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Analysis of phosphorus forms on different soil types using heterogeneous isotope exchange reaction

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ABSTRACT

Pot experiments and soil incubation were carried out to follow the processes of added phosphorus (P) fertilizer using radioisotope tracer technique, heterogeneous isotope exchange. Four types of soils were investigated (Dystric Arenosol (Humic), Calcic Vertisol (Gleyic), Rendzic Phaeozem (Hyperhumic), Calcic Gleysol(Arenic, Humic)). The changes of phosphate fractions (water-soluble (P_w), isotopically exchangeable (P_{IE}), ammonium lactate soluble phosphorus (P_{AL}), and tightly sorbed phosphorus ($P_{tightly}$)) in soil were determined as a function of P supply, incubation time and the plant culture. The soil samples were incubated at different times (1, 3, 13 weeks) and phosphate quantities. 0, 40, 80, 160, 320 mg P/kg soil doses of P fertilizer were added to the soils. After incubation, perennial ryegrass (Lolium perenne L.) was sown. A significant correlation was found between the sum of P_w and P_{IE} and P_{uptake} of plant proving that these values can be good indicators of plant available phosphorus. During plant culture, the P_w and P_{IE} values always decrease showing that plant directly utilizes these phosphate forms. The transformation of added phosphate to tightly sorbed phosphate is the highest for soils with great humus content.

1. Introduction

In crop production, it is important to use phosphorus fertilizer to achieve adequate yields. Excessive use of fertilizers can lead to economic losses, as well as eutrophication (Schindler, 1977) and depletion of limited resources (Cordell et al., 2009). Phosphorus plays an important role in plant development and also influences yield (Mengel et al., 2001; Vance et al., 2003). Phosphorus is a non-renewable resource and has no substitute (Goll et al., 2012). Determining the amount of P required for the maximum yield with minimal negative impact is essential. It would be advisable to use a method that accurately, quickly and reliably measures the amount of phosphorus that can be taken up by the plant. Currently, acidic ammonium acetate lactate (P_{AL}) extraction is used in Hungary (Egnér et al., 1960). In several countries (eg Norway, Sweden, Slovenia) this method is accepted for the determination of plant available phosphorus (Braun et al., 2019). There are other extraction methods (Olsen (Olsen et al., 1954), Mehlich 3 (Mehlich, 1984), Bray (Bray and Kurtz, 1945)), which are accepted to determine the amount of phosphorus available for plants (Daly et al., 2015; Audette et al., 2016; Duminda et al., 2017; Miller and Arai, 2017; Yan et al., 2017). However, P_{AL} may overestimate the amount of phosphorus that can be taken up by plants, because these method cannot accurately determine the weakly bound phosphate and do not take into account the slow release of phosphorus into the soil solution (Morel and Plenchette, 1994; McLaren et al., 2014; Braun et al., 2020).

In our previous works, a model was proposed (Kónya and Nagy, 2015) for the evaluation of the heterogeneous isotope exchange of the radioactive ^{32}P isotope through which the quantity of phosphate dissolved in the soil solution as well as the weakly sorbed phosphate on soil can be studied under a steady state and equilibrium. These two quantities together are considered to be available for plants. In addition, the transport rate of phosphorus between the soil and solution, also under a steady state or equilibrium can be determined. The heterogeneous isotope exchange clearly illustrates the most important aspects and advantage of the radiotracer methods: the soil and soil solution under a steady state or equilibrium can be studied without disturbing the exchange process. So the developed method shows the actual amount of P species at the time of sampling (Nagy et al., 2019).

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Table 1
The main properties of Dystric Arenosol (Humic), Calcic Vertisol (Gleyic), Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic).

	Dystric Arenosol (Humic)	Calcic Vertisol (Gleyic)	Rendzic Phaeozem (Hyperhumic)	Calcic Gleysol (Arenic, Humic)
pH (H_2O)	6.95	8.14	7.49	8.22
pH (KCl)	5.81	6.97	6.95	7.37
Plasticity according to Arany[a]	< 30	45	–	–
Humus content %	1.22	3.25	14.52	6.39
Hydrolytic acidity	0.6	2.85	–	–
Organic C (g/kg)	7.00	18.9	84.2	37
N (g/kg)	0.65	2.00	8.40	3.70
Clay and silt %	12	52	28.36	28.32
$CaCO_3$%	–	3.00	11.51	14.39
Ammonium-lactate extractable-P (mg/kg)	81	70	13	118
Ammonium-lactate extractable-K (mg/kg)	261	201	164	142
Total P (digestion with H_2SO_4) (mg/kg)	491	842	1518	1044

[a] Plasticity index according to Arany: water quantity taken up by soil to reach plasticity capacity (cm^3/100 g).

In this paper, four types of soils were investigated (Dystric Arenosol (Humic), Calcic Vertisol (Gleyic), Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic)). The changes of phosphate fractions (water-soluble (P_w), isotopically exchangeable (P_{IE}), and ammonium lactate soluble phosphorus (P_{AL}), tightly sorbed phosphorus ($P_{tightly}$)) in soil were determined as a function of P supply, incubation time and the plant culture. Moreover, the correlations between P fractions and P uptake of the plant (P_{uptake}) were also investigated. In this work, we investigate the changes of phosphate fractions (P_W, P_{IE}, $P_{tightly}$, P_{AL}) in the soil in dependence on P supply, incubation time and the plant growing. In addition, we study the correlations between P fractions and P uptake by the plant. A better understanding of P sorption processes in soil-plant systems will allow contributing to achieving nutrient management more effectively. This work is the continuation of our previous work (Balla et al., 2021), where two soil types (Chernozem and Calcaric Arenosol (Humic)) were investigated. On the basis of previous results, it can be stated that the P fractions by increasing P rates and preincubation periods changed differently on Chernozem and Calcaric Arenosol (Humic), due to the different organic matter and clay content as well as the P transformation to tightly sorbed form. In this study, P fractions of other soil types were also investigated, samples with and without plant growing were compared, and it was examined how P species change in different soils taking into account the properties of the soil.

