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The Response of Photosynthetic Parameters of *Triticum aestivum* L. and *Pisum sativum* L. to Different Levels of Salinity Stress at Different Climate Conditions

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The aim of this work was to investigate the response of physiological parameters of winter wheat (*Triticum aestivum* L. cv. 'Ada') and pea (*Pisum sativum* L. cv. 'Early onward') to different levels of salinity stress under changing climate conditions. Experimental plants were grown in growth chambers under the conditions of current climate (400 μmol mol^{-1} CO_2 and day/night temperatures of 21/14 °C) and warmed climate (800 μmol mol^{-1} CO_2 and day/night temperatures of 25/18 °C). Under both climate conditions, plants were exposed to 100 mM and 200 mM concentrations of sodium chloride. Exposure to salinity stress was initiated when plants developed the second true leaf or their pair. After salinity treatments, which lasted 2 weeks, the response of photosynthetic and transpiration rates, intracellular content of CO_2, water use efficiency, II photosystem quantum efficiency rate and photosynthesis performance index were measured. Physiological parameters of pea plants were mostly affected at current climate conditions. The biggest reductions were found in the photosynthetic and the transpiration rate, which decreased by 57.0% and 71.5% under 200 mM effect, respectively. Assessment of intensity of fluctuation of wheat and pea physiological parameters at current climate showed that changes were bigger in pea plants (51.3%) in comparison with wheat (29.3%). While at warmed climate conditions, the changes in physiological parameters of peas and wheat were the opposite: for wheat, they increased and became 52.6%, and for peas, they decreased to 41.1% in comparison with control plants ($p < 0.05$).

Keywords: wheat, pea, climate change, salinity, photosynthesis.

Introduction

Carbon dioxide is estimated to continually increase from currently 400 ppm to between 750 and 1300 ppm by the end of this century (IPCC 2014). The global mean surface air temperature is predicted to increase about 1.8–6.0 °C by the end of the 21st century (IPCC 2014). It is well known that rising [CO_2] has a direct effect on plant metabolism and growth because CO_2 is the carbon substrate of photosynthesis, and there is also a common assumption that the rising temperature will directly increase the release of CO_2 via respiration both in light and darkness (Zhang et al. 2017, Kacienė et al. 2017).

Many recent papers and reviews emphasize how the enhancement of atmospheric CO_2 directly impacts the physiology of plants, generally accelerates the photosynthetic rate and increases plant growth and yield (He et al. 2010, Sun et al. 2010, de la Mata et al. 2012, Zhang et al. 2012). Moreover, elevated CO_2 can lead to reallocation of carbon and nitrogen resources among plant organs, and change the secondary metabolites content of plant tissues (Sun et al. 2010). Because the two environmental factors are concomitant, the worst issue is that any rise in the atmospheric CO_2 level will lead to an increase in air temperature. Not surprisingly, some researchers have focused on understanding of the interactions between CO_2 and temperature on plant growth and development (Morison and Lawlor 1999). In theory, a progressive increase in CO_2 could decrease the ratio of photosynthesis to photorespiration and the ratio of gross photosynthesis to dark respiration at a higher temperature. Long (1991) has demonstrated that an increase in CO_2 from 350 to 650 micro mol mol^{-1} could raise the optimum temperature of light-saturated leaf photosynthesis by 5 °C and the relative stimulation of light compensation point by elevated CO_2 was reduced at a high temperature (Chang et al. 2016).

Salt damage is one of the most important abiotic stresses in global agricultural regions. Twenty percent of global land area and over 50% of agricultural irrigated land is influenced by salt damage. It is estimated that over 25% of agricultural land in the next 25 years, and even 50% by 2050, will be rendered unusable for farming due to salt (Mahajan and Tuteja 2005, Yan et al. 2005). Soil salinization not only affects the ecological environment balance, but also seriously restricts sustainable development of the economy and world agriculture (Niu and Wang 2002). Salt stress simultaneously leads to other secondary stresses of crops, moderate salt-stress results in ion and osmotic stress in plants, and severe stress will destroy the ion balance leading to nutritional deficiency and oxidative stress (Wang et al. 2003).

