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Tolerance of wheat and lettuce plants grown on human mineralized waste to high temperature stress

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Abstract

The main objective of a life support system for space missions is to supply a crew with food, water and oxygen, and to eliminate their wastes. The ultimate goal is to achieve the highest degree of closure of the system using controlled processes offering a high level of reliability and flexibility. Enhancement of closure of a biological life support system (BLSS) that includes plants relies on increased regeneration of plant waste, and utilization of solid and liquid human wastes. Clearly, the robustness of a BLSS subjected to stress will be substantially determined by the robustness of the plant components of the phototrophic unit. The aim of the present work was to estimate the heat resistance of two plants (wheat and lettuce) grown on human wastes. Human exometabolites mineralized by hydrogen peroxide in an electromagnetic field were used to make a nutrient solution for the plants. We looked for a possible increase in the heat tolerance of the wheat plants using changes in photosynthetically active radiation (PAR) intensity during heat stress. At age 15 days, plants were subjected to a rise in air temperature (from 23 ± 1 °C to 44 ± 1 °C) under different PAR intensities for 4 h. The status of the photosynthetic apparatus of the plants was assessed by external CO_2 gas exchange and fluorescence measurements. The increased irradiance of the plants during the high temperature period demonstrated its protective action for both the photosynthetic apparatus of the leaves and subsequent plant growth and development. The productivity of the plants subjected to temperature changes at 250 W m^{-2} of PAR did not differ from that of controls, whereas the productivity of the plants subjected to the same heat stress but in darkness was halved.

Keywords: BLSS; Human waste recycling; Wheat; Lettuce; Heat resistance; Air temperature step

1. Introduction

It is established that any biological life support system (BLSS) must integrate the biological and physicochemical principles of environment regeneration (Meleshko et al., 1995; Gitelson et al., 2003; Gros et al., 2003). Importantly, it must include human waste in mass exchange to significantly reduce its storage and disposal. A phototrophic unit represented by crops can achieve regeneration of water, air and the plant-based part of a human diet (Tikhomirov et al., 2007; Tikhomirova et al., 2008). BLSS tolerance to stress will substantially depend on the resistance of the higher plants present in the phototrophic unit. Air temperature is one environmental factor that significantly influences processes of plant growth and development. Breakdown of a thermoregulation system could result in an elevation of air temperature beyond the tolerance range of the plants.

Plants response to an air temperature increase depends on temperature value and stress duration, on conditions of cultivation before stress and also on the development

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stage: reproductive organs are the most sensitive to heat stress (Wahid et al., 2007). Most works are devoted to the problem of the effect of elevated temperature on heat tolerance of plants at molecular, cellular and entire plant levels (Kepova et al., 2005; Mishra and Singhal, 1992; Wahid et al., 2007). But thermotolerance of one plant cannot be directly extrapolated to the plant community, i.e. to the crop level. Unfortunately, there is little information about heat tolerance of plants at a crop level in the literature. For example, it was observed that, from cellular to entire plant and crop levels, there is an increase in the time before damage appears. For isolated chloroplasts this time is 5 min (Yamane et al., 1998; Bukhov et al., 1999). For a leaf this time varies from 5 min to one hour (Belikov and Melekhov, 1975). At the crop level this time increases to 5 h (Sarieva et al., 2010) and even 7 h (Tikhomirov and Ushakova, 2001; Ushakova and Tikhomirov, 2002).

We had previously assessed the heat tolerance of some plants grown on standard mineral solutions at optimal temperatures (Ushakova and Tikhomirov, 2002; Zavorueva and Ushakova, 2004). A possible increase in the heat tolerance of plants at elevated intensity of photosynthetic active radiation (PAR) was shown during stress. The work reported here set out to assess the heat tolerance of plants grown on human mineralized exometabolites and determine whether heat tolerance could be improved by increasing PAR intensity during heat stress.

2. Materials and methods

Spring wheat *Triticum aestivum* L., line 232 selection by G.M. Lisovsky (Lisovsky and Shilenko, 1979), one of the main foodstuffs of the European diet type, and lettuce *Lactuca sativa* L. var. Vitaminny as a representative of leaf lettuce cultures were used.

The plants were grown under 24 h illumination (no photoperiod) at a PAR intensity of 150 W m^{-2} (690 μmol m^{-2} s^{-1}), a value close to the maximal photosynthesis efficiency of this culture (Tikhomirov and Sidko, 1983). DRI-2000 lamps (high-pressure mercury lamps with added metal iodides) served as the PAR light source. Planting density was 1000 m^{-2} for the wheat and 180 m^{-2} for the lettuce. Air temperature was 23 ± 1 °C, humidity was 70%. The plants were grown hydroponically on expanded clay aggregate; CO_2 concentration was 350–400 ppm, humidity 60–70%. After the heat stress treatment, the plants were grown to harvest maturity (wheat to 60 days, lettuce to 30 days).

Tap water to which was added human liquid and solid wastes mineralized by the "wet incineration" method was used as a nutrient solution (Kudenko et al., 2000; Zolotukhin et al., 2005). The process of growing the plants has been described elsewhere (Ushakova et al., 2009). The human mineralized exometabolites included reduced nitrogen (amide and ammonia). At age 15 days the wheat plants were subjected to an air temperature of 44 ± 1 °C for 4 h. PAR intensity during this period was set at 0 (light out), 150 W m^{-2} or 250 W m^{-2} (690 μmol m^{-2} s^{-1} or 1150 μmol m^{-2} s^{-1}).

