PHOTOSYNTHETIC PARAMETERS AND ABSCISIC ACID LEVELS OF PEA PLANTS INFLUENCED BY ORGANIC POLLUTANTS

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Abstract: Growth and development of plants is affected by many biotic and abiotic factors. Current problem is the presence of environmental pollutants. Contamination of soil and air can cause stress reaction and some fatal changes in metabolism of affected plants. The important monitored pollutants include polycyclic aromatic hydrocarbons and active substances of some herbicides. Pea plants (*Pisum sativum* L.) were cultivated in Richter nutrient solution (control) and two variants, nutrient solution with 5µM fluoranthene (FLT) or with 5µM flurochloridone (FLU). Changes of photosynthetic apparatus, especially in damaged leaves of treated plants were evaluated. The main differences between variants have been observed in the levels of photosynthetic pigments. Both substances (FLT and FLU) decreased the content of chlorophylls and carotenoids. But FLT treatment caused only slight decrease of the quantum yield of electron transport of PS II compared to control. Significant differences in the level of abscisic acid (ABA) in leaves between the variants and between the damaged leaves and green leaves were observed. The changes of pigment content and damage of photosynthetic apparatus were visible on the plants, especially on the colour of the leaves.

Key Words: fluoranthene, flurochloridone, photosynthetic pigments, Φ PS II

INTRODUCTION

Growth and development of plants is affected by many biotic and abiotic factors – e.g. nutrients, humidity, pathogens and presence of environmental pollutants. Plants are not able to move from place to place and therefore contamination of environment can cause stress reaction and some fatal changes in metabolism of affected plants. The most important group of pollutants are polycyclic aromatic hydrocarbons (PAHs), which plants absorb mainly from air, but also from soil or water. These substances are accumulated in lipid rich parts of plants (Wild, Jones 1991). Fluoranthene (FLT) is a polycyclic aromatic hydrocarbon, which belongs to industrial pollutants. The origin of these pollutants is from oil processing and chemical industry. FLT is also a component of exhaust fumes. Its negative influence on plants, especially their photosynthetic apparatus is known (Kummerová et al. 2010).

The residues of active substances of some herbicides belong to substances that can have also a negative effect on plant life. Because nowadays the use of herbicides rises, it is necessary to know their effect on plants. Flurochloridone (FLU) is the active substance of some pre-emergence herbicides. FLU has an inhibitory effect on the synthesis of carotenoids, which protect chlorophylls against oxidative stress.

The aim of this study was to follow the changes of photosynthetic apparatus of the FLT and FLU treated plants.

MATERIAL AND METHODS

Cultivation of plants

Pea plants (*Pisum sativum* L. var. Oskar) were used as a model plants. They were cultivated in Richter nutrient solution (Richter 1926) – control and two variants – nutrient solution with 5µM FLT or with 5µM FLU. The nutrient solution was exchanged in the time of collecting samples. Plants were placed in a climabox, under controlled conditions photoperiod 18/6 h (day/night) and temperature 22/16° C.

Sampling and measurement of photosynthetic parameters

The samples were collected in five or six growth phases, what is indicated in graphs. Minimal number of evaluated samples for every parameter was three plants for every variant. Level of photosynthetic pigments in damaged leaves was analyzed from acetone extract by spectrophotometeric analysis, at λ = 663 nm, 645 nm and 440 nm. The levels of the pigments were calculated by equations: chlorophyll $a = 12.7{*}A_{663} - 2.69{*}A_{645}$, chlorophyll $b = 22.9{*}A_{645} - 4.68{*}A_{663}$, carotenoids = $4.968{*}A_{440} - 0.268{*}$(content of chlorophyll *a* + content of chlorophyll *b*), where A main absorbance for the said λ. TLC separation of pigments for illustration of differences between experimental variants was done. Mobile phase petrol : acetone : diethyl ether (5:2:1). Quantum yield of electron transport of PS II was measured by Fluor Pen FP 100 (Photo System Instruments).