2. Material and methods

Soil incubation and pot experiments (Balla et al., 2021) were carried out on Dystric Arenosol (Humic), Calcic Vertisol (Gleyic), Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic). The soils were classified according to World Reference Base (WRB) for Soil Resources (European Soil Bureau Network European Commission, 2005; IUSS Working Group WRB, 2014). The soil samples were collected from the upper 0–0.3 m layer of arable areas in the eastern part of Hungary. Table 1 shows the main properties of soils.

The soil samples were incubated at different times (1, 3, 13 weeks) and phosphate quantities. 0, 40, 80, 160, 320 mg P/kg soil doses of P fertilizer were added to the soils. Then soil samples were divided into 2 groups. In the first group of pots, perennial ryegrass (*Lolium perenne L.*) was sown, while in the second group no plant was sown, both groups were followed by another 9 weeks of plant growing or incubation, respectively (Balla et al., 2021). After incubation and plant growing, heterogeneous isotope exchange was examined by adding carrier-free radioactive ^{32}P-labeled phosphate to determine the relative quantity of phosphate species and the exchange rate between these species under steady state conditions (Kónya and Nagy, 2015). The heterogeneous isotope exchange studies can be done only all phosphorous species are in equilibrium in the system. By adding carrier-free phosphorous isotope to such system, the concentration of which is below 10–10 mol/dm^3, the sorption fee energy is not affected by this isotope just because of the very low concentration. Only the mixing entropy is changed; directing the isotope exchange. In our system, the phosphorous content is by several orders of magnitude higher than the concentration of radioactive phosphorous, there is no other reaction, only isotope exchange occurs. obviously, if no phosphorous were in the system, the sorption of radioactive phosphorous adsorption was studied, not the heterogeneous isotope exchange. As a conclusion, the criterion of the heterogeneous isotope exchange studies is that the total concentration/quantity of inactive phosphorous in the system must be at least 3–4 orders of magnitude higher than that of radiotracer.

The grass P uptake (P_{uptake}), water-soluble (P_w), isotopically exchangeable (P_{IE}), and ammonium lactate soluble phosphorus (P_{AL}) fractions of soils were measured at the end of the pot experiment from soil samples with and without plants. P_w phosphorus was determined by

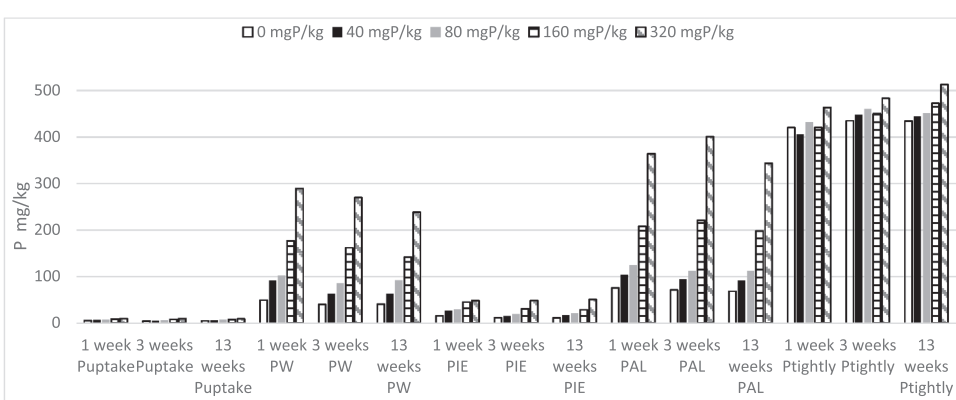

Fig. 1. Phosphate fractions after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Dystric Arenosol (Humic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, p < 0.05 %).

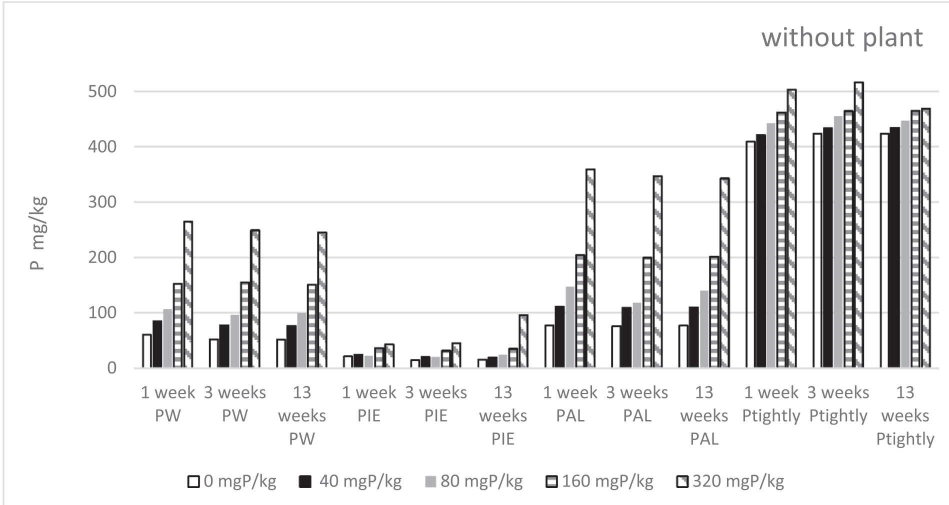

Fig. 2. Phosphate fractions without plant growing (P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Dystric Arenosol (Humic), (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

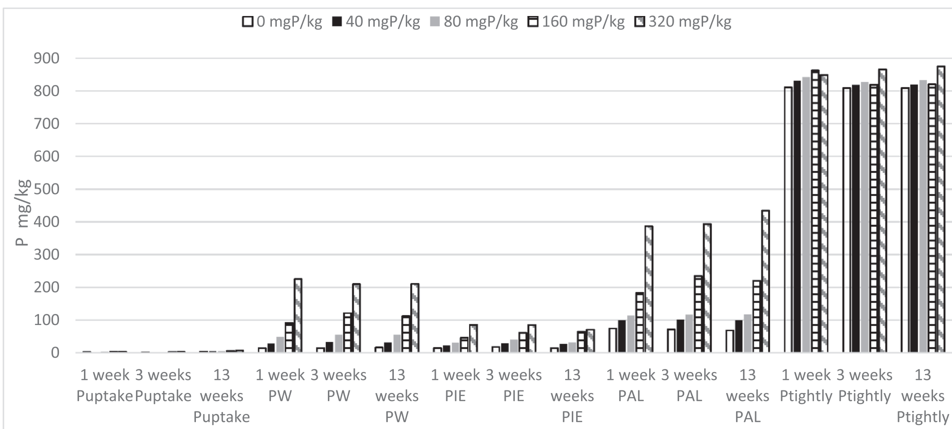

Fig. 3. Phosphate fractions after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Vertisol (Gleyic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

