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Impact of Long-Term Conservation Agriculture Practices on Phosphorus Dynamics under Maize-Based Cropping Systems in a Sub-Tropical Soil

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Abstract: Over the past decade, scientific studies have increasingly concentrated on the effects of global phosphorus (P) scarcity on food security. A comprehensive strategy that considers demand reduction and recycling possibilities is needed to address the global P scarcity. Reduced tillage along with crop residue retention could decrease fixation of P in soil, improve labile P content and enhance organic-P (Po) buildup and its mineralization by phosphatases; this could be an extra benefit of conservation agriculture (CA) in soils. To study the impact of long-term CA on soil organic and inorganic P fractions and their distribution, a long-term field trial was conducted under a maize-based cropping system with different tillage (zero tillage (ZT), permanent bed (PB) and conventional till (CT) and cropping system (maize–wheat–mungbean (MWMb), maize–chickpea–sesbania (MCS), maize–mustard–mungbean (MMuMb) and maize–maize–sesbania (MMS)). Phosphorus dynamics were studied through sequential fractionation (organic and inorganic P) at 0–5 and 5–15 cm soil depth. The findings showed that a higher amount of soluble and loosely bound P (SL-P) was detected in ZT among the inorganic P fractions, whereas iron-bound P (Fe-P), aluminum-bound P (Al-P), reductant soluble P (RES-P) and calcium-bound P (Ca-P) were found higher in CT in both soil depths. Among Organic-P fractions, moderately labile and non-labile Po was found higher in PB and ZT but, in the case of labile Po, it was found insignificant with respect to tillage operations. Significant synergistic effects of winter legume (chickpea) with summer legumes (sesbania and mungbean) in crop rotation were observed on SL-P, Labile Po, Humic acid-Po, Alkaline phosphatase and MBP at 0–5 and 5–15 cm soil depths. Given the potential relevance of understanding P dynamics for efficient P management in long-term conservation agriculture practices, our findings offers critical new insight for the P management for sustainable development.

Keywords: conservation agriculture; crop residue; organic P; phosphorus fraction; zero tillage

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1. Introduction

One of the biggest difficulties in the twenty-first century is the sustainable production of additional food (50% more food) from scarce and limited land and water resources to feed the predicted 9.1 billion people on the planet by 2050 [1]. Concerns about the sustainability of agriculture around the world led to the development of the concept of conservation agriculture (CA), which has progressively increased to encompass 12.5% of the world's agriculture land (180 M ha) [2]. Conservation agriculture is an approach to growing different crops, based on no-tillage practices with crop residue recycling, crop rotation and cover crops, in order to offer an enduring soil cover and a naturally rising soil organic matter content in surface horizons [3]. The CA-based different tillage and initial crop practices, such as permanent raised beds (PB) and zero tillage (ZT), hold potential to improve nutrient cycling in soil along with sustainability in soil health. The use of CA has been shown to provide benefits, including soil structure improvement, water retention of soil and higher yield benefit from crops [4,5]. The North-Western Indo-Gangetic Plains of India have historically practiced the rice–wheat cropping system, which is to account for the loss of soil carbon and deterioration in soil health. This problem can be reduced by substituting out rice for an aerobic crop, such as maize, with legumes in the crop rotations [6].

Soil organic matter (SOM) is a key determinant of soil quality by improving soil structure, soil fertility, productivity and sustainability under a dry-land farming system [7]. Agricultural management techniques, such as tillage, mulching, management of crop residue and the use of organic and mineral fertilizers, have an impact on the dynamics of SOM. In order to control the release and storage of nutrients from SOM, tillage plays an essential role. Rapid mineralization of SOM during CT resulted in the loss of nitrogen (N) and carbon (C) from soil. Continuous long-term experiments under CA provide evidence of enhanced yields, farm profits and soil properties of the maize–chickpea rotation and, thereby, could sustain production in the long run [8]. Dominating species and their indexes provide the necessary information about various nutritional deficits; hence, understanding different cropping systems is crucial for the sustainable management of the crop [9]. The type of chemical reaction in phosphate ions occurring after fertilizer application and the intensity of P accumulation under different fractions are mostly determined by the texture and mineralogy of the soil [10]. Among the management techniques, the soil tillage practices [11], the fertilizer's physical and chemical composition [12], the extent of soil–fertilizer contact [13], the rate of application [14] and plant biomass's contribution, are the major prominent factors. Together, these factors may have an impact on the soil residual P properties.

According to Dhillon et al. [15], in total, 5.7 billion hectares of agricultural land are P deficient globally. Actually, in practice, only a small portion of applied P fertilizers are taken up by plants. On the contrary, the majority is fixed in soil in different less-available forms [16]. Usually, the amount of P in the additional residue is crucial for controlling how quickly P is immobilized or mineralized in soil [17]. No till (NT) has reduced soil disturbance, which increases the buildup of nutrients in the upper layer, particularly those with low mobility, such as P applied through fertilizer and crop residues. By changing the diversity in the soil's microbial population and its enzyme activity, which, in turn, impacts the soil's P availability, CA-based techniques have the potential to increase P availability [18]. Through microbial activities, these techniques can control the buildup and depletion of SOM, C-sequestration and soil aggregation, which contribute to better crop growth [19]. P fixation can be decreased by organic matter inputs under residue retention due to more organic anion competition for P-binding sites [20]. Improvement in the available P concentration without increasing P leaching and fixation improves sustainable production, with a higher crop yield.

Retention of OM and reduced tillage can improve the structure or aggregation of weathered soils (MWD) [21] and possibly contribute to better availability of P. In these soils, enhanced aggregation can decrease the quantity of soluble P inorganic (Pi) that is exposed to potential sorption sites by reducing the total soil surface area [22]. Excessive fertilization following ineffective farming techniques, as shown in monocultures with CT [23] and in the crop rotation system without a cover crop [24], may encourage even greater residual P buildup, because these manufacturing systems have very poor P-usage efficiency.

