of a society.

From the seventies the combined effect of droughts and of economic and political crises has promoted desertification among the major and global concerns of the world. Changes in sub-Saharan Africa and other tropical countries still produce today a dramatic decrease of the biological diversity and a general rural exodus. The new ecosystem that takes over is very poor and incapable to sustain human activities. Far before the desertification trend was fully analyzed and mastered (even poorly), deforestation came up as an other major plague of tropical areas with again a procession of natural diversity reduction, of threats on biological resources, of erosion acceleration and of social difficulties and conflicts. In temperate and polar countries pollution is the major threat for living organisms both on land (chemicals...) and at sea (oil spills...). Nations got aware of the threats continuous expansion places on biological resources that are used up much faster than they get renewed. The powerful media explained that some kind of global settlement between humans and nature had to be reached. Some UN sessions were converted into forums to advocate for sustainable development and the face-à-face between North and South took a new dimension. The question of biological diversity which had been until the end of the eighties a research problem for biology and a source of innovations and profits for agriculture, forestry and industry became a global political stake (11). From now on the term **Biodiversity** was definitely coined and appeared in the text of an International Convention elaborated in 1992 (Rio).

Biodiversity is actually a loose concept. It is generally used to characterize the living load on a specific location and includes the diversity of ecosystemic organization, the multiplicity of abundance of species, the intraspecific genetic diversity of individuals and their rate of activity and reproduction. In clear biodiversity is complex. It cannot be reduced to a simple figure and a thorough scientific analysis is necessary to provide its valid evaluation. The assessment is complicated by the value and the hierarchical position given to the various criteria used. Difficulties arise at various levels : Scientists debate about the concept of species and the possible genetic exchanges between them, indeed these rare events are carefully looked for by breeders who take advantage of them to transfer economically interesting characters. Scientists debate also between the respective role of ecosystems adaptability and genetic diversity in the biological transformation processes. In the short run the first brings some answers to

changing environmental conditions, but the spectrum of the response, its maintenance and its evolution depend on the second. In other terms should the ecological functions of a community be preferred to its genetic resources or the reverse ? Biology *per se* does not provide unique answers to such questions but the information is necessary to think , decide and accept what society needs. And the debate rebounds at the social level : should the protection of rare and endangered species be more important than the identification and genetic enrichment of potentially useful ones or the reverse ? Are people more interested by the strict conservation and the myth of a lost paradise, by man-managed transformations of the environment in the course of a lifetime or by a new evolution crisis similar to the ones earth has already faced in the geological past, for instance at the dinosaurs extinction ?

Obviously pompous assertions and definitive opinions will not resolve the situation and on the other hand, as time is running fast, immobility, indifference and selfish interests are dangerously short-sighted attitudes. In any case conflicts about the appropriation and the management of biological resources have already arisen and will certainly get more acute. Many situations some of them quite dramatic have been popularized by the media. The La Réunion or Pfälzerwald-Vosges du Nord examples have brought up four pieces of evidence :

- 1)- A thorough research effort is needed in each case and must be adapted to the level and the geographical dimension of the specific problem ;
- 2)- Human activities and human goals are essential determinants ;
- 3)- The ratio between the speed of imposed changes and that of the natural renewal of the biological resources is critical ;
- 4)- Risks inherent to human activities are unavoidable but can be evaluated, discussed, and eventually accepted if society is thoroughly informed.

In essence biological research and knowledge have to be integrated in the decision-making process. Although of general application this objective is analyzed further with details in the case of plants and plant-breeding.

The first objective in temperate and developed countries is to build up frugal plants bringing crops with higher yields and reduction of the costly inputs of agriculture (time, energy, chemicals...). The second goal of plant-breeders is to diversify their products genetically in order to anticipate (or sometimes initiate) changes in the consumers demands. Achievements in the field come up when the following three questions are solved : it is possible to conceive new genes or new genetic combinations in the considered species ? Where to find them ? How to build up a new plant ? By the end of this twentieth century breeders dispose of two sources of innovations : the recent molecular engineering techniques and the traditional reservoir of natural diversity. In view of the biodiversity concern society is liable to ask whether biotechnology is a salvage or a scourge and whether stringent conservation measures should be opposed to free exploitation or cautious management of biological resources.

