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Study on Effect of Planting Date and Nitrogen Levels on Yield, Yield Components and Fatty Acids of Linseed (*Linum usitatissimum* L.)

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Abstract: In order to determine the optimum planting date and nitrogen fertilizer on yield, yield components and fatty acids profile of linseed, an experiment was conducted using split plot on the basis of RCB design with three replications in Experimental Field of Islamic Azad University of Yasooj during 2006-2008. Main plots were consisted of five planting dates (Mar. 5, Mar. 20, Apr. 3, Apr. 18 and May. 4) and sub plots including nitrogen fertilizer at rates of 0, 50, 100 and 150 kg ha^{-1}. The results of two-year analysis of data indicated that the highest plant height, capsule per plant and kernel weight obtained by Mar. 50, 20 and 100 and 150 kg N ha^{-1} and the lowest value were belonged to May.4 and Apr.18. At later planting date these three traits severely decreased. Mar. 5 and 100 kg N ha^{-1} was the best combination to produce the highest straw yield and harvest index. Among the different planting dates, Mar. 5 produced the highest grain yield at either N level. Grain yield after Mar.5 was reduced by 8 to 130 %. Planting date × N interaction interaction was also significant in this trait. The oil content ranged 24.4 to 32. 6% and was average 30%. Delay in planting date after Mar. 5 decreased oil seed about 33% at May.4. Increasing in nitrogen fertilizer decreased oil seed content. However, oil yield increased with applying in nitrogen fertilizer, due to increase in grain yield. Apr. 18 + 150 kg N ha^{-1} and Mar.5 and 150 kg N ha^{-1} were the best combinations for seed protein and seed protein yield, respectively. The unsaturated fatty acid contents varied from 14.6 to 52.6 %. The best combination in order to obtain the highest linolenic acid and oleic acid was Mar. 5+150 kg N ha^{-1} and Mar. 5+0 kg N ha^{-1}, respectively. In general, results of this study revealed that Mar.5 planting date was the best date for increasing all traits except seed protein percent. 100 or 150 kg N ha^{-1} also increased all traits except oil percent and oleic acid of linseed.

Key words: Linseed · Planting date · Nitrogen · Yield · Yield components · Fatty acids

INTRODUCTION

Flax (*Linum usitatissimum* L.) is a member of the genus *Linum* in the family Linaceae. It is an erect annual herb plant growing to about 1.2 m tall, with slender stems. The leaves are glaucous green, slender lanceolate, sessile, 20-40 mm long and 3 mm broad. The blue or white flowers are arranged in erect terminal panicles. The fruit is a globose capsule with shiny, flattened, brown seeds with a short blunt beak [1].

Flax is now unknown in the wild but originally it may have been a native of Asia. It has been cultivated since at least 5000 BC, probably first by the ancient Mesopotamians and later by the Egyptians who rapped their mummies in linen cloth. The Romans spread flax cultivation to Northern Europe and now the plant is grown all over the world for the oil extracted from the seeds and for its fibers, which are made into linen and other cloths. Various parts of the plant have been used to make fabric, dye, paper, medicines, fishing nets, hair gels and soap. It is also grown as an ornamental plant in gardens [2].

The seeds are widely used medicinally. Their contains about 36-48% oil content which is high in

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unsaturated fatty acids, with esters of linoleic acid, linolenic acid, stearic acid and oleic acid; also mucilage, proteins, a cyanogenic glycoside (linamarin) and enzymes [3-5]. Crushed seeds mixed to a paste with water are used to make hot poultices to relieve pain and to heal septic wounds, skin rashes and ulcers. The extracted oil is used in the pharmaceutical industry to make liniments for burns and rheumatic pain. The oil is also important in the manufacture of paints, soap and printer's ink. Inclusion of linseed in the diet results in increased polyunsaturated fatty acids [6, 7], which protect the body against cancer [8], reduce the chances of cardiovascular diseases and certain other health related problems [9]. So, linseed may play a vital role in reducing these health risk problems due to its nutritive value which depends upon different factors like cultivar, locality, planting date, fertilizers and year of production [10].

In Iran, flax is cultivated as a dual purpose (seeds for oil and stems for fiber). The cultivated area through the last 20 years was decreased due to the great competition of other economic winter crops resulting in a gap between production and consumption. Therefore, it is necessary to increase flax productivity per unit area which could be achieved by using high yielding cultivars and improving the agronomic factors [11,12]. One of these factors is planting date which is important factor for vigorous growth and consequently higher yield of different plant species [13,14,11].

