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Determination of oil content and fatty acids profile in sunflower seeds through near infra-red spectroscopy under various treatments of potassium nitrate, zinc sulphate and gibberellic acid

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# Abstract

The effects of potassium nitrate (KNO<sub>3</sub>), zinc sulphate (ZnSO<sub>4</sub>) and gibberellic acid (GA<sub>3</sub>) foliar spray on oil content and fatty acid profile in sunflower varieties were evaluated through Near Infra-Red Spectroscopy (NIRS). Eight varieties of sunflower (Rising Sun, SMH-0907, Ausigold-7, SMH-0939, US-444, Hysun-33, SMH-0917 and HS-K6) were grown in glass house. Different concentration of KNO<sub>3</sub> (10 mg/L and 5 mg/L), ZnSO<sub>4</sub> (2 mg/L and 1 mg/L) and 100 ppm of gibberellic acid solution were prepared. Four doses of each treatment were applied to the plants and for each dose fresh solutions were prepared. Potassium nitrate and Zinc sulphate significantly increased the oil % and unsaturated fatty acid level in sunflower varieties. In T1 treatment (10 mg/L of KNO<sub>3</sub>), the highest oil was found in the SMH-0917 (43.35 %), oleic acid (16.50 %) in Rising Sun and linoleic acid in SMH-0939 (77.25 %). In T3 treatment (2 mg/L of ZnSO<sub>4</sub>) the maximum oil % was noted in SMH-0917 (41.25 %), oleic acid in SMH-0917 (15.40 %) and linoleic acid in Hysun-33 (77.25 %). Gibberellic acid foliar application (100 ppm of GA3) significantly enhanced the oil content. The maximum oil % was observed in Hysun-33 (36.00 %), oleic acid in Rising Sun (14.30 %) and linoleic acid (75.60 %) in SMH-0917. Conclusively, oil content and unsaturated fatty acid could be increased by the application of K, Zn and GA3 in sunflower.

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### Introduction

Sunflower is the most important oilseed crop which ranks third after peanut, soybean and contributes significantly to edible oil in the world. It can be successfully cultivated in rain-fed and irrigated across the world (Susan *et al.*, 1993). Sunflower is a short duration crop, having the capability to acclimatize to a wide range of environment and soil conditions (Thavaprakash *et al.*, 2003).

Their seed contains vitamins B1, B5, E, folate and a good source of amino acids (tryptophan), dietary fiber, minerals (manganese, potassium, copper, iron, magnesium, selenium, calcium phosphorus and zinc) and cholesterol lowering phytosterols (Bibi *et al.*, 2003; Arshad *et al.*, 2012). It also consists of high levels of linoleic acid (polyunsaturated fatty acid) which cannot synthesize in the human cells (4). The high ratio of unsaturated to saturated fatty acid in the food will reduce the amount of cholesterol in the blood serum and thus prevents the coronary heart disease (Booyens *et al.*, 1988).

Sunflower production potential can be altered by using nutrients, plant growth hormones and good quality of seeds. For an optimum production, the nutrient should be supplied in a proper amount to the crops (Barker and Pilbeam, 2007). Sunflower needs high amount of nutrients for their normal growth. The physiological function will be badly affected when nutrients under optimum level applied to the plant (Ahmad *et al.*, 2001). Among the nutrients, potassium and zinc (K and Zn) are very essential nutrients and it is found in very low amount in crops (Welch and Graham, 2004).

It plays a key role in the plant growth and biomass (Cakmak *et al.*, 2008). Potassium takes part in many essential processes in plants, i.e. regulation of plant osmotic pressure of the cells and control the closing and opening of the stomata. It also has a role in the fats, protein and formation of ATPs. It can contribute to the action of approximately 60 enzymes (Page *et al.*, 1982). Zinc is an important regulatory cofactor and structural constituent of different enzymes and protein in many biochemical processes like photosynthesis,

conversion of sugar to starch, carbohydrate metabolism, auxin metabolism, protein synthesis and integrity of bio-membranes (Suresh *et al.*, 2013). Plants needed zinc in very small amount, but critically, whenever proper amount of zinc is not present, plants will suffer from physiological stress (Suresh *et al.*, 2013). Zinc is required for different biological process and more than 100 enzyme functions (Baloch *et al.*, 2010).

Among the naturally occurring plant growth hormones, Gibberellic acid (GA3) is considered of specific importance in promoting the plant growth. Exogenous application of GA3 not only increases the vegetative growth, but also, promotes crop yield, 1000 seed weight and seed number/capitulum (Madrap *et al.*, 1992). This study aimed to find out the effect of exogenous application of nutrients (Potassium and Zinc) and plant growth regulator (Gibberellic acid) on the level of oil content and unsaturated fatty acids in sunflower varieties.

### Material and methods

#### Plant materials

This experiment was carried out in the glass house, Department of Biotechnology, University of Malakand during spring 2014. Eight sunflower varieties (Rising Sun, SMH-0907, Ausigold-7, SMH-0939, US-444, Hysun-33, SMH-0917 and HS-K6) were collected from National Agricultural Research Centre (NARC) Islamabad. From the obtained seeds only healthy seeds were selected for the sowing.

