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CANOLA GROWTH AND PHOSPHORUS AMENDMENTS. I. YIELD AND QUALITY RESPONSE OF CANOLA TO DIFFERENT PHOSPHORUS AMENDMENTS

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A little is known about the brassica species response to different phosphorus (P) amendments and its effects on accumulation of P, proteins, oil and fatty acids in canola (*Brassica napus* L.) seeds. The two years field study was conducted to evaluate the yield and quality of canola hybrids (Hyola-401 and Hyola-43) in response to different P management strategies. The two canola hybrids were tested under different P application rates (@ 30 kg ha^{-1} or 60 kg ha^{-1}) applied through broadcasting and side-drilling, alone and in combination with farmyard manure (FYM). Phosphorus applications have significant positive linear (p>0.05%) effects on yield and yield components in the both canola hybrids. The maximum seed yield (2749±6.96 kg ha^{-1}; 2698±7.67 kg ha^{-1}), oil contents (43.73±0.18%; 42.20±0.20%), protein contents (26.23±0.12%; 26.37±0.12%) and P accumulated in grains (0.52±0.01%; 0.45±0.02%) were recorded in Hyola-43 when fertilized @ 60 kg P ha^{-1} through side drilling along with FYM during 2004-05 and 2005-06, respectively. Hyola-43 appeared a good crop yielding canola hybrid as compared to Hyola-401. The high oleic and low linolenic acid provide good quality oil during frying process. Two folds increase was observed P accumulation in grain and this increase in endogenous grain P may have significant effects on next crop growth on soils having low inherent P. The study result suggests incorporating of FYM along with P fertilizer to overcome the issues of world inorganic P reserves utilization and for the sustainable higher quality canola production.
Keywords: Canola, oil quality, phosphorus, grain, nutrition

INTRODUCTION

High demands for food due to high annual population growth rate (2.05%) and changing dietary habits are increasing pressure on agriculture (Anonymous, 2013). Changing food patterns in Pakistan have resulted in a rapid increase in edible oil consumption and forced the researchers to find good quality edible oil. Canola oil having high oleic acid contents and low erucic acid contents sure the neutral taste and provides excellent stability. The high oleic canola oils are among the major healthful oils replacing *trans* fat in food processing and packaging (Bomgardner, 2012). High contents of monounsaturated fatty acids, especially oleic acid (18:1) are associated with a low incidence of coronary heart disease because it decreases total cholesterol (up to 10%) and low-density lipoprotein cholesterol (Cintra *et al*., 2006).

Domestic production of edible oils meets only 34% of the national need (Anonymous, 2013) while the rest is imported by spending large amount of foreign exchange. This huge drain on our resources can be minimized by increasing productivity of domestic canola oil crop through adopting better nutrient management practices. Out of various agronomic factors responsible for reduced final crop harvest and poor quality in developing countries, phosphorus (P) plant nutrition is most important one (Ahmad *et al*., 2011; Ghosh *et al*., 2006; Grant *et al*., 2001; Nadeem *et al*., 2011). Phosphorus is an important macronutrient comprising approximately 0.2% of a plant's dry weight (Schachtman *et al*., 1998) and is a key component of all the basic cell structures for improving the quality of food. For optimum crop yield, plants require adequate P from very early growth stages (Grant *et al*., 2001; Nadeem *et al*., 2011) and a significant P uptake is noted in seedling roots from 5th day after sowing (Nadeem *et al*., 2011).

