

STUDIES REGARDING THE INFLUENCE OF NITROGEN FERTILIZING DOSE ON SOME ECOPHYSIOLOGICAL PARAMETERS FOR *TRITICUM AESTIVUM*

Adina-Daniela DATCU^{1,2*}, F. SALA¹

¹Banat's University of Agricultural Sciences and Veterinary Medicine „King Michael I of Romania” from Timișoara, Soil Science and Plant Nutrition

²West University of Timișoara, Faculty of Chemistry, Biology, Geography, Biology-Chemistry Department

*E-mail address: dana_datcu19@yahoo.com

Abstract. This paper presents data obtained from a physiological study conducted in the spring of 2018 on *Triticum aestivum* spp. vulgare, Ciprian cv. The species was cultivated on a slightly gleized cambic chernozem specific for Banat Area, also found in the Didactic Station of BUASVM Timișoara, Romania. Due to the large scale use and different doses and combinations of chemical fertilizers in the agricultural practice, we chose to fertilize the wheat with nitrogen in five experimental variants: 0, 50, 100, 150 and 200 kg active substance ha⁻¹. Balancing the N rate, water use efficiency and yield are important problems in our country. Thus a proper use of fertilizers in general, and N in particular became a necessity. The plants were collected and transported in the laboratory where the leaves were separated using a scalpel. These were weighted, dried and incinerated. Some gravimetric parameters including leaves fresh weight, dry weight and ash content were determined. Next, organic and mineral content, but also OC / MC ratio were calculated. FW of the leaves increased proportionally with N dose. Leaf fresh weight is directly impacted by technological factors like N fertilizer rates. Consequently, regarding this parameter, significant differences between the five variants were observed, FW being significantly higher for N 200 probes when compared with other leaves from the plots fertilized with a lower amount of N. Same trend was observed for DW and AC, respectively. MC values varied between the N doses. From this point of view, the smallest mineral content in leaves was obtained for N 100 and N 150 variants, suggesting a possible remobilization of minerals in wheat plant with respect to N dose. Regarding OC, the maximum value was obtained for N 200. Despite the fact that OC and MC varied without a clear trend, OC / MC ratios had followed a clear polynomial increase together with the increase of N dose, varying between 4.9236 for N 0 and 10.2368 for N 200 variant. ANOVA revealed some significant differences. AC was the only parameter with homogenous data, FW, DW, OC, MC and OC/MC ratio being heterogeneous.

Keywords: OC/MC ratio, mineral content, wheat leaves, N fertilization, organic content.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is grown worldwide under diverse climatic conditions (CZYCZYŁO-MYSZA et al., 2018). It is a crop of major importance and together with other staple cereals supply the bulk of calories and nutrients in the diets of a large proportion of the world population (CAKMAK, 2008).

Fertilizers play a significant role in obtaining agricultural yield, with a variable contribution which vary between 40-60% depending on the crop, the climatic conditions and agricultural technology (STEWART et al., 2005). Grain yield is a particularly complex trait, being the end product of many processes in the plant and, in consequence, is very environmentally dependent (QUARRIE et al., 2005). Between all metabolic elements which plants use from soil, nitrogen is needed in the largest amounts (TUCKER, 2004; BOJOVIĆ AND MARKOVIĆ, 2009). High grain yield represents a higher crop nitrogen (N) requirement in total (CASSMAN et al., 2002). TILMAN et al. (2001) forecasted that global N fertilizer consumption

would need to increase by 2.7 times by 2050 to meet the food demand. Ammonium azotate is a fertilizing substance used in Romania in a wide range of areas (SALA, 2011). It is quite cheap when compared with NPK, this being a reason for the extended use of it by farmers.

N fertilization is well known to improve soil fertility (MALHI et al., 2011). However, using excessive N fertilizer can decrease the N utilization rate, which not only causes a huge waste of resources and economic losses, but can also adversely impact the environment (SCHINDLER AND HECKY, 2009; HVISTENDAHL, 2010). Due to the fact that all of fertilizers (mostly ammonium nitrate) are well soluble in water, only 40-50% of the introduced fertilizer is assimilated by plants, 20% is washed-off into soil, which conditions a global contamination of the environment (rivers, seas, water reservoirs, ground waters) (GUGAVA AND KOROKHASHVILI, 2018).

