

Sulphur doses and application times on yield and oil quality of canola grown in calcareous soil

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SUMMARY: Pakistan has been constantly deficient in its oil seed production and it is very difficult to meet the edible oil requirement of its ever-increasing population. A field experiment was conducted at the Agronomy Research Farm, The University of Agriculture Peshawar, Northern Pakistan during winter (2013–14). Five sulphur levels (15, 30, 45, 60 and 75 kg·ha⁻¹) and times of application (at seedling, bolting and flowering stages) were used for the canola variety Abasin-95. The experiment was laid out in a randomized complete block design replicated four times on a 5 m × 3.2 m plot size. The results showed that the sulphur-applied plots gave the highest seed yield, biological yield, glucosinolate, erucic acid, oil content, protein content, oleic acid and linoleic acid compared to the control plots. Sulphur applied at the rate of 60 kg·ha⁻¹ and applied at the bolting stage increased seed yield, biological yield, oil content, and protein content.

KEYWORDS: *Canola; Linoleic acid; Oil; Oleic acid; Production; Protein; Sulphur*

RESUMEN: *Dosis de azufre y tiempos de aplicación en el rendimiento y la calidad del aceite de canola cultivada en suelo calcáreo.* Pakistán ha sido constante y crónicamente deficiente en producción de semillas oleaginosas y es muy difícil cumplir con la demanda de aceites comestibles para una población cada vez mayor. Se realizó un experimento de campo en la granja de investigación agronómica de la Universidad de Agricultura Peshawar, en el norte de Pakistán, durante el invierno (2013–14). Los cinco niveles de azufre (15, 30, 45, 60 y 75 kg·ha⁻¹) y su tiempo de aplicación (en las etapas de plántula, floración y floración) se utilizaron para la variedad de canola (Abasin-95). El experimento se realizó en un diseño de bloques completos al azar que se replicó cuatro veces con un tamaño de parcela de 5 m × 3,2 m. Los resultados mostraron que las parcelas aplicadas con azufre dieron el mayor rendimiento de semilla, rendimiento biológico, glucosinato, ácido erúico, contenido de aceite, contenido de proteína, ácido oleico y ácido linoleico en comparación con las parcelas de control. El azufre se aplicó en una tasa de 60 kg·ha⁻¹ y se aplicó en la etapa de empernado, incrementando el rendimiento de las semillas, el rendimiento biológico, el contenido de aceite y el contenido de proteínas.

PALABRAS CLAVE: *Aceite; Ácido linoleico; Ácido oleico; Azufre; Canola; Producción; Proteína*

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1. INTRODUCTION

Canola is an important oil seed crop of Cruciferae (Holmes, 1980). Rapeseed is grown as an oil seed crop and also as a fodder crop in the Indus Valley c. 300 BC (Wiess, 1983). Vegetable oils are rich sources of erucic acid and glucosinolate (sulphur compounds) in the tissues, which give a bitter taste which can be unpleasant and even toxic (Muhammad *et al.*, 1991). The desirable range of erucic acid and glucosinolate in canola oil are 2% erucic acid and 30 $\mu\text{mol}\cdot\text{g}^{-1}$ glucosinolates in oil (Grombacher and Nelson, 1992). After palm and soybean, Canadian plant breeders have reduced these compounds to an acceptable level by converting rapeseed species to canola as the world's third most commonly consumed vegetable oil (Rękas *et al.*, 2017). In agriculturally advanced countries, economic conditions demand higher crop yields to meet the high profit that comes from oil crops and population growth (Sawan *et al.*, 2007).

The average yield of canola in Pakistan is very low (922 $\text{kg}\cdot\text{ha}^{-1}$) and its average yield in northern Pakistan is 452 $\text{kg}\cdot\text{ha}^{-1}$ (MNFS&R, 2013–14). In Pakistan, canola is cultivated in an area of 238900 ha and in northern Pakistan in an area of 18800 ha. The annual seed production of canola is 220300 tonnes in Pakistan, while in northern Pakistan 8500 tonnes (MNFS&R, 2013–14). Canola was cultivated in an area of 586 thousand acres with seed production of 218 thousand tonnes and 68 thousand tonnes oil yield (GOP, 2014). In the north-west frontier of Pakistan (Khyber Pakhtunkhwa Province) canola mainly cultivated on marginal lands or river bank fields. Therefore, farmers cannot obtain maximum yield due to nutrient deficiencies and low soil fertility.

