HISTO-ANATOMICAL ASPECTS REFERRING TO THE VEGETATIVE ORGANS ON SOME HALOPHYTES FROM ROMANIA

ASPECTE HISTO-ANATOMICE REFERITOARE LA ORGANELE VEGETATIVE ALE UNOR SPECII DE HALOFITE DIN ROMÂNIA

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Abstract. *The vegetative organs of halophyte species from Romanian flora have been histo-anatomically investigated in this study. Limonium gmelinii (Willd.) O. Kuntze (Plumbaginaceae), Bassia sedoides (Pall.) Asch. (Chenopodiaceae) and Spergularia media (L.) C. Presl (Caryophyllaceae) species have been collected and their organs subjected to standard procedure used in anatomical investigation. These species were collected from different saline habitats from Dobrogea. In this work, anatomical features were described and some peculiar adaptations were also evidenced: successive cambia phenomenon, succulence and typical salt glands. These adaptations found in halophytes organs attest the affinity with botanical families, and the very close relationships between plants and environmental conditions.*
Key words: *halophytes, salt glands, succulence, adaptation.*

Rezumat. *În acest studiu, am investigat din punct de vedere anatomic organele vegetative de la câteva specii de halofite din România. Speciile Limonium gmelinii (Willd.) O. Kuntze (Plumbaginaceae), Bassia sedoides (Pall.) Asch. (Chenopodiaceae) şi Spergularia media (L.) C. Presl (Caryophyllaceae) au fost colectate, iar organele lor supuse procedurii standard folosite în investigaţiile de anatomie vegetală. Speciile au fost colectate din diferite habitate salinizate din Dobrogea. În această lucrare am descris trăsăturile anatomice ale organelor vegetative evidenţiind unele adaptări particulare: fenomenul de policambie, suculenţa, glande saline tipice. Aceste adaptări atestă afinitatea speciilor de halofite faţă de familiile botanice, precum şi strânsa inter-relaţie dintre plante şi factorii de mediu.*
Cuvinte cheie: *halofite, glande saline, suculenţă, adaptare.*

INTRODUCTION

Plants that naturally grow on soils affected by salinity have developed over time a variety of morphological and anatomical adaptive features in order to considerable extend the time in which they are exposed to a high pressure of the habitat selection like: high salinity and aridity (Hameed and Ashraf, 2008).

Generally, during the development cycle the halophytes are facing two problem situations: the tolerance to high concentrations of salt in the habitat and the ability to absorb water from the soil with low potential (Grigore and Toma, 2008).

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The histo-anatomical characteristics can be theoretical and presumptive considered, adaptations to the abiotic stress conditions. These features can be one important point in the addition of halophyte species to an ecological group. The adaptation and evolution process has occurred under the consistent and prolonged action of the abiotic factors. The combination of high soil salinity and humidity was remarked chronologically with a major impact on anatomical structures.

MATERIAL AND METHOD

The plant material was represented by the halophyte species from the Romanian flora, adapted to complete their life cycle on the saline soils.

The investigated species were collected in anthesis, during the summer of 2012, from different saline habitats from Dobrogea, along the Black Sea shore (*Limonium gmelinii* Willd. O. Kuntze, *Plumbaginaceae*) and near salt marshes from Sulina (*Spergularia media* (L.) C. Presl, *Caryophyllaceae*) and Histria (*Bassia sedoides* Pall. Asch., *Chenopodiaceae*). Fixing the collected biological material was carried out in 70% ethyl alcohol, being preserved until the sectioning moment. The collected plant material has been fixed and preserved in ethanol 70 %.

The cross sections through the vegetative organs were performed using the hand microtome and the botanical razor. The sections obtained were then subjected to immersion in sodium hypochlorite for 20-30 minutes, washed with tap water, then staining: in the first stage with iodine green (1 minute) and then washed with ethanol; in the second step the sections were stained with carmine red (for 20 minutes), washed with water and, finally, fitting them in glycerol gelatin (Toma et al., 2000).

Subsequently, after obtaining the permanent slides were performed microphotographs using the NOVEX photon microscopy (Holland) and the digital camera Canon.

RESULTS AND DISCUSSION

The species investigated have special mechanisms of adaptation that allow them to cope with the harsh life conditions (increased salinity of the soil).

The cross sections through the root of all investigated species reveals a secondary structure resulting from the activity of both lateral meristems: cambium and phellogen.

In *B. sedoides,* the central cylinder is extremely thick, derived both from the activity of normal cambium, as well from that of successive cambia (Fig 1). Successive cambia phenomenon is a typical characteristic of many halophytic species (Grigore and Toma, 2006; Grigore andToma, 2010a).