Lettuce was grown in the same conditions. At age 21 days the plants were subjected to an air temperature of 44°C under a PAR intensity of 150 W m^{-2} for 4 h. After termination of the stress the plants continued to grow in the initial conditions.

Plants were enclosed in a chamber for photosynthesis measurements. The 350 L volume chamber was made of stainless steel, with an organic glass ceiling, which was cooled with running tap water. The lamps were attached to the ceiling. We used the same lamps as those used to grow plants. Relative humidity was 55–60%. If CO_2 concentration decreased during the experiment, it was adjusted to 320–450 ppm. Net photosynthesis was measured at CO_2 concentration 350 ppm. If CO_2 concentration increased, we did not interfere in the process.

Reaction of the plants to heat stress was estimated on the basis of rate changes in external CO_2 gas exchange of wheat and lettuce canopy and those of intact wheat leaves. CO_2 gas exchange of intact wheat leaves was measured in a leaf chamber using an Li-7000 differential gas analyzer (LICOR, USA). The canopy CO_2 gas exchange was estimated from changes in CO_2 concentration in a closed volume using an Li-820 gas analyzer (LICOR, USA). Respiration was measured based on the rate of change in CO_2 concentration in a closed space for the first 20 min after the light was turned out (a conventional technique) (Bykov, 1962; Tikhomirova et al., 2009).

The state of the photosynthetic apparatus of the leaves (PSA) was estimated from the intensity of lipid peroxidation (LPO) and the parameters of pulse-modulated chlorophyll fluorescence in PS 2. (Krause and Weiss, 1991; Nesterenko et al., 2006). Measurements were made during cultivation along the middle third of undetached leaf blades of well-illuminated upper tiers of wheat, and in lettuce by means of a PAM 2100 portable fluorometer (Walz, Germany). Maximal quantum yield of charge separation in photosystem 2 (PS2) was estimated by the ratio F_v/F_m of variable fluorescence (F_v) to maximal fluorescence (F_m). From the yield value we estimated the effective quantum yield of PS2 during constant illumination to give an approximate characteristic of total quantum photosynthesis yield. From the q_P value (photochemical quenching of chlorophyll fluorescence in PS2) we estimated the proportion of energy–power employment of electron-induced chlorophyll in photochemical reactions of chloroplasts with constant light, and from the q_N value (nonphotochemical quenching of chlorophyll fluorescence) the proportion of absorbed energy dissipated as heat (Roháček, 2002; Roháček and Bartak, 1999; Lazàr, 1999).

Photosynthetic pigments were extracted from fresh plant biomass (150–200 mg) by boiling in 100% acetone. The green and yellow pigment contents were determined using a UV 1700 spectrophotometer (Shimadzu, Japan) in acetone extraction solution at 662 nm, 644 nm (chlorophyll) and 470 nm (carotenoids) (Gavrilenko and Zhigalova, 2003).

Accumulation of malonic dialdehyde (MDA) serves as an indicator of lipid peroxidation associated with oxidation of mainly unsaturated fatty acids of membranes. So we can use MDA content to judge integrity of membrane cells. The intensity of lipid peroxidation (LPO) was determined by the reaction of thiobarbituric acid (TBA) with malonic dialdehyde (Lukatkin, 2002). Optical density was recorded using a UV 1700 spectrophotometer (Shimadzu, Japan) at 532 nm against medium with reagent. The MDA content was expressed in nmol/g of dry weight.

Experiments were repeated twice. In each experiment there were 16 lettuce plants and 60 wheat plants. Standard errors are shown on the graphs. Significant difference between two comparative groups was estimated by the Student's test. The critical value of *t*-test was found by the number of degrees of freedom at 95% significance level ($\alpha = 0.95$). Differences of comparative groups were considered significant at probability of error $P < 0.05$.

3. Results and discussion

3.1. Effect of light and temperature stress on CO_2 gas exchange in wheat and lettuce

Elevation of air temperature in darkness resulted in a considerable increase in plant respiration intensity (*R*) (Fig. 1). Before the temperature elevation *R* was 17.6 μmol·CO_2 m^{-2} s^{-1} of crops. By 40 min. after the start of the heat stress *R* had risen to 25.4 μmol·CO_2 m^{-2} s^{-1} of crops. During the following 1.5 h *R* further increased to 34.2 μmol·CO_2 m^{-2} s^{-1} of crops, after which it reverted to values close to those measured before the stress.

The air temperature elevation at PAR 150 W m^{-2} significantly accelerated oxidizing processes: in light, CO_2 release at the intensity of 16.4 μmol·CO_2 m^{-2} s^{-1} of crops was observed 40 min from the beginning of the stress (Fig. 1). At the end of the stress, the difference between CO_2 uptake and release rates narrowed, with a reduction in CO_2 release intensity of up to 3.9 μmol·CO_2 m^{-2} s^{-1} of crops. Increasing the PAR intensity to 250 W m^{-2} before applying the stress caused a more than 2.5-fold elevation of net photosynthesis intensity (P_{net}) at the air temperature of 23 °C (from 10 to 26.9 μmol·CO_2 m^{-2} s^{-1} of crops). Thus under heat stress during the first 2.5 h CO_2 uptake was observed, the intensity of which gradually fell. At 2.5 h the intensity of oxidizing and photosynthetic processes equalized. At the end of exposure, the intensity of CO_2 release even slightly exceeded its uptake (Fig. 1). We note that a strong increase in PAR intensity when the air temperature was raised can increase but can also decrease the tolerance of the plants to the heat stress.

