

## ***Ex situ* conservation and rare plants propagation in the Lecce Botanical Garden: reproductive biology problems**

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**Abstract** — Among nature conservation strategies, the cultivation *ex situ* in Botanical Gardens is the more immediate and allows the middle and long term survival of risk extinction and rare species. The conservation of such species imposes, first of all, direct observation of principal phases of development *in situ* and the correct collecting of the propagation material. Choosing the fitter and faster multiplicative technique surely offers greater opportunities of conservation *ex situ*, with elevated phenotypic and genotypic expressiveness populations, and it allows to get an high number of individuals to be re-introduced in origin places, with the purpose to strengthen the consistence of the natural populations and to reduce its vulnerability degree.

**Key words:** Botanical Garden, conservation, reproductive biology, vegetable biodiversity.

### **INTRODUCTION**

The Lecce Botanical Garden, born only few years (1992) as “Center of studies for Mediterranean botanical species protection and conservation, with attached botanical garden”, proposes itself to the public and the scientific world with finality that aims above all to conservation *ex situ* of threatened species, (in captivity propagation) to scientific culture divulging, to environmental retraining projects (MARCHIORI and DEDEJ 2000).

The most important goal in a modern Botanical Garden is protecting the biodiversity, attentively appraising the phyto-geographical context where it is inserted and privileging the flora, particularly that autochthonous and threatened one. With these premises, the Botanical Garden of Lecce shapes him as a modern structure not only directed to maintenance, but also to sensitization and active sharing in programs of recovery and environmental safeguard. The principal activities of such Institution essentially pursue the followings objectives: territorial biodiversity analysis; propagation material harvest; propagation and cultivation protocols debugging; principal natural habitats structuring species multiplication; alive thematic collections coltivation; back-to-nature and restauration interventions in degraded environments with reintroduction in nature environment of species that characterizes, from the physiognomy point of view the principal habitats of

Salento; didactic-divulging activity (ACCOGLI *et al.* 2000).

The Lecce Botanical Garden proposes safeguard programs which foresee a direct intervention in restauration projects of seriously threatened or impoverished habitat or environments, through reconstruction or expansion of natural populations; therefore common Mediterranean Stain, hinterland principal vegetable associations, coast and wetland typical species are multiplied in special collections.

The vegetable species retrieval and harvest activity allowed to individualize possible bio-genetic reserves on the territory, it motivated studies on phenology and on seeds germinating potentiality, and last but not least, the local eco-types recognition.

Annual species, whose collections are maintained by renewing every year the seeding, are picked up and preserved with a simple deposit for a short or middle term maintenance and in a Germplasm Bank for a long-term conservation.

The Botanical Garden therefore represent a studies and monitoring center of the territory of its pertinence, which effects Nature Conservation programs, calibrated, according to different local realities, according to a net that can also interacts to a regional or national level, pursuing a global safeguard program.

The retrieved and multiplied vegetable species in the Garden represent an ample part of Apulia vegetable biodiversity and they constitute a mothers plants reserve from which retrieving material for multiplications. The principal alive collections coltivated in the Garden are articulated along a demonstrative path:

- rare and endemic species collection inserted in the Regional and/or National Red List and phyto-geographical elevated value species collection;
- Catalogue Field of spontaneous species of pharmacological or officinal interest;
- Catalogue Field of wild species ancestors of those of agronomic interest;
- Fruit trees species and horticultural species, ancient local cultivar in extinction danger collection;
- Idrophyte species of handicraft interest (*Juncus* sp.pl., *Typha latifolia* L., *T. angustifolia* L., *Phragmites australis* (Cav.) Trin.) collection;
- Mediterranean maquis and dunes structuring species collection;
- Collection of aromatic spontaneous plants of gastronomic or industrial interest.

Among the alive collections, greater scientific importance surely assumes that one constituted by rare and extinction risk species, for which the Botanical Gardens have the institutional assignment to adopt opportune maintenance strategies, both in situ that ex situ (IUCN-BGCS & WWF 1989).

Particular attention has been turned to the propagation and maintenance *ex situ* of the species distributed on the territory of Apulia and included in the Red Lists (National and Regional) of which few is still known regarding their ecological role, to their real distribution and reproductive biology.

The few preceding knowledge regarding the multiplication forms of such species motivated the attempts of propagation and protocols debugging of cultivation, maintenance and management of alive collections with calendars layout of fruits harvest (ACCOGLI *et al.* 1999). In Apulia, the plants exposed to risk are 180 (CONTI *et al.* 1992; CONTI *et al.* 1997). Further updating, gave a regional "checklist" that includes 184 "taxa", of which 19 already extinct in nature and 5 they represent wrong signalings, for which the real contingent to be preserved *ex situ* amounts to 160 species; which mostly (almost 50%) are distributed on the Gargano, in pedo-climatic conditions difficult to be reproduced in the Botanical Garden of Lecce. The investigations on distribution of sur-

vival risk species, made accessions record of propagated material related to 76 species, that represent almost the 48% of the total. The 88% part are represented by "salentine" species, confirming the garden appointment in the biodiversity safeguard of the pertinence territory (ACCOGLI *et al.* 2004).

