

Split nitrogen sources effects on nitrogen use efficiency, yield and seed quality of safflower (*Carthamus tinctorius* L.)

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Abstract

The effects of nitrogen (N) on crop yields have historically been assessed with field trials, but selection and use of the best sources and optimal timing N applications have a significant role in realizing the maximum potential of oilseeds quality and quantity. This study was conducted to determine the combine effects of N sources [ammonium nitrate (AN), ammonium sulphate (AS), sulphur coated urea (SCU), and urea (U)] and split N fertilisation [($1/4, 3/4, 0$), ($1/3, 1/3, 1/3$), ($1/2, 1/2, 0$), and ($1/3, 2/3, 0$)] on safflower (*Carthamus tinctorius* L.) some growth characters, yield and seed quality, and N use efficiency based on a split plot design with three replications at the experimental research station, Shiraz University in 2015 and 2016. The highest safflower dry matter ($5140.93 \text{ kg ha}^{-1}$), seed yield ($3303.52 \text{ kg ha}^{-1}$) and protein yield ($694.95 \text{ kg ha}^{-1}$) were achieved with the application of AN fertiliser in a split pattern of $1/2, 1/2, 0$ (applying half of the N at sowing time and the rest at stem elongation), while the highest oil yield ($753.09 \text{ kg ha}^{-1}$) was observed by U fertiliser and similar split pattern. Applying AN fertiliser and split patterns of $1/3, 2/3, 0$ (applying one third of the N at sowing and two thirds of the N at stem elongation) and $1/4, 3/4, 0$ (applying one quarter of the N at

sowing and three quarters at stem elongation) maximised safflower N uptake efficiency (NUpE) (0.78 kg kg^{-1}). However, the highest N utilisation efficiency (NUE) (43.70 kg kg^{-1}) was obtained when AN fertiliser in a split pattern of $1/2, 1/2, 0$ was applied. On the contrary, applying AS and SCU fertilisers was less effective on safflower performance by all split patterns. It is concluded that applying AN fertiliser in a split pattern of $1/3, 2/3, 0$ and or U fertiliser in a split pattern of $1/2, 1/2, 0$ not only enhanced safflower growth, yield and seed quality improved, but also increased the N use efficiency of safflower.

Introduction

Oilseeds are of great value in nutritional demands of mankind, animal feeding, and medicine. Among them, safflower (*Carthamus tinctorius* L.) is cultivated for many purposes: its achenes are commonly used as birdseeds and for the extraction of edible or industrial oil; corollas can be used for dyeing fabrics as food colourings and cosmetics and in painting or for producing medicines as pointed out by Danieli *et al.* (2011) and also as forage crop as reported by Cazzato *et al.* (2011). An important characteristic of this crop is its adaptation to semi-arid growing conditions owing to a deep root system (2-3 m in depth) enabling it to obtain moisture from unavailable levels for most crops (Ashkani *et al.*, 2007; Yeilaghi *et al.*, 2012). Quoted by Siadat *et al.* (2011) and Cazzato *et al.* (2013), the efficient use of fertilisers is one of the most important factors in maximizing crop yield and sustainability. Rathke *et al.* (2005) have underlined the significance of higher soil nutrient and particularly N availability in determining the yield quantity and quality of oilseeds. Moradi-Telavat *et al.* (2008) reported that nitrogen (N) increased rapeseed yield through increased silique number and seed weight. But a significant negative effect of N on oil content has been observed (Malhi and Gill, 2004; Siadat *et al.*, 2011). Fimes *et al.* (2000) and Li *et al.* (2007) noted that the response of crop to N fertiliser is influenced by the formulation, fertiliser management, soil properties, and seasonal trends. Muharnmad *et al.* (2007) reported that the highest seed protein was obtained when rapeseed (*Brassica napus* L.) treated by calcium ammonium nitrate. Similarly, Ozturk (2010) found that ammonium sulphate (AS) and urea (U) fertilisers gave higher crop yield than ammonium nitrate (AN) fertiliser, while Osman *et al.* (2014) showed that AN increased growth and yield parameters of rapeseed compared to other N fertiliser sources.