Salt stress leads to a decrease in the efficiency of photosynthesis (Sayed 2003) and is known to influence the chlorophyll content of plant leaves (Kahn 2003). PS II is more sensitive to all types of stresses as compared with PS I (Apostolova et al. 2006). However, photosynthesis may decrease by the direct effect of salt stress on photosynthetic electron transport (Borsani et al. 2001). Salt stress involves the known component of osmotic stress and ionic toxicity. Salinity reduces plant growth through both nonspecific (osmotic) effects and ion specific mechanisms (Tavvakkoli et al. 2008, Mehta et al. 2010, Negrao et al. 2017).

Crops under salt stress often adapt to it through adjusting related metabolic processes and morphological characteristics. This includes inducing expression of cell membrane and cell wall related proteins; changing cell shape; inducing various kinds of osmoprotectants, late embryogenesis abundant (LEA) proteins, chaperones and detoxification enzymes; protecting cell, cell organelle and proteins; as well as stimulating related stress signal transduction pathways (Seki et al. 2003, Shinozaki et al. 2003, Cheng et al. 2016).

These problems could be partially alleviated by utilization of low quality irrigation water such as saline groundwater or seawater on appropriate wastelands for production of non-conventional crops, especially in arid regions. Most of the conventional crops cannot tolerate salinity even at low concentrations. It is, therefore, necessary to develop sustainable biological production systems for brackish or high salinity water irrigation. The development of suitable halophytic crops has been considered for the production of food, forage, oil, wood, timber, ornamental, medicine and biofuel (Koyro and Eisa 2008, Koyro et al. 2011). Halophytes are extremophiles and are equipped with physiological and biochemical mechanisms enabling them to cope with high soil salinity (Koyro et al. 2013). Thus, the aim of this work was to investigate the response of physiological parameters of winter wheat (*Triticum aestivum* L. cv. 'Ada') and pea (*Pisum sativum* L. cv. 'Early onward') to different levels of salinity stress under changing climate conditions.

Materials and methods

Experimental plants were grown in the control chamber under the conditions of current climate (400 μmol mol^{-1} CO_2 and day/night temperatures of 21/14 °C) and warmed climate (800 μmol mol^{-1} CO_2 and day/night temperatures of 25/18 °C). Under both climate conditions, plants were exposed to various intensity of salinity stress. Plants were affected by 100 mM and 200 mM concentrations of sodium chloride. Exposure to salinity stress was initiated when plants developed the second true leaf or their pair (BBCH 12) (Meier 2001). After salinity treatments, which lasted 2 weeks, the response of physiological parameters such as photosynthetic and transpiration rate, intracellular CO_2 concentration, water use efficiency, photosynthesis performance index (PIabs), and maximum II photosystem quantum efficiency rate (Fv/Fm) were measured.

Gas exchange parameters were measured with a portable photosynthesis system LI-6400 (LI-COR, USA). Net photosynthetic rate (μmol CO_2 $m^{-2}s^{-1}$), intercellular CO_2 concentration (μmol CO_2 mol air^{-1}), transpiration (mmol H_2O m^{-2} s^{-1}) and water use efficiency (μmol CO_2 mmol H_2O^{-1}) of the 3rd fully expanded leaves were registered every 3 seconds for 5 minutes in 5 randomly selected plants of each treatment variant; from these data, a mean of daily measured indices was calculated. The CO_2 level and temperature were set depending on the individual treatment on plants. During the measurements, the CO_2 concentration in the chamber of leaves was controlled with the LI-COR CO_2 injection system. The airflow rate through the assimilation chamber was maintained at 400 μmol s^{-1}, and PAR was about 170 μmol m^{-2} s^{-1}.

Chlorophyll *a* fluorescence parameters were determined by using 'Hansatech' fluorimeter – Handy Pea. Measurements were performed by applying special clips to healthy leaves and darkening them for 15 min. Then, the measuring sensor was applied to a clipper and the measurement was taken (Hansatech Instruments 2006).

Measured parameters:

- Fv/Fm – maximum II photosystem quantum efficiency rate,
- PIabs – photosynthesis performance index,

All measurements were carried out in 3 replications.

