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COMPARATIVE HISTO-ANATOMICAL ANALYSIS OF THE VEGETATIVE ORGANS OF SEDUM TELEPHIUM L. SSP. MAXIMUM (L.) KROCK. IN VITRO AND FROM NATURE

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Abstract. In a histo-anatomic analysis of the exemplars of Sedum telephium L. ssp. maximum (L.) Krock. from nature and *in vitro*, the root presents a secondary structure, protected by a quite thin peridermis. The central cylinder bears vascular bundles, less numerous in vitro; the phloemic elements are grouped in small isles in both in vitro and from nature exemplars. The stem presents a few vascular bundles, where the xylem vessels bear thickened and lignified walls. The petiole reveals three vascular bundles. The foliar limb is amphistomatic, bearing anizocytic stomata in both in vitro and from nature exemplars, with homogenous mesophyll.

Key words: Sedum telephium L. ssp. maximum, in vitro

Introduction

Sedum gender belongs to Crassulaceae family [STEFAN & OPREA, 2007; METCALFE & CHALK, 1972] and consists of almost 400 species with succulent leaves. Sedum telephium ssp. maximum (L.) Krock. is frequent spread in the Romanian flora as spontaneous species, as well as ornamental cultivated species. More than that, the Romanian traditional medicine considers that this plant might have therapeutic (vulnerary, antiseptic, wounds) effects.

In the middle sixteenth century, Hieronymus Bock had reported that extracts of Sedum telephium ssp. maximum were used in Rhine valley to treat internal injuries like ulcers of the lungs. Now, medical researchers are isolating the active ingredients from those traditional medicine plants and testing their efficacy. In the early 1990's, researchers in Munich identified two polysaccharides in Sedum telephium ssp. maximum that were antiinflammatory [MULINACCI & al., 1993]. A few years later, Italian scientists observed the ways the polysaccharides and flavonols operated on cells during wound healing.

Material and methods

The medium where individuals of Sedum telephium L. ssp. maximum (L.) Krock. grew up was prepared after Murashige - Skoog prescription [MURASHIGE & SKOOG, 1962], by adding vitamins (1 mg/l HCl thiamine, 1 mg/lHCl pyridoxine and 1 mg/l nicotinic acid), growing regulators: 1 mg/l benzyl adenine (BA) and 1 mg/l β -indole acetic acid (AIA), 20g/l sacharose and only 7 g/l agar - agar; medium pH was adjusted to 5,6, then the culture medium was sterilised in the autoclave for 30 minutes, at 121°C.

Subcultures of Sedum telephium ssp. maximum have been done in glass jars of 10 cm high and 4,7 cm diameter. After inoculation, the jars were obturated with polyethylene

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sheet, fixed with rubber bands, then they were transferred to the growing chamber, illuminated with fluorescent white light, with light intensity of 1700 lux and a photoperiod of 16h/day (23°C) and 8h/night (21°C \pm 2°C).

In order to carry out the histological analysis, the vegetal material represented by the vegetative organs of *Sedum telephium* ssp. *maximum*, *in vitro* and from nature, came through the following steps:

The material has been fixed and preserved in 70% ethylic alcohol.

The sections were cut manually, by microtome, using elder pith as support. The histological sections were washed in sodium hypochlorite for 20-35 minutes, and then washed in acetic acid and distillate water [ANDREI & PARASCHIVOIU, 2003; ŞERBĂNESCU-JITARIU & al., 1983]. The sections were coloured with iodine green (1 minute), washed in 90% ethylic alcohol and distilled water then coloured with ruthenium red (1 minute) and washed in distilled water again. They were mounted in gel and analyzed in an Optika light microscope. The light micrographs were performed by means of the same light microscope, using Canon A540 camera. Drawings were obtained by employing a Romanian Projektionszeichenspiegel MC1 light microscope.

