Assessing Organic Phosphorus Contributions for Predicting Soybean Response to Fertilization

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Core Ideas

- **The contribution of P indices that include organic P to predict soybean P fertilization response was evaluated.**
- **Bioavailable P and MA-P indices correlated with soybean relative yield.**
- **MA-P index correlated with labile organic P fractions.**
- **MA-P index improved the accuracy to predict soybean yield response to applied P.**

The use of organic P fraction in soil test P for soybean [*Glycine max* (L.) Merr.] under no-till could improve the accuracy to predict crops response to P fertilization. The research objectives were to assess the contribution and accuracy of P indices that include organic-P fractions to predict soybean yield response to P fertilization in comparison with Bray soil test P (BP). The study included P fertilization experiments conducted in the Pampas Region of Argentina during three growing seasons. We selected sites considering Cate and Nelson quadrants (I, II, III), established by a critical BP concentration of 9 mg P kg^{-1} and a relative soybean yield (RY) of 85%. Bray P, bioavailable (BioP), particulate (POM-P), and mineral-associated (MA-P) soil P fractions were determined in soil samples from the 0- to 5-cm and 0- to 20-cm depths. Bray P varied from 4.8 to 31 mg P kg^{-1} and BioP from 8 to 29 mg P kg^{-1} at the 0- to 20-cm depth. Phosphorus content in POM ranged from 19 to 171 mg P kg^{-1}, and MA-P ranged from 208 to 446 mg P kg^{-1}. Bioavailable P and MA-P correlated with RY, whereas POM-P did not. Bioavailable P performed similarly to BP test to predict soybean yield response to P fertilization. Phosphorus content in MA fraction reduced Cate–Nelson classification errors, improving the accuracy to predict soybean yield response to applied P in comparison with BP test. Sampling at the 0- to 5-cm depth did not improve soil test P performance compared with the 0- to 20-cm depth.

Abbreviations: BP, Bray phosphorus; BioP, bioavailable phosphorus; IP, inorganic phosphorus; MA-P, phosphorus in the mineral-associated fraction; MicP, microbial phosphorus; NT, no-till; OP, organic phosphorus; POM-P, phosphorus in the particulate organic matter fraction; RY, relative soybean yield; SOM, soil organic matter; ToP, total extractable phosphorus.

Phosphorus is an essential nutrient for plant growth (Raghothama, 2005) and is commonly deficient in soils of extensive agriculture areas (Batjes, 1997). Accordingly, the development of different diagnostic tools to improve crop and pasture nutrient management has been promoted. Soil total-P concentration exceeds plant requirements, but available P forms for plant uptake commonly are in low concentrations because soil total-P is comprised mainly of insoluble forms (Beegle, 2005).

Soil P concentrations commonly show stratification due to soil P extraction by crops from deeper horizons (Selles et al., 1995), P residue deposition and recycling in the soil surface, and broadcast-applied P fertilizers. Topsoil total-P is accumulated mainly in organic forms that could be mineralized by soil microorganisms, which increases P availability for crops (Damon et al., 2014; Selles, 2003; Selles et al., 1997; Tracy et al., 1990). Under no-till (NT) systems, organic P contribution for crops may be higher in comparison to tilled systems in the long term because soil organic matter (SOM) stratification in surface layers increases organic P stock, soil microbial activity, and nutrient turnover (Hedley et al., 1982; Selles, 2003; Stewart and Sharpley, 1987).

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Organic-P mineralization has been indicated as more relevant in soils with low available P levels (Sharpley, 1985; Stewart and Sharpley, 1987; Thien and Myers, 1992) and could reduce soil test P accuracy to predict crop response to P fertilization (Salas et al., 2003). This could be associated with lack of grain yield response to P fertilization in sites with low Bray P levels, a frequent error of soil test P reported by Heckman et al. (2006).

Bray soil test P (BP) is the most commonly used and calibrated soil test in the Pampas Region of Argentina (Sainz Rozas et al., 2012). Research performed in this region showed some cases with small or no soybean yield response to P fertilization in sites with BP below critical levels calibrated for the region (i.e., 9–12 mg P kg^{-1}) (Gutiérrez Boem et al., 2010; Melchiori et al., 2008). This lack of response could be due to P supply from organic fractions (Ciampitti et al., 2011b; Suñer et al., 2002; Wyngaard et al., 2013). Mallarino (2003) and Heckman et al. (2006) reported higher P critical levels for corn (*Zea mays* L.) when they measured an organic-P fraction by ICP method, improving soil test P accuracy of corn yield response.

Different methods to quantify organic-P fractions and determine bioavailability indices have been reported (Ciampitti et al., 2011b; Hedley et al., 1982; Thien and Myers, 1992). Particularly, Thien and Myers (1992) proposed a methodology to measure bioavailable P (BioP) fractions and calculate an index mainly based on microbial and labile organic-P supply. This BioP fraction was suggested as an index to improve fertilization management, but it has not been evaluated as soil test P. Others indexes, such as total-P and organic-P content in the soil particulate organic matter (POM) fraction, have showed sensitivity to changes in management practices (i.e., P fertilization and tillage system) (Ciampitti et al., 2011b; Wyngaard et al., 2013, 2016). Both POM and mineral-associated fraction (MA) are comprised of inorganic and organic P fractions (Wyngaard et al., 2013, 2016). Phosphorus content in POM (POM-P) was related to P uptake in corn and rice (*Oryza sativa* L.) (Ciampitti et al., 2011b; Wei et al., 2016) and has been indicated as a possible predictor of P nutritional status of crops (Ciampitti et al., 2011b) and an estimator of potential P mineralization (Wyngaard et al., 2013). Therefore, soil test P methods that consider some organic P fraction could improve the accuracy of recommendations of current soil test P, such as BP.

Soybean is the most extensively cultivated and economically important crop in Latin America Southern Cone and particularly in the Pampas Region of Argentina (Ministerio de Agroindustria, 2016). Soybean has higher P requirements than others crops (Garcia and Correndo, 2013). Hence, the improvement of soil test P for soybean crops may contribute to develop better P fertilization practices. We hypothesized that the prediction of soybean yield response to P fertilization may be improved by taking into account P supply from organic fractions. The research objectives were to assess the contribution and accuracy of P indices that include organic-P fractions to predict soybean yield response to P fertilization in comparison with Bray soil test P in the Pampas Region of Argentina under NT systems.

