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Qualitative Analysis of Spring Planted Sunflower Hybrids as Influenced by Varying Nutritional Area

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Abstract: A field experiment was conducted at the Agronomic Research Area of PMAS Arid Agriculture University, Rawalpindi to evaluate the quality parameters of spring planted sunflower hybrids as influenced by varying potassium application doses during two consecutive years i.e. 2008 and 2009. Experiment was quadruplicated using randomized complete block design with split plot arrangement keeping different levels of nutritional area in main plots and sunflower hybrids in subplots. Protein and achene oil contents were determined by Nuclear Magnetic Resonance Technique, where as fatty acid composition was determined by GC-9A Fatty Acid Analyzer. Different levels of nutritional area significantly increased protein content and palmitic acid concentration in achene but reduced oil content when levels of nutritional area vary from 60 x 20 cm^2 to 60 x 60 cm^2 (2 plants/hill). However, the concentration of oleic, linoleic and linolenic remained un-affected by varying levels of nutritional area. Hybrid Hysun-33 produced significantly higher protein content (18.89%) in achene as compared to S-278. Conversely, hybrid S-278 accumulated significantly higher oil content (43.48 %) as compared to hybrid Hysun-33. No proper pattern was noticed regarding stearic, oleic, linoleic and linolenic acid accumulation in achenes. It is concluded that sunflower hybrids exhibited differential genotypic response to different levels of nutritional area by increasing oil contents, palmitic acid concentration and reducing protein contents in achenes without affecting stearic, oleic, linoleic and linolenic acid concentration.

Key words: Sunflower hybrids, nutritional area, protein, oil contents, fatty acid composition

INTRODUCTION

Sunflower (*Helianthus annuus* L.) is an important oilseed crop in our country. It has shown its potential to contribute its share in domestic edible oil requirements. Its has lion's share (34.1 g) in per capita vegetable oil consumption of daily oil intake (83 g) in our country (Anonymous, 2008-09).

Pakistan is bestowed with various ecologies, where sunflower can be cultivated because of its wide range of adaptability. Here sunflower can be grown twice in a year during spring and winter (Ahmad, 1993). Sunflower grown in spring is usually slower in growth than during autumn (Kaleem *et al*., 2010), but the response for the quality parameters remains significant. Protein, oil contents and fatty acid profile composition (Flagella *et al*., 2002) in sunflower are significantly influenced by season mainly by the temperature, growth durations which are particular characteristics of seasonal changes (Killi, 2004). Longer reproductive phase and warmer temperature at the time of seed development (spring sown) of sunflower is favorable for high oil contents (Kaleem *et al*., 2010) than during season with high temperature and low relative humidity at the time of pollination (autumn season) that affects pollen vigor, causing poor pollination, produces less weight and infertile achene ultimately leading to head infertility and low achene yield (Jose *et al*., 2004; Omidi *et al*., 2010).

Sunflower hybrids available in Pakistani markets exhibited diversity in their response to nutritional area (Ahmad, 1993). These hybrids, because of their difference in their root system and penetration capacity, vary in their response to protein, oil contents in achene (Omidi *et al*., 2010) as well as palmitic, oleic, linoleic, linolenic acid and stearic acid concentration in achene (Flagella *et al*., 2002; Hassan *et al*., 2003) and unsaturated/saturated ratio of fatty acids; have distinguished them in the world of sunflower hybrids (Kaleem *et al*., 2010). Usually, 2:1 ratio of unsaturated fatty acids to saturated fatty acids is the best for normal human utilization (Bukhsh, 2010). Sunflower cooking oil is extensively used by heart patients because of very low cholesterol concentration and high fatty acid concentration (Flagella *et al*., 2002; Omidi *et al*., 2010). So, there was an urge to screen out sunflower hybrids keeping in view their ability to manufacture achene oil and fatty acid production (Kaleem *et al*., 2009; Gustavo and Luis, 2007; Roche *et al*., 2010).

