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EFFECT OF TIME NITROGEN FERTILIZER APPLICATION ON THE SENESCENCE AND GRAIN YIELD OF FIVE DURUM WHEAT CULTIVARS (TRITICUM DURUM DESF.)

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Abstract

The experiment was conducted to evaluate the effects of time nitrogen fertilizer application on the senescence of durum wheat and its association with grain yield and its components. The experimental treatments were set up in a split-plot design with five durum wheat cultivars (Adnham, Grecale, Duma, Sardar, Svevo) as the main plots and the time nitrogen fertilizer application (100 % tillering, 100 % stem elongation and 50% tillering + 50 % stem elongation stages) under two locations in Northern Iraqi conditions. The results of this study indicate that late senescence cultivars (Grecale and Svevo) produced the highest flowering date, No head/m2, No. grain/10 head, grain yield g/m2 and harvest index %. Moreover, time nitrogen fertilizer application at the tillering stage (early application) influenced most of the studied traits. In conclusion, extending photosynthesis capacity during the grin-filling phases by delaying the rate of senescence might be a good approach to improve grain yield of wheat cultivars.

Keywords: cultivars, rate of senescence (RS), time nitrogen fertilizer application (TNFA)

Introduction

Proper usage of fertilizer, particularly nitrogen (N), is critical to minimize input cost and harmful effects on the environment. Wheat is considered as one of third important crops behind maize and rice; however, the environment substantially impacts wheat yield and quality. This can help to explain why different regions of the world cultivated many different kinds of wheat (Stone and Savin, 2000). In the Mediterranean climate, wheat is grown in an area characterized by most temperature and rainfall favourable at early growing stage, followed by heat stress and water shortage at the developmental stages (Lo´pez-Bellido et al., 2005). In wheat, the grain yield (GY) and quality are mainly affected by the quantity of N that remobilized at the filling stages from accumulated N in vegetative parts at the early stage (Kichey et al. 2007). Generally, the GY and its components are often influenced by genetic source and environmental conditions, such as the application of N rate and time, water deficit, and heat stress especially at the developmental stages (Abedi et al. 2010; Farooq et al. 2011; Altaf et al. 2021). According to these studies, selecting the proper application of nitrogen rate and time is a critical process to maximize the crop production and minimize the losses of N by leaching. In cereal crops, the N recovery at the sowing is lower than the recovery at late applications between tillering stage (TS) and stem elongation stage (STES) (López-Bellido et al. 2005; Abedi et al. 2011). The low recovery of N application at the early stage can be explained due to the high nitrate leaching and denitrification. A possible explanation for this might be the low N demand of the small plants (root and shoot) during the early stages. On the other hand, many studies mentioned that early N application between early TS and end of STES stages could achieve the maximum GY in wheat. This may be explained by the highest N demand is generally occurred during TS when the biomass accumulated quickly (Malhi et al. 2006). Nitrogen availability, light intensity, temperature and water deficit have a significant influence on the grain-filling duration in wheat. All those conditions lead to increased grain-filling duration, which results in well-filled grain (Garrido- Lestache et al. 2004); whereas, under stress conditions, drought and heat, as in Mediterranean climate, where the grain-filling period is short, leads to lower GY.

Senescence is a last developmental process when the plant tissues turned to yellow and mostly influenced by genetic control as well as environmental conditions. Generally, it begins about 10-24 from the beginning of anthesis, which allows more refilling of nutrients from the tissue into the sink (Gregersen et al. 2013; Chapman et al. 2021). All environmental factors influencing plant senescence could be classified into two factors: abiotic such as water stress and nutritional deficiency, and biotic such as pathogen infection. Nitrogen availability in the plant strongly influences rate of leaf senescence (RS). An example, the low N condition could accelerate the onset and RS; however, the high availability of N condition can lead to delays in the onset and RS in wheat (Havé et al. 2017; Chen et al. 2020). Early leaf senescence is linked to high grain protein content and somewhat low GY; however, delayed leaf senescence is usually associated with high GY and relatively low grain protein content in wheat. Together, early and late leaf senescence is regulated mainly by the senescence regulation associated genes and environmental conditions (Alhabbar et al. 2018a; Sultana et al. 2021). Therefore, delaying the RS can increase the grain-filling phase, leading to extended duration of CO2 assimilation and photosynthesis, leading to high GY, especially under favourable situations.