MATERIALS AND METHODS

Site Characteristics

The study was performed using an on-farm network of P fertilization experiments under soybean crops at the east of the Pampas Region of Argentina during the 2003–2004, 2004–2005, and 2005–2006 cropping seasons. The climate of the region is temperate, and the rainfall regime is humid. All sites were cropped under NT for at least 5 yr before each experiment was setup with a continuous crop sequence that included four crops in 3 yr as corn–soybean–double cropped wheat (*Triticum aestivum* L.)/soybean under rainfed conditions. The experiments were located on Hapluderts, Argiudolls, and Hapludolls (Table 1). Treatments included four P fertilization rates (0 [no-P control], 10, 20, and 30 kg P ha^{-1}) and were laid out in a randomized complete block design with three replications for Sites 4, 6, 8, and 12 and four replications for the others. Fertilizers were hand broadcasted before planting as calcium triple superphos-

Table 1. Site characteristics and location, soil taxonomic subgroup, and averages of soybean yield (SY) of no-P control, soybean relative yield (RY), and soil properties (0- to 20-cm depth): soil organic matter content (SOM), pH, and texture (clay, silt, and sand), and Bray P (BP), classified by Quadrants I, II, and III of Cate and Nelson graphic procedure.

Quadrant	Site	Experimental location	Taxonomic subgroup†	Year	SY	RY	BP	SOM	pH	Clay	Silt	Sand
					kg ha^{-1}		mg kg^{-1}	g kg^{-1}			%	
I	1	Don Cristóbal	Argd	2003	1613	0.69	4.8	45	6.7	39.9	57.5	2.6
	2	Videla	Argd	2004	3094	0.75	7.8	37	5.9	21.5	73.1	5.3
	3	Oro Verde	Hapt	2004	3002	0.66	4.9	32	7.1	41.2	48.1	10.8
	4	Larroque	Hapt	2005	1440	0.77	4.5	51	7.5	37.8	58.0	4.3
II	5	María Grande	Hapt	2004	2996	0.91	6.8	26	7.6	42.9	55.4	1.8
	6	Junín	Hapd	2004	3459	0.90	6.5	45	6.6	18.2	28.3	53.4
	7	Conesa	Argd	2004	3434	0.96	4.8	40	6.4	17.1	73.1	9.8
	8	María Grande	Argd	2003	2640	0.89	7.4	42	6.9	38.4	58.8	2.7
III	9	Alcorta	Argd	2003	4091	0.95	11.1	33	6.0	22.7	58.5	18.8
	10	Pergamino	Argd	2004	3701	0.96	15.2	36	6.2	23.2	58.5	18.3
	11	Pergamino	Argd	2005	5291	0.96	12.3	41	6.2	21.6	53.6	24.8
	12	Oro Verde	Argd	2004	4949	0.99	33.8	57	6.4	28.0	66.1	5.9

† Argd, Argiudolls; Hapd, Hapludolls; Hapt, Hapluderts.

phate (200 g P kg^{-1}). Other crop management practices (e.g., cultivar selection; seeding dates and rates; and weed, insect, and disease control methods) were those commonly used by farmers in the region, as described by Ciampitti et al. (2011a).

Composite soil samples (15–20 cores) were collected on each experiment and replication before P fertilization from the 0- to 5-cm and 5- to 20-cm depths, air dried, ground, and passed through a sieve with 2-mm mesh openings. For each sample, we determined SOM content (Walkley and Black, 1934), pH (Van Lierop, 1990), Bray P (Bray and Kurtz, 1945), and soil texture (Gee and Bauder, 1986). Soybean grain yield was determined using a plot combine to harvest a 10-m^2 area in each plot, and yield was corrected to 135 g kg^{-1} moisture. Relative soybean yield (RY) was calculated as the ratio between the grain yield of the no-P control plot and the grain yield of maximum P rate treatment (Dahnke and Olson, 1990) for each replication of the experiments.

This work was performed by selecting sites of the on-farm network according to their P availability concentrations and relative yields and their relative location with respect to the quadrants of the graphical Cate–Nelson classification method (Cate and Nelson, 1965). We selected four sites from each quadrant considering a critical BP concentration of 9 mg kg^{-1} and a soybean relative yield of 85% determined by Melchiori et al. (2008). We considered the relative position of each site in quadrants of the graphical Cate–Nelson classification method according to Dahnke and Olson (1990) as follows: Quadrant I: bottom left, BP <9 mg kg^{-1} and RY <85%; Quadrant II: top left, BP <9 mg kg^{-1} and RY >85%; and Quadrant III: top right, BP >9 mg kg^{-1} and RY >85% (Table 1; Fig. 1). We calculated two types of classification errors: (i) data indicating P fertilization need when it was not necessary (misclassification errors in Quadrant II) and (ii) data indicating no need of P fertilization when it was needed (misclassification errors in Quadrant IV). We determined classification errors as the percentage of cases in Quadrants II and IV related to total number of data.

Bioavailable P Index

The BioP index was determined as suggested by Thien and Myers (1992) using sets of soil samples stimulated with carbon and nitrogen, incubated, and extracted. Bioavailable P index is comprised of inorganic P (IP), organic P (OP), and microbial P (MicP) fractions. Each BioP fraction was determined in triplicate at the 0- to 5-cm and 5- to 20-cm soil sampling depths. Values at the 0- to 20-cm depth were calculated by weighting the 0- to 5-cm and 5- to 20-cm P values:

$$\mathrm{Px}\left(0\text{–}20\ \mathrm{cm}\right)=\frac{\left(0\text{–}5\ \mathrm{cm}\times 5\right)+\left(5\text{–}20\ \mathrm{cm}\times 15\right)}{20}$$

$$\mathrm{Px}\left(0\text{–}20\ \mathrm{cm}\right)=\text{different P fractions}$$

Briefly, 4 g soil samples were incubated with 2 mL of 0.44 mol L^{-1} glucose and 0.24 mol L^{-1} NH_4Cl solution (C/N, 10:1) at 35°C for 7 d. After incubation, soil samples were grouped in two sets: Set A (unfumigated samples) and Set B (fumigated samples). In Set A, inorganic extractable P (IP) was determined by modified Watanabe and Olsen (1965) procedure, and total extractable P (ToP) content in the $NaHCO_3$ extract was determined by acid digestion