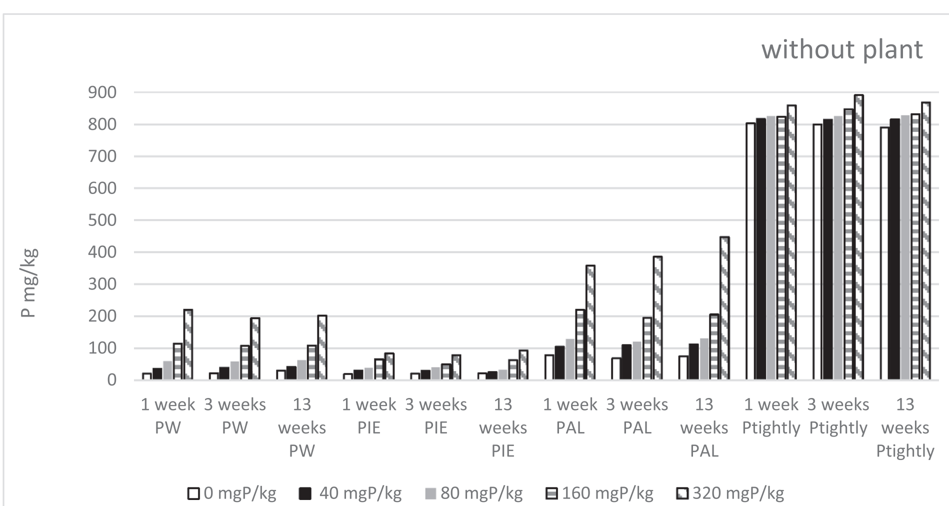

Fig. 4. Phosphate fractions without plant growing (P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Vertisol (Gleyic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

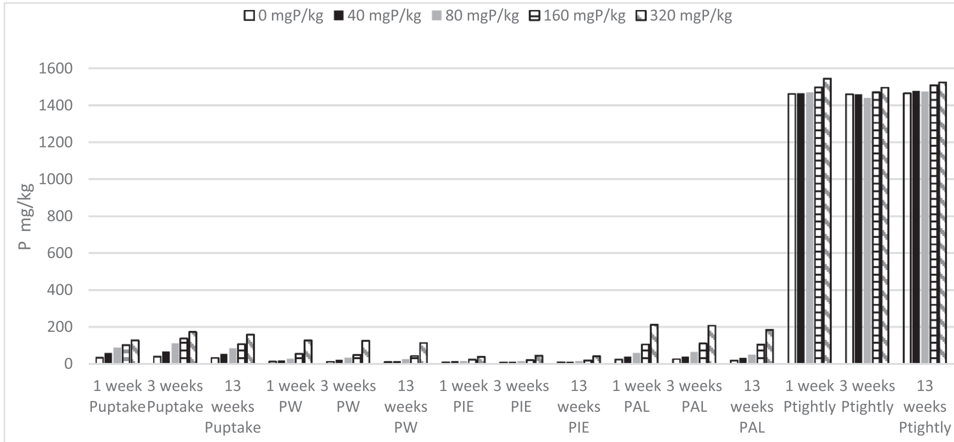

Fig. 5. Phosphate fractions after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Rendzic Phaeozem (Hyperhumic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

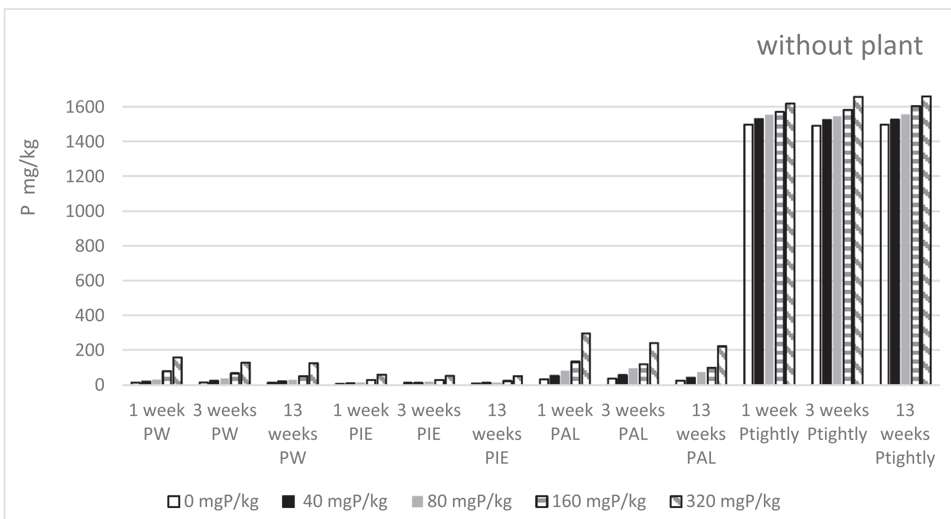

Fig. 6. Phosphate fractions without plant growing (P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Rendzic Phaeozem (Hyperhumic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

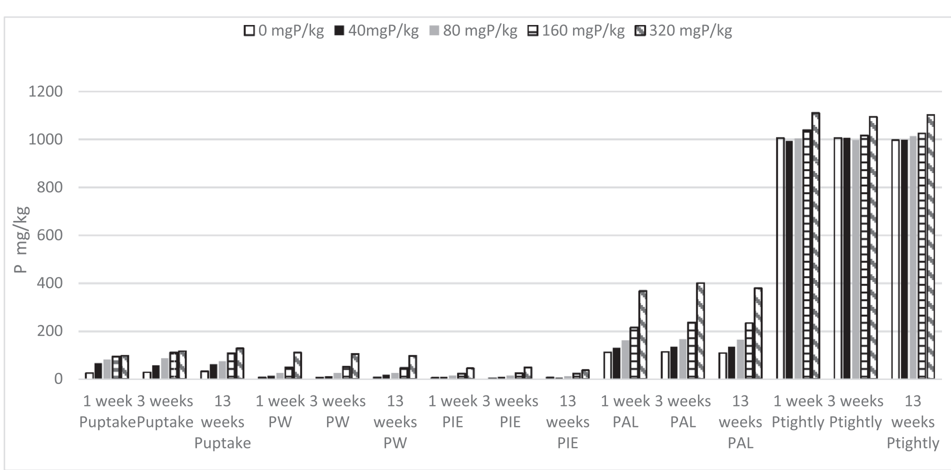

Fig. 7. Phosphate fractions after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Gleysol (Arenic, Humic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