Materials and Methods

Site and Crop Management

Field experiments were conducted in the Mosul region (North Iraq) in two locations for the winter agricultural season 2021-2022. The first experiment was performed at Nimrud (Balawat village), which lies 25 kilometers southeast of the city of Mosul, and the second experiment was conducted at Bashiqa, which lies 12 kilometers Northeast of the city of Mosul. Both locations are classified as the Mediterranean region; information about monthly rainfall and minimum and maximum temperatures are shown in (Supplementary Table 1).

The experiment was set up as a randomized complete block designed with a split-plot and three replications. Main plots included the time nitrogen fertilizer application (TNFA) (100 % tillering

stage (TS), 100 % stem elongation stage (STES) and 50% TS + 50 % Stem Elongation stages (STES). The sub-plots were five durum wheat cultivars (Adnham, Grecale, Duma, Sardar, Svevo). The sub- plot consists 5 lines with a length of (1 m), and the distance between a line and another (18 cm) with (30 cm) a gap between an experimental unit. Sowing date was 1st December in 2021, which is the recommended dates for wheat crop in the northern Iraqi farming districts.
TNFA was synchronized to Zadoks scale (Z) of wheat growth stages (Zadoks et al. 1974). Hence, these were T1= 80 kg N /ha (topdressing) was supplied at early TS (Z21 - Z23); T2= 80 kg N /ha (topdressing) supplied at early STES (Z31- Z32), and T3= 40 kg N /ha supplied at TS, and 40 kg N/ha applied at early STES. Urea fertilizer (46% N) was utilized as a main source of Nitrogen. Additionally, recommended dose (160 kg/ha) of Diammonium phosphate was utilized by furrow placement prior to sowing. The soil sample was collected at depth of 0–60 cm prior to wheat sowing. Soils were analyzed for nitrate content, as shown in (Supplementary Table 2). During the two locations, the rainfall season was only about 160 - 270 mm (Supplementary Table 1), so three separate times irrigation applications (50mm every time) were supplied in February and March.

Measurements

The flowering date was recorded when around 50% of the heads in each plot own visible stamens (Z 61-65). Total AGB (grain and straw) samples were collected on the 3rd of June 2022, when all plants were visually inspected to be fully mature. Number of grains/10 head was calculated by counting No. of grains of 10 heads/plot using grains counter machine. Number of heads was estimated by counting the heads in the middle three rows/plot. Harvest index (HI) was calculated by the grain ratio to above-ground biomass (AGB) at harvest (Richards et al. 1987).

Stay-green cultivars (late of senescence)

Chlorophyll Concentration Meter, IC-CCM-200 (Apogee Instruments) has been used to estimate amount of chlorophyll in the flag leaf. The IC-CCM-200 measures the Chlorophyll Content Index by dividing the transmission of radiation (931 nm) by the transmission of radiation (653 nm) (Padilla, 2019). Values of five plants were taken per plot at the middle position on the flag leaf at every stage. The CCI measurements were recorded at seven developmental growth stages [Ear emergence from boot (Z55-Z59), anthesis (Z61-Z65), 7,14,21,28 and 35 days after anthesis (DAA) based on the zadoks scale. The RS of flag leaf was estimated when the chlorophyll content decreased rapidly after anthesis stage (Ommen et al. 1999; Gelang et al. 2000). The maximum chlorophyll content index was observed at 7 days after anthesis (7DAA) in all five durum wheat cultivars. Therefore, RS was calculated as (MaxCCI – XCCI) / MaxCCI x 100, where MaxCCI is the plant's maximum CCI value before declines, and XCCI is the CCI value at the specific stage used to calculate flag leaf RS reduction (Alhabbar et al. 2018b).