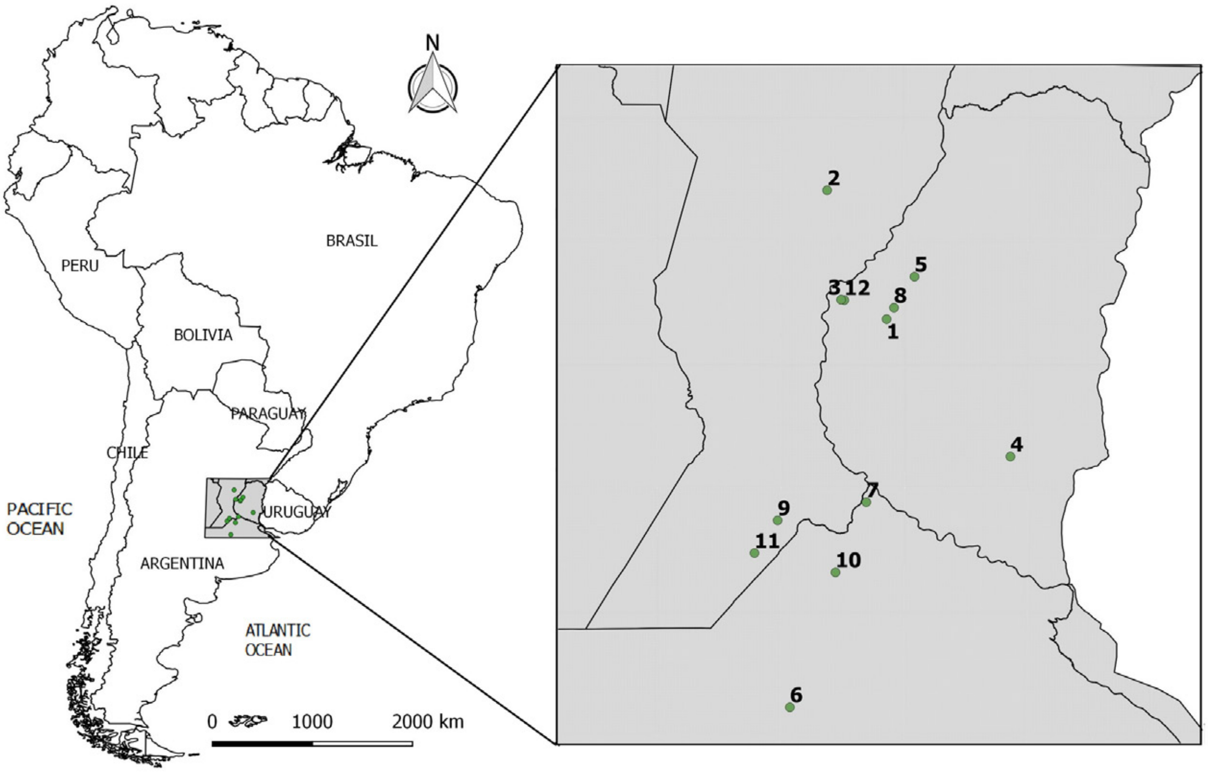

Fig. 1. Geographical location of selected sites from the trial network conducted in 2003 to 2004, 2004 to 2005, and 2005 to 2006 at the Pampas region of Argentina.

with ammonium persulfate (Bowman, 1989). The soil/solution ($0.5\ \text{mol L}^{-1}\ \text{NaHCO}_3$) ratio used was 1:10.

In Set B, after incubation, soil samples were fumigated with 2 mL of hexanol for 36 h at room temperature in capped containers and then evaporated for 24 h. For each sample, IP was extracted with $0.5\ \text{mol L}^{-1}\ \text{NaHCO}_3$. The extracted IP after fumigation represents initial inorganic P plus microbial P (IP+m) released after microbial cell lysis with hexanol. Microbial P was calculated as the difference between IP+m (determined in Set B) and IP (determined in Set A) using a recovery factor of 0.4 according to McLaughlin et al. (1986).

For the two soil sample sets, P concentration was measured colorimetrically by the method of Murphy and Riley (1962), and different bioavailable P fractions were calculated as:

$$\text{OP} = \text{ToP} - \text{IP}$$

$$\text{MicP} = \frac{(\text{IP+m}) - \text{IP}}{0.4}$$

$$\text{BioP} = \text{OP} + \text{MicP} + \text{IP}$$

P Content in Soil Fractions

Soil samples were fractioned using the method proposed by Cambardella and Elliott (1992), modified using glass balls as a dispersing agent (Irizar et al., 2010). Briefly, 10-g soil samples were shaken with 30 mL of distilled water and two glass balls for 18 h. Then, the soil fraction that passed through the 53-mm sieve (soil mineral-associated fraction [MA]) was dried at 105°C for 48 h, homogenized, and ground by hand with a mortar and pestle. The amount of POM-P was determined as the difference between the value of total P (TP) content and the one obtained in MA (MA-P) corrected by sand content (Cambardella and Elliott, 1992).

Total-P content of soil was determined by $\text{HNO}_3\text{–HClO}_4$ digestion and colorimetric analysis (Kuo, 1990).

Statistical Analysis

Statistical analyses were performed with Infostat software (Di Rienzo et al., 2011). Descriptive statistics and correlations for the complete data set and for each quadrant of the Cate–Nelson method were calculated. The averages for each quadrant of BP, BioP fractions, TP, MA-P, and POM-P were compared by *t* test.

The critical concentration for each P fraction and soil sampling depth were determined by the Cate–Nelson (Cate and Nelson, 1971) statistical procedure using RY as the dependent variable. Critical concentrations were estimated with the Cate–Nelson graphic procedure when the statistical procedure failed. We calculated classification errors in Quadrants II and IV using all the replications of each experiment ($n = 44$) as the total number of points in those quadrants with respect to all points in the graphic.

RESULTS

Site Characteristics and Soybean Yield

Bray soil test P contents varied around the critical concentration (9 mg P kg^{-1}) from 4.8 to 31.1 mg kg^{-1} of P. Soil pH ranged from 5.9 to 7.6, and SOM content varied from 26 to 57 g SOM kg^{-1} (Table 1). Bray soil test P content averaged 5.5 mg P kg^{-1} in Quadrant I and 6.4 mg P kg^{-1} in Quadrant II; these values were statistically lower ($P < 0.05$) than the average BP content of 18.1 mg P kg^{-1} in Quadrant III.

