

Effects of nitrogen fertilizer on photosynthetic light energy transfer and use in hybrid fescue under drought stress

Januškaitienė I.^{1*}, Dikšaitytė A.¹, Sujetovienė G.¹, Žaltauskaitė J.¹, Kacienė G.¹, Miškelytė D.¹, Juknys R.¹

¹ Vytautas Magnus University, Universiteto 10, Kauno raj, Lithuania

*corresponding author: e-mail: irena.januskaitiene@vdu.lt

Abstract. The aim of this study was to investigate the response of chlorophyll a fluorescence parameters of hybrid fescue (× Festulolium loliaceum (Huds.) P. Fourn.) under partly regulated environment and drought stress effect. The results showed that the increasing nitrogen content increased the values of the studied indicators. Meanwhile drought stress reduced them. At the highest fertilization level N90, the photosynthetic performance (PI_{total}) index increased by 89 % compared to N0, the effect of drought reduced this indicator by 50 %, but remained higher than PI_{total} of plants of N0 level. After the recovery period, these differences became even more pronounced. Similar regularities were found for other investigated parameters such as quantum yield of electron transport (phi(Eo)) and quantum yield for the reduction of end acceptors of PSI per photon absorbed (phi(Ro)). Total electron carriers per reaction center (Sm) also increased with increasing nitrogen content in the soil. The higher nitrogen content significantly increased the values of the mentioned indicators after the recovery period. These aforementioned changes in photosynthetic energy transfer also lead to higher plant biomass and lower losses due to drought stress.

Keywords: drought stress, nitrogen fertilization, chlorophyll fluorescence, forage crop

1. Introduction

Changing climatic conditions under the current scenarios will affect agricultural productivity worldwide and therefore have a significant impact on global food supply (Nazir et al., 2017). Since drought inhibits or slows down the fixation of photosynthetic carbon, mainly by limiting the entry of CO_2 into the leaf or by directly inhibiting metabolism (Zhuang et al., 2020). Thus, the search for measures to mitigate the negative effects of these stressors is very relevant. Nitrogen absorption and utilization in drought stress conditions are known to be critical for plant growth and productivity, and the use of N can contribute to drought resistance in many plants (Wanaich et al., 2011; Elkelish et al., 2020). In the absence of water, N

reserves can help increase crop resistance to drought by protecting the photosynthetic apparatus, activating antioxidant protection systems, and improving osmoregulation (Gou et al., 2017; Wang et al., 2020).

Chlorophyll a fluorescence measurements delivers data on physical changes in pigment-protein complexes, excitation energy transfer, primary photochemistry and the operating quantum efficiency of electron transport through PSII (Rosenqvist, 2004). Drought stress can not only cause structural damage to PSII and lightharvesting complexes directly, but also affect the process of photosynthetic electron transport and photophosphorylation (Hura et al., 2007). Nitrogen plays an important role in transfer and dissipation of excess light energy, which can relieve damage of excess excitation energy to photosynthetic apparatus (Wang et al., 2016).

2. Material and methods

To study the response of chlorophyll a fluorescence parameters of hybrid fescue (× Festulolium loliaceum (Huds.) P. Fourn.) under partly regulated environment and drought stress effect plants were grown in pots filled with a mixture of field soil, perlite and fine sand (volume ratio 5:3:2) in the greenhouse. Before sowing, the commercial fertilizer (NPK 8-19-29) (Achema, Lithuania) was applied at a rate of 310 kg ha⁻¹ to reach background levels of nitrogen (N). Three different treatments were conducted to imitate different nitrogen fertilization rates: background level (N0), moderate (N60) and high (N90) fertilization. In the treatment N60 and N90, additional nitrogen doses of 60 and 90 kg ha⁻¹ were applied as ammonium nitrate. When plants reached three leaves development stage, drought stress was imposed. Drought was applied by withholding watering for 7 days and after this period plants were re-watered to 30% of SWC and left for 7 days for recovery. Chlorophyll a fluorescence parameters measurements were taken with the Plant Efficiency Analyser, PEA (Hansatech Instruments, Ltd., King's Lynn, Norfolk,

England) with randomly selected youngest fully expanded leaves on the last (7^{th}) day of the exposure of drought and after recovery.