According to Nunes et al. [25], soil tillage practices and P-fertilization management have an impact on the residue P that collected in the following 17 years of fertilizer use and grain crop cultivation. In comparison to natural soil, Pi and Po accumulated under tillage techniques. Conventional tillage reduced the effects of P-fertilizer management by distributing Pi fractions more uniformly across the profile. The available P was modified by the residue regime systems, but not the residual or total P. In the top 5 cm layer, complete CA considerably raised total organic P by 6.3%. Haokip et al. [26], given the aforementioned information, suggested that a thorough understanding of soil P dynamics is necessary in a maize-based cropping system that received fertilizer P over an extensive period under conservation agriculture.

In the context of all of the above, the present study examines how long-term practices of different tillage and cropping systems affected soil organic and inorganic P fractions and their distribution. Understanding how different factors are directly or indirectly related to the P availability, as well as depth distribution in the soil, may be strongly impacted by various reactions of P in the soil.

2. Materials and Methods

2.1. Experimental Site

The long-term field experiment started in the research field of the Indian Institute of Maize Research, Pusa Campus, New Delhi, India (28°40′ N, 77°12′ E and 229 MSLelevation) during the wet (*Kharif*) season of 2008. The climate in this area is semi-arid, with 650 mm of annual rainfall (average during the past 30 years) with annual mean evaporation of 850 mm. During the experimentation years (2008–2020), the mean daily minimum temperature in January was 0–4 °C, the average maximum temperature was 40–46 °C, and the average relative humidity ranged from 67 to 83%. The soil in research field is classified as Typic Haplustept [27], sandy loam in texture, with basic alkaline pH (7.8) and non-saline in nature (EC value of 0.32 dS m^{-1}). Table 1 shows the initial physico-chemical and biological characteristics in the soil.

Table 1. Initial soil properties of the experimental site.

Soil Properties	Value
Sand (%)	64.3
Silt (%)	13.8
Clay (%)	22.0
pH (1:2 soil: water)	7.80
Bulk density (Mg m^{-3})	1.65
Soil organic carbon (g kg^{-1})	4.31
Available N (kg ha^{-1})	158
Available P (kg ha^{-1})	11.6
Available K (kg ha^{-1})	248
Microbial biomass carbon (mg C g^{-1} soil)	340
Alkaline phosphatase (mg p-NP Rel g^{-1} 24 h^{-1})	34.0

2.2. Experimental Details

The research study was set up in split-plot design with different tillage practices in main plot (permanent raised bed (PB), zero tillage (ZT) and conventional tillage (CT)) and crop rotations are in sub plots maize–wheat–mungbean (MWMb), maize–chickpea–sesbania (MCS), maize–mustard–mungbean (MMuMb) and maize–maize–sesbania (MMS). Before initiation of the experiment, the field was tilled (up to 30 cm depth) using a chisel plough, pulverized and then laser leveled. The sub-plot size was set at 16.5 m × 4.0 m for the duration of the trial. The CT included using a spring-type cultivator, rotavator and one disc harrow ploughing per field. Using a zero-till planter with an inverted T- type, different crops were directly drilled in ZT plots. The PB was 67 cm wide from mid-furrow to mid-furrow, with 37 cm wide flat tops and 15 cm furrow depth. The PB was reformed with a disc coulter without significantly burying residues at once at the end season, while the raised-bed multi-crop planter was used for crop planting. The crop establishment and management details of each crop are mentioned in Table 2 and the layout of the experimental field is depicted in Figure 1.

Table 2. Crop establishment and management details.

Crop	Variety	Seed Rate	Spacing	Fertilizer Dose
Kharif maize	HQPM–1	20 kg ha^{-1}	67 × 25 cm	150 kg N + 60 kg P_2O_5 + 40 kg K_2O + 25 kg $ZnSO_4$ ha^{-1}
Rabi maize	HQPM–1	20 kg ha^{-1}	67 × 20 cm	180 kg N + 80 kg P_2O_5 + 60 kg K_2O + 25 kg $ZnSO_4$ ha^{-1}
Wheat	PBW–343	100 kg ha^{-1}	Row spacing 22.5 cm (ZT and CT) 18.5 cm (PB)	120 kg N + 60 kg P_2O_5 + 40 kg K_2O ha^{-1}
Chickpea	Pusa–547	80 kg ha^{-1}	30 × 20 cm (ZT and CT) 18.5 × 20 cm (PB)	30 kg N + 40 kg P_2O_5 + 40 kg K_2O ha^{-1}
Mustard	NRCDR–2	5 kg ha^{-1}	Row spacing 30 cm	90 kg N + 40 kg P_2O_5 + 30 kg K_2O ha^{-1}
Mungbean	Pusa Vishal	25 kg ha^{-1}	Row spacing 30 cm	30 kg N + 40 kg P_2O_5 ha^{-1}
Sesbania	Local cultivar	35 kg ha^{-1}	Broadcasted	Not applied

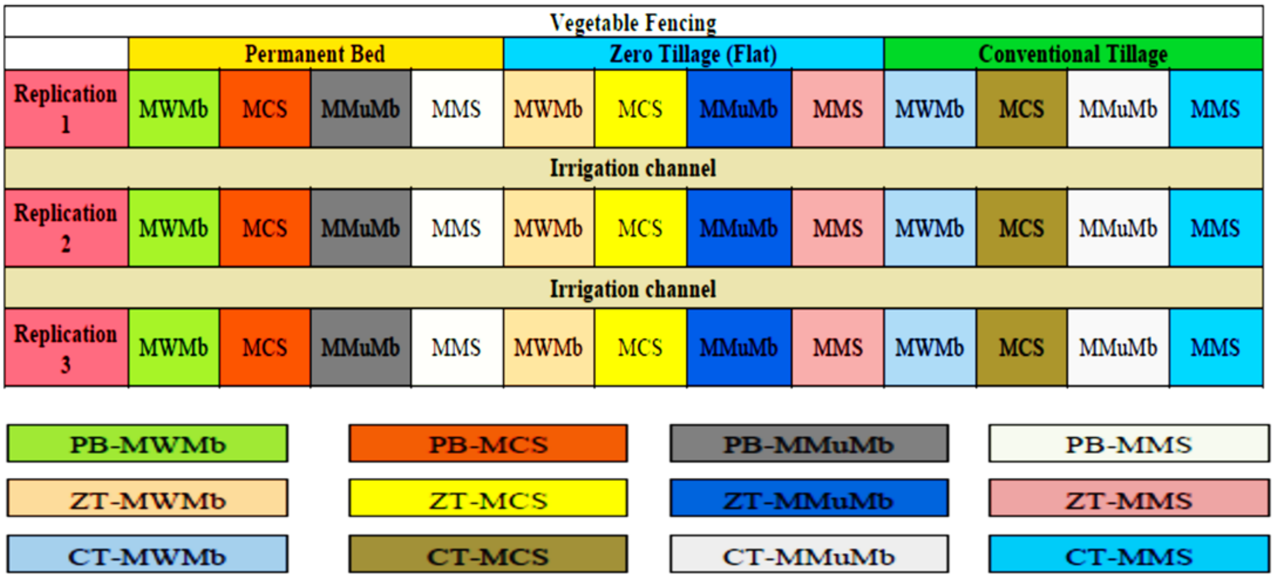

Figure 1. Layout of the experimental field.