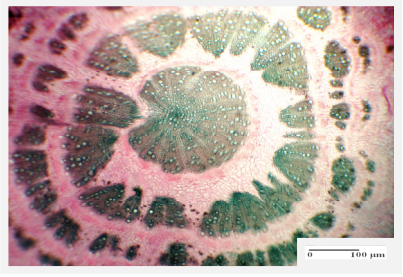

Fig.1 - Cross-section through the root of *Bassia sedoides*

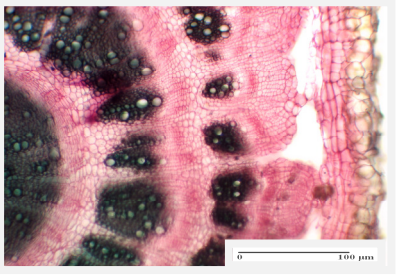

Fig.2 - Cross-section through the root of *Bassia sedoides*

From the activity of the additional cambia have resulted three rings of vascular bundles, of collateral type, close to each other, separated by parenchymatic- cellulosic rays. The size-of bundles decreases from the internal to the external ring (in which the tracheogenesis process is still noticeable).

The salinity can influence the degree of lignification in the root affected by additional cambia, while in the stem this lignification is less important (Grigore and Toma, 2006).

The primary root cortex in the species *Bassia sedoides* and *Spergularia media* is parenchymatic-cellulosic, with many large air-storing cavities (aerenchyma), more or less developed in the underground organs (Fig 2., Fig 3.). In *Limonium gemelinii* root, sclerenchymatic elements, sclereides-isolated or grouped can be observed (Fig 4.).

Fig.3 - Cross-section through the root of *Spegularia media*

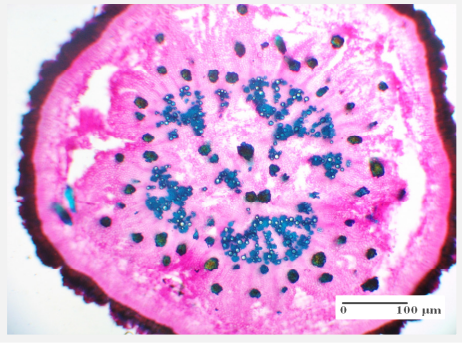

Fig.4 - Cross-section through root at *Limonium gmelini*

For the species *Limonium gmelinii* some cells that are part of the suberin roots contain tannin. The thickening degree of the walls and cell size decreases from the periphery to the center. This particular type of suberin is rarely mentioned in the literature (Metcalfe, 1972).

The rhizome of the species *Limonium gmelinii* has a root epiderma with a secondary structure both the to the periphery and the in the central cylinder which is very thick, having the conductive elements arranged in the form of concentric rings (Fig.5.)

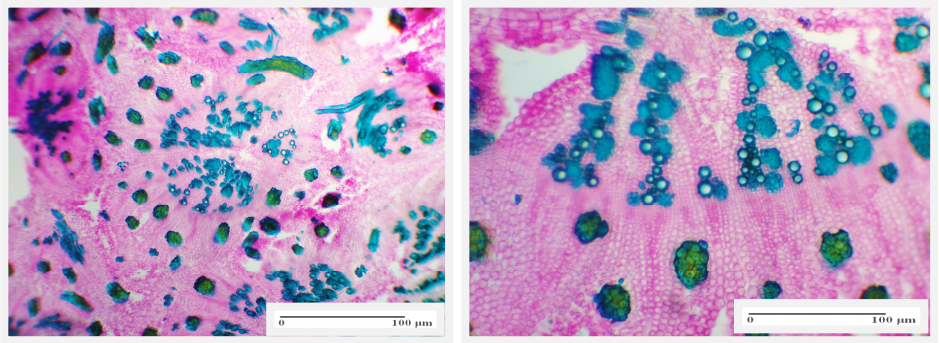

Fig.5 - Cross sections through the rhiyome of *Limonium gmelini*

The contour of the cross section of the stem of *Bassia sedoides* is circular, with protuberances. The epidermis has small cells, isodiametrical, with the external wall bulging. Tri-cellular protecting hairs, with very long terminal cell can be observed at this level. The cortex and medulla contain many cells with oxaliferous sand or druses of calcium oxalate (Fig.6.).

Fig.6 - Cross section through the stem of *Bassia sedoides*

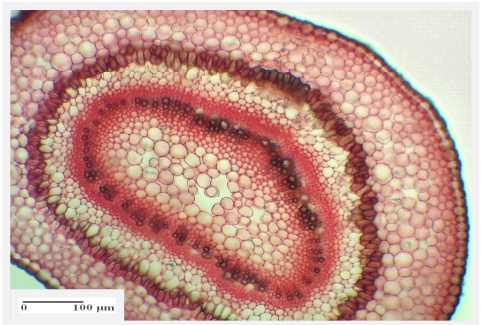

Fig.7 - Cross section through the stem of *Spegularia media*

The contour of the cross section of the stem of *Spergularia media* is rectangular-trapezoidal, and the structure is primary. This presents a sclerenchymatic pericycle in its outer side (Fig 7.).

The outline of the leaf lamina section for the species *Bassia sedoides* and *Spergularia media* is elliptic. In all the investigated species the midrib is slightly prominent on the underside of the lamina (Fig. 8., Fig. 9.). The epidermis has, in addition to normal, isodiametrical cells.

