Improving Phosphorous Uptake Efficiency and Quality of Maize Through Optimization of Basal Application

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Abstract

Phosphorous (P) dearth in alkaline calcareous soils of Pakistan renders P unavailable for plant uptake. Consequently, affects grain yield and is major constraint to achieve desired yield potential of field grown maize (*Zea mays* L.). The objectives of this study were to enhance phosphorus use efficiency (PUE) by determining thresholds of basal applied P. Treatments comprised of five rates of basal applied P viz. 0, 30, 60, 90, 120, and 150 kg ha^{-1}. The experiment was laid out in randomized complete block design (RCBD) with a net plot size of 2.7 m × 5 m, during autumn season 2013. Improved agronomic attributes; i.e., plant height, cob length, grains per cob, grain-pith ratio, 1000-grain weight, biomass and grain yield were observed at 120 kg P ha^{-1}. Likewise, higher agronomic use efficiency of P, grain P content and total P uptake were recorded. Conclusively, application of P at 120 and 150 kg ha^{-1} would improve grain yield and quality of maize over control. However, 120 kg P ha^{-1} manifested more promising results than 150 kg P ha^{-1}.

Introduction

Maize, as being C4 plant crop, involves use of the high amount of nitrogen (N) and P; maize is extensively grown in Pakistan over an area of 1.33 million hectares with a total production of 6.13 million tons (Govt. of Pakistan, 2017). Hence, maize provides the raw material for numerous agro-based industries. Contrarily, maize productivity is hampered under alkaline calcareous soils of Pakistan. Furthermore, maize yield gaps can be connected to poor resource availability, higher input costs and poor infrastructure of the marketing system. Crops requiring continuous and sufficient amounts of major nutrients like N and P often suffer soil nutrient imbalances (e.g. nutrient output not replaced by inputs) (Bindraban et al, 2000). Phosphorous is required by the plants to store and transfer high energy compounds for growth and to form some vital compounds (Kronvang et al, 2007). Plants are able to initiate the synthesis of complex compounds (for example the secretion of root exudates; organic acids, phosphatases, and the bringing on some inorganic transporters of phosphate (Pi) as well as establishing mycorrhizal symbiotic associations with fungi that promotes P acquisition) when prone to early season P scarcity (Yang and Finnegan, 2010).

Phosphorus deficiency is invariably a common crop growth and yield-limiting constraint in calcareous soils of Pakistan. These soils containing high calcium carbonate content with pH ranging from 7 to 9, favour a series of chemical processes such as adsorption and precipitation and reduces P solubility (Khan et al, 2005). Furthermore, soils with greater fixation capacity augment fertiliser use (Khalil et al, 2010). Continuous cropping without proportionate nutrient replenishment is reported to contribute to low P content of many soils (Bünemann, 2003).

Early season plant growth and development is solely dependent on soil applied P. Thus, any limitation in the soil P availability can adversely affect root initiation and growth of younger tissues (Grant et al, 2001). Adequate P ensures rapid growth, favours earlier maturity and improves vegetative growth parameters i.e. biomass yield, plant height, grain yield, the number of rows and grains per ear and P uptake efficiency of maize (Almeida et al, 2005). Deficiency of P causes abnormal rows and grain starch accumulation. Moreover, deficiency symptoms are not apparent unless soil P reserves completely devoid of P. Maize seeds contain both inorganic and organic P. However, inorganic P

is more stable than organic while the concentration of inorganic P varies sharply due to soil P availability (Zakirullah and Khalil, 2012).

When seed P is depleted, plant growth is essentially supported by the acquisition of P through soil medium (Hammond et al, 2009; Vázquez et al, 2009). Phosphorous deficiency in maize can be attributed to the unavailability of external P, impaired seed-phytate hydrolysis and abiotic stresses (Parentoni and De Souza Júnior, 2008). Increase in seed P contents depicts advantageous effects at early seedling growth stages and thus supplements plant P needs. Therefore, higher seed assimilated P and optimal uptake can buffer against short-term variations in soil P supply at later stages of the plant development (Grant et al, 2001). Although, the seed P contents mark in little to the final P content of the mature plant, however, contribute significantly to the P nutrition of young seedlings. Greater seed P provides higher yields output by upregulating synthesis of sugar-phosphates (Grant et al, 2001; Zhu and Smith, 2001).