2.3. *Soil Sampling and Processing*

The soil was sampled in 2020 from fixed plots. The soil samples (0–5 and 5–15 cm) were collected from each treatment using an auger. They were broken after drying in the shade using gentle strokes from a wooden hammer and soil was air dried under shade and crushed to pass through a 2 mm sieve prior to analyses. The fresh soil samples were stored at 4 °C for assaying biological activity. Bulk soil sample was collected before the establishment of experiment and used to analyze physico-chemical properties.

2.4. Analytical Methods

2.4.1. Inorganic Phosphorus Fraction

To determine the P content in different fractions of soil, the Chang and Jackson [28] and Kuo [29] modified sequential P fractionation scheme was used. This five-step sequential extraction scheme utilized for extraction of different fractions of P consisted of different extraction conditions and different extracting solutions. Following each stage, soils were twice centrifuged and washed with a saturated sodium chloride (NaCl) solution. After centrifugation, the filtrate from the previous step was added to the supernatant to make up the volume. Available P in soil was extracted through Olsen method [30] and quantification performed by colorimetry. After the soil samples were digested in the diacid mixtu re (HNO_3^-–H_3PO_4 mixture), total P (TP) was determined using spectrophotometer. The sum of all inorganic P fractions was used to calculate total inorganic P. The phosphomolybdo-blue color method was used to determine the phosphorus contents in extracts [31].

2.4.2. Organic Phosphorus Fraction

The procedure of organic-P (Po) fractionation modified by Ivanoff et al. [32] and described by Kovar and Pierzynski [33] was used to analyze soil samples. Through this process, we determined labile, fulvic acid P, humic acid P, moderately labile and non-labile organic P fraction. In the same extract total P and total inorganic P (Pi) concentrations were measured after digestion and before digestion, respectively, and their differences were used to determine Po in each fraction. The difference between the humic plus fulvic acid fraction and the fulvic acid fraction was used to compute the humic acid-Po fraction. Calculating moderately labile Po involved adding the HCl-Po and fulvic acid fractions. Humic acid Po and residual Po were combined to give non-labile Po.

2.4.3. Soil Biological Properties

Before the microbiological parameter study, the field-moist soil was given 24 h to acclimate to room temperature. Chloroform fumigation–extraction method [34] was used to determine microbial biomass phosphorus (MBP). Alkali phosphatase enzyme activities (APA) in soil samples were measured in modified buffer (pH 11) using p-nitrophenol method [35].

2.4.4. Statistical Analysis

Using SAS 9.3 software (SAS Institute, Cary, NC, USA), the data collected for various soil parameters were evaluated using the analysis of variance (ANOVA) method (Gomez and Gomez [36] for split-plot design). At a ≤5% level of significance ($p \leq 0.05$), the least significant difference test was employed to determine the interaction effects of the treatments.

3. Results

3.1. Soluble and Loosely Bound P

Tillage and crop establishment methods significantly improve SL-P after twelve years at 0–5 and 5–15 cm soil depths. The concentration of SL-P at both soil depths (0–5 and 5–15 cm) was maximum in ZT flat (16.3 and 15.01 mg kg^{-1}, respectively) and was statistically on par with PB at 0–5 cm soil depth. The increase in SL-P was more in 0–5 cm soil depth (10.8–19.3%) than 5–15 cm soil depth (6.7–13.9%). The effect of crop rotation on SL-P was also significant at 0–5 and 5–15 cm soil depths. The MCS and MWMb plots had the highest levels of SL-P and the values for the two crop rotations were significantly similar for both soil depths. Among various crop rotations, the lowest SL-P was observed in MMuMb plots at 0–5 and 5–15 cm soil depths (12.5 and 12.6 mg kg^{-1}, respectively). The MCS crop rotation had 35.2 and 19% higher SL-P than MMuMb at 0–5 and 5–15 cm soil depths, respectively (Table 3).

Table 3. Long-term impact of different tillage practices and maize-based crop rotation on inorganic P fractions (mg kg^{-1}) at different soil depths.

Treatments	SL-P	Al-P	Fe-P	RES-P	Ca-P	SL-P	Al-P	Fe-P	RES-P	Ca-P
	0–5 cm					5–15 cm				
Tillage Practices (T)										
PB	15.1 ^{AB}	26.1 ^{B}	28.9 ^{B}	107 ^{B}	218 ^{B}	14.0 ^{B}	21.5	28.1 ^{B}	98.9 ^{B}	220
ZT-flat	16.3 ^{A}	26.5 ^{B}	31.1 ^{B}	111 ^{B}	223 ^{B}	15.0 ^{A}	18.5	31.3 ^{A}	103 ^{B}	225
CT-flat	13.6 ^{B}	28.9 ^{A}	33.6 ^{A}	124 ^{A}	247 ^{A}	13.2 ^{B}	22.6	31.8 ^{A}	116 ^{A}	221
SEm (±)	0.63	0.82	0.88	4.13	6.87	0.33	1.28	0.77	3.68	7.12
LSD ($p \leq 0.05$)	1.76	2.27	2.44	11.5	19.0	0.91	NS	2.13	10.2	NS
Cropping systems (CS)										
MWMb	16.4 ^{A}	27.9 ^{A}	30.9	114 ^{AB}	233	14.9 ^{AB}	21.3	29.8	111	221
MCS	16.9 ^{A}	23.1 ^{B}	30.7	123 ^{A}	226	15.0 ^{A}	20.5	30.4	111	219
MMuMb	12.5 ^{C}	28.3 ^{A}	31.7	108 ^{B}	230	12.6 ^{C}	20.8	30.9	97.8	226
MMS	14.2 ^{B}	29.4 ^{A}	31.4	111 ^{B}	228	13.8 ^{B}	20.7	30.3	105	223
SEm (±)	0.66	1.27	0.88	5.16	6.72	0.53	1.28	1.07	6.08	5.62
LSD ($p \leq 0.05$)	1.39	2.68	NS	10.8	NS	1.12	NS	NS	NS	NS
T × CS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Note: SL-P: soluble and loosely bound P; Al-P: aluminum bound P; Fe-P: iron bound P; Ca-P: calcium bound P; RES-P: reductant soluble P; The similar upper-case letters within a column are not significantly different at $p \leq 0.05$ according to the Duncan's multiple range test. NS: Non-significant.

