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ANATOMICAL COMPARISONS AMONG *HELIANTHUS* SPECIES DIFFERING IN RESISTANCE TO *PHOMOPSIS HELIANTHI*

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Anatomical differences have been observed between the inbred line S-200 of *Helianthus annuus*, and the wild populations PHS-72272 of *H. rigidus* and PHS-2059 of *H. tuberosus*, which are much employed in to-day breeding for resistance work.

Introduction

A high degree of susceptibility to the stem canker of the cultivated sunflowers caused by *Diaporthe helianthi* Munt. Cvet. *et al.* has been found in all the hybrids and in most of the varieties of *Helianthus annuus* L. According to Škorić (8), several wild species, especially *H. rigidus* Dest. and *H. tuberosus* L., are prospective sources of resistance to this and other fungal diseases. In a more intensive inclusion of wild sunflower populations in breeding programmes, the histological studies would be useful. This assertion is supported by the fact that, in the case of the sunflower stem canker, the systemic nature and the invasion route of the causative agent were demonstrated through histological analysis (2, 3). However, although comparative anatomical studies on different sunflower varieties exist in the literature (4, 5), the reports on this subject are not abundant.

This paper presents the results obtained in the investigations carried out through 300 permanent, microtome-cut, counterstained histological plant part sections of the above mentioned *Helianthus* species, which are much used in to-day breeding work.

Material and Methods

This investigation was conducted with three *Helianthus* species comprising the following genotypes: *H. annuus*, inbred line S-200, and the wild sunflower populations of *H. rigidus* PHS-72272, and *H. tuberosus* PHS-2059. The sowing was manual; it was performed at the optimum time for the location (mid-April), on well prepared chernozem soil and plant arrangement of 70 x 30 cm. The cultural practices were adapted to the local conditions. Leaf segments, such as main vein, lateral veins, petiole and stem parts, were used for the histological work. The material was collected at the anthesis stage of the plants and consisted of small fragments of the 5th, 10th, 15th, 20th and apical leaves, corresponding stem parts of *H. annuus* and *H. rigidus* and in the case of *H. tuberosus* the 10th, 20th, 30th, 40th, and 50th leaves. The selected plant material was immediately preserved in a water-formaldehyde-ethyl alcohol solution when collected in the field. Several pieces of each collection were afterwards placed in FAA fixing solution, where they remained until they sank. Samples were embedded in paraffin and sectioned according to the usual techniques. Microtome-cut, 5-15 µm sections were stained with Safranine O and counterstained with Light Green SF Yellowish. Several sections of each selected part were mounted in Canada balsam. Light microscopy and photomicrographs were done using a Reichert microscope Diastar™.

Results

Differences between the species included in this study have been remarked in the stem and leaf cross-sections of the plants investigated.

Stems. The epidermis is constituted of rectangular cells, covered with a thin cuticle, below which a layer of tabular collenchyma cells is found. The cortex consists of several layers of parenchymatous cells; its last layer, the endodermis, separates the cortex from the vascular cylinder. No major distinctions were observed in these parts. In the central cylinder there are numerous collateral bundles. In *H. rigidus* only initial stages of secondary thickening are present. By contrast, in *H. tuberosus* the secondary thickening is fully developed. In *H. tuberosus* the sclerenchyma is also more developed than in *H. rigidus*, and the pith rays are narrower due to the development of the concentric rings of the secondary tissues. The pith of parenchyma is found at the central part of the stem. Both in the cortex and in the pith area, the secretory canals show some differences concerning to their number, which is higher in *H. rigidus*.

Leaves. The symmetrical, deltoid, bifacial lamina is covered by a 1-layered epidermis. The epidermal cells are tabular in shape and covered with a thin cuticle. The external walls of the epidermal cells are thicker than all the others. Trichomes are present on the adaxial and abaxial surfaces of the leaves, but they are not found abundant in any of the investigated species. The palisade tissue, which occurs only on the adaxial side of the lamina, consists of one layer in *H. annuus* and *H. tuberosus*, and of two layers of cells in *H. rigidus*. Palisade cells are not appressed and intercellular spaces are present, especially in *H. annuus*. The spongy tissue occurs just above the lower epidermis; it occupies about half of the thickness of the mesophyll and it is composed of loosely arranged cells irregular in shape and size. The vascular system in the midrib, which is composed of several separate collateral bundles, is surrounded by a parenchymatous and a collenchymatous bundlesheath. In *H. annuus* these bundles are more numerous than in the other two *Helianthus* species, and the midrib relief is especially conspicuous.

D i s c u s s i o n

The plant anatomy has shown its value in ecological, systematic, phylogenetic and other studies (1, 2, 3, 6, 7). Anatomical and histological studies could be worthy also in breeding programmes. Besides, the genes controlling the biochemical aspects of the plant resistance, the anatomy and histology of the host plant could play a role in the pathogenic processes. This especially applies to the structures of the leaves, through which many parasitic organisms gain entrance into the host (for instance *Alternaria* spp., *Phomopsis helianthi*, *Septoria helianthi* Ell. & Kell., and others). Since in interspecific hybridization, wild sunflower species are donors of valuable characteristics. Further histological studies of the parental vegetative organs (i.e. selected wild species and susceptible *H. annuus* genotypes), as well as of the later generations and progenies of successive back-crosses, might show important changes in the structure of the plant organs undergoing the interference of parasites.

Comparative histological studies of plant material obtained in the course of interspecific hybridization and during the pathological processes resulting from experimental infections could sustain or refute this hypothesis.

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