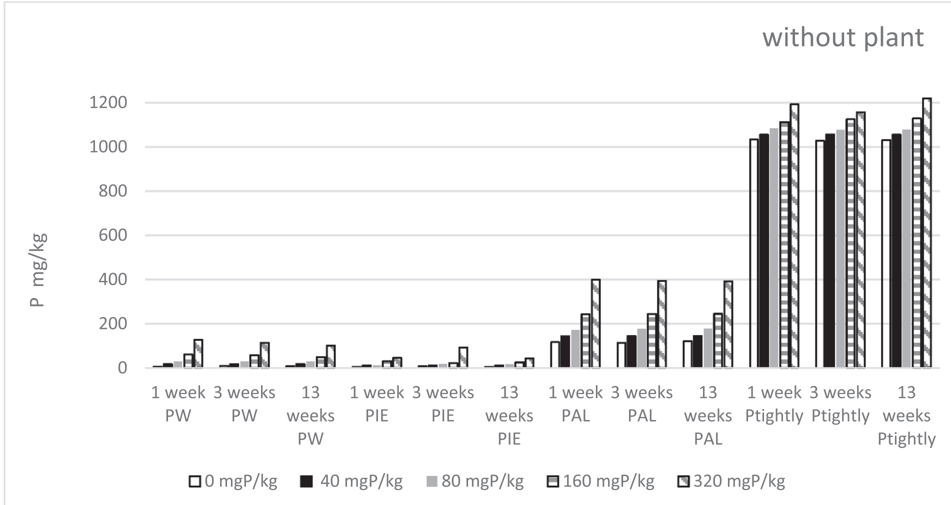

Fig. 8. Phosphate fractions without plant growing (P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Gleysol (Arenic, Humic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

Table 2
Phosphate fractions without plant growing and after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Dystric Arenosol (Humic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

Without plant									
	P_W			P_{IE}			P_{AL}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	60.3	52.6	51.7	21.4	14.6	15.6	77.2	76.2	77.1
40 mg/kg	85.2	77.5	76.8	24.6	20.0	19.5	111.0	109.3	110.1
80 mg/kg	106.5	96.2	99.7	22.0	19.9	24.3	147.4	117.9	140.0
160 mg/kg	152.4	154.9	150.9	36.4	31.7	35.1	204.4	199.9	201.6
320 mg/kg	264.75	249.04	245.48	43.21	45.43	96.10	359.10	346.80	343.00
	$P_{tightly}$								
	1 week	3 weeks	13 weeks						
0 mg/kg	409.3	423.8	423.7						
40 mg/kg	421.3	433.5	434.8						
80 mg/kg	442.5	454.9	447.0						
160 mg/kg	462.3	464.4	465.0						
320 mg/kg	503.0	516.5	469.4						
After plant growing									
	P_{uptake}			P_W			P_{IE}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	5.5 a^{A}	4.6 a^{A}	4.9 a^{A}	49.6 a^{A}	40.2 a^{A}	40.6 a^{A}	15.7 a^{A}	11.5 a^{A}	11.1 a^{A}
40 mg/kg	6.9 ab^{A}	5.3 ab^{A}	6.1 ab^{A}	91.8 a^{A}	63.0 a^{A}	63.3 ab^{A}	27.2 ab^{B}	15.2 a^{A}	16.8 a^{A}
80 mg/kg	7.4 ab^{A}	6.4 ab^{A}	7.3 b^{A}	102.2 a^{A}	85.2 a^{A}	91.9 b^{A}	29.4 ab^{A}	19.2 ab^{A}	21.0 ab^{A}
160 mg/kg	8.7 b^{B}	8.1 bc^{A}	7.9 bc^{A}	176.6 b^{A}	161.9 b^{A}	141.6c^{A}	45.4 b^{A}	30.9 b^{A}	28.9 b^{A}
320 mg/kg	9.9 b^{A}	9.8 c^{A}	9.2 c^{A}	289.2 c^{A}	269.6 c^{A}	238.3 d^{A}	48.5 b^{A}	48.6 c^{A}	50.8 c^{A}
	P_{AL}			$P_{tightly}$					
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks			
0 mg/kg	75.6 a^{A}	71.1 a^{A}	68.5 a^{A}	420.3 a^{A}	434.7 a^{A}	434.4 a^{A}			
40 mg/kg	104.0 a^{A}	94.5 a^{A}	91.6 a^{A}	405.1 a^{A}	447.5 a^{A}	444.8 ab^{A}			
80 mg/kg	124.2 ab^{A}	112.3 a^{A}	112.0 a^{A}	432.0 a^{A}	460.2 a^{A}	450.8 ab^{A}			
160 mg/kg	207.7 b^{A}	221.1 b^{A}	197.8 b^{A}	420.3 a^{A}	450.1 a^{A}	472.5 b^{A}			
320 mg/kg	364.2 c^{A}	400.4 c^{A}	343.7 c^{A}	463.4 a^{A}	482.9 a^{A}	512.7c^{A}			

shaking 1:200 soil to water suspension for two hours and the P concentration of the extract was measured by ammonium phosphomolibdate blue photometric method (EN ISO 6878). P_{IE} was determined by heterogeneous isotope exchange (Kónya and Nagy, 2015), where to soil suspension with 1:200 soil and distilled water carrier-free ^{32}P (as KH_2PO_4) was added. The detailed experimental technique can be found in Kónya and Nagy, 2015, which will be briefly discussed here. The suspension was shaken for 2 h, samples were filtered at different times, and the intensity was measured by liquid scintillation method. The amount of exchangeable P was calculated from ratio of the radioactivities of the soil and the solution. In the case soils with and without plant, the amount of tightly sorbed P forms was calculated in the following way (Nagy and Kónya, 2018):

$$P_{tightly} \text{ without plant} = P_{total} - (P_{IE} + P_w) \quad (1)$$

$$P_{tightly} \text{ with plant} = P_{total} - (P_{IE} + P_w + P_{uptake}) \quad (2)$$

