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A COMPARATIVE ANALYSIS OF THE HISTOLOGICAL STRUCTURE OF THE AERIAL ORGANS OF PLANTS GROWN ON STERILE HEAPS AND RESPECTIVELY IN ORDINARY SOIL

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ABSTRACT. The living conditions offered by the sterile heaps are not favourable to the development of plants, especially if the heaps have not been placated with phytosoil. But there are a small number of species showing various degrees of tolerance towards these conditions, and that manage to grow in the respective areas (Gh. Bârlea, 2008).

Keywords: *Robinia pseudacacia* L., *Centaurea cyanus* L., *Polygonum cuspidatum* Sieb. et Zucc. (*Reynoutria japonica* Houtt.), sterile heaps, petiole, stem, aerial stem

INTRODUCTION

Our paper is going to make a comparison between the microscopic structure of certain aerial organs of plants that grew on the sterile heaps in Sasar and witness plants that grew in ordinary farming soil.

MATERIALS AND METHODS

Materials consist in: petiole of ***Robinia pseudacacia*** L. prevailed from a plant that grew in normal soil, and another plant that grew on sterile soil (from the heap in Sasar), a stem of ***Centaurea cyanus*** L.prevailed from a plant that grew in normal soil and a plant that grew on sterile soil (from the Sasar heap), an aerial stem of ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.), prevailed from a plant that grew in normal soil and from a plant that grew on sterile soil (from the Sasar heap), methyl red dye, optic microscope.

Methods consists in identifying the differences between the histological structure of the plants that grew on the sterile heaps (devoid of soil), as compared to the ones that grew in ordinary farming soil. In order to do this we have cut cross-sections of the aerial organs that were then dyed in red with methyl, and have taken comparative photos on the optic microscope, at first with a small lens (4X), then with a more powerful one (10X or 40X), in order to highlight the inner tissues. The witness-plants had been picked from farming areas not far from the Sasar sterile heap, but on the other shore of the river with the same name, so as not to be susceptible of the same kind of pollution. The identification of the species was done in accordance with „*Flora ilustrată a României*" (Ciocârlan V., 2000) and, "*A Magyar flora kepekben*" (Javorka and Csadody, 1968). These species had been decided on bearing in mind the fact that they can be found in large numbers in the Baia-Mare Depression and on sterile heaps.

RESULTS AND DISCUSSIONS

On analysing the microcopic samples of fresh plants we were able to conclude that differences exist between the plants that grow in ordinary farming soil and the plants from sterile heaps. These differences were analysed under the microscope and will be presented in the microscopic pictures that follow:

1. Petiole of ***Robinia pseudacacia*** L.

By comparison we can observe that:

- With the plant that grew on the sterile heap the cuticle is thicker which shows a tendency of self-defence against the polluting agents, especially the dust that goes up from the substratum, and then sets down on the leaves and the petioles (Fig.5).
- With the plant that grew on the sterile heap the outer bark is thicker (3 layers) also as a form of self-defence against the pernicious factors from the outside, whereas with the plant that grew in normal soil, and which does not need these alterations, the outer bark is thinner (2-3 layers).
- With the plant that grew on the sterile heap the inner bark is thinner (2-3 layers of cells) compared to the plant that grew in normal soil (with which the inner bark has 3-4 layers of cells), which shows a poorer growth on the polluting sub-stratum.
- No essential alterations can be observed in the structure of the strenghthening tissue in the plant that grew on the sterile heap, the tissue contains 3-4 layers that contain rare cells with chloroplasts that come from the bark.
- With the plant that grew on the sterile heap the area of the liber includes medullary rays, rare metaphloem. These are cells that contain chloroplasts that come from the bark.
- With the plant that grew on the sterile heap, most of the ligule is metaphloem, except that with the plant that grew in normal soil the ligule ring is larger, as the metaphloem ocupies most of the space.

