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PHYSIOLOGICAL ACTIVITY OF WHEAT SEEDLINGS UNDER CHEMICAL STRESS INDUCED BY VOLATILE ETHYL BENZENE

The effect of the ethyl benzene vapours in air at the concentration ranging from 81 to 6570 mg·m^{-3} on the physiological response and anatomical changes of winter wheat seedlings, cv. "Tonacja" was examined. This effect, intensifying CO_2 assimilation and increasing photosynthetic water-use efficiency, was observed at the lowest concentration. At the concentrations of 730 to 6570 mg·m^{-3}, ethyl benzene vapours suppressed both germination and development of plants as well as decreased physiological activity of the seedlings. They frequently induced chlorotic and necrotic areas on leaf surface, as well as dehydration of cells and changes in their shape.

1. INTRODUCTION

Along with the development of civilisation, noxious emissions of harmful substances into natural environment occur more frequently. One of environmentally nuisant compounds is ethyl benzene, which contributes to the formation of ground-level ozone in air. Its permissible concentration in atmospheric air is 0.5 mg·m^{-3} in 30 minutes, 0.2 mg· m^{-3} in 24 hours, and 0.038 mg·m^{-3} per year [1]. In the vicinity of emitters, under unfavourable conditions for air spreading (inversion) and with low discharge stacks, ethyl benzene concentration may be, however, considerably higher. This results from its relatively high permissible concentrations in the workplace, i.e. 100–350 (STEL) mg·m^{-3} [2], causing in turn that ethyl benzene concentrations in the ventilation air can be close

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to the higher one, or even exceed it. Furthermore, ethyl benzene, with vapour density higher than that of air, easily concentrates in land depressions, thus actively contributing to the increase of environmental pollution [3]. Living organisms, including the plants from ecosystems situated close to ethyl benzene emission sources, can be therefore exposed to its toxic effect.

The study aimed at examining the physiological response of seedlings to the stress induced by ethyl benzene and their morphological and anatomical changes.

2. MATERIAL AND METHODS

The experiment was carried out in 25 dm^3 phytotrone chambers with controlled atmosphere and photoperiod (80% relative humidity, 20 °C, 200–300 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PhAR intensity). The ethyl benzene concentrations in air applied, i.e. 81, 243, 730, 2190 and 6570 $mg \cdot m^{-3}$, were obtained by inserting into each chamber a bottle with ethyl benzene–silicone oil solution of a required concentration (its vapour pressure equilibrium at 20 °C was 930 Pa [4]). The ethyl benzene concentration assumed to stabilise after about 6 hours, which was found by chromatographic analyses. Concentration deviations from the expected values ranged from +10 to –20%. Next, the pots with winter wheat seeds of cv. "Tonacja" (10 seeds per each pot) sowed into a soil of light loamy clay granulometric composition and pH_{H_2O} 7.0 were placed in the chambers. The experiment was done in three series with four replications. The seeds and the seedlings developing from them were exposed to regular effect of the air containing ethyl benzene at the concentrations given above for a time period of about three weeks. On the 18th day the wheat seedlings were subjected to the measurements listed below.

Plant physiological response descriptors were determined by measuring gaseous exchange parameters, such as CO_2 assimilation intensity (A, $\mu mol \cdot m^{-2} \cdot s^{-1}$) and transpiration ($E$, $mmol \cdot m^{-2} \cdot s^{-1}$), with a portable infrared gas analyser LCA-4 (ADC Bioscentific Ltd. Hoddesdon, Great Britain). The measuring camera covered a 0.5-cm^2 leaf fragment. Also the stomatal index (SI, i.e. the number of stomata per 1 mm^2) was determined under a CX21 SD/SF series microscope (Olympus Optical Co. Ltd, Tokio, Japan). Photographs were taken with an Olympus digital camera C5050Z (Olympus Optical Co. Ltd, Tokio, Japan). Biometric parameters, i.e. plant height, were determined with a measuring rule (in cm). Water-use efficiency (WUE, $\mu mol\ CO_2 \cdot mmol^{-1}\ H_2O$) was calculated as CO_2 assimilation to transpiration ratio.