3.2. Aluminum-Bound P

Twelve years of CA practices with crop rotations focused on maize had a big impact on the Al-P fraction in the 0–5 cm soil depth. However, CA practices significantly decreased Al-P by 4.8 and 18% under PB and ZT-flat, respectively, over CT-flat at the 5–15 cm soil layer. Among all the maize-based crop rotations, the MMS resulted in the significantly highest Al-P, which was statistically at par with MMuMb and MWMb at 0–15 cm soil layer. Crop rotation and tillage practices have no effect on Al-P at 5–15 cm soil depth (Table 3).

3.3. Iron-Bound P

Phosphorus fertilization had a significantly substantial impact on the Fe-P fraction, increasing Fe-P (Table 3). Long-term CA practices decreased Fe-P fraction significantly from 33.6 to 29 mg kg^{-1} at 0–5 cm over CT flat. The Fe-P was significantly lower in PB than CT at 0–5 and 5–15 cm soil depths. The tillage practices PB and ZT flat were statistically on par with Fe-P at 0–5 cm soil depths. Different maize-based crop rotations have no statistically significant effect on Fe-P at both soil depths.

3.4. Calcium-Bound P

The Ca-P in the research field was considerably reduced from 9.7 to 11.7% by ZT flat and PB, respectively, over CT flat at 0–5 cm soil depths. The treatments PB and ZT flat were statistically on par for Ca-P in 0–5 cm soil depths. Various cropping systems have no effect on Ca-P at 0–5 and 5–15 cm soil depths (Table 3).

3.5. Reductant Soluble-P

At all soil depths over CT, CA tillage techniques had a detrimental impact on the RES-P percentage. Reductant soluble P under PB and ZT flat were 13.7 and 10.5% lower, respectively, over CT flat at 0–5 cm. However, at 5–15 cm soil depths, RES-P under PB and ZT decreased by 17.1 and 11.2%, respectively, as compared to CT-flat plots. Different maize-based crop rotation significantly influenced RES-P at the 0–5 cm soil depth only and the highest RES-P (123 mg kg^{-1}) was found for the MCS cropping system (Table 3). Distribution of inorganic P fractions follows the order; SL-P < Al-P < Fe-P < RES-P < Ca-P.

3.6. Labile Po

The labile Po ranged from 27.2 to 29.7 mg kg^{-1} and 18 to 20.9 mg kg^{-1} in 0–5 and 5–15 cm soil depths, respectively. Twelve years of MCS crop rotation significantly increased the labile Po by 23.9 and 37.3% over MMuMb in 0–5 and 5–15 cm soil depths. Long-term tillage practices have no effect on labile Po (Table 4).

Table 4. Long-term impact of different tillage practices and maize-based crop rotation on organic P fractions (mg kg^{-1}) at different soil depths.

Treatments	Labile Po		Moderately Labile Po				Non-Labile Po			
	$NaHCO_3$-Po		HCl-Po		Fulvic Acid Po		Humic Acid Po		Residual Po	
	0–5 cm	5–15 cm	0–5 cm	5–15 cm	0–5 cm	5–15 cm	0–5 cm	5–15 cm	0–5 cm	5–15 cm
Tillage practices (T)										
PB	29.7	20.9	49.5	36.5 ^{A}	79.8 ^{B}	71.8 ^{A}	48.9	35.6	139 ^{A}	128 ^{A}
ZT-flat	28.4	20.8	50.0	36.9 ^{A}	81.4 ^{A}	70.2 ^{B}	50.0	36.3	146 ^{A}	127 ^{A}
CT-flat	27.2	18.1	39.5	26.2 ^{B}	68.1 ^{C}	62.2 ^{C}	43.8	29.4	115 ^{B}	101 ^{B}
SEm (±)	2.18	1.06	4.09	2.98	0.29	0.53	3.72	2.94	4.74	0.80
LSD ($p \leq 0.05$)	NS	NS	NS	8.27	0.82	1.48	NS	NS	13.1	2.22
Cropping systems (CS)										
MWMb	29.9 ^{A}	20.8 ^{B}	53.0	40.2	81.5 ^{A}	72.9 ^{A}	53.0 ^{A}	39.3 ^{A}	149 ^{A}	134 ^{A}
MCS	31.1 ^{A}	22.8 ^{A}	50.6	38.1	77.8 ^{B}	67.7 ^{B}	52.2 ^{AB}	38.5 ^{A}	141 ^{B}	125 ^{B}
MMuMb	25.1 ^{B}	16.6 ^{C}	41.4	27.6	75.5 ^{B}	66.9 ^{B}	45.9 ^{B}	29.6 ^{B}	124 ^{C}	112 ^{C}
MMS	27.5 ^{AB}	19.5 ^{B}	40.3	26.8	70.9 ^{C}	64.6 ^{C}	39.2 ^{C}	27.7 ^{B}	118 ^{C}	105 ^{D}
SEm (±)	1.94	0.91	5.84	5.69	1.16	0.68	3.11	1.73	3.42	2.62
LSD ($p \leq 0.05$)	4.08	1.91	NS	NS	2.43	1.44	6.53	3.64	7.18	5.50
T × CS	NS	NS	NS	NS	NS	NS	NS	NS	NS	9.53

Note: The similar upper-case letters within a column are not significantly different at $p \leq 0.05$ according to the Duncan's multiple range test. NS: Non-significant.

