G. SERRATO VALENTI (*), F. RIVEROS (**)

PERFORMANCE OF *PROSOPIS* SPECIES (LEGUMINOSAE) IN SALINE HABITATS AND THEIR UTILIZATION IN A PLANT INTRODUCTION PROGRAMME IN THE HANBURY BOTANIC GARDENS (LA MORTOLA, VENTIMIGLIA)

Abstract — One of the FAO's objectives is to develop strategies aimed at trasforming desert ecosystem into productive agricultural land. To this purpose, several studies on *Prosopis* species were started several years ago and are in progress. These species were chosen owing to their stress tolerance and their nutrizional value.

P. juliflora is more salt tolerant than *P. tamarugo* which in turn is more so that *P. cineraria*. Establishing these species in the Hanbury Botanic Gardens would have a double aim: the cultivation and preservation of very beautiful plants and the transfer to the field of the laboratory tests.

Riassunto — Specie del genere Prosopis (Leguminosae) in habitats salini e loro introduzione nei Giardini Botanici Hanbury (La Mortola, Ventimiglia). Uno degli obiettivi della FAO è la ricerca di strategie atte a trasformare ecosistemi desertici in terre agrarie produttive. In questo ambito, alcune specie del gen. Prosopis sono state e sono tuttora oggetto di studi, per la loro straordinaria stress-tolleranza e per la loro valenza nutrizionale. P. juliflora è più resistente alla salinità di P. tamarugo a sua volta più resistente di P. cineraria. Queste specie verranno introdotte nei Giardini Botanici Hanbury (La Mortola) allo scopo di conservare bellissime piante e di trasferire in campo le esperienze di laboratorio.

Key words — Prosopis sp. - salt-tolerance - Hanbury Botanic Gardens (La Mortola).

For over 100 years the Hanbury Botanic Gardens have housed in Italy, together with free-growing untouched luxuriant specimens of Mediterranean scrub, exotic plants from all over the world. Indeed, one of the specific aims of these gardens is to preserve interesting species. These include not only rare endemic species, relicts, endangered plants, but also, in particular, plants that for reasons which may differ

^(*) Istituto Botanico «Hanbury» dell'Università, Genova.

^(**) Crop and Grassland Service, FAO, Roma.



Prosopis juliflora, a highly versatile tree. Pod harvest.

greatly from one case to another, are the subject matter of studies. It is obvious that special attention is paid to essences useful to man. In the Hanbury Gardens, bordering on the sea shore and featuring a dry climate, many plants can find a habitat that is favourable for their introduction. In this context, the Gardens would like to plant some species of *Prosopis*, specifically in the area located closest to the sea and least protected from the South winds. These species are of great interest for the development of pauperized arid and semi-arid lands. This genus, in fact, is recognized as being a highly significant resource for the present and future welfare of mankind, providing an insurance against world starvation (RIVEROS, 1988).

It is well-known that cutting of forests, the degradation of the soil, and the consequent desertification of the land have produced and continue to produce all over the world, albeit to very different extents, enormous losses in terms of agricultural and forestry production, with serious — in some areas extremely so — conseguences for man, who caused them. In some countries (in India and Latin America, for example), projects for recovering the land have been drafted and implemented for some time now, however it is only in recent years that awareness of the serious nature of this issue has begun to surface.

One of FAO's most urgent objectives is therefore to develop strategies aimed at transforming desert ecosystems into productive agricultural land. With this in mind, the choice of stress-tolerant plants is of fundamental importance. What are needed are plants able to withstand extreme conditions but which, at the same time, can be of use to man both directly as a source of nutrition or as raw materials for essential activities or vital needs (for building houses, as a source of warmth, medication, and so on), or indirectly as animal fodder, making it possible to build up an efficient food chain. Several features typical of the Prosopis genus give it a unique standing in this respect. Although its species also grow near water, some have been found to survive in very dry climates where few other plants could. Prosopis can live and even prosper in infertile soils with a high salt content. It normally requires a minimum yearly rainfall of 250 mm, but some species will adapt to a mean of 75 mm or even less. Even in prolonged periods of drought they are resistant enough to continue producing plenty of pods. These are most important part of the plant, and indeed Prosopis pods are among the oldest known foods, used by prehistoric man in the western hemisphere and still, today, a source of proteins and carbohydrates for many North and South American desert populations. What is more, in Hawaii, Perù, Chile and Argentina the pods from Prosopis forests are used to feed livestock. Their nutritional value is comparable to that of barley and maize. Yet another interesting aspect is that some Prosopis species are an excellent mean for keeping erosion under control, for maintaining the shape of coastal dunes, for use as windbreaks, for reclamation of degraded grasslands and wastelands, where no other valuable tree species would easily grow, etc. (HABIT et al., 1981; MUTHANA, 1988; DUTTON et al., 1992).