In the same experiments, while the PAR intensity effect on the heat resistance of intact leaves was estimated, we also studied the kinetic dependence of CO_2 gas exchange (Fig. 2). The strongest inhibition of the net photosynthesis in the leaves was observed during heat stress at 44 °C for 4 h in darkness (Fig. 2). The leaves displayed the highest tolerance to the stress under PAR 150 W.m^{-2}. Under 250 W m^{-2}, in the interval between 1.5 h and 2.5 h of the stress, CO_2 release was even observed. However, at the end of the stress time, at both 150 W m^{-2} and 250 W m^{-2}, full inhibition of net CO_2 uptake by leaves was observed.

Reaction differences in wheat canopy and separate leaves were approached by determining PAR intensity for the canopy using a light-receiving sensor placed perpendicular to incident light. Here, essentially depending on the canopy architecture, the wheat leaves received 74% of the PAR value measured, i.e. 110 or 185 W m^{-2}. For estimation of the leaf heat resistance, leaves were set perpendicular to the incident light. Hence they were more intensely illuminated than real crops.

A reason for lower wheat heat tolerance can be that the plants only respire in darkness and perhaps it is more stressful due to more depletion of energy reserves of the plant in darkness than in the light. Another reason is the decrease of transpiration intensity in darkness that results in leaves overheating. Probably leaves overheating results in a decrease of photosynthetic processes during heat stress at the intensity of 250 W m^{-2}.

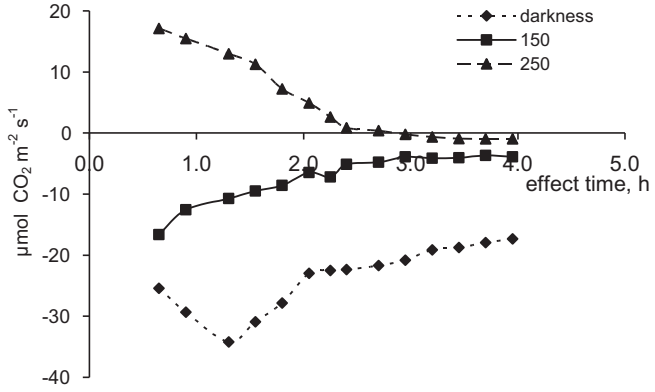

Fig. 1. Kinetics of CO_2 exchange of 15-day wheat canopy during the effect of air temperature of 44 °C under different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}).

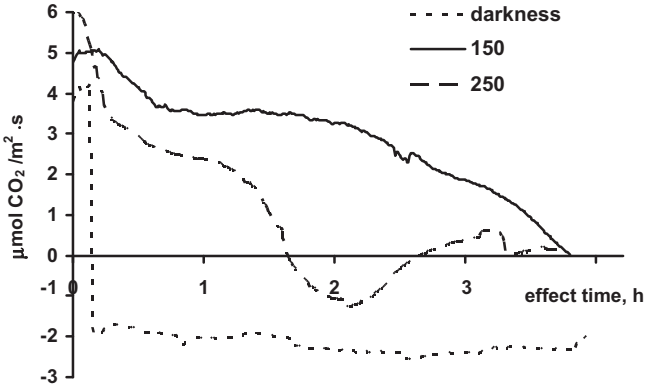

Fig. 2. Kinetics of CO_2 exchange of intact upper leaves of 15-day wheat canopy during heat stress at 44°C under different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}).

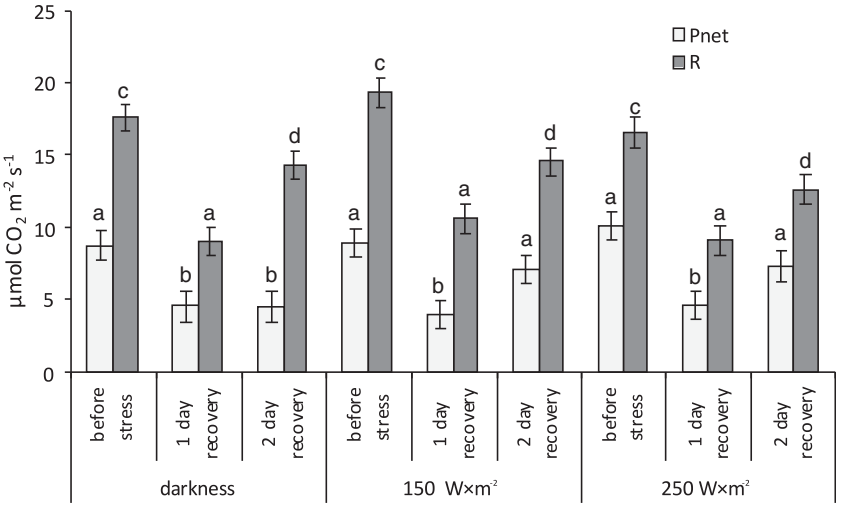

Fig. 3. Effect of PAR intensity (darkness, 150 W m^{-2} and 250 W m^{-2}) on net photosynthesis P_{net} and respiration R of wheat canopy before and after heat stress ($n = 3$). Vertical bars represent the standard errors, and the different letters above the columns indicate significant difference at $P < 0.05$ according to t-test.