Early planting of flax is enough to allow the plants to become well established before freezing temperatures occur. After plants have branched at the crown, cold-hardy varieties can withstand much lower temperatures without serious injury. Avoid planting flax too early in the fall or late spring frosts may damage the crop at the bloom stage. In South Texas, when moisture conditions permit, plant between November 10 and December 10. In this area, flax seeded after January 1 usually produced lower yields than December seeding. If moisture is available, October 15 to October 30 is an ideal time for planting flax in the temperate area.

In western Canada, flax (*Linum usitatissimum* L.) is often sown in early May when soils are usually cool and moist [15]. This practice has been adopted so that the crop flowers early to escape the hot, dry conditions generally experienced in mid to late July. Cool temperatures combined with a long photoperiod during and after flowering increases both seed yield and the production of linolenic acid. Unfortunately, seeding flax into cool wet soils can delay germination and result in a poor stand due to the invasion of seeds by soil fungi. Plants emerging under these conditions have reduced vigourity and yield potential. Also, if emergence is delayed due to low soil temperatures and/or deep seeding, soil incorporated herbicide residues such as trifluralin or triasulfuron from a previous crop can injure and reduce the emerging seedlings [15]. The importance of early stand establishment has been documented in several studies. Ford [16] demonstrated that the number of seeds per capsule as well as the number of capsules per plant was greatly reduced if seeding was delayed. A delay in seeding also resulted in a reduction in oil content, iodine value and linolenic acid [17]. Sosulski and Gore [18] reported that oil content, iodine number and linolenic acid increased under long photoperiods whereas short photoperiods resulted in delayed flowering and reduction in both oil and fatty acid content.

Responses to fertilizer applications depend upon moisture conditions, previous crop and fertility status of the soil. Experimental data generally show fertilizer applications to be profitable under good management and favorable moisture. When large amounts of low nitrogen residue, such as grain sorghum stubble, are returned to the soil before planting flax, apply 1.5 Kg of nitrogen per expected bushel of yield. A strong interaction of S and N for seed yield was found in rapeseed and mustard [19-25], sunflower [26] linseed [27] Groundnut [28,29] and Soybean [30,31].

Due to the environmental conditions of Iran, oil flax crop is more adaptable to fiber one. High temperatures and intense radiation in fact favor seed production and oil accumulation. Moreover, at high temperatures the oil is richer in saturated glycerides while at low temperatures and less intense radiation the plants tend to remain in the vegetative phase yielding more fiber of better quality. The average crop yields about 1 t ha^{-1}, but experimental trials proved that the best varieties can produce up to 3 t ha^{-1} of seed with 40% of oil content and 20% of proteins. The objectives of this study were to determine the effects of optimum nitrogen fertilizer and planting date on various plant and seed characteristics for a new flax cultivar.

MATERIAL AND METHODS

The studies were conducted at Agricultural Researches Station of Islamic Azad University of Yasouj, (51°, 41' E, 30°50'*N*, 1832 m above sea level) during 2006-2008 (Fig 1). The experiment was complete randomized block, split plot design with three replications. Main plots, planting date, were Mar. 5, Mar. 20, Apr. 3, Apr. 18 and

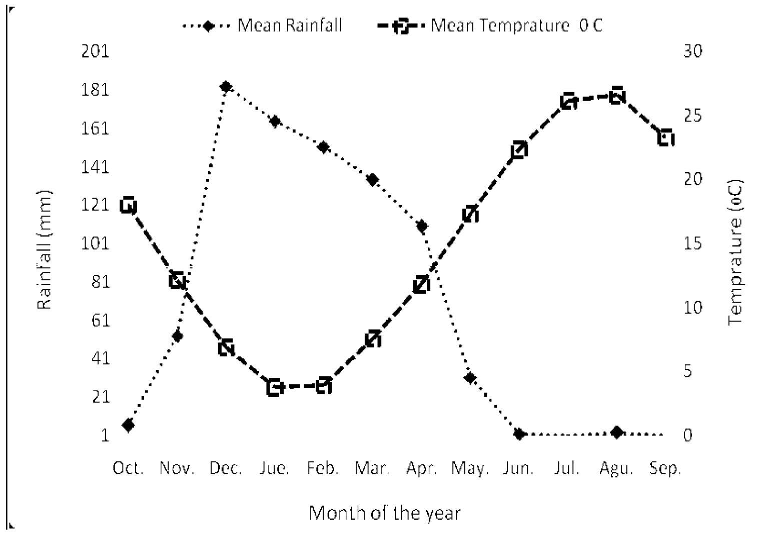

Fig. 1: Monthly values of mean temperature and precipitation from planting to the end of Sep. for 2006-2008.