Because N availability is an important factor in determining crop productivity, managing fertiliser rate and application timing can be a suitable strategy to improve crop growth and yield when crops need it or when water is available to enhance nutrient uptake (Barlo'g and Grzebisz, 2004; Corbellini *et al.*, 2006). Some studies have shown that, depending on the level of fertility of the initial soil, split applications of N fertiliser result in higher rates of plant recovery and higher seed yields than under single applica-

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tions (Mossedaq and Smith, 1994; Dordas and Sioulas, 2008; Tedone *et al.*, 2014). Corbellini *et al.* (2006) and Blandino *et al.* (2016) reported that the rate and timing of N applications are crucial factors, not only to obtain high yields, but also to increase protein, gluten and the rheological parameters of wheat. Sheibani and Ghadiri (2012) observed that the application of U at a rate of 304 kg ha⁻¹, half at the sowing time and the other half at the end of rosette stage of rapeseed, enhanced seed yield up to 35%. Timing of N fertiliser also increases the efficiency of N use efficiency, with a higher plant recovery of this mineral and an increase in seed yield and quality, compared to a single application (López-Bellido *et al.*, 2012). Tedone *et al.* (2014) found that N application efficiency and N recovery efficiency in wheat crops increased when N fertiliser was applied at the stem elongation phase, whereas high amounts of N at sowing time and tillering, resulted in poor efficiency.

Therefore, it is important to develop strategies to enhance efficiency in uptake and utilisation of mineral nutrients by crops. Although N fertilisation has been effectively used in agricultural ecosystems' management, the effect of split N sources on oil crop production is unclear. In order to achieve the maximum potential of the safflower yield quantity and quality, this study was conducted to determine the combined effects of N sources and splitting N fertilisation on growth, yield and seed quality of safflower yield, and changes in N use efficiency.

Materials and methods

Field experiment

A 2-year field experiment was conducted at the experimental research station (Badjgah), Shiraz University (52° 46' E, 29° 50' N and 1810 m), Iran in 2015 and 2016. The soil was silty clay loam with a pH of 7.25 and an EC of 0.475 dS m⁻¹. The site was under fallow before cultivation. Total N, mean phosphorus (P) and mean potassium (K) were 0.07%, 12 mg kg⁻¹ and 250 mg kg⁻¹, respectively. Monthly average temperature, rainfall, and relative humidity for the last 30 years including the years 2015 and 2016 are shown in Table 1. The experiment was lay-out as split plot based on randomised complete block design with three replications. The treatments consisted of four levels of N fertiliser sources: AN (25% N), AS (21% N), sulphur coated urea (SCU; 34% N), and U (46% N) as main plot and split patterns of N were top-dressed (broadcast method) in four levels [(1/4, 3/4, 0), (1/3, 1/3, 1/3), (1/2, 1/2, 0), and (1/3, 2/3, 0)] as sub plot. Nitrogen fertilisation was carried out at three stages of safflower growing season including sowing time, stem elongation, and flowering (Flemmer *et al.*, 2015). Safflower N requirement (at a rate of 100 kg pure N ha⁻¹ during growing sea-

son) was determined according to the soil test analysis. The numbers in each treatment represent the amount of N fertiliser applied at each stage. For example treatment of 1/4, 3/4, 0 indicated that one quarter of the N (25 kg N ha⁻¹) at sowing, three quarters (75 kg N ha⁻¹) at stem elongation, and no-N application at flowering stage, respectively. Land preparation practices included plowing, disking and ridging plots (sized 3 by 3 m). Each plot was separated by two ridges to avoid cross contamination among plots. The seeds of safflower (Zendehrood cultivar) were sown two cm deep in rows spaced 15 cm apart (30 plants m⁻²) on March 25th in both years. Triple super phosphate fertiliser was applied at the sowing time at a rate of 50 kg ha⁻¹ according to the soil test analysis. Other management practices, such as pest control, were conducted according to local agronomic practices unless otherwise indicated. Weeds were controlled by hand hoeing throughout the growing season. The irrigation period was set at 10 day intervals for all treatments. Soil samples were taken at three depths (30, 60 and 90 cm) at the time of irrigation using the gravimetric method, and moisture content of soil was measured each time before irrigation and each plot was uniformly irrigated by siphon.

Trait measurements

At the end of the growth period (approximately middle of July in both years), safflower samples were randomly hand harvested from the middle 1 m² of each plot at maturity and partitioned into stems, leaves, and capitulum. After drying at 76°C for 48 h, plant samples were weighed, and dry matter, plant height, capitulum number per m², number of seeds per capitulum, 1000-seed weight, seed yield, oil yield (Soxhelt method according to Jensen, 2007), protein yield (semi micro-Kjeldahl digestion according to Bremner and Mulvaney, 1982), biological yield, and harvest index:

$$(HI) = \frac{\text{Seed yield}}{\text{Biological yield}} \quad (1)$$

were calculated.