3.7. Moderately Labile Po

Long-term tillage and diversified maize-based cropping systems significantly influence moderately labile Po at 0–5 and 5–15 cm soil depths. Moderately labile Po consisted of HCl-Po and fulvic acid Po. Fulvic acid Po among different tillage practices was found to be highest in ZT flat (81.4 mg kg^{-1}) in 0–5 cm and PB (71.8 mg kg^{-1}) in 5–15 cm soil depths. However, among cropping systems, the highest moderately labile Po (81.5 and 72.9 mg kg^{-1}) was found for the MWMb cropping system in 0–5 and 5–15 cm soil depths, respectively. It was observed that the cropping systems MWMb and MCS were statistically on par for fulvic acid Po and moderately lablile Po (Table 4).

3.8. Non-Labile Po

Non-labile Po consisted of humic acid Po and residual Po. The effect of different tillage and maize-based crop rotation and their interaction was significant for non-labile Po. The highest non-labile Po was highest on ZT flat in 0–5 and 5–15 cm soil depths, which is statistically on par with PB. Among various cropping systems, MWMb had the highest non-labile Po (202 and 173 mg kg^{-1}, respectively) at 0–5 and 5–15 cm soil depths, respectively. Moreover, the lowest non-labile Po (169 and 142 mg kg^{-1}) was found for the MMuMb cropping system at 0–5 and 5–15 cm soil depths, respectively (Table 4). The distribution of organic soil P fractions increases in Labile Po < Moderately Labile Po < Non-Labile Po.

3.9. Total P

Total P at different soil depths was affected by long-term tillage practices and maize-based crop rotation (Figure 2). Tillage and crop establishment methods significantly influenced total organic P in both 0–5 and 5–15 cm soil depths (Figures 3 and 4). The concentration of total organic P in 0–5 cm was maximum in ZT flat, which was statistically on par PB; a similar result was also found at 5–15 cm soil depths. Among cropping systems, MWMb was found to be highest in total organic P at 0–5 and 5–15 cm soil depths (367 and 307 mg kg^{-1}, respectively). Total inorganic P was found to be significantly lower in PB and ZT flat (11.6 and 8.9%) compared to CT flat (Figure 3). Different cropping systems were found non-significant in total inorganic P in both 0–5 and 5–15 cm soil depths (Figure 4).

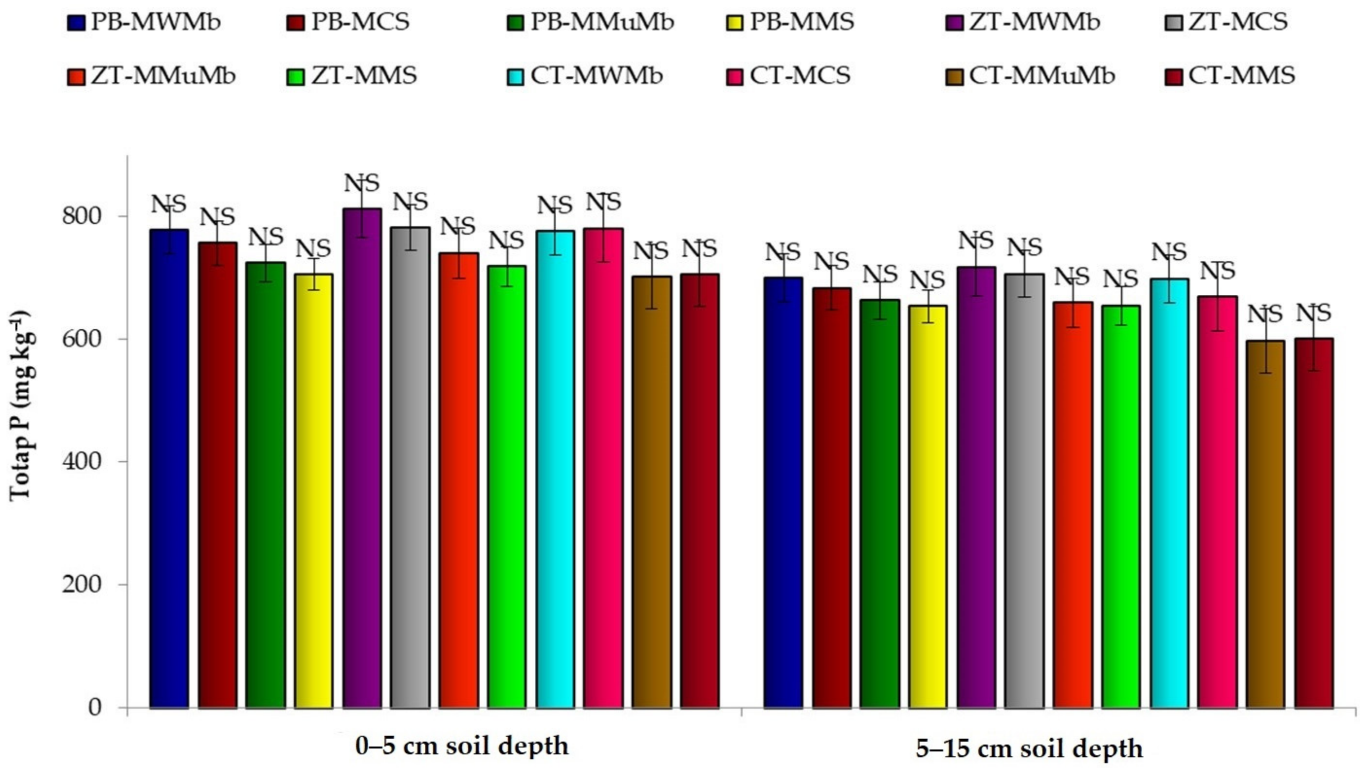

Figure 2. Effect of long-term tillage practices and maize-based crop rotation on total P (mg kg^{-1}) content at different soil depths. NS: Non-significant.