Average soybean yields ranged between 1495 and 5291 kg ha^{-1}, and were 2287 and 3132 kg ha^{-1} in Quadrants I and II, respectively; both values were significantly lower ($P < 0.01$) than the average soybean yield in Quadrant III, which was 4508 kg ha^{-1}. Phosphorus fertilizer response, calculated as RY, ranged from 0.69 to 0.96 (Table 1), and the average relative yield of Quadrant I was significantly lower ($P < 0.01$) than the average of Quadrants II and III.

Bioavailable P Content

Bioavailable P concentrations ranged from 8.3 to 38.8 mg P kg^{-1} at the 0- to 5-cm depth, from 4.2 to 26.1 mg P kg^{-1} at the 5- to 20-cm depth, and from 8.1 to 29.1 mg P kg^{-1} at the 0- to 20-cm depth (Fig. 2). Bioavailable P fractions in all cases showed a higher average P content and a wider range of variation at the 0- to 5-cm depth compared with the 5- to 20-cm depth (Fig. 2). Average IP, OP, and BioP contents were similar in Quadrants I and II ($P > 0.05$) and significantly lower ($P < 0.05$) than those in Quadrant III at the 0- to 5-cm and 5- to 20-cm soil sampling depths.

Soil test IP and OP (0–20 cm) showed similar concentrations in Quadrants I and II ($P > 0.05$) and significantly lower concentrations ($P < 0.05$) than those in Quadrant III. However, MicP fractions were significantly higher ($P < 0.05$) in Quadrants II and III compared with Quadrant I.

At the 0- to 20-cm soil sampling depth, BioP contents were <13 mg P kg^{-1} for all sites placed in Quadrant I, ranged from 15 to 20 mg P kg^{-1} in Quadrant II, and were >20 mg P kg^{-1} in Quadrant III (Fig. 2). The average BioP content was 11.3 mg P kg^{-1} in Quadrant I, which was significantly ($P < 0.05$) lower than the average in Quadrants II and III. Bioavailable P content in Quadrant II was lower compared with Quadrant III (17.3 and 24.9 mg P kg^{-1}, respectively; $P < 0.05$). Bioavailable P fraction content increased from Quadrant I to III, and organic P was the largest fraction, representing around 50% of the BioP content (Fig. 2).

P Content in Soil Fractions

Soil total P contents ranged from 281 to 597 mg P kg^{-1} at the 0- to 5-cm soil sampling depth, from 245 to 540 mg P kg^{-1} at the 5- to 20-cm depth, and from 257 to 547 mg P kg^{-1} at the 0- to 20-cm depth. Phosphorus contents in the MA ranged from 232 to 503 mg P kg^{-1} at the 0- to 5-cm depth, from 199 to 434 mg P kg^{-1} at the 5- to 20-cm depth, and from 208 to 446 mg P kg^{-1} at the 0- to 20-cm depth. Phosphorus content in the POM ranged from 30 to 183 mg P kg^{-1} at the 0- to 5-cm depth, from 15.7 to 167 mg P kg^{-1} at the 5- to 20-cm depth, and from 19.3 to 171 mg P kg^{-1} at the 0- to 20-cm depth (Fig. 3). Total P content at the three different depths were similar in sites of Quadrants II and III ($P < 0.05$) and significantly higher than those in Quadrant I. Phosphorus content in MA was significantly different in sites of Quadrants I, II, and III ($P < 0.05$), and POM-P content was not different among quadrants (Fig. 3).

Relationships among Soil Proprieties, P Fractions, Soybean Yield, and Relative Yield

Correlations determined at the 0- to 20-cm soil sampling depth showed that P indices did not correlate with SOM content and pH (Table 2), except ToP, which showed a negative correlation with pH ($r = -0.59$). Bray P significantly correlated only with IP ($r = 0.82$) and MA-P ($r = 0.66$). At the same soil sampling depth, BioP showed a highly significant correlation with IP, ToP, OP, MA-P, and TP but showed no correlation with POM-P (Table 2). Phosphorus content in the POM was not correlated with evaluated P fractions, but MA-P was highly correlated with all BioP fractions. Phosphorus content in the POM showed a positive correlation ($r = 0.74$) with sand content of soils and a negative correlation with silt content ($r = -0.85$) (Table 2). Correlations between POM-P and sand and silt content were similar at the 0- to 5-cm depth (Table 3).

Relationships determined at the 0- to 5-cm depth showed similar correlation coefficients among P indices and SOM compared with the 0- to 20-cm depth. We noted a negative correlation of OP and BioP with pH (Table 3). Bray P was highly correlated with IP, ToP, BioP, TP, and MA-P. Bioavailable P and POM-P showed similar correlations among P indices measured at both depths (0–5 and 0–20 cm). Phosphorus content in MA was highly correlated with all BioP fractions except MicP.

Bioavailable P and BP indices were similarly correlated with soybean yield. However, RY showed higher correlation with BioP ($r = 0.81$) than BP ($r = 0.54$) (Table 2). At the 0- to 5-cm depth, BP showed higher correlation than BioP with soybean yield (Table 3) and similar correlation with RY. Phosphorus in