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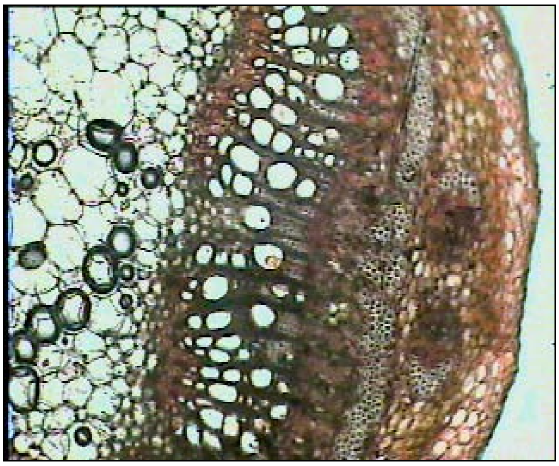

Fig. 1 Cross section in the petiole of *Robinia pseudacacia* L. A plant that grew in normal soil, x 10

Fig. 2 Cross section in the petiole of *Robinia pseudacacia* L. A plant that grew on sterile substratum, x 10

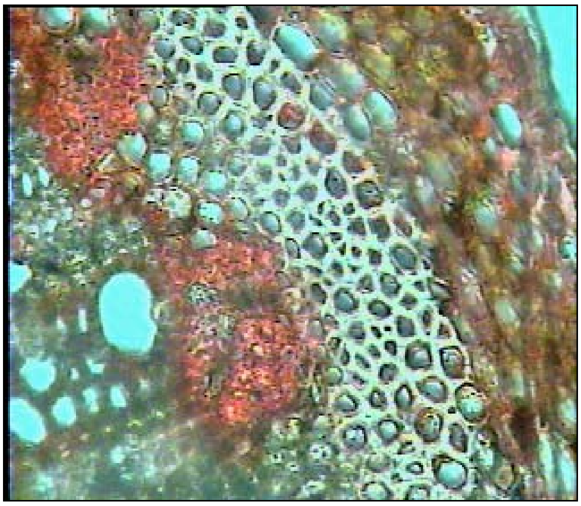

Fig. 3 Cross section in the petiole of *Robinia pseudacacia* L., a plant that grew in normal soil. Detail, x 40

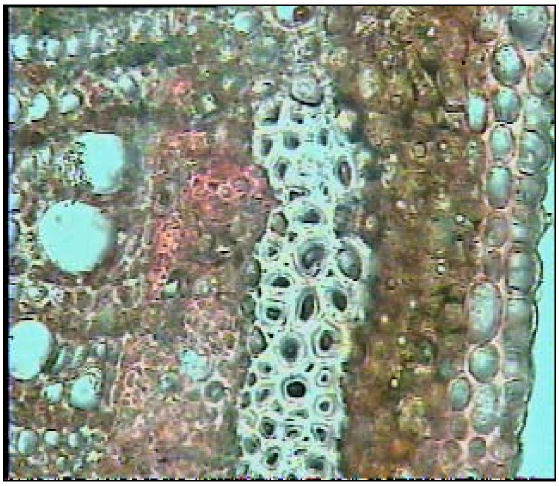

Fig. 4 Cross section in the petiole of *Robinia pseudacacia* L., a plant that grew on sterile substratum. Detail, x 40

Fig. 5 Polluting dust set down on the leaves of *Robinia pseudacacia* L. on the Săsar sterile heap

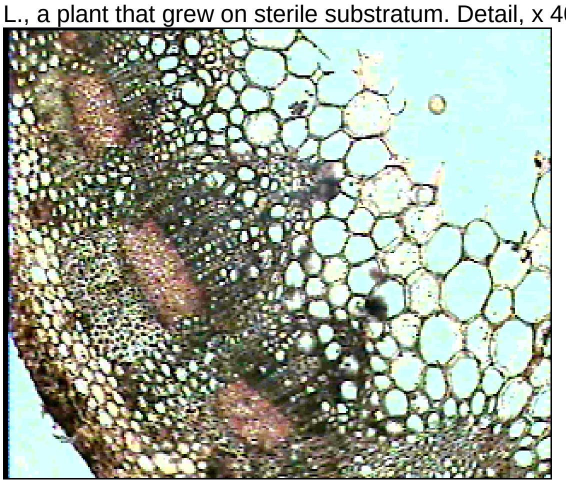

Fig. 6 Cross-section in a stem of *Centaurea cyanus* L., a plant that grew in normal soil, x 10

- With the plant that grew on the sterile heap, the ligule is not so well developed, probably because of the pollutant agents that hinder the normal growth of the tissues.

- With the plant that grew on the sterile heap there are no substantial alterations of the medulla and the cambium, which look normal both in the plant that grew on the sterile heap and the one that grew in normal soil, although the sclerenchyma, the collenchyma, and the liber tissue seem less developed.