Table 1: Soil physical and chemical characteristics of the experimental site

Variable	Year 2006-2007	2007-2008
Texture	Si-C	S-C-L
PH	7.5	7.85
EC dS m^{-1}	0.9	1.0
Organic matter (%)	0.92	0.69
N(%)	0.088	0.076
P mg Kg^{-1}	13.4	16
K mg Kg^{-1}	340	206
Fe	3.7	5.4
Zn	0.64	0.1
Mn	5.8	9.9
Cu	0.48	0.98

May. 4. Sub plots, fertilizer treatment, were N at rates of 0, 50,100 and 150 kg ha^{-1}. The cultivar used in this experiment was Olay ozon. To determine the soil characteristics 15 samples from 30 cm depth were collected and analyzed by Shiraz Soil Testing Laboratory for basic soil physical and chemical properties (Table 1). P and K fertilizer were applied according to recommendations of Soil Testing Laboratory of Shiraz Agricultural Researches Station in forms of superphosphates and potassium sulfate, respectively. Plots were sown with a cone seeder and were 6 m long and 2.4 m wide, with 12 rows 0.2 m apart. Plant density was 570000 plants ha^{-1}. Plots were plowed and disked before planting in July. The plots were disked again before seeding in February. Irrigation of each main plot was measured volumetrically by field calibrated gypsum block. The Ψsoil at 30-cm depth was kept-0.025 MPa to maturity. The irrigation system was operated so that runoff did not occur. At maturity, 10 plants were taken randomly from each subplot for recording the following morphological, yield components and yield.

The various traits on the linseed plant that are discussed in this paper were evaluated as fallows:

- Plant height (cm); from the cotyledonary node till the top of the plant.
- Number of capsules per plant.
- Specific seed weight (average weight of 1000 seeds in grams); recorded on 10 random samples from each sub-plot.
- Straw yield (kg/ha); weight of air dried straw.
- Seed yield. Center eight rows (of 12 rows) of each plot were harvested for grain yield and converted to grain yield per hectares.
- The oil content of a sample of whole seeds from each plot was determined by Near-Infrared Reflectance Spectroscopy as described by Bhatty [32].
- Oil yield (kg/ha); calculated by multiplying seed oil percentage ×seed yield per ha.
- Grain protein percent was estimated by Kjeldhal method and crude protein (CP) was calculated by multiplying nitrogen (%) with factor 6.25 [33].

Samples were dried in a forced-air oven at 70° C for 48 h.

After determination of the oil yield (in the dry matter), fatty acids were esterified as methyl esters [33] and analyzed by Agilent 6890 N GC equipped with a DB-23 capillary column (60 m x 0.25 µm) and a FID (Flame Ionization Detector) detector. The carrier gas was helium, at a flow rate of 1.2 mL/min. Both injector and detector temperatures were kept at 250°C. Column temperature was initially kept at 165°C for 15 min and then increased to 200°C at a rate of 5°C/min, where it was maintained for 15 min. Samples of 1 μl were injected by auto-sampler, in the split mode (1:50). The fatty acid identification was performed by retention time comparisons with corresponding fatty acid methyl ester standards. The standards were purchased from Sigma-Aldrich Ltd.

Data were analyzed by analysis of variance [34].When significant differences were found ($P=0.05$) among means, Duncan's multiple range test (DMRT) were applied.

RESULTS AND DISCUSSION

Plant Height: There was significant difference observed in year, planting dates, nitrogen fertilizer and interaction between them in this trait (Table 2). The highest plant height was related to 150 kg N ha^{-1} and Mar. 5 and 20, resulting to lodging in some plots (Table 3 and 4). Early planting date with applying 150 kg N ha^{-1}, plants are expected to have a long growth period and continuous supply of N during growth stage. This condition is probably led to more plant height, thus increasing in lodging. Garsid [35] observed that delay in planting date resulted in significant decrease in the stem length.

Number of Capsule per Plant: Both planting date and nitrogen fertilizer were significant in this trait (Table 2). The data (Table 3) indicated that the highest capsule per plant obtained by Mar. 5, 20 and 100 or 150 kg N ha^{-1}. Apr.18 and May.4, with the same statistical groups, produced the lowest values. Significant interactions at 5% probability level were detected in year × nitrogen fertilizer and planting date × nitrogen fertilizer (Table 4).