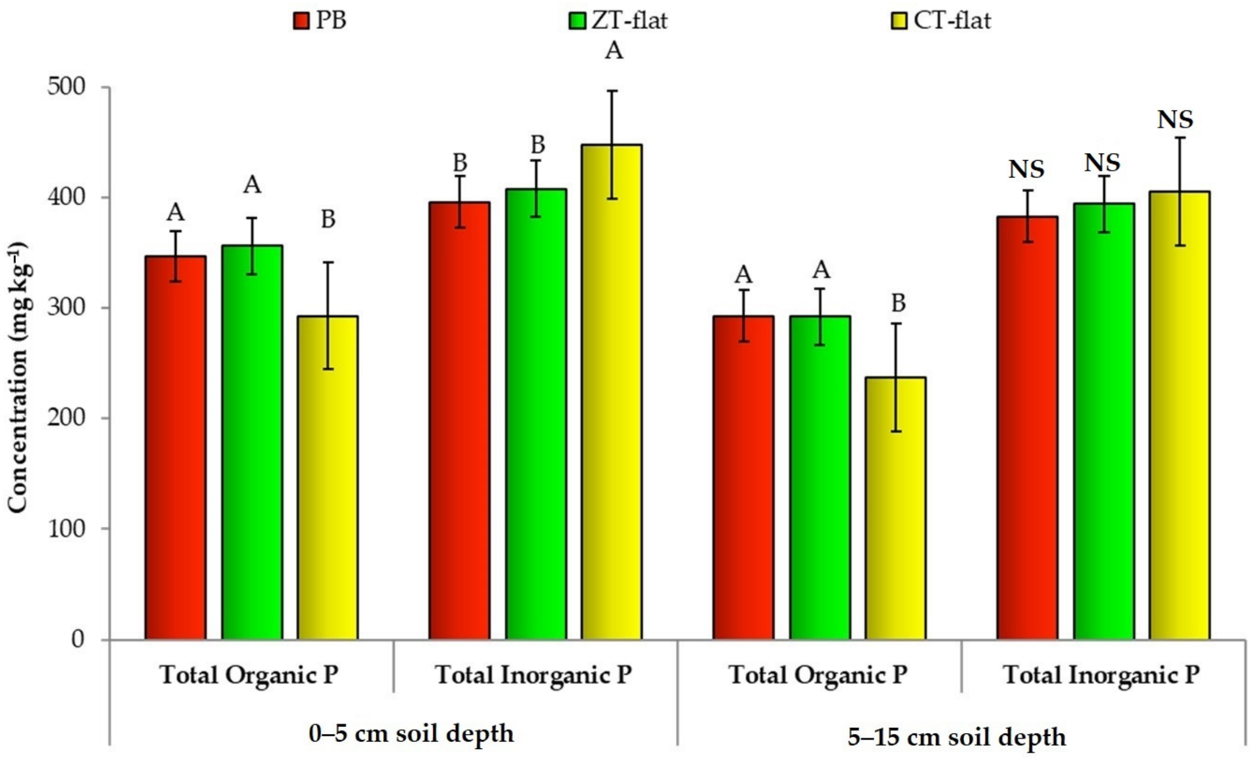

Figure 3. Effect of long-term tillage practices tillage practices on total organic and total inorganic P (mg kg^{-1}) content at different soil depths. Different letters (A and B) for each parameter show significant difference at $p \leq 0.05$ by Duncan's Multiple Range Test. NS: Non-significant difference.

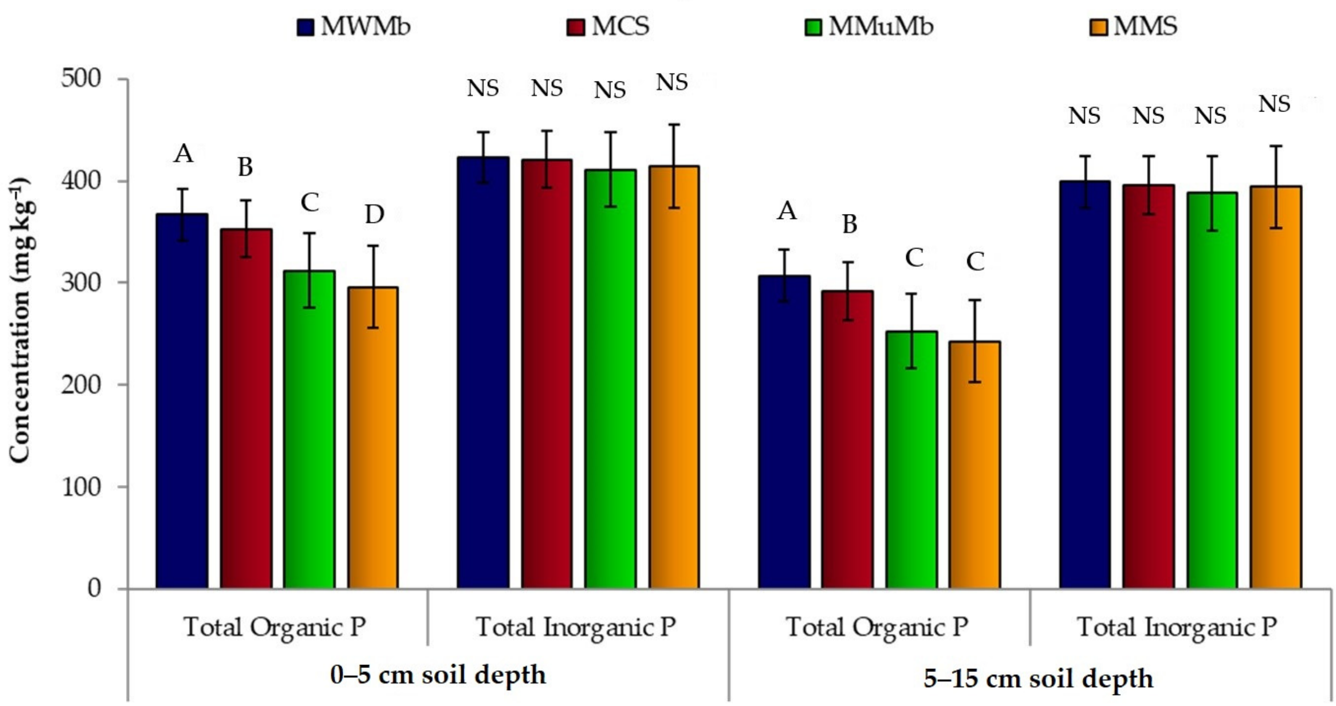

Figure 4. Effect of long-term maize-based crop rotation on total organic and total inorganic P (mg kg^{-1}) content at different soil depths. Different letters (A, B, C and D) for each parameter show significant difference at $p \leq 0.05$ by Duncan's Multiple Range Test. NS: Non-significant.

