Humic Substances Decrease Water Deficiency Stress of Wheat Seedlings

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1. INTRODUCTION

Drought is a serious problem that affects many regions of the world decreasing the growth rate of crops and limiting the productivity worldwide. Therefore, water availability is an essential factor influencing agriculture. Humic substances (HS) are known to increase tolerance of the plants towards different stresses including water deficiency (1,2). However, the mechanism of this phenomenon is still poorly understood. To gain deeper insight into this process, an investigation of mitigating activity of HS differing in molecular properties have to be conducted. This study was aimed to evaluate the influence of different HA samples on wheat seedlings exposed to water deficiency stress.

2. MATERIALS AND METHODS

Isolation and characterization of humic materials: Leonardite humic acid (LHA) was a commercially available preparation Powhumus (Humintech, Germany) desalted using dialysis before the experiments. Peat humic acids (PHA) were isolated from highland peat using 0.1 M NaOH extraction procedure. The obtained preparations were characterized using elemental analysis and 13C NMR spectroscopy as described in (3). The elemental composition and content of carbon in the structural fragments of humic materials used are summarized in Table 1.

Table 1: Elemental composition and content of carbon in the structural fragments of humic materials used in this study.

HA	Atomic ratios[1]			Content of carbon in the structural fragments, %[2]						
	H/C	O/C	C/N	C=O	COO	Ar-O	CAr	OCO	Alk-O	Alk
LHA	0.87	0.50	53	5.7	19.0	12.8	49.9	1.1	0.9	10.5
PHA	0.93	0.50	57	2.3	12.8	6.7	32.2	4.3	19.9	22.0

[1] – H/C, O/C, and N/C ratios were calculated on ash- and water-free basis.

[2] – Content of carbon in the structural fragments was determined by ^{13}C NMR spectroscopy as the integral intensity (%) of the following spectral regions, ppm: 220-185 (C=O), 185-167 (COO), 167-145 (Ar-O), 145-108 (Ar), 108-90 (OCO), 90-50 (Alk-O) 50-5 (Alk).

Bioassay: Bioassays were conducted using seedling technique. Wheat plants (*Triticum aestivum* L. var. Inna) were used as a biotarget. Distilled water was used as a blank. To invoke water stress in plants, the osmotic solutions of polyethylene glycol of molecular weight 6000 D (PEG 6000) was used. The seeds were placed for germination in the solution of PEG 6000 (10 %) and LHA or PHA preparations were added to distilled water or to the osmotic solution at the concentrations of 10 to 50 mg/L if required. Wheat seeds were germinated in the test solutions in the dark at 25°C for 72 hours. Shoot and root length of seedlings was used as a response.

3. RESULTS AND DISCUSSION

There was no growth stimulating effects of HA detected when seeds were germinated in the distilled water without PEG 6000. Root and shoot length in the presence of HA did not exceed (98±5)% and (100±5)% respectively of blank for both HA at all the concentrations studied.

A use of PEG 6000 osmotic solution allowed to evoke a substantial water stress in the shoots of wheat seedlings: in the presence of PEG 6000 shoot length decreased to (78±3)% of blank value (Fig. 1). For the roots of treated seedlings, the stress was less pronounced: the corresponding value was (96±7)% of blank value and did not differ significantly from the non-treated seedlings.

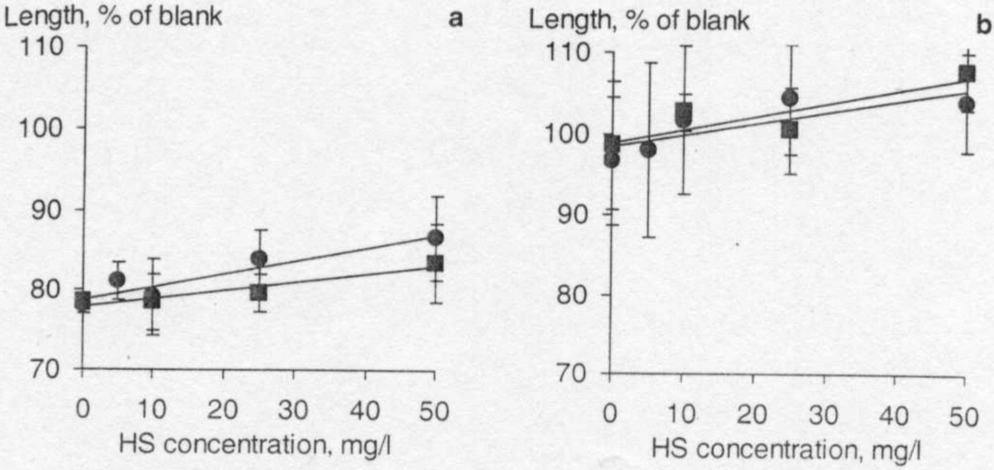

Figure 1: Influence of coal and peat HA on the shoot and root length of wheat seedlings exposed to water deficiency stress using PEG 6000. Bars show confidence intervals.

The substantial stimulating effects were detected when HA were added to the osmotic solutions of PEG 6000. Both shoot and root length of seedlings germinated in the osmotic solutions in the presence of HA increased considerably. In case of coal HA, the presence of humics compensated considerably for water deficiency stress. The shoot length of seedlings exceeded (81±2)% of blank at all examined concentrations reaching its maximum (87±3)% at the highest concentration studied. For peat HA mitigating effect was less appreciable and the analogous values of length were (79±2)% and (83±3)% of blank, respectively. Despite of relatively slight effect of HA application (the length of seedlings shoots under water deficiency stress in the presence of HA did not reach the blank value), the observed trend of increasing shoot length alone with HA concentration allowed concluding on mitigating activity of HA.

The ability to mitigate water deficiency stress was shown by both HA preparations used in this study. A little bit higher mitigating effect was observed for coal HA which were characterized with higher aromatic carbon content and higher H/C atomic ratio (Table 1). This finding indicated that hydrophobicity of HS might play an important role in determining humics mitigating activity against water deficiency stress. However, to establish structure-activity relationship for water-stress mitigating activity of HS, the set of humic fractions should be expanded on the account of HS samples largely differing from the used in this study.

The given results indicated clearly that HA could effectively mitigate the water stress of the plants at early stage of their development.

4. CONCLUSIONS

Mitigating activity of coal and peat HA preparations was observed towards wheat seedlings under water deficiency stress induced by PEG 6000. The given results indicated clearly that HA could effectively mitigate the water stress of the plants at early stage of their development.

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