Fertilizers always play an important role in increasing many crop yields as a result of sufficient nutrition availability to the crops. Sulphur has prime importance in the synthesis of chlorophyll and oil in canola. It plays an important role in chemical composition as well as seed oil content (Hassan *et al.*, 2007). During deficient conditions of sulphur the crop production may not be sustainable and seriously affected by applied NPK fertilizer efficiency (Ahmad *et al.*, 1994). Previous studies have shown sulphur nutrition to have different effects on the seed production and oil quality of the canola crop when applied at different growth stages. The application of 20 $\text{kg}\cdot\text{ha}^{-1}$ sulphur at sowing time improved the oil content, glucosinolate and protein content of canola (Ahmad *et al.*, 1994; Jan *et al.*, 2008). Non destructive methods for the oil determination of rapeseed have the advantages of being rapid and relatively more environmentally friendly since they require minimum sample preparation (Uncu *et al.*, 2019).

Keeping in mind the importance of sulphur both for improving the seed production and oil quality of canola, the present research was aimed to assess the effects of sulphur nutrition levels and sulphur application timing on the yield and oil quality of canola in the calcareous soils of northern Pakistan.

2. MATERIALS AND METHODS

2.1. Experimental treatments and design

In order to study the effect of sulphur nutrition levels and time of application on seed yield and oil quality of canola, an experiment was undertaken at the Agronomy Research Farm, The University of Agriculture Peshawar (2013–2014). The experiment consisted of five sulphur levels (S1 = 15, S2 = 30, S3 = 45, S4 = 60, S5 = 75; $\text{kg}\cdot\text{ha}^{-1}$) and three different application times (AT1 = application at seedling growth stage, AT2 = application at bolting growth stage and AT3 = application at flowering growth stage). In addition, a control was maintained with each of the replicated treatments (no sulphur application was made). A randomized complete block design was used and replicated four times. Ammonium sulphate (NH_4SO_4) from the Fuji fertilizer private Ltd, Lahore, Pakistan was used as the source of sulphur. Canola seeds (Abasin-95) were provided by the Nuclear Institute for Food and Agriculture, Pakistan and sown in winter. 5 m \times 3.2 m plots were made and replicated, consisting of 8 rows with 0.4 m row-to-row distance. Phosphorus (50 $\text{kg}\cdot\text{ha}^{-1}$) was applied during sowing in the form of single super phosphate, and nitrogen (75 $\text{kg}\cdot\text{ha}^{-1}$) was applied in the form of urea (half dose during sowing time and half dose during flowering stage) after subtracting the amount of nitrogen supplied through ammonium sulphate. After complete emergence, hand thinning was done at the four leaf stage, maintaining 50 plants per row. All other cultural practices, including irrigation, weeding and hoeing etc. were carried out uniformly in all the plots. An average soil analyses of the experimental site showed a sandy loam texture, pH (7.4), EC (0.204 $\text{dS}\cdot\text{m}^{-1}$), bulk density (1.52 $\text{g}\cdot\text{cm}^{-3}$), moisture (7.8%), organic matter (0.50 $\text{mg}\cdot\text{kg}^{-1}$), phosphorous (0.45 $\text{mg}\cdot\text{kg}^{-1}$) and low sulphur (0.035 $\text{mg}\cdot\text{kg}^{-1}$) at 0–15 cm depth. Mean maximum, minimum temperature ($^{\circ}\text{C}$), humidity (%) and rainfall (mm) for the growing period of the canola crop are presented in Figure 1. The experiment was carried out from October 2013 to May 2014. Seed yield ($\text{kg}\cdot\text{ha}^{-1}$) was determined in each of the replicated plots where the four central rows were harvested at maturity and dried under sun light, manually threshed, weighed and converted to $\text{kg}\cdot\text{ha}^{-1}$.

Biological yield ($\text{kg}\cdot\text{ha}^{-1}$) was calculated after harvesting the four central rows in each of the replicated plots at harvest maturity of the canola crop, dried under sun light, weighed and converted in $\text{kg}\cdot\text{ha}^{-1}$.

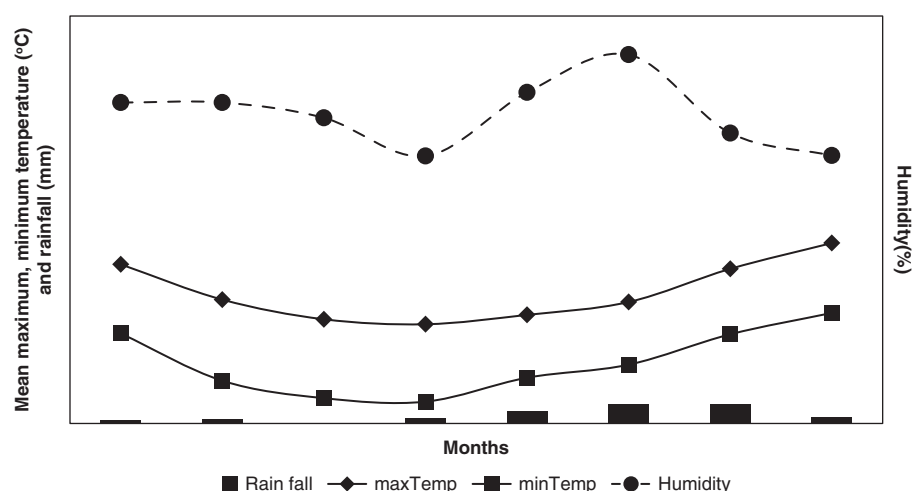


FIGURE 1. Mean minimum, maximum temperature (°C), humidity (%) and rainfall (mm) for the growing period of canola (2013–14).