Statistical analysis

Analysis of variance (ANOVA) was carried out by Genstat software to investigate the effects of location, cultivar and TNFA When F-values were significant, the least significant difference (LSD) test and standard deviation were used to compare treatment differences in means. In all cases the differences were deemed to be significant if ($p<0.05$). The correlation analysis was done to examine the relationship between the RS and GY and its component.

Results

The climate data showed acceptable rain amounts in April and May, which is unexpected in the study region (Mediterranean climate) as presented in the (Supplementary Table 1). The stay- green cultivars possess a longer grain-filling phase allowing additional CO_2 assimilation and photosynthesis. These advantages for stay-green cultivars could lead to high GY, especially under favourable conditions during grain filling period.

Identification of early-senescence and stay-green cultivars

The identification of early-senescence and stay-green cultivars was based on the Chlorophyll Concentration Meter after 7 DAA (Maximum CCI). The RS showed a wide-ranging difference among the five cultivars during the developing stages (Chart 1a). Among growing stages, the RS significantly increased during stages (Chart 1b). Averaged over the TNFA, location and stages, the RS was significantly different between the five cultivars. Grecale and Svevo cultivars (stay-green) have less senescence rate than the other cultivars (early-senescence). TNFA was not significant; however, the late application (T2 and T3) achieved less RS than the early application (Chart 1c). The interaction of cultivars and growth stages significantly impacted the RS ($P<0.001$). At the 14 DAA, all five cultivars have the same RS However, at later stages, 21, 28, and 35 DAA, both cultivars Grecale and Svevo (stay-green) have less RS compared to the Sardar, Duma and Adnham (early-senescence), as shown in the (Chart 1d). For most of the rest of the interactions, the was no significant difference influence on the rate of senescence.

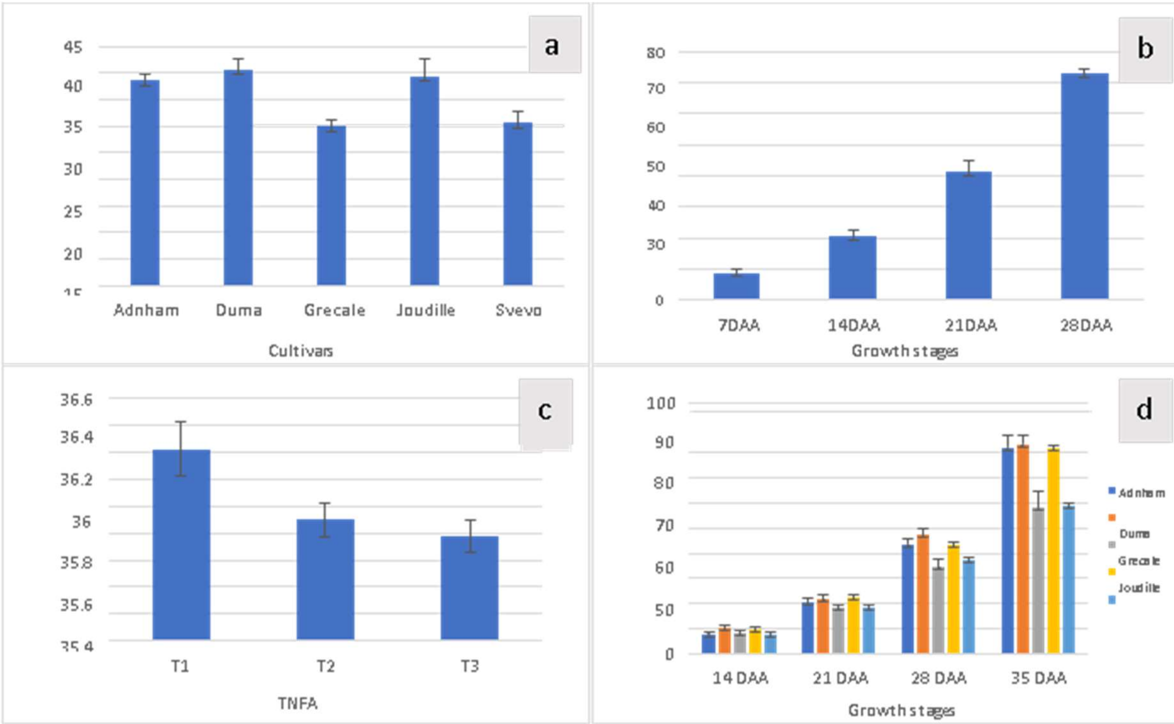

Chart 1 shows the effect of cultivars (a), growth stages (b), TNFA (c), and the interaction between five durum wheat cultivars and different growth stages on the RS (d)