2. A stem of ***Centaurea cyanus*** L.

The ***Centaurea*** genus is well represented on the sterile heaps, especially on the one from Sasar, not so much in the number of species, but mainly in the number of plants and the area on which they grow. This is the reason why I have decided to have microscopic samples of the stem, and to compare them with plants that grow in normal soil. Thus, one can observe that the plants from this species as well as other species that grow on the sterile, show a higher degree of weathering and ageing than the plants that grow in normal soil. Thus, at the level of the stem we can observe in the cross-section that:

- With the plant that grew on the sterile heap the epiderm is badly exfoliated compared to the plant from the normal soil, which shows a higher degree of weathering, a thing which can be seen with the naked eye through macroscopic analysis. This exfoliation may be due to the anorganic dust which causes the dehydration of the cells leading to the detaching of some of them and thus creating flaws.
- With the plant that grew on the sterile heap the outer bark is also more exfoliated than with the one that grew in normal soil (which also has 3-4 layers of cells), thus continuing the process started in the epiderm.
- With the plant that grew on the sterile heap the inner bark shows no flaws, but shows a slight tendency towards growing thinner (2-3 layers) compared to the plant that grew in normal soil (3-4 layers) most probably as a manifestation of the lack of the nourishing substances that the sterile soil has not been able to offer.
- With the plant that grew on the sterile heap the medullary rays are narrower compared to the plant that grew in normal farming soil with which the rays are normal up to the bark.
- With the plant that grew on the sterile heap the ligule (which contains more metaxylem) and the liber are narrower than normal, having denser fascicles, although it shows no other visible alterations, but the majority of tissues show the same direction determined by the scarcity of nourishing substances, which the sterile heap is incapable of ensuring if there is no adequate phyto-soil placating. In the witness plant, the ligule has metaxylem at the inside and protoxylem at the outside, the liber representing one third of the ligule thickness.
- With the plant that grew on the sterile heap as a reaction of defence against pollutant factors, and the poor nourishing conditions, the strenghthening tissue has a tendency of growth (being almost uninterrupted except for some very thin medullary rays) taking up some of the space of the medullary rays,too, a manifestation which can be observed in many other plants that grow in a polluted environment. With the witness plant, the strenghthening tissue is continual,set among the perixyl.
- With the plant that grew on the sterile heap the medulla is normal, but has smaller cells than the witness plant.

3. Aerial stem of ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.).

The aerial stem of ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.) is at least similar at the plant from the sterile soil to the plant grown in normal soil. Thus:

- With the plant that grew on the sterile heap, (Fig. 11), the cuticle is somehow thicker than with the plant grown in normal soil (fig. 12), against the background of the perrenial self-defence against an aggressive environment.
- With the plant that grew on the sterile heap the bark has several chloroplasts, 7-8 layers of cells similar to the one from the witness plant, showing no clear decelable microscopic alterations,except for the fact that it is thinner.
- With the plant that grew on the sterile heap the strenghthening tissue is better developed and more continual, rarely interrupted by medullary rays, against the same tendency of defence against the environment factors, whereas with the witness plant it is discontinual due to the perixyls (Fig.10).
- With the plant that grew on the sterile heap, the liber is more clearly fragmented than with the plant from the normal soil, a character which we have generally encountered at the plants from sterile soils, probably because of a sudden dehydration due to the loss of water at the level of the sterile substratum on the hill side.
- With the plant that grew on the sterile heap, the ligule contains less metaxylem, that is the ducts have not been able to grow to this stage, probably because of the lack of nutrients, besides the ample dehydration, whereas with the witness plant the ligulw has less than 20 % very compact metaxylem.
- With the plant that grew on the sterile heap the medullary rays are unaltered compared to normal (Fig.9), being clear, up to the strenghthening tissue.

With the plant that grew on the sterile heap the medulla has somewhat smaller cells than the normal plant, but no major differences are evident, except that the cells are larger in the centre, with fewer amyloplasts, the cells being smaller at the exterior and having more amyloplasts.