Numerous other strategies are employed to improve seed P content. However, basal applied P is more economically feasible and useful from perspectives of farmers. There is scarce information available about the association of seed P contents with emergence, growth, yield and quality of maize traits . Moreover, optimisation of basal applied P regulates available P for plant uptake under alkaline calcareous soils of Pakistan. Hence experiment was conducted with the objectives (i) to study basal applied P rates as potential resource to affect agronomic performance of maize, (ii) to determine threshold level of basal applied P to optimise yield, agronomic P use efficiency and P-mediated quality maize traits.

Materials and Methods

Agronomical trails

An experiment was conducted at the Agronomic Research Area, University of Agriculture, Faisalabad, Pakistan (31° 25′ 45″ N, 73° 4′ 44″ E) in randomized complete block design (RCBD) with a net plot size of 2.7 m × 5 m (total area of 243 m^2 for all plots) by using three replications during Autumn season, while sowing was performed on 25th July, 2013. Soil samples were taken at 20 cm depth before sowing and composite samples (no = 3) were made.

A synthetic maize cv. MMRI-Yellow (Maize and Millet Research Institute, Yusafwala-Sahiwal) was sown by using seed rate of 25 kg ha^{-1} with planting distance of 30 cm in 45cm apart rows and 5 rows per plot. Thinning

was done to maintain 16 plants per row

Soil analyses

Physico-chemical analyses showed soil texture as sandy clay loam, determined by hydrometer method (Ishaq et al, 2001). Total nitrogen (0.23%) in the soil was determined by Kjeldahl method (Pandiaraj et al, 2015). The available P (5.51 mg kg^{-1}) was determined by sodium bicarbonate method (Parnell et al, 2002a). Available potassium (171 mg kg^{-1}) in the soil was determined with ammonium acetate solution (Khan et al, 2014) and organic matter content (0.91%) was determined by using the method of Wahid et al (2014). Calcium carbonate content (4.7%) was measured by acid dissolution (Allison and Moodie, 1965). Available zinc (0.62 mg kg^{-1}) in soil was extracted with 0.1 N HCl extraction solution and determined with atomic absorption spectrophotometer (Whitney, 1998). The hot-water extraction procedure was used to extract available B in the soil and was measured (0.45 mg kg^{-1}) colorimetrically using azomethine-H (Tesfahunegn et al, 2016).

Fodder crop was harvested before collecting the soil samples and the seed bed was prepared by applying presoaking irrigation. When soil field capacity moisture was attained, a tractor mounted cultivator was used for ploughings followed by rotavator. After each cultivation, planker was used to break the clods and levelling the field.

P treatments and nutrient fertilization

Diammonium phosphate (DAP) was applied to provide P (0, 30, 60, 90, 120, 150 kg ha^{-1}). Remaining N was applied through urea to fulfil 130 kg ha^{-1} requirement and sulphate of potash (SOP) was used to provide 62 kg ha^{-1} potassium (K) (Ijaz-ul-Hassan et al, 2012). All nutrients were applied at planting except N which was applied in three splits i.e., 44.5 kg ha^{-1} at the time of planting, 28 kg ha^{-1} at V8 (stage at which plant bearing 8th fully opened leaf) and 57 kg ha^{-1} at V15 (stage at which plant bearing 15th fully opened leaf). Overall, 8 canal water irrigations exclusive of presoaking irrigation, were applied. The field was kept free from all kind of weeds during the crop season by adopting necessary cultural practices and chemical application. Similarly, mandatory plant protection measures regarding insects and pests' control of the maize crop were adopted.

Plant sample collection, P analysis and P uptake evaluation

For analysis of P, plants were selected from middle two rows of each experimental treatment plot and stem and leaf tissue samples were taken at sixth node

from base of the plants as described by Deleers et al (1985), at crop maturity (i.e., 105 days after sowing) on 7th November, 2013. Cobs were separated from plants and dried to 12% moisture content by placing in the field and then seeds were collected after threshing. Homogenous seeds were then separated from whole seed lot on basis of seed size (Nadeem et al, 2014). Threshed grains and vegetative samples were dried in an oven at 70°C for 72 hours to attain a constant dry weight. Dried samples were finely ground with a Wiley mill fitted with a stainless-steel chamber and blades. Grains and vegetative samples were digested separately by using 2:1 (v/v) (HNO$_3$:HClO$_4$). Analysis of total P was carried out by the vanadate-molybdate yellow colour method (Chapman and Pratt, 1961) and total P concentration was calculated with the help of spectrophotometer (Shimadzu, UV-1201, Kyoto, Japan) at 410-nm wavelength and standard curve was achieved by using Microsoft Excel program. After plotting the absorbance curve, same procedure was followed for unknown samples. Phosphorus concentration in the unknown samples was calculated from the calibration curve. However, percentage of P can also be calculated

$$\% \; P \; in \; plant \; material = ppm \; P \; (from \; calibration \; curve) \times \frac{R}{Wt} \times \frac{100}{1000}$$

by using the given formula below:

Where: R = Ratio between total volume of the digest/aliquot and the digest/aliquot volume used for measurement.

