



Influence of Urea Fertilization with and without Inhibitors on Growth and Yield of Safflower (*Carthamus tinctorius* L.) under Different Tillage Practices

Panteleimon STAVROPOULOS^{1*}, Ioannis ROUSSIS^{1*}, Ioanna KAKABOUKI¹, Antonios MAVROEIDIS¹, Anastasios ZOTOS², Vassilios TRIANTAFYLLIDIS³, Chariklia KOSMA², Dimitrios BESLEMES⁴, Evangelia TIGKA⁴, Georgios PAPADOPOULOS¹, Aikaterini TSELA¹

¹ Laboratory of Agronomy, Department of Crop Science, Agricultural University of Athens, 11855 Athens, Greece ² Department of Biosystems and Agricultural Engineering, University of Patras, 30200 Mesolonghi, Greece ³ Department of Biosystems Administration of Food and Agricultural Enterprises, University of Patras, 30100 Agrinio, Greece

⁴ Hellenic Agricultural Organization Demeter, Institute of Industrial and Forage Crops, 41335 Larissa, Greece * Corresponding author: I.Roussis e-mail: iroussis01@gmail.com

RESEARCH ARTICLE

Abstract

Nitrogen fertilization is important for plant development. Because of the problems caused by urea, which is mainly used in nitrogen fertilizers, new types of fertilizers have inhibitors, that control the fertile disposal in soil. In addition, tillage practice is important in order to maintain soil productivity and prepare a good seedbed. This study aimed to evaluate the effects of implications of urea fertilizer with and without nitrification (MPA) and urease inhibitor (NBPT), and conventional and no-tillage systems on plant growth and yield of safflower (*Carthamus tinctorius* L.) crop. A field experiment was laid out in a split-plot design with four replications, two main plots (conventional and no-tillage system), and three sub-plots (control, urea with and without nitrification and urease inhibitors). The results indicated that fertilization significantly affected plant height, dry weight, seed yield and number of capitula and the highest values observed with urea with urease inhibitor. Tillage practice influenced the number of capitula and number of seeds per capitulum, and the higher numbers were found under conventional tillage. The findings of the present study imply that urea fertilizer with nitrification and urease inhibitors was very efficient and contributed a notable impact on the plant growth and yield of safflower.

Keywords: conventional tillage; nitrification inhibitor; no-tillage; plant development; urease inhibitor; seed yield.

Received: 18 October 2022 Accepted: 31 October 2022 Published: 15 November 2022

DOI: 15835/buasvmcn-hort:2022.0036

© 2022 Authors. The papers published in this journal are licensed under the Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License

INTRODUCTION

Safflower (*Carthamus tinctorius* L. Asteraceae) constitutes a versatile minor oilseed crop that can be utilized for edible oil production, as a vegetable crop, cut flower, forage crop for animal feed, industrial crop for dye production, as well as medicinal crop. The safflower's oil can be also used in biodiesel production (Golzafar et al., 2012; Bilalis et al., 2017). It is natively grown in Asia, middle East and Africa and has been cultivated in China, India, Iran and Egypt (Asgarpanah and Kazemivash, 2013). Nitrogen (N) fertilization is important for plant growth, development, and quality. The most common nitrogen fertilizer is urea. In order to avoid nitrogen losses due to ammonia volatilization caused by the use of ureabased fertilizers, a number of chemical compounds that can be added to urea to postpone the transition of N have been discovered. These slow-release products are classified as (a) urease and (b) nitrification inhibitors. Urease inhibitors

reduce NO₃⁻ and NH₄⁺ production in soil, slowing urea hydrolysis. Furthermore, the presence of urease inhibitors in the soil affects the efficacy of NH₃ loss management (Wang et al., 2020). Nitrification inhibitors have a significant effect on the enzymatic activity of NH₃ oxidizing bacteria, and their addition to urea delays the conversion of ammonium ions (NH₄⁺) to NO₃⁻, potentially lowering N₂O emissions from soil denitrification (Karydogianni et al., 2022). Several studies have found that adding urease inhibitors to urea reduces ammonia loss, increasing crop yield and N uptake compared to a single urea application (Karydogianni et al., 2020; Krol et al., 2020). Moreover, because N is a component of the chlorophyll structure, the addition of nitrification inhibitors enhances the chlorophyll concentration in the leaves, and hence the crop yield and quality (Wang et al., 2020).