Phosphorus is considered least mobile and available nutrient in rhizosphere and therefore applied in the form or organic or inorganic fertilizer products. Phosphorus deficiency is particular in low input agricultural systems (Raghothama and Karthikeyan, 2005) especially in Pakistani soils where 90% soils are deficient in available P (Bajwa, 1990; Anonymous, 2013) and this low P availability caused lower cell production and ultimately low final crop harvest (Assuero *et al*., 2004; Colomb *et al*., 2000; Plénet *et al*., 2000). Problems of P fixation and ultimately of deficiency, limit the crop production on >90% soils of the Pakistan due to high pH, calcareous nature and presence of P in relatively insoluble form of tri-calcium phosphate (Aziz *et al*., 2005; Aziz *et al*., 2006; Gill *et al*., 2004). Availability of P under such conditions can be improved through adopting certain management practices like using its high rates, better placement, integrated use of its organic and inorganic sources and growing P solublizing cultivars. High costs and environmental concerns of inorganic fertilizers and slow release of nutrients from organic sources are pushing the farmers for integrated use of these sources. Therefore judicious use of these nonrenewable resources is most critical for the sustain productivity of current cropping systems (Vance *et al*., 2003). Inorganic sources even at half dose of their recommendation in combination with organic sources can produce yields similar to those of full inorganic source dose (Mandal and Sinha, 2004). Banding of P also reduces fixation due to minimal contact between the soil and banded fertilizer.

Phosphorus mobilizing process may differ with plant species and even among cultivars (Gahoonia *et al*., 2000) due to their relative differences in morphological and physiological characteristics. These features make these plants or cultivars successful even on low P soils (Gahoonia *et al*., 1997; Neumann *et al*., 1999). On the other hand, inefficient cultivars retain more proportion of total P in their roots instead of its translocation from roots to shoots and ultimately to the portions of economic importance (Nadeem *et al*., 2013). Exploitation of differences for P acquisition and utilization among cultivars is a promising strategy for increasing P use efficiency of applied as well as native soil P (Aziz *et al*., 2005). A little knowledge is available about the genotypic differences among canola hybrids for their capacity to grow on soils with low P availability (Marschner *et al*., 2007). Recently introduced canola hybrids have good genetic makeup with high yield potentials but the test of their performance and response to P fertilizers "considering the non renewable nature of P (Vance *et al*., 2003)" need immediate attention for realizing their full potential. Therefore a breeder must already test fertilizer effects on the hybrids.

Though plenty of literature is available on P management in field crops, nonetheless little work has been done to understand the management of P and its impact on yield and quality of canola in Pakistan. We hypothesized that integrated use of organic and inorganic P sources have better effects on canola yield and oil quality. Therefore, this study was planned to unravel the impact of integrated use of organic and inorganic P sources and genetic diversity among canola hybrids for better P acquisition on yield and quality of canola under agro climatic conditions of Faisalabad Pakistan.

MATERIALS AND METHODS

***Site description*:** Two years (2004-2006) field experiment was conducted during winter seasons (October to April) at Agronomic Research Area, University of Agriculture, Faisalabad, Pakistan (31° 25′ lat. N; 73° 09′ long. E, 185 m). Climatic data during the both growing seasons including monthly average temperature (°C), relative humidity (%) and rainfall (mm) are presented in Figure 1.

***Experimental details*:** Before sowing the canola crop, soil

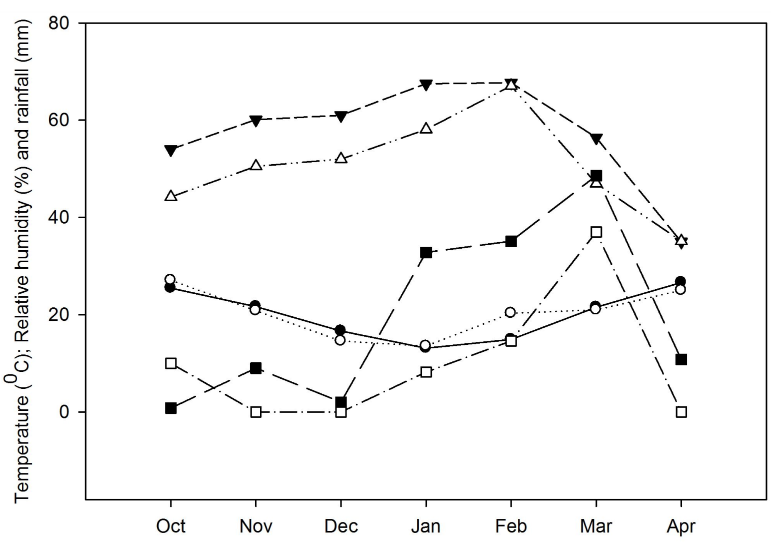

Figure 1. Average temperature (in °C; solid circles "●" and hollow circles "○"); relative humidity (in %; solid triangle "▼" and hollow triangle "∆") and rainfall (in mm; solid square "■" and hollow circles "□") during 2004-05 and 2005-06, respectively.

