Correlation between macronutrient content and sunflower resistance to *Sclerotinia sclerotiorum* measured by sclerotia infection of stem

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ABSTRACT

Nutrition of plants has a substantial impact on the predisposition of plants to be attacked or affected by pests and diseases. Since availability and nutrient quantity in the host plants is a limiting factor for *Sclerotinia sclerotiorum* infection development in sunflower, the aim of this work was to determine macronutrient content in photosynthetic tissue (leaves) of sunflower plants before and after development of fungus infection, and to find the correlation between macronutrient content and resistance to sclerotium infection. The study was carried out on eight sunflower inbred lines. Macronutrient (N, P, K, Ca and Mg) content was determined in dry plant material of control and sclerotium infected plants. There was a high positive correlation between N content in infected plants and resistance, which points to the important role of this nutrient in sunflower defence from *Sclerotinia* attack. Moderate negative correlation between K and Ca content in control plants showed that the content of these nutrients in healthy plants could be used as an indicator of cultivated sunflower resistance/susceptibility to *Sclerotinia* infection.

Key words: macronutrients – resistance – *Sclerotinia sclerotiorum* – sunflower.

INTRODUCTION

Nutrition of plants has a substantial impact on the predisposition of plants to be attacked or affected by pests and diseases. By affecting the growth pattern, the anatomy and morphology and particularly the chemical composition, the nutrition of plants may contribute either to an increase or decrease of the resistance and/or tolerance to pests and diseases (Krauss, 2001).

White rot, caused by *Sclerotinia sclerotiorum* (Lib.) de Bary and *S. minor* Jager, is the most important sunflower (*Helianthus annuus* L.) disease in the regions with moderate climate (Masirevic and Gulya, 1992; Vasic et al., 2004). The nutrition of *Sclerotinia* during all stages of disease development is probably the most important factor in determining the extent of infection development (Lumsden, 1979). Smirnova (1967) determined that, for normal development of *Sclerotinia*, all nutrients are needed (N, S, K, Mg, and Fe), while Purdy and Grogan (1954) observed that growth and sclerotium formation only occur when the inorganic macronutrients P, K, Mg and S are present in the medium. Availability and nutrient quantity in the host plants is a limiting factor for *Sclerotinia* infection development in sunflower (Acimovic, 1998).

Because of the importance of nutrients for *S. sclerotiorum* infection development, the aim of this work was to determine the nutrient content in photosynthetic tissue (leaves) of sunflower plants before and after development of fungus infection, and to find the correlation between nutrient content and resistance to sclerotium infection.

MATERIALS AND METHODS

Six cms inbred lines (PR-ST-3A, CMS_3-8A, PH-BC_1-40A, Ha-48A, Ha-74A, and CMS_1-50A) and two restorer lines (RUS-RF-OL-168 and RHA-583), all selected in Institute of Field and Vegetable Crops, Novi Sad, Serbia, were used in the study.

The experiment was set in two variants - SC_1 – non-infected control, and SC_2 – stem infection with sclerotium. In each variant, four rows with 12 plants per genotype were sown. Plants in SC_2 plot were inoculated by incorporation of sclerotia into the middle part of the stem, at the stage of bud appearance (E4). Wounds with sclerotia were covered with wet cotton and aluminium foil as described by Vasic et al. (2002). After the infection, plants in both variants were irrigated three times a week, for three hours.

Screening was done at the stage of physiological maturity (M0), and resistance was determined as percentage of healthy plants.

Leaf samples for physiological analyses were taken 28 days after the inoculation in both SC_1 and SC_2 variants. In both cases, three healthy leaves from the upper part of the plant were taken from 10 plants from the inner rows. In SC_2 variant, leaves were taken only from the plants showing symptoms of the white rot. Macronutrient (N, P, K, Ca and Mg) content was determined in dry plant material.

Nitrogen content was determined by the Kjehdal method (Nelson and Sommers, 1973) and phosphorus content spectrophotometrically by the ammonium molybdate-vanadate method (Gericke and Kurmies, 1952). Leaf samples were dry-ashed and dissolved in 25% HCl and analyzed for potassium content by flame photometry and for calcium and magnesium content by atomic absorption spectroscopy.

Resistance of genotypes to sclerotia infection was correlated with macronutrient content in infected and non-infected plants in order to estimate their relationship.

RESULTS AND DISCUSSION

Results obtained showed that there was a difference between tested genotypes in resistance to *Sclerotinia* sclerotium infection. The most resistant genotype was Ha-48A (>90%), and the most susceptible ones were RUS-RF-OL-168 (60.0%) and Ha-74A (60.0%). However, all tested genotypes could be considered tolerant since they all had resistance over 50% (Table 1).

Table 1. Macronutrient content in tested sunflower inbred lines.