## MATERIALS AND METHODS

The propagation material, picked on the territory, consist in: fruits in maturation, seeds, radical tiller, scions.

The complete maturation of the fruits has been completed in checked environment.

In the receptacle of the capolinis of the composite ones, often larvas and parasites develop and damage seriously the fruits ("cipsele"), therefore their extraction must be effected immediately after the harvest. The same problem occurs for many not dehiscent fruits, which are allowed to dry for facilitating the seeds extraction: the larvas that develop feeding themselves with flower organs, jeopardize more seriously fertile seeds production.

When possible, for the same specie, both the vegetative propagation that generative has been effected, using natural or universal substrata, the more possible similar to those of origin habitats. For seeding, scions and transplantations, it has been used PVC containers, maintained in cold tunnel or situated in full field, trying to get the optimum yield, under water contribution or atmospheric agents exposure diversified conditions. Annual repetitions of propagation and germinating seeds tests brought to define, almost for every specie, the optimal period of harvest of generative and vegetative material.

We tried to establish the optimum of propagation yield under not checked conditions and we recorded events that normally occur in nature, as the competition with infesting species plantule, phytophagous insects preys (aphids and snails), conditions of water stress (prolonged aridity or damp) that jeopardize the germinating yield.

## RESULTS

The adopted multiplication technical success also depends by correctly collecting the propagation material you, by the exact knowledge on the species biological form (Fig. 1) and as well as by the harvest timing.

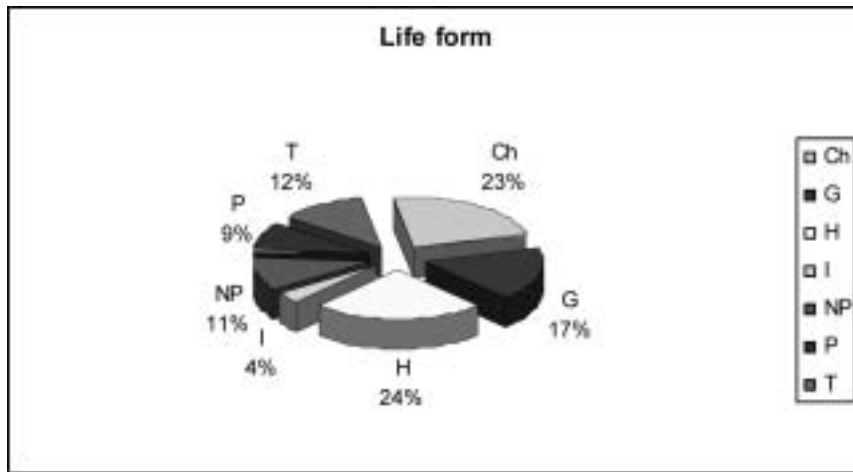


Fig. 1 — Life form spectrum of the propagated species.

The geophytes (G) can be propagated both by generative or vegetative way. They, in fact, generally produce seeds in abundance, but with very different germination percentages, according to the species. For orchids, for instance, a generative propagation is practically impossible, because of difficulties in seeds harvest at the opportune moment and the problems related to germination, often tied up to the presence of micorrhizas.

In general, for the bulbs, the vegetative propagation is easier, because of the short bulbs that are produced at the base of the principal bulbs or inside the inflorescence from which they detach themselves when they reach such dimensions to be able to conduct autonomous life. This propagation type, even if it is successful, is rather slow and it doesn't allow to have mature individuals in brief times.

Other biological forms (excluded the terofites that must be sowed every year), the withdrawn material for vegetative propagation consisted in scions, propagules, tiller, stolons, rhizomes, tufts, in short, portions of plant that contain gems able to reconstruct new complete individuals with well formed radical and aerial apparatus.

When seeds or propagules were not available (for instance, for *Isoetes histrix* Bory, *Marsilea strigosa* Willd., *Cheilanthes vellea* (Aiton) F.Muell. and all collected Orchidacees) it was necessary transplantation of the whole plant and the continuation of its reproductive cycle in the Garden (Fig. 2).

For some cases (for instance: *Nymphaea alba* L. ssp. *alba*, *Ranunculus baudotii* Godr, and others) we proceeded only by vegetative propagation, for of seeds production lacking in the planted out