and FYM samples were collected for different chemical analysis. Soil samples were also collected from the field with the help of a soil auger to a depth of 0-30 cm prior to fertilizer application. Farmyard manure samples were air-dried, ground and passed through a 2 mm sieve and were analyzed for the physico-chemical properties. Soil was analyzed for its various chemical properties by using the methods as described by (Chapman and Pratt, 1962). Different soil chemical analyses showed that experimental soil have 0.71% organic matter, 7.9 pH, 0.052% N contents, 5.42 ppm available Olson P and 175 ppm K contents.

Percentage of sand, silt and clay was determined by Bouyoucos hydrometer method using one percent sodium hexametaphosphate as a dispersing agent. Textural class was determined by using the international textural triangle (Moodie *et al*., 1959). Soil belonged to Lyallpur soil series (aridisol-fine-silty, mixed, hyperthermic Ustalfic, Haplarged in USDA classification and Haplic Yerrmosols in FAO classification) with these average characteristics in 0-30 cm depth: 66% sand, 16% silt and 18% clay. Farm yard manure and NPK analysis during study period showed 0.54% and 0.56% N, 0.24% and 0.23% P, 0.36% and 0.34% K during 2004-05 and 2005-06, respectively.

Experiments were laid out in split plot design and were replicated three times. Canola hybrids were kept in main plots and P application treatments in the sub-plots. Net sub plot size was 2.4 m × 5 m.

The treatments comprised of two canola hybrids (Hyola-401 and Hyola-43) which were tested under different P application rates applied either through broadcasting or through side-drilling, alone and in combination with FYM. Individual treatments are composed on T_1 (control), T_2 (only FYM @ 10 t ha^{-1}), T_3 (P @ 30 kg ha^{-1} broadcast), T_4 (P @ 60 kg ha^{-1} broadcast), T_5 (P@ 30 kg ha^{-1} + FYM @ 10 t ha^{-1}), T_6 (P@ 60 kg ha^{-1} + FYM @ 10 t ha^{-1}), T_7 (P@ 30 kg ha^{-1} side drilling), T_8 (P@ 60 kg ha^{-1} side drilling), T_9 (P@ 30 kg ha^{-1} side drilling + FYM @ 10 t ha^{-1}) and T_{10} (P@ 60 kg ha^{-1} side drilling + FYM @ 10 t ha^{-1}).

Pre-soaking irrigation of 10 cm was applied before seedbed preparation. When soil reached to proper moisture level, seedbed was prepared. The crop was cultivated on October 11 and 08 and was harvested on April 7 and 10 in 2004-05 and 2005-06, respectively. Sowing was done with the help of single row hand drill in 30 cm apart rows using seed rate of 5 kg ha^{-1}. Plant population was maintained by thinning. Nitrogen fertilizer was applied @ 90 kg ha^{-1}. Half dose of N and all the P according to treatment were applied at sowing, while remaining N was applied with first irrigation. Pyrethroide was sprayed at the rate of 1 liter ha^{-1} once for the control of aphids.