Moreover, N uptake efficiency:

$$(NUpE) = \frac{\text{Total N uptake}}{\text{Pure N requirement of crop}} \quad (2)$$

and N utilisation efficiency:

$$(NUtE) = \frac{\text{Seed yield}}{\text{Total N uptake}} \quad (3)$$

Table 1. Some monthly weather parameters during the 30-year period from 1986-2016 (the years 2015 and 2016 have been presented separately).

Months	Average temperature (°C)			Precipitation (mm)			Average relative humidity (%)		
	2015	2016	30-Year	2015	2016	30-Year	2015	2016	30-Year
April	13.90	10.20	11.23	39.50	33.50	45.82	43.10	43.16	51.85
May	17.60	17.30	16.15	10.00	0.50	11.70	34.56	33.81	48.41
June	23.00	20.30	20.49	0.00	0.00	0.76	24.65	27.29	39.47
July	26.00	25.29	25.43	0.00	0.00	0.30	24.48	25.31	37.49
August	24.00	25.04	24.23	0.00	0.00	0.27	26.37	27.05	37.96
Average/Total	20.90	19.62	19.33	9.90	6.80	11.77	30.63	31.32	43.04

were estimated according to Rathke *et al.* (2006). Uptake efficiency is the ability of the plant to remove N from the soil as nitrate and ammonium ions, while the utilisation efficiency is the ability to use N to produce seed yield.

Statistical analysis

Differences between means were tested using SAS 9.1 software (SAS Institute, 2003). Statistical tests included one-way ANOVA (general linear model) followed by least significant difference (LSD) test at 5% probability level (Petersen, 1994), assuming a normal distribution of the dependent variable data and homogeneity of variances. The effect of year and interaction between year and all treatments were not significant, so the combined data were reported.

Results

Growth and yield response

Safflower growth and yield were significantly influenced by split N fertiliser sources (Table 2). Results showed that the highest plant height (121 cm) and total dry matter (5140.93 kg ha⁻¹) were obtained by AN fertiliser and split pattern of ¹/₂,¹/₂,0. Applying AN fertiliser and split pattern of ¹/₂,¹/₂,0 increased stem and leaf dry

matter approximately 18 and 12%, respectively compared to U fertiliser in a similar split pattern. Likewise, the highest capitulum number (101 per m²), seeds number (88), 1000-seed weight (39.25 g) (Table 3), seed yield (3303.52 kg ha⁻¹), protein yield (694.95 kg ha⁻¹), biological yield (8443.60 kg ha⁻¹), and harvest index (39.14%) were obtained by AN fertiliser and split pattern of ¹/₂,¹/₂,0, while the highest oil yield (753.09 kg ha⁻¹) was obtained when U fertiliser was applied in a split pattern of ¹/₂,¹/₂,0 (Table 4). Applying AN fertiliser and split pattern of ¹/₂,¹/₂,0 increased seed and protein yields (approximately 22 and 63%, respectively) compared to U fertiliser in a similar split pattern. On the contrary, applying U fertiliser in a split pattern of ¹/₂,¹/₂,0 increased oil yield (approximately 5%) compared to AN fertiliser in a similar split pattern. On the contrary, applying AS and SCU fertilisers increased safflower growth and yield to a lower extent compared to other N sources (Tables 3 and 4).

Nitrogen use efficiency changes

The effects of N sources and split patterns were significant on safflower N use efficiency (Table 2). AN fertiliser and split patterns of ¹/₄,³/₄,0 and ¹/₃,²/₃,0 maximised safflower NUpE (0.78 kg kg⁻¹), however the highest safflower NUtE (43.70 kg kg⁻¹) was obtained when AN fertiliser was applied in a split pattern of ¹/₂,¹/₂,0. In contrast, applying U fertiliser increased both of them N efficiencies, when split pattern of ¹/₂,¹/₂,0 was applied. The lowest NUpE (0.40 kg kg⁻¹) and NUtE (12.31 kg kg⁻¹) was observed

Table 2. The summary of the source of variation and the mean square of plant height, total, stem and leaf dry matters, capitulum number, seeds number, 1000-seed weight, seed, oil, protein and biological yields, harvest index, NUpE, and NUtE as affected by split N fertilisers application.