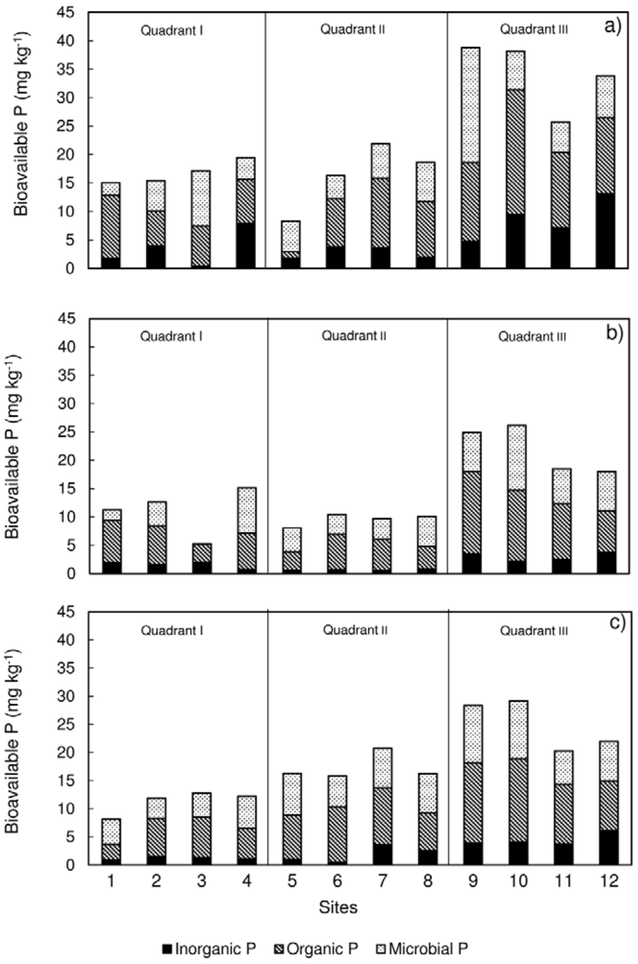

Fig. 2. Bioavailable P fractions. Inorganic P (IP), organic P (OP), and microbial P (MicP) in each site by quadrant of the graphical Cate–Nelson method at (a) 0- to 5-cm soil sampling depth, (b) 5- to 20-cm soil sampling depth, and (c) 0- to 20-cm soil sampling depth. Sites classified in Quadrants I, II, and III considering their relative position of the Cate–Nelson graphical procedure.

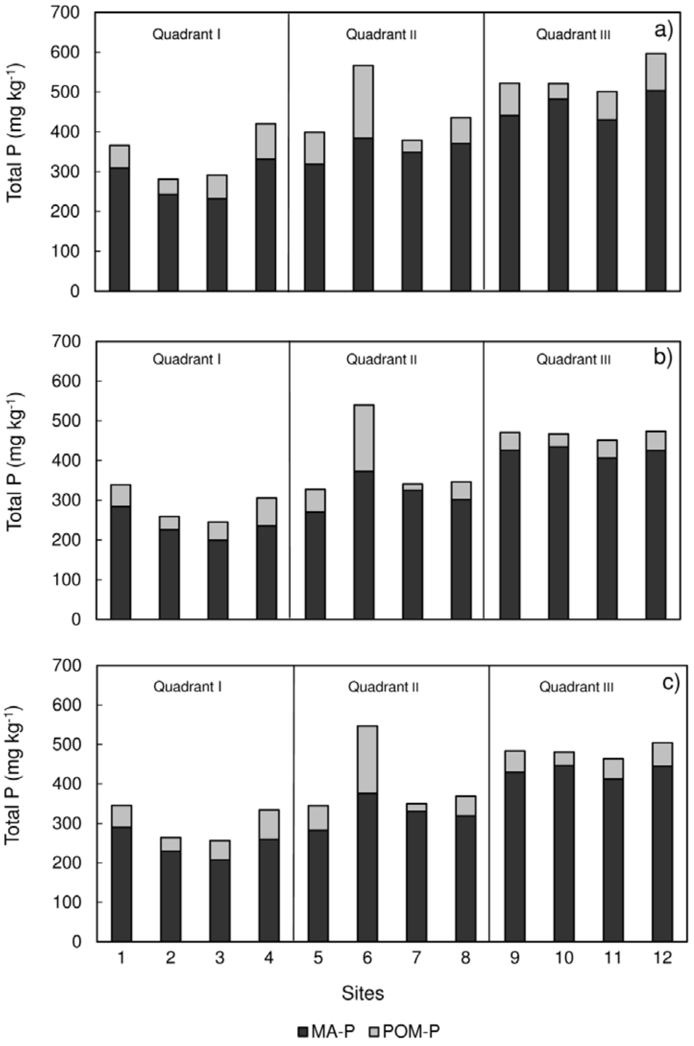

Fig. 3. Total P contents in the particulate organic matter fraction (POM-P) and in the mineral-associated fraction (MA-P) in each site by quadrant of the graphical Cate–Nelson method at (a) the 0- to 5-cm soil sampling depth, (b) the 5- to 20-cm soil sampling depth, and (c) the 0- to 20-cm soil sampling depth. Sites classified in Quadrants I, II, and III considering their relative position of the Cate–Nelson graphical procedure.

Table 2. Simple correlations (*r* Pearson coefficient) at the 0- to 20-cm depth between soybean yield (SY) and relative soybean yield (RY), Bray P (BP), soil organic matter content (SOM), and fractions: inorganic P (IP), total extractable P (ToP), organic P (OP), microbial P (MicP), bioavailable P (BioP), total-P (TP), P content in mineral-associated fraction (MA-P), and P content in particulate organic matter fraction (POM-P).

	SY	RY	BP	SOM	pH	IP	ToP	OP	MicP	BioP	Clay	Silt	Sand	TP	MA-P	POM-P
SY	1	**	*	ns†	*	**	**	*	ns	**	*	ns	ns	*	**	ns
RY	0.71	1	ns	ns	ns	**	**	**	**	**	ns	ns	ns	**	**	ns
BP	0.67	0.54	1	ns	ns	**	ns	ns	ns	ns	ns	ns	ns	ns	*	ns
SOM	0.01	0.08	0.47	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
pH	-0.59	-0.34	-0.35	-0.06	1	ns	*	ns	ns	ns	**	ns	ns	ns	ns	ns
IP	0.73	0.70	0.82	0.29	-0.53	1	**	ns	*	**	ns	ns	ns	ns	**	ns
ToP	0.77	0.80	0.54	-0.11	-0.59	0.79	1	**	**	**	*	ns	ns	*	**	ns
OP	0.67	0.73	0.32	-0.29	-0.53	0.56	0.95	1	**	**	*	ns	ns	*	**	ns
MicP	0.36	0.73	0.34	-0.22	-0.21	0.57	0.81	0.80	1	**	ns	ns	ns	ns	**	ns
BioP	0.68	0.81	0.50	-0.15	-0.49	0.75	0.98	0.94	0.90	1	ns	ns	ns	*	**	ns
Clay	-0.60	-0.55	-0.22	-0.12	0.81	-0.41	-0.63	-0.64	-0.23	-0.53	1	ns	*	ns	ns	ns
Silt	0.02	0.09	0.22	0.05	-0.29	0.47	0.13	-0.06	0.10	0.12	-0.08	1	**	ns	ns	**
Sand	0.38	0.30	-0.03	0.04	-0.30	-0.10	0.32	0.48	0.07	0.26	-0.60	-0.75	1	*	ns	**
TP	0.61	0.74	0.55	0.33	-0.38	0.48	0.65	0.63	0.56	0.65	-0.53	-0.4	0.67	1	**	ns
MA-P	0.71	0.84	0.66	0.25	-0.53	0.74	0.82	0.74	0.72	0.83	-0.56	-0.07	0.43	0.92	1	ns
POM-P	-0.05	0.01	-0.08	0.26	0.22	-0.43	-0.2	-0.05	-0.19	-0.2	-0.09	-0.85	0.74	0.49	0.11	1