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Although this is assumed that equal amount of fertilizer applied was absorbed uniformly to every plant, but the availability of the nutritional area to every plant determines the extent of availability of the nutrients to plants (Vallalobos *et al*., 1994; Jose *et al*., 2004) and vary their availability (Zarea *et al*., 2005). In its consequence, variation of nutritional area varies availability of nutrients; even they are equally and uniformly applied. Generally, more nutritional area gives more nutrition to plants, but the extent where maximum uptake of nutrients occurs again gives the question to be quenched (Narwal and Malik, 1985; Allam *et al*., 2003). Variation in availability of nutrients to plants definitely influences the protein, oil and fatty acid profile composition in achene (Killi, 2004; Omidi *et al*., 2010; Hassan *et al*., 2003). It is well documented that there is antagonistic relationship between protein and oil contents in achene, but still there is ambiguity about the availability of nutritional area and palmitic, oleic, stearic, linoleic and linolenic acid (Jose *et al*., 2004; Zarea *et al*., 2005; Flagella *et al*., 2002).

Very lithe literature was available in this regard, so a study was conducted to find out the optimum nutritional area in terms of protein oil contents in achene as well as fatty acid profile for different sunflower hybrids under the agro ecological conditions of Rawalpindi district.

MATERIALS AND METHODS

The study was conducted at the experimental area, Department of Agronomy, PMAS Arid Agriculture University, Rawalpindi during spring 2008 and 2009. Before sowing the crop, soil samples were collected to a depth of 30 cm from the experimental area and analyzed for physico-chemical properties (Table 1).

The experiment was laid out an irrigated sandy clay loam soil with four replications. The experiment was laid out in split plot design keeping different nutritional levels i.e. 60 x 20, 60 x 30, 60 x 40, 60 x 50, 60 x 60 (with 1 plant/hill) and 60 x 60 cm^{-2} (with 2 plants/hill) in main plots and sunflower hybrids in subplots. Net plot size was 3.60 m x 7.20 m. Crop was sown manually using a dibbler on a well prepared bed. Both hybrids Hysun-33 and S-278 were sown on February 4 and 7 during 2008 and 2009, respectively, using recommended seed rate of 7.5 kg/ha. A basal dose of fertilizer @ 100 kg N+100 kg P_2O_5 ha^{-1} and K in the form of urea DAP and potassium sulfate respectively, was applied. All the P, K and half dose of the N were applied at the time of sowing, while rest of the urea was applied with first irrigation (4 weeks after sowing). Subsequent irrigations were given when needed. In addition to soaking irrigation for seed bed preparation 4 and 6 irrigations of 7.5 hectare centimeters each were given to the crop during spring 2008 and 2009. Thinning was done at 4-5 leaf stage to maintain an intra row plant to plant distance of 20-60 cm. Row to row distance was kept constant i.e 60 cm. Crop was kept free of weeds by providing interculture and hand weeding as required to avoid competition between weeds and sunflower crop. Crop was harvested manually on June 6 and 10 during 2008 and 2009, respectively. Achene yield was recorded at 15% moisture content.

Table 1: Pre-sowing physico-chemical analysis of soil

		Value	
Determination	Unit	2008	2009
Physical analysis			
Sand	%	67.00	64.00
Silt	%	15.00	15.00
Clay	%	18.00	21.00
Textural class		Sandy clay loam	
Chemical analysis			
Saturation	%	35.00	34.00
pH		7.80	7.40
EC_e	dS m^{-1}	2.20	2.18
Organic matter	%	0.80	0.83
Total nitrogen	%	0.041	0.040
Available phosphorus	ppm	7.10	7.00
Available K	ppm	169.00	165.00

Achene samples were taken randomly from each experimental unit which were analyzed for oil and protein contents, extracted by NMR (Nuclear Magnetic Resonance system), Model MQA-7005, Oxford Institute, USA, by standardizing the equipment with six different oil contents having the samples previously analyzed, thus oil content in each treatment were recorded (Wamsely, 1998). The fatty acids in oil were analyzed by a gas chromatograph (AIML-NUCON) after intersterilification with methanolic KOH. In this method, fatty acids were converted to methyl esters prior to analysis by Gas Chromatography (GC). Oil samples (50 µL) were methylated in 4 ml 1 M KOH for one hour at room temperature. The resultant Fatty Acid Methyl Esters (FAME) were extracted with High Performance Liquid Chromatography grade hexane and analyzed by GC using a fused capillary column (WCOT fused silica 30m x 0.25 mm coating CPWAX 52 CBDF = 0.25 µM, CP8713, a Flame Ionization Detector (FID) and nitrogen gas as carrier (3.5 ml/min). GC split ratio was 100%. Injector and Detector temperatures were 260°C and column oven temperature was 222°C for 7.5 min. FAMEs were injected manually. Fatty acids were detected by chromatographic retention time by comparison with authentic standards (Paquot, 1988).