$$\text{Biological yield (kg ha}^{-1}\text{)} = \frac{\text{Biological yield of the harvested rows}}{\text{Row - row distance} \times \text{row length} \times \text{no. of rows}} \times 10000$$

2.2. Seed quality parameters

To determine the glucosinolate ($\mu\text{ mol g}^{-1}$), erucic acid (%), oil content (%), protein content (%), oleic acid (%) and linoleic acid (%) in the canola seed, a five (5) gram sample obtained from each treatment plot was analyzed by a Near Infra-red Reflectance Spectroscopy System, (TR-3657-C Model 6500) as described by Ahmad *et al.*, (2015) at Oilseed Quality Laboratory, Crop Breeding Division, Nuclear Institute for Food and Agriculture Peshawar, Pakistan.

2.3. Statistical analysis

The recorded data was statistically analyzed according to the analysis of variance techniques used for randomized complete block design and least significant difference (LSD) was applied at a 5% level of significance ($P \leq 0.05$) upon significant F-test through the procedure described by Jan *et al.*, (2009).

3. RESULTS AND DISCUSSION

3.1. Weather data

During the growing season the temperature ranged from 11.8 °C minimum to 25.8 °C maximum (Figure 1). Relative humidity ranged from 52.5 to

72.4%, while total rainfall was recorded as 14.1 mm. It is clear that minimum and maximum temperature gradually decreased from October to January and then increased. Maximum rainfall was recorded in March (3.9mm) and April (3.8mm).

3.2. Seed yield (kg·ha⁻¹)

Seed is the ultimate output of a crop which determines the profitability of the crop production enterprise. Sulphur levels and time of sulphur application had significant effects on the seed yield of canola and their interaction was found to be non significant (Table 1). Sulphur application increased seed yield compared to the control. Higher seed yield (2452 kg·ha⁻¹) was recorded in plots where 60 kg·ha⁻¹ sulphur were applied as compared to the 15 kg·ha⁻¹ sulphur plots (1376 kg·ha⁻¹). These results are in agreement with Chauhan *et al.*, (1996), who reported that sulphur increased the seed yield of rapeseed. The findings of Begum *et al.*, (2012) revealed that sulphur up to 60 kg·ha⁻¹ application produced a higher seed yield of canola. Sulphur application timings had significantly affected the seed yield. More seed yield (1998 kg·ha⁻¹) was obtained at the bolting stage, while lower seed yield was obtained when sulphur was applied at seedling and bolting stages (1907 and 1922 kg·ha⁻¹, respectively). The crops grown in sulphur fertilized plots obtained higher seed yield (1942 kg·ha⁻¹), while lower seed yield (1134 kg·ha⁻¹) was recorded from the control plots. Canola seed yield was increased when sulphur was applied at the rate of 40 kg·ha⁻¹ (Varényiová *et al.*, 2017). The findings of Malhi and Gill (2002); Malhi and Leach (2000) indicated that the seed yield of canola was lower when sulphur was applied at the flowering stage compared to that obtained when sulphur was applied at the bolting stage. Hocking

et al., (1996) also found that sulphur applied at bolting resulted in the same yield although there was significant reduction in seed yield when sulphur application was delayed until the flowering stage. The interaction between S x AT revealed non significant effects on seed yield (Table 6).

3.3. Biological yield ($\text{kg}\cdot\text{ha}^{-1}$)

The data revealed that sulphur levels have a significant effect on the biological yield of canola (Table 1). Increasing sulphur rates up to $60 \text{ kg}\cdot\text{ha}^{-1}$ significantly increased the biological yield. Maximum biological yield ($11461 \text{ kg}\cdot\text{ha}^{-1}$) was obtained from $60 \text{ kg}\cdot\text{ha}^{-1}$ followed by $75 \text{ kg}\cdot\text{ha}^{-1}$ (Table. 3). A higher coefficient of variation (12.03) was seen in the biological yield (Table 7). Collectively, the highest biological yield ($10574 \text{ kg}\cdot\text{ha}^{-1}$) was recorded in the sulphur-treated plots compared to the control plots ($8469 \text{ kg}\cdot\text{ha}^{-1}$). The application timings and interaction of S x AT had no significant effect on biological yield. These results are comparable to those of Khandkar *et al.*, (1991) in that biological yield was enhanced significantly with the application of sulphur levels. Malik

et al., (2004) also reported that biological yield was significantly affected by sulphur levels.