## Soil preparation and seed sowing

Before seeds sowing fertile loamy soil (mixture of sand, silt, clay and organic matter with a proportion of (3: 3: 2:2 respectively) were added to the glass house and mixed gently. A small size plots (2 m<sup>2</sup>) were made and well irrigated. After three days of irrigation, seeds were sowed in a line. All seeds were sown with three replicates and line to line space was kept 30 cm. After seeds germination thinning were carried out and plant to plant distance were kept 12 cm. During plant growth all agronomic practices were carried out.

## Electrical conductivity (EC%)

Soil electrical conductivity is an important indicator of soil health. Soil sample (10 gm) was taken and dissolved in the 100 ml distilled water. Through conductivity meter the soil EC % was measured. The unit of EC is deci Siemens per meter.

### Treatments

Different treatments of nutrients (potassium and Zinc) and plant growth regulator (Gibberellic Acid) were exogenously added to the plants. Different concentration of  $KNO_3$  (10 mg/L and 5 mg/L), ZnSO<sub>4</sub> (2 mg/L and 1 mg/L) and 100 ppm of gibberellic acid (GA<sub>3</sub> was taken and dissolved in 5 ml of methanol then finally volume was reached to1000 ml with dH<sub>2</sub>O) solution were prepared. When plant reached four to five leaves stage, foliar spray was applied at the interval of seven days. Three doses of each treatment were applied to the plants and for each dose fresh solutions were prepared.

## Harvesting

When plant reached to their physiological maturity, the capitulum of each plant was cut from the plants. They were kept for the sun dried for two days, to remove the excess water. Then seeds from each heads were hand threshed properly and seeds were preserved.

### **Biochemical analysis**

All the seeds of treated and control plants were analysed for oil % and fatty acid composition through Near Infrared Spectroscopy (NIRS) at the Nuclear Institute for Food and Agriculture (NIFA) Peshawar. The NIRS analysis was carried out using a Near– Infrared scanning monochromator (NIR System model 6500, Foss NIR Systems Inc., MD, USA). Seeds of sunflower (5 -7 seeds) were taken, placed in a standard ring cup and then scan.

The scanning process of each sample could be finished in 1.5 minutes. Reflectance data were stored as  $\log (1/R)$  at 2 nm Intervals. Samples were scanned once (no repeated spectral measurements were made) and were not rotated when spectra collection was made.

Two pairs of lead sulfide detectors collected the reflectance spectra. Reflectance energy readings were referenced to corresponding readings from an internal ceramic disk provided by the instrument manufacturer.

The spectrum of each sample was the average of 32 successive scans. Computer operation and spectral data collection were manipulated using ISI version 3.1 software (InfraSoft International, Port Matilda, PA). The NIRS calibration equation was developed for oil content, palmatic acid ( $C_{16:0}$ ), stearic acid ( $C_{18:0}$ ), oleic acid ( $C_{18:1}$ ), linoleic acid ( $C_{18:2}$ ) and protein. Fatty acids were expressed as the percentage of total fatty acids. For the calibration only the spectra data from 1100 to 2500 nm were used.

## Statistical analysis

The obtained data were analysed through MS excel and GraphPad Prism 5. The mean value of replicate were calculated through MS excel 2007 and significant differences among varieties and treatment were analysed through GraphPad Prism 5.

## Results

# Effect of different treatments (potassium, zinc and gibberellic acid) on oil (%)

Different treatments of potassium, zinc and gibberellic acid significantly ( $p \le 0.05$ ) increased the oil % in selected sunflower varieties as compared to Control C (Table 1). In control C (without treatments of K, Zn and GA3), the maximum oil % was found in Ausigold-7 (35.15 %) while the minimum oil % was noted in SMH-0917 (30.45 %).

The potassium, zinc and GA3 treatments increased the oil content in SMH-0917 up to (43.35 %, 41.25 % and 35.09 %) respectively. In T1 treatment (10 mg/L of K), the maximum oil % was observed in SMH-0917 (43.35 %) while the lowest oil % was revealed by US-444 (37.20 %) Table (1). A significant variation ( $p \le$ 0.05) was also found among varieties under the treatments of potassium (Table 1). **Table 1.** Effect of different treatments of potassium nitrate (KNO<sub>3</sub>), zinc sulphate (ZnSO<sub>4</sub>) and gibberellic acid (GA<sub>3</sub>) on production of Oil % in sunflower varieties. Control is compared with treatments in each column.  $\pm$  denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for oil % in selected sunflower varieties.