Analysis of ABA content

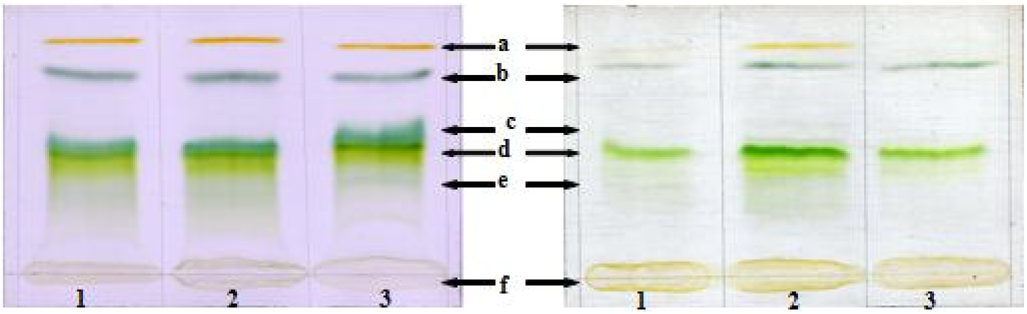
The content of abscisic acid (ABA, marker of stress) in green and damaged leaves, was determined by radioimmunoassay (RIA), which uses the specific affinity of monoclonal antibody MAC 252 (Quarrie et al. 1988) to the ABA molecule. The principle of the method is competitive reaction between native ABA from the sample and radioactively marked ^{3}H-ABA. The ^{3}H-activity was measured by spectrophotometer PACKARD 2900 TR. Results were analyzed by special program Securia PACKARD.

RESULTS AND DISCUSSION

Photosynthetic pigments and quantum yield of PS II

The content of photosynthetic pigments was analyzed in leaves with visible symptoms of damage. TLC separation (Figure 1) illustrates decrease of all of pigments during cultivation of plants for all variants. The same picture demonstrates differences between variants, especially in level of β-carotene and chlorophyll *b*.

Figure 1 TLC separation of pigments, left 1 week old plants (2 – 3 leaves), right 1 month old plants (maturation of pods)



Legend:1 – FLT, 2 – control, 3 – FLU, a – β-carotene, b – pheophytin, c – chlorophyll a, d – chlorophyll b, e – xathophylls, f – start

During cultivation the level of chlorophylls in leaves in all variants including control plants decreased. But the influence of both xenobiotics (FLT, FLU) decreased the level of chlorophylls to the half of the value of control plants (Figure 2, 3). The main differences compared with control were noticeable since the 8 leaves growth phase. The content of carotenoids (Figure 4) in control plants was identical during the cultivation period. FLT caused decrease of the level of carotenoids in leaves similarly as FLU, the inhibitor of carotenoid synthesis. The decrease of the levels of photosynthetic pigments in leaves by FLT and FLU treatment is similar with the results of recent studies (Kummerová et al. 2010, Oguntimehin et al. 2010).