Source of variation	df	Plant height	Total dry matter	Stem dry matter	Leaf dry matter	Capitulum number	Seeds number	1000-seed weight
Year	1	176.04 ^{ns}	93,543.86 ^{ns}	47,530.27 ^{ns}	7714.99 ^{ns}	13.50 ^{ns}	68.34 ^{ns}	22.98 ^{ns}
Error (a)	4	107.71 ^{ns}	50,675.80 ^{ns}	27,490.88 ^{ns}	3520.01 ^{ns}	3.33 ^{ns}	121.34 ^{ns}	9.63 ^{ns}
Nitrogen source (N)	3	828.25 ^{**}	5,876,466.65 ^{**}	4,006,696.02 ^{**}	185,611.31 ^{**}	1754.34 ^{**}	8400.28 ^{**}	531.67 ^{**}
Year * N	3	126.56 ^{ns}	48,661.31 ^{ns}	25,458.35 ^{ns}	3730.41 ^{ns}	31.80 ^{ns}	26.17 ^{ns}	4.85 ^{ns}
Error (b)	12	22.68 ^{ns}	31,336.26 ^{ns}	16,818.60 ^{ns}	2246.66 ^{ns}	37.93 ^{ns}	175.94 ^{ns}	15.82 ^{ns}
Nitrogen split (S)	3	665.36 ^{**}	802,840.98 ^{**}	433,629.22 ^{**}	56,428.16 ^{**}	324.06 [*]	2434.64 ^{**}	73.41 ^{**}
N * S	9	221.13 ^{**}	279,727.07 ^{**}	151,058.04 ^{**}	19,721.97 ^{**}	210.41 [*]	261.02 [*]	24.25 [*]
Year * S	3	2.79 ^{ns}	1002.38 ^{ns}	536.38 ^{ns}	72.30 ^{ns}	2.41 ^{ns}	8.81 ^{ns}	2.91 ^{ns}
Year * N * S	9	18.02 ^{ns}	336.83 ^{ns}	175.93 ^{ns}	25.98 ^{ns}	5.09 ^{ns}	20.94 ^{ns}	3.66 ^{ns}
Error (c)	48	54.57	58,209.23	30,858.85	4314.73	59.82	105.18	11.04
Source of variation	df	Seed yield	Oil yield	Protein yield	Biological yield	Harvest index	NUpE	NUtE
Year	1	129,492.54 ^{ns}	17,943.40 ^{ns}	6252.06 ^{ns}	444,912.1 ^{ns}	24.65 ^{ns}	0.01 ^{ns}	2.71 ^{ns}
Error (a)	4	9639.54 ^{ns}	3337.43 ^{ns}	156.02 ^{ns}	49,970.0 ^{ns}	9.69 ^{ns}	0.004 ^{ns}	21.18 ^{ns}
Nitrogen source (N)	3	18,053,031.56 ^{**}	1,339,878.29 ^{**}	696,560.46 ^{**}	44,455,591.0 ^{**}	1817.71 ^{**}	0.43 ^{**}	1874.95 ^{**}
Year * N	3	12,808.69 ^{ns}	4312.89 ^{ns}	474.63 ^{ns}	97,302.3 ^{ns}	2.44 ^{ns}	0.0004 ^{ns}	11.50 ^{ns}
Error (b)	12	284,342.42 ^{ns}	39,855.10 ^{ns}	8476.76 ^{ns}	326,068.5 ^{ns}	39.78 ^{ns}	0.005 ^{ns}	96.19 ^{ns}
Nitrogen split (S)	3	3,741,045.90 ^{**}	139,663.11 ^{**}	196,570.70 ^{**}	7,382,675.9 ^{**}	370.91 ^{**}	0.11 [*]	526.31 ^{**}
N * S	9	708,925.42 ^{**}	39,517.14 ^{ns}	57,518.75 ^{**}	1,618,282.8 ^{**}	44.43 [*]	0.06 [*]	161.72 [*]
Year * S	3	3673.19 ^{ns}	588.51 ^{ns}	434.90 ^{ns}	5840.5 ^{ns}	0.46 ^{ns}	0.002 ^{ns}	6.39 ^{ns}
Year * N * S	9	20,020.43 ^{ns}	2262.57 ^{ns}	1099.99 ^{ns}	18,986.3 ^{ns}	2.79 ^{ns}	0.002 ^{ns}	8.62 ^{ns}
Error (c)	48	173,045.92	14,342.54	3980.01	274,979.1	19.60	0.005	44.33