Fig. 7 Cross-section in a stem of *Centaurea cyanus* L., a plant that grew on sterile soil, x 10

Fig. 8 Cross-section in a stem of *Centaurea cyanus* L., *a* plant that grew in normal soil. Detail, x 40

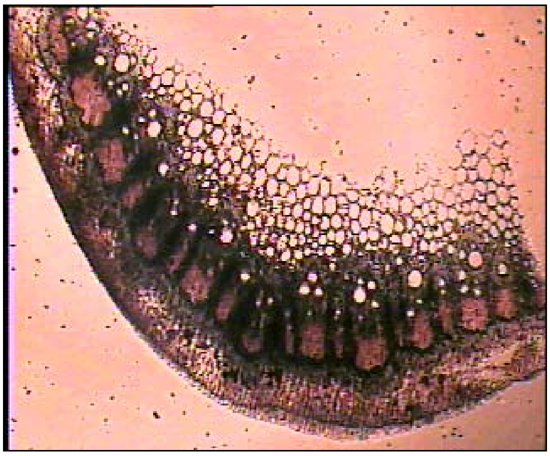

Fig. 9 Cross section in aerial stem of *Polygonum cuspidatum* Sieb. et Zucc. (*Reynoutria japonica* Houtt.). A plant that grew in normal soil, x 10

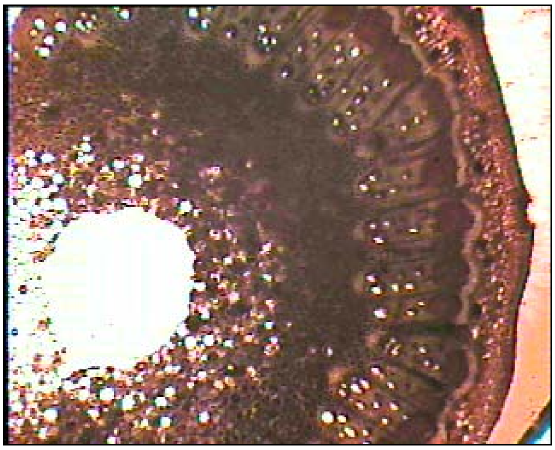

Fig. 10 Cross section in aerial stem of *Polygonum cuspidatum* Sieb. et Zucc. (*Reynoutria japonica* Houtt.). Plant grown in normal soil, x 10

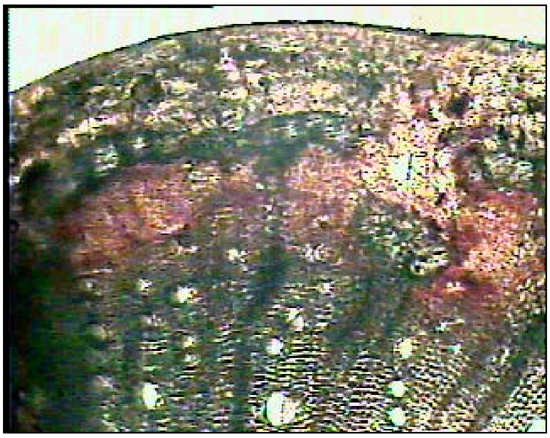

Fig. 11 Cross section in aerial stem of *Polygonum cuspidatum* Sieb. et Zucc. (*Reynoutria japonica* Houtt.). Plant grown in normal soil, x 10

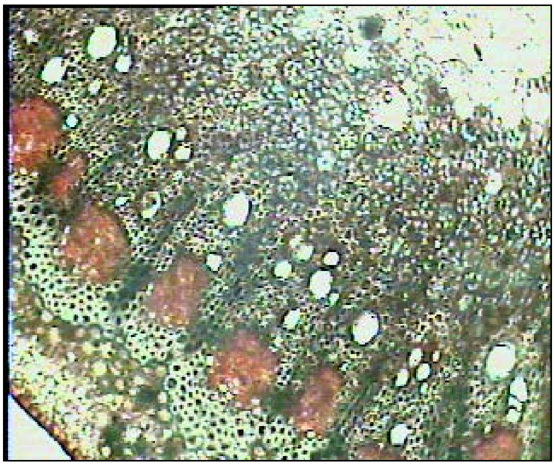

Fig. 12 Cross section in aerial stem of *Polygonum cuspidatum* Sieb. et Zucc. (Reynoutria japonica Houtt.). Plant grown on sterile soil x 10

Fig. 13 Plantlet of dried out *Robinia pseudacacia* L. on the southern side of the Săsar sterile heap

CONCLUSIONS

Considering the things we have presented above, we can say that:

1. ***Robinia pseudacacia*** L., is a very resistant species to the environment factors, including the ones from the sterile heaps. There exist certain alterations on the sterile substratum, however they are not very ample, and above all not significant enough to prevent the species to grow in these areas. The acumulation of pollutants in this plant does, of course, hinder its development, however,even if its size and even its vigour are smaller than in normal substratum, this species qualifies itself very well for becoming an important element in the settling of the ground and regenerating of the soil on the sterile heaps, especially after it has been placated with phyto-soil.

