Bulletin of University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca. Horticulture
Open Access

EFFECT OF FERTILIZERRS WITH UREASE AND NITRIFICATION INHIBITORS ON DURUM WHEAT CROP ON YIELD AND QUALITY (*Triticum turgidum* L. subsp. durum)

Antigolena FOLINA[1], Ioanna KAKABOUKI[1], Ioannis ROUSSIS[1], Antonis MAVROEIDIS[1], Stella KARYDOGIANNI[1], Varvara KOYNELI[1], Aspasia EFTHIMIADOU[1], Nikolaos KATSENIOS[1], Vasilis TSOUKANAS[2], Dimitrios BILALIS [1*]

[1]*Department of Crop Science, Laboratory of Agronomy, Agricultural University of Athens, Greece*
[2] *EuroChem Hellas S.A.*
* *Corresponding author: D. Bilalis e-mail: bilalis@aua.gr*

RESEARCH ARTICLE

Abstract

Conventional agricultural has a significant role in climate change. For this reason, farmers choose more innovative practices such as fertilizers inhibitors. Durum wheat (*Triticum turgidum* L. subsp. durum) is the most cultivated winter crop in the Mediterranean basin. The scope of this study is to determine the improvement of the fertilizer yield by adding nitrification (DMPSA) and urease (NBPT) inhibitor in urea in durum wheat crop. Meridiano variety was evaluated for one growing period through 2019–2020 under two basic fertilization (20-20-0 and 12-40-0 (+10S +Zn). The experiments were designed according to split-plot design, 2 main plots (basic fertilization) and 7 subplots (top fertilization). The top fertilization were the various urea combination treatments (urea, urea + urease inhibitor thiophosphoric-triamide (NBPT) (UI), urea + nitrogen inhibitor 3,4-dimethylpyrazole succinic (DMPSA) and control. Nitrogen markers, such as nitrogen use efficiency (NUE), nitrogen harvest index and nitrogen agronomic efficiency (NAE) were used to evaluate nitrogen release. The length of the inflorescences was over 20 cm for all fertilizations. Regarding basic fertilization, larger inflorescences were recorded with 12-40-0 (+10S +Zn). The grain protein content and nitrogen were higher by 1-2% under basic basic fertilization 20-20-0. Grain and biomass production were increased with both fertilizers' inhibitors (NBPT and DMPSA). Between two inhibitors, urease inhibitor (NBPT) yielded higher than DMPSA.

Keywords: durum wheat; DMPSA; NBPT; nitrogen management; NUE; NAE.

Received: 8 October 2021
Accepted: 22 October 2021
Published: 15 November 2021

DOI:
15835/buasvmcn-hort:2021.0029

INTRODUCTION

In the coming years, global population growth is projected to escalate. FAO estimates that the world population in 2050 will exceed 9.1 billion, up from 7.8 billion todays. As a consequence, in order to meet the food needs that will arise, the total food production should be increased by 70%, the annual grain production by 42% and the annual animal production by 135% (Trostle et al., 2010· Hertel et al., 2011·Valizadeh et al., 2020). Therefore, the improvement of agricultural sustainability in the future will depend on the optimization of crop production, the more efficient use of arable land, the more environmental conservation. Wheat (*Triticum spp*) is the third most important cereal crop in the world, as it is a staple in the diet of at least 36% of the world's population. Nowadays, the cultivated areas with wheat surpass any other crop (Fatima et al., 2014). Basic fertilization in the fall, before or during sowing, aims to meet the needs of the plants up to the twinning stage. The requirements of wheat up to this stage in nitrogen reach 4 Kg

N str-1 (from 12-16 Kg N str-1) that the crop needs to complete its biological cycle) (Katsenios et al, 2016). The high nitrogen absorption in cereals is located from the stage of twinning until the end of flowering, stages in which the plant forms the structures of its production (formation of siblings, elongation of stem, leaf area intensity, flowering, number and size of grains in the ear). Significant nitrogen deficiency can lead to a reduction in leaf area index and leaf area growth duration and consequently to a lower uptake of sunlight and lower photosynthetic rates (Fageria et al.2003). That is why it is necessary to use slow release fertilizers (Folina et al., 2021).