3.4. Glucosinolate ($\mu\text{mol}\cdot\text{g}^{-1}$)

The glucosinolate content of canola as influenced by various sulphur levels and application times is presented in Table 2. The statistical analysis of data showed a significant effect of sulphur levels on the glucosinolate of canola. Average values for the data showed that significantly higher glucosinolate ($31.2 \mu\text{mol}\cdot\text{g}^{-1}$) was obtained from $75 \text{ kg}\cdot\text{ha}^{-1}$ treated plots, whereas minimum glucosinolate ($19.7 \mu\text{mol}\cdot\text{g}^{-1}$) was obtained from $15 \text{ kg}\cdot\text{ha}^{-1}$ plots. Application times and interaction of sulphur x application times was found non significant (Table 5). The sulphur levels showed a positive influence on the glucosinolate content of canola. Mailer (1989) reported that sulphur was essential up to some level for normal plant growth and affected the glucosinolate content of canola. It was also elaborated that a sulphur deficiency lowered glucosinolate content in control plots. Wang *et al.*, (1997) also determined that an increase in sulphur application enhanced the glucosinolate content of canola.

TABLE 1. Seed yield ($\text{kg}\cdot\text{ha}^{-1}$) and biological yield ($\text{kg}\cdot\text{ha}^{-1}$) of canola as affected by sulphur levels and application times

Application times (AT)	Seed yield ($\text{kg}\cdot\text{ha}^{-1}$)						Biological yield ($\text{kg}\cdot\text{ha}^{-1}$)					
	Sulphur ($\text{kg}\cdot\text{ha}^{-1}$)						Sulphur ($\text{kg}\cdot\text{ha}^{-1}$)					
	15	30	45	60	75	Mean	15	30	45	60	75	Mean
Seedling	1318	1540	1979	2421	2278	1907 ^b	9363	9121	10600	11548	10788	10284
Bolting	1469	1630	2023	2526	2342	1998 ^a	10198	10863	10025	11513	11850	10890
Flowering	1340	1581	1974	2408	2306	1922 ^b	9796	10125	10713	11325	10788	10549
Mean	1376 ^c	1584 ^d	1992 ^c	2452 ^a	2309 ^b		9785 ^b	10036 ^b	10446 ^{ab}	11462 ^a	11142 ^a	
Control	1134	Lsd	Sulphur	AT	SxAT		Control	8469	Lsd	Sulphur	AT	SxAT
Fertilized	1942	0.05	35.67	27.63	ns		Fertilized	10574	0.05	1033	ns	ns

S=Sulphur, AT= Application times, SxAT= Sulphur x Application times, ns=non significant, Lsd=Least significant difference test

TABLE 2. Glucosinolate ($\mu\text{mol}\cdot\text{g}^{-1}$) and erucic acid (%) of canola as affected by sulphur levels and application times

Application times (AT)	Glucosinolate ($\mu\text{mol}\cdot\text{g}^{-1}$)						Erucic acid (%)					
	Sulphur ($\text{kg}\cdot\text{ha}^{-1}$)						Sulphur ($\text{kg}\cdot\text{ha}^{-1}$)					
	15	30	45	60	75	Mean	15	30	45	60	75	Mean
Seedling	19.7	23.6	25.5	25.5	29.8	24.8	1.3	1.4	1.5	1.7	2.1	1.6
Bolting	20.0	23.8	23.8	25.5	31.3	24.9	1.3	1.5	1.5	1.6	2.1	1.6
Flowering	19.5	23.4	23.3	24.2	32.4	24.6	1.4	1.5	1.6	1.6	2.1	1.6
Mean	19.7 ^c	23.6 ^b	24.2 ^b	25.1 ^b	31.2 ^a		1.3 ^d	1.4 ^{cd}	1.5 ^{bc}	1.6 ^b	2.1 ^a	
Control	18.9	Lsd	Sulphur	AT	SxAT		Control	1.2	Lsd	Sulphur	AT	SxAT
Fertilized	24.7	0.05	1.68	ns	ns		Fertilized	1.6	0.05	0.11	ns	ns

S=Sulphur, AT= Application times, SxAT= Sulphur x Application times, ns=non significant, Lsd=Least significant difference test
Means of same category followed by different alphabets reveal significant differences among mean values ($P < 0.05$) using least significant difference test.

The 60 kg·ha⁻¹ sulphur application enhanced the glucosinolate content of canola, the safe and acceptable limit, which is 30 µmol·g⁻¹. Sulphur is involved in the synthesis of chlorophyll and also required in Cruciferae for the synthesis of volatile oil which accumulates as glucosinolate (Marschner 1986 and Ahmad *et al.*, 2007).