The ammonium-lactate–acetic acid soluble P content (P_{AL}) was determined according to Egnér et al. (1960). At the end of the experiment the total biomass was determined by harvesting and drying the ryegrass at 60 °C. The total plant P was determined after the digestion with H_2SO_4-H_2O_2 by ammonium molybdate vanadate spectrophotometric method (Thamm et al., 1968). The P uptake of plant was calculated from the P concentration in the shoot and dry matter yield of plant

Table 3
Phosphate fractions without plant growing and after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Vertisol (Gleyic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %).

Without plant

	P_W			P_{IE}			P_{AL}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	20.3	21.7	30.0	18.8	20.3	21.7	78.1	68.6	74.6
40 mg/kg	35.1	38.8	41.2	30.1	28.8	25.2	104.6	109.2	112.0
80 mg/kg	59.2	57.6	62.3	38.0	39.4	32.5	128.6	119.8	130.0
160 mg/kg	114.2	106.4	108.5	64.8	49.3	62.6	220.4	194.7	204.9
320 mg/kg	220.1	193.6	201.7	83.5	77.8	92.5	357.7	385.6	447.5

	$P_{tightly}$		
	1 week	3 weeks	13 weeks
0 mg/kg	802.9	800.0	790.3
40 mg/kg	816.8	814.3	815.6
80 mg/kg	824.8	825.0	827.2
160 mg/kg	823.0	846.3	830.9
320 mg/kg	858.4	890.6	867.8

After plant growing

	P_{uptake}			P_W			P_{IE}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	1.6 a^{A}	1.4 a^{A}	3.1 a^{A}	14.2 a^{A}	14.3 a^{A}	15.8 a^{A}	15.2 a^{A}	17.6 a^{A}	14.3 a^{A}
40 mg/kg	2.1 ab^{A}	1.5 a^{A}	4.1 a^{B}	27.6 a^{A}	33.1 a^{A}	31.5 a^{A}	21.5 a^{A}	29.1 ab^{A}	26.9 ab^{A}
80 mg/kg	2.5 ab^{A}	1.8 a^{A}	4.4 a^{A}	47.8 b^{A}	54.2 a^{A}	54.7 a^{A}	29.8 a^{A}	39.4 b^{A}	30.7 abc^{A}
160 mg/kg	2.8 ab^{AB}	2.3 a^{A}	5.8 a^{B}	91.3 c^{A}	121.1 b^{B}	111.8 b^{B}	45.5 a^{A}	60.6 c^{A}	64.3 bc^{A}
320 mg/kg	3.3 b^{A}	2.8 a^{A}	7.0 a^{A}	224.8 d^{A}	209.5 c^{A}	210.3 c^{A}	85.6 b^{A}	84.6 d^{A}	70.4 d^{A}

	P_{AL}			$P_{tightly}$		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	73.9 a^{A}	71.4 a^{A}	68.4 a^{A}	810.9 a^{A}	808.7 a^{A}	808.9 a^{A}
40 mg/kg	99.7 b^{A}	100 ab^{A}	99.3 ab^{A}	830.8 ab^{A}	818.4 a^{A}	819.5 a^{A}
80 mg/kg	113.8 c^{A}	116.2 b^{A}	117 b^{A}	841.9 ab^{A}	826.6 a^{A}	832.1 a^{A}
160 mg/kg	183.3 d^{A}	234.7 c^{B}	219.6 c^{AB}	862.4 b^{A}	818.1 a^{A}	820.1 a^{A}
320 mg/kg	386.7 e^{A}	393.1 d^{A}	434.1 d^{A}	848.3 ab^{A}	865.1 a^{A}	874.3 a^{A}

shoot and was expressed in mg P/kg soil (Balla et al., 2021).

Statistical analysis was carried out with SPSS 13.0 software. One-way ANOVA analysis was carried out to evaluate the effect of phosphorus rates and incubation period separately on plant P uptake and on soil P fractions. Statistical analysis was carried out on the first group of pot experiments, because there were two repetitions with plant. Significant differences are indicated by different letters ($p < 0.05$ ANOVA and Tukey tests). The relationships between various phosphorus forms were developed by regression analysis.

3. Results and discussion

Figs. 1–8 show the phosphate fractions on different soil types with and without plants. The figures are interpreted separately for each soil type.

3.1. P species changes on Dystric Arenosol (Humic)

3.1.1. After plant growing

After plant growing the P_{uptake} values ranged between 4.6 and 9.9 mg/kg. The values did not change significantly with the incubation time, but increased as the amounts of P added was increased (Fig. 1, Table 2). Plant with a higher mass was grown on the Dystric Arenosol (Humic) as the added phosphorus increases, so a higher amount of phosphorus was taken up by plant.

The P_w values ranged between 40.2 and 289.2 mg/kg on Dystric Arenosol (Humic) (Fig. 1, Table 2). The P_w values after plant decreased with incubation time, however it increased with added phosphate. Thus, the addition of water-soluble phosphorus fertilizer significantly increased the P_w contents in this soil type.

After plant growing the P_{IE} values ranged between 11.1 and 50.8 mg/kg (Fig. 1, Table 2). The amount of P_{IE} was significantly less at 3 and 13 weeks incubation period than at one week of incubation except for 320 mg/kg.

The $P_{tightly}$ values ranged between 405.1 and 512.7 mg/kg (Fig. 1, Table 2). A small increase was observed with increasing incubation period. This fact, together with the decrease of P_w and P_{IE} suggests the transformation of water-soluble and isotopically exchangeable/weakly sorbed phosphate to tightly sorbed phosphate species (Nagy et al., 2019).

The P_{AL} values ranged between 68.5 and 400.4 mg/kg (Fig. 1, Table 2). The values did not change significantly with the incubation time, but increased with the amounts of P added.