Nitrogen use efficiency (NUE) has been described in many different ways (Cormier et al., 2013), but the most accepted definition is the production of grain or biomass, per unit of available nitrogen (N) in the soil (including of residual N in the soil and N inputs through fertilization), or otherwise the economically maximum production per unit of nitrogen applied, absorbed, or used by the plant to produce seed or tissue (Moll et al., 1982).

The NUE is the net result of N binding (uptake efficiency) and its conversion (utilization efficiency) (Moll et al., 1982; Sylvester-Bradley & Kindred, 2009). Not only helps to minimize production costs in wheat crops (up to 40%) (Bock et al., 1984) with fewer fertilizer applications but also to reduce the environmental impact of the use of nitrogen fertilizers (Good et al., 2004).

Nitrogen Agronomic Efficiency (NAE) is related to the ability of the crop to increase its economic yield (seed/ grain yield) in relation to the amount of applied nitrogen. This indicator therefore describes the effectiveness of fertilization in the production of grain / kg N fertilizer.

The experiment aims to make sustainable use of applied nitrogen through urease and nitrification inhibitors though evaluation of nitrogen indices. There is limited research about the effect of new types of nitrogen fertilizers on durum wheat crop. The main goal of this study is to evaluated the effect of nitrogen fertilization on the agronomic features of durum wheat under semiarid Mediterranean conditions.

MATERIALS AND METHODS

The trial was carried out in central Greece, area of Aliartos. The area is located at 95 m above sea level and the coordinates are 38 ° 23'52 "N, 23 ° 6'35" E during 2019-2020. The soil is alkaline and is distinguished by a satisfactory content of organic matter (3.47%). The soil pH is 8, EC 1,11 mS/cm, P 21,25 mg kg^{-1} soil, $CaCO_3$ 11,38% and K 319,09 mg kg^{-1}. A soil sampling was performed before sowing throughout the experiment. Soil was collected with the soil extractor at a depth of 30 cm from the soil surface. The cylinder had a diameter of 20 cm. The soil analysis was performed at the Agricultural University of Athens. Temperatures and precipitation in growing period are reported in Figure 1. This year was characterized by high temperatures and low rainfall in the last stage of the crop cycle. The total precipitation was 426,7 mm.

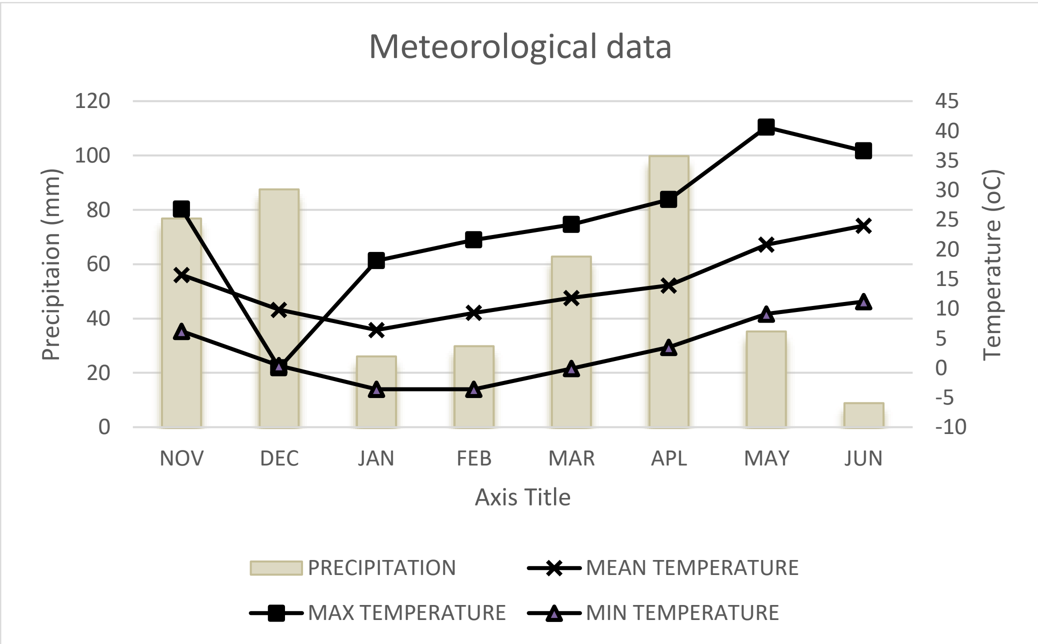

Figure 1. Meteorological data at the experimental area for growing season 2019-2020.