Effect of five cultivars, TNFA and locations on the agronomic traits

Most vegetative traits had significant differences in locations, cultivars and TNFA. The mean plant height in Nimrud (74.3 cm) was higher than that in Bashiqa (68.19 cm). The Duma cultivar had the highest plant height compare to Sardar (Table 1). The average of TNFA, the application at T1 gave the maximum plant height, whereas applying the N at T3 produced the lowest plant height.

Table 1) Effects of locations, durum wheat cultivars and TNFA on Plant height, Flag Leaf Area, Flowering, AGB and Chlorophyll Content Index

Locations (L)	Plant height (cm)	Flag Leaf Area (cm^2)	Flowering (Day)	AGB (g/m^2)	Chlorophyll Content Index(CCI)
Bashiqa	68.19	20.54	118.2	1365	44.46
Nimrud	74.3	30.99	128.51	1269	44.14
Test F	<.001***	<.001***	<.001***	0.022*	0.610
LSD	1.775	1.292	0.466	81.4	N.S
Cultivars (CV)					
Duma	75.06	23.96	123.28	1266	43.04
Grecale	71.61	26.07	123.89	1387	45.16
Adnham	68.11	26.72	123.56	1365	43.64

Sardar	66.89	25.04	122.44	1237	44.89
Svevo	74.57	27.03	123.61	1328	44.77
Test F	<.001***	0.021*	0.003**	0.110	0.164
LSD	2.806	2.042	0.736	NS	NS
Time of N applications (TNFA)					
T1	72.28	26.53	123.10	1410	44.67
T2	72.21	25.52	123.40	1269	44.01
T3	69.26	25.25	123.57	1271	44.22
Test F	0.008**	0.241	0.259	0.008**	0.682
LSD	2.174	NS	NS	99.7	NS
L x CV	0.003**	0.061 NS	<.001***	0.191 NS	0.052 NS
L x T	0.139 NS	0.666 NS	0.114 NS	0.009**	0.010*
CV x T	0.039*	0.162 NS	0.936 NS	0.191 NS	0.144 NS
L x CV x T	0.002**	0.577 NS	0.457 NS	0.023*	0.908 NS

- *, **, *** indicate significance at the 0.05, 0.01and 0.001 resp.

Flowering date significantly influenced by the locations, cultivars and TNFA (Table 1). The plants of Bashiqa have an early flowering date (118.2 days) compared to Nimrud location (128.51 days). On average, Sardar cultivar reached the flowering date earlier than other cultivars. The AGB was affected by the locations, cultivars and TNFA. The plants of Bashiqa produce maximum AGB (1365 g/m2) than Nimrud (1269 g/m2). TNFA contributes to the AGB; the highest value was reached when N added at the TS (1410 g/m2). Overall, there were no significant differences in most agronomic traits between interaction among locations, cultivars and TNFA (Table 1).

Effects of locations, durum wheat cultivars and TNFA on the GY and its components

The GY and its components were significantly variance among locations, cultivars and TNFA, as shown (Table 2). Across the cultivars and TNFA, the plants of Bashiqa gave maximum thousand-grain weight than Nimrud. Among the five cultivars, Duma has the highest thousand-grain weight (54.77 g), while Adnham has the lowest (44.85 g). Among the TNFA, the late application (STES) produced the highest thousand-grain weight (54.77 g) compared to TS (early application) (45.80 g). Regarding the mean of locations, the plants of Bashiqa produced a greater No. of heads (247.9 head/m2) than Nimrud (231.1 head/m2). The highest No. heads was obtained by Grecale, with an average of (267.2 head/m2). The mean of cultivars and TNFA indicated the superiority of the plants of Nimrud location with maximum No. of grains (399.4 grain/10 head) over the plants of Bashiqa location (213.8 grain/10 head). The mean values for the cultivars, Grecale was produced a high No. of grains compared Duma. Apply N fertilizer at TS achieved biggest No. of grains (320.1 grain/10 head). The GY was a difference among the five wheat cultivars and TNFA (Table