3.5. Erucic acid (%)

The mean values for the data showed that sulphur application enhanced erucic acid (1.6%) compared to the control (1.2%). Significantly higher erucic acid (2.1%) was recorded for 75 kg sulphur·ha⁻¹ (Table. 2). The application times and the interaction of sulphur x application times was found to be non significant for the erucic acid (%) of canola seeds. However, erucic acid gradually increased with increasing doses of sulphur (Table 2). Manaf and Fayyaz (2006) reported that erucic acid was at its maximum when the highest dose of sulphur (25 kg·ha⁻¹) was applied. Non-essential fatty acids like palmitic, stearic and erucic acid were increased in the canola seed with decreased sulphur levels (Begum *et al.*, 2015).

3.6. Oil content (%)

The sulphur levels significantly affected the oil content in canola seeds (Table 3). The comparison of sulphur fertilized plots with the control plots showed that the sulphur fertilized plots obtained significantly higher oil contents (44.1%) than the control plots (41.1%) (Table 3). The oil content was enhanced from 42.8 to 45.2% as the sulphur rate was boosted up to 60 kg·ha⁻¹. Kumar and Trivedi, (2012) recorded increasing oil percentages with increasing sulphur applications. However, a further sulphur application of up to 75 kg·ha⁻¹ did not enhance the oil content. Application timings and the interaction of S x AT had no significant affect on the oil content. Similar results were obtained in earlier

studies by Mailer *et al.*, (1989), who concluded that sulphur application enhanced the oil content of canola. The oil contents of canola were enhanced by sulphur application up to 30–50 kg·ha⁻¹ (Subhani *et al.*, (2003); Malhi and Leach, 2000). The control plots showed reduced oil contents compared to the 60 kg·ha⁻¹ sulphur-applied plots. Higher oil content was in sulphur-applied plots, as oil contains many fatty acids and these fatty acid structures contain sulphur compounds (Malik *et al.*, 2004).

3.7. Protein content (%)

The seed protein content (%) of canola is the basic parameter with respect to the quality. The statistical analysis of the data indicated that sulphur levels showed a positive influence on protein content (%) (Table 3). The 60 kg·ha⁻¹ sulphur application resulted in higher seed protein content (24.8%) in canola. Protein content was increased from 22.4% to 24.8% when sulphur levels increased from 15 to 60 kg·ha⁻¹, while protein content decreased (23.6%) when the sulphur level was further increased up to 75 kg·ha⁻¹. Canola seed protein and oil content were increased with increasing doses of sulphur (Kandil and Nadia, 2012). The crops grown without sulphur had lower protein contents (20.6%) than the sulphur-treated plots (23.4%). The time of application and interaction of S x AT had no significant effect on the protein content in the seeds (Table. 5). The sulphur application enhanced the seed protein contents of canola due to the protein content of rapeseed containing large amounts of amino acids such as cysteine and methionine which are constituents of sulphur (Holmes, 1980). These results confirm the findings of Rashid (1996) in that amino acids (methionine, cystine and cysteine) contain sulphur as an important structural part and therefore essential protein synthesis occurs due to sulphur. The results obtained regarding the response of sulphur application to the protein content of canola seed are also in line with the earlier findings of Wang

TABLE 3. Oil content (%) and protein content (%) of canola as affected by sulphur levels and application times

Application times (AT)	Oil content (%)						Protein content (%)					
	Sulphur (kg·ha ⁻¹)						Sulphur (kg·ha ⁻¹)					
	15	30	45	60	75	Mean	15	30	45	60	75	Mean
Seedling	43.6	43.4	45.6	45.9	44.2	1907 ^b	22.6	23.5	23.3	24.9	23.5	23.5
Bolting	41.6	43.5	45.0	44.8	43.8	1998 ^a	22.6	22.4	23.7	24.6	23.3	23.3
Flowering	43.3	43.5	44.0	45.0	44.2	1922 ^b	22.0	22.4	23.3	24.9	24.1	23.3
Mean	42.8 ^c	43.5 ^{bc}	44.8 ^{ab}	45.2 ^a	44.0 ^{abc}		22.4 ^c	22.8 ^{bc}	23.4 ^{bc}	24.8 ^a	23.6 ^{ab}	
Control	41.1	Lsd	Sulphur	AT	SxAT		Control	20.6	Lsd	Sulphur	AT	SxAT
Fertilized	44.1	0.05	1.45	ns	ns		Fertilized	23.4	0.05	1.22	ns	ns

S=Sulphur, AT= Application times, SxAT= Sulphur x Application times, ns=non significant, Lsd=Least significant difference test

et al., (1997) who stated that sulphur applied at up to 40 kg·ha⁻¹ increased the protein content of canola seeds.