The Least Significant Differences (LSD) test procedure was applied to estimate the difference between different treatment values in all parameters and p value < 0.05 was the threshold for significance.

3. Results and discussion

Nitrogen is an important constituent in plant biomolecules, such as amino acids, amides, proteins, ribonucleic acid, chlorophyll, enzymes, vitamins, and the like. Nitrogen enables the plant to be faster establishment and produce more photosynthesis compounds (Pishva et al., 2020). The results of this study showed that the increasing nitrogen content increased the values of the studied indicators (Fig. 1-2). Meanwhile drought stress reduced them. Drought is thought to be the main factors restricting a gricultural crop yields because it has negative effects on photosynthesis, protein content, leaf expansion, and respiration (Chaves et al. 2003; Gao et al., 2020).

Under elevated N supply, plants can present stimulated growth and physiological performance, and show

different nutrient status (Xu et al., 2015). Nitrogen application might promote the physiological responses to drought via elevating N and chlorophyll concentrations, and enhancing PSII photochemical activity (Song et al., 2019). In the present study, at the highest fertilization level N90, the photosynthetic performance index (PI_{total}) increased by 89 % compared to N0, the effect of drought reduced this indicator by 50 %, but remained higher than PI_{total} of plants of N0 level. After the recovery period, these differences became even more pronounced when a difference of 2.1 (control plants) and 2.8 (drought-stressed plants) times was found between the N0 and N90 treatment variants.

Norma lized total complementary area above the OJIP (reflecting multiple turnover Q_A reduction events) (Sm) also increased with increasing nitrogen content in the soil, and the increases in optimal irrigation conditions were statistically insignificant, while in the presence of drought, higher nitrogen content statistically significantly increased the number of total electron carriers per reaction center (Sm). These changes were particularly pronounced after the recovery period. The parameter Sm, assessing of the electron transporter PQ pool between PS II and PSI, increase in Sm drought stress might be also realted to an increased electron transport between these photosystems (Stirbet, 2011).

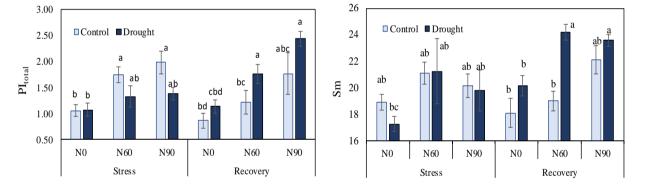


Figure 1. The changes of photosynthetic performance index (PI_{total}) and normalized total complementary area above the OJIP (Sm) of hybrid fescue under drought stress effect and after recovery period at different nitrogen concentrations in soil. The statistically significant differences among the treatments within each period (stress and recovery) are labeled with different lowercase letters at p < 0.05 (Fisher's LSD). The error bars represent standard errors of means (n = 5).

Similar regularities were found for other investigated parameters such as quantum yield of electron transport (phi(Eo)) and quantum yield for the reduction of end acceptors of PSI per photon absorbed (phi(Ro)). The higher nitrogen content significantly increased the values of the mentioned indicators after the recovery period, when the energy fluxes and use efficiency of drought-stressed plants increased significantly and became even higher than that of properly irrigated control plants. Drought stress reduced the quantum yield, which indicates photosystem II efficiencies. The loss of this trait represents the occurrence of light inhibitory effect by environmental stresses (Kocheva et al., 2004). During electron transport to photosystem I, drought stresses may cause interferences in Hill's reaction and thereby quantum yield of photosystem II can be reduced (Zelatev and Yordanov, 2004; Killi et al., 2020). Nitrogen increases fluorescence quantum efficiency through the development of leaf area, assimilates distribution and keeps up the photosynthetic efficiency (Arduiniet et al., 2006; Movludi et al., 2014).

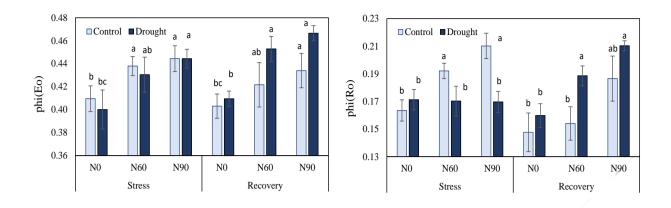


Figure 2. The changes of quantum yield of electron transport (phi(Eo)) and quantum yield for the reduction of end acceptors of PSI per photon absorbed (phi(Ro)) of hybrid fescue under drought stress effect and after recovery period at different nitrogen concentrations in soil. Designations as in Fig. 1.