Genotype	Variant	Resistance (%)	N	P	K	Ca	Mg
			mg/100g				
$PH\text{-}BC_1\text{-}40A$	Control		3820	246.7	3223	3069	1250.0
	Infection	75.0	3582	313.0	3210	3299	1157.0
Ha-48A	Control		4520	295.7	3478	2873	758.7
	Infection	93.9	4425	254.3	3112	2883	1043.0
$CMS_3\text{-}8A$	Control		3854	288.0	2916	2921	1299.0
	Infection	77.8	3310	293.7	2969	2572	1265.0
PR-ST-3A	Control		3691	307.0	3576	2893	866.7
	Infection	73.7	3834	249.0	2914	2745	1075.0
$CMS_1\text{-}50A$	Control		4065	266.7	3646	2796	856.7
	Infection	60.6	3888	273.7	3516	2623	950.0
RUS-RF-OL-168	Control		4133	284.0	4552	2991	887.7
	Infection	60.0	3385	253.0	3181	3694	1397.0
Ha-74A	Control		3093	323.7	3462	3103	1095.0
	Infection	60.0	3290	309.0	3620	3646	1271.0
RHA-583	Control		3412	290.3	4096	2359	619.3
	Infection	71.9	3643	268.7	3645	2132	580.3

Nutrient content in sunflower leaf tissue depended both on genotype and the presence or the absence of infection (Table 1). Genotype dependence of nutrient content in sunflower tissue was also observed by other authors (Saric et al., 1991; Vasic et al., 2001).

Nitrogen concentration in leaves of tested genotypes ranged from 4472 mg/100g (average for both variants – infection, control) in genotype Ha-48A, which was at the same time the one most resistant to sclerotium infection (93.9%), to 3192 mg/100g in genotype Ha-74A (Table 1). There was a low positive correlation between resistance and N content in leaves of control plants, and a high positive correlation between resistance and N content in leaves of infected plants (Table 2). This is not in accordance with the conclusions presented in the work of HuiLian (2004). This author connected increased susceptibility of field crop plants to pathogen attack with the increased nitrogen compound content in the plant. However, facultative parasites, such as *Sclerotinia*, require weak plants to infest and kill in order to survive (Marchner, 1995). Vigorous plant growth stimulated by ample N would suppress infestation by this group of pathogens. This may explain the differences in expression of plant diseases in relation to the nutrition of the host and N content.

Phosphorus content in leaves of control plants was in positive correlation with resistance (Table 2). This is in accordance with the results of Sindhan and Parashar (1996), who found that leaves of groundnut (*Apios americana* Medic.) cultivars resistant to *Cercospora arachidicola* contained more phosphorus than leaves of susceptible cultivars.

Potassium concentration in leaves of control plants ranged from 4552 mg/100g in genotype RUS-RF-OL-168 to 2916 mg/100g in genotype CMS_3-8A (Table 1). Although the results varied, a negative correlation between resistance and K content in leaves of the control, as well as in leaves of infected plants was found (Table 2). Correlation coefficients were moderate and low, respectively, but results clearly showed that a higher K content in leaves has as a consequently lower resistance to sclerotium infection. This is in contrast with the data obtained by Perrenoud (1990) concerning the relationship of K and plant health. This author reviewed some 2450 references and showed that in 70% of all quoted cases K induced a significant reduction in fungal disease incidences. However, in a more recent publication, Mondal et al. (2001) found a negative correlation between K content and disease incidence in soybean (*Glycine max* (L.) Merrill) and sesame (*Sesamum indicum* L.).

Table 2. Correlation of resistance of inbred lines to sclerotia infection with macronutrient content in infected and non-infected sunflower plants.

Macronutrient	Variant	Resistance (%)	Correlation coefficients
N	Control		0.1137
	Infection	75.0	0.5064
P	Control		0.2140
	Infection	93.9	-0.0554
K	Control		-0.3582
	Infection	77.8	-0.1485
Ca	Control		-0.3161
	Infection	73.7	-0.4121
Mg	Control		-0.1917
	Infection	60.6	-0.4040

There was a moderate negative correlation between Ca content in leaves of control and infected plants and resistance to sclerotium infection, meaning that plants with a higher Ca content were more susceptible to the infection (Table 2). This is in contrast with the results obtained by other authors regarding the role of Ca in sunflower resistance and reaction to *Sclerotinia* infection. Antonova et al. (1984) tested chemical composition of leaves and flowers of sunflower plants resistant and susceptible to *Sclerotinia sclerotiorum* and found that flowers of resistant plants contain more Ca.

Infection led to increase in Mg content in leaves of five genotypes (Table 1). Increases in Mg content in infected plants could be a consequence of chlorophyll degradation and disturbed processes of re-translocation and reutilization of Mg ions, since a pathogen attack causes chlorotic to necrotic changes on all plant parts, which are a consequence of chlorophyll degradation (Singh et al., 1998).

Similarly to the results obtained by other authors, nutrient content in leaves of tested sunflower inbred lines was genotype specific. It has also depended on the development of *Sclerotinia* infection, i.e. genotype resistance. There was a high positive correlation between N content in infected plants and resistance, which points to important role of this nutrient in sunflower defence from *Sclerotinia* attack. Moderate negative correlations between K and Ca content in control plants showed that the content of these nutrients in healthy plants could be used as an indicator of cultivated sunflower resistance/susceptibility to *Sclerotinia* infection. Further studies are in progress in order to find out more on the role of nutrients in sunflower response to *Sclerotinia* attack and to test the value of the results of this study on a larger number of sunflower genotypes.

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