1000-kernel Weight: Year and planting dates had significant effect on kernel weight in 1 % probability level (Table 2). The highest value of kernel weight was obtained in Mar. 5 and 20 and the lowest value were belonged to May.4. At later planting date, because of confronting the grain filling period to high temperature at the end of growing season kernel weight severely decreased (Table 3). Significant planting date × nitrogen fertilizer interactions were detected at $P \geq 0.01$ in this trait (Table 4). Dubey [36] concluded that delay in planting date caused the decrease in 1000-kernel weight, oil rate and grain yield.

Straw Yield: Main effects of treatments had significant effect on straw yield in 1 % probability level (Table 2). Mar. 5 and 100 and 150 kg N ha^{-1} produced the highest straw yield (Table 3). After this planting date, straw yield decreased to 60% at May.4. Straw yield showed significant interactions between year × nitrogen fertilizer and planting date × nitrogen fertilizer. As, Mar. 5 and 100 kg N ha^{-1} was the best combination to produce the highest straw yield. Hassan and Leitch [37] reported that early planting date of flax produced high amount of dry matter due to severe competition between plants.

Harvest Index: There was significant difference between year and planting dates treatments in this trait (Table 2). Mar.5 planting date was more successful than other treatments to transport of assimilate from sources to plant sinks (Table 3). Significant planting date × year and planting date × nitrogen fertilizer interactions were detected at $P \geq 0.05$ on harvest index. Mar. 5 and 100 kg N ha^{-1} produced the highest value.

Table 2: Summary of combined analysis of variance across years, planting date and N fertilizer on flax

Sources	df	Plant height (cm)	Number of capsule $plant^{-1}$	1000-Kernel weight	Straw yield	Grain yield	Harvest Index	Oil percent	Oil Yield	Grain protein	Grain protein yield	Linolenic acid	Linoleic acid	Oleic acid
Year (Y)	1	**	**	**	NS	**	*	NS	**	NS	NS	NS	NS	**
Planting Date (S)	4	**	**	**	**	**	**	**	**	**	**	**	NS	NS
Y×S	4	NS	NS	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS
Nitrogen (N)	3	**	**	NS	**	**	NS	**	**	**	**	**	**	**
Y×N	3	**	*	NS	**	NS	NS	**	NS	NS	NS	NS	**	*
S×N	12	**	*	*	**	**	*	*	**	**	**	**	NS	*
Y×S×N	12	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
CV, %		8.9	10.7	7.5	15.2	10.7	12.5	9.35	13.5	9.3	18.2	13.1	14.8	11.5

* and, ** Significant at 0.05 and 0.01 probability levels, respectively. NS=nonsignificant at $P > 0.05$.

Table 3: Mean comparison of traits of linseed cultivar at five planting date and four N fertilizers to both years

Treatments	Plant height (cm)	Number of capsule plant^{-1}	1000-Kernel weight (g)	Straw yield	Grain yield	Harvest Index (%)	Oil percent	Oil Yield Kg ha^{-1}
Planting date								
Mar. 5	67 A	51 A	6.1 A	385 A	1371 A	41 A	32.6 A	431 A
Mar. 20	67 A	50 A	5.9 A	375 B	1277 B	36 B	32.2 A	403 B
Apr. 3	60 B	40 B	4.7 B	316 C	1134 C	27 C	29.8 B	317 C
Apr.18	54 C	26 C	3.9 C	254 D	706 D	23 D	26.7 BC	187 D
May. 4	54 C	25 C	3.0 D	238 E	603 E	19 E	24.4 C	141 E
Nitrogen(N) Kg ha^{-1}								
0	49 D	27 C	4.7 A	227 C	459 C	27 A	30.5 A	151 C
50	56 C	34 B	4.7 A	287 B	584 B	28 A	30.5 A	181 B
100	66 B	46 A	4.7 A	370 A	1518 A	29 A	27.7 B	426 A
150	71 A	46 A	4.8 A	369 A	1511 A	29 A	26.8 B	425 A