Data obtained from were analyzed by Fisher's analysis of variance techniques using least significant difference test at 5% level of probability to compare the differences among treatment means (Steel *et al*., 1997). Weather data for both years were obtained from Meteorological Center, Rawalpindi situated in the University premises (Table 2).

Table 2: Meteorological data taken during 2008 and 2009

	Max. Temp. (°C)	Min. Temp. (°C)	RH (%)	Rainfall (mm)
February, 2008	26.70	10.60	55.00	45.00
March, 2008	29.90	12.92	60.00	81.00
April, 2008	34.00	15.90	44.00	18.00
May, 2008	37.30	19.80	42.00	80.60
June, 2008	37.60	23.00	51.00	22.30
February, 2009	27.80	10.40	50.00	30.00
March, 2009	30.05	12.02	51.57	15.00
April, 2009	29.70	15.77	59.33	92.90
May, 2009	37.16	20.76	40.00	10.10
June, 2009	35.57	22.29	62.43	225.0

Temp. = Temperature; RH = Relative Humidity

RESULTS AND DISCUSSION

Protein content: There was significant variation among the treatments regarding nutritional area for protein content in achene. There was general trend that protein content in achene increased with increase in nutritional area up to level of nutritional area of 60 x 60 cm^2 (with 1 plant/hill) then it tended to decline. Although the maximum protein content (18.80%) was recorded when crop was grown under the nutritional area of 60 x 60 cm^2 (with 1 plant/hill), yet it was statistically at par with many other treatments of nutritional area (Table 3). Minimum protein content (17.45%) was observed when crop was grown under the nutritional area of 60 x 20 cm^2. Similar promotive effect of increase in nutritional area on sunflower protein content was also reported by Ahmad (1993), Ahmad *et al.* (2001), Flagella *et al.* (2002), Gustavo and Luis (2007) and Omidi *et al.* (2010).

Sunflower hybrids had a significant effect on protein content in achene (Table 3). Hybrid Hysun-33 attained significantly higher protein content (18.89.11%) than hybrid S-278 (17.25%) which may be due to varying genetic potential of the hybrids. These results are in confirmed with the findings of Roche *et al.* (2010) and Allam *et al.* (2003) that different sunflower hybrids exhibit the differential response to protein content in achene due to their difference of genetic make up. Interactive effects of nutritional area and sunflower hybrids on protein content in achene were found to be significant. Maximum protein content in achene (19.60%) were recorded when sunflower hybrid Hysun-33 was grown under nutritional area of 60 x 60 cm^2 (with 1 plant/hill) which was statistically at par with many other treatments. Correspondingly, minimum protein content in achene (16.70%) was recorded when sunflower hybrid S-278 was grown under the nutritional area of 60 x 30 cm^2 which was statistically similar with many other treatments. These results are in line with the findings of Ahmad (1993) and Kaleem *et al.* (2010). They reported that sunflower hybrids respond differentially to varying nutritional area in terms of protein contents in achene.

Oil content: There was significant variation among the treatments regarding nutritional area for oil content in achene. Oil content in achene decreased with increase in nutritional area although, the maximum oil content (41.95%) was recorded when crop w as grown under the nutritional area of 60 x 20 cm^2, yet it was statistically at par with many other treatments of nutritional area (Table 3). Minimum oil content (39.83%) was observed when crop was grown under the nutritional area of 60 x 50 cm^2. Similar effect of increase in nutritional area on sunflower oil content was also reported by Ahmad (1993) and Ahmad *et al.* (2001).

Sunflower hybrids had a significant effect on oil content in achene (Table 3). Hybrid S-278 attained significantly higher oil content (43.48%) than in hybrid Hysun-33 (37.70%) which may be due to varying genetic potential of the hybrids. These results are in confirmed with the findings of Allam *et al.* (2003), Gustavo and Luis (2007) and Roche *et al.* (2010) those reported that different sunflower hybrids exhibited the differential response to oil content due to their difference of genetic make up. Interactive effects of nutritional area and sunflower hybrids on oil content were found to be significant. Maximum oil content (44.85%) was recorded when sunflower hybrid S-278 was grown under nutritional area of 60 x 20 cm^2 which was statistically at par with many other treatments. Correspondingly, minimum oil content (36.35%) was recorded when sunflower hybrid Hysun-33 was grown under the nutritional area of 60 x 50 cm^2 which was statistically similar with many other treatments. These results are in line with the findings of Ahmad (1993), Flagella *et al.* (2002) and Kaleem *et al.* (2010). They reported that sunflower hybrids respond differentially to varying environments and nutritional area in terms of oil contents in achenes.