For comparison of independent variables, the Student t and U tests were used. All analyses were performed by STATISTICA and EXCEL and the results were expressed as mean values and respective confidence intervals ($CI_{0.05}$) with a significance level of 5% ($p < 0.05$).

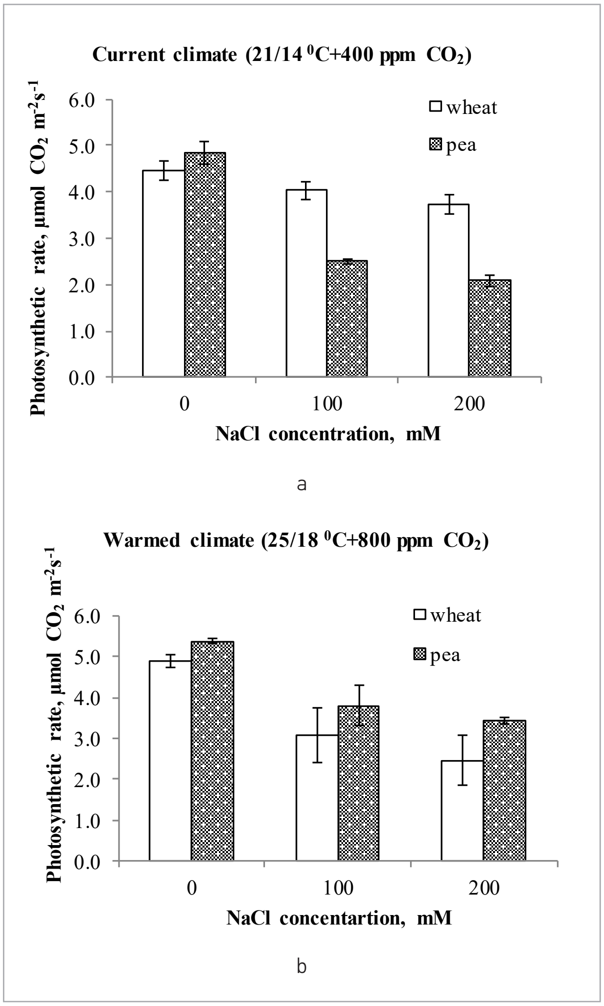
Results and discussion

The effect of NaCl on plants at the current climate conditions is presented in Figure 1A; 100 mM and 200 mM concentrations reduced the photosynthetic rate of wheat by 9.5% and 16.3%, respectively, compared with control plants ($p < 0.05$). Meanwhile, the photosynthetic rate of peas tended to decrease with increasing NaCl concentration ($p < 0.05$). The effect of 100 mM and 200 mM decreased it by 48.4% and 57.0%, respectively, compared with control plants ($p < 0.05$). At warmed climate conditions (Fig. 1B), 100 mM decreased the photosynthetic rate in wheat leaves by 37.0% compared with control plants ($p > 0.05$). The effect of the 200 mM NaCl concentration on the photosynthetic rate in wheat decreased by 50.0% compared with control plants ($p < 0.05$). As the NaCl concentration increased from 100 to 200 mM, the photosynthetic rate of peas decreased by 29.3% and 35.9%, respectively, compared with control plants ($p < 0.05$).

Investigations conducted with salinity tolerant and salinity sensitive wheat varieties showed that the salt tolerant varieties had a higher photosynthesis rate, and the photosynthetic rate of salinity sensitive varieties was significantly lower (Subrahmanyam et al. 2006). A study done with peas showed that a decrease in the potential of photosynthesis may be due to a decrease

Fig. 1

The changes of the photosynthetic rate under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)