Figure 2 Changes of chlorophyll a content during the cultivation

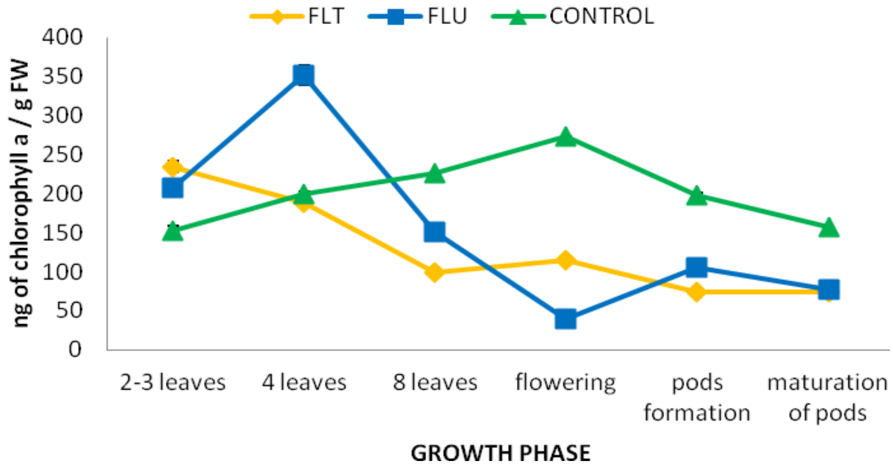


Figure 3 Changes of chlorophyll b content during the cultivation

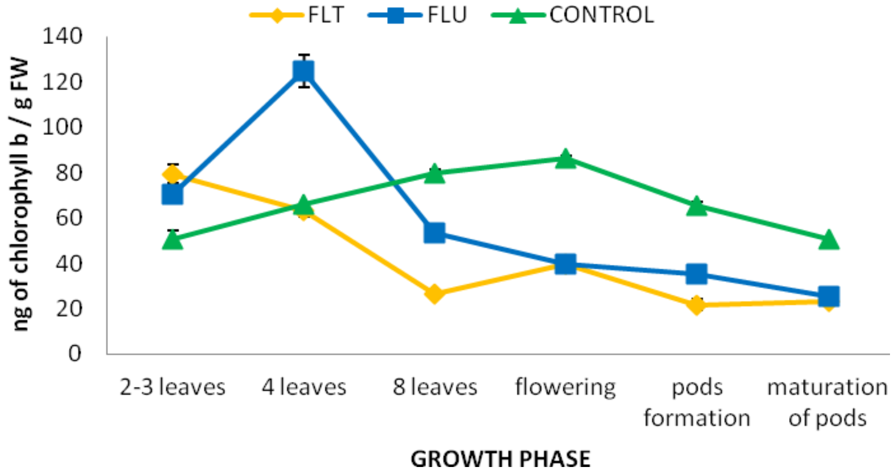
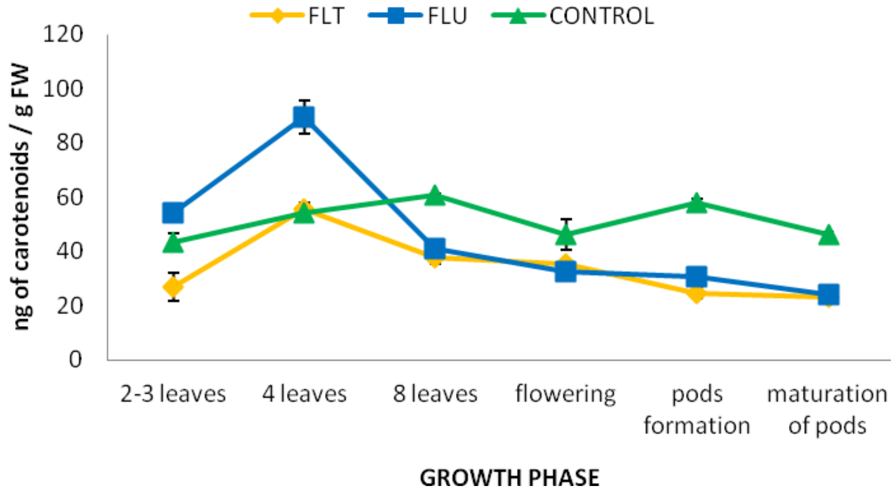
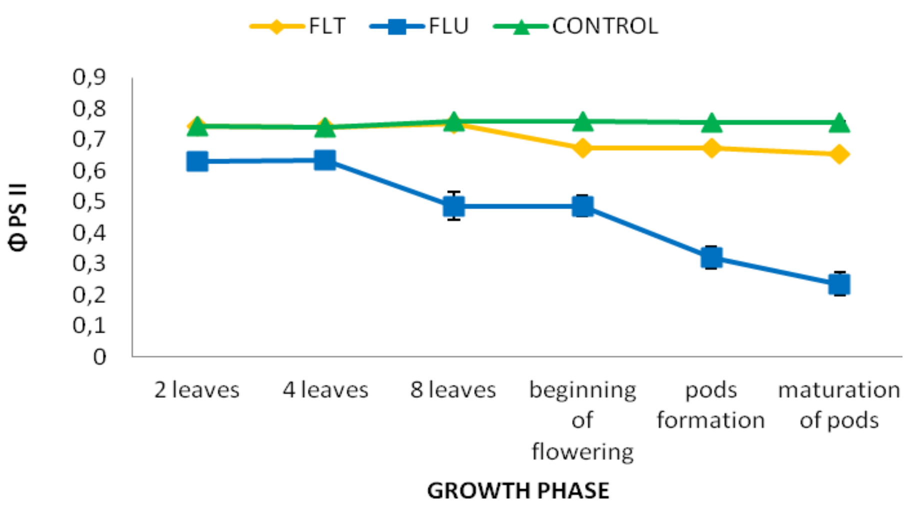


Figure 4 Changes of content of carotenoids during the cultivation



The quantum yield of electron transport of PS II (Φ PS II) was measured on green leaves without visible symptoms of damage. Nevertheless there were differences in values between the variants. Control plants showed the same values throughout the cultivation. FLT influenced a decrease of the values, but only slightly compare with FLU effect (Figure 5). The decrease of Φ PS II relate with the loss of the pigments but obviously with the change of chloroplasts ultrastructure too (Popova 1996, Klíčová et al. 2002).

Figure 5 Dynamics of Φ PS II



Level of ABA

Relating to level of carotenoids, important as oxidative stress protection for chlorophylls and as precursors for ABA synthesis, level of abscisic acid (ABA) in leaves was determined too. Level of abscisic acid was analysed in green leaves and in leaves with symptoms of damage caused by xenobiotics treatment (Figure 6, 7). Level of ABA in control plants rises during cultivation. Treatment by FLU significantly reduced ABA in green leaves but in damaged leaves the results were similar to control. Different situation was observed after FLT treatment. In green leaves level of ABA was lower but in damaged leaves increased shortly in the phase of flowering and then decreased to the control value. There is probably some correlation with the decreased level of ABA precursors – carotenoids (Moore, Smith 1984), and the decreased level of ABA in green leaves. But on the other hand the higher content of ABA in damaged leaves could be caused by translocation of this phytohormone to these parts (Gapin et al. 2000).