The effect of two basic and 7 top fertilizations were investigated in Meridiano variety (the characteristics of which are presented in the table below (Table 1). The ground fertilization factor was set in large plots, and top fertilization in randomized subplots (Table 2) (4 m wide × 5 m long). The previous crop was cotton. Sowing took place in late November (19/11/2019) at a density of 300 seeds m^{-2} and harvest 213 days after sowing (DAS). Basal fertilization was applied one day before and top fertilization was applied 64 DAS.

The experimental design was split-plot, where the whole-plot factor was two different ground fertilization types (CROPLEX 12-40-0 + 10S, NP 20-20-0) and the split-plot factor was seven different top fertilization (Urea 46% N, Urea 46% N + NBPT, Urea 40% N, Urea 40% N + NBPT, Can 27% N, Can 27% N + DMPSA and control). The fertilizer Can 27% is a quality granular calcium ammonium nitrate-based nitrogen fertilizer. The design had 2 whole plots (560 m^2 each), each of which was split into subplots with two varieties (20 m^2) with four replications. The total applied fertilizer dose was 160 kg N ha^{-}; 100 kg N ha^{-1} as a basal dressing and 60 kg N ha^{-a} as a top dressing. The nitrification inhibitor was 3,4-Dimethylpyrazole-succinic acid (DMPSA) and urease inhibitor was N-(n-butyl) thiophosphoric triamide (NBPT).

Table 1. Traits of Meridiano variety

Vareity characteristics	
Genealogy	Simeto / WB881 / Duilio / F21
Registered	1999
Property	ALFA SEEDS SA
Plant characteristics	
Head emerge	Middle
Height	Medium
Hood color	Light brown
Production potential	Very high
Grain quality characteristics	
Specific weight	High
Yellow color indicator	(b Minolta) 24-26
Protein content	Medium - high
Gluten quality	Good
Resistance to disease- conditions	
Rust, septoria, mildew	Good
Cold, lodging	Very good

Table 2. Fertilization treatments, ground/ basic (CROPLEX 12-40-0 + 10S and NP 20-20-0) and top fertilizers (Urea 46% N, Urea 46% N + NBPT, Urea 40% N, Urea 40% N + NBPT, Can 27% N, Can 27% N + DMPSA, Control)

Fertilization	
Ground	**Top**
CROPLEX 12-40 -0 + 10S	Control
	Urea 46% N
	Urea 46% N + NBPT
	Urea 40% N
	Urea 40% N + NBPT
	Can 27% N
	Can 27% N + DMPSA
NP 20-20-0	Control
	Urea 46% N
	Urea 46% N + NBPT
	Urea 40% N
	Urea 40% N + NBPT
	Can 27% N
	Can 27% N + DMPSA

Note: Analysis of variance on data was set up using the Statistic (Stat Soft, 2011) logistic package as a Completely Randomized Design. Differences between means were separated using the LSD test, at the 5% level of significance ($p < .05$).