From the comparison of kinetics of net photosynthesis of canopy and leaves, we find that the heat resistance of the plants on elevation of PAR intensities during the stress can increase, but obviously only within certain limits.

After 1 day in normal conditions after the heat stress in darkness, the P_{net} and R intensities were, respectively, 53% and 51% of the initial values. After 2 days, the P_{net} intensity had not changed but the respiration intensity increased and approached its initial value. Consequently in total gas exchange, the respiration part (R/P_{gross}) increased from 0.64 before the heat stress to 0.77 after the 2-day recovery (Fig. 3).

After heat stress at 150 W m^{-2} and one-day recovery, P_{net} and R were 45% and 55% of the initial values, but after 2-day recovery, P_{net} and R intensities increased, and became practically equal to their initial values (0.68 and 0.67 correspondingly) (Fig. 3). The same trend was observed after heat stress at 250 W m^{-2}, but after 2 days, the recovery was lower than after heat stress at 150 W m^{-2} (Fig. 3).

The reaction of lettuce to heat stress at 44 °C at PAR 150 W m^{-2} was also characterized by an increase of respiration, which resulted in CO_2 release in the light conditions (Fig. 4). At the end of the stress period the intensity of CO_2 release decreased. Within 24 h after the heat stress, CO_2 external gas exchange rates were returned to the initial values (Fig. 5).

Comparison of wheat and lettuce crops after heat stress at 150 W m^{-2} showed that lettuce possesses a higher heat resistance of photosynthesis and respiration processes.

3.2. Effect of light and temperature stress on the photosynthetic apparatus of wheat and lettuce grown on mineralized exometabolites

3.2.1. Lipid peroxidation

The effects of different stresses induce the formation of oxygen-active forms (OAF) in cells. At a high level of stress, pro/antioxidant balance is upset. Accumulation of malonic dialdehyde is one indicator of lipid peroxidation (LPO) increase. The LPO activation results in membrane structure and function breakdown (Lukatkin, 2002).

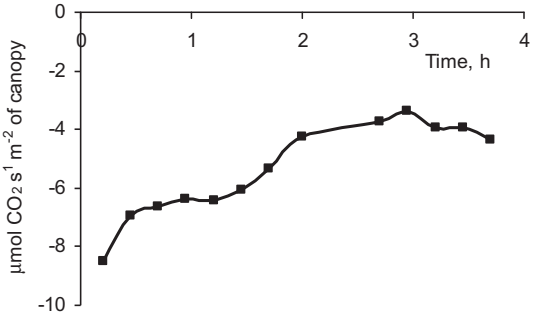

Fig. 4. Kinetics of CO_2 exchange of lettuce canopy during the effect of air temperature of 44 °C (150 W m^{-2} of PAR intensity).

Estimation of MDA level showed that in the leaves of the control plants the value of this coefficient was 85 nmol/g of dry weight (Fig. 6). Although the control plants demonstrated a stable tendential MDA accumulation, the heat stress did not cause any changes in LPO activity, indicating preservation of pro/antioxidant balance.

3.2.2. Photosynthetic pigments

Concentration of green and yellow pigments in the control wheat leaves was on average 11.3 and 2.6 mg/g of dry weight respectively. The values of chlorophyll and carotenoid contents varied in the range 4.0–4.5.

At the end of 4 h heat exposure the green pigment concentration in plant leaves stressed in darkness and at PAR 250 W m^{-2} decreased non-significantly and in light with PAR 150 W m^{-2} by 31% ($t > t_\alpha$ at $P < 0.05$) (Fig. 7A). The same trend was observed for carotenoids: the highest concentration of yellow pigments was in the leaves stressed

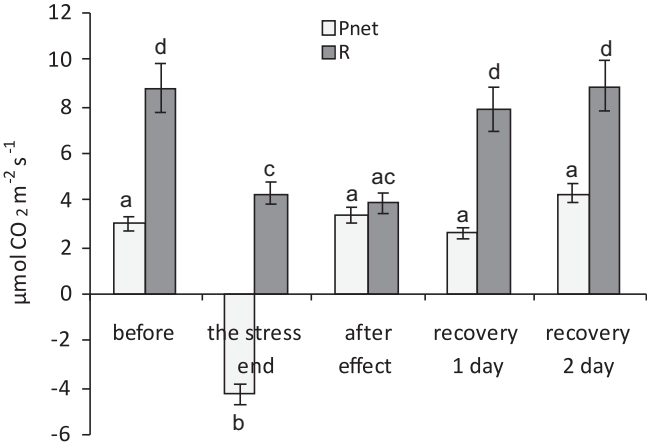

Fig. 5. Lettuce net photosynthesis P_{net} and respiration R before and after the effect of air temperature of 44 °C at 150 W m^{-2} of PAR intensity ($n = 3$). Vertical bars represent the standard errors, and the different letters above the columns indicate significant difference at $P < 0.05$ according to t-test.