Figure 6 Changes of ABA level in green leaves

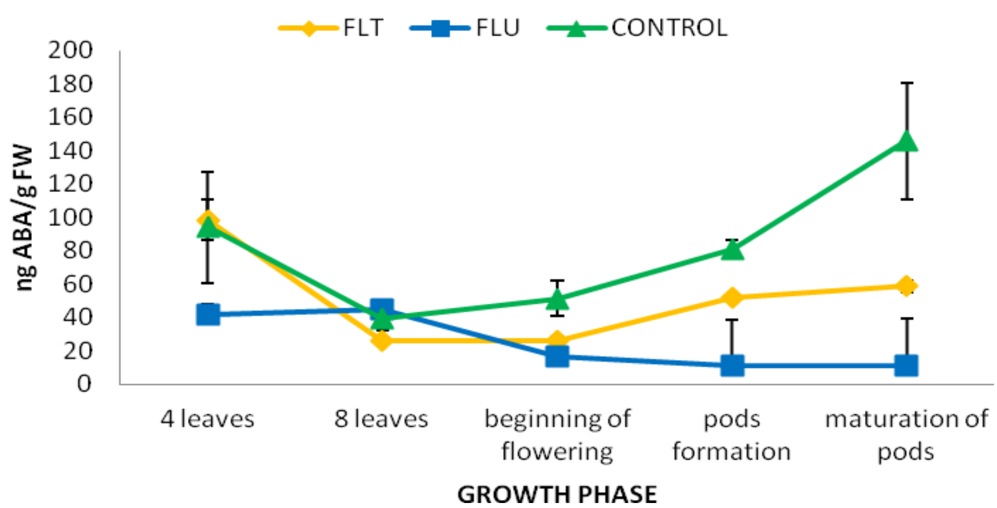
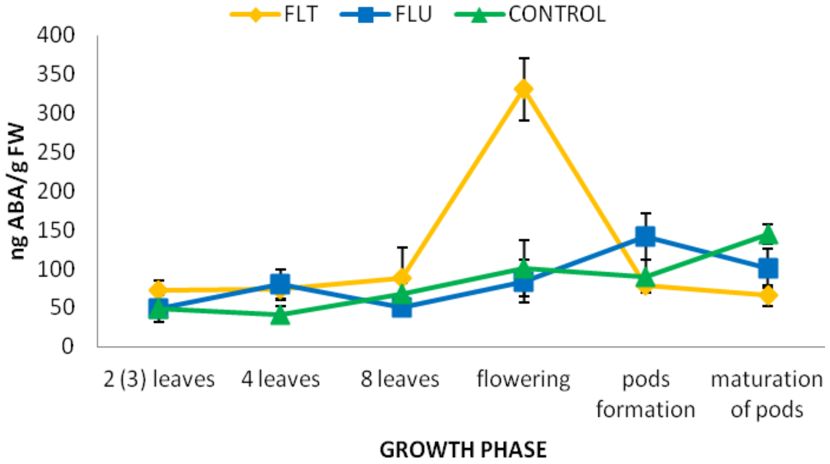


Figure 7 Changes of ABA level in damaged leaves



Visual symptoms

The damage of the photosynthetic apparatus caused by FLT and FLU treatment was visible on the changes of the colour of leaves. Chlorosis caused by FLT treatment was observed on the entire leaf blade of young leaves. But changes of colour caused by FLU were visible mainly on stems and leaf veins (Figure 8). In the case of FLU symptoms occurred first on old leaves. These changes visible on the aboveground parts of plants are the evidence of root uptake and xylem transport of both used substances (Klíčová et al. 2002).

Figure 8 Colour changes of leaves of treated plants (from left FLT, control, FLU)

CONCLUSION

FLT can negatively affect the plant photosynthetic apparatus (Kummerová, Váňová 2007). Its influence decreases the level of photosynthetic pigments and content of ABA in green leaves. The typical symptoms of damage by FLT are chlorosis on leaf blades, especially on young leaves.

FLU is a carotenoid synthesis inhibitor (Klíčová et al. 2002). Carotenoids are important as oxidative stress protection for chlorophylls and as precursors for ABA synthesis (Yamazaki et al. 1999). Therefore FLU has detrimental effect on photosynthesis and level of ABA. Loss of pigments causes the formation of “albinotic” plants.

The results of changes of plant colour show that both pollutants are taken up from roots to the shoot by xylem transport.

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