2). Nimrud location provided the highest mean of GY (478.8 gm), but it was not significant as that of Bashiqa location (464.9 gm). The values of cultivars over location and TNFA resulted in a 20.8% higher GY of Grecale than to Sardar. As for the TNFA, the highest GY occurred when N fertilizer was added at TS (523.0 gm). Regarding HI, the plants of Nimrud location produced higher HI (37.74 %) than Bashiqa location (34.43 %). Svevo cultivar has a higher HI than Sardar cultivar. Apply N 50% at TS + 50% STES has better HI compared to STES.

Overall, there were significant differences in most GY and its component traits over the interaction between locations, cultivars and TNFA (Table 2)

(Table 2) Effects of locations, durum wheat cultivars and TNFA on thousand grain weight, No. Head, No. grain, GY and harvest index

Locations (L)	Thousand grain weight (g)	No Head (Head/m^2)	No. Grain (10 Head)	GY (g/m^2)	Harvest Index (%)
Bashiqa	52.32	247.9	213.8	464.9	34.43
Nimrud	49.68	231.1	399.4	478.8	37.74
Test F	0.010**	0.029*	<.001***	0.297	0.002**
LSD	1.991	14.98	11.82	N.S	2.004
Cultivars (CV)					
Duma	54.77	222.5	282.6	423.9	33.59
Grecale	49.60	267.2	330.5	532.2	38.72
Adnham	44.85	223.6	290.1	469.9	34.90
Sardar	54.27	218.6	306.1	421.6	33.36
Svevo	51.51	265.6	323.6	511.6	39.86
Test F	<.001***	<.001***	<.001***	<.001***	<.001***
LSD	3.148	23.69	18.68	41.55	3.168
Time of N applications (TNFA)					
T1	45.80	240.0	320.1	523.0	36.85
T2	54.45	231.2	300.5	417.9	33.25
T3	52.75	247.3	299.2	474.6	38.15
Test F	<.001***	0.219	0.008**	<.001***	<.001***
LSD	2.438	NS	14.47	32.18	2.454
L x CV	0.791 NS	0.511 NS	0.083 NS	0.188 NS	0.040*
L x T	0.333 NS	0.013*	0.016*	0.007**	0.743 NS
CV x T	0.072 NS	0.007**	<.001***	0.117 NS	0.305 NS
L x CV x T	0.166 NS	0.781 NS	<.001***	0.025*	0.868 NS

- *, **, *** indicate significance at the 0.05, 0.01and 0.001 resp.

Correlation between RS with agronomic traits and GY components

Results of the correlation analysis between RS, agronomic traits and GY components of early cultivars (Duma, Adnham and Sardar) and late cultivars (Grecale and Svevo) under different TNFA are presented in (Table 3).

Overall, results of correlation indicate that, there was a negative association among RS at 7, 14, 21, and 28 DAA and AGB (r= -0.06, -0.14, -0.12, -0.36 resp.), harvest index (r= -0.50, -0.33, -0.34, -0.31 resp.), and GY (r= -0.30, -0.07, -0.50, -0.48 resp.) under early maturity cultivars (Duma, Adnham and Sardar). However, a positive association was found among RS and AGB (r= 0.38, 0.56, 0.33, 0.19 resp.), harvest index (r= 0.22, 0.30, 0.05, 0.48 resp.), and GY (r= 0.51, 0.26, 0.54, 0.58 resp.) under late maturity cultivars (Grecale and Svevo).