In addition to fertilization, tillage practice is important, can alter the physical and chemical qualities of soil, resulting in changes in plant establishment, root growth, aerial cover, and, ultimately, crop yield. The most common tillage practice is conventional tillage, but it causes several problems such as erosion and loss of topsoil. To prevent these problems, practices like no-tillage is nowadays used in sustainable agriculture (Kakabouki et al., 2018).

The aim of this study was to evaluate the effects of using different types of fertilizers (control, urea, urea with inhibitors) and different type of tillage practices (conventional tillage and no tillage), on the plant growth and yield of safflower (*C. tinctorius* L.) crop.

MATERIALS AND METHODS

A field experiment was conducted at the Agricultural University of Athens (AUA) (Athens, Attica region, Central Greece; 37.983759' N, 23.701959' E, 29 m above sea level) from November 2020 to June 2021. The soil was a clay loam with pH (1:2 H₂O) 7.29, 104.3 ppm NO₃⁻, a limit - sufficient supply of phosphorus (P Olsen: 9.95 ppm) and 1.37% organic matter content. The weather data throughout the experimental period were obtained from the automatic weather station (Davis Vantage Pro2 Weather Station; Davis Instruments Corporation, Hayward, CA, USA) of the AUA and are shown in Figure 1. Total precipitation from November 2020 to May 2021 was 204.4 mm, while the mean temperature at the same period was 14.8 °C.

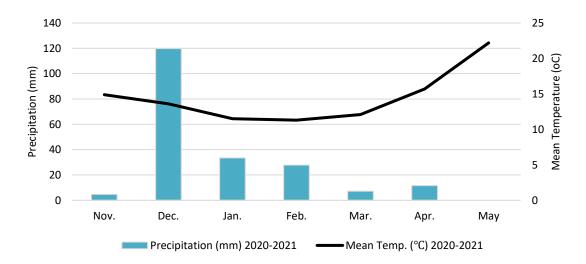


Figure 1. Weather data (mean monthly air temperature and precipitation) for experimental site throughout the experimental period (November 2020 - May 2021)

The experiment was set up on an area of 418 m² according to the split-plot design with four replicates, two main plots (conventional tillage and no-tillage), and three sub-plots [fertilization type: untreated (control), urea, urea with inhibitors]. Specifically, the application of conventional tillage was achieved by mouldboard ploughing at 25 cm, followed by one rotary hoeing at 10-15 cm, while the plots of no-tillage were no-tilled. The total applied fertilizer dose for urea fertilizers with and without inhibitors was 100 kg N ha⁻¹. The type of urea fertilizer was 46-0-0. The nitrification inhibitor was N-((3(5)-methyl-1H-pyrazol-1-yl) methyl) acetamide (MPA; 0.07%) and the urease inhibitor was N-(2-Nitrophenyl) phosphoric triamide (2-NPT; 0.035%) for the fertilizer with urea with double inhibitors (46-0-0). The plot size was 15 m². Sowing distances were 40 cm between rows. Safflower (*Carthamus tinctorius* L. cv. GW9022) was sown by hand at a depth of 2-3 cm. The field was sown on 24th November 2020. Weeds were controlled by hand hoeing when it was necessary.

For plant height and dry weight per plant, five plants were randomly selected from each plot at 130 days after sowing (DAS). Leaf area was measured using an automatic leaf area meter (Delta-T Devices Ltd, Burwell, Cambridge,

UK). Thus, the measurements on plant basis were converted into a LAI by divining by the average crop density of each plot. For the determination of seed yield components, ten plants were randomly selected at the harvest time (2nd June 2021; 190 DAS). Moreover, the safflower seed yield was determined by manually harvesting all the plants of each plot.

The experimental data were subjected to statistical analysis according to the split-plot design. The statistical analysis was performed with SigmaPlot 12 statistical software (Systat Software Inc., San Jose, CA, USA). Differences between means were separated using the Tukey's test. All comparisons were made at the 5% level of significance.