Stomata and long tri-cellular protecting hairs, with very long terminal cell in *Bassia sedoides*, while in *Spergularia* and *Limonium* the presence of the protecting hairs is not observed (Fig. 9., Fig. 10).

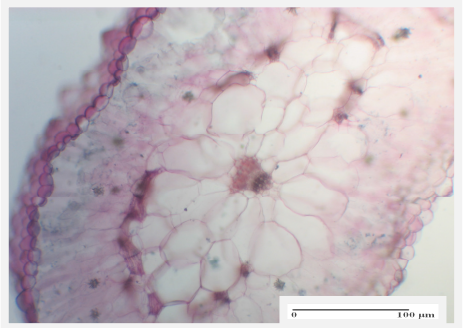

Fig. 8. Cross-section through the lamina of *Bassia sedoides*

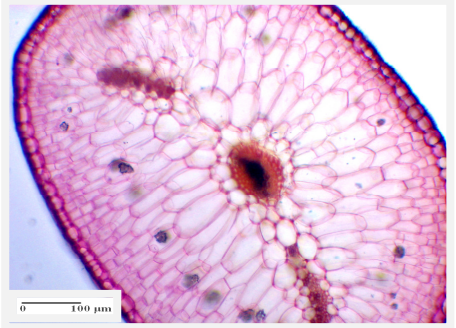

Fig. 9. Cross-section through the lamina of *Spergularia media*

For the investigated species (*Bassia, Spergularia*) the mesophyll is differentiated into palisade tissue, composed of cells whose size increases towards the center of the lamina (is observed islands with druses of calcium oxalate) and water storage parenchyma (3-4 layers of isodiametric cells, reduced for the

species *Limonium gmelini*). In the thickness of the mesophyll there are large mechanical idioblasts (Fig. 9). In the epidermis of *Limonium gmelini* Licopoli organs (secreting $CaCO_3$) can be noticed; these have a eight cells structure, consisting of four internal cells that transcribe a meatus and four external cells involved in the salt secretion adjusting also the transpiration (Fig. 11., Fig. 12.).

The halophyte species *Limonium gmelini* is adapted to uptake large amounts of salts from the soil, but it eliminates them on the surface of the leaf area, due to the presence of a large numbers of salt glands.

On dry weather, on the leaf surface, a layer of dry salt can be observed, blown by the wind or washed by rain (Grigore M.N.,Toma C., 2010b).

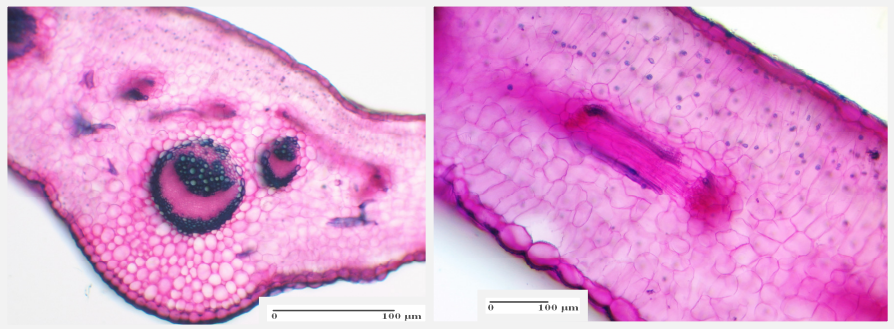

Fig. 10 - Cross-section through the lamina of *Limonium gmelini*

Fig. 11 - Lower epidermis of lamina of *Limonium gmelini* (surface view)

Fig. 12 - Image of the uper epidermis of *Limonium gmelini*

The succulence is an adaptive feature, present in the structure of the lamina of the investigated species (*Bassia* and *Spergularia*) and represents, generally, one of the main features involved in tolerance of plants on saline soils.

The halophytes accumulate large amounts of salts in the air organs; therefore they have the highest degree of succulence.

Further, the succulence has a dilution of the salts accumulated in plants, of toxic ions from cells, allowing the plant to cope with the large quantities of salt.

CONCLUSIONS

The investigated species have well established mechanisms that allow them to cope with the harsh conditions of life (excessive salinity of the soil, dryness).

The analysis of all these observations could lead to some preliminary ecological conclusions: the species *Bassia sedoides* and *Spergularia media* are meso-hygro halophyte and the species *Limonium gmelini* is xero-mesohalophyte.

The presence of mechanical elements (sclereids) in the organs structure for the species *Limonium gmelini* was also evidenced.

The anatomical characteristics of the analyzed species can be correlated with the environmental features in which plants grow. Therefore, the investigated species can be nominated as obligatory halophytes, being adapted to survive to high salinity of the soil.

The analyzed species have differences in the anatomical structure, but their adaptations may be considered converged to the environmental factors. *Bassia* presents the successive cambia phenomenon in the root level (typical for the most species of the *Chenopodaceae* family), the succulence of the lamina (particularly to *Bassia* and *Spergularia*) and the presence of the salt glands at *Limonium* (typical for the species of this genus). Therefore, the analyzed species in terms of histo-anatomical point of view show different strategies to adapt to the same stressful factors.

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