Of course, as one can see, ***Robinia pseudacacia*** L is sensitive to certain conditions. Thus, when planted on the southern side of the hill, in full sun, and on a steep slope, which does not retain water, the plants went dry pretty soon (Fig.13); but when planted on the northern or western side, or on less steep slopes, which retain more water (although the sterile maintains a high degree of moist), this plant grew very well, rooted well-enough not to be blown down by the wind, grew enough foliage, which through putrefaction contributed to the forming of soil,be that rather slowly, indeed.

The species is present on all the sterile heaps in the Baia-Mare Depression (including the one in Bozânta), growing better when planted in areas with more moist and slopes not too steep, and with less exposure to the sun. This plant can reach in such areas a good development (even for normal soils), such as the north-western side of the sterile heap in Sasar, but its use as melliferous plant is very limited, even negligible in droughty conditions, because of the polluting dust which sets down on the leaves (Fig.5), and which sends away the bees or even kills them if the dust sets on the flowers during a droughty period when the plants are blooming. Rain may wash away at least some of the dust, even to the point at which the bees can work. These are the main reasons why all the sterile heaps, at least in the Bia-Mare Depression have ***Robinia pseudacacia*** L., as main arboricolous fixing element. Even though on smaller areas can be seen ***Populus alba*** L., ***Populus nigra*** L., ***Populus tremula*** L., ***Quercus petraea*** (Mattuschka) Liebl., ***Quercus robur*** L., and even ***Castanea sativa*** Miller (*Castanea vesca* Gärtn.), the acacia remains by far the most wide-spread, and the most constant plant on the sterile heaps.

To conclude with, ***Robinia pseudacacia*** L. is a species which, even though on the sterile heaps has a growth which is hindered, as compared to normal, this plant easily adapts itself to settle and regenerate the soil on the sterile heaps, the histological alterations being insignificant for the development of the plants.

2. The ***Centaurea*** genus is well-represented on the sterile heaps (especially on the one in Săsar), but has a low degree of coverage due to the rare distribution of the plants. ***Centaurea cyanus*** L. is a species which, as we have shown above is not greatly affected by the sterile substratum, but then, it can neither be considered essential for the forming of soil directly on the sterile, at least because of its reduced density. However, after a previous placating of the heaps with phyto-soil in a thin layer, ***Centaurea cyanus*** L. might help a lot to cut down on the cost of ecologising, as it is a true settler of soil, at least till the natural development of other species of plants.

3. Out all the species dealt with in this paper, ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.) is the one that is least affected by what kind of sterile substratum it grows on, and this is why it may be considered as a pioneer of vegetal expansion on the sterile heaps, and mainly in the farming fields in the area. Although the species is recognised as being spread at the level of Romania mainly in Maramures (V. Ciocârlan, 2000), it grows on the sands in the north-west of the country (G. Ardelean, C. Karácsonyi, 2005), up to the Ieru valley (G. Ardelean, C. Karácsonyi, 2002), but cannot be found south of the Bihor County (A. Ardelean, 2006). This plant was initially cultivated to be eaten and as ornament, but it spread and grew wild, covering larger and larger expanses of land, without the intervention of man, who tried then to reduce its expansion because it was threatening the cultivated plants. ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.) can be grown either after rooting, or directly as cuttings, even in sterile substratum, but, if only a very thin layer of soil is used, through its rich network of roots, it fixes the soil very well, and the rich foliage consistently contributes to the generating of new soil. Taking into account everything we have stated above, we can conclude that ***Polygonum cuspidatum*** Sieb. et Zucc. (*Reynoutria japonica* Houtt.) is a species that might become one of the biological materials that can be used for settling and ecologising the sterile heaps.

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