RESULTS AND DISCUSSIONS

The tillage system and fertilization effects on the plant height of safflower are shown in Table 1. Plant height was not affected by soil tillage; however, the different fertilization regimes had a significant effect on this trait. During the experiment, plant height increased linearly with the increasing rate of available nitrogen. Specifically, the highest mean plant height (102.00 cm) was achieved in urea with inhibitors treatment, followed by urea (90.82 cm) and control (84.38 cm). The increased height of safflower plants with increased levels of available nitrogen to plants was mostly due to nitrogen's role in promoting metabolic activity, which contributed to an increase in the number of metabolites, resulting in internode elongation and enhanced plant height by raising levels of available nitrogen (Abbadi and Gerendás, 2009; Kakabouki et al., 2019).

Concerning dry weight per plant, the effect of the different tillage systems was found not to be statistically significant. Despite that, the study data revealed that the highest values (16.27-18.86 g) were observed in the conventional tillage plots (Table 1). In response to the fertilization effect, this had a significant effect on this trait, and the highest values were demonstrated in the case of urea with inhibitors treatment with the values being 18.86 and 17.89 g per plant in the conventional and minimum tillage plots, respectively. A similar trend has been reported by (Abbadi et al., 2008), (Dordas and Sioulas 2008; Koutroubas et al., 2021), who they recorded a positive impact of nitrogen fertilization on the plant dry matter of safflower. According to the analysis of variance, neither the tillage system nor the fertilization regimes had a significant impact on the leaf area index (LAI). At this point, it is worth noting that the plants of urea with inhibitors treatment, presented slightly higher values (4.83-4.98 m m⁻²; Table 1). In general, it has been demonstrated that supply with increased nitrogen rates results in higher total leaf area, which in turn increases light absorption and carbon fixation by plants (Field and Mooney, 1986)

	TILLAGE								
	СТ	NT	СТ	NT	СТ	NT			
	Plant height (cm)		Dry weight per plant (g)		LAI (m m ⁻²)				
FERTILIZATION									
UREA	94.25 ^{Ab}	87.39 ^{Ab}	16.73 ^{Ab}	16.67 ^{Ab}	4.48 ^{Aa}	4.69 ^{Aa}			
UREA +	99.25 ^{Aa}	104.75 ^{Aa}	18.86 ^{Aa}	17.89 ^{Aa}	4.83 ^{Aa}	4.98 ^{Aa}			
CONTROL	88.00 ^{Ac}	80.75 ^{Ac}	16.27 ^{Ac}	14.44 ^{Ac}	4.25 ^{Aa}	4.44 ^{Aa}			
	$P_{\rm fert} = 0.041$		$P_{\rm fert} = 0.025$		$P_{\rm fert} = 0.324$				
	$P_{\rm tillage} = 0.457$		$P_{\text{tillage}} = 0.290$		$P_{\rm tillage} = 0.123$				

Table 1. Effects of tillage systems (conventional tillage and no-tillage: CT and NT, respectively) and fertilization (urea, urea with inhibitors and untreated: UREA, UREA + INHIBITORS, and CONTROL, respectively) on plant height, plant dry weight and leaf area index (LAI).

Note:*P*-values are from ANOVA. Values with different lowercase letters are significantly different among fertilization treatments in the same tillage system at P < 0.05. Different capital letters show statistically significant difference among tillage systems at P < 0.05.

The results for the number of capitula, seeds per capitulum and seed yield are presented on Table 2. Number of capitula was affected by both tillage practice and fertilization regime. The number of capitula was greater in the conventional than in no-tillage plots during the experimental period (10.72 and 9.51 for conventional and no-tillage system, respectively). This result is on contrary with (Banjara et al., 2015) reported increased number of capitula in no-tillage system. In response to fertilization effect, the highest values were observed in urea with inhibitors (13.39) treatment followed by urea without inhibitors (9.98). Such results were also demonstrated in previous studies reported that increasing levels of available nitrogen to plants produced better results in terms of the number of capitula per plant (Abbadi et al., 2008; Dordas and Sioulas, 2008; Eryigit et al., 2021). In contrast, (Strasil and Vorlicek 2002), demonstrated that nitrogen fertilization did not influence the number of capitula per plant under rainfed conditions.