Five plants were randomly selected from each plot for recording average plant height plant^{-1} and seed and biomass yield was measured on plot^{-1} basis and converted to kg ha^{-1}. The crop was harvested and left in respective sub plots for almost one week for sun drying at physiological maturity when more than 50% siliquae turned brown. Sun dried crop was threshed manually. Oil contents were determined by Soxhlet Fat Extraction method (AOAC, 1990) while protein contents of seeds were calculated by determining nitrogen contents using Kjeldahl method (Bremner, 1964). Gas liquid chromatography was done for determining relative composition of different fatty acids in oil (Martin, 1979). Phosphorus concentration in seeds and straw was determined from digested samples using vanadate-molybdate colorimetric method (Van Veldhoven and Mannaerts, 1987).

***Statistical analysis*:** All the data collected were statistically analyzed using computer package SAS, 9.1 by using the Fisher's analysis of variance technique and LSD test at 5% probability was used to compare the differences among treatments' means (SAS, 9.1). To determine the significance of treatments, least significant differences (LSD) were estimated at 5% probability level and Tukey's test was used. Simple correlation co-efficient and regression equations for yield components and grain yield as well as for grain quality parameters and grain yield were also computed.

RESULTS

***Seed yield*:** A linear increase was observed in seed yield of both canola hybrids treated with different P amendments during both growing seasons (2004-05 and 2005-06) as shown in Figure 2. Overall Hyola-43 (H_1) performed better (p>0.05%) in terms of final seed yield as compared to Hyola-401 in both growing seasons (Fig. 2). Hyola-43 produced 2387±14.92 and 2178±17.92 kg ha^{-1} seeds during 2004-05 and 2005-06, respectively. Phosphorus amendments had significant effects on final seed yield and Hyola-43 produced higher seed yield (2650±55.44 and 2523±66.65 kg ha^{-1}) during both years when 60 kg P ha^{-1} was side drilled along with FYM during 2004-05 and 2005-06, respectively) as shown in Fig. 2. Minimum seed yield (1915±55.44 and 1500±66.65 kg ha^{-1}) was recorded with Hyola-401 was treated with control (No P and No FYM) treatment during 2004-05 and 2005-06, respectively.

***Seed oil and protein contents*:** The primary objective of canola cultivation is oil production, so the seed oil content is its most desired parameter. On the other hand protein obtained in the form of seed cake as a byproduct after oil extraction. Hybrids and P applications had significant effect (P>0.05%) on seed oil contents during both years (Fig. 3). Hyola-43 produced significantly more seed oil contents (41.40 ±0.33% and 39.97±0.34%) as compared to Hyola-401 (39.76 ±0.33% and 37.86±0.34%). As far as P applications are concerned, maximum seed oil contents were produced by Hyola-43 when 60 kg P ha^{-1} was side drilled along with FYM (42.40±1.23% and 40.89±1.29%) during both years. Although the maximum oil contents were recorded when 60 kg P ha^{-1} was applied along with FYM, but it was statistically similar to 30 kg P ha^{-1} along with FYM during both years (Fig. 3).

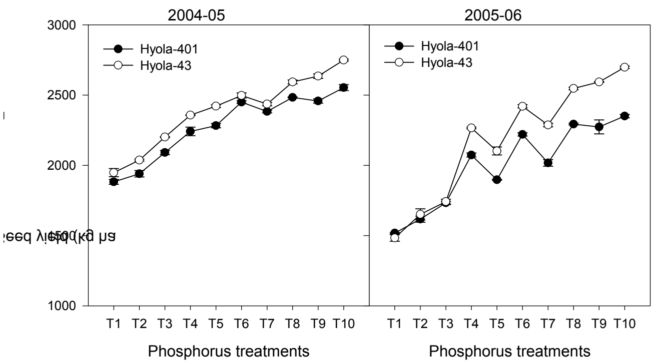

Figure 2. Seed yield (kg ha^{-1}) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