Treatments	Rising Sun		SMH-0907		Ausigold	-7	SMH-0	939		US-444		Hysun-33		SMH-0917		HS-K6	
	Oil	Column	Oil	Column	Oil	Column	Oil	(	Column	Oil	Column	Oil	Column	Oil	Column	Oil	Column
	(%)	"A"	(%)	"A"	(%)	"A"	(%)	,	"A"	(%)	"A"	(%)	"A"	(%)	"A"	(%)	"A"
Control C	32.40 ±	d	33.35 ±	с	$35.15 \pm$	a	32.60	± (	d	32.25	± d	34.70 ± 2.97 <sup>e</sup>	b	$30.45 \pm 0.07$ <sup>e</sup>	e	$33.30 \pm 3.25$ <sup>e</sup>	с
(without treatments)	2.55 <sup>e</sup>		1.48 f		1.48 <sup>e</sup>		$5.32^{\text{ f}}$			2.33 <sup>e</sup>							
T1 Treatment	38.15 ±	d	39.10	± c	40.65 ±	b	40.50	±Ι	b	37.20	± e	$40.35 \pm 4.45^{a}$	b	43.35 ± 5.73 <sup>a</sup>	a	40.30 ± 8.91 <sup>a</sup>	b
(10 mg/L of KNO3)	3.04 <sup>a</sup>		2.26 a		2.26 a		0.71 <sup>a</sup>			4.95 <sup>a</sup>							
T2 Treatment	37.80 ±	d	37.10	± d	$39.25 \pm$	b	38.10	± (	с	36.65	± e	$38.30 \pm 2.40$ b	с	$41.25 \pm 3.61$ b	a	$38.30 \pm 2.55$ b	с
(5 mg/L of KNO3)	5.66 <sup>b</sup>		0.14 <sup>b</sup>		0.14 <sup>b</sup>		4.24 <sup>b</sup>			2.33 <sup>c</sup>							
T3 Treatment	36.15 ±	d	36.60	± d	$38.85 \pm$	b	36.85	± (	d	37.35	± c	$38.50 \pm 2.40$ b	b	$41.25 \pm 3.61$ b	a	$38.30 \pm 1.84$ b	b
(2 mg/L of ZnSO4)	0.07 <sup>c</sup>		0.99 <sup>c</sup>		0.99 <sup>c</sup>		0.64 <sup>c</sup>			$5.87 {}^{\mathrm{b}}$							
T4 Treatment	34.80 ±	e	35.10	± d	$38.10 \pm$	b	34.60	± (	е	37.15	± c	$37.70 \pm 1.56$ °	с	40.40 ± 4.67 °	a	$37.40 \pm 7.21^{\circ}$	с
(1 mg/L of ZnSO4)	5.66 <sup>d</sup>		0.14 <sup>d</sup>		0.14 <sup>c</sup>		5.23 <sup>d</sup>			0.92 <sup>b</sup>							
T5 Treatment	34.22 ±	с	34.84 ±	с	36.84 ±	a	33.65 ±	E (	d	$34.12 \pm$	с	36.00±	a	35.09 ±	b	$35.11 \pm$	b
(100 ppm of GA3)	2.11 <sup>d</sup>		1.99 <sup>e</sup>		1.99 <sup>d</sup>		2.44 <sup>e</sup>			1.12 <sup>d</sup>		2.87 <sup>d</sup>		3.44 <sup>d</sup>		1.08 <sup>d</sup>	

**Table 2.** Effect of different treatments of  $KNO_3$ ,  $ZnSO_4$  and  $GA_3$  on production of protein % in selected sunflower varieties. Control is compared with treatments in each column.  $\pm$  denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for protein % in selected sunflower varieties.

Treatments	Rising Sun	ing Sun		SMH-0907 A		Ausigold-7		39	US-444		Hysun-33		SMH-0917		HS-K6	
	Protein (%)	-	Protein		Protein (%)	Column	Protein	Column	Protein (%)		Protein (%)	Column	Protein (%)		Protein (%)	Column
		"A"	(%)	"A"		"A"	(%)	"A"		"A"		"A"		"A"		"A"
Control C	19.65 ± 1.48 <sup>d</sup>	d	$20.35 \pm$	b	$22.00 \pm$	а	19.45 ±	с		± b	$20.50 \pm 2.12$ d	b	$18.30\pm0.28^{e}$	e	19.80 ±	с
(without treatments)			0.49 <sup>d</sup>		0.21 <sup>f</sup>		3.32 <sup>f</sup>		1.06 <sup>d</sup>						1.48 <sup>e</sup>	
T1 Treatment	$22.55 \pm 1.63$ <sup>a</sup>	e	$23.65 \pm$	d	$32.05 \pm$	a	25.40 ±	b	23.45 ± 1.6	3 d	$24.30 \pm 2.55^{a}$	с	25.75± 2.47 <sup>a</sup>	b	24.90 ±	с
(10 mg/L of KNO3)			1.63 <sup>a</sup>		0.07 <sup>a</sup>		0.57 <sup>a</sup>		a						5.09 <sup>a</sup>	
T2 Treatment	$20.40 \pm 2.97$ <sup>c</sup>	f	$21.05 \pm$	e	28.45 ±	А	24.90 ±	b	22.90	± d	22.30 ± 1.84 b	b	24.30 ± 2.55 b	b	23.65 ±	с
(5 mg/L of KNO3)			1.20 °		1.77 <sup>b</sup>		2.26 <sup>b</sup>		0.14 <sup>b</sup>						1.48 <sup>b</sup>	
T3 Treatment	$21.80 \pm 0.28$ b	d	$22.25 \pm$	с	24.00 ±	b	22.10 ±	с	22.45 ± 3.4	6 с	$22.40 \pm 1.13$ <sup>b</sup>	с	25.30 ± 2.55 <sup>a</sup>	a	24.90 ±	b
(2 mg/L of ZnSO4)			1.06 <sup>b</sup>		0.00 <sup>c</sup>		0.71 <sup>c</sup>		b						5.09 <sup>a</sup>	
T4 Treatment	20.40 ± 2.97 °	d	$21.35 \pm$	с	22.60 ±	b	21.45 ±	с	21.65	± c	$21.05 \pm 0.35$ <sup>c</sup>	с	23.35 ± 0.64 °	a	23.60 ±	а
(1 mg/L of ZnSO4)			0.49°		0.85 <sup>e</sup>		3.32 <sup>d</sup>		0.07 <sup>c</sup>						4.38 °	
T5 Treatment	20.34 ±	e	21.98 ±	с	23.76 ±	a	$20.88 \pm$	d	20.88 ±	d	$21.00 \pm$	d	22.98 ±	b	$22.00 \pm$	с
(100 ppm of GA3)	1.87 °		1.09 °		1.08 <sup>d</sup>		1.23 <sup>e</sup>		0.09 <sup>d</sup>		0.89 °		1.09 <sup>d</sup>		2.09 <sup>d</sup>	