*Significant at the 0.05 probability level.

**Significant at the 0.01 probability level.

† Not significant.

MA showed higher correlation than BP and BioP with soybean yield and RY at both depths considered.

Relationships between Relative Soybean Yield and P Indices

Relationships between BP and BioP indices and soybean RY allowed us to determine a critical soil test concentration of 8.5 mg kg^{-1} for BP and 18.5 mg kg^{-1} for BioP by the Cate–Nelson procedure at the 0- to 20-cm depth (Table 4; Fig. 4). When we considered points that were out of Quadrants I and III of the Cate–Nelson graphic taking into account both BP and BioP critical concentrations, we calculated similar total classification errors of 18% (Fig. 4). We also studied the relationship between BP+BioP, BP+OP, BP+MicP, and BP+OP+MicP indices with RY as complementary indices, but they showed higher classification errors than BP alone (Table 4).

We calculated a critical concentration of 50 mg P kg^{-1} at the 0- to 20-cm depth using POM-P as independent variable by Cate–Nelson (Fig. 5a) and 62 mg P kg^{-1} for BP+POM-P content. Classification errors were 39 and 50%, respectively (Fig.

Table 3. Simple correlations (*r* Pearson coefficient) at the 0- to 5-cm depth between soybean yield (SY) and relative soybean yield (RY), Bray P (BP), soil organic matter content (SOM), and fractions: inorganic P (IP), total extractable P (ToP), organic P (OP), microbial P (MicP), bioavailable P (BioP), total-P (TP), P content in mineral-associated fraction (MA-P), and P content in particulate organic matter fraction (POM-P).

	SY	RY	BP	SOM	pH	IP	ToP	OP	MicP	BioP	Clay	Silt	Sand	TP	MA-P	POM-P
SY	1	**	*	ns†	*	ns	ns	ns	ns	*	ns	ns	ns	*	*	ns
RY	0.71	1	ns	ns	ns	ns	ns	ns	ns	*	ns	ns	ns	**	**	ns
BP	0.67	0.54	1	ns	ns	**	*	ns	ns	*	ns	ns	ns	*	**	ns
SOM	0.01	0.08	0.47	1	ns	*	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
pH	-0.59	-0.34	-0.35	-0.06	1	ns	ns	*	ns	*	**	ns	ns	ns	ns	ns
IP	0.50	0.54	0.82	0.60	-0.32	1	**	*	ns	**	ns	ns	ns	*	**	ns
ToP	0.50	0.55	0.67	0.44	-0.55	0.85	1	**	ns	**	ns	ns	ns	*	**	ns
OP	0.41	0.46	0.44	0.24	-0.61	0.58	0.92	1	ns	**	ns	ns	ns	ns	**	ns
MicP	0.37	0.26	0.17	-0.36	-0.35	0.01	0.16	0.24	1	*	ns	ns	ns	ns	ns	ns
BioP	0.58	0.57	0.63	0.19	-0.60	0.69	0.88	0.86	0.60	1	ns	ns	ns	*	**	ns
Clay	-0.53	-0.47	-0.17	-0.19	0.84	-0.31	-0.49	-0.53	-0.08	-0.44	1	ns	ns	ns	ns	ns
Silt	0.03	0.10	0.23	0.05	-0.29	0.24	0.20	0.14	0.08	0.20	-0.09	1	**	ns	ns	**
Sand	0.32	0.22	-0.08	0.09	-0.30	0.00	0.15	0.23	-0.02	0.11	-0.57	-0.77	1	ns	ns	**
TP	0.57	0.76	0.64	0.43	-0.25	0.67	0.66	0.53	0.20	0.63	-0.40	-0.33	0.53	1	**	*
MA-P	0.64	0.84	0.73	0.36	-0.39	0.76	0.83	0.73	0.27	0.80	-0.42	-0.02	0.30	0.93	1	ns
POM-P	0.09	0.15	0.08	0.31	0.20	0.08	-0.11	-0.23	-0.06	-0.12	-0.11	-0.80	0.71	0.57	0.22	1

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

† Not significant.

5b). Considering the high correlation between MA-P and RY (Table 2), we calculated a critical concentration of 293 mg kg^{-1} for MA-P and classification errors of 7% using Cate–Nelson (Table 4; Fig. 6).

We also studied the relationship between each bioavailable P fraction (IP, OP, MicP), TP soil test methods, and RY (not shown), but in all cases we obtained higher classification errors compared with BP, BioP, and MA-P indices. We evaluated these relationships in the 0- to 5-cm depth (Table 4), but no major improvement in relationships or classification errors were found compared with the 0- to 20-cm depth.

DISCUSSION

This study considers a wide range of variation in BP contents that are within the typical values reported in the Pampas Region (Boschetti et al., 2003; Sainz Rozas et al., 2012). Taking into account soil availability BP contents, all the sites were classified in low (<10 mg P kg^{-1}) and medium (10–20 mg P kg^{-1}) categories, considering the classification levels established by Sainz Rozas et al. (2012) for the Pampas region, except Site 12, which had a high BP content (>20 mg P kg^{-1}).