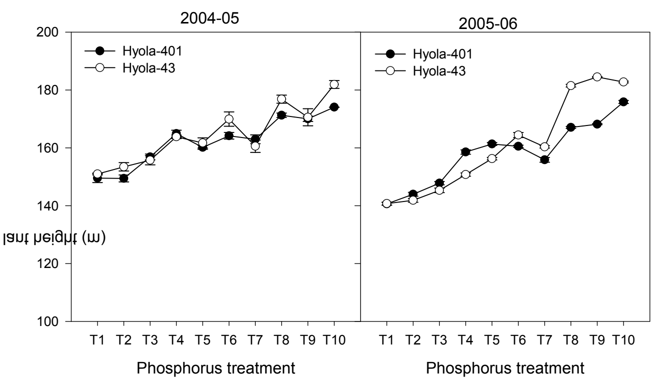

Figure 3. Seed oil (%) production in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

Although significant effect of hybrids and P applications was observed on seed proteins contents (Fig. 4). Proteins contents were statistically higher (26.15±0.07%; 26.40±0.31% in Hyola-401 and 26.23±0.12%; 26.10±0.20% in Hyola-43 during 2004-05 and 2005-06, respectively) when 60 kg P ha^{-1} was applied along with FYM (10 t ha^{-1}) which is statistically at par with the treatment where 30 kg P ha^{-1} along with FYM during both growing seasons. Although there were significant differences among protein contents among both canola hybrids in T_1, T_2 and T_3 treatments; however these variations were disappeared with P amendments during both growing seasons (Fig. 4).

Oleic acid contents**:** Canola hybrids and P applications significantly affect the oleic acids contents (Fig. 5a). During first growing season (2004-05), Hyola-43 significantly produced higher oleic acid contents as compared to Hyola-401 with all P applications as shown in Figure 5a. A slight decrease was observed in oleic acid contents at T_7 in both canola hybrids. Similar oleic acid production trend was observed during 2005-06; however, there were not much pronounced effects of canola hybrids as observed during 2004-05. The maximum oleic acid contents (62.12±0.39%) were produced by Hyola-43 when 60 kg P ha^{-1} was side drilled along with FYM, whereas minimum oleic acid (57.77±0.03%) was produced by Hyola-401 at control treatment (T_1).

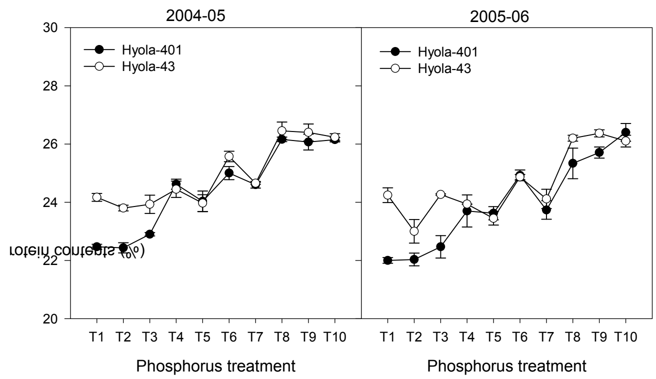

Figure 4. Seed protein contents (%) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

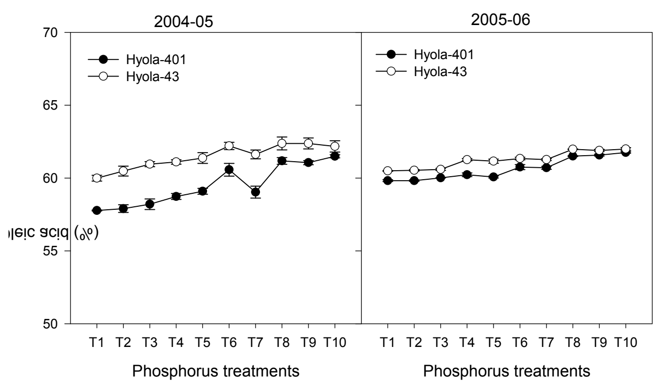

Figure 5a. Oleic acid contents (%) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