As the concentration of potassium decreases, the level of oil is also decreased. In T3 treatment (2 mg/L of Zn), the highest oil % was revealed by SMH-0917 (41.25 %) while the lowest oil % was observed in Rising Sun (36.15 %). Similarly the T4 treatment (1 mg/L of Zn) also increased the oil percent as compared to control C. The proper amount of zinc increases the oil level while decreasing the level of zinc below the optimum the percent of oil is also decreased. Zinc treatments showed significant ( $p \le 0.05$ ) differences among selected sunflower varieties for oil % (Table 1).

The plant growth regulator (GA3) also increases the oil % in the selected sunflower varieties. In T5 treatment (100 ppm of GA3) the highest oil % was revealed by Ausigold-7 (36.84 %) while the lowest oil % was observed in the SMH-0939 (33.65 %).

**Table 3.** Effect of different treatments of potassium nitrate (KNO<sub>3</sub>), zinc sulphate (ZnSO<sub>4</sub>) and gibberellic acid (GA<sub>3</sub>) on production of Palmatic acid in selected sunflower varieties. Control is compared with treatments in each column.  $\pm$  denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for Palmatic acid % in selected sunflower varieties.

Treatments	Rising Sun		SMH-090	07	Ausigold-	7	SMH-093	9	US-444		Hysun-33	3	SMH-0917		HS-K6	
	Palmatic acid (%)	Column "A"	Palmatic acid (%)	Column "A"	Palmatic acid (%)	Column "A"	Palmatic acid (%)	Column "A"	Palmatic acid (%)	Column "A"						
Control C (without treatments)	4.80 ± 1.98 ª	b	4.65 ± 0.21 <sup>a</sup>	с	5.60 ± 0.34 ª	a	4.65 ± 0.21 <sup>a</sup>	с	4.45 ± 1.06 <sup>a</sup>	c	3.85 0.21 <sup>a</sup>	± d	4.35 ± 1.48 ª	с	4.30 ± 0.05 <sup>a</sup>	с
T1 Treatment (10 mg/L of KNO3)	$2.55 \pm 0.78$ d	с	3.25 0.49 <sup>c</sup>	± a	3.35 ± 0.14 °	a	3.25 = 0.49 <sup>c</sup>	± a	3.00 ± 0.49 °	b b	2.40 0.57 <sup>b</sup>	± c	$2.50 \pm 0.57$ °	с	$2.15 \pm 25.24$ <sup>c</sup>	d
T2 Treatment (5 mg/L of KNO3)	3.15 ± 0.07 ° f	с	3.50 0.57 <sup>c</sup>	ΕC	4.10 ± 0.14 <sup>b</sup>	a	3.50 = 0.57 °	± b	3.30 ± 0.42°	: C	3.05 0.21 <sup>a</sup>	± c	3.70 ± 0.14 <sup>b</sup>	b	3.50 ± 0.28 <sup>b</sup>	с
T3 Treatment (2 mg/L of ZnSO4)	2.65 ± 0.78 <sup>d</sup>	d	3.55 0.07 °	E D	3.80 ± 0.28 °	a	3.55 = 0.07 <sup>c</sup>	⊧ b	3.35 ± 0.07 °	b b	3.15 1.20 <sup>a</sup>	± b	2.70 ± 0.14 °	d	3.00 ± 0.71 <sup>b</sup>	с
T4 Treatment (1 mg/L of ZnSO4)	3.90 ± 0.06 ° f	b	4.00 : 0.21 <sup>b</sup>	⊧ b	3.20 ± 0.57 °	c	4.00 = 0.21 <sup>b</sup>	E a	3.85 ± 0.35 °	: b	3.25 0.07 <sup>a</sup>	± c	3.20 ± 0.57 b	с	2.75 ± 0.07 °	d
T5 Treatment (100 ppm of GA3)	4.00 ± f 0.09 <sup>b</sup>	с	4.60 ± 1.00 <sup>a</sup>	b	5.00 ± 0.92 <sup>a</sup>	a	4.60 ± 1.00 <sup>a</sup>	b	4.10 ± 1.23 <sup>b</sup>	с	3.60 ± 0.24 ª	d	4.00 ± 0.56 <sup>a</sup>	с	3.98 ± 0.78 <sup>b</sup>	с

Effect of Potassium, zinc and gibberellic acid on protein (%)

Different treatments (K, Zn and GA3) showed significant ( $p \le 0.05$ ) effect on the protein % in sunflower varieties as compared to control C (Table 2). In control plants (without treatment) the

maximum protein was found in Ausigold-7 (22.00 %) while the lowest was obtained in SMH-0917 (18.30 %). In T1 treatment (10 mg/L of K), the highest amount of protein was found in Ausigold-7 (32.05 %) while the lowest protein % was observed in Rising Sun (22.55 %).