Several studies on three species of *Prosopis* (*P. iuliflora*, *P. tamarugo*, and *P. cineraria*), were started several years ago and are in progress. Their aim is to screen these species in order to identify the best-suited to withstand the desertification process. The research activities covering these three species are part of a broader programme developed by the FAO Grassland Group, and their purpose is to enhance their use in those areas where the *Prosopis* species can be integrated into the existing farming and livestock production systems (RIVEROS, 1992).

Prosopis iuliflora, is native to South and Central America (MAYDELL, 1986), *P. tamarugo* comes from Chile (HABIT *et al.*, 1981), and *P. cineraria* is indigenous to south Persia, Arabia, Afghanistan, Pakistan (Punjab, Sind, Baluchistan) and India (RECHINGER, 1986).

In the course of our investigations, we have examined mainly the effects of salinity on the anatomical structure of the roots, leaves and stems of seedlings grown in the presence of 0-600 mM NaCl (SERRATO-VALENTI *et al.*, 1989; SERRATO-VALENTI *et al.*, 1991; SERRATO-VALENTI *et al.*, 1992). It is well known, in fact, that in arid lands one of major problem is very often the high salt content of the soil.

The seeds of *Prosopis* species were obtained from the FAO seed bank, Rome (Accession numbers: 73500 for P. iuliflora, 72339 for P. tamarugo and 73499 for P. cineraria) and were stored at 4°C. The seeds were surface sterilized with 0.5% (w/v) sodium hypochlorite for 1 min and then washed two or three times in distilled water. They were sown in flasks on 250 ml of 0.25 strength Hoagland's nutrient solution (NOGGLE and FRITZ, 1976) solidified with 0.9% Difco Bacto agar. The pH of the medium was adjusted to 5.8 before autoclaving. The cultures were kept in a growth chamber at 25 ± 1°C and exposed to 3000 lx by means of daylight fluorescent tubes under light/dark cycles of 12/12h. The salinity treatments consisted of 200, 400 and 600 mM NaCl added to the basic medium and a control without added salt was also included. In order to observe them under the light microscope, the root, stem and leaflet segments were fixed for 24 h in FAA, then dehydrated in a graded ethanol series and embedded in JB4 resin (Polyscience Inc., Warrington, PA) in BEEM capsules (BRINN and PICKETT, 1979). All tissue blocks were sectioned at 5-7 μ m on a Reichert Om U2 ultramicrotome equipped with a glass knife. The sectioned material was stained with Toluidine Blue 0 (TBO) 0.05% in acetate buffer 0.1 M at pH 4.4 for 1 min, as metachromatic stain (FEDER and O'BRIEN, 1968). The section were photographed on Ilford FP4 film. We are giving here a table summarizing the most significant salinity data of three species investigated, at stage of 45 d after emergence of the radicle.

P. iuliflora, P. tamarugo and *P. cineraria* showed differential anatomical changes in the roots, stems and leaves when were grown in presence of increasing NaCl concentrations. In *P. iuliflora* the roots of the seedlings grown in 200 and 400 mM NaCl had a more advanced secondary structure than the controls. In the stem, as salinity rose, an increase of the cambium, of the phloem and of the sclerenchyma was noted. At 400 mM NaCl the formation of deuteroxylem started. Also at 400 mM NaCl, in the leaves, there was an increased number of water storage cells as compared to seedlings grown in the absence of NaCl or with a lower molarity. The main vein was considerably more developed. In *P. tamarugo* the anatomical features of the root and stem were differently affected by salinity. In the root, the differentiation of the secondary structure occurred later in the salt-grown plants than in