Linoleic acid contents**:** The maximum linoleic acid contents (20.30±0.11%) in Hyola-401 and (20.66±0.12%) in Hyola-43 were recorded in T_8 (side drilled P @ 60 kg ha^{-1}) which was statistically at par with T_9 (side drilled P @ 30 kg ha^{-1} along with FYM at the rate of 10 t ha^{-1}) and T_{10} (side drilled P @ 60 kg ha^{-1} along with FYM) in 2004-05 (Fig. 5b). Minimum linoleic acid contents (19.20±0.16% and 19.18±0.04%) was recorded in control (T_1) which was statistically at par with T_2, T_3, T_4, T_5 and T_7. During 2005-06, maximum linoleic acid contents (20.54±0.09% in Hyola-401 and 20.97±0.06% in Hyola-43) were observed in T_{10} (side drilled P @ 60 kg ha^{-1} along with FYM at the rate of 10 t ha^{-1}) that was statistically at par with T_9 (side drilled P @ 30 kg ha^{-1} along with FYM at the rate of 10 t ha^{-1}) and T_8 (side drilled P @ 30 kg ha^{-1}) treatments. Minimum linoleic acid content was produced in T_1 (control) that was statistically at par with T_2.

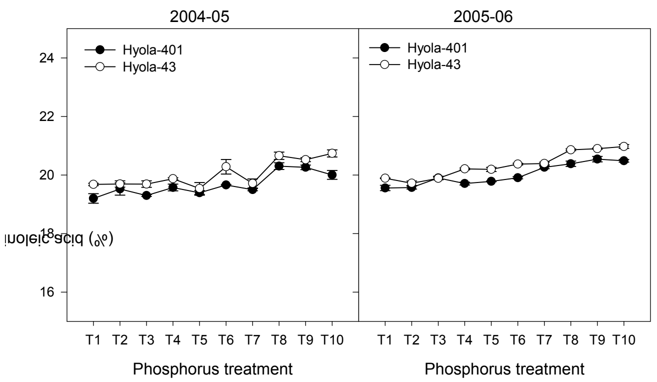

Figure 5b. Linoleic acid contents (%) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

Linolenic acid contents: Canola hybrids and P applications showed significant effects on linolenic acid contents. Conversely to oleic and linoleic acid, linolenic acid contents were decreased with the application of P during both growing seasons as shown in Fig. 5c. More linolenic acid contents were recorded in Hyola-401 as compared to Hyola-43 with all P application treatments (Fig. 5c). Minimum linolenic acid contents (9.41±0.08%; 9.30±0.05% in Hyola-401 and 8.75±0.03%; 8.47±0.13% in Hyola-43 during 2004 and 2005, respectively) were recorded in T_{10} treatment and these contents increased with the decrease in P application levels. Maximum linolenic acid contents were recorded in control (T_1) (9.89±0.04%; 10.16±0.11% in Hyola-401 and 9.01±0.10%; 9.80±0.05% in Hyola-43 during 2004 and 2005, respectively) as shown in Figure 5c.

Straw and grain P contents: A significant effect of P applications was observed on accumulated P contents in straw and grain contents during both growing seasons as shown in Figure 6a & b. Maximum grain P contents were recorded at T_{10} (0.50±0.01%; 0.45±0.02% in Hyola-401 and 0.52±0.01%; 0.45±0.02% in Hyola-43 during 2004-5 and 2005-06, respectively). Similarly, maximum straw P contents were recorded at T_{10} (0.21±0.01%; 0.18±0.02% in Hyola-401 and 0.22±0.01%; 0.20±0.01% in Hyola-43 during 2004-5 and 2005-06, respectively). Minimum grain and straw P contents were observed in control treatment (T_1) as shown in Fig. 6a & b respectively. During 2005-06 both canola hybrids showed non-significant differences for straw P contents.