Table 2. Effects of tillage systems (conventional tillage and no-tillage: CT and NT, respectively) and fertilization(urea, urea with inhibitors and untreated: UREA, UREA + INHIBITORS, and CONTROL, respectively) on number of
capitula, seeds per capitulum and seed yield.

	TILLAGE							
	СТ	NT	СТ	NT	СТ	NT		
	Number of capitula		Seeds per capitulum		Seed yield (kg ha ⁻¹)			
FERTILIZATION								
UREA	10.46 ^{Ab}	9.49 ^{Bb}	25.03 ^{Aa}	23.12 ^{Ba}	1222.5 ^{Ab}	1195.9 ^{Bb}		
JREA + INHIBITORS	14.25 ^{Aa}	12.53 ^{Ba}	27.43 ^{Aa}	22.35 ^{Ba}	1469.4 ^{Aa}	1333.7 ^{Ba}		
CONTROL	7.46 ^{Ac}	6.51 ^{Bc}	17.24 ^{Aa}	19.07^{Ba}	858.9 ^{Ac}	770.2 ^{Bc}		
	$\frac{P_{\text{fert}} = 0.013}{P_{\text{tillage}} = 0.025}$		$P_{\rm fert} = 0.170$		$P_{\rm fert} = 0.002$			
			$P_{\rm tillage} = 0.019$		$P_{\rm tillage} = 0.009$			

Note: P-values are from ANOVA. Values with different lowercase letters are significantly different among fertilization treatments in the same tillage system at P < 0.05. Different capital letters show statistically significant difference among tillage systems at P < 0.05.

The number of seeds per capitulum was affected only by tillage practice (Table 2) and the greatest value was observed in the conventional tillage plots (23.23). The result of this study also agreed with the results reported by (Kucuk and Akbolat 2013), in which safflower presented higher seeds per capitulum with conventional tillage practice. Concerning the seed yield, it was affected by both tillage practice and fertilization (Table 2) and the highest value was obtained in conventional tillage plots fertilized with urea with inhibitors (1469.4 kg ha⁻¹). In a previous multi-year study conducted in the Mediterranean environment, safflower did not respond to N treatment in a no-till system (Yau and Ryan, 2010). In another study, (Dordas and Sioulas 2008) used nitrogen fertilizer rates of 100 and 200 kg ha⁻¹ and produced higher safflower seed production despite higher soil NO₃⁻ levels. Taking into account the abovementioned, the difference in nitrogen fertilizer requirements for the safflower crop may be due to the soil management strategy used, different genotypes, the preceding crop, the residual nitrogen concentration in the soil, as well as climatic conditions.

CONCLUSIONS

To sum up, most of the plant characteristics were affected by fertilization. More specifically, plant height and dry weight were not affected by tillage practice, but by fertilization, and the best results were obtained from the urea with inhibitors treatment. Seeds per capitulum were affected by tillage practice, and the highest values were observed in conventional tillage. The number of capitula and seed yield were affected by both tillage practice and fertilization and the highest value was found in the combination of conventional tillage and urea with inhibitors. Finally, the present study indicated that urea with inhibitors fertilization and conventional tillage greatly improved plant productivity. There is a clear need to continue this research study as a long-term experiment in order to evaluate the effect of seasonality.

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

Funding Source: This research did not receive any funding.

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.