3.10. Soil Biological Properties

The APA and MBP activities were significantly enhanced under ZT flat and PB compared with CT flat under both soil depths. Under Zt flat, the APA was 18.3–38.0% higher compared with CT flat under 0–5 and 5–15 cm soil depths, respectively (Table 5). MBP was improved by 45.1% upon adoption of ZT flat and the effect was more prominent at 5–15 cm soil depths (104%). Among cropping systems, MCS resulted in the highest level of APA and MBP at 0–5 cm soil depths, whereas at 5–15 cm soil depth, APA and MBP were found highest in MMuMb and MMS cropping systems, respectively. At 0–5 and 5–15 cm soil depths, the interaction between tillage and crop rotation was significant for soil alkaline phosphatase and MBP.

Table 5. Long-term impact of different tillage practices and maize-based crop rotation on alkaline phosphatase activities (μg p-NP g^{-1} soil h^{-1}) and microbial biomass phosphorus (mg g^{-1}) at different soil depths.

Treatments	**Alkaline Phosphatase Activities (μg *p*-NP g^{-1} soil h^{-1})**		**Microbial Biomass Phosphorus (mg g^{-1})**	
	0–5 cm	**5–15 cm**	**0–5 cm**	**5–15 cm**
Tillage practices (T)				
PB	282.7 ^{B}	165.6 ^{B}	30.4 ^{A}	27.2 ^{B}
ZT-flat	298.8 ^{A}	185.3 ^{A}	31.5 ^{A}	29.1 ^{A}
CT-flat	252.5 ^{C}	134.2 ^{C}	21.7 ^{B}	14.2 ^{C}
SEm (±)	2.28	2.19	1.12	0.44
LSD ($p \leq 0.05$)	6.34	6.08	3.12	1.22
Cropping systems (CS)				
MWMb	289.4 ^{B}	143.6 ^{D}	28.6 ^{B}	25.0 ^{B}
MCS	308.6 ^{A}	173.0 ^{B}	33.6 ^{A}	24.2 ^{B}
MMuMb	228.5 ^{C}	180.1 ^{A}	20.6 ^{C}	17.5 ^{C}
MMS	285.4 ^{B}	150.1 ^{C}	28.7 ^{B}	27.3 ^{A}
SEm (±)	2.41	1.77	1.14	0.74
LSD ($p \leq 0.05$)	5.06	3.72	2.39	1.55
T × CS	8.77	6.45	4.14	2.68

Note: The similar upper-case letters within a column are not significantly different at $p \leq 0.05$ according to the Duncan's multiple range test.

4. Discussion

The larger ($p \leq 0.05$) SL-P at 0–5 and 5–15 cm depths of ZT flat and PB plots, then in CT plots, showed that CA practices effectively improve available P fraction in soil. When residue was applied to the soil, Ohno and Erich [37] discovered a comparable rise in SL-P in the soil to our results. The decreased P adsorption in specific surface of the mineral resulted in improved soil-P status, which was primarily responsible for the rise in P availability. The greater value of SL-P was achieved at a soil depth of 0–15 cm, as opposed to 15–30 cm. This might be as a result of the native P being redistributed over time toward the surface in the CA practices, such as residue retention. Hence, the rise in available P is maximally concentrated in surface soil. Due to P fixation via leaching and low mobility of phosphate ions, the SL-P was similarly lower for samples of lower soil depths, as stated [38,39]. Comparable findings were obtained by Weil et al. [40], who concluded that the accumulation of soil organic matter is what causes the increase in SL-P. An increase in the available N through N fixation helps a synergistic effect on available P, which results in higher SL-P in an MCS cropping system.

Conservation agriculture practices show a significant ($p \leq 0.05$) decrease in Al-P fraction compared to CT at 0–5 cm soil depths. Therefore, this could be as a result of residue retention, increasing soil microbial activity, which improves the solubilization of Al-P. Al-P was shown to decrease an incubation experiment after the application of organic manure, due to dissolution of Al–P upon application of manure [41]. The Al was present in this soil in a reactive state, which bound to the additional P by forming a covalent Al-P bond on the surface of Al-oxide in soil after fertilizer application and may be responsible for the rise in Al-P [42]. Root exudates from winter and summer legumes are the reason for low-AL-P contents in the MCS cropping system at 0–5 cm soil depths.

The result of different tillage for Fe-P suggests that the quantity of Fe-P reduced with CA at 0–5 cm soil depths. These fractions are Fe-phosphate, tightly bound to the Fe-oxide mineral, decreasing the availability of P to the plants [43]. By reducing or chelating Fe, increased microbial activity brought on by the addition of CR may aid in the breakdown of Fe-P or P associated with Fe-oxide minerals. Different cropping system treatments show a non-significant relationship with Fe-P due to low-Fe content in these soils. Others found an opposite pattern, with manure application Fe-P fraction rising [41]. These observations were found due to manure application triggering an Fe-P synthesis pathway.

Different CA practices (PB, ZT-flat) reduce Ca-P in surface soil (0–5 cm) compared to CT plots. Due to the soil's higher pH, this fraction held the majority of the P. In the sandy loam soil, about 40% of P was in forms that were bound to calcium. Due to the higher stability of Ca-P under a pH above 7, alkaline soils typically maintain a considerable quantity of P as Ca-P, regardless of the kind of applied fertilizers. Comparable results on the inorganic P fraction in prolonged fertilization studies in calcareous soils were also reported by Song et al. [44]. The majority of the P in the current experiment may be stabilized as Ca-P due to the high soil pH. The bulk of P was present in the Ca-bound fraction, since a redox reaction does not directly affect the solubility of Ca-P [45]. Kumawat et al. [46] also found that retaining crop residues greatly decreased the Ca-P in the soil, whereas P fertilization at various soil depths significantly increased it.