The below nitrogen indices were calculated in order to be measured the nitrogen efficiency.

$$NUE = \frac{[\{Nuptake\ of\ fert\ plot\ (kg) - Nuptake\ of\ unfert\ plot\ (kg)\}]}{applied\ N\ fertilizer\ (kg)}$$

$$NAE = \frac{[\{yield\ of\ fert\ plot\ (kg) - yield\ of\ unfert\ plot\ (kg)\}]}{quant\ of\ N\ applied\ (kg)}$$

RESULTS AND DISCUSSIONS

Plant height was significantly affected by fertilization. Higher plants were noticed under the ground fertilization with Sulphur. The highest plant height for both conventional and fertilizers with nitrification and urease inhibitor is observed in the treatments with CAN fertilizers, however between the treatments CAN 27% N and CAN 27% N + DMPSA. The urease inhibitor at 40 nitrogen units did not have statistically significant effects with basic sulfur fertilization. In contrast, in urea surgeries with 46% N there is a significant difference with urea surgery 46% N (95.88 cm) showing higher height than urea plants 46% N + NBPT (92.13 cm) (Table 3).

For the basic 20-20-0 fertilization the dry weight reached levels between 565 Kg ha^{-1} (Control) and 1052 Kg ha^{-1} (Urea 40% N + NBPT), and there were statistically significant differences between the treatments Urea 46% N + NBPT (800 Kg ha^{-1}) and Urea 40% N + NBPT (1052 Kg ha^{-1}) as well as between Can 27% N (870 Kg / ha) and Can 27% N + DMPSA (1044 Kg ha^{-1}), while no significant differences were observed between Urea 40% N + NBPT (1052 Kg ha^{-1}) and Can 27% N + DMPSA (1044 Kg ha^{-1}) (Table 3). For basic CROPLEX 12-40-0 + 10S fertilization, the maximum dry weight is 1511,837 kg ha^{-1} (Urea 40% N + NBPT).

This result agrees with (Bahmanyar & Ranjbar, 2008) that nitrogen increases plant height and dry weight. On the other hand, with the use of nitrogen inhibitor was given to the plants gradually. The plant did not take advantage of the extra nitrogen units given, probably because all the nitrogen units were too high. In the present study, number of leaves was significantly affected from both kind of fertilizations (basic and top).

Table 3. Height, dry weight and number of leaves as effected by fertilizer treatments.

Ground	Top	Height 183 DAS (cm)	Dry weight 183 DAS (kg ha^{-1})	Number of leaves 161 DAS
CROPLEX 12-40-0 + 10S	Control	90,677^{a}	565,087^{a}	7,37^{b}
	Urea 46% N	95,674^{b}	813,087^{b}	7,332^{b}
	Urea 46% N + NBPT	92,286^{a}	800,337^{c}	11,145^{c}
	Urea 40% N	94,567^{b}	929,087^{d}	8,395^{a}
	Urea 40% N + NBPT	95,708^{b}	1052,088^{e}	9,395^{a}
	Can 27% N	97,521^{c}	870,587^{f}	9,332^{a}
	Can 27% N + DMPSA	100,568^{c}	1044,088^{g}	8,707^{a}
NP 20-20-0	Control	81,005^{a}	892,087^{a}	5,457^{a}
	Urea 46% N	93,599^{b}	1320,088^{b}	6,832^{b}
	Urea 46% N + NBPT	94,661^{b}	1160,088^{c}	7,832^{c}
	Urea 40% N	90,005^{c}	1280,088^{d}	5,707^{a}
	Urea 40% N + NBPT	94,63^{b}	1511,837^{e}	5,707^{a}
	Can 27% N	88,005^{c}	1360,088^{f}	7,395^{c}
	Can 27% N + DMPSA	93,005^{b}	1248,338^{g}	6,707^{b}
F_{ground}		799,528***	2183551,794***	266,834***
F_{top}		267,242***	249118,985***	29,584***
$F_{ground*top}$		114,213***	24219,638***	8,123***

Means within a column followed by the different letters are significantly different at P=0.05 ('ns': not statistically significant; *: statistically significant for a significance level of p <0.05; **: statistically significant for a significance level of p <0.01;***: statistically significant for a significance level of p <0.001).

The spike length was significantly affected by top fertilization. Under 20-20-0 both inhibitors were enhanced spike length. Other the others hand, ground fertilization of CROPLEX 12-40-0 + 10S, there was not noticed differences between pairs of conventional and slow release fertilizers (Table 4).