(Table 3) Correlation between RS during grain-filling stages with agronomic traits of early-senescence and late senescence under locations and TNFA

	Early-senescence				Late senescence			
	Sen. 7	Sen. 14	Sen. 21	Sen. 28	Sen. 7	Sen. 14	Sen. 21	Sen. 28
Plant height	-0.36	-0.38	-0.09	-0.08	0.06	-0.43	-0.27	0.17
Chlorophyll	-0.04	-0.03	0.26	0.19	-0.06	0.01	-0.19	-0.43
Flowering	-0.41	-0.54*	-0.35	-0.33	0.09	-0.22	-0.20	0.38
Flag Leaf Area	-0.38	-0.45	-0.36	-0.27	0.10	-0.28	-0.20	0.47
AGB	-0.06	-0.14	-0.12	-0.36	0.38	0.56*	0.33	0.19
No. Head	0.004	0.49*	0.26	0.30	0.04	0.40	-0.26	-0.25
No. Grain	-0.60**	-0.50*	-0.25	-0.25	0.01	-0.50*	-0.35	0.31
thousand-grain weight	0.30	0.02	0.05	0.02	-0.55*	-0.20	0.11	-0.21
Harvest Index	-0.50*	-0.33	-0.34	-0.31	0.22	0.30	0.05	0.48
GY	-0.30	-0.07	-0.50*	-0.48*	0.51*	0.26	0.54*	0.58*

- *, ** indicate significance at the 0.05 and 0.01 resp. Senescence rate at 7DAA (Sen 7), senescence rate at 14DAA (Sen. 14), senescence rate at 21DAA (Sen. 21), senescence rate at 28DAA (Sen. 28)

Discussion

Both stay-green and early-senescence traits could be a valuable approach for improving the GY of wheat under specific area. Usually, under Mediterranean climates such as northern Iraq, the growing season of wheat when both temperature and rainfall are appropriate through vegetative stage (i.e. January, February And March), whereas with regular heat stress and lack of rainfall through grain-filling stage (i.e. April and May). However, in the current study, there was an acceptable amount of rainfall during the maturity stage in both locations. The stay-green trait describes the plant's ability to maintain its leaves green for a longer duration, often but not always exhibiting higher GY than early-senescence plants (Luche al., 2015 Alhabbar al., 2018b). Many studies found that stay-green plants with slow RS possess high N uptake and remobilization by extending grain-filling periods (Lopes et al. 2012; Hitz et al. 2017; Alhabbar 2018b). Our results indicated that the RS (Chart 1) completely differs for the five durum wheat cultivars. Duma, Adnham and Sardar (early-senescence) cultivars own an early and fast RS; however, delayed RS within the cultivars Grecale and Svevo (stay-green). Therefore, the two cultivars Grecale and Svevo have more chance to uptake and accumulate N during the post-anthesis growth stages. Taken together, this information could be used to select cultivars with an extending capacity of photosynthesis during the grin-filling phases by delaying the RS and led to increasing the potential GY of wheat.

Effects of TNFA on GY and its component

Optimum TNFA is a good approach for improving recovery of applied N up to 70% and thus raise the GY and quality of wheat. In the current study, the TNFA improved most GY and its component traits. The early applications (TS) increased the Plant height, Flag Leaf Area, AGB, No. Grainl10 heads, GY and Harvest Index; however, the late application (STES) improved the thousand-grain weight. A synchronization N application between time supply and plant demand increases crop productivity and reduces N losses by leaching (Chen et al. 2006; Gentile et al. 2009). In wheat, the greatest N demand is ranged between TS and STES phase when plant targets are producing high AGB and GY, whereas application at later phase around flowering stage when plant targeting high protein in grains.

Correlation between RS with GY components

Table (3) shows the correlations between the RS with studied traits among the early and late senescence cultivars. Taken together, the results suggest that a negative association between the RS at 7, 14, 21, and 28 DAA and AGB, harvest index and GY under early cultivars, while a positive association under late cultivars. However, the thousand-grain weight correlated positively under early cultivars and negatively under late cultivars.

A possible explanation for this might be that early-senescence could increase nutrient remobilization from tissue to grain, leading to an increase in thousand-grain weight compared to stay-green (Gregersen et al. 2008; Alpuerto et al. 2021; Sultana et al. 2021). However, stay-green maintains the capacity of N absorption and photosynthesis at the developmental stages, which leads to higher biomass accumulation (Borrell et al. 2001).

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