Treatments	Grain protein (%)	Grain protein yield Kg ha^{-1}	Linolenic acid (%)	Linoleic acid (%)	Oleic acid (%)
Planting date					
Mar. 5	22.2 C	320 A	52.4 A	15.1 A	20.3 A
Mar. 20	22.1 C	302AB	52.2 AB	15.1 A	20.1 A
Apr. 3	23.7 B	284 B	52.1 B	14.9 A	19.9 A
Apr. 18	25.6 A	188 C	50.1 C	14.8 A	20.2 A
May. 4	25.4 A	158 D	50.6 C	14.6 A	19.9 A
Nitrogen(N) Kg ha^{-1}					
0	19.9 D	89 D	50.3 D	14.6 B	20.6 A
50	22.7 C	130 C	51.1 C	14.7 B	20.3 B
100	24.7 B	355 B	52.2 B	15.1 A	19.8 C
150	28.1 A	427 A	52.6 A	15.4 A	19.5 D

Table 4: Interaction between planting date and N fertilizers on mean values traits in both years

Treatments		Plant height (cm)	Number of capsule plant^{-1}	1000-Kernel weight (g)	Straw yield --------Kg ha^{-1}	Grain yield ----------	Harvest Index (%)	Oil percent
Planting date	Nitrogen Kg ha^{-1}							0
	54 E	37 C	6.2 A	281 G	625 EFG	40 A	35 A	
Mar. 5	50	63 D	46 B	6.1 A	354 D	736 E	41 A	35 A
	100	73 B	60 A	6.1 A	453 A	2087 A	41 A	32 ABCD
	150	79 A	60 A	6.1 A	451 B	2039 A	41 A	29 DEFG
	0	52 E	37 C	6.0 A	264 H	605 FG	36 B	34 AB
Mar. 20	50	66 CD	45 B	6.0 A	341 E	734 E	36 B	34 ABC
	100	71 B	59 A	6.0 A	451 AB	1901 B	36 B	31 BCDE
	150	79 A	59 A	6.0 A	444 B	1867 B	37 B	29 DEFG
	0	50 E	29 D	4.8 B	222 IJ	520 GH	27 C	30 CDEF
Apr. 3	50	55 F	37 C	4.7 B	301 F	675 EF	27 C	31 BCDE
	100	64 D	47 B	4.7 B	371 C	1646 C	27 C	28 EFGD
	150	70 BC	47 B	4.7 B	369 C	1696 C	27 C	27 FGHI
	0	43 F	17 F	4.0 C	194 K	198 I	23 D	27EFGHF
Apr. 18	50	50 E	23 E	4.0 C	224 I	464 H	23 D	28 DEFG
	100	61 D	31 D	3.9 C	296 F	1001 D	23 D	26 GHID
	150	62 D	33 CD	4.1 C	303 F	1008 D	23 D	26 GHI
	0	43 F	17 F	2.9 D	175 L	198 J	19 E	26 GHI
May. 4	50	49 E	20 EF	3.0 D	215 J	314 I	19 E	25 GHIF
	100	62 D	30 D	3.1 D	281 G	954 D	19 E	23 I
	150	54 E	33 CD	3.2 D	280 G	946 D	19 E	24 HI

Table 4: Continued

Treatments		Oil Yield Kg ha^{-1}	Grain protein (%)	Grain protein yield Kg ha^{-1}	Linolenic acid (%)	Linoleic acid (%)	Oleic acid (%)
Planting date	Nitrogen Kg ha^{-1}						
	0	225 FG	18.1 I	113 GHI	51.4 DE	14.8 A	21 A
Mar. 5	50	250 EF	21.5 GH	156 G	52 CD	15.2 A	20.6 AB
	100	644 A	22.7 FG	460 BC	52.9 AB	15.1 A	20 BCDE
	150	607 AB	26.3 CDE	551 A	53.3 A	15.5 A	19.5 DEF
	0	216 FG	17.9 I	108 GHI	50.7 EF	14.9 A	20.6 AB
Mar. 20	50	245 EFG	21.5 GH	158 G	51.7 CD	14.8 A	20.3 ABC
	100	587 BC	22.9 FG	435 C	52.9 AB	15.3 A	19.7 CDE
	150	563 C	26.1 CDE	507 AB	53.4 A	15.5 A	19.3 FG
	0	164 HI	20.0 HI	104 HI	50.5 F	14.5 A	20.5 ABC
Apr. 3	50	202 GH	22.2 FGH	150 GH	51.4 DE	14.8 A	20.2ABCD
	100	443 D	24.2 DEF	388 D	52.3 BC	14.9 A	19.6 DEF
	150	457 D	28.5 ABC	492 B	52.9 AB	15.3 A	19.2 G
	0	99 JK	21.6 FGH	75 IJ	49.3 G	14.2 A	20.5 AB
Apr. 18	50	128 IJ	24.1 DEF	111 GHI	50.7 EF	14.7 A	20.3 ABC
	100	250 EF	26.6 BCD	256 F	51.7 CD	15.2 A	20.1 BCD
	150	272 E	30.0 A	311 E	51.9 CD	15.4 A	19.9BCDE
	0	53 L	21.8 FGH	43 J	49.6 G	14.4 A	20.3 ABC
May. 4	50	79 KL	24.1 EFG	76 IJ	49.8 G	13.9 A	20.1BCDE
	100	206 FGH	26.9 BC	237 F	51.3 DE	14.9 A	19.7 CDE
	150	225 FG	29.0 AB	274 EF	51.7 CD	15.2 A	19.4 EFG