*Significant at the 0.05 probability level; **Significant at the 0.01 probability level; ^{ns} not significant.

when AS fertiliser was applied in split patterns of $1/3, 1/3, 1/3$ and $1/2, 1/2, 0$, respectively. Additionally, applying SCU fertiliser and split patterns of $1/2, 1/2, 0$ and $1/3, 2/3, 0$ enhanced NUpE up to 0.50 kg kg^{-1} , while NUtE increased up to 21 kg kg^{-1} when SCU fertiliser was applied in a split pattern of $1/2, 1/2, 0$ (Table 4).

Yield, yield components, and nitrogen efficiency correlation

Correlation results showed that increasing yield components tended to enhance safflower yield and seed yield changes are closely related to the capitulum number (0.86**), seeds number

Table 3. Effects of split N fertilisers application on plant height, total, stem, and leaf dry matters, capitulum number, seeds number, and 1000-seed weight.

Nitrogen sources	Split pattern			Plant height (cm)	Total dry matter (kg ha ⁻¹)	Stem dry matter (kg ha ⁻¹)	Leaf dry matter (kg ha ⁻¹)	Capitulum number per m ²	Seeds number per capitulum	1000-seed weight (g)
	ST ^a	SE ^b	FC ^c							
Ammonium sulfate (AS)	1/4	3/4	0	100.83±8.61	3440.78±163.38	2442.95±116.00	997.82±47.38	78.50±5.29	40.16±6.33	24.25±3.49
	1/3	1/3	1/3	101.83±3.01	3570.98±300.63	2535.39±213.45	1035.58±87.18	69.83±9.86	33.50±7.56	26.56±1.65
	1/2	1/2	0	109.83±6.52	3404.58±137.40	2417.25±97.55	987.32±39.84	71.83±3.32	40.16±7.50	24.88±2.38
	1/3	2/3	0	92.16±4.85	3732.53±173.25	2650.09±123.01	1082.43±50.24	77.33±4.61	39.16±12.71	26.75±4.00
Urea (U)	1/4	3/4	0	110.50±4.92	3955.38±107.65	2887.42±78.59	1067.95±29.06	79.33±4.04	73.50±19.50	29.02±2.19
	1/3	1/3	1/3	104.17±3.33	4002.81±19.70	2922.05±14.38	1080.76±5.32	84.83±6.52	40.00±8.76	30.56±3.22
	1/2	1/2	0	115.16±7.81	4415.95±256.79	3223.64±187.45	1192.30±69.33	95.83±11.01	75.66±14.01	34.49±3.89
	1/3	2/3	0	104.33±6.75	4163.90±226.58	3039.64±165.40	1124.25±61.17	90.00±3.27	52.16±8.80	35.35±3.64
Ammonium nitrate (AN)	1/4	3/4	0	109.50±8.52	4199.75±96.60	3107.81±71.48	1091.93±25.11	85.33±6.75	88.00±9.00	33.91±5.05
	1/3	1/3	1/3	105.33±8.73	4225.10±141.60	3126.57±104.83	1098.52±36.83	85.50±7.50	60.16±10.69	32.72±2.38
	1/2	1/2	0	121.00±5.77	5140.93±426.63	3804.29±315.70	1336.64±110.92	101.33±8.97	88.66±11.40	39.25±2.48
	1/3	2/3	0	118.17±6.25	4576.81±432.57	3386.84±320.10	1189.97±112.46	95.00±6.72	76.16±8.89	37.44±2.27
Sulphur coated urea (SCU)	1/4	3/4	0	88.33±13.53	3482.76±184.01	2507.59±132.49	975.17±51.52	71.33±5.05	43.50±11.35	27.62±1.85
	1/3	1/3	1/3	104.67±3.40	3411.83±133.51	2456.52±96.13	955.31±37.38	80.16±10.53	31.16±5.53	25.46±3.39
	1/2	1/2	0	110.00±6.24	3699.78±286.95	2663.84±206.61	1035.93±80.34	78.33±7.28	48.16±6.82	29.97±4.05
	1/3	2/3	0	104.17±2.51	3468.40±117.10	2497.24±84.31	971.15±32.78	71.83±2.92	40.33±6.04	25.74±1.62
LSD (5%) ^d			11.00	382.90	279.00	104.04	11.95	17.62	5.23	

±standard deviation with indication of significant differences. ^aSowing time; ^bStem elongation stage; ^cFlowering stage; ^dLeast significant difference at P=0.05.