**Table 4.** Effect of different treatments (KNO<sub>3</sub>, ZnSO<sub>4</sub> and GA<sub>3</sub>) on production of Stearic acid in sunflower varieties. Control is compared with treatments in each column.  $\pm$  denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for Stearic acid % in selected sunflower varieties.

Treatments F	Rising Sun		SMH-0907		Ausigold-7		SMH-0939		US-444		Hysun-3	3	SMH-0917		HS-K6	
S	Stearic	Column	Stearic	Column	Stearic	Column	Stearic	Column	Stearic	Column	Stearic	Colu	Stearic	Column	Stearic	Column
А	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	mn	Acid (%)	"A"	Acid (%)	"A"
												"A"				
Control C 5	$5.15 \pm 0.92^{a}$	b	$4.40 \pm 0.14$ <sup>a</sup>	d	$4.70 \pm 0.45^{a}$	с	4.40 ± 1.56 <sup>a</sup>	d	$5.20 \pm 0.85$ <sup>a</sup>	b	5.05	± b	$3.85 \pm 0.49$ <sup>a</sup>	e	$5.60 \pm 0.28$ <sup>a</sup>	a
(without											0.07 <sup>a</sup>					
treatments)																
	3.60 ± 0.00 °	b	$2.80\pm0.00\ ^{c}$	d	$2.75\pm0.07^{c}$	d	3.25 ± 2.05 b	с	$3.60 \pm 0.14$ <sup>c</sup>	b	3.85	±a	$3.15 \pm 0.49$ <sup>a</sup>	с	$3.35 \pm 0.07$ <sup>c</sup>	с
(10 mg/L of											0.78 <sup>c</sup>					
KNO3)																
T2 Treatment 4	4.85 ± 0.78 <sup>b</sup>	а	$3.70 \pm 0.42$ b	с	$3.35 \pm 0.21$ b	с	$4.10 \pm 0.14^{a}$	b	4.05 ± 0.64 b	b	4.10	± b	$3.50 \pm 0.57$ <sup>a</sup>	с	$4.20 \pm 0.42^{b}$	b
(5 mg/L of											1.27 b					
KNO3)																
T3 Treatment 3	3.60 ± 0.00 °	b	$2.80 \pm 0.20$ c	d	$2.75 \pm 0.07$ c	d	$2.90 \pm 4.04$ <sup>c</sup>	d	3.60 ± 0.42 °	b	3.05	± c	$3.60 \pm 1.13^{a}$	b	$4.20 \pm 0.42$ b	а
(2 mg/L of											0.21 <sup>c</sup>					
ZnSO4)																
T4 Treatment 4	1.45 ± 0.35 <sup>b</sup>	b	$4.10 \pm 0.14^{a}$	с	$3.95 \pm 0.21$ b	с	4.40 ± 1.56 a	b	$3.70 \pm 0.14$ <sup>c</sup>	d	4.05	± c	3.85 ± 0.49 a	d	4.75 ± 1.77 b	а
(1 mg/L of											1.91 <sup>b</sup>					
ZnSO4)																
T5 Treatment 5	5.00 ±	а	3.90 ±	с	4.00 ±	b	4.10 ±	b	4.32 ±	b	3.99 ±	с	3.70 ±	с	5.12 ±	a
(100 ppm of 1	.00 a		0.09 <sup>b</sup>		1.21 <sup>b</sup>		1.23 <sup>a</sup>		2.10 <sup>b</sup>		1.00 <sup>c</sup>		1.56 <sup>a</sup>		2.00 <sup>a</sup>	
GA3)																

The potassium treatments showed significant differences among selected varieties of sunflower (Table 2). In T3 treatment (2 mg/L of Zn), the highest protein was

revealed by SMH-0917 (25.30 %) while lowest protein was observed in Rising Sun (21.80 %). Among the varieties significant ( $p \le 0.05$ ) differences in protein % were found under the treatments of zinc.

GA3 also enhanced the level of protein % in sunflower varieties. The maximum protein % was revealed by Ausigold-7 (23.76 %) while the minimum protein was observed in Rising Sun (20.34 %). Variation in protein % among sunflower varieties was found under the treatment of GA3 (Table 2).

# Effect of treatments of K, Zn and GA3 on palmatic acid (%)

The exogenous application of potassium (K) zinc (Zn) and Plant growth regulator (GA3) significantly ( $p \le 0.05$ ) decreased the level of palmatic acid as compared to control C (Table 3). In control C (without treatments of K, Zn and GA3), the maximum palmatic acid % was found in the Ausigold-7 (5.60 %) while minimum palmatic acid was noted in the Hysun-33 (3.85 %).

**Table 5.** Effect of different treatments of potassium nitrate, zinc sulphate and gibberellic acid on production of Oleic acid in selected sunflower varieties. Control is compared with treatments in each column. ± denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for Oleic acid % in selected sunflower varieties.