Balancing the N rate, water use efficiency and yield is an important problem in farming systems (ZHONG AND SHANGGUAN, 2014; SALA et al., 2016). In addition, world food demand is expected to double during 2005–2050 (BORLAUG, 2009), thus it is important to increase food production with lower water use, particularly in water shortage regions (ZHONG AND SHANGGUAN, 2014). The small net changes in soil-N during the course of the long-term experiments are consistent with the fact that a near steady-state is typically achieved in fields under a continuous cropping regime (JOHNSTON et al., 2009). Nitrogen nutrition played an important role on growth, yield components and grain quality of cereal crops (MAQSOOD et al., 2014). On the other the hand, balanced fertilizing is the application of essential plant nutrients in the right proportion and in optimum quantity for a specific crop condition (SHARMA et al., 2015).

The aim of this research was to compare leaves fresh and dry weights, ash content, as gravimetric parameters, but also to calculate and determine if significant differences could be found for organic and mineral contents. Moreover the ratio between the last two parameters was also of interest.

MATERIALS AND METHODS

The biological material to be analyzed was represented by samples belonging to the *Triticum aestivum* (L.) ssp *vulgare*, *Ciprian* cv. The wheat was cultivated in the Didactic Station of BUASVM Timișoara, Romania. The specific soil in Banat Area can be characterized as a slightly gleized cambic chernozem with neutral reaction (pH = 6.95-7.1) (PIȘLEA AND SALA, 2012).

The plots were fertilized with nitrogen in five variants: N 0, N 50, N 100, N 150, N 200 kg active substance ha⁻¹. The samples were collected in May 2018 during the morning.

An analytical balance (Kern model) with precision of 0.0001 g was used for all mass measurements. After separating the leaves, fresh weight was measured (FW). Afterwards, all the samples were dried in an oven (Sauter model) at 100 °C for 2 hours and weighed, resulting dry weight (DW). Dried probes were incinerated to ash in a furnace (Nabertherm model) at 500 °C for 2 h. After completion of the cooling process, the mass of the ash content (AC) was determined. Ash content is the inorganic residue left after burning the organic matter and it is the reflection of the mineral content of biomass. Organic matter was calculated by subtracting crude ash weight from dry matter weight (IANOVICI, 2016). Using this parameter OC was determined, as following:

$$OC = (100 \times OM) / FW.$$

MC was determined using next formula:

$$MC = (100 \times AC) / FW.$$

OC / MC ratio was also determined.

Statistical processing was realized using Microsoft Office Excel 2013 and PAST Software 3.20 (HAMMER ET AL., 2001). Probability (p) values less than 0.05 were considered significant. Data had a normal distribution. Variance between the five experimental variants was analyzed by one-way ANOVA between groups, Levene's test for homogeneity of variance (based on means) and Welch F test in the case of unequal variances (CIOBANU, 2016).

RESULTS AND DISCUSSION

FW presented the lowest values for the probes fertilized with N 0. The mean value of this parameter was 0.0374 g (Figure 1). As the N dose increased, FW of the leaves also increased ($r^2 = 0.9885$). The highest mean value of FW was obtained for N 200 samples (0.3731 g). At the maximum level of fertilization, the leaves were almost 10 times bigger when compared to the ones from the N 0 variant.