Table 4. Effects of split N fertilisers application on seed, oil, protein, and biological yields, harvest index, NUpE, and NUtE.

Nitrogen sources	Split pattern			Seed yield (kg ha ⁻¹)	Oil yield (kg ha ⁻¹)	Protein yield (kg ha ⁻¹)	Biological yield (kg ha ⁻¹)	Harvest index (%)	NUpE ^e (kg ha ⁻¹)	NUtE ^f (kg ha ⁻¹)
	ST ^a	SE ^b	FC ^c							
Ammonium sulfate (AS)	1/4	3/4	0	754.74±59.24	171.25±23.30	80.71±5.48	4195.54±180.97	17.95±1.26	0.48±0.05	15.91±3.21
	1/3	1/3	1/3	628.43±212.12	143.59±46.93	60.23±20.09	4199.45±394.78	14.82±4.03	0.40±0.01	15.67±5.60
	1/2	1/2	0	713.88±82.52	184.24±13.61	88.09±6.26	4118.40±70.10	17.28±2.19	0.57±0.01	12.31±1.26
	1/3	2/3	0	834.42±354.52	231.92±83.82	81.23±29.67	4566.98±524.91	17.78±5.88	0.51±0.03	16.00±6.26
Urea (U)	1/4	3/4	0	1688.06±459.98	485.86±166.65	249.95±65.74	5643.47±567.91	29.52±5.26	0.70±0.02	24.07±6.10
	1/3	1/3	1/3	1039.35±261.63	393.89±181.40	124.86±36.19	5042.12±267.46	20.45±4.22	0.65±0.04	15.80±3.46
	1/2	1/2	0	2708.77±956.20	753.09±229.27	425.29±147.61	7124.73±1206.27	37.19±7.05	0.73±0.13	38.99±18.20
	1/3	2/3	0	1664.46±359.32	512.04±143.29	255.87±61.70	5828.38±133.01	28.46±5.53	0.70±0.05	24.07±7.00
Ammonium nitrate (AN)	1/4	3/4	0	2599.97±796.02	720.79±270.63	377.51±106.73	6799.73±892.56	37.55±7.11	0.78±0.05	33.01±8.59
	1/3	1/3	1/3	1685.22±393.04	446.03±93.06	186.65±55.21	5910.33±514.62	28.28±4.03	0.67±0.09	24.69±2.18
	1/2	1/2	0	3303.52±154.97	715.65±31.93	694.95±125.74	8443.60±297.06	39.14±3.05	0.76±0.05	43.70±4.72
	1/3	2/3	0	2715.58±475.33	673.88±188.72	508.24±47.48	7292.34±551.05	37.15±4.94	0.78±0.08	35.25±10.40
Sulphur coated urea (SCU)	1/4	3/4	0	840.49±204.72	177.90±28.63	90.21±18.05	4321.90±386.53	19.27±2.91	0.47±0.03	17.42±2.94
	1/3	1/3	1/3	640.37±189.83	164.46±38.96	76.42±21.00	4052.13±317.23	15.62±3.50	0.44±0.02	14.56±5.00
	1/2	1/2	0	1134.83±258.20	238.31±34.22	114.22±25.87	4834.63±51.92	23.39±5.38	0.53±0.02	21.31±4.25
	1/3	2/3	0	743.26±102.44	167.82±42.185	98.98±18.96	4211.70±210.31	17.59±1.64	0.53±0.10	14.10±1.36
LSD (5%) ^d			706.02	221.40	111.35	865.05	7.78	0.10	10.06	

±standard deviation with indication of significant differences. ^aSowing time; ^bStem elongation stage; ^cFlowering stage; ^dLeast significant difference at P=0.05; ^eNitrogen uptake efficiency; ^fNitrogen utilisation efficiency.

(0.95**), and 1000-seed weight (0.92**). Likewise, protein yield increased as N use efficiency increased especially NUtE (0.82**), while increasing NUpE tended to decrease oil yield (-0.55*). Additionally, oil yield was negatively correlated with protein yield (-0.65*), while positive correlation between NUpE and NUtE was observed (Table 5).