Wt = Weight of dry plant material (g)

After that P concentration was calculated as mg g^{-1} of dry weight by conversion method.

Phosphorus uptake was calculated by using the following formulas (Yaseen and Malhi, 2009) and agronomic use efficiency of P (AEP) was calculated by Memon et al (2011) method:

P uptake (kg ha^{-1}= P concentration in grain /straw (%) × grain/straw yield (kg ha^{-1})

Total P uptake (kg ha^{-1}) =P uptake in grain+P uptake in straw

$$Agronomic \; efficiency \; (kg \; kg^{-1}P) = \frac{Yield \; (fertilized \; treatment) - Yield \; (control \; treatment)}{(Fertilizer \; P \; applied \; (kg \; ha^{-1})}$$

Statistical analysis

Data regarding agronomic traits were analysed by using Statistix 8.1 statistical software (Software, Tallahassee FL 32317. The USA) and means comparison was done by Tukey's HSD (honest significant difference) test while graphs regarding agronomic and quality parameters were formulated by employing Microsoft Excel program.

Results and discussion

Maize production requires rational management of P under limited P environments that become even more challenging under calcareous soils. To increase P acquisition and internal use efficiency of field crops, incremental higher soil P application is the only way to achieve desired yield targets. Therefore, this study was

Table 1 - Effect of soil applied P levels on different agronomic traits of maize

Treatments	Plant height (cm)	Cob length (cm)	Grains per cob	Grain-pith ratio	1000-grain weight (g)
P rates (kg ha^{-1})					
0 (Control)	142 c	15.0 c	298 d	2.18 c	164 c
30	149 bc	17.5 bc	396 c	2.63 bc	189 b
60	183 abc	19.1 b	444 bc	3.38 abc	198 ab
120	213 a	24.6 a	630 a	4.22 a	218 a
150	217 a	24.6 a	608 a	3.70 ab	218 a
HSD	**63.5**	**3.43**	**76.8**	**1.218**	**22.6**

Any two means not sharing a letter in common within a column differ significantly at p≤0.01.
HSD= Honestly Significant difference

conducted to explore the benefits of increased P supply to maize plants in improving its reproductive growth, agronomic performance and in peculiar P allocation towards grains under varying levels of soil applied P.

Agronomic traits

Data regarding agronomic traits of maize i.e. plant height, cob length, grains per cob, grain-pith ratio and 1000-grain weight (Table 1), showed significant difference (P ≤ 0.01) under different soil applied P levels. An increasing trend of plant height was observed irrespective of homogenous emergence of plants followed by soil applied P levels (data are not shown). Maximum values of plant heights were observed at 120 and 150 kg P ha^{-1} , and least plant height was calculated at control. Cob length also increased significantly (P ≤ 0.01) by increasing soil P supply to plants at 150 and 120 kg ha^{-1} as compared to lower levels of P; whereas minimum cob length was observed both under control and 30 kg P ha^{-1}. Similarly, maximum number of grains per cob was obtained (P ≤ 0.01) at higher soil applied P levels followed by lower P levels; least at the control. Grain-pith ratio is a determinant of grains' number to the size of pith, and increased P supply tended to affect this yield trait also where significant response was attained under 120 kg P ha^{-1} which was the same as all other applied levels; least effect was at 0 kg P ha^{-1} (control). Biomass yield and grain yield resulted

highest under 120 and 150 kg P ha⁻¹ followed by other P levels except for 30 and 0 kg P ha⁻¹ where values were minimum (Figure 1-2).
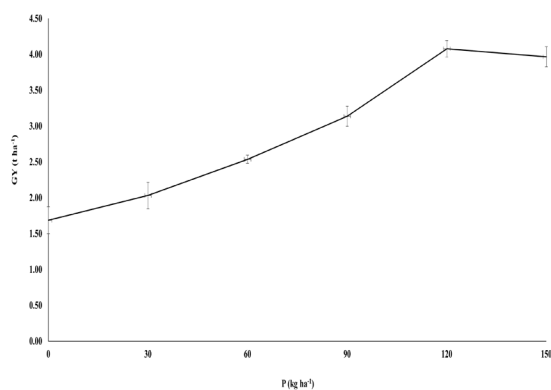