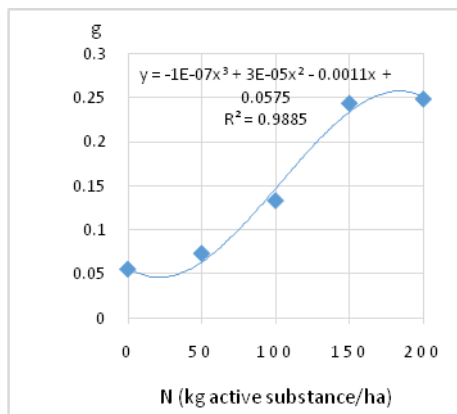


Figure 1. N dose influence on FW

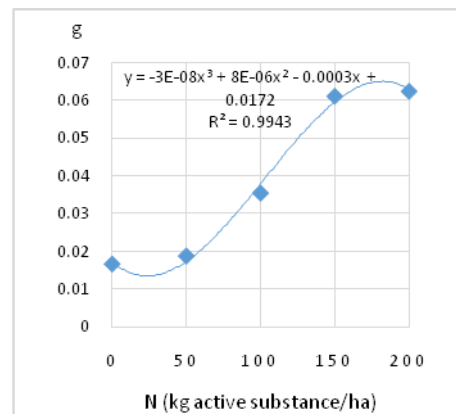


Figure 2. N dose influence on DW

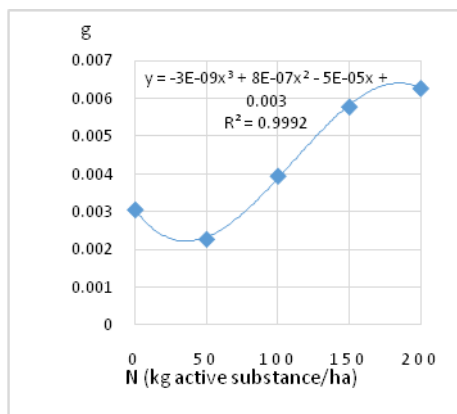


Figure 3. N dose influence on AC

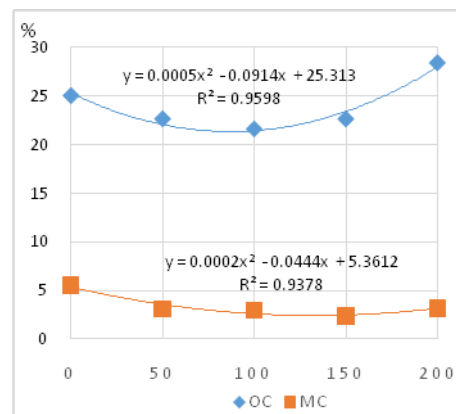


Figure 4. N dose influence on OC and MC

Same trend was observed for DW ($r^2 = 0.9943$) (Figure 2). The lowest mean value was obtained for N 0 probes (0.01679) and the highest mean value was recorded for N 200 leaves (0.06247).

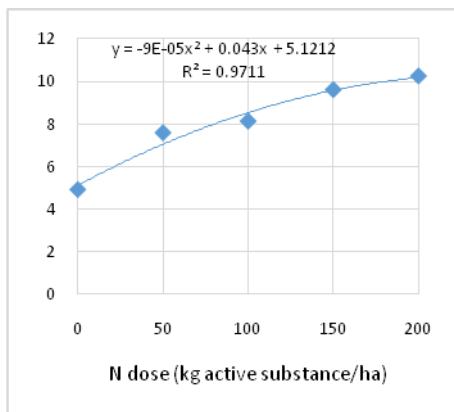


Figure 5. N dose influence on OC/MC ratio

Minimum and maximum values of the studied parameters determined for each experimental variant, but also the results obtained after the completion of analysis of variance, with significant values being in bold, can be seen in Table 1.

Table 1.

Analysis of variance for FW, DW and AC calculated for wheat leaves for the five experimental variants

		N 0	N 50	N 100	N 150	N 200	Levene's test	One-way ANOVA	Welch F test
FW - in g	Min	0.0374	0.0579	0.0738	0.1812	0.1684	0.002616		F=54.11, df=20.8, p=1.066E-10
	Max	0.0667	0.092	0.1967	0.3732	0.3731			
DW - in g	Min	0.0112	0.0133	0.0206	0.0402	0.0427	0.002766		F=39.3, df=21.53, p=1.334E-09
	Max	0.0274	0.0288	0.044	0.0945	0.0898			
AC - in g	Min	0.0016	0.0013	0.0023	0.0037	0.0038	0.1458	F = 18.8, p= 3.79E-09	
	Max	0.0055	0.0033	0.0054	0.0075	0.009			
OC	Min	18.1667	20.7006	19.4204	19.8124	21.0747	0.009181		F=1.028, df=21.77, p= 0.4152
	Max	34.3939	27.8689	27.7311	25.0579	28.3658			
MC	Min	3.1873	2.2453	2.3233	1.8820	2.2565	1.44E-07		F =9.63, df=21.84, p=0.0001204
	Max	9.9819	4.1401	3.2936	2.8902	3.1293			
OC/MC ratio	Min	2.7091	5	7.0741	7.4154	7.5111	0.0385		F= 19.52, df = 20.8, p= 5.505E-07
	Max	7.1875	10.8125	10	11.9495	10.2368			

AC had the lowest average for N 50 probes (0.00228) and the highest average for N 200 probes (0.00627). A polynomial increase of ash content was noticed proportionally with N dose ($r^2 = 0.9992$) (Figure 3).