Palmitic acid: Results in Table 3 revealed that variation in nutritional area significantly influenced palmitic acid content of sunflower oil. According to results, palmitic acid concentration in achene increased with increase in nutritional area although, the maximum palmitic acid concentration (6.34%) was recorded when crop was grown under the nutritional area of 60 x 60 cm^2 (with 1 plant/hill), yet it was statistically at par with many other treatments of nutritional area then it tended to decline (Table 3). Minimum palmitic acid concentration (5.78%) was observed when crop was grown under the nutritional area of 60 x 20 cm^2. Similar promotive effect of increase in nutritional area on sunflower palmitic acid concentration of achene was also reported by Ahmad (1993) and Ahmad *et al.* (2001).

Sunflower hybrids had a significant effect on palmitic acid concentration in achene (Table 3). Hybrid Hysun-33 attained significantly higher palmitic acid concentration (6.27%) than in hybrid S-278 (5.87%) which may be due to varying genetic potential of the hybrids (Omidi *et al.*, 2010; Roche *et al.*, 2010). Interactive effects of nutritional area and sunflower hybrids on palmitic acid concentration in achene were found to be significant.

Table 3: Qualitative analysis of spring planted sunflower hybrids as influenced by different levels of nutritional area (Mean of two years)

Treatments	Achene protein content (%)	Achene oil content (%)	Palmitic acid (%)	Stearic acid (%)	Oleic acid (%)	Linoleic acid (%)	Linolenic acid (%)
A- Nutritional Area (N) (cm^2)							
P_1 = 60 x 20	17.45bc	41.95a	5.78c	2.18ab	45.37	44.07	0.23
P_2 = 60 x 30	17.75c	40.98ab	5.95bc	2.26a	44.35	42.85	0.24
P_3 = 60 x 40	18.05bc	40.77ab	5.95bc	2.14ab	44.71	43.79	0.23
P_4 = 60 x 50	18.37ab	39.83b	6.16ab	2.09ab	44.02	42.59	0.22
P_5 = 60 x 60 (1 plant/hill)	18.80ab	39.87b	6.34a	1.94b	44.34	43.74	0.23
P_6 = 60 x 60 (2 plants/hill)	18.02a	40.17b	6.29ab	1.96ab	44.41	42.83	0.23
LSD (a)	0.87*	1.55*	0.376*	0.301*	NS	NS	NS
B- Sunflower Hybrids (H)							
H_1 = Hysun-33	18.89a	37.70b	6.27a	2.09	43.98b	43.72	0.23
H_2 = S-278	17.25b	43.48a	5.87b	2.08	45.58a	42.89	0.23
LSD (b)	0.60*	0.53*	0.254*	NS	1.93*	NS	NS
C- Interaction (H x N)							
P_1 x H_1	18.19abcd	38.25c	5.93bc	2.20a	43.75	44.87	0.23
P_1 x H_2	16.72d	44.85a	5.63c	2.15ab	46.99	43.26	0.23
P_2 x H_1	18.80abc	38.22cd	6.20abc	2.24a	43.70	43.36	0.23
P_2 x H_2	16.70d	43.74ab	5.70c	2.27a	45.00	42.35	0.25
P_3 x H_1	18.65abc	36.80cde	6.13abc	2.21a	43.47	43.90	0.23
P_3 x H_2	17.45cd	43.75ab	5.78c	2.06ab	45.96	43.68	0.23
P_4 x H_1	19.09ab	36.35e	6.43ab	2.15ab	42.08	42.67	0.22
P_4 x H_2	17.65bcd	42.80b	5.90bc	2.02ab	45.97	42.51	0.22
P_5 x H_1	19.60a	36.81e	6.57a	1.99ab	43.47	44.92	0.23
P_5 x H_2	18.00bcd	42.93b	6.10abc	1.93ab	45.21	42.55	0.23
P_6 x H_1	19.01ab	37.50de	6.43ab	1.79b	43.43	42.60	0.22
P_6 x H_2	17.01cd	42.85b	6.15abc	2.10ab	45.39	43.05	0.23
LSD (c)	1.49*	1.31*	0.623*	0.387*	NS	NS	NS