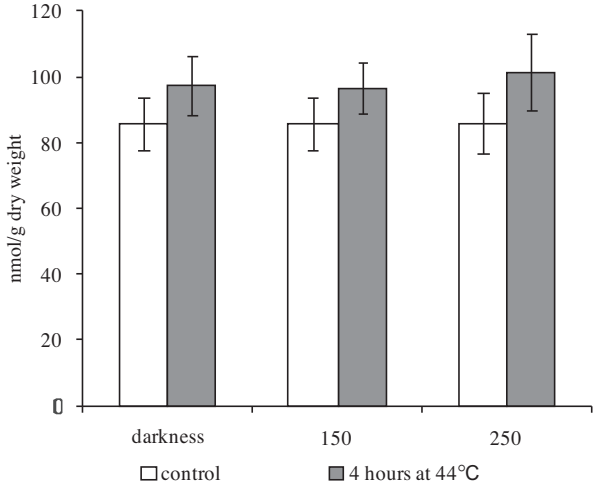

Fig. 6. Malonic dialdehyde (MDA) content in wheat leaves in nmol/g of dry weight ($n = 3$). Effect of PAR intensity (darkness, 150 W m^{-2} and 250 W m^{-2}). Vertical bars represent the standard errors.

in darkness, the lowest at 150 W m^{-2} ($t > t_\alpha$ at $P < 0.05$) (Fig. 7B).

In Fig. 7A and B, we clearly see that the return of the plants to normal growing conditions did not bring about full regeneration of photosynthetic pigment concentration in the leaves, which was still close to 70% of the control ($t > t_\alpha$ at $P < 0.05$) after 24 h and 48 h.

Thus in all the treatments, the heat stress disturbed the equilibrium between processes of regeneration and degradation of photosynthetic pigments, favoring their net destruction.

3.2.3. Estimation of PSA status by chlorophyll fluorescence

The value of maximum quantum yield of PS2 (F_v/F_m) in two-week wheat leaves varied normally in the range of 0.72–0.75 relative units. At the end of the 4 h heat stress in light with PAR 150 W m^{-2}, a barely noticeable (3–5%)

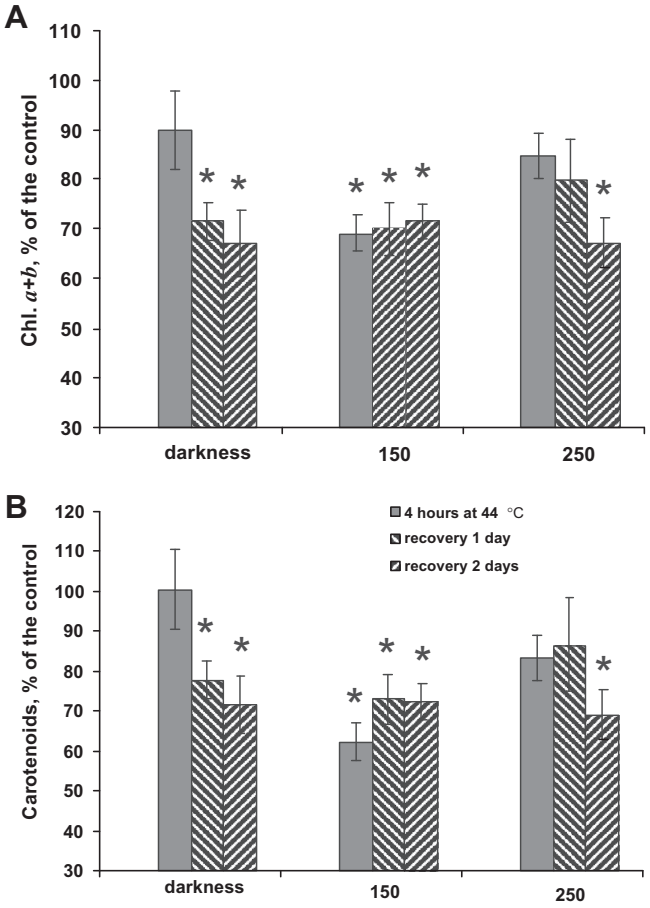

Fig. 7. Effect of heat stress under different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}) on photosynthetic pigment content (A – Chl. $a + b$; B – carotenoids) in wheat leaves, in percentage of the control ($n = 5$). Vertical bars represent the standard errors. In the t-test, * refers to a significant difference in relation to the control at $P < 0.05$.

($t > t_\alpha$ at $P < 0.05$) decrease in the value of this coefficient was observed (Fig. 8A). The return to normal conditions changed the general situation: after the heat stress under PAR 150 W m^{-2} there was no difference in the first 24 h in normal conditions; after two days a recovery process had begun, the value of maximum quantum yield of PS2 exceeding that of controls. However, for wheat in the treatment under PAR 250 W m^{-2}, only the end of the 4 h heat stress saw a noticeable ($t > t_\alpha$ at $P < 0.05$) fall in the maximum quantum yield of PS2, and there was no difference in the other cases (Fig. 8A).

More significant changes in maximum quantum yield of PS2 were observed when two-week wheat underwent heat stress in darkness. Already after 2 h of heat stress in these conditions the value F_v/F_m of experimental plants was 30% lower than controls ($t > t_\alpha$ at $P < 0.01$). When the stress was terminated, the maximum quantum yield of PS2 in the experimental plant leaves did not reach 50% of the control value ($t > t_\alpha$ at $P < 0.01$) (Fig. 8A). However, the subsequent return to normal conditions enabled gradual recovery, with the elevation of maximum quantum yield values to 80% of the control ($t > t_\alpha$ at $P < 0.05$).