a

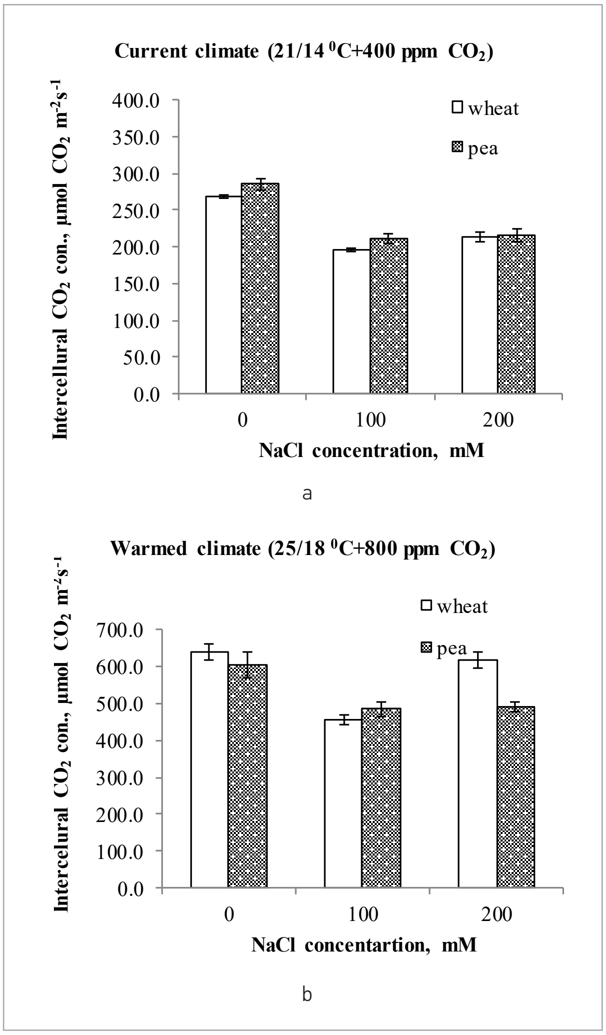
b

in the leaf area (Farouk et al. 2011). The results of this study confirm that the photosynthetic rate of sensitive plants (peas) was significantly lower, and the climate change softened the effect of salinity on the intensity of photosynthesis of peas. The gas exchange parameters, such as the leaf CO_2 assimilation rate, transpiration and others, were strongly affected by salt treatments, principally high external NaCl, in other researches too (Rodrigues et al. 2014). A reduction in photosynthesis associated with low stomatal conductance is a strong indicator of stomatal limitations, as demonstrated in cotton cultivars and Jatropha cur-cas plants (Meloni et al. 2003, Silva et al. 2011).

Under the effect of the 100 mM NaCl concentration, the wheat intercellular CO_2 concentration decreased by 26.7% compared with control plants ($p < 0.05$) at current climate conditions (Fig. 2A). The effect of the 200 mM NaCl concentration resulted in a decrease in the intercellular CO_2 concentration of 20.1% compared with control plants ($p < 0.05$). The intercellular CO_2 concentration in pea leaves by the increase in salinity also changed unevenly. Besides, 100 and 200 mM NaCl concentrations decreased the intercellular CO_2 concentration by 26.2% and 24.7%, respectively compared with control counts ($p < 0.05$). At warmed climate conditions, the intercellular CO_2 concentration of

Fig. 2

The changes of intercellular CO_2 concentrations under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)