3.8. Oleic acid (%)

The data on the oleic acid (%) of canola as affected by sulphur levels, application timings and their interaction are given in Table 4. The control plots resulted in less oleic acid (52.65%) as compared to the sulphur fertilized plots (55.82%). The statistical analysis of the data showed a non

significant effect of sulphur levels and application timing on the oleic acid (%) of canola. Ahmed and Abdin (2000), concluded that sulphur levels had no significant effect on oleic acid. However, oleic acid (55.23% to 56.18%) was increased with increasing sulphur doses from 15–75 kg·ha⁻¹. Maximum oleic acid (229.6 mg·g⁻¹) was obtained from sulphur-treated plots as compared to the control (Shoja *et al.*, 2018). Maximum oleic acid contents were observed at 60 kg·ha⁻¹ when applied at the bolting stage. Oleic acid was increased when sulphur doses were increased from 30–60 kg·ha⁻¹ (Ray *et al.*, 2015).

TABLE 4. Oleic acid (%) and linoleic acid (%) of canola as affected by sulphur levels and application times

Application times (AT)	Oleic acid (%)						Linoleic acid (%)					
	Sulphur (kg·ha ⁻¹)						Sulphur (kg·ha ⁻¹)					
	15	30	45	60	75	Mean	15	30	45	60	75	Mean
Seedling	54.33	56.40	55.63	55.05	56.20	55.52	10.7	10.6	10.4	10.5	10.4	10.5
Bolting	55.90	55.40	56.08	56.38	56.78	56.11	10.3	10.5	10.6	10.9	10.5	10.5
Flowering	55.48	54.85	56.38	56.98	55.55	55.85	9.9	10.1	10.8	10.9	10.3	10.4
Mean	55.23	55.55	56.03	56.13	56.18		10.3	10.4	10.6	10.8	10.4	
Control	52.65	Lsd	Sulphur	AT	SxAT		Control	9.5	Lsd	Sulphur	AT	SxAT
Fertilized	55.82	0.05	ns	ns	ns		Fertilized	10.5	0.05	ns	ns	ns

S=Sulphur, AT= Application times, SxAT= Sulphur x Application times, ns=non significant, Lsd=Least significant difference test. Means of same category followed by different alphabets reveal significant differences among mean values ($P < 0.05$) using least significant difference test.

TABLE 5. Analysis of variance for seed yield (kg·ha⁻¹), biological yield (kg·ha⁻¹), glucosinolate (μmol·g⁻¹), erucic acid (%), oil content (%), protein content (%), oleic acid (%), and linoleic acid (%) of canola as affected by sulphur levels and application times

Source of variance	D.F.	Seed yield (kg·ha ⁻¹)	Biological yield (kg·ha ⁻¹)	Glucosinolate (μmol·g ⁻¹)	Erucic acid (%)	Oil content (%)	Protein content (%)	Oleic acid (%)	linoleic acid (%)
Rep	3	1078.52	1745287.04	4.19	0.01	6.04	2.44	2.39	0.08
Treatments (T)	15	847831.51**	3518813.50*	64.92**	0.33**	6.50*	5.07*	4.54**	0.52*
Control Vs Fertilized	(1)	2450235.16**	16611081.67**	128.12**	0.54 ^{ns}	33.71**	28.53**	37.76**	3.58**
Sulphur Levels (S)	(4)	2537405.12**	6112988.54**	204.12**	1.06**	11.46*	10.29**	2.05 ^{ns}	0.42 ^{ns}
Application timings (AT)	(2)	47777.06**	1843977.92 ^{ns}	0.46 ns	0.01 ^{ns}	3.33 ^{ns}	0.37 ^{ns}	1.72 ^{ns}	0.13 ^{ns}
Sulphur x Application time	(8)	2757.86 ^{ns}	1003901.35 ^{ns}	3.53 ^{ns}	0.01 ^{ns}	1.42	0.71	2.34 ^{ns}	0.28
Error	45	1881.54	1578991.76	4.20	0.02	3.11	2.20	1.84	0.25
Total	63								

* = Significant at 5% level of probability, ** = Significant at 1% level of probability, ns = non significant, all parameter presents their sum of squares

TABLE 6. Co-efficient of variance (C.V) for seed yield (kg·ha⁻¹), biological yield (kg·ha⁻¹), glucosinolate (μmol·g⁻¹), erucic acid (%), oil content (%), protein content (%), oleic acid (%), and linoleic acid (%) of canola as affected by sulphur levels and application times

Parameters	Seed yield	Biological yield	Glucosinolate	Erucic acid	Oil content	Protein content	Oleic acid	Linoleic acid
C.V %	2.29	12.03	8.40	8.65	2.44	6.38	2.44	4.83