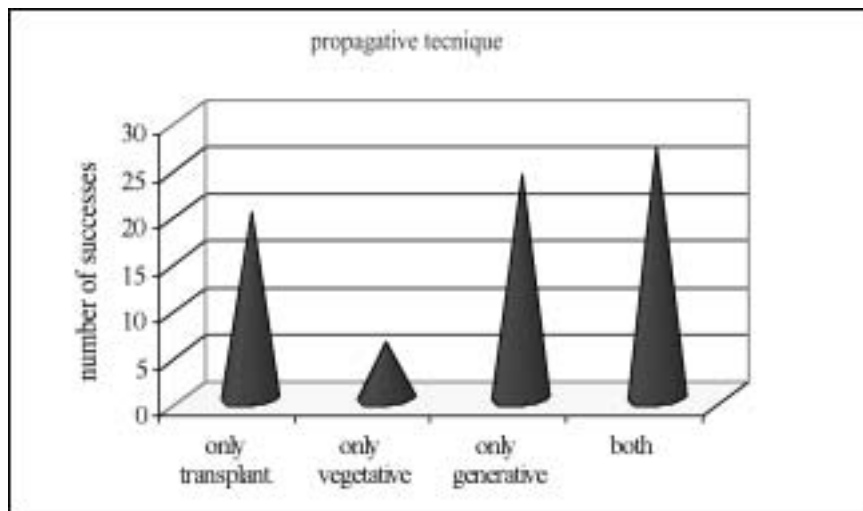


Fig. 2 — Propagative technique.

individuals; instead, the seeds have very often been so numerous to immediately guarantee a large number of individuals (*for instance: Echinops spinosissimus* Turra, *Prunus webbii* (Spach) Vierh., *Umbilicus cloranthus* Heldr. et Sart., *Quercus ithaburensis* Decaisne ssp. *macrolepis* (Kotschy) Hedge). In these cases the vegetative propagation was not necessary.

For having a general picture of multiplication activity course regarding under-risk species, we effected quantitative survey of obtained successes both for vegetative and generative ways, dividing them in classes, according to recorded percentage (Fig. 3). We did not considered cases when species did not survived to transplantation.

In the greater part of the cases, both vegetative and generative propagation shown a success superior to 50%, that could decidedly be improved by effecting the propagation under checked conditions (of temperature, light and damp).

The scarce success cases individualization, above all for generative propagation (for instance: *Limoniastrum monopetalum* (L.) Boiss, *Iris revoluta* Colas., *Linum tommasinii* Rchb. and others), underlines necessity for further investigations on why of the reproductive potential reduction, that in these cases is transferred in ovulis missed fertilization because of incomplete development of the embryonic sack or for non-arrival of pollinic tube. It is known  $Ca^{2+}$  role during germination and extension of pollinic tube and on the stigma, on ovary, in vital seeds formation: the correct exogenous and endogenous concentration is fundamental for a correct reproductive cycle (GRILLI CAIOLA 1995). The safeguard necessity of so rare

and spotted distribution species, makes necessary further investigations about reproductive processes which are dependent on exogenous factors as temperature, photoperiod, damp, nourishing and substratum oligo-elements, but also on endogenous factors as chromosomal order, that determines the gametofitic compatibility (ACCOGLI *et al.* 2006)

## CONCLUSIONS

Territory species and habitats recovery passes through the study of the reproductive cycles in animals and vegetables organisms, but, above all, through the study of genetic and epigenetic factors which regulate their themselves cycles, as well also through the study of microclimatic factors and anthropic action. These factors determine not only the number of individuals in the populations, but also their ability to sustain themselves in course time with production of fertile seeds.

The success levels recorded during effected propagation tests, can help deciding to use more generative rather than vegetative propagation, according to necessity to have fertile and well developed individuals to re-introduce in origin sites; we must however consider that vegetative multiplication in brief time well developed individuals determines even if it already give an omozygotic.

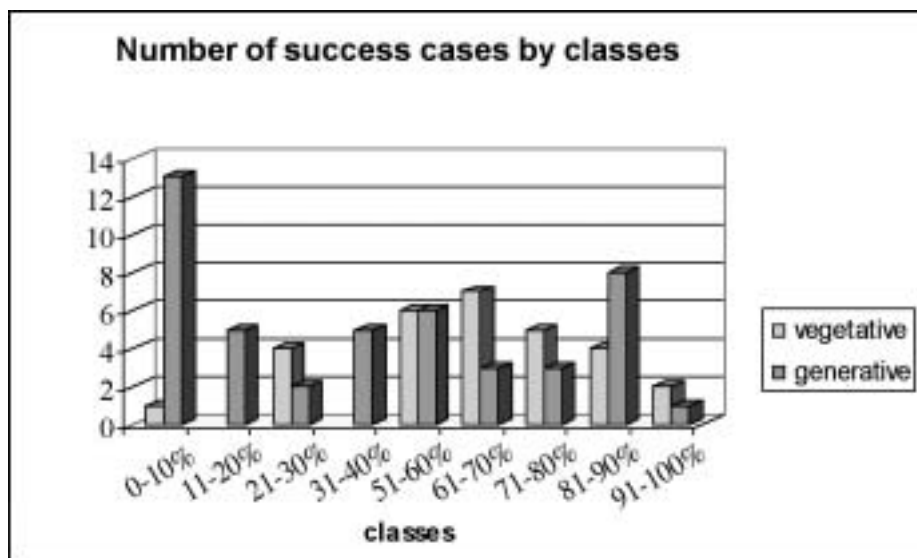


Fig. 3 — Number of success cases by classes.

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