a

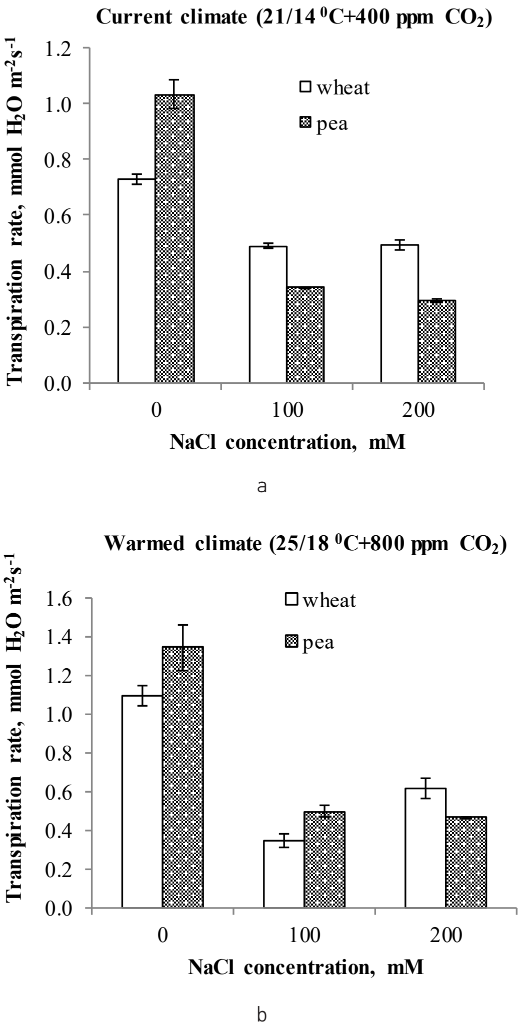
b

wheat in the presence of 100 mM NaCl decreased by 28.61% compared with control plants ($p < 0.05$) (Fig. 2.B). The effect of the 200 mM NaCl concentration was insignificant ($p > 0.05$). The intercellular CO_2 concentration of peas was decreasing with increasing salinity compared with control plants ($p < 0.05$).

The transpiration rate of wheat at current climate conditions decreased by 32.9% ($p < 0.05$) under the effect of 100 mM NaCl (Fig. 3A). In wheat, the 200 mM NaCl concentration decreased the transpiration rate by 32.5% compared with control plants ($p < 0.05$). The pea transpiration rate decreased more than 2 times more than wheat, i.e., by 66.9% and 71.5%, under the impact of 100 and 200 mM NaCl, respectively, compared with control plants ($p < 0.05$). At warmed climate conditions (Fig. 3B), the effect of 100 and 200 mM NaCl reduced the wheat transpiration rate by 68.3% and 43.9%, respectively, compared with control plants ($p < 0.05$). The effect of 100 and 200 mM NaCl concentrations resulted in a decrease by 62.9% and 65.3%, respectively, in the transpiration rate of peas compared with control plants ($p < 0.05$).

Fig. 3

The changes of the transpiration rate under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)



a

b

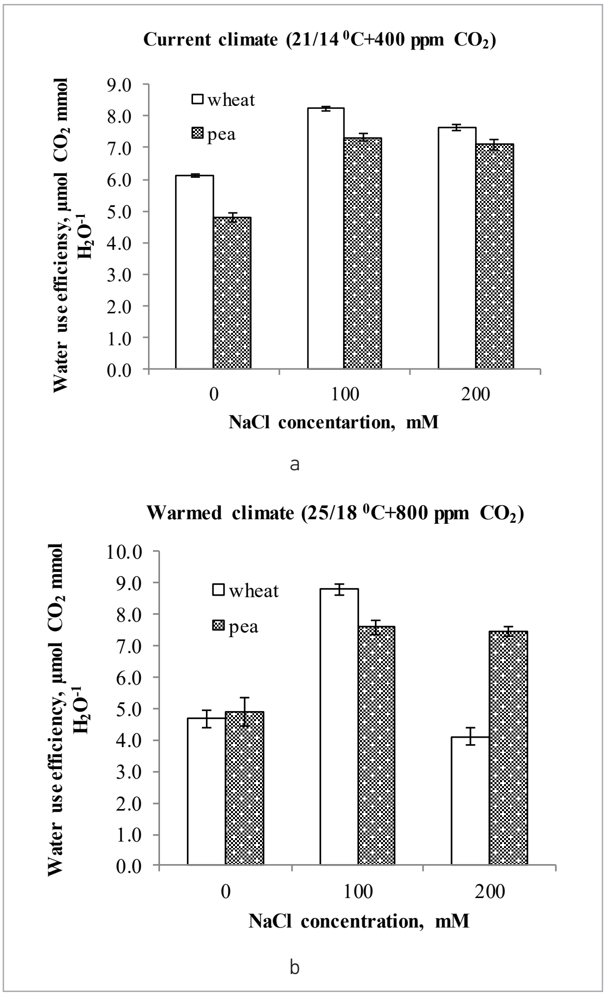
The gas exchange parameters, such as leaf CO_2 assimilation rate, transpiration, stomatal conductance and carboxylation instantaneous efficiency were strongly affected by salt treatments, principally high external NaCl. A reduction in photosynthesis associated with low stomatal conductance is a strong indicator of stomatal limitations (Meloni et al. 2003, Silva et al. 2011, Rodrigues et al. 2014). A study with *Brassica* plants has shown that the intensity of transpiration also decreases with increasing salinity. Such changes may be related to decreased stomatal conductance, low density of stomata in plant leaves, and a reduced leaf area under salinity stress (Omamt et al. 2006).