		200 mM NaCl	400 mM NaCl	600 mM NaCl
	root	very thick exodermis, advanced secondary structure	very thick exodermis, advanced secondary structure, lignified pith	rarely emerged
Prosopis juliflora	stem (epicotyl)	advanced phloem, cambium and sclerenchyma, no secondary xylem	even more advanced phloem, cambium and sclerenchyma, beginning of secondary xylem	rarely emerged
	leaf	more water storage cells	even more water-storage cells, larger mid-vein	rarely emerged
	root	delay in the differentiation of the vascular system	marked delay in the differentiation of vascular system	serious structural alteration
Prosopis tamarugo	stem (hypocotyl)	early differentiation of the seconday xylem	disorganisation of the vascular system	stunted or not emerged
	leaf	delay in the structural differentiation	probable beginning of tissue disorganisation	stunted or not emerged
	root	large amount of secondary xylem probable beginning of periderm	rarely emerged	not emerged
Prosopis cineraria	stem (epicotyl)	larghe quantity of secondary xylem	not emerged	not emerged
	leaf	very thick mesophyll, trend towards isolateral organisation	not emerged	not emerged

PERFORMANCE OF PROSOPIS SPECIES ECC.

All the results reported above concern the seedling stage of 45 d after emergence of the radicle. The comparisons were made with the controls of the same species, not between the different species.

those grown without salt. In the stem, secondary lignification started earlier in the salt-grown plants (200 mM) than in the control plants, and the vascular system became disorganised at 400 mM NaCl. In the leaflets, the structure differentiation was delayed in the seedlings growing in 200 mM NaCl, and the structure disorganisation seemed to start at 400 mM. The *P. cineraria* seedlings grown in 200 mM NaCl showed an increase in the thickness of the secondary xylem, both in the root and in the epicotyl, a greater thickness of the leaf mesophyll and a trend towards isolateral leaf organisation. The roots rarely emerged at 400 mM NaCl.

The above findings suggest that differential sensitivity to salt stress exits among organ types of the same species and among different species of the same genus. The stimulating effect of salinity on the differentiation of the secondary xylem is in any case frequent. Early lignification is often observed in halophytes and also non-halophytes grown in salt (MILLNER, 1934; SAADEDDIN and DODDEMA, 1986; SOLOMON *et al.*, 1986), and our own observations support this view. This development pattern could be interpreted as an adaptive change to facilitate water transport. Also leaf structure is especially plastic to saline stress and the literature shows that there is a conflict of opinion about it (NIEMAN, 1965, WIGNARAJAH *et al.*, 1975, LONGSTRETH and NOBEL, 1979, ROBINSON *et al.*, 1983; CURTIS and LÄUCHLI, 1987).

It is clear, in any case, that *P. cineraria* is more sensitive than *P. tamarugo* which in turn is more so than *P. iuliflora*. This fact concurs with previously published data on *P. iuliflora* (MUTHANA, 1988) according to which this species has proved to be the most versatile plant for harsh environmental conditions.

Establishing these species in the Hanbury Botanic Gardens would have a double aim: the cultivation and preservation of these extraordinary and very beautiful plants and the transfer to the field of the laboratory tests concerning the response of these plants to the environmental characteristics, in particular to the salinity of the medium.

REFERENCES

- BRINN N.T. and PICKETT J.P. (1979) Glycol methacrylate for routine, special stains, histochemistry, enzyme histochemistry and immunohistochemistry. J. Histochem. Cytochem., 2, 125-130.
- CURTIS P.S. and LÄUCHLI A. (1987) The effect of moderate salt stress on leaf anatomy in *Hibiscus cannabinus* (kenaf) and its relation to leaf area. *Am. J. Bot.*, **74**, 538-542.
- DUTTON R.W., POWELL M. and RIDLEY R.J. (1992) *Prosopis* species. Aspects of their value, research and development. CORD, Durham, 320 pp.
- FEDER N. and O'BRIEN T.P. (1968) Plant microtechnique: some principles and new methods. Am. J. Bot., 55, 123-142.