The range of BioP index determined in our work for sites in a wide agricultural region of Argentina (i.e., 8.1–29.1 mg P kg^{-1}) was lower than values reported in other studies (Duda et al., 2013; Thien and Myers, 1992). This result were probably attributable to soils with a narrower range of soil property variations (i.e., SOM and P), with a similar crop rotation, and that were cultivated under NT system. Duda et al. (2013) reported a wider range of BioP content (i.e., 7.9–115.7 mg P kg^{-1}) because their study included grassland soils with larger SOM content. Likewise, Thien and Myers (1992) reported sites where BioP increased associated with SOM.

In our work, IP was the fraction with the smaller contribution to BioP, whereas OP was the major component, as shown

Table 4. Critical concentrations (CC) for soybean estimated at two sampling depths with different soil test P values by Cate–Nelson statistical (CNs) or graphical (CNg) models.

Sampling depth	Soil test†	Model	CC	R^2	CE§
			mg kg^{-1}		%
0–5 cm	BP	CNs	13.2	0.38	20
	BioP	CNs	20.4	0.36	23
	BP+OP	CNs	19.0	0.37	23
	BP+MicP	CNs	19.5	0.33	18
	BP+OP+MicP	CNs	28.2	0.34	18
	POM-P	CNg	66	na‡	32
	BP+POM-P	CNg	84	na	36
	MA-P	CNs	303	0.63	7
0–20 cm	BP	CNs	8.5	0.38	18
	BioP	CNs	18.5	0.33	18
	BP+OP	CNs	16.6	0.33	20
	BP+MicP	CNg	13.5	na	18
	BP+OP+MicP	CNs	21.7	0.33	20
	POM-P	CNg	50	na	39
	BP+POM-P	CNg	62	na	50
	MA-P	CNs	293	0.64	7

† BioP, bioavailable P; BP, Bray P; MA-P, P content in mineral-associated fraction; MicP, microbial P; OP, organic P; POM-P, P content in particulate organic matter fraction.

‡ Not applicable.

§ CE, classification errors of Cate–Nelson (total number of points in Quadrants II and IV with respect to all points in the data set).

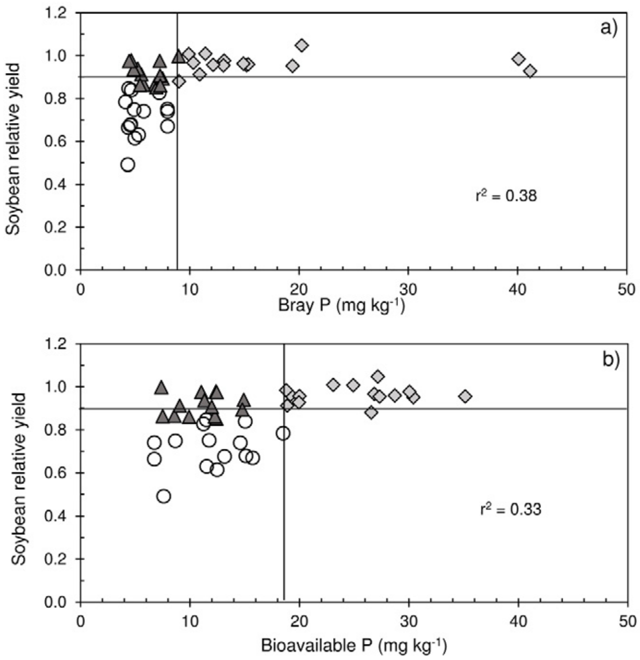

Fig. 4. Relationship between soybean relative yield and (a) Bray P and (b) bioavailable P at the 0- to 20-cm depth. Phosphorus contents in sites of Quadrant I (circle), Quadrant II (triangle), and Quadrant III (rhombus) (n = 44).

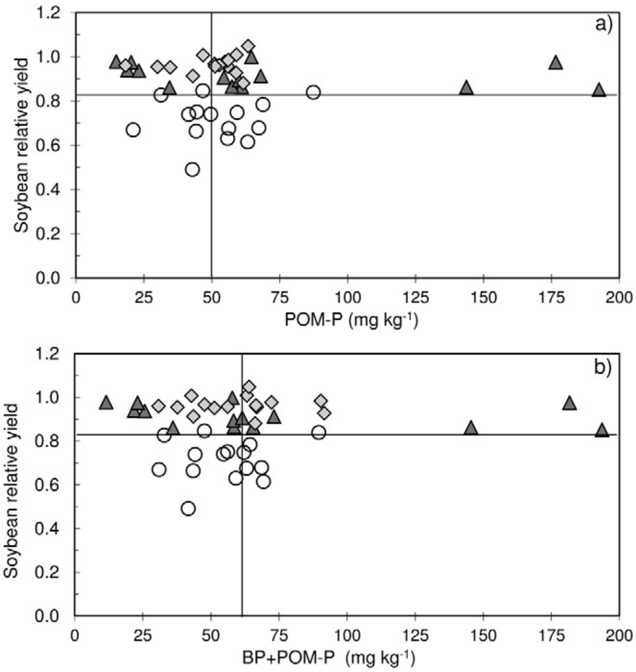

Fig. 5. Relationship between soybean relative yield and (a) P content in particulate organic matter fraction (POM-P) and (b) Bray P (BP) plus P in particulate organic matter fraction (POM-P) at the 0- to 20-cm depth. Phosphorus contents in sites of Quadrant I (circle), Quadrant II (triangle), and Quadrant III (rhombus) (n = 44).

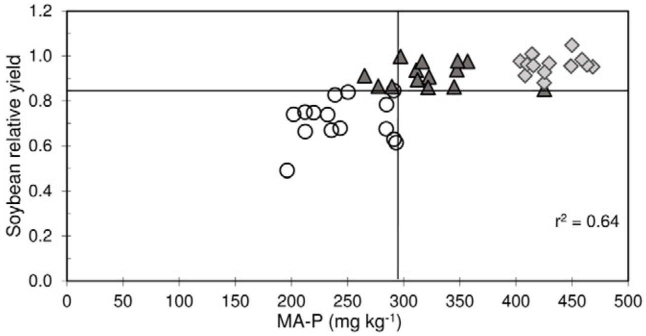

Fig. 6. Relationship between relative soybean yield and mineral-associated P index (MA-P) at the 0- to 20-cm depth. Phosphorus contents in sites of Quadrant I (circle), Quadrant II (triangle), and Quadrant III (rhombus) (*n* = 44).

by Thien and Myers (1992). However, Duda et al. (2013) found a higher contribution of MicP fraction to BioP, an effect that was more important in soils with low IP, suggesting that microorganisms could access P fractions adsorbed on inorganic soil particles (He and Zhu, 1998) and assimilate them in microbial biomass (Thien and Myers, 1992). In fact, in our work the minor contribution of the MicP fraction to BioP could be due to the lower microbial activity associated with the lower temperatures of our temperate region compared with the tropical region of the Duda et al. (2013) study. Also, they studied grasslands where microbial biomass is the main fraction involved in P recycling (Stevenson, 1986).