The weight of 1000 grains in 213 DAS ranged between 45,175 g (Control) and 48,505 g (Can 27% N + DMPSA). After the statistical analysis, no significant differences were found between the operations Urea 46% N + NBPT

(48.1 g) and Can 27% N + DMPSA (48.54 g) and between Urea 40% N (47.81 g) and Can 27% N (47.50 g), while statistically significant differences were observed between Can 27% N (47.50 g) and Can 27% N + DMPSA (48.54 g) and between Urea 46% N + NBPT (48,10 g) and Urea 40% N + NBPT (47.90 g). Under basic fertilization 20-20-0, treatments Urea 46% N + NBPT and Can 27% N + DMPSA did not statistically differ. The 1000-seed weight with nitrification inhibitor was higher than conventional fertilizers (Table 4).

According to table 4, grain yield was significantly affected by top fertilization. Higher yields were revealed with Can 27% N + DMPSA for both basic fertilizers (5652,20 in NP 20-20-0 and 5665,00 CROPLEX in 12-40-0 + 10S) (Table 4).

Garrido-Lestache et al. (2005) investigated the effect of nitrogen fertilization level on durum wheat cultivation below Mediterranean regions (Spain). Grain yield significantly increased only between the control and the operation with 10 Kg N str^{-1}, while no significant increase was observed when the fertilization level rose to 15 and 20 Kg N str^{-1}.

Table 4. Spike length (with awns), 1000-seed weight and grain yield as effected by fertilizer treatments.

Ground	Top	Spike length 183 DAS (cm)	1000-seed weight 213 DAS (g)	Grain yield (kg ha^{-1})
CROPLEX 12-40-0 + 10S	Control	21,688^{a}	45,175^{a}	4616,67^{a}
	Urea 46% N	22,813^{b}	48,1^{c}	5506,67^{c}
	Urea 46% N + NBPT	22,906^{b}	48,175^{c}	5636,67^{d}
	Urea 40% N	23,175^{c}	47,708^{b}	5265,00^{b}
	Urea 40% N + NBPT	22,856^{b}	47,775^{b}	5425,00^{b}
	Can 27% N	24,706^{d}	47,475^{b}	5605,00^{d}
	Can 27% N + DMPSA	24,831^{d}	48,505^{c}	5665,00^{d}
NP 20-20-0	Control	20,300^{a}	44,175^{a}	4605,01^{a}
	Urea 46% N	21,145^{b}	47,85^{b}	5505,00^{c}
	Urea 46% N + NBPT	22,906^{b}	48,200^{c}	5625,00^{d}
	Urea 40% N	22,175^{c}	47,683^{b}	5215,80^{b}
	Urea 40% N + NBPT	23,706b	47,895b	541,5^{b}
	Can 27% N	23,87d	47,554b	561,523^{d}
	Can 27% N + DMPSA	24,831d	48,608c	565,220^{d}
F_{ground}		ns	ns	ns
F_{top}		19,636***	16,234***	5391,33***
$F_{ground*top}$		ns	ns	ns

Means within a column followed by the different letters are significantly different at P=0.05.('ns': not statistically significant; *: statistically significant for a significance level of p <0.05; **: statistically significant for a significance level of p <0.01;***: statistically significant for a significance level of p <0.001).

For the basic fertilization CROPLEX 12-40-0 + 10S the NUE index ranged from 0.14 (CAN 27% N) to 0.31 (Urea 46% N + NBPT). Statistically significant differences were observed both between CAN 27% N (0.14) and CAN 27% N + DMPSA (0.18), and between Urea 40% N (0.17) and Urea 40% N + NBPT (0.24). Also the difference between the operations Urea 46% N (0.24) and Urea 46% N + NBPT (0.31) is evaluated as statistically significant.

For the basic fertilization NP 20-20-0 the NUE index ranged from 0.23 (Urea 40% N + NBPT) and 0.3 (Urea 46% N + NBPT). For CAN 27% N (0.24) and CAN 27% N + NBPT (0.26) the difference is not evaluated as statistically significant. In contrast, differences were observed between both Urea 40% N (0.25) and Urea 40% N + NBPT (0.23) and between Urea 46% N (0.23) and Urea 46% N + NBPT (0.3) are evaluated as statistically significant.