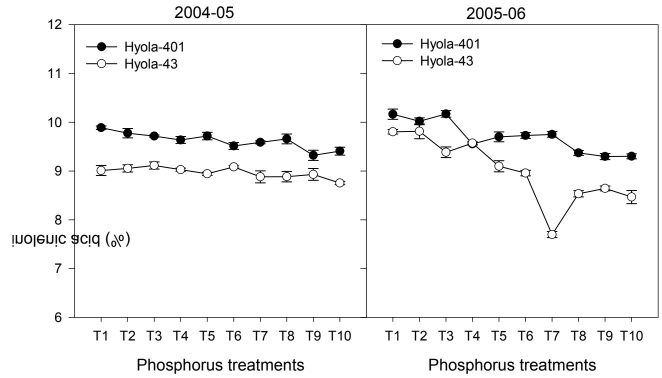

Figure 5c. Linolenic acid contents (%) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

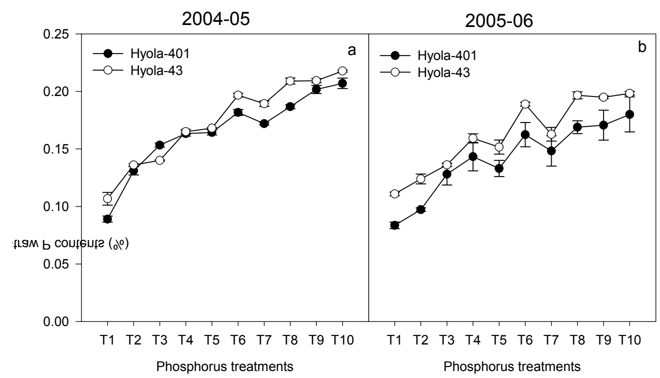

Figure 6. Straw P contents (%) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

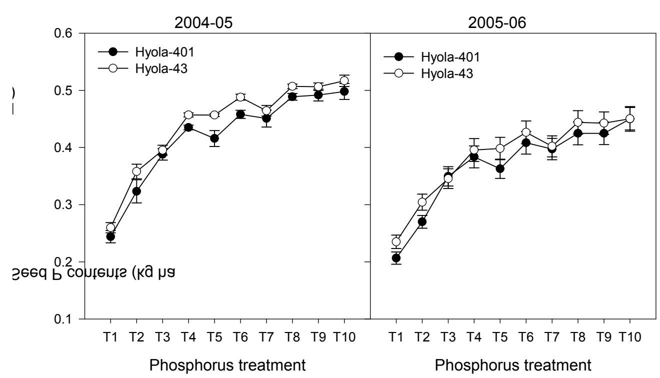

Figure 7. Seed P accumulation (mg) in canola hybrids as affected by different phosphorus amendments. Data are means and vertical bars indicate ± SE for *n*=3.

DISCUSSION

Crop growth trend and seed yield: Higher seed yield by Hyola-43 in T_{10} (side drilled P @ 60 kg ha^{-1} along with FYM) was due to the better performance for agronomic parameters like number of siliquae plant, seeds siliqua and 1000-seed weight (data not shown) in response to better availability of phosphorus during very early growth stages. Newly growing seedling start to uptake external P from 5^{th} day after sowing (Nadeem *et al*., 2011) and early seedling P nutrition has significant effects on final crop harvest (Grant *et al*., 2001). Genotypic differences in canola was observed for nutrient use efficiency (Shenoy and Kalagudi, 2005) and this is correlated with relative ability of genotypes for solubilizing and acquisition of P. High P application rates (Cheema *et al*., 2001), FYM (Mandal and Sinha, 2004) and side-drilling of fertilizers play a vital role for increasing final crop harvest in brassicas. Better seed yield from inorganic fertilizers in combination with FYM is mainly due to numerous positive effects of FYM on inorganic fertilizers; like improving water holding capacity, blockage of P absorbing sites in the soil, stimulation of soil microbial biomass thus increasing the rate of mineralization and supplying more nutrients (Ayaga *et al*., 2006; Blake *et al*., 2000; Ghosh *et al*., 2006; Ghosh *et al*., 2004; Jiang *et al*., 2006; Mandal and Sinha, 2004; Whalen *et al*., 2002). Farmyard manures also increase the availability of P by reduction in P sorption (Damodar *et al*., 1999). Higher rates and better placement of P fertilizers is chiefly responsible for increased availability of P thus resulting in higher seed yield (Gokmen and Sencar, 1999; Lickfett *et al*., 1999; McKenzie *et al*., 2003; Turk and Tawaha, 2002).