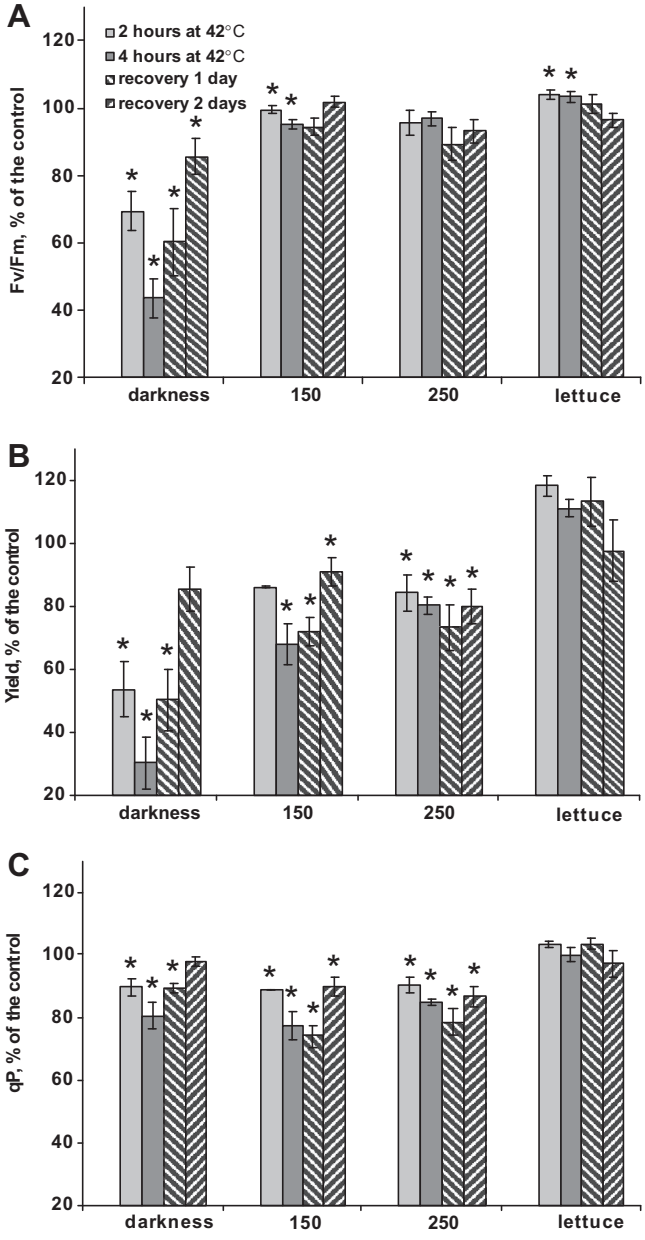

Fig. 8. Effect of heat stress (44 °C) on variables of impulse-modulated fluorescence (A – F_v/F_m, value of maximum quantum yield of PS2; B – Yield, the effective quantum yield of PSA; C – q_P, photochemical fluorescence quenching) of wheat (levels of illumination at the time of stress: darkness, PAR 150 W m^{-2} and 250 W m^{-2}) and lettuce leaf chlorophylls ($n = 5$). Vertical bars represent the standard errors. In the t-test, * refers to a significant difference in relation to the control at $P < 0.05$.

The value of the effective quantum yield of PSA of the control 15-day wheat plant leaves adapted to the PAR intensity of 150 W m^{-2} under which the plants were grown was 0.50 (Fig. 8B). This was about 30% lower than the maximum quantum yield.

After 2 h at 44 °C under the different light conditions, the wheat yield values (effective quantum yield) decreased on average by 15% of controls ($t > t_\alpha$ at $P \leqslant 0.05$). The effect in darkness, where in 2 h the given coefficient dropped significantly (by 50% of the control, $t > t_\alpha$ at $P < 0.05$), was exceptional. At the end of 4 h of heat stress applied to wheat plants the values of effective quantum yield of plant leaves in the treatment when PAR was raised to 250 W m^{-2} were practically the same (15% less, $t > t_\alpha$ at $P < 0.05$) as in controls. When the heat stress on two-week wheat was not accompanied by exposure changes in the chamber the yield value continued to drop and decreased by 30% of controls ($t > t_\alpha$ at $P < 0.05$). When the heat stress was applied in darkness, by 4 h the effective quantum yield of wheat leaves had decreased by 70% ($t > t_\alpha$ at $P < 0.05$) (Fig. 8B).

Once the wheat plants were back in normal conditions they reacted differently to the different experimental treatments. In darkness, the effective quantum yield displayed the same recovery dynamics of maximum quantum yield, and in 2 days reached the control value. Similar dynamics were observed for the treatment at PAR 150 W m^{-2}, where on the second day the recovery reached 90% of the control value ($t > t_\alpha$ at $P < 0.05$). However, after two days in normal conditions, for the treatment at 250 W m^{-2}, no clear recovery was observed, and the values of effective quantum yield remained steady ($t > t_\alpha$ at $P < 0.05$) (Fig. 8B).

The photochemical quenching (q_P) of the control wheat plants was 0.84 relative units (Fig. 8C). The heat stress caused a significant ($t > t_\alpha$ at $P < 0.05$) decrease in the photochemical quenching coefficient; after 2 h of stress the q_P value of wheat in all the treatments had decreased by about 15% of the control. At the end of 4 h exposure of two-week wheat to heat stress in darkness and at PAR 150 W m^{-2}, the q_P value was 20–23% lower than the control value ($t > t_\alpha$ at $P < 0.05$). For 250 W.m^{-2} of PAR, the q_P value decreased by 15% compared with controls ($t > t_\alpha$ at $P < 0.05$) (Fig. 8C).