3. RESULTS AND DISCUSSION

Based on the findings, with a simultaneous scarcity of literature references in this field, it is difficult to precisely explain changes in the physiological activity of wheat seedlings and the stomata aperture induced by ethyl benzene vapours. For several reasons, including purely ecological ones, such efforts are required; this is why many authors [5]–[8] have already pointed to the usefulness of carrying out studies on the phytotoxic effect of volatile organic compounds, including ethyl benzene.

The study carried out proves that ethyl benzene at the concentrations applied clearly affects the germination time of wheat seeds. Wheat seeds used as the control and those at the ethyl benzene concentration in air of 81 mg·m^{-3} germinated after 7 days from the sowing time, while those at the concentrations from 730 to 2190 mg·m^{-3} – after 10 days and of 6570 mg·m^{-3} – after 12 days. This indicates that ethyl benzene at higher concentrations has an inhibiting effect on wheat germination. Furthermore, the wheat seedlings growing in the atmosphere with ethyl benzene concentration ranging from 2190 to 6570 mg·m^{-3} were characterised by clearly slower elongation growth compared to the control and showed morphological changes, in particular the reduction of their leaf surfaces. At ethyl benzene concentration of 6570 mg·m^{-3}, wheat leaf blades were clearly thicker, more rigid and saturated with a larger amount of sclerenchyma with a thick layer of waxy cuticle. These are the characteristics of xeromorphic plants which allow them to reduce transpiration.

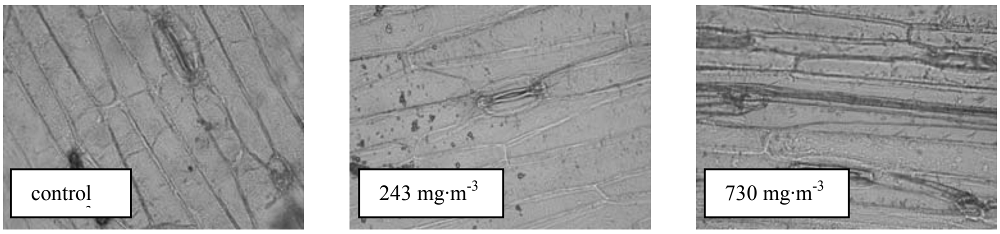


Fig. 1. Microscopic picture of stomatal apparatuses of wheat growing in ethyl benzene vapours-polluted atmosphere

Under the conditions of high ethyl benzene concentration, i.e. from 730 to 6570 mg·m^{-3}, also a progressing plasmolysis of epidermis cells was observed, as well as dehydration of stomatal cells and deformation of stomata (figure 1) In the opinion of MCLACHLAN [9], volatile organic compounds are absorbed by stomatal apparatuses, this being manifested itself, among others, as changes of stomatal index. This phenomenon was also confirmed by STOLARSKA et al. [10], who examined the effect of volatile styrene on plant growth. It was found in the present study that ethyl benzene vapours at higher concentrations (above 2190 mg·m^{-3}) contributed to the dehydration of stomatal cells and to changes in their shape, but they did not have a significant effect on the number of stomatal appartuses (table 1). Together with the increase of ethyl benzene concentration, progressing chloroses and necroses were observed (figure 2). Similar symptoms of toxic effect of benzene at a concentration of 10000 mg·m^{-3}on crop plants were observed by MILLER [11].

Table

Effect of different ethyl benzene concentrations on the number of stomatal apparatuses per 1 mm^{-2} of epidermis

Concentration of ethyl benzene (mg·m^{-3})	Number of stomatal apparatuses
0.0 (control)	100
81	110
243	90
730	100
2190	98
6570	124
LSD $_{0.05}$	40

LSD $_{0.05}$ – insignificant differences at $\alpha = 0.05$.