Plant molecular biology is a rapidly expanding chapter of our biological knowledge. Basic science has already been very efficiently transferred into applications (specific resistance of corn to herbicides for instance). All the major crops of this planet can be and have been genetically engineered in the laboratory, (Table III). Genes (as DNA fragments) are readily isolated and sequenced from

plant genomes. They can be manipulated by mutations in vivo or in vitro according to computerized models. Subsequently they are reintroduced by the recombination into plants and the function of the newly created genetic forms is analyzed in laboratories and in fields. The techniques are universal and it appears that plants are very tolerant to the introduction of new genes. Consequently many new biological objects are created, most of them never occurred during evolution such as plants with bacterial or animal genes. The rate of authorized experiments in the field has grown exponentially the last ten years and indicates the importance of this activity in a profit oriented economy (figure 1).

The advances in plant sciences often allow the complete mastery of the plant reconstruction after the genetic transformation and that of the expression of the introduced gene according to the will of the experimentator. This has obviously considerable merits : it creates molecular diversity which may evolve rapidly, it opens vast possibilities of innovations and indeed succeeds in providing new plants for agriculture. An impenitent optimist may now dream of a complete mastery of man over nature and consider that in some ways the major problems arisen about biodiversity may be solved by technology. However the situation is not as bright as to sustain such wonders. Expanding molecular biology is still an analytical process that goes gene by gene and, even with clever shortcuts, the time has not come when all the some fifty thousands different genes of a plant genome can be manipulated nor their networks of functional and topographical interactions. Moreover by nature the technique takes into account the relationships between the plant, its neighbours and its environment only very modestly.

Developments of molecular engineering for plants require considerable financial investments and involve economic risks. These can only be assumed by large companies and the essence of power over the main vital and strategic crops is consequently taken out the hands of farmers, of local plant-breeders and even of nations. At last the very philosophy of the approach is still considered as a cultural shock by some individuals in our societies.

For hundreds of years since the infancy of agriculture in the Neolithic times farmers, and later plant-breeders, have drawn on the resources of natural diversity. Indeed plants have often solved problems identical or similar to those encountered by producers. Wild barley *Hordeum spontaneum* has developed through evolution more than fifteen different genes conferring resistance to the blight *Puccinia horde*. On the contrary *Hordeum vulgare*, the cultivated species, is sensitive to the pathogen. Breeders trying to create resistant lines have only used a few resistant genes (one to three) and a large panel of ready-made solutions is still available in nature (12). Examples of the kind are hundreds, and moreover all the spontaneous forms elaborated through evolution have the advantage of having successfully resisted the test of natural selection. Only a restricted number of genetic combinations have been used in the domestication process where, especially in the last hundred years, efforts were concentrated on selection for purity and unique properties that would support the need for higher yields, large homogeneous fields and mechanical farming techniques.

In general a vast reservoir of genetic resources is accessible and most of the time we do not even know its size and its diversity !.

This trend has not always been prevalent. In Western Africa local farmers have developed for long techniques that have, on the contrary, maintained a low-level but constant genetic exchange between their cultivated rice *Oryza glaberrima* and the wild species *Oryza breviligulata* which grows as a weed in the same locations (13, 14). This flow of genes was even controlled and, according to their needs and their culture, farmers took advantage of it to draw recurrently new and better lines (as in Casamance) or to maintain heterogeneous but adaptable populations (as in Mali). On doing so traditional agriculture was able to identify original genetic forms and turn them into domestic lines when they would have disappeared in the wild through natural selection... Much is to be found in the genetic diversity of wild plants and much is to be learned from the traditional and local cultures of farmers (15).