For the basic fertilization CROPLEX 12-40-0 + 10S the NAE index ranged from 4.06 (Urea 40% N) to 6.58 (CAN 27% N + DMPSA). The difference between CAN 27% N (6.18) and CAN 27% N + DMPSA (6.58) is not evaluated as statistically significant, in contrast to the differences observed between Urea 40% N (4, 06) and Urea 40% N + NBPT (5.04) and between Urea 46% N (5.54) and Urea 46% N + NBPT (6.36).

For the basic fertilization NP 20-20-0 the NAE index ranged between 6.93 (Urea 40% N) and 10.56 (Urea 46% N + NBPT). The difference between CAN 27% N (7,28) and CAN 27% N + DMPSA (7,64) is not evaluated as statistically significant, in contrast to the differences observed between Urea 40% N (6, 93) and Urea 40% N + NBPT

(8.49) and between Urea 46% N (8.32) and Urea 46% N + NBPT (10.56).

According to NAE, urease and nitrification inhibitors application yielded 1 kg of extra grain. Optimization of inorganic nitrogen fertilization to increase nitrogen utilization efficiency (NUE) is a key issue and goal of applied research in agricultural systems. Nitrogen use efficiency in cereal crops worldwide has been estimated at around 33% (Raun & Johnson, 1999), ranging from 14 to 59% in wheat (Melaj et al, 2003; Lopez-Bellido et al., 2005), suggesting that the current strategies followed in nitrogen fertilization are extremely ineffective.

The efficiency of using nitrogen in cereals crop may be low due to the loss of nitrogen to the environment (evaporation, leaching, etc.) (Lu et al., 2021). To reduce these losses, fertilizer application should aim to match the plant's nitrogen requirement as much as possible with the available nitrogen in the soil (Heinemann, P., & Schmidhalter, 2021). Therefore, the application of nitrogen should be done during the growth stage that allows the plant to absorb it rapidly (Raun et al., 2008). Guardia et al. (2018) applied urea fertilizers with wheat with nitrification and / or urease inhibitors (12 Kg N str^{-1}). This study showed a small increase in seed yield (of the order of 1.48%) in the urea treatment with NBPT urease inhibitor compared to plain urea, while the grain nitrogen increased by 3.85% and the efficiency of use nitrogen (NUE) by 7.2% (Figure 2).