The return of the plants to normal conditions enabled various degrees of recovery of photochemical fluorescence quenching depending on the light treatment. After 24 h in normal conditions after the heat stress in darkness, q_P was 90% ($t > t_\alpha$ at $P < 0.05$); after two days it did not appreciably differ from controls; 24 h after the heat stress at PAR 150 W m^{-2} the q_P value was unchanged, but after two days it increased to 90% of the control ($t > t_\alpha$ at $P < 0.05$). However, for the treatment at PAR 250 W m^{-2} the q_P value continued to fall 24 h after the heat stress, and in the normal conditions of cultivation decreased further by 6% ($P < 0.01$); two days later it had returned to its level immediately after the stress ($t > t_\alpha$ at $P < 0.05$) (Fig. 8C).

The value of maximum quantum yield of PS2 in lettuce was normally 0.76 relative units.

For lettuce, this variable increased by 4%, ($t > t_\alpha$ at $P < 0.05$) at the end of 4 h of the high temperature effect (Fig. 8A). The maximum quantum yield practically reverted to the control values in 24 h; a slight non-significant fall in the values of this variable was observed on the second day after the heat stress (Fig. 8A).

The value of the effective quantum yield of PSA of the control 15-day lettuce plant leaves adapted to the PAR

intensity of about 150 W m^{-2} under which the plants were grown was 0.54 (Fig. 8B). For lettuce the yield after 2 h of heat stress increased non-significantly compared with controls. The effective quantum yield of lettuce had completely returned to the control values by the second day of recovery (Fig. 8B).

The photochemical quenching (q_P) of the control lettuce plants was 0.87 relative units (Fig. 8C). This coefficient increased slightly for lettuce under the heat stress. The q_P value of lettuce returned to the control value after the heat stress. The changes of lettuce q_P values in the recovery period were not reliable (Fig. 8C).

3.3. Effect of temperature stress on productivity of wheat and lettuce grown on human mineralized wastes

The heat stress for 4 h at PAR 150 W m^{-2} impaired growth and development processes in both wheat and lettuce: the lettuce edible biomass decreased by 40% and inedible biomass by 33% compared with controls (Fig. 9).

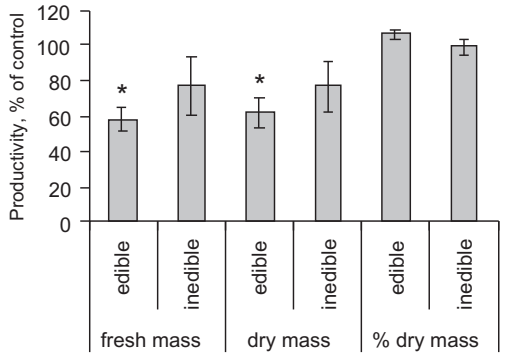

Fig. 9. Productivity of lettuce, in percentage of the control, after a 4-h stress at 44 °C at 150 W m^{-2} of PAR intensity ($n = 5$). Vertical bars represent the standard errors. In the t-test, * refers to a significant difference in relation to the control at $P < 0.05$.

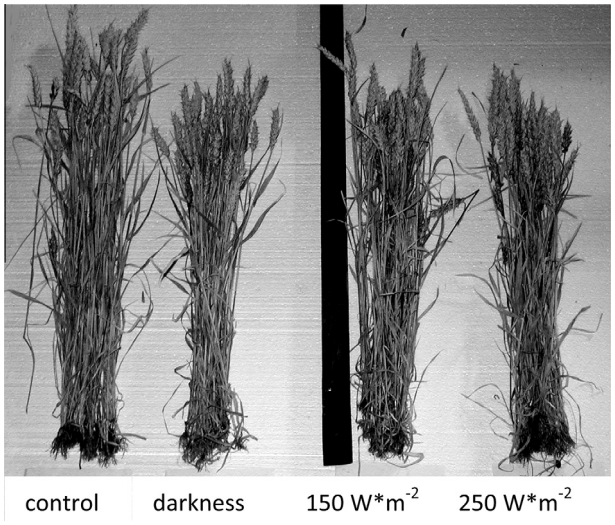

Fig. 10. Wheat after a 4-h stress at 44 °C at different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}), after harvesting.

The structure and productivity of stressed wheat in the same conditions also changed (Figs. 10–12). Here the wheat reaction to the heat stress depended on the PAR intensity. The stem heights that decreased most were those of plants that underwent the heat stress at 250 W m^{-2} (Figs. 10 and 11). At the same time the number of productive shoots and seeds in the ears of the plants were highest. We note that PAR intensity did not markedly affect the heat resistance of the wheat leading shoot. Differences in the seed numbers of the leading shoot lay within the limits of experimental error. However, an increase in the shoot seed number with PAR intensity elevation during the stress at 44 °C was observed. The differences observed were essentially linked to the development of tillers and the numbers of seeds in them.