At current climate conditions, the water use efficiency of wheat under the effect of the 100 mM NaCl concentration increased by 34.6% compared with control plants ($p < 0.05$) (Fig. 4A). The effect of the 200 mM NaCl concentration resulted in a statistically significant increase ($p < 0.05$) in the water use efficiency by 24.7% compared with control plants. The water use efficiency of peas, as well as that of wheat, was mostly impacted by a concentration of 100 mM NaCl, which resulted in a significant increase in WUE up to 52.0% compared with control ($p < 0.05$). In the effect of 200 mM NaCl, the water use efficiency of peas also increased by 47.2% ($p < 0.05$) compared with control plants.

The effect of the 100 mM NaCl concentration at warmed climate resulted in a significant increase by 87.7% ($p < 0.05$) of the wheat water use efficiency compared with control plants (Fig. 4B). When wheat was affected by the 200 mM NaCl concentration, the water use efficiency decreased significantly compared with 100 mM NaCl ($p < 0.05$), but remained unchanged compared with control plants ($p > 0.05$). The effect of the 100 mM NaCl concentration reduced the water use efficiency of peas by 54.8% compared with control plants ($p < 0.05$). The 200 mM concentration increased the water use efficiency of peas by 52.1% ($p < 0.05$) compared with control plants.

Fig. 4

The changes of water use efficiency under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)



a

b

Salinity significantly reduced the photosynthetic and transpiration rates of both investigated plants, and the reduction was proportional to the increase in the NaCl level. A reduction in photosynthetic carbon assimilation was similarly noted in *Phaseolus* species (Bayuelo-Jimenez et al. 2003) and tobacco (Sifolaand Postiglione et al. 2002), and this decrease was attributed to reduced stomatal conductance. It has been reported that the reduction in net CO_2 assimilation by increased salinity could be owing to a limitation of CO_2 supply because of stomatal closure (Steduto et al. 2000), to non-stomatal factors related to the toxic effect of salts on photosynthetic apparatus, or inhibition in photochemical capacity or a combination of these factors. Some results show a close relationship between the photosynthetic rate, stomatal conductance and stomatal density, indicating that the reduction in net CO_2 assimilation could be explained by the reduction in stomatal conductance and stomatal density (Omamt et al. 2006).

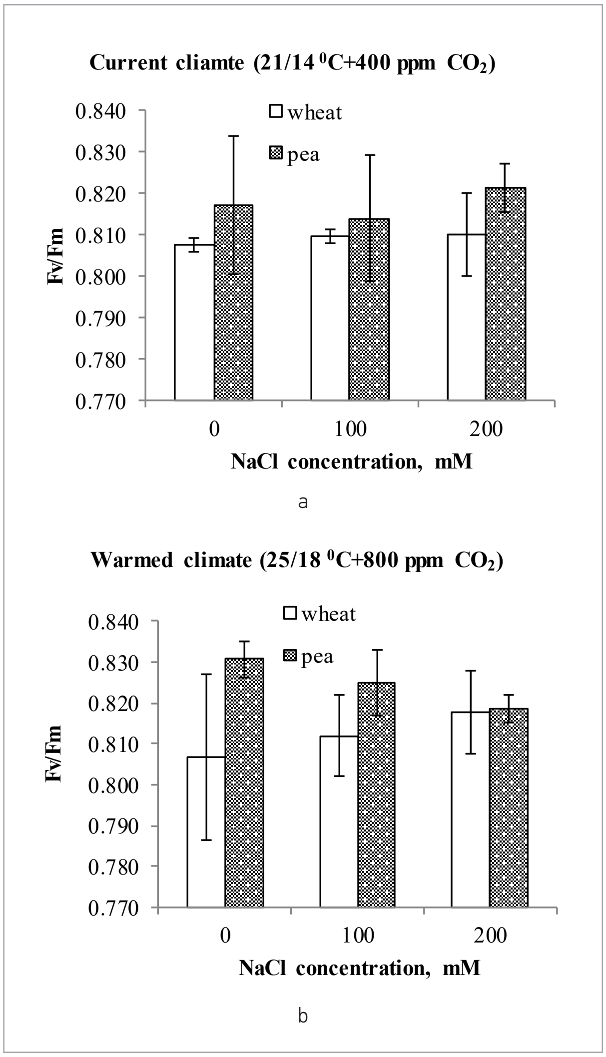
At current climate conditions, the changes of the efficiency of the II photosystem (Fv/Fm ratio) of wheat and peas were not significantly different from control plants ($p > 0.05$) (Fig. 5A). In the case of warmed climate conditions, the Fv/Fm ratio of wheat and peas remained almost constant as in current climate, only the concentration of 200 mM NaCl caused a statistically significant decrease in the efficiency of the II photosystem of peas by 1.4% compared with control ($p < 0.05$) (Fig. 5B). These results are explained by the fact that salinity does not affect the number of quanta absorbed per unit time (Mehta et al. 2010). Such insignificant changes in the ratio of Fv/Fm are also possible due to the relative differences in water content in plants that can mitigate the negative effect of salinity on the Fv/Fm ratio (Slabbert and Kruger, 2011).