The data were homogeneous only for AC. Analysis of variance revealed that the data are significantly different for this parameter ($p = 3.79E-09$). The Tukey's pairwise comparisons for AC indicated significant differences for samples: N 0 – N 150 ($p = 0.0001405$), N 50- N

150 ($p = 1.31E-06$), N 100 – N 150 ($p = 0.01699$), N 0 – N 200 ($p = 7.63E-06$), N 50- N 200 ($p = 6.66E-08$), N 100 - N 200 ($p = 0.001356$), N 50 – N 100 ($p = 0.0356$). Thus, a significant increase was noticed for this parameter proportionally with the mass of active substance used.

OC values presented a polynomial trend ($r^2 = 0.9598$) (Figure 4). This parameter had the lowest values for medium fertilized variants. MC had the highest values for N 0 variant, but a trend was also observed ($r^2 = 0.9378$) (Figure 4). Regarding organic content, this parameter showed variations on N gradient. The highest value was obtained for N 200 probes and the lowest for N 100 probes. MC presented only few variations. A higher mineral deposition in leaves was observed for N 0 samples.

When the data were heterogeneous, we applied Welch F test. In this case, F was significantly different for FW, DW, MC and OC/MC ratio. Regarding OC, no significantly differences were noticed after the completion of Welch test (Table 1).

OC/MC ratio is a parameter which presented a trend proportionally with the N gradient ($r^2 = 0.9711$) (Figure 5). The lowest value for this parameter was for N 0 probes (OC/MC = 4.9236) and the maximum was reached for N 200 probes (OC / MC = 10.23680). Thus, for the fertilized wheat samples OC has the biggest value and MC the smallest, when compared to OC. Significant increase of this parameter was observed when the N dose increased.

CONCLUSIONS

This study aimed to determine some gravimetric and physiological parameters for leaves samples from *Triticum aestivum* subsp. *vulgare*, *Ciprian* cv. FW, DW and AC were determined after collecting, drying and incinerating wheat leaves. Significant differences were obtained between variants. These parameters increased proportionally with the N gradient. ANOVA test revealed that only AC samples were homogenous. For this parameter one-way ANOVA results were taken into consideration. FW, DW, OC, MC and OC/MC ratio data were heterogeneous. OC and MC presented different trends, but OC/MC ratio increased proportionally with the N fertilizers amount.

ACKNOWLEDGEMENTS

The authors wish to thank Didactic Station of BUASVM Timișoara for providing equipment and support.

We also thank SCDA Lovrin for the wheat seed and West University of Timișoara, Faculty of Chemistry, Biology and Geography for research equipment and tools.

BIBLIOGRAPHY

- BOJOVIĆ B., MARKOVIĆ A. 2009. Correlation between nitrogen and chlorophyll content in wheat (*Triticum aestivum* L.). *Kragujevac J. Sci.* 31: 69-74.
- BORLAUG N.E. 2009. Foreword. *Food Sec.* 1: 1–11.
- CAKMAK I. 2008. Enrichment of cereal grains with zinc: agronomic or genetic biofortification? *Plant and Soil* 302, 1–17.
- CASSMAN K.G., DOBERMANN A., WALTERS D.T. 2002. Agroecosystems, nitrogen-use efficiency, and nitrogen management. *Am-bio.* 31: 132–140.
- CZYCZYŁO-MYSZA I.M., MARCIŃSKA I., SKRZYPEK E., BOCIANOWSKI J., DZIURKA K., RANČIĆ D., RADOŠEVIĆ R., PEKIĆ-QUARRIE S., DODIG D., QUARRIE S.A. 2018. Genetic analysis of water loss of excised leaves associated with drought tolerance in wheat. *PeerJ* 6:e5063.
- CIOBANU L.A. 2016. *Bellis perennis* - variations of physiological responses in urban conditions. *Annals of West University of Timișoara, ser. Biology*, 2016, vol. 19 (1): 77-86.