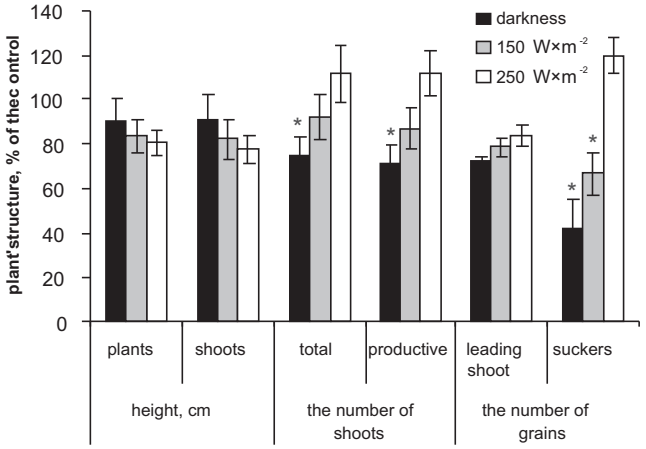

Fig. 11. Structure of wheat after a 4-h effect of air temperature at 44°C at different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}), after harvesting ($n = 5$). Vertical bars represent the standard errors. In the t-test, * refers to a significant difference in relation to the control at $P < 0.05$.

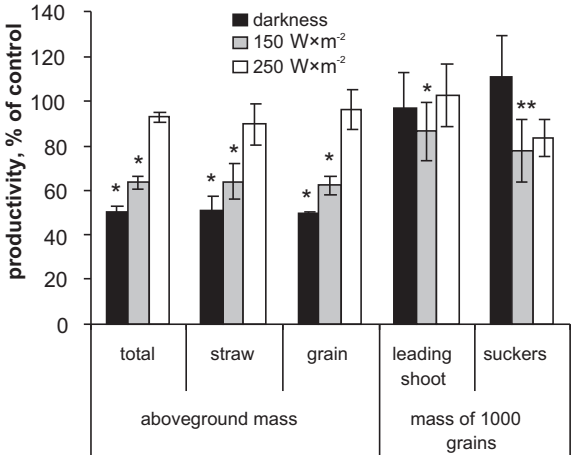

Fig. 12. Productivity of wheat after a 4-h effect of air temperature at 44 °C, at different PAR intensities (darkness, 150 W m^{-2} and 250 W m^{-2}), after harvesting ($n = 5$). Vertical bars represent the standard errors. In the t-test, * refers to a significant difference in relation to the control at $P < 0.05$.

The protective function of increased PAR intensity during heat stress was clearly observed when comparing wheat productivity (Fig. 12).

Total above-ground mass and seed mass of the plants stressed under PAR 250 W m^{-2} did not differ from controls. The above-ground mass and seed mass of plants stressed in darkness were half those of controls. The productivity of the plants stressed under PAR 150 W m^{-2} decreased by almost 40% compared with controls.

4. Conclusion

These experiments demonstrate that heat stress due to an air temperature of 44 °C held for 4 h did not result in any considerable disturbance of the photosynthetic apparatus of wheat and lettuce plants. An average decrease in photosynthetic pigments of 25% was observed during heat stress and afterwards, which may be a sign of stress.

The heat stress under illumination did not markedly affect the photochemical processes of PS2, whereas in darkness it significantly disturbed this function. On the whole, the data obtained by the impulse-modulated method of chlorophyll fluorescence showed a high level of PS2 immunity of wheat leaf chloroplasts and the activation of prompt adaptation mechanisms of the photosynthetic apparatus under heat stress. By 24 h after return to initial conditions only non-significant after-effects of the stress were observed.

The data obtained reflect the complex functional interconnection between the energy and metabolic systems. According to the literature, about half of all adenosine triphosphate (ATP) formed in the process of photophosphorylation is used for carbonic acid assimilation (Semikhatova, 1990). Insofar as heat stress resulted in inhibition of net photosynthesis at the effectively functioning energy systems, we may assume that while the stress lasts, the energy consumption for the support and for the recovery of the photosynthetic apparatus and other cellular structures increases dramatically. In addition, the energy is consumed for activation of adaptation processes, including the antioxidant response. In support of this conclusion is the fact that the disturbing effect of heat stress on photosystems was greater when the plants were in darkness during the stress.

The wheat reproductive organs were the most responsive to the heat stress. At the beginning of the stress, the leading shoots were in the flower organ formation phase. As expected, the lack of material available for the support and recovery of the structures resulted in some irreversible disturbance in the formation of reproductive organs. However, for the plants stressed under the increased light intensity, the disturbance of the leading shoot stimulated tiller formation. However, in the plants stressed in darkness an irreversible disturbance evidently occurred, resulting in the inhibition of shoot formation.

Comparison with heat resistance of wheat grown on the Knop solution shows that cultivation on human mineralized exometabolites resulted in a decrease in the heat resistance of the plants (Ushakova and Tikhomirov, 2002; Zavorueva and Ushakova, 2004). In 7 h with the air temperature at 45 °C under both 150 W m^{-2} and 250 W m^{-2}, no disturbance of the growth or development processes cultivated on the Knop solution was observed. The decreased tolerance is probably linked to the form of the nitrogen nutrition: the Knop solution contains nitrogen in nitrate form and mineralized human metabolites contain it in ammonium and amide (urea) forms. The mineralized exometabolites apparently contain compounds that lower the tolerance of the plants to adverse environmental factors. In further work, it will be necessary to clarify the immediate cause of the decreased tolerance of the plants in cultivation with the solutions based on mineralized exometabolites. This is important to ensure the robustness of the vegetative unit as a BLSS component. Through the experiments carried out, it has been shown that the plants are indeed resilient and that they can survive an event like loss of temperature control, for at least 4 h.

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