Results and discussions

The histo-anatomic cutting revealed the following aspects:

The root (Figs. 1-8)

The cross section evidences a thin peridermis with 4-6 layers of flattened cells, with thin walls. The cortex is thick, compact, not differentiated in exodermis, cortical parenchyma and endodermis, formed by 10-12 layers of cells with thin, cellulosed walls which form meatus of various dimensions. Here and there, division walls can be seen.

The central cylinder is compact, represented by 7-8 xylemic bundles (only 3-4 xylemic bundles in the *in vitro* exemplars), formed by vessels with lignified, but quite thin walls and numerous phloemic small isles, consisting of few elements (sieved tubes and guard cells, with cellulosed walls). Cambium is thin, formed by a few layers of cells where differentiation is in progress.

The aerian stem (Figs. 9-16)

The cross section has a circular profile. The epidermis is suberified, consisting of 2-3 layers of cells with thickened walls. The cortex is dense, formed by numerous layers of cells (only 6-8 layers of cells in the *in vitro* exemplars).

The central cylinder bears vascular bundles of various dimensions (4-5 in the exemplars from *in vitro*); the xylem has vessels with lignified and thickened walls, while the phloem consists of sieved tubes and guard cells. The medullary parenchyma bears big cells with cellulosed walls, which form meatus.

The leaf (Figs. 17-24)

The petiole (Figs. 17 and 21) has a crescent form in cross section. The epidemis bears izodiametric cells, having the external wall stucked out and covered by a thin cuticle (in the exemplars collected from nature). In both *in* vitro and from nature exemplars, the fundamental parenchyma is quite homogenous, formed by cells of various dimensions, with thin cellulosed walls. There are three vascular bundles, the median bundle is bigger than the others. The xylem consists of vessels with thickened walls, while the phloem is formed by sieved tubes and guard cells.

The foliar limb

Front side epidermis view (Figs. 19, 20, 23, 24):

The upper epidermis bears cells with waved walls and anyzocitic stomata (stomata surrounded by cells of various dimensions). The lower epidermis has a similar structure to that of the upper epidermis, but the cells bear more waved walls; as stomata are present in both epidermis, the foliar limb is amphistomatic.

The cross section through the foliar limb dispays common elements: the epidermis consists of big cells; their external wall is covered by thin cuticle; the mesophyll is homogenous, so the lamina has a normal bifacial-izofacial structure. There are a few vascular bundles, the one from the center is bigger than the others. The xylem bears vessels with thickened but unlignified walls, while the phloem bears sieved tubes and guard cells.

Conclusions

The root, the stem and the leaf present similar histological characteristics in both studied exemplars. The main differences regard quantitative aspects (various number of layers in the root cortex, various number of vascular bundles- xylemic and phloemic, different organization of the mixed vascular bundles in the leaf).

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Explanation of plates

PLATE I

- Cross section through the root (Fig. 1: from nature; Fig. 2: in vitro);
- Cross section through the root- peridermis (Fig. 3: from nature; Fig. 4: in vitro);
- Cross section through the root- vascular bundles (Figs. 5 and 7: from nature; Fig. 6 and 8: in vitro); PLATE II

Cross section through the stem (Fig. 8: from nature; Fig. 10: in vitro)

- Cross section through the stem- epidermis (Fig. 11: from nature; Fig. 12: in vitro)
- Cross section through the stem- central cylinder (Fig. 13: from nature; Fig. 14: in vitro)

Cross section through the stem- detailed vascular bundle (Fig. 17: from nature; Fig. 18: in vitro)

PLATE III

Anatomy of the leaf (petiole and foliar limb)

Legend: ep = epidermis, ep = epidermic cell, f pr = fundamental parenchyma, l ep = lower epidermis, mzph =mezophyll, phl = phloem, st = stomata, u ep = upper epidermis, xy = xylem; $Bar = 100 \ \mu m$







6

Fig. 7





7

Fig. 15

vlem