The P_w and $P_{IE} + P_w$ show good correlation with P_{AL} as well as plant P_{uptake} proving that these values can be good indicators of plant available phosphorus (Table 1). This will be discussed later for all studied soil types.

3.1.2. Without plant

The P_w values ranged between 51.72 and 264.75 mg/kg on Dystric Arenosol (Humic) (non calcareous) (Fig. 2, Table 2). The P_w changes with and without plant are similar considering the effect of the incubation time and P rates. It is important to note, however, that the P_w values are always higher without plant than after plant growing, showing that plant takes up a portion of P_w.

The P_{IE} values without plant ranged between 14.63 and 96.1 mg/kg on Dystric Arenosol (Humic) (Fig. 2, Table 2). A high increase is observed with increasing phosphate doses at 13 weeks incubation. Similar to P_w values, P_{IE} values are also higher than those of with plant growing. This shows that a portion of P_{IE} is also taken up by plant.

The $P_{tightly}$ values ranged between 409.29 and 516.52 mg/kg (Fig. 2, Table 2). There is no change in increasing the incubation time, while a slight increase is observed as P doses increase. The change of $P_{tightly}$ values with and without plant are less consequent because of the competing effects of plant phosphorous uptake and the transformation of water-soluble and isotopically exchangeable/weakly sorbed phosphate to tightly sorbed phosphate species.

The P_{AL} values ranged between 76.2 and 359.1 mg/kg (Fig. 2, Table 2). The values did not change significantly with the incubation time, but increased due to the amounts of P added.

Table 4
Phosphate fractions without plant growing and after plant growing (P_{uptake}, P_w, P_{IE}, P_{AL}, $P_{tightly}$) on Rendzic Phaeozem (Hyperhumic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %.

Without plant									
	P_W			**P_{IE}**			**P_{AL}**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	13.6	14.9	11.8	6.7	11.9	9.8	33.8	36.7	25.2
40 mg/kg	19.3	23.0	20.7	10.9	12.3	12.8	53.2	56.3	41.9
80 mg/kg	31.8	36.5	28.5	12.9	18.3	14.5	82.4	94.3	73.8
160 mg/kg	79.3	67.5	51.1	29.4	28.9	23.5	134.4	119.3	99.6
320 mg/kg	159.2	128.6	126.2	60.6	52.5	52.0	296.6	240.6	222.5
	$P_{tightly}$								
	1 week	3 weeks	13 weeks						
0 mg/kg	1497.7	1491.2	1496.4						
40 mg/kg	1527.8	1522.7	1524.4						
80 mg/kg	1553.3	1543.2	1555.1						
160 mg/kg	1569.3	1581.6	1603.5						
320 mg/kg	1618.1	1657.0	1659.8						
After plant growing									
	P_{uptake}			**P_W**			**P_{IE}**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	34.3 a^{A}	39.8 a^{A}	32.7 a^{A}	12.6 a^{A}	11.9 a^{A}	10.9 a^{A}	9.0 a^{A}	6.8 a^{A}	10.0 a^{A}
40 mg/kg	59.6b^{A}	68.2 a^{A}	55.9 b^{A}	18.6 a^{A}	20.9 ab^{A}	14.8 a^{A}	14.2 a^{A}	10.2 ab^{A}	9.8 a^{A}
80 mg/kg	87.7 c^{A}	111.5 b^{A}	82.9 c^{A}	26.4 a^{A}	32.9 b^{A}	26.3 a^{A}	15.2 a^{A}	14.4 ab^{A}	14.4 a^{A}
160 mg/kg	102.5 c^{A}	137.3 bc^{A}	107.6 d^{A}	55.1 a^{A}	49.0 c^{A}	42.5 b^{A}	23.4 ab^{A}	20.9 b^{A}	19.4 a^{A}
320 mg/kg	127.8 d^{A}	172.9 c^{B}	158.3 e^{B}	128.0 b^{A}	125.3 d^{A}	113.9c^{A}	38.1 b^{A}	44.7c^{A}	41.4 b^{A}
	P_{AL}			**$P_{tightly}$**					
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks			
0 mg/kg	25.0 a^{B}	26.0 a^{B}	17.8 a^{A}	1462.1 a^{A}	1459.5 a^{A}	1464.4 a^{A}			
40 mg/kg	41.0 a^{B}	39.1 a^{B}	31.4 b^{A}	1465.6 a^{A}	1458.7 b^{A}	1477.4 b^{A}			
80 mg/kg	58.5 a^{AB}	65.7 b^{B}	51.2 c^{A}	1468.7 ab^{B}	1439.2 c^{A}	1474.4 c^{B}			
160 mg/kg	105.5 b^{A}	110.7 c^{A}	105.1 d^{A}	1496.9 b^{A}	1470.8 d^{A}	1508.6 d^{A}			
320 mg/kg	212.1 c^{A}	207.3 d^{A}	183.8 e^{A}	1544.1 c^{A}	1495.0 e^{A}	1524.5 e^{A}			

The P_{AL} values without plant are significantly higher than with plant at 0, 40 and 80 mg/kg phosphorous rates, but they are very similar at high doses (160 and 320 mg/kg). The great added P amounts (the large doses of P added) cover the effect of plant growing. The isotope exchange method, however, clearly shows this.

3.2. P species changes on Calcic Vertisol (Gleyic)

3.2.1. After plant growing

On Calcic Vertisol (Gleyic) the P_{uptake} of ryegrass was the lowest, due to the considerably lower biomass production. P_{uptake} values ranged between 1.37 and 7 mg/kg. The values increased with increasing the P rates (Fig. 3, Table 3), however, the addition of P and the increase of incubation time did not change the plant P uptake to such an extent as in the other studied soil types (Dystric Arenosol (Humic), Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol(Arenic, Humic)). In the case of Calcic Vertisol (Gleyic) the P taken up by the plant approximately doubled compared to the one week incubation. This soil was to only one to show such an increase in P uptake under the effect of long incubation period.

After plant growing, the P_w values ranged between 14.2 and 224.8 mg/kg on Calcic Vertisol (Gleyic) (Fig. 3, Table 3). The addition of water-soluble phosphorus fertilizer significantly increased the P_w contents in the soil solutions; the increase was greatest on Calcic Vertisol (Gleyic) of all soils.