Fig. 1 - Effect of soil applied P levels on grain yield of maize

The reduction in plant growth has been observed in terms of decreased plant height in addition to other retardation effects on plant developmental stages under P stress which showed that optimum soil P supply provided better vegetative growth. This helped to explain increased phosphorus use efficiency; (PUE) in terms of better vegetative development of the crop. Other studies showed that plants start to economise on Pi under reduced P supply and utilise leaf P stored in vacuoles well before its depletion thus reduced their shoot growth (Rouached et al, 2011; Veneklaas et al, 2012). Ai et al (2009) also observed the increase in plant heights for hybrid maize when followed by both organic and inorganic sources of P. This showed that sufficient P supply during early season favours higher P acquisition through vigorous root growth and similar results were witnessed by Grant et al (2001). But the results of Onasanya et al (2009) were contradictory with our findings as they calculated higher plant heights at
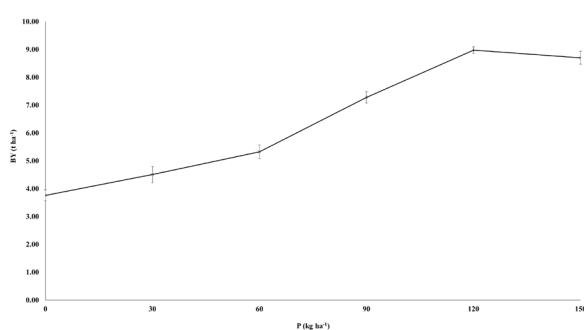
maximum doses of N without P application. Moreover, roots density and their spatial distribution in the soil may also vary and explain better nutrient acquisition and water uptake by the plants which were not the part of this study.

Grain yield contributing parameters were improved under increased P levels and depicted greater cob length, grains per cob and higher grain-pith ratio as compared to control. This might be due to the additive influence of higher P levels and improved PUE by plants during reproductive development. In addition, more N availability during vegetative growth along with sufficient P supply implicated for synergistic effects on higher photosynthates accumulation in attaining more grains and cobs' girth. Other studies also found an incremental increase in plant height, a number of cobs per plant and grains per cob followed by higher applied P levels (50 and 75 kg ha⁻¹) but no benefits were observed with excessive application of P under saline conditions (Khan et al, 2005). This shows that crop nutrient management practices need thorough consideration based on different soil chemical properties. It is also observed that a number of grains are directly related to soil P deficiency levels as fewer seeds are the result of low P in plants, particularly in cereals and legume crops (Fageria and Santos, 2008). Reduction in seed numbers occurred through reduced numbers of fertile spikes and reduced numbers of kernels per spike. Moreover, 1000-grain weight and grain-pith ratio were also boosted up due to better photosynthesis rate and assimilate partitioning in grains at maturity. In the present study, retarding effects on growth and reproductive development of crop were also observed under selected soil conditions where soil P was found deficient thus reduced the cob length and number of grains per cob. Recently, Rehim et al (2016) explained positive results under optimum P nutrition that enabled plants to maintain their reproductive growth through better photosynthesis rate and transformation of sugars and starches due to improved mineral nutrition and higher P availability. Ultimately, maximum dry matter and grain yield were obtained as a result of better agronomic PUE by the crop under optimum P availability.

Phosphorus Uptake Traits

Agronomic efficiency of P (AEP) was computed to estimate the yield response of maize per kg of P applied to the soil which was obtained maximum where 120 kg P ha⁻¹ was applied and almost similar effects were observed at 150, 90 and 60 kg P ha⁻¹ and minimum at 30 kg P ha⁻¹ (Figure 3). Similarly, grain P contents increased from 1.5 to 3.2 mg kg⁻¹ in response to soil
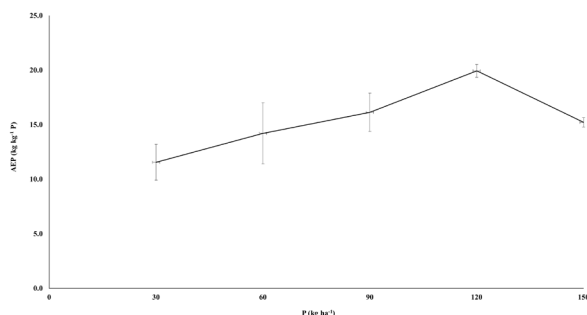