- GUGAVA E., KOROKHASHVILI A. 2018. Technologies for obtaining nitrogen fertilizers prolonged effect in wheat. *Annals of Agrarian Science* 16: 22-26.
- HAMMER Ø. HARPER D.A.T., RYAN P.D. 2001. PAST: Paleontological Statistics software package for education and data analysis. *Paleontologica Electronica* 4 (1): 9 pp.
- HVISTENDAHL M. 2010. China's push to add by subtracting fertilizer. *Science* 327: 801–801.
- IANOVICI N. 2016. *Taraxacum officinale* (Asteraceae) in the urban environment: seasonal fluctuations of plant traits and their relationship with meteorological factors. *Acta Agrobot.* 69 (3): 1677.
- JOHNSTON A.E., POULTON P. R., COLEMAN K. 2009. Soil organic matter: its importance in sustainable agriculture and carbon dioxide fluxes. *Adv. Agron.* 101: 1–57.
- QUARRIE S.A., STEED A., CALESTANI C., SEMIKHODSKII A., LEBRETON C., CHINOY C., STEELE N., PLJEVLJAKUSIĆ D., WATERMAN E., WEYEN J., SCHONDELMAIER J., HABASH D.Z., FARMER P., SAKER L., CLARKSON D.T., ABUGALIEVA A., YESSIMBEKOVA M., TURUSPEKOV Y., ABUGALIEVA S., TUBEROSA R., SANGUINETI M-C., HOLLINGTON P.A., ARAGUÉS R., ROYO A., DODIG D. 2005. A high-density genetic map of hexaploid wheat (*Triticum aestivum* L.) from the cross Chinese Spring SQ1 and its use to compare QTLs for grain yield across a range of environments. *Theor Appl Genet* 110: 865–880.
- MALHI S., NYBORG M., GODDARD T., PUURVEEN D. 2011. Long-term tillage, straw management and N fertilization effects on quantity and quality of organic C and N in a Black Chernozem soil. *Nutr Cycl Agroecosys* 90 (2): 227–241.
- MAQSOOD M., SHEHZAD M.A., RAMZAN Y., SATTAR A. 2014. Effect of nitrogen nutrition on growth, yield and radiation use efficiency of different wheat (*Triticum aestivum* L.) cultivars. *Pak. J. Agri. Sci.* 51:451-458.
- PÎSLEA D., SALA F. 2012. Changes in soil reaction under the influence of mineral fertilization. *Research Journal of Agricultural Science* 44 (3): 102-107.
- SALA F., 2011. *Agrochimie*. Ed. Eurobit, Timisoara, pp 249-267.
- SALA F., RUJESCU C., CONSTANTINESCU C. 2016. Causes and solutions for the remediation of the poor allocation of P and K to wheat crops in Romania. *AgroLife Scientific Journal* 5(1): 184-193.
- SCHINDLER D., HECKY R. 2009. Eutrophication: more nitrogen data needed. *Science* 324: 721–722.
- SHARMA V.K., PANDEY R.N., SHARMA B.M. 2015. Studies on long term impact of STCR based integrated fertilizer use on pearl millet (*Pennisetum glaucum*) - wheat (*Triticum aestivum*) cropping system in semi arid condition of India. *Journal of Environmental Biology* 36(1): 241-247.
- STEWART W.M., DIBB D.W., JOHNSTON A.E., SMYTH T.J. 2005. The contribution of commercial fertilizer nutrients to food production. *Agron. J.* 97: 1-6.
- TILMAN D., FARGIONE J., WOLFF B., D'ANTONIO C., DOBSON A., HOWARTH R., SCHINDLER D., SCHLESINGER W.H., SIMBERLOFF D., SWACKHAMER D. 2001. Forecasting agriculturally driven global environmental change. *Science*. 292: 281–284.
- TUCKER M. 2004. Primary Nutrients and Plant Growth. - In: *Essential Plant Nutrients* (SCRIBD, Ed.). North Carolina Department of Agriculture.
- ZHONG Y., SHANGGUAN Z. 2014. Water consumption characteristics and water use efficiency of winter wheat under long-term nitrogen fertilization regimes in Northwest China. *PLOS One* 9 (6): e98850.