The P_{IE} values ranged between 14.3 and 85.6 mg/kg on Calcic Vertisol (Gleyic) (Fig. 3, Table 3). The amount of P_{IE} was significantly less at 13 weeks incubation period than at one week of incubation.

The $P_{tightly}$ values ranged between 808.7 and 874.3 mg/kg (Fig. 3, Table 3). A small increase is observed with increasing incubation period. As mentioned earlier, this suggests the transformation of water soluble and isotopically exchangeable/weakly sorbed phosphate to tightly sorbed phosphate species (Nagy et al., 2019).

The P_{AL} values ranged between 68.4 and 434.1 mg/kg (Fig. 3, Table 3). The values did not change significantly with the incubation time, but increased to the amounts of P added.

The P_w and $P_{IE} + P_w$ show good correlation with P_{AL} but no correlation was observed with plant P_{uptake}.

3.2.2. Without plant

The P_w without plant values ranged between 20.25 and 220.1 mg/kg (Fig. 4, Table 3). The P_w value decreases with incubation time. The P_w changes with and without plant are similar as a function of incubation time and P rates. The values without plant are higher than with plant in the case of small P rates (0, 40 and 80 mg/kg), showing the phosphorus uptake by plant.

The P_{IE} values without plant ranged between 18.85 and 92.54 mg/kg (Fig. 4, Table 3). According to the paired T test probe results (at $p < 0.05$), higher P_{IE} values were obtained without plant (Fig. 4, Table 3) growing than after plant growing at 0 and 40 mg/kg P rates (Fig. 4), which can be explained by the P uptake of plant. Similar results were obtained according to Cabeza et al. (2013).

The $P_{tightly}$ values ranged between 790.32 and 890.58 mg/kg (Fig. 4, Table 3). Both the incubation time and the added P dosage increased the $P_{tightly}$ values only slightly. As mentioned earlier, this suggests the transformation of isotopically exchangeable/weakly sorbed phosphate to tightly sorbed phosphate species (Nagy et al., 2019).

The P_{AL} values ranged between 68.6 and 385.6 mg/kg (Fig. 4, Table 3). The P_{AL} values increase with incubation time. The P_{AL} changes with and without plant are nearly the same as a function of incubation time and P rates. Similarly to Dystric Arenosol (Humic), the P_{AL} values without plant are significantly higher than with plant.

3.3. P species changes on Rendzic Phaeozem (Hyperhumic)

3.3.1. After plant growing

After plant growing the P_{uptake} values ranged between 32.7 and 172.9 mg/kg on Rendzic Phaeozem (Hyperhumic) (Fig. 5, Table 4), increasing with the P rates. Considering the incubation time, the P_{uptake} at 3 weeks of incubation has a maximum. After plant growing the P_w

Table 5
Phosphate fractions without plant growing and after plant growing (P_{uptake}, P_{w}, P_{IE}, P_{AL}, $P_{tightly}$) on Calcic Gleysol (Arenic, Humic) (Different letters denote significant differences between treatments, capital letters refer to incubation time, while lowercases refer to P incubation, Tukey, $p < 0.05$ %.

Without plant

	P_{W}			P_{IE}			P_{AL}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	6.6	9.0	8.4	5.4	7.9	4.8	116.5	114.3	120.8
40 mg/kg	17.6	17.2	18.1	11.9	11.6	11.4	144.1	144.6	145.7
80 mg/kg	28.7	29.1	29.2	11.8	18.5	16.5	171.7	175.5	178.0
160 mg/kg	61.5	57.8	50.1	30.8	22.4	26.1	242.6	243.6	244.9
320 mg/kg	127.1	114.5	102.4	45.4	93.6	42.2	398.8	394.1	391.8

	$P_{tightly}$		
	1 week	3 weeks	13 weeks
0 mg/kg	1032.0	1027.0	1030.7
40 mg/kg	1054.5	1055.2	1054.5
80 mg/kg	1083.5	1076.4	1078.4
160 mg/kg	1111.6	1123.8	1127.7
320 mg/kg	1191.4	1156.0	1219.4

After plant growing

	P_{uptake}			P_{W}			P_{IE}		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	25.4 a^{A}	28.7 a^{A}	33.1 a^{A}	7.0 a^{A}	6.2 a^{A}	7.0 a^{A}	5.4 a^{A}	3.5 a^{A}	6.8 a^{A}
40 mg/kg	67.0 b^{A}	56.8 ab^{A}	61.3 a^{A}	13.9 a^{A}	12.3 a^{A}	18.1 ab^{A}	9.2 ab^{A}	8.6 ab^{A}	5.7 a^{A}
80 mg/kg	81.4 b^{A}	87.0 bc^{A}	73.7 ab^{A}	24.2 b^{A}	25.2 b^{A}	25.4 b^{A}	14.4 b^{A}	14.6 b^{A}	11.6 ab^{A}
160 mg/kg	94.0 b^{A}	110.8 c^{A}	108.5 bc^{A}	48.7c^{A}	51.4 c^{A}	47.3 c^{A}	22.8 c^{A}	25.1 c^{A}	23.5 ab^{A}
320 mg/kg	97.5 b^{A}	117.1 c^{A}	128.2 c^{A}	110.6 d^{B}	104.7 d^{B}	97.0 d^{A}	45.2 d^{A}	48.2 d^{A}	36.7 b^{A}

	P_{AL}			$P_{tightly}$		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
0 mg/kg	112.0 a^{A}	113.9 a^{A}	108.7 a^{A}	1006.2 a^{A}	1005.7 a^{A}	997.1 a^{A}
40 mg/kg	131.2 b^{A}	135.7 ab^{A}	135.1 b^{A}	994.0 b^{A}	1006.2 b^{A}	998.9 b^{A}
80 mg/kg	161.2 c^{A}	165.2 b^{A}	164.6 c^{A}	1004.0 c^{A}	997.2 c^{A}	1013.3 b^{A}
160 mg/kg	215.5 d^{A}	236.7 c^{B}	233.9 d^{AB}	1038.5 d^{A}	1016.7 d^{A}	1024.7 c^{A}
320 mg/kg	367.6 e^{A}	401.1 d^{A}	379.2 e^{A}	1110.6 e^{A}	1094.0 e^{A}	1102.2 d^{B}

values ranged between 10.9 and 128 mg/kg (Fig. 5, Table 4). The addition of water-soluble phosphorus fertilizer significantly increased the P_{w} contents in the soils but the increase was the lowest on Rendzic Phaeozem (Hyperhumic) of all soils.