3.9 Linoleic acid (%)

Sulphur levels, time of application and their interaction have no significant effects on the linoleic acid of canola (Table 4). A lower linoleic acid content (9.5%) was found in the control plots than the plots fertilized with sulphur (10.5%). The results are in line with those of Malhi and Gill (2002), and Malhi and Leach (2000). Increasing doses of sulphur from 15–60 kg·ha⁻¹ increased linoleic acid (10.3–10.8). However, further a increase beyond 60 kg sulphur ha⁻¹ decreased the linoleic acid content. Ray *et al.*, (2015) reported that increasing sulphur doses from 30–60 kg·ha⁻¹ increased linoleic acid percentage. Manaf and Hassan 2006 concluded that linolenic acid was increased when sulphur fertilization increased from 15–25 kg·ha⁻¹. Sulphur applied at seedling and bolting stage induced more linoleic acid (10.5) compared to flowering stage.

4. CONCLUSIONS

The application of sulphur at the rate of 60 kg·ha⁻¹ produced higher yield and better oil quality in canola seeds. Sulphur application at the bolting stage produced a higher yield and oil components in canola seeds. It is recommended that sulphur be applied at the rate of 60 kg·ha⁻¹ in the bolting stage to enhance the yield and oil quality of canola.

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REFERENCES

- Ahmed A, Abdin MZ. 2000. Effect of sulphur application on lipid, RNA and fatty acid content in developing seeds of rapeseed (*Brassica campestris* L.). *Pl. Sci.* **150**, 71–76. [https://doi.org/10.1016/S0168-9452\(99\)00167-3](https://doi.org/10.1016/S0168-9452(99)00167-3)
- Ahmad G, Jan A, Arif M, Jan MT, Khattak RA. 2007. Influence of nitrogen and sulphur fertilization on quality of canola (*Brassica napus* L.) under rainfed conditions. *J. Zhejiang Univ. Sci.* **B 8**, 731–737. <https://doi.org/10.1631/jzus.2007.B0731>
- Ahmad N, Muhammad A, Munir I, Shah Z, Basir A. 2015. Characterization of rapeseed germplasm for various quality parameters. *American-Eurasian J. Agric. Environ. Sci.* **15**, 2466–2472.
- Ahmad N, Saleem MT, Rashid M, Jalil A. 1994. *Sulphur status and crop response in Pakistan soils*. National Fertilizer Development Center. Pub. No. 7/94. Planning and Development Division, Islamabad. PP. 1–5.
- Begum F, Hossain F, Mondal MRI. 2012. Influence of sulphur on morphophysiological and yield parameters of rapeseed (*Brassica campestris* L.). *Bangladesh J. Agric. Res.* **37**, 645–652.
- Begum F, Hossain F, Islam MM, Mondal MRI. 2015. Effect of sulphur on biochemical parameters of rapeseed (*Brassica Campestris* L.) in grey terrace soil. *Archives Agro. Soil Sci.* **62**, 261–276. <https://doi.org/10.1080/03650340.2015.1045496>
- Chauhan DR, Paroda S, Ram M. 1996. Response of Indian mustard (*Brassica juncea*) to biofertilizers, S and N fertilization. *Indian J. Agron.* **41**, 621–623.
- GOP. 2014. *Economic Survey Report*. (2013–14). Govt. of Pak. Finance Division, Islamabad. P. 1–18.
- Grombacher A, Nelson L. 1992. *Canola Production*. A University of Nebraska Neb Guide Publication, No. G92-1076-A. Cooperative Extension, Institute of Agriculture and Natural Resources, University of Nebraska-Lincoln.
- Kandil H, Gad N. 2012. Growth and oil production of canola as affected by different sulphur sources. *J. Basic. Appl. Sci. Res.* **2**, 5196–5202.
- Hassan FU, Manaf A, Qadir G, Basra SMA. 2007. Effects of sulphur on seed yield, oil, protein and glucosinolates of canola cultivars. *Int. J. Agric. Biol.* **9**, 504–508.
- Hocking PJ, Pinkerton A, Good A. 1996. Recovery of field-grown canola from sulphur deficiency. *Aust. J. Exp. Agric.* **36**, 79–85.
- Holmes MRJ. 1980. *Nitrogen; In Nutrition of the oilseed rape crop*. Applied Sci Pub Barking Essex England. P21–67.
- Jan A, Ahmad G, Jan T, Jamal M, Subhan F. 2008. Oil yields of canola as affected by N and S levels and methods of application under rainfed conditions. *Sarhad J. Agric.* **24**, 1–10.
- Jan MT, Shah P, Hoolinton PA, Khan MJ, Sohail Q. 2009. *Agriculture research: Design and Analysis*. Deptt. of Agronomy, NWFP Agric. Uni. Peshawar, Pakistan.
- Khandkar MR, Malviya PK, Nigam KB. 1991. Influence of N and S applications on mustard (*Brassica juncea* L.) in vertisol. *Res. Devel. Rep.* **8**, 189–190.
- Kumar R, Trivedi S.K. 2012. Effect of levels and sources of sulphur on yield, quality and nutrient uptake by mustard (*Brassica juncea*). *Progressive Agriculture, Internat. J.* **12**, 69–73.
- Mailer RJ. 1989. Effects of applied sulphur on glucosinolate and oil concentration in the seeds of rape (*Brassica napus* L.) and turnip rape (*Brassica rapa* L.). *Aust. J. Agric. Res.* **40**, 617–624.
- Malhi H, Leach D. 2000. *Restore canola yields by correcting sulphur deficiency in the growing season*. Proc. 12th annual meeting and conference “Sustainable farming in the new millennium”. Saskatchewan Soil Conservation Association, Regina, SK., Canada.
- Malhi SS, Gill KS. 2002. Effectiveness of sulphate-S fertilization at different growth stages for yield, seed quality and S uptake of canola. *Can. J. Plant Sci.* **82**, 665–674.
- Malik MA, Aziz I, Khan HZ, Wahid MA. 2004. Growth, seed yield and oil content response of canola (*Brassica napus* L.) to varying levels of sulphur. *Internat. J. Agric. Biol.* **6**, 1153–1155.
- Marschner H. 1986. *Functions macronutrients & micronutrients, Mineral nutrition of higher plants*. Academic Press Inc. London, U.K. pp. 1–674.
- Varényiová M, Ducsay L, Ryant P. 2017. Sulphur nutrition and its effect on yield and oil content of oilseed rape (*Brassica Napus* L.). *Acta Univ. Agric. Silv. Mende. Brun.* **65**, 555–562. <https://doi.org/10.11118/actaun201765020555>
- Manaf A, Fayyaz UH. 2006. Effects of sulphur on fatty acid accumulation in brassica cultivars. *Int. J. Agric. Biol.* **8**, 588–592.
- MNFS&R. 2013–14. *Ministry of National Food Security and Research, Agric. Statistics of Pakistan*. Govt. of Pak, Economic Wing, Islamabad, pp. 1–166.
- Muhammad S, Khalil IA, Khan S. 1991. Fatty acid composition of rape and mustard oilseed cultivars. *Sci. Khyber.* **4**, 29–36.
- Ray K, Sengupta K, Pal AK, Banerjee H. 2015. Effects of sulphur fertilization on yield, S uptake and quality of Indian mustard under varied irrigation regimes. *Plant Soil Environ.* **61**, 6–10. <https://doi.org/10.17221/860/2014-PSE>
- Rashid A. 1996. *Secondary and micronutrients*. In A. Rashid, Q. S. Memon, E. Bashir and R. Band (Eds.) Soil Science. National Book Foundation; Islamabad, p. 341–372.
- Rękas A, Siger A, Wroniak M, Scibisz I. 2017. Chemical composition and resistance to oxidation of high-oleic rapeseed