These aforementioned changes in photosynthetic energy transfer also lead to higher plant biomass and lower losses due to drought stress (Fig. 3). Plants adapted to dry conditions generally have higher roots mass allocation, and lower leaf petiole length and SLA, which is beneficial for water absorbance, and promotes water conductance (Xu et al. 2009). Additionally, drought stress may cause structural injuries of the photosystem II (PSII) and lightharvesting complex, and diminish electron transport through PSII, thereby inhibiting the photosynthetic process (Quero et al. 2006). Under both optimum and limited water conditions, N input had significant effects on total biomass accumulation (Fig. 3). However, the influence of N supply on chlorophyll *a* fluorescence indicators was not so clear. Some studies indicated that improved N supply strengthens the adverse effects of drought stress (Friedrich et al. 2012), because fertilized plants develop high shoots for aboveground resources, increasing the transpiring surface and further water shortage. Other studies have shown distinct outcomes for the combined effects of drought stress and N deposition, such as synergism (DaMatta et al. 2002) and antagonism (Wu et al. 2008; Wang et al., 2016), or separate influences (Wang et al., 2020; Pishva et al., 2020). However, a consistent conclusion is still lacking even in the newest researches (Xu et al., 2015; Wang et al., 2020; Pishva et al., 2020).

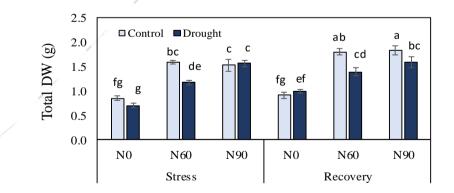


Figure 3. The changes of total dry weight of hybrid fescue under drought stress effect and after recovery period at different nitrogen concentrations in soil. Designations as in Fig. 1.

4. Conclusion

This study highlights that different level of nitrogen application changes hybrid fescue photosynthetic light energy use transitions and growth response to drought stress.

References

Arduini I., Masoni A., Ercoli L. and Mariotti M. (2006), Grain yield, and dry matter and nitrogen accumulation and remobilization in durum wheat as affected by variety and seeding rate, *Eur J Agron*, **25**, 309-318.

- DaMatta F.M., Loos R.A., Silva E.A., Loureiro M.E. and Ducatti C. (2002), Effects of soil water deficit and nitrogen nutrition on water relations and photosynthesis of pot-grown Coffea canephora Pierre, *Trees*, **16**, 555–558.
- Elkelish A., Ibrahim M.F., Ashour H., Bondok A., Mukherjee S., Aftab T., and El-Gawad H.G.A. (2021), Exogenous application of nitric oxide mitigates water stress and reduces natural viral disease incidence of tomato plants subjected to deficit irrigation, *Agronomy*, **11**(1), 87.
- Friedrich U., von Oheimb G., Kriebitzsch W.U., Schlesselmann K., Weber M.S. and Hardtle W. (2012), Nitrogen deposition increases susceptibility to drought - experimental evidence with the perennial grass Molinia caerulea (L.), *Moench. Plant Soil*, **353(1–2)**, 59–71.
- Gao L., Su J., Tian Q., and Shen Y. (2020), Contrasting strategies of nitrogen absorption and utilization in alfalfa plants under different water stress, *Journal of Soil Science and Plant Nutrition*, **20(3)**, 1515-1523.
- Gou W., Zheng P.F., Tian L., Gao W., Zhang L.X., Akram N.A. and Ashraf M. (2017), Exogenous application of urea and a urease inhibitor improves drought stress tolerance in maize (*Zea mays L.*), *J. Plant Res*, **130**, 599–609.
- Killi D., Raschi A. and Bussotti F. (2020), Lipid Peroxidation and Chlorophyll Fluorescence of Photosystem II Performance during Drought and Heat Stress is Associated with the Antioxidant Capacities of C3 Sunflower and C4 Maize Varieties, International journal of molecular sciences, 21(14), 4846.
- Kocheva K., Lambrev P., Georgiev G., Goltsev V. and Karabaliev M. (2004), Evaluation of chlorophyll fluorescence and membrane injury in the leaves of barley cultivars under osmotic stress, *Bioelectrochemistry*, 63(1), 121-124.
- Li S., Zhou L., Addo-Danso S.D., Ding G., Sun M., Wu S. and Lin S. (2020), Nitrogen supply enhances the physiological resistance of Chinese fir plantlets under polyethylene glycol (PEG)-induced drought stress, *Scientific reports*, **10**(1), 1-8.
- Movludi A., Ebadi A., Jahanbakhsh S., Davarı M. and Parmoon G. (2014), The effect of water deficit and nitrogen on the antioxidant enzymes' activity and quantum yield of barley (Hordeum vulgare L.)., *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 42(2), 398-404.
- Nazir U., Anjum S.A., Farooq A., Nawaz U., Nawaz M., and Samiullah M. (2017), Climate change and agricultural system: potential impacts and soil management strategies - a review, *Journal of Agricultural Research* (03681157), 55(3).
- Pishva Z.K., Amini-Dehaghi M., Bostani A. and Naji A.M. (2020), Biological and chemical nitrogen fertilizer