REFERENCES

1. Abbadi J, Gerendás J. Nitrogen use efficiency of safflower as compared to sunflower, J Plant Nutr. 2009; 32:929-45.

- 2. Abbadi J, Gerendás J, Sattelmacher B. Effects of nitrogen supply on growth, yield and yield components of safflower and sunflower. Plant Soil 2008; 306:167-80.
- 3. Asgarpanah J, Kazemivash N. Phytochemistry, pharmacology and medicinal properties of *Carthamus tinctorius* L. Chin J Integr Med. 2013; 19(2):153-9.
- 4. Banjara TR, Pali GP, Purame BP. Effect of tillage practices on growth and yield of safflower under rainfed midland condition of Chhattisgarh. Ecoscan 2015; 7:423-8.
- 5. Bilalis D, Roussis I, Fuentes F, Kakabouki I, Travlos I. Organic agriculture and innovative crops under Mediterranean conditions. Not Bot Horti Agrobot Cluj-Napoca 2017; 45:323-31.
- 6. Dordas CA, Sioulas C. Safflower yield, chlorophyll content, photosynthesis, and water use efficiency response to nitrogen fertilization under rainfed conditions. Ind Crops Prod. 2008; 27:75-85.
- 7. Eryigit T, Aldemir R, Kaya AR, Tuncturk M, Yildirim B, The influence of nitrogen doses on yield and yield properties of safflower (*Carthamus tinctorius*) varieties under micro-climate conditions of Igdir plain Turkey. J Plant Nutr. 2021; 44:2840-8.
- 8. Field C, Mooney HA. The photosynthesis-nitrogen relationship in wild plants. In: Givnish TJ, editor. On the economy of plant form and function. Cambridge, London: Cambridge University Press; 1986; p. 25-55.
- 9. Golzarfar M, Shirani Rad AH, Delkhosh B, Bitarafan Z. Safflower (*Carthamus tinctorius* L.) response to different nitrogen and phosphorus fertilizer rates in two planting seasons. Žemdirbystė (Agriculture) 2012; 99(2):159-66.
- 10. Kakabouki IP, Hela D, Roussis I, Papastylianou P, Sestras A, Bilalis DJ. Influence of fertilization and soil tillage in quinoa crop (*Chenopodium quinoa* Willd.). Nitrogen uptake and utilization efficiency. Expression of nitrogen indices. J Soil Sci Plant Nutr. 2018; 18(1):220-35.
- 11. Kakabouki IP, Roussis I, Hela D, Papastylianou P, Folina A, Bilalis D. Root growth dynamics and productivity of quinoa (*Chenopodium quinoa* Willd.) in response to fertilization and soil tillage. Folia Hort. 2019; 31(2): 277-91.
- 12. Karydogianni S, Darawsheh MK, Kakabouki I, Zisi C, Folina AE, Roussis I, Tselia Z, Bilalis D. Effect of nitrogen fertilizations, with and without Inhibitors, on cotton growth and fiber quality. Agron Res. 2020; 18:432-49.
- 13. Karydogianni S, Roussis I, Mavroeidis A, Kakabouki I, Tigka E, Beslemes D, Stavropoulos P, Katsenios N, Tsiplakou E, Bilalis D. The influence of fertilization and plant density on the dry matter yield and quality of black mustard [*Brassica nigra* (L.) Koch]: An alternative forage crop. Plants 2022; 11:2683.
- 14. Koutroubas SD, Damalas CA, Fotiadis S. Safflower assimilate remobilization, yield, and oil content in response to nitrogen availability, sowing time, and genotype. Field Crops Res. 2021; 274:108313.
- 15. Krol DJ, Forrestal JP, Wall D, Lanigan JG, Sanz-Gomez J, Richards GK. Nitrogen fertilizers with urease inhibitors reduce nitrous oxide and ammonia losses, while retaining yield in temperate grassland. Sci Total Environ. 2020; 725:138329.
- 16. Kucuk H, Akbolat D. Investigation of different tillage and seeding methods in safflower (*Carthamus tinctorius* L.) cultivation. Sci. Papers Ser. A, Agron. (Univ. Agron. Sci. Vet. Med. Buchar.) 2018; 61(1):481-6.
- 17. Strasil Z, Vorlicek Z. The effect of nitrogen fertilization, sowing rates and site on yields and yield components of selected varieties of safflower (*Carthamus tinctorius* L.). Rost Vyroba 2002; 48(7):307-11.
- 18. Wang H, Köbke S, Dittert K. Use of urease and nitrification inhibitors to reduce gaseous nitrogen emissions from fertilizers containing ammonium nitrate and urea. Glob Ecol Conserv. 2020; 22: e00933.
- 19. Yau SK, Ryan J. Response oh rainfed safflower to nitrogen fertilization under Mediterranean conditions. Ind Crops Prod. 2010; 32:318-23.