Discussion

It seems that applying AN and U fertiliser sources are able to stimulate safflower growth by means of an enlarged leaf canopy and a greater rate of leaf expansion, which increased light interception and enhances photosynthesis (Ozturk, 2010). Additionally, a higher availability of N in the form of nitrate (NO_3^-) and ammonia (NH_4^+) increased cations uptake such as potassium (K), calcium (Ca) and magnesium (Mg) (Rathke *et al.*, 2005; Narits, 2010). It seems that increasing safflower growth plays a vital role in enhancing crop yield (Ozturk, 2010). These results are in agreement with those Osman *et al.* (2014), who reported that in oilseed rape, seed yield is closely related to the capitulum number and the physiological restrictions to capitulum formation are related to poor crop growth and limited leaf expansion. Bagawan and Ravikumar (2001) found a positive correlation between capitulum number and safflower seed yield. Tuncturk and Yildirim (2004) observed that the highest yield response of safflower was obtained by AN and U fertilisers application.

Furthermore, we found that the highest growth and yield responses obtained when safflower was treated by split pattern of $1/2$ at sowing time, $1/2$ at stem elongation. This high crop growth and performance are the result of high application rate of N in early growing season as it helps plants to avoid competing for N (Ribaud *et al.*, 2011; Jaynes, 2013). Additionally, Zong *et al.* (2014) found that the high application rate of N in mid-growing season resulted in relatively higher plant production compared with the early growing season and this increase was associated with N enrichment during vegetative-reproductive transition. Narits (2010) observed that the highest oilseed rape growth and yield response was obtained when N source was split into three

period times (sowing time, stem elongation, and flowering stage). Additionally, physiological investigations have shown that the stem elongation stage is important in the N nutrition of crop when a high N-supply is needed for yield formation (Behrens *et al.*, 2001; Barlo'g and Grzebisz, 2004). Kaefer *et al.* (2015) reported that oilseed rape growth and yield response were not influenced by N fertiliser sources, but were significantly influenced by split N pattern of $1/3$ at sowing and $2/3$ at top-dressing.

Meanwhile, results showed that adding AN and U fertilisers can increase N use efficiency due to greater access of safflower root to the mass flow of N especially NO_3^- (Dawson *et al.*, 2008). NUpE indicates effectiveness of fertiliser N-recovery due to N uptake by the plant (Hirel *et al.*, 2007). Furthermore, high NUtE is resulted from effective remobilisation and translocation of N from vegetative parts (especially stem) of the plant to developing tissues representing strong sinks for N during the seed-filling period (Masclaux-Daubresse *et al.*, 2010). The positive effect of adding N sources (especially AN fertiliser) in over production of protein yield and a reduction in oil content is consistent with the findings of Tuncturk and Yildirim (2004) and Farahbakhsh *et al.* (2006) that increasing N uptake by plants resulted in production of amino acid and other nitrogenous compounds during peptide bands. When amino acid and other compounds increased, the percentage of fatty acids decreased.

Conclusions

To enhance the productivity of safflower, different sources and application timing of N adapted to site conditions are remarkable growing strategies, which could help to realise the maximum potential of safflower quality and quantity. Our results showed that timing of N fertiliser application has the potential to change safflower agronomical parameters, alter productivity and quality of safflower yield, and influence safflower N uptake and utilisation efficiency. Overall, our findings suggested that applying AN fertiliser in a split pattern of $1/3, 2/3, 0$ and/or U fertiliser in a split pattern of $1/2, 1/2, 0$ not only improves safflower growth, yield and seed quality, but also enhances the N use efficiency of safflower.

Table 5. Correlation between yield, yield components, and N efficiency of safflower.

	Seed yield	No. Capitulum	No. Seeds	1000-seed weight	Oil yield	Protein yield	Harvest index	NUpE	NUtE
Seed yield	1.00								
No. Capitulum	0.86**	1.00							
No. Seeds	0.95**	0.72**	1.00						
1000-seed weight	0.92**	0.86**	0.82**	1.00					
Oil yield	0.64*	0.48*	0.54*	0.44*	1.00				
Protein yield	0.86**	0.74**	0.79**	0.69*	-0.65*	1.00			
Harvest index	0.98**	0.84**	0.97**	0.91**	0.58*	0.81**	1.00		
NUpE	0.89**	0.80**	0.88**	0.85**	-0.55*	0.79**	0.92**	1.00	
NUtE	0.98**	0.86**	0.92**	0.90**	-0.65*	0.82**	0.96**	0.81**	1.00

*Significant at the 0.05 probability level; **Significant at the 0.01 probability level.

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