Of course with the birth of genetics at the turn of this century plant-breeders were in a position to rationalize and expand very considerably the hybridization and the selection methods. They built up a whole new domain of biological science. Results were stunning and they reinforce the feeling for a careful management of genetic resources of wild plants in the future. New varieties appeared and the gains on many crops supported the outbreak of human demography. The rate of renewal of varieties also increased and innovations matched changing needs, changing techniques, changing marketing procedures.... The search for specificity and homogeneity has been a constant concern all along this evolution. However the idea of looking in nature for new and different genetic resources made its way in the second part of the century. Characters were transferred from wild relatives to domestic plants and the culmination was the elaboration of completely new species which combined properties initially separated in their genitors, triticale being one prominent example (16). Obviously the future of the traditional approach of plant-breeding is still bright. However this process suffers some drawbacks : first it needs time (often many years of ungratifying selection efforts). Second it relies on the two unproven hypotheses that nature has already devised in the wild a genetic answer to the question of the breeder and that the expected character is empirically transferable to cultivated varieties. At last it requires more and more financial investments and consequently this activity has progressively shifted from the authority of individuals to that of companies and recently to very large multinationals in the developed Western world.

Common sense says that the traditional approach using natural genetic diversity and the modern molecular biotechnology should be envisaged as complementary (17, 18). Indeed one of the first and most achieved example of recent progress on tomato supports this view. Improving tomato crops and elaborating varieties that would satisfy the needs of the fruit consumers and of the transport, conditioning and marketing systems, or the imperatives of soup, sauce and concentrates industrial productions has been the goal of an intense efforts all along this century. The genetic diversity of tomato is very large distributed among nine species in many populations, lines or DNA banks (Table IV). Genetic engineering and traditional methods have been combined and new lines, new products are about to be transferred to agriculture and general consumption. The resistance to early rotting and a savoury taste are natural

characters that biotechnology has assembled into a new variety : Flavr Savr (brand name McGregor), which passed satisfactorily all the tests imposed by the US of administration. Flavr Savr is now accepted and has entered a career in agriculture and supermarkets. The way is paved for tens and hundreds of transgenic plants that are waiting for official green light and public acceptance. The vast majority of them is composed of plants where introduced genes come from natural biodiversity (Table V). Society has now to face and hopefully solve three key-questions : Are these new plants and the approach that have generated them culturally acceptable ? Will they enter the developed market and economy that have induced their elaboration ? Will this procedure be accessible for poor countries where most of present genetic and biological resources reside ?

In any case the path has been well traced for the coming years. More knowledge is necessary to contribute to the decision making process and this can be obtained through molecular biology, physiology and ecology research and through a intensive effort of inventory of the presently living diversity.

A careful management of genetic resources is necessary to protect, conserve and follow the evolution of biological diversity. But a controversy has sprung up on this point : a conservationist and well-wishing movement often opposes ruthless exploitation and even measures that take in consideration the inevitable and sometimes necessary intervention of humans.

Biology of conservation is a branch of life sciences that is presently in full expansion and integrates advances in basic knowledge with technical and engineering progress. Historically since the eighteenth century collections, museums, botanical gardens,... were the repositories of biological diversity. This *ex situ* conservation is obviously fragile, analytical, physically and financially limited. *In situ* conservation was conceived as a step forward in the form of parks, restricted and protected areas.... This is still an excellent approach to maintain specific ecosystems Germ banks. DNA banks, cryoconservations of tissues are the modern versions of collections and have multiplied the potential of conservation. More recently the advances in the field of population genetics open the way to a dynamic management of collection. Theories were elaborated and tested by experimentation that can predict, knowing the reproductive properties of a species, what procedures should be followed to maximize genetic diversity in a conserved population and how to maintain it through generations. In the case of wheat programs have been successfully devised through which with a minimized investment genetic diversity of a collection will increase with time (19). It is clear that such procedures are difficult to standardize and must take in consideration the peculiarities of each species (lettuce and oak are not amenable to the same operations during the life-time of an experimentator or a manager...). But on the whole management of biodiversity is accessible provided the three golden rules of conservation are obeyed : identify biological diversities before launching a possibly devastating exploitation of a resource, set up protection procedures, let biological changes and evolution follow their ways and maintain a man-guided intervention in these processes.