Grain Yield: Grain yield after Mar.5 was reduced by 8 to 130 % (Table 3). Grain yield increased in response to applied N, with the grain yield of the crop that no received N being three fold than for that receiving 150 kg N ha^{-1} (Table 3). However, Increasing in grain yield beyond 100 kg N ha^{-1} was marginal. This dose not implies that there was no response to the application of higher rates of N at different planting dates. Among the five planting dates, Mar. 5 produced the highest grain yield at either N level (Table 4) and the doubling of N fertilization increased grain yield by 1351 kg ha^{-1}. A hundred kilograms of N ha^{-1} was adequate for maximum grain yields with Mar.5 planting date. The N response data may be used in determining N fertilizer rates for specific yield goals. Planting date × N interaction was only significant in this trait. High temperatures during the period of reproductive growth lead to decline of reproductive growth period, failure in the number of crop and finally decrease the number of seed and then the decline comes to grain yield. Therefore, in most studies the grain yield decreases with the delay in planting [38].

Oil Percent and Oil Yield: Planting date, nitrogen fertilizer and interaction between them were significant in oil percent and oil yield (Table 2). Mar. 5 produced the highest oil seed and oil seed yield. Delay in planting date after Mar. 5 decreased oil seed about 33% at May.4 (Table 3). Increasing in nitrogen fertilizer decreased oil seed content (Table 3). However, oil seed yield increased with applying in nitrogen fertilizer, due to increase in grain yield. The oil content ranged 24.4 to 32. 6% and was average 30% in the approximately 50% of the material used. Generally, linseed contains about 40% oil in the seed [39]. Oil content of linseed recorded in this study was lower compared to those reported by Hume [40] and Kouba [4].

Grain Protein and Grain Protein Yield: Both planting date and N fertilizer had significant effect of grain protein percent and grain protein yield (Table 2). Delay in planting date resulted in increase grain protein percent up to 15 % at May.4 than Mar. 5 planting date (Table3). Because of shorter growth period at late planting dates, starch accumulation was low. Therefore, grain protein content increased. Conversely, the highest grain protein yield, obtained at Mar. 5 due to increase in grain yield. The crop received 150 kg N ha^{-1} produced 41 % more protein than the crop received 0-N (Table 3). Planting date × N interaction was only significant in this trait. Apr. 18 + 150 kg N ha^{-1} and Mar.5 and 150 kg N ha^{-1} were the best combinations for grain protein and grain protein yield, respectively.

Fatty Acids: Table 3 and Table 4 show summary of the fatty acid composition and percentage of unsaturated fatty acids. The unsaturated fatty acid contents varied from 14.6 to 52.6 %. Generally, linseed oil contains approximately 9-11% saturated (5-6% palmitic acid and 4-5% stearic acid) and 75-90% unsaturated fatty acids (50-55% linolenic acid, 15-20% oleic acid) [41]. The highest linolenic acid obtained in Mar. 5 with average 52.4%. However, there were no differences in linoleic acid and oleic acid with change in planting date (Table 2). linolenic acid and linoleic acid increased with increasing in nitrogen fertilizer. Conversely, oleic acid content decreased with increasing in nitrogen fertilizer (Table 3). The best combination in order to obtain the heighest linolenic acid and oleic acid was Mar.5 +150 kg N ha^{-1} and Mar.5 +0 kg N ha^{-1}, respectively (Table 4). Temperature is generally considered to be the most critical climatic factor affecting oil yields and quality [42]. Carder [43 has shown that day length (photoperiod) is also an important factor responsible for the differential growth behavior of grain crops when grown at two diverse latitudes.

CONCLUSION

The results of this study demonstrate that Mar. 5 planting date was the best date for increasing all traits except grain protein percent. 100 or 150 kg N ha^{-1} also increased all traits except oil percent and oleic acid of linseed. It recommends carry out the experiment under irrigation levels, soil types and other conditions.

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