The most popular parameter of the JIP test is the performance index (PI). The photosynthetic performance index is an indicator of sample vitality. It is the combined measurement of the amount of photosynthetic reaction centres (RC/ABS), the maximal energy flux that reaches the PS II reaction centres and the electron transport at the onset of illumination (Mehta et al. 2010).

At current climate conditions with increasing concentration of salinity, the performance index (PIabs) of wheat and peas was changing unevenly (Fig. 6A). At warmed climate conditions, the performance index increased by 36.5% as compared with control plants ($p < 0.05$) when wheat was affected by the 100 mM NaCl concentration, and it increased by 81.2% ($p < 0.05$) under the 200 mM effect (Fig. 6B). The performance

Fig. 5

The changes of the II photosystem efficiency (Fv/Fm) under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)

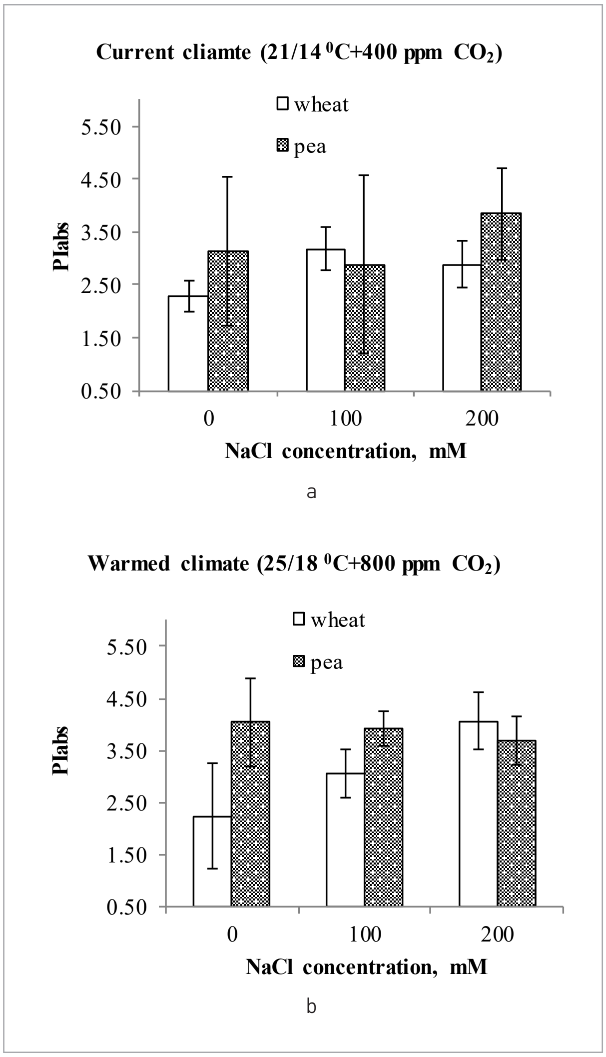


a

b

Fig. 6

The changes of the performance index (PIabs) under the effect of NaCl at different climate conditions (mean±$CI_{0.05}$)



a

b

index of peas with steady growth of NaCl remained constant, with a slight but insignificant decrease in the trend ($p > 0.05$). Under salt stress, there is a loss in chlorophyll protein (47 kDa) and a core membrane linker protein 94 kDa that can attach phycobilisome to thylakoid (Garnier et al. 1994). Effects of salt stress in cyanobacterium *Spirulina platensis* (Sudhir et al. 2005) showed a decrease in PS II mediated oxygen evolution activity and an increase in PS I activity. It was ascribed to changes in the thylakoid membrane protein profile, which led to the decreased energy transfer from light harvesting antenna to PS II (Mehta et al. 2010).