Treatment	Rising Sun		SMH-0907		Ausigold-7		SMH-0939		US-444		Hysun-33		SMH-0917		HS-K6	
	Oleic	Column	Oleic	Column	Oleic	Column	Oleic	Colum	Oleic	Colum	Oleic	Column	Oleic	Column	Oleic	Colum
	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	n	Acid (%)	n	Acid (%)	"A"	Acid (%)	"A"	Acid (%)	"A"
								"A"		"A"						
Control C (without	14.10 ± 1.84 °	a	13.30± 0.42 <sup>b</sup>	b	11.95± 1.77 °	c	8.70 ± 0.34 <sup>d</sup>	d	13.60± 1.27 °	b	$12.95 \pm 0.35$ <sup>d</sup>	c	11.10 ± 1.70 <sup>e</sup>	c	13.15 ± 0.35 °	b
treatment s)																
T1	$16.50 \pm 0.35$ <sup>a</sup>	a	14.70± 0.71 <sup>a</sup>	c	12.95± 0.07 <sup>b</sup>	d	$14.70 \pm 0.28$ a	с	$15.10 \pm 0.99$ <sup>a</sup>	b	$15.30 \pm 0.14$ <sup>a</sup>	b	14.60 ± 0.99 <sup>b</sup>	с	$14.85\pm0.07^{\rm \ b}$	с
Treatment																
(10 mg/L																
of KNO3)																
T2	$15.50 \pm 0.35$ b	а	$14.00 \pm 0.41^{a}$	b	$12.70 \pm 0.14$ b	d	$12.85 \pm 1.63$ °	d	$14.50 \pm 0.71^{b}$	b	$13.65 \pm 0.35$ °	с	$13.20 \pm 0.42$ <sup>c</sup>	с	$13.95 \pm 1.34$ °	с
Treatment																
(5 mg/L of																
KNO3)																
Т3	$15.00 \pm 0.28$ b	а	$14.10 \pm 1.13^{a}$	b	14.70± 0.28 a	b	$14.80 \pm 0.42$ a	b	$15.20\pm 0.99$ a	а	$14.45 \pm 2.05$ b	b	$15.40 \pm 0.85$ a	а	$15.10 \pm 0.14$ a	а
Treatment																
(2 mg/L of																
ZnSO4)																
T4	$14.35 \pm 0.21 ^{\circ}$	а	13.90± 0.42 b	b	$14.30\pm 0.14^{a}$	а	14.00 ± 0.28	а	$14.50\pm 0.71^{b}$	а	$13.65 \pm 0.35$ °	с	$14.00 \pm 1.84$ b	а	$14.85 \pm 1.06$ b	a
Treatment							a									
(1 mg/L of																
ZnSO4)																
Т5	14.30 ±	a	13.40 ±	b	$12.50 \pm$	c	13.00 ±	b	14.00 ±	a	13.00 ±	b	12.09±	с	14.00 ±	a
Treatment	0.12 <sup>c</sup>		1.20 <sup>b</sup>		1.11 <sup>b</sup>		1.43 <sup>b</sup>		0.34 <sup>b</sup>		1.01 <sup>c</sup>		2.00 <sup>d</sup>		1.00 <sup>b</sup>	
(100 ppm																
of GA3)																

Potassium and zinc treatments significantly decreased the level of palmatic acid % in Ausigold-7 up to (3.35 and 3.20 % respectively) Table (3).

In T1 treatment (10 mg/L of K) the highest palmatic acid was exhibited by Ausigold-7 (3.35 %) while lowest palmatic acid was observed in HS-K6 (2.15 %). In T3 treatment (2mg/L of Zn) the highest Palmatic acid was observed in Ausigold-7 (3.80 %) while lowest Palmatic acids was showed by Rising Sun (2.65 %). The foliar application of GA3 also slightly decreases the amount of palmatic acid as compared to control plant. All treatment showed highly significant differences among varieties for palmatic acid in selected varieties (Table 3).

# Effect of different treatment of potassium, zinc and gibberellic acid on stearic acid %

The exogenous application of different treatments (K, Zn and GA3) showed a significant ( $p \le 0.05$ ) effect on Stearic acid % as compared to control (Table 4).

In control C (without treatments of potassium, zinc and gibberellic acid), the highest Stearic acid % was found in HS-K6 (5.60 %) while the lowest was noted in SMH-0917 (3.85 %). The potassium and zinc treatment significantly ( $p \le 0.05$ ) reduce the level of stearic acid in selected sunflower varieties. In T1 treatment (10 mg/L of K) the maximum stearic acid was found in Hysun-33 (3.85 %) while lowest stearic acid was observed in Ausigold-7 (2.75 %) The potassium treatments showed highly significant differences for stearic acid in selected varieties (Table 4). In T3 treatment (2 mg/L of Zn) the maximum stearic acid was revealed by HS-K6 (4.20 %) while lowest stearic acid was noted in Ausigold-7 (2.75 %). The application of zinc reduces the level of stearic acid in selected varieties. It showed highly significant variation among varieties for stearic acid.