Higher BioP contents observed at the 0- to 5-cm soil sampling depth may be associated with organic matter stratification commonly observed in NT systems (Díaz-Zorita et al., 2002) and with the accumulation and recycling of more available organic-P fractions (Selles, 2003). This stratification was also observed for TP, MA-P, and POM-P (Ciampitti et al., 2011b). The topsoil organic-P fraction accumulation in NT systems could become an important source of available P for crops (Tiessen et al., 1994).

Although a higher topsoil P concentration could contribute to crop nutrition due to a greater P turnover (Selles et al., 1995), no P indices measured at the 0- to 5-cm depth performed better to predict soybean yield response to added P compared with the 0- to 20-cm depth (Table 4). Therefore, the use of the standard 0- to 20-cm soil sampling depth (Sainz Rozas et al., 2012) was the most suitable for soil test P evaluated.

Total P content determined in our work was in the range reported in the Pampas Region (Gutierrez Boem et al., 2008; Suñer and Galantini, 2015). Likewise, MA-P content was similar to that determined by Suñer and Galantini (2015), although they considered MA in a coarser fraction (<100 mm). Phosphorus content in POM was higher than that reported by Ciampitti et al. (2011b) and Wyngaard et al. (2013). The differences can be associated with the higher clay content of our studied soils, where organic matter could be protected (Hassink and Whitmore, 1997). However, our POM-P values were lower compared with those reported by Suñer and Galantini (2015), probably because they evaluated grassland soils located in a region with lower mean temperatures. In addition, we obtained a negative correlation between POM-P and silt content, which was in agreement with Suñer and Galantini (2015), who reported that POM-P levels decreased as silt+clay content increased.

Sites with high BP values also showed high BioP, similarly to the relationship between inorganic P fractions and BioP index reported by Thien and Myers (1992) and Duda et al. (2013). The BioP critical level was, as expected, higher than locally reported BP critical levels (Cencig et al., 2003; Gutiérrez Boem et al., 2010; Melchiori et al., 2008) because the former include sources of labile organic-P that are not measured by BP (Suñer et al., 2005; Wyngaard et al., 2013).

The inclusion of organic-P has been suggested as a way to improve soil test P accuracy, mainly in NT systems (Ciampitti et al., 2011b; Selles, 2003; Tiessen et al., 1992). In fact, BioP index could assess P availability in soils, as reported by Duda et al. (2013) and Thien and Myers (1992). To our knowledge, only one study has reported a positive correlation between BioP and P accumulation in plant tissues (Duda et al., 2013).

Our research supports the statement that BioP could be a useful index to assess soil P supply, as was suggested by Thien and Myers (1992), because it was better correlated to RY than BP (Table 2). However, the BioP index as soil test P did not improve the accuracy to predict soybean yield response to P fertilization and did not reduce classification errors compared with BP (Fig. 4). Although the BioP index in sites from Quadrant II had higher content than sites in Quadrant I, the BioP index per se, or as a complementary index to BP, was not good enough to explain why soybean response to P fertilization was very low in some sites with low P availability levels. This type of error of soil test yield response prediction is frequent, showing crops that do not respond to P fertilization even if the site test is below the critical concentration (Heckman et al., 2006). Nevertheless, Mallarino (2003) and Heckman et al. (2006) obtained better precision to predict corn yield response to P fertilization when methods to quantify P in traditional extractants included organic P fractions.

Phosphorus content in POM was not related to soybean yield or relative yield and did not improve soil test P accuracy, in contrast with the idea suggested by Ciampitti et al. (2011b) and Wyngaard et al. (2013). Phosphorus in POM is comprised of inorganic and organic forms (Wyngaard et al., 2013), and it is a potential P source for crops, which could be mineralized during the growing season (Ha et al., 2008). However, P supply from organic fractions depends on suitable conditions for P mineralization (Condron et al., 2005). Therefore, not all POM-P would be available for crops. In addition, Wyngaard et al. (2013) determined that POM has the capacity to adsorb P, and some portion of POM-P would remain in residues or immobilized by soil microbes (Ha et al., 2008).

Phosphorus in the MA fraction was related with RY and showed a better fit using the Cate–Nelson procedure, suggesting a contribution from this soil fraction to supply P for crops. We showed that some sites initially classified in Quadrant II of Cate–Nelson using BP were reclassified in Quadrant III when

MA-P was used as independent variable, revealing a differential P contribution from MA-P index. We found a high correlation between MA-P and OP, MicP, and BioP, probably because MA-P is composed of 47 to 89% organic P (Wei et al., 2016; Wyngaard et al., 2013, 2016).

The better performance of the MA-P index to determine soil test P in sites initially located on Quadrant II of Cate–Nelson compared with BP or BioP showed that these indices may not measure all crop-available P fractions. Phosphorus in the MA fraction showed higher correlation with RY and improved soil test P accuracy compared with BP, suggesting that the MA-P fraction supplies P for soybean crops. Further research is required to determine the potential contribution of organic P concentration in POM-P and MA-P fractions to predict soybean yield response to P fertilization.

CONCLUSIONS

Bioavailable P was related to soybean relative yield and reached a critical concentration of 18.5 mg P kg^{-1} at the 0- to 20-cm sampling depth. However, BioP did not improve the accuracy to predict soybean yield response to P fertilization compared with soil test BP. Our data indicate that sampling at the 0- to 20-cm depth was more consistent than at the 0- to 5-cm depth.

Phosphorus in POM was not related with soybean relative yield but was highly related with MA-P. We determined a critical concentration of 293 mg P kg^{-1} in MA-P at the 0- to 20-cm sampling depth. Phosphorus in MA performed better than BP and BioP to accurately classify sites according to their P concentration and RY, reducing Cate–Nelson classification errors, and improving P diagnosis.

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