Fig. 2 - Effect of soil applied P levels on biological yield of maize

Fig. 3 - Effect of soil applied P levels on agronomic efficiency of P (AEP) in maize

applied P from 30 to 150 kg P ha^{-1}, respectively (Figure 4). Total P uptake also increased linearly at increased P levels and maximum values were observed under 120 and 150 kg P ha^{-1} (Figure 5). While 30 and 60 kg P ha^{-1} levels performed negatively to reduce this attribute including control treatment.
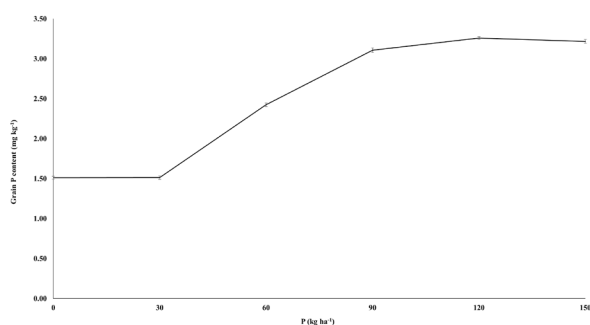

Fig. 4 - Effect of soil applied P levels on grain P content of maize

Greater AEP responses at increasing P levels clearly demonstrated better PUE by plants and might be the effect of better P acquisition by roots thus helped the plants to produce grains with more grain weight and yield as compared to lower P availability. Ultimately, maximum dry matter and grain yield were obtained as a result of better agronomic PUE by the crop (Figure 1-2). More AEP values at lower P levels indicated more roots competition and thereby enhanced the P uptake
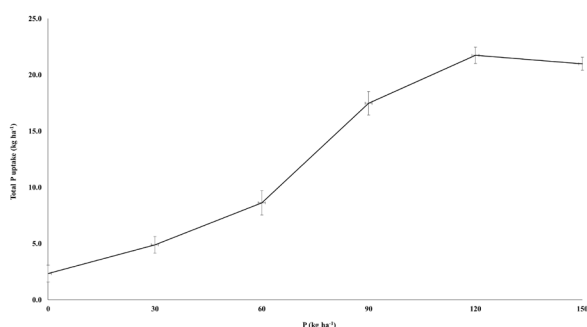

Fig. 5 - Effect of soil applied P levels on total P uptake in maize

to some extent and efficient utilization of soil applied P. The results of this study clearly demonstrated that higher PUE by plants might be the effect of better P acquisition by roots at higher applied P levels as compared to lower P availability.

Although P concentration from senescing vegetative tissues is very helpful for maintaining the growth of young leaves by fueling high P but this benefit is very small (Onasanya et al, 2009; Veneklaas et al, 2012). During leaf senescence, plants start to remobilize their vegetative P towards reproductive tissues (grains) which is another way to utilise the vegetative P efficiently and to store it in the grains. In this experiment, grain P content varied significantly with soil applied P levels and the possible reason was higher P uptake by roots which accumulated in plant tissues and further remobilized in grains at crop maturity. Moreover, the similar grain P content under control and 30 kg P ha^{-1} treatments, might be due to increased roots signaling function for more P uptake under deficient P conditions (Chiou et al, 2011; Lei et al, 2011) which was also observed in terms of similar AEP values under lower soil applied P levels. Hence, soil applied P is very important in determining plants' needs for its uptake and accumulation in different sink tissues of plant, particularly grains.

Conclusions

Phosphorus application necessitates rational management under limiting soil P conditions in maize crop. To optimize P availability, basal application of P at higher levels should be adjusted according to agronomic needs of the crop which might have substantial effects on grain yield due to better PUE. In the present study, basal application of P higher than threshold level (120 kg ha^{-1}) did not prove more economical in terms of grain yield per unit of P fertiliser. Keeping in view the importance of P-enriched seeds which were harvested during this study, further studies should be conducted for highlight their possible participatory role in the seedling establishment during next maize growing season under field conditions.

Acknowledgements

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