**Table 6.** Effect of different treatments of potassium nitrate, zinc sulphate and gibberellic acid on production of linoleic acid in selected sunflower varieties. Control is compared with treatments in each column.  $\pm$  denote standard deviation and different letter represent the significant difference. The columns "A" shows comparison of significant differences among varieties for linoleic acid % in selected sunflower varieties.

Treatments	Rising Sun		SMH-0907		Ausigold-7		SMH-093	39	US-444		Hysun	-33	SMH-0917		HS-K6	
	Linoleic Acid (%)		Linoleic Acid (%)		Linoleic Acid								Linoleic Acid (%)		Linoleic Acid (%)	Column
		"A"		"A"	(%)	"A"	Acid (%)	"A"	Acid (%)	"A"	Acid	"A"		"A"		"A"
											(%)					
Control C	73.10± 3.25 °	C	$2.10 \pm 0.14$ <sup>e</sup>	d	$67.85 \pm 0.21$ e	f	70.10 ±	e	70.70 ±	e	4.11	± b	$5.10 \pm 0.45$ <sup>c</sup>	а	$2.50 \pm 0.71$ d	b
(without							2.97 <sup>f</sup>		2.83 d		1.56 <sup>c</sup>					
treatments)																
T1 Treatment	75.75± 0.49 <sup>a</sup>	С	6.15 ± 0.46 <sup>a</sup>	b	76.55 ± 1.06 <sup>a</sup>	b	77.25±	a	75.70 ±	c	5.65	± c	7.15 ± 1.91 <sup>a</sup>	a	5.55 ± 0.78 <sup>a</sup>	с
(10 mg/L of	f						1.06 a		0.60 <sup>a</sup>		1.77 <sup>b</sup>					
KNO3)																
T2 Treatment	74.35± 1.20 <sup>b</sup>	С	5.70 ± 0.28 <sup>b</sup>		74.35 ± 0.49 °	с	76.30 ±	a	74.65 ±	c	5.35	± b	$6.40 \pm 0.28$ b	а	$4.30 \pm 0.42^{b}$	с
(5 mg/L of	f			b			2.40 <sup>b</sup>		1.91 <sup>b</sup>		2.05 b					
KNO3)																
T3 Treatment	5.45 ± 0.64 <sup>a</sup>	С	$4.10 \pm 0.14$ <sup>c</sup>	d	75.90 ± 0.42	с	73.45 ±	e	75.30 ±	c	7.25	± a	$6.85 \pm 2.33$ b	b	4.65 ± 2.90 <sup>b</sup>	d
(2 mg/L of	f				b		2.33 °		6.08 a		0.64 <sup>a</sup>					
ZnSO4)																
T4 Treatment	$5.10 \pm 2.97$ <sup>a</sup>	В	3.00 ±	d	75.00 ± 0.42	b	72.60 ±	e	74.15 ±	с	5.45	± b	6.00 ± 2.19 <sup>b</sup>	a	$4.10 \pm 0.87$ b	с
(1 mg/L of	f		1.98 <sup>d</sup>		b		2.69 <sup>d</sup>		0.92 <sup>b</sup>		4.03 <sup>b</sup>					
ZnSO4)																
T <sub>5</sub> Treatment	4.00 ±	В	3.32 ±	с	71.43 ±	e	71.00 $\pm$	е	72.12 $\pm$	d	$4.21 \pm$	b	5.60 ±	а	3.00 ±	с
(100 ppm of	f 1.11 <sup>b</sup>		0.98 <sup>d</sup>		1.21 <sup>d</sup>		1.21 <sup>e</sup>		0.21 <sup>c</sup>		2.11 <sup>c</sup>		1.98 <sup>c</sup>		1.43 <sup>c</sup>	
GA3)																

The gibberellic acid treatment (T5 treatment) also minimizes the stearic acid as compared to control plants. The maximum stearic acid was observed in the HSK-K6 while minimum stearic acid was noted in SMH-0917 (3.70 %) Table (4).

# Effect of different treatments (K, Zn and GA3) on oleic acid %

The treatments of potassium, zinc and gibberellic acid showed significant ( $p \le 0.05$ ) effect on the production of oleic acid in different selected varieties as compared to control C (Table 5). These treatments increased the level of oleic acid (monounsaturated fatty acid) in selected varieties. In control C (without treatments of K, Zn and GA3), the high oleic acid was observed in Rising Sun (14.10 %) while lowest oleic acid was noted in SMH-0939 (8.70 %). In T1 treatment (10 mg/L of K) the maximum oleic acid was observed in Rising Sun (16.50 %) while minimum oleic acid was revealed by Ausigold-7 (12.95 %). Potassium treatments showed significant differences for oleic acid among selected varieties.

In T3 treatment (2 mg/L of Zn), the high oleic acid was observed in SMH-0917 (15.40 %). In T5 treatment (100 ppm gibberellic acid foliar spry), the maximum oleic acid was noted in the Rising Sun (14.30 %) while minimum oleic acid noted in the SMH-0917 (12.09 %). Among the varieties highly significant differences were found in oleic acid (Table 5).