- oil pressed from microwave pre-treated intact and de-hulled seeds. 2107. *Grasas Aceites* **68**, e225. <https://doi.org/10.3989/gya.0775171>
- Sawan ZM, Hafez SA, Basyony AE, Alkassas AR. 2007. Nitrogen, potassium and plant growth retardant effect on oil content and quality of cotton seed. *Grasas Aceites* **58**, 243–251.
- Shoja T, Majidian M, Rabiee M. 2018. Effects of zinc, boron and sulphur on grain yield, activity of some antioxidant enzymes and fatty acid composition of rapeseed (*Brassica napus* L.). *Acta Agri. Slov.* **111**, 73–84. <https://doi.org/10.14720/aas.2018.111.1.08>
- Subhani A, Shabbir G, Fazil M, Mahmood A, Khalid R, Cheema NM. 2003. Role of sulphur in enhancing the oil contents and yield of rapeseed under medium rainfed conditions. *Pak. J. Soil Sci.* **22**, 50–53.
- Uncu O, Ozen B, Tokatli F. 2019. Mid-infrared spectroscopic detection of sunflower oil adulteration with safflower oil. *Grasas Aceites* **70**, e290. <https://doi.org/10.3989/gya.0579181>
- Wang QR, Pinkerton A, Hocking P, Wang QR. 1997. Effects of sulphur top-dressing on yield and quality of a double-low rapeseed (*Brassica napus* L.). *Oil Crop. China.* **19**, 53–57.
- Weiss EA. 1983. *Rapeseed*. In *Oilseed Crops*, Longman Group, New York, P.161–215.