NS = No Significant, * = Significant Means followed by different letters in a column are significantly different at p>0.05

Maximum palmitic acid concentration in achene (6.57%) were recorded when sunflower hybrid Hysun-33 was grown under nutritional area of 60 x 60 cm^2 (with 1 plant/hill) which was statistically at par with many other treatments. Correspondingly, minimum palmitic acid concentration (5.63%) was recorded when sunflower hybrid S-278 was grown under the nutritional area of 60 x 30 cm^2 which was statistically similar with many other treatments. These results are in line with the findings of Ahmad (1993), Gustavo and Luis (2007) and Kaleem *et al.* (2010). They reported that sunflower hybrids respond differentially to varying nutritional area in terms of palmitic acid concentration.

Stearic acid: There was statistically significant variation among different treatments of nutritional area regarding stearic acid concentration. Maximum stearic acid concentration (2.26%) was recorded when crop was grown under nutritional area of 60 x 30 cm^2 which was statistically at par with many other treatments. Likewise, minimum stearic acid concentration (1.94%) was recorded when crop was grown under nutritional area of 60 x 60 cm^2 (with one plant/hill).

Sunflower hybrids were statistically similar with respect to stearic acid concentration under different levels of nutritional area. Similar trends were reported by Vallalobos *et al.* (1994) and Allam *et al.* (2003) Interactive effects of different levels of nutritional area and sunflower hybrids on stearic acid concentration in achene were found to be significant. Sunflower hybrid S-278 grown under nutritional area of 60 x 30 cm^2 recorded maximum stearic acid concentration in achene (2.27%) which was statistically at par with many other treatments. Like wise, sunflower hybrid Hysun-33 grown under nutritional area of 60 x 60 cm^2 (with 1 plant/hill) recorded minimum stearic acid concentration in achene (1.79%) which was statistically similar to many other treatments. These results are in line with the findings of Kaleem *et al.* (2010) and Ahmad *et al.* (2001) that sunflower hybrids are similar in their response to different levels of nutritional area in terms of their quantification of stearic acid concentration in achenes.

Oleic acid: Concentration of oleic acid was not significantly influenced by varying levels nutritional area (Table 3). Variation in unsaturated/saturated fatty acids ratio by different levels of nutritional area was reported by Ahmad *et al.* (2001), Killi (2004) and Jose *et al.* (2004). Sunflower hybrid Hysun-33 was statistically at par with S-278 in terms of comparison of oleic acid concentration in achene. Likewise trends were observed by Flagella *et al.* (2002) and Roche *et al.* (2010). Interactive effects of K levels and sunflower hybrids on oleic acid concentration in sunflower oil contents were also non significant. These results are in agreement with the findings of Weiss (2000) and Kaleem *et al.* (2010) those found that sunflower hybrids did not respond differentially to varying levels of nutritional area in terms of oleic acid concentration in achene.

Linoleic acid: According to results, different levels of nutritional area did not influence linoleic acid concentration of sunflower achene and statistically non significant results were found for linoleic acid (Table 3). Similar trends were recorded by Narwal and Malik (1985) and Zarea *et al.* (2005) for the sunflower hybrids. Interactive effects of different levels of nutritional area and sunflower hybrids on concentration of linoleic acid in sunflower achene were also found to be non significant. These results are in line with the findings of Ahmad *et al.* (2001) and Hassan *et al.* (2003) that sunflower hybrids showed similar responses to varying levels of nutritional area in terms of linoleic acid.

Linolenic acid: According to results, statistically non significant results were found for linolenic acid (Table 3) and different levels of nutritional area did not influence the linolenic acid concentration in sunflower achene. However, it varied from 0.23-0.24%. Sunflower hybrids exhibited similar trends. Interactive effects of different levels of nutritional area and sunflower hybrids on linolenic acid concentration in were found to be non significant. However, it ranged from 0.22-0.25%. Similar findings were recorded by Hassan *et al.* (2003), Narwal and Malik (1985) and Zarea *et al.* (2005) that sunflower hybrids did not respond differentially to varying levels of nutritional area in terms of linolenic acid concentration.

Conclusion: Sunflower hybrids exhibited differential genotypic response to different levels of nutritional area by increasing oil contents, palmitic acid concentration and reducing protein contents in achenes without affecting stearic, oleic, linoleic and linolenic acid concentration in achenes.

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