# Effect of different treatments (potassium, zinc and gibberellic acid) on linoleic acid %

The foliar spray of K, Zn and GA3 showed a significant ( $p \le 0.05$ ) effect on linoleic acid as compared to control C (Table 6). In control plants, maximum linoleic acid was noted in SMH-0917 (75. 10 %) while lowest linoleic acid was observed in Ausigold-7 (67.85 %). In T1 treatment (10 mg/L of K) the maximum linoleic acid was observed in the SMH-0939 (77.25%) while the lowest was found in HS-K6 (75.55 %). Among varieties, treatments showed highly significant differences for linoleic acid (Table 6). The exogenous application of zinc also increased the level of linoleic acid in sunflower varieties. In T<sub>3</sub> treatment (2 mg/L of Zn), maximum linoleic acid was noted in Hysun-33 (77.25 %) while minimum linoleic acid was noted in the SMH-0939 (73.45 %). The GA3 treatment also slightly enhanced the linoleic acid in selected varieties. All treatments showed highly significant ( $p \le 0.05$ ) differences among varieties for linoleic acid production (Table 6).

#### Discussion

#### Oil and protein contents

Sunflower occupies an important position in oilseed crops in the world due to short life span, wide range of adaptability to climate and high yielding potential (Thavaprakash et al., 2003). It gives positive responses to the nutrients and for an optimum production the nutrients should be supplied in a proper amount to the crops. Among the nutrients, potassium plays a key role in plant growth and metabolism. It plays an essential role in photosynthesis, energy transfer, osmoregulation of stomatal movement, enzyme activation and cationanion (Marschner, 2012). It can enhance the transportation of amino acid to the seeds and thus increased the metabolism of protein. Potassium showed significant effect on protein and oil content in sunflower seeds. Similar results have been reported by (Bellaloui et al., 2013; Abasiyeh et al., 2013). Our results are also in agreement with (Adnan, 2011). Zinc is a micronutrient and plays an important role as a regulatory co-factor or structural constituent of several enzymes and

different proteins in biochemical pathways (photosynthesis, carbohydrate metabolism, protein metabolism, conversion of sugar to starch, pollen formation, auxin metabolism (Ahmad et al., 2011). Zinc is required in a small amount, but the critical concentration is very essential for an optimum growth and development of the crops (Suresh et al., 2013). The exogenous application of Zn increased the oil and protein content in the sunflower seeds. Zinc demonstrated significant effect on protein and oil content. It enhanced the oil contents and protein in oilseed crops. The similar results also reported by (Sawan et al., 2006; Shaker et al., 2012). Among the naturally occurring growth hormones, gibberellic acid is considered of specific importance in promoting the plant growth. Exogenous application of GA3 not only increases the vegetative growth, but also 1000 seed weight, seed number/capitulum and promotes crop yield (Madrap et al., 1992; Bibi et al., 2003). Gibberellic acid treatment also enhanced the oil content in sunflower. Similar increment in oil % by gibberellins application also reported by (Bibi et al., 2003).

### Fatty acid profile

The treatments (K, Zn and GA3) showed a significant effect on palmatic acid and stearic acid in sunflower as compared to control C. The potassium and zinc significantly reduce the level of saturated fatty acids (Palmatic acid and Stearic acid). The saturated fatty acids are decreased as the K and Zn concentrations increased. These fatty acids are found in low level in the sunflower oil, which is very essential for a good health. These treatments reduce the saturated fatty acid and have a beneficial impact on oil composition (Ahmad et al., 2001; Sawan et al., 2006). A low level of saturated fatty acid oil is strongly recommended for edible purposes (Sawan et al., 2006). Oleic acid is a mono-unsaturated fatty acid and high amount of oleic acid is present in the sunflower seeds. The potassium treatments showed significantly increased the level of oleic acid in sunflower seeds. Our results are also in line with findings of (Salama et al., 1987; Mekki et al., 1999; Bellaloui et al., 2013) while it is contradictory to (Ahmad et al., 1999).

The variation of results may be due to ecological circumstances (heat, drought, soil conditions and genotypes). Zinc treatment revealed a significant effect of unsaturated fatty acids (Oleic acid) in oilseed crops. Zn application decreased saturated to unsaturated fatty acids in oil crops. A similar beneficial impact of improved Zn nutrition on oil composition was reported by (Sawan *et al.*, 2006). The linoleic is the most abundant fatty acid in the sunflower oil. Potassium is very essential nutrient and vital component of many important compounds of the cells.

It can increase the level of unsaturated fatty acid. It also involved in biochemical pathways in plants where potassium can activate many enzymes (Taiz and Zeiger, 1991). These characteristics of K may reveal changes in the quality of seed oil (Mekki *et al.*, 1999). Potassium can increase the degree of unsaturation in the final oil (Froment *et al.*, 1998).

These results are in agreement with (20) and contradictory to (Adnan, 2011). Froment *et al.*, 1998 also suggested that K treatments enhance linoleic acid in oil content. The exogenous application of zinc can lead to high production and oil yield due to increase in chlorophyll concentration, auxin biosynthesis, increase the level of phosphorus and nitrogen (Moinuddin and Imas, 2008). Zn increased unsaturated fatty acids in oilseed crops (Marschner, 2012).

### Conclusion

It is concluded from the present research works that optimum nutrients (potassium and zinc) and plant growth regulator (gibberellic acid) play a key role in the increment of oil % and unsaturated fatty acid. Sunflower crop is very responsive to these treatments and can be improved the quality of oil through these treatments.

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