- HABIT M.A., CONTRERAS D.T. and GONZALES R.H. (1981) *Prosopis tamarugo*: fodder tree for arid zones. FAO, Plant Production and Protection Paper, 25, FAO, Rome, 110 pp.
- LONGSTRETH D.J. and NOBEL P.S. (1979) Salinity effects on leaf anatomy. *Plant Physiol.*, **63**, 700-703.
- MAYDELL H.J. (1986) Trees and shrubs of the Sahel. Their characteristics and use. **196**, 356-358. Schriftenreihe der GTZ, Eschborn, 525 pp.
- MILLNER M.E. (1934) Anatomy of *Silene vulgaris* and *S. maritima* as related to ecological and genetical problems. I. Root structure. *New Phytol.*, **33**, 77-95.
- MUTHANA K.D. (1988) Prosopis juliflora (Schwartz) DC, a fast growing tree to blossom the desert. In: HABIT M.A. and SAAVEDRA J.C., ed., The current state of knowledge on Prosopis juliflora: 133-143. FAO, Plant Production and Protection Division, Brazil, 554 pp.
- NIEMAN R.H. (1965) Expansion of bean leaves and its suppression by salinity. *Plant Physiol.*, **40**, 156-161.
- NOGGLE G.R. and FRITZ G.J. (1976) Introductory plant physiology. Prentice-Hall, Englewood Cliffs, NJ. 688 pp.
- RECHINGER K.H. (1986) Mimosaceae. In: RECHINGER K.H., ed., Flora Iranica, **161**, 12-13. Akademische Druck-u. Verlagsanstalt. Graz.
- RIVEROS F. (1988) Foreword. In: HABIT M.A. and SAAVEDRA J.C., ed., The current state of knowledge on *Prosopis juliflora*, FAO, Plant Production and Protection Division, Brazil, 554 pp.
- RIVEROS F. (1992) The genus *Prosopis* and its potential to improve livestock production in arid and semi-arid regions. In: DUTTON R.W., POWELL M. and RIDLEY R.J., ed., *Prosopis* species. Aspects of their value, research and development: 237-248. CORD, Durham, 320 pp.
- ROBINSON S.P., DOWTON W.J.S. and MILLHOUSE J.A. (1983) Photosyntesis and ion content of leaves and isolated chloroplasts of salt-stressed spinach. *Plant Physiol.*, **73**, 238-241.
- SAADEDDIN R. and DODDEMA H. (1986) Anatomy of the 'extreme' halophyte Arthrocnemum fruticosum (L.) Moq. in relation to its physiology. Ann. Bot., **57**, 531-544.
- SERRATO-VALENTI G., GHEZZI F. e PESCE M., 1989 Modificazioni strutturali ed istochimiche in plantule di *Prosopis iuliflora* (SW.) DC. (Leguminosae) indotte dalla salinità del mezzo. *Atti Soc. Tosc. Sc. Nat. Mem.*, Serie B, **96**, 63-76.
- SERRATO-VALENTI G., FERRO M., FERRARO D. and RIVEROS F. (1991) Anatomical changes in *Prosopis tamarugo* Phil. seedlings growing at different levels of NaCl salinity. *Ann. Bot.*, 68, 47-53.
- SERRATO-VALENTI G., MELONE L., ORSI O. and RIVEROS F. (1992) Anatomical changes in Prosopis cineraria (L.) Druce seedlings growing at different levels of NaCl salinity. Ann. Bot., 70, 399-404.
- SOLOMON M., GEDALOVICH E., MAYER A.M. and POLJAKOFF-MAYBER A. (1986) Changes induced by salinity to the anatomy and morphology of excised pea roots in culture. *Ann. Bot.*, **57**, 811-818.
- WIGNARAJAH K., JENNINGS D.H. and HANDLEY J.F. (1975) The effect of salinity on growth of *Phaseolus vulgaris* L.I. Anatomical changes in the first trifoliate leaf. *Ann. Bot.*, **39**, 1029-1038.

(m. pres. il 14 luglio 1994; ult. bozze il 12 settembre 1995)