Conventional tillage results in accumulation of RES-P at 0–5 and 5–15 cm soil depths. Soil mixing through CT practices results in more accumulation of P as an RES-P fraction. The alkali-insoluble P was revealed as reductant soluble, but the character of this fraction was only partially characterized [47]. While varied P-fertilization rates considerably raised RES-P at both soil levels, increasing CR retention had a detrimental impact on the reductant soluble-P fraction at all soil depths [46].

The result of tillage and cropping systems for organic-P fraction suggests that the proportion of labile, moderately labile and non-labile fractions decrease with soil depth. After using manure continuously for 11 years, [48] also noted comparable outcomes, with labile Po concentrations ranging from 22 to 23 mg kg^{-1}. Due to the fact that labile Po is the main unstable Po fraction and converts to plant availability the earliest [49], this random fluctuation occurred. This is primarily because it is easily accessible to soil microbes [50]. This concentration of labile Po fraction can change depending on the kind of soil, microbial population, enzymatic activity and weather conditions [51]. The phospholipids, nucleic acids and sugar phosphate were the main sources of the labile Po obtained in this extraction. Since these substances are easily mineralizable and quickly release accessible P, plants or soil microbes can use them [52]. According to Ramphisa [53], applying manure promotes microbial activity, which first immobilizes P and it is then mineralized and released to plants. Po instability was similarly shown by Hao et al. [54], who found higher manure addition without proper irrigation increased labile Po values but claimed there was no such tendency during irrigation. Depending on the amount of manure used, the labile Po fraction either remained unchanged or decreased throughout irrigation. Po is, hence, an active fraction and labile Po is quickly mineralized or converted into more stable molecules rather than accumulating [55].

With HCl solution as the extractant, the lowest Po recovery was achieved. To avoid interfering with the subsequent extractions, the samples were pre-treated with the primary goal of removing Pi [33]. However, the acidic HCl solution removes Po from the OM-associated humic acid fraction and Po bound to organic Al, Ca compounds. This extraction is taken into consideration when calculating the moderately labile Po [33]. However, as observed by Motavalli and Miles [48] and Sharpley et al. [56], there was a considerable impact of manure on the organic-P fractions, such as moderately labile and non-labile Po. The humification of manure by microbes may be the cause of higher Po in these fractions; furthermore, these changes are time related. Schroeder and Kovar [57] found that the fulvic acid Po quantity was around four-times greater than the humic acid Po content. According to these researchers, fulvic acid Po were the main types of organic P in the soil. According to Soltangheisi et al. [58], more moderately labile P could be used by cover crops, which also increased the amount of labile P components in the soil. The primary components in this fraction are fulvic and humic acids, with fulvic acids being more labile molecules and having elevated P concentrations [57]. On the other hand, under CT, labile and moderately labile fractions of Po are more simply mineralized and these could be the primary source of P to plants [11].

The residual P fraction acts as a sink in the soil systems when treated with P through fertilizers or crop residues [54]. Reddy et al. [59] observed that residue incorporation and retention increased $NaHCO_3$-Pi and $NaHCO_3$-Po.

Total P was found to be insignificant with different tillage practices in 0–5 cm soil depths, whereas it was found significantly higher in ZT flat and PB (6.7% and 5.3%, respectively) at 5–15 cm soil depths. Among different cropping systems, MWMb was found to be highest in total P, which was statistically similar to the MCS cropping system at both soil depths. Increased P concentrations in the non-labile and moderately labile Po fractions helped to raise overall Po. This organic-P increase was seen regardless of the method used to calculate Po; however, for the majority of soil samples, with the incineration technique, lower Po values were attained. It was expected and described by previous authors that the plots treated with manure would show an increase in organic P contents [54,56]; despite the low amounts, P is present in all types of manure. Only a small portion of applied P fertilizers is taken up by plants, while the majority is fixed in soil in different, less-available forms [16,17].

A higher abundance of organic substrate under ZT-flat and PB treatment resulted in a significant increase in MBP and APA activities. The supply of carbon and nutrients through mineralization is a key factor for the proliferation of microbes. Alkaline phosphatase activity was higher under CA, owing to the alkaline reaction in the present experimental soil and easily decomposable substances. The microbial population, particularly bacteria and fungi proliferated extensively due to the high C, N and P supply. The differential C:N ratio and lignin content resulted in different microbial activity in each sub-plot treatment. The allelopathic effect of residue from the mustard crop also affected the rate of decomposition. Alkaline phosphatase and MBP were significantly impacted by the synergistic interactions between winter legumes (chickpea) and summer legumes (sesbania and mungbean) in crop rotation [60].

5. Conclusions

The results of this study reveal that long-term conservation agriculture, under different tillage and cropping systems, influences phosphorus dynamics in the soil. Twelve years of permanent beds and zero-tillage-flat practices have improved the phosphorus fertility in soil by improving available phosphorus fraction and organic phosphorus. The zero-tillage-flat and permanent beds with crop residue retention and inclusion of two legumes, chickpea and sesbania, enhanced the availability of phosphorus in soil. The additional beneficial effect of legume crop rotation on soil phosphorus fertility was further improved with zero-tillage-flat and permanent-bed-tillage practices. Zero-tillage flat and permanent beds significantly enhanced organic phosphorus in surface and subsurface soil; however, zero-tillage flat and permanent beds at 0–5 and 5–15 cm soil depths significantly decreased the inorganic phosphorus proportion. Among inorganic phosphorus fractions, soluble and loosely bound phosphorus was found higher in zero-tillage flat, whereas aluminum-bound phosphorus, iron-bound phosphorus, reductant soluble phosphorus and calcium-bound phosphorus were found higher in conventional tillage at both 0–5 and 5–15 cm soil depths. Among organic phosphorus fractions, moderately labile and non-labile Po was found to be higher in permanent beds and zero-tillage flat but, in the case of labile organic phosphorus, it was found insignificant with respect to tillage operations. Additionally, it was found that ZT flat followed by PB significantly improved MBP and alkaline phosphatase levels in soil compared to CT flat. When comparing crop rotations, MCS emerged as the most effective maize-based rotation for enhancing the soil's biological activities (APA, MBP) and available P fractions. Thus, adoption of conservation agriculture will help in improving phosphorus availability and, at the same time, offer a viable opportunity for crop residue disposal. The results of this study could enhance the understanding of phosphorus dynamics in soil and prove useful in rationalizing nutrient management practices under different conservation agriculture practices.

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