Summarizing all changes of the investigated parameters, the average of percentage changes of the investigated parameters was calculated. And the following results were obtained: at current climate, the average changes in physiological parameters of peas were 51.3% and those of wheat were 29.3%; and at warmed climate, the average changes in physiological parameters of wheat increased and became 52.6%, and those of peas decreased to 41.1%, in comparison with the current climate control plants ($p < 0.05$).

Conclusions

Higher changes of physiological parameters of wheat were detected at warmed climate conditions, as the biggest reduction was detected in the transpiration rate, the photosynthesis performance index and the water use efficiency, when the mentioned parameters fluctuated by 68.3%, 81.2% and 87.7%, respectively, over control.

The biggest reductions of the photosynthetic and the transpiration rate of peas was found after the 200 mM NaCl effect at current climate conditions, when this salinity level decreased the mentioned parameters by 57.0% and 71.5%, respectively ($p < 0.05$).

At current climate, changes in physiological parameters of peas were significantly higher (51.3%) compared with wheat (29.3%). While at warmed climate, the changes in physiological parameters of investigated plants were opposite: for wheat, they increased and became 52.6%, and for peas, they decreased to 41.1% in comparison with current climate control plants ($p < 0.05$).

At current climate conditions, salinity more strongly affected physiological parameters of peas, and at warmed climate conditions the opposite was detected – wheat became more sensitive than peas.

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Triticum aestivum L. ir *Pisum sativum* L. fotosintezės rodiklių atsakas į druskingumo stresą esant skirtingomis klimato sąlygomis

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Šio darbo tikslas – ištirti žieminių kviečių (*Triticum aestivum* L. veislė ‘Ada’) ir žirnių (*Pisum sativum* L. veisė ‘Early-onward’) fiziologinių rodiklių pokyčius dėl skirtingo druskingumo poveikio esant skirtingoms klimato sąlygomis. Tirti augalai auginti klimato kamerose esant dabartiniam klimatui (400 μmol mol^{-1} CO$_2$ ir dienos/nakties temperatūra 21/14 °C) ir atšilusiam klimatui (800 μmol mol^{-1} CO$_2$ ir dienos/nakties temperatūra 25/18 °C). Abiejose klimato sąlygose augalai buvo paveikti 100 mM ir 200 mM natrio chlorido koncentracijomis. Augalams išleidus antrą tikrąjį lapą ar jų porą pradėtas druskingumo poveikis, kuris tęsėsi dvi savaites. Eksperimento pabaigoje išmatuota fotosintezės ir transpiracijos intensyvumai, viduląstelinis CO$_2$ kiekis, vandens naudojimo efektyvumas, II fotosistemos darbo efektyvumas ir fotosintezės gyvybingumo indeksas. Tyrimo metu stipriausiai buvo paveikti žirnių fiziologiniai rodikliai esant dabartinėms klimato sąlygoms, kuomet nustatytas didžiausias 57,0 % ir 71,5 % fotosintezės ir transpiracijos intensyvumų sumažėjimas atitinkamai, esant 200 mM druskingumo poveikiui. Įvertinus vidutinį kviečių ir žirnių fiziologinių rodiklių pokyčių intensyvumą dabartiniame klimate, didesni buvo žirnių ir siekė 51,3 %, o kviečių sudarė tik 29,3 %. Tuo tarpu atšilusio klimato sąlygomis tirtų augalų fiziologinių rodiklių pokyčiai buvo priešingi: kviečiams nuostoliai padidėjo ir tapo 52,6 %, o žirnių sumažėjo iki 41,1 %, palyginti su kontroliniais augalais ($p < 0,05$).

Raktiniai žodžiai: kviečiai, žirniai, klimato kaita, druskingumas, fotosintezė.