Yield and quality attributes of canola: There existed a highly significant relationship of yield and quality attributes with seed yield during both the years (Table 1). The correlation coefficient values (r) varied between 2.9% and 92.5% during 2004-05 and between 73.2% and 97.3% during 2005-06. Non-significant correlation coefficient in case of number of branches per plant in 2004-05 indicated no relationship with seed yield. Negative value of correlation coefficient for linolenic acid during 2005-06 shows its inverse relationship with seed yield. All the other yield and quality parameters had strong positive correlation with seed yield. Study results are regarding correlation of different growth and yield parameters are in confirmation with other researchers (Ali *et al*., 2003; Chongo and McVetty, 2001) who observed a significant positive correlation between harvest index and grain yield.

Canola oil and protein content response to P amendments: Our results are in line with other researchers (Lickfett *et al*., 1999; Sawan *et al*., 2006), who found increased oil concentration at higher P levels. Increase in oleic acid and linoleic acid contents could be due to the better availability of N and S from FYM and due to high P rates (Ahmad and Abdin, 2000). Higher P rates produced more oleic and linoleic acid contents (Sawan *et al*., 2006). These results clearly indicate the role of high P levels for improving the quality of canola oil, because at higher P levels increased oleic and linoleic acid contents and reduced linolenic contents ensures high quality of oil. In addition to different fatty acids accumulation in canola seeds, protein contents also increased due to higher rates of P and these results are in confirmation with other researchers (Lickfett *et al*., 1999; Sawan *et al*., 2006) who reported that higher levels of P produced higher protein contents.

Table 1. Correlation of seed yield (kg ha^{-1}) with yield and quality parameters of canola.

Character	Year	
	2004-05	2005-06
Number of plants per m^2	0.255*	0.894**
Plant height (cm)	0.841**	0.916**
Number of branches / plant	0.029^{NS}	0.894**
Number of siliquae per plant	0.888**	0.944**
Number of seeds per siliqua	0.841**	0.926**
1000-achene weight	0.925**	0.960**
Biological yield (kg ha^{-1})	0.913**	0.973**
Harvest index (%)	0.885**	0.948**
Oil content (%)	0.866**	0.846**
Protein content (%)	0.620**	0.791**
Oleic acid content (%)	0.866**	0.877**
Linoleic acid content (%)	0.804**	0.872**
Linolenic acid content (%)	0.709^{NS}	-0.732 **
Seed phosphorus content (%)	0.862**	0.914**
Straw phosphorus content (%)	0.813**	0.873**

** = Highly Significant at 1% probability

Phosphorus accumulation in canola seeds due to P amendments: Availability of P increases with integrated use of FYM and inorganic fertilizers that can be linked to positive effects of FYM (Ayaga *et al*., 2006; Mohanty *et al*., 2006). Broadcasting of fertilizer resulted in reduced P accumulation in crop (Lu *et al*., 1987). Differential P accumulation by canola genotypes can be attributed to their genetic variation for P acquisition possible due to differences in root lengths. Canola genotypes differ for shoot P concentration and genotypic differences among brassica cultivars for shoot P concentration are well documented (Aziz *et al*., 2006; Gill *et al*., 2004; Greenwood *et al*., 2005; Marschner *et al*., 2007; Mohanty *et al*., 2006).

Conclusion: High P application in combination with FYM significantly affect the quality of canola oil as well as P accumulation in seeds. Higher accumulation of P in seeds favors the next crop by fulfilling the inorganic P requirements of the developing seedlings on soils low in inherent available P during few weeks of early growth. Study results suggest that P should be applied in combination with FYM for getting good quality canola crop and to sustain the fertility of soil in agro climatic conditions of Faisalabad, Pakistan.

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