impact on cumin (*Cuminum cyminum* L) under different irrigation regimens, *Journal of Herbmed Pharmacology*, **9**(**3**), 209-217.

- Quero J.L., Villar R., Maranon T. and Zamora R. (2006), Interactions of drought and shade effects on seedlings of four Quercus species: physiological and structural leaf responses, *New Phytol*, **170**, 819–833.
- Song J., Wang Y., Pan Y., Pang J., Zhang X., Fan J., and Zhang Y. (2019), The influence of nitrogen availability on anatomical and physiological responses of *Populus alba*×P. glandulosa to drought stress, *BMC plant biology*, **19**(1), 1-12.
- Stirbet A. (2011), On the relation between the Kautsky effect (chlorophyll a fluorescence induction) and photosystem II: basics and applications of the OJIP fluorescence transient, *Journal of Photochemistry and Photobiology B: Biology*, **104(1-2)**, 236-257.
- Wang X., Wang L. and Shangguan Z. (2016), Leaf gas exchange and fluorescence of two winter wheat varieties in response to drought stress and nitrogen supply, *PLoS One*, **11**(**11**), e0165733.
- Wang Y., Huang Y., Fu W., Guo W., Re, N., Zhao Y. and Ye Y. (2020), Efficient physiological and nutrient use efficiency responses of maize leaves to drought stress under different field nitrogen conditions, *Agronomy*, **10**, 523.
- Waraich E.A., Ahmad R. and Ashraf M.Y. (2011), Role of mineral nutrition in alleviation of drought stress in plants, *Australian Journal of Crop Science*, 5(6), 764-777.
- Wu F., Bao W., Li F. and Wu N. (2008), Effects of drought stress and N supply on the growth, biomass partitioning and water-use efficiency of Sophora davidii seedlings, *Environ Exp Bot*, 63, 248–255.
- Xu F., Guo W.H., Wang R.Q., Xu W.H., Du N. and Wang Y.F. (2009), Leaf movement and photosynthetic plasticity of black locust (*Robinia pseudoacacia*) alleviate stress under different light and water conditions, *Acta Physiol Plant*, **31**(3), 553–563.
- Xu N., Guo W., Liu J., Du N. and Wang R. (2015), Increased nitrogen deposition alleviated the adverse effects of drought stress on *Quercus variabilis* and *Quercus mongolica* seedlings, *Acta Physiologiae Plantarum*, **37(6)**, 107.
- Zhuang J., Wang Y., Chi Y., Zhou L., Chen J., Zhou W., Song J., Zhao N. and Ding J. (2020), Drought stress strengthens the link between chlorophyll fluorescence parameters and photosynthetic traits, *PeerJ* 8:e10046 https://doi.org/10.7717/peerj.10046.
- Zlatev Z.S. and Yordanov I.T. (2004), Effects of soil drought on photosynthesis and chlorophyll fluorescence in bean plants, *Bulg J Plant Physiol*, **30**(**3-4**), 3-18.