Since science and technology provide information and procedures to carry on a careful management of biological diversity, whether plants, animals or microorganisms are concerned, why has **Biodiversity** become a global political

stake ? Why is the impression that time is running out so pervasive ? Reasons are many and one of them is that the majority of still unknown resources reside in the tropics where local or foreign exploitation precedes knowledge, where poverty creates imperative necessities. The second major difficulty biodiversity management faces is the level of people's awareness. Information and education certainly need improvement. But the bottom question is the appropriation of biological resources (private or common ?) and the fair recognition of the responsibilities and the rights of all the social partners involved locally, nationally and internationally. Of course the rate of usage over renewal of living forms is the crucial figure. In order to survive societies will have to act simultaneously in the future on usage and on a man-guided renewal. But biology alone will not tell decision-makers whether large and immediate profits are preferable to sustainable development or whether some putative future is a good reason to prevent poor people from drawing today their food and fuel from nature...

Acknowledgments :

The authors are supported by grants from CNRS, Université Paris-Sud and EPHE.

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Table I

NUMBERS OF IDENTIFIED LIVING SPECIES IN 1991

Viruses	5 000
Bacteria	5 000
Protoctists	30 000
Ferns, fungi,...	174 000
Plants	250 000
Invertebrates	990 000
Vertebrates	45 000

The ratio between known species and those still to be identified is around 1/5. It varies considerably from some 4 % for viruses to almost 100 % for mammals.

Table II

GENETIC DIVERSITY

Species diversity

1.6 x 10⁶ species identified in 1991
10⁷ species on earth
10³ in man vicinity

Turn-over

Mean half-life : 10⁵ - 10⁶ years
3.7 x 10⁹ years of Life on earth

Intraspecific diversity

Mutations and recombination :
individual diversity

Mean individual half-life : 1 year

Table III

MOST OF THE ESSENTIAL CULTIVATED PLANTS HAVE BEEN GENETICALLY ENGINEERED

Rice, Corn and Wheat

Cotton

Rape, Sunflower and Soya

Potato and Beet

Tomato, Lettuce, Cucumber and Melon

Rose and Chrysanthemum

Tobacco

Alfalfa

Poplar

Table IV

TOMATO GENETIC DIVERSITY

9 related species :

Lycopersicum esculentum (the cultivated plant),
L. pimpinellifolium, *L. cheesmanii*,
L. hirsutum, *L. parviflorum*,
L. chmielewkii, *L. chilense*,
L. peruvianum, *L. pennellii*.

Hundreds of populations sampled by farmers, collectors, plant breeders.

Thousands of varieties in collections and traditional plant breeding programs.

Hundreds of lines from genetic engineering in plant molecular biology laboratories.

Finally one commercial line authorized in the US on a large scale: Flavr Safr.

Table V

DIVERSITY OF GENE SOURCES FOR MOLECULAR ENGINEERING OF PLANTS

Transgenic plants have been constructed
with genes from all the living kingdoms

Origins of introduced genes	Examples of character transferred
The same species or close relatives	Lectin genes for insect resistance...
Very different plant species	Genes for proteins rich in methionine...
Bacteria	Herbicide resistance genes...
Viruses	Capsid genes for virus resistance...
Animals	Immunoglobulin genes, anti-freeze proteins genes...

Figure 1

AUTHORIZED FIELD TESTS OF
GENETICALLY ENGINEERED PLANTS