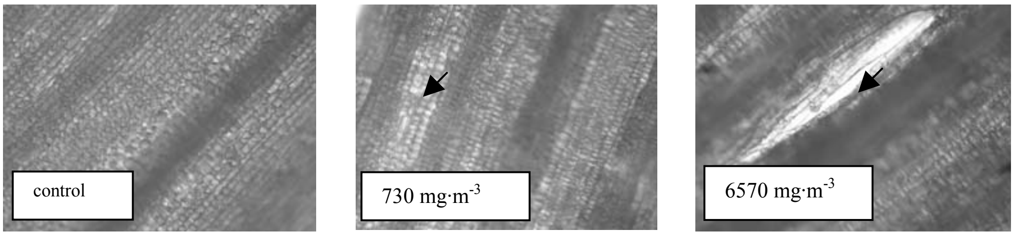
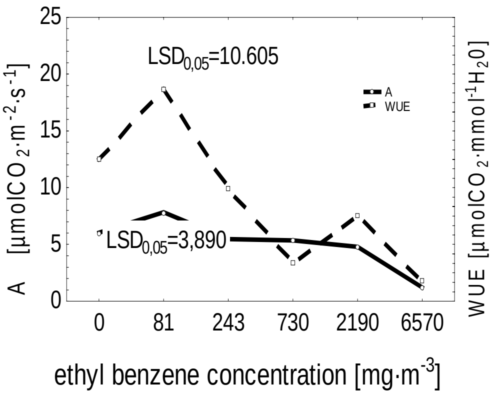


Fig. 2. Microscopic picture of epidermis with marked necroses and chloroses induced by ethyl benzene vapours

a)



b)

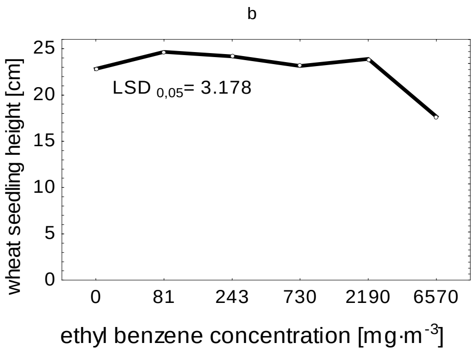


Fig. 3. Effect of different ethyl benzene concentrations in atmospheric air on CO_2 assimilation intensity (A) and photosynthetic water-use efficiency (WUE) (a) and wheat seedling height (b). LSD $_{0.05}$ – insignificant differences at $\alpha = 0.05$

In the present study, a strong physiological response of wheat seedlings to ethyl benzene vapours was also shown (figure 3a). At its lowest concentration applied, the seedlings intensively assimilated carbon dioxide and effectively used water in this process. The values of these parameters were significantly higher from those in the seedlings exposed to ethyl benzene concentrations ranging from 2190 and 6570 $mg \cdot m^{-3}$. This trend in the changes in the efficiency of photosynthetic water use was reflected by the changes in the height of seedlings (figure 3b).

4. CONCLUSIONS

1. Ethyl benzene vapours significantly affected the physiological activity of wheat seedlings, cv. "Tonacja"; they stimulated the photosynthetic activity of the seedlings at the lowest concentration (81 $mg \cdot m^{-3}$), but inhibited it at the concentrations above 730 $mg \cdot m^{-3}$.

2. Ethyl benzene at its concentration in air of 2190 and 6570 $mg \cdot m^{-3}$ induced a clear suppression in the germination of wheat seeds and contributed to the formation of chloroses and necroses on the leaves of wheat seedlings.

3. Wheat seedlings growing in the atmosphere containing ethyl benzene at a concentration of 6570 $mg \cdot m^{-3}$ were characterised by the thick layers of both waxy cuticle and sclerenchyma, which are typical of xeromorphic plants.

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AKTYWNOŚĆ FIZJOLOGICZNA SIEWEK PSZENICY W WARUNKACH STRESU CHEMICZNEGO WYWOŁANEGO LOTNYM ETYLOBENZENEM

Badano wpływ par etylobenzenu w powietrzu w zakresie stężeń 81–6570 mg·m^{-3} na reakcję fizjologiczną oraz zmiany anatomiczne siewek pszenicy ozimej odmiany ‘Tonacja’. Przy najniższym stężeniu obserwowano działanie intensyfikujące asymilację CO_2 oraz zwiększające efektywność wykorzystania wody w fotosyntezie. Stężenia par etylobenzenu od 730 do 6570 mg·m^{-3} ograniczały kiełkowanie i rozwój roślin oraz obniżały aktywność fotosyntetyczą siewek. Powodowały często chlorotyczne oraz nekrotyczne plamy na powierzchni liści, odwodnienie komórek i zmiany ich kształtu.