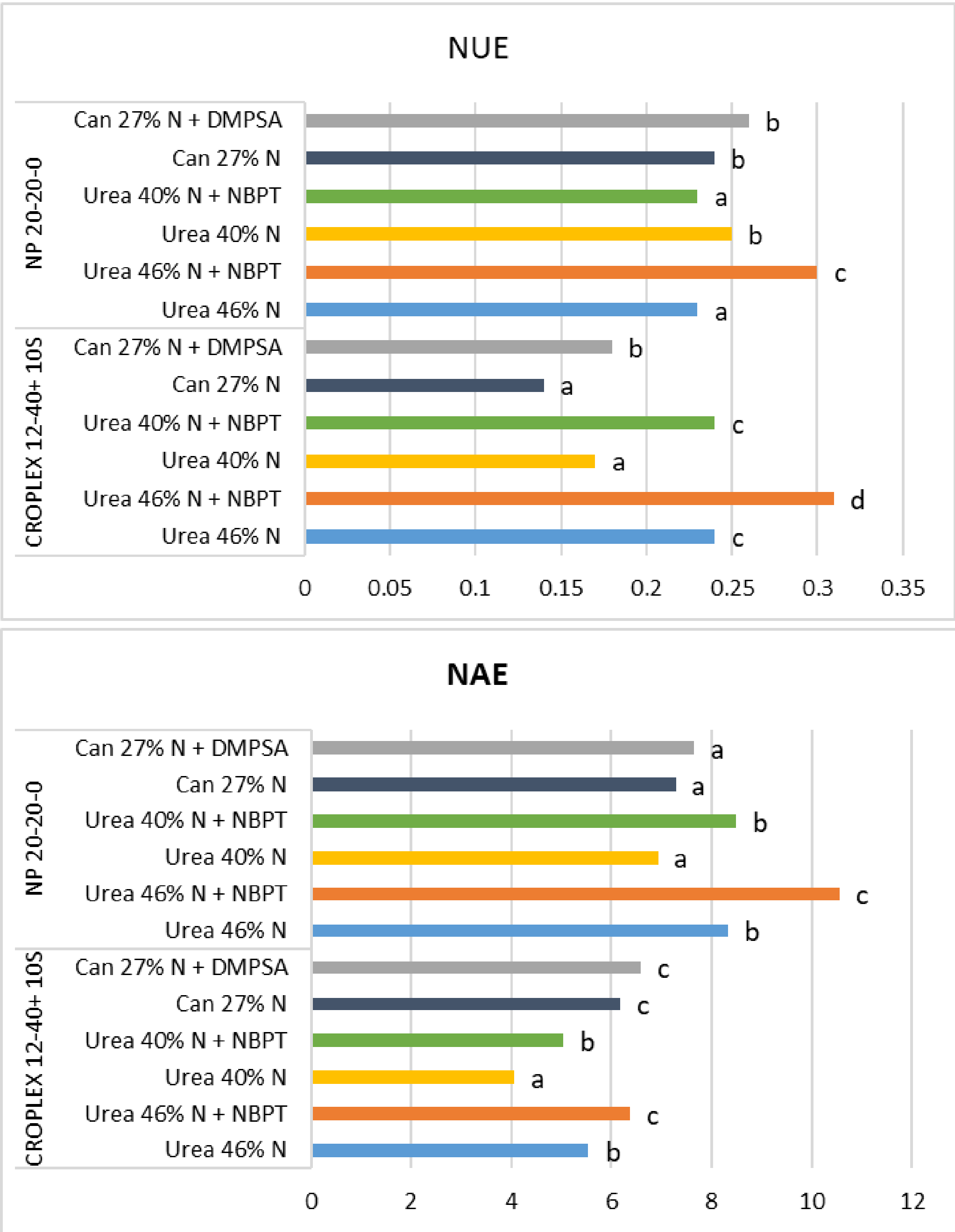

Figure 2. Nitrogen Use Efficiency and Nitrogen Agronomic Efficiency (NUE & NAE) for the top fertilizers

CONCLUSIONS

Regarding dry weight, it was observed higher under urea than inhibited fertilizers. Number of leaves was higher under CROPLEX 12-40-0 + 10S ground fertilization. The number of leaves is considered as a genetic trait and what's why the difference between the two ground fertilizations is up to two leaves. Spike length was 3% difference in favor of CROPLEX 12-40-0+ 10S was revealed under top fertilization of Urea 46% N + NBPT. Although yields were noticed higher under basic fertilization CROPLEX 12-40-0 + 10S, the nitrogen agronomic efficiency index (NAE) was higher values under a basic 20-20-0. The application of urea with inhibitor (Urea 46% N + NBPT) yield 6.36 more kg under basic fertilization CROPLEX 12-40-0 + 10S compared to 10.56 kg (NP 20-20-0) 4 more grain kg. To sum up, DMPSA (nitrification inhibitor) was performed better than NBPT (urease inhibitor) in durum wheat cultivation in Greek conditions.

Author Contributions:G.G., A.M., I.I., I.R., A.T.,D.B., V.K .Conceived and designed the analysis. G.G., A.M., I.I., I.R., A.T.,D.B., V.K.Collected the data; G.G., A.M., I.I., I.R., A.T.,D.B., V.K. Contributed data or analysis tools; .G., A.M., I.I., I.R., A.T.,D.B., V.K. Performed the analysis; .G., A.M., I.I., I.R., A.T.,D.B., V.K .Wrote the paper. All authors read and approved the final manuscript.

Funding Source: This research received funding from EuroChem Hellas S.A.

Acknowledgments

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflicts of Interest

The authors declare that they do not have any conflict of interest.

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