After plant growing the P_{IE} values ranged between 6.8 and 44.7 mg/kg (Fig. 5, Table 4). In the case of Rendzic Phaeozem (Hyperhumic) the results were lower than in Dystric Arenosol (Humic) Calcic Vertisol (Gleyic). The amount of P_{IE} was less at 13 weeks incubation period than at 1 and 3 weeks of incubation.

The $P_{tightly}$ values ranged between 1439.2 and 1544.1 mg/kg (Fig. 5, Table 4). As a function of incubation period, the $P_{tightly}$ values were practically the same, in some P rates, a very small increase is observed.

The P_{AL} values ranged between 17.8 and 212.1 mg/kg (Fig. 5, Table 4). The values did not change significantly with the incubation time, but increased to the amounts of P added, a higher value was observed at 320 mgP/kg P added. The lowest P_{AL} values were obtained on Rendzic Phaeozem (Hyperhumic) after 13 weeks of incubation time.

The general experience on Rendzic Phaeozem (Hyperhumic), the changes in the values of all studied quantities (plant P uptake, P_{IE}, P_{w} and P_{AL}) were rather small. The ratios of the different phosphate forms in this soil practically remained unchanged; this suggests that plant took the added phosphate up.

3.3.2. Without plant

The P_{w} without plant values ranged between 11.8 and 159.23 mg/kg on Rendzic Phaeozem (Hyperhumic) (Fig. 6, Table 4), decreasing with incubation time and increasing with P rate.

The P_{IE} values without plant ranged between 6.69 and 60.65 mg/kg (Fig. 6, Table 4), showing similar tendencies to P_{w} as a function of P rate and incubation time. According to the paired T test probe results (at $p < 0.05$), higher P_{w} and P_{IE} values were obtained without plant (Fig. 5, Table 4) than after plant growing (Fig. 6, Table 4), which can be explained by the P uptake of plant. Similar results were obtained according to Cabeza et al. (2013).

The $P_{tightly}$ values without plant ranged between 1491.24 and 1659.77 mg/kg (Fig. 6, Table 4). The values changes with and without plant are similar in function incubation time and effects of P rates. $P_{tightly}$ values with and without plant are almost the same.

The P_{AL} values without plant ranged between 25.17 and 296.57 mg/kg (Fig. 6, Table 4). The values changes with and without plant are similar in function incubation time and effects of P rates. A slight decrease is observed with increasing incubation time. At the 320 mg/kg added doses of P, the values for each incubation time are approx. doubled compared to the values of 160 added P doses. Values without plant are higher than values with plants.

3.4. P species changes on Calcic Gleysol (Arenic, Humic)

3.4.1. After plant growing

After plant growing the P_{uptake} values ranged between 25.3 and 128.2 mg/kg on Calcic Gleysol (Arenic, Humic) (Fig. 7, Table 5). At higher P doses the P uptake of ryegrass increased with incubation time and with P rates.

After plant growing the P_{w} values ranged between 6.2 and 110.6 mg/kg (Fig. 7, Table 5). The addition of water-soluble phosphorus fertilizer significantly increased the P_{w} contents in the soils.

After plant growing the P_{IE} values ranged between 3.5 and 48.2 mg/kg on Calcic Gleysol (Arenic, Humic) (Fig. 7, Table 5). A small maximum was observed at 3 weeks at high P doses.

The $P_{tightly}$ values ranged between 994 and 1110.6 mg/kg on Calcic Gleysol (Arenic, Humic) (Fig. 7, Table 5).

As a summary, there were only small changes in P forms (P_{w}, P_{IE} and $P_{tightly}$) as the incubation period increased, suggesting less intense P transformation to tightly sorbed form.

The P_{AL} values ranged between 108.65 and 401.05 mg/kg on Calcic Gleysol (Arenic, Humic) (Fig. 7, Table 5). The values did not change significantly with the incubation time, but increased to the amounts of P added. The largest difference between P_{w} and P_{AL} was observed on Calcic Gleysol (Arenic, Humic), at low P_{w} values, P_{AL} values are high.

Table 6
The relative portion of added P transformed to tightly sorbed species on the studied soils as a function of P rates and incubation time (%).

Without plant									
	Calcaric Arenosol (Humic)			**Chernozem**			**Calcic Vertisols (Gleyic)**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
40 mg/kg	39.0	53.0	39.2	64.2	67.6	97.3	34.8	35.8	63.2
80 mg/kg	33.5	43.3	40.3	31.7	68.4	34.4	27.4	31.3	46.1
160 mg/kg	34.2	29.1	35.4	22.5	66.6	68.9	12.6	29.0	25.3
320 mg/kg	29.4	25.9	33.2	31.9	76.0	76.0	17.4	28.3	24.2
	Dystric Arenosols (Humic)			**Rendzic Phaeozem Hyperhumic)**			**Calcic Gleysols (Arenic, Humic)**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
40 mg/kg	29.9	24.2	27.7	75.3	78.8	70.2	56.2	70.4	59.5
80 mg/kg	41.5	38.9	29.2	69.5	64.9	73.4	64.3	61.7	59.5
160 mg/kg	33.1	25.4	25.8	44.7	56.5	66.9	49.8	60.5	60.6
320 mg/kg	29.3	29.0	14.3	37.6	51.8	51.1	49.8	40.3	59.0
After plant growing									
	Calcaric Arenosol (Humic)			**Chernozem**			**Calcic Vertisols (Gleyic)**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
40 mg/kg	54.2	52.0	58.4	46.4	55.4	98.6	50.9	24.5	29.0
80 mg/kg	49.4	41.6	53.3	22.3	60.2	97.7	39.8	22.9	30.7
160 mg/kg	0	29.5	33.3	36.7	18.3	95.6	32.9	6.4	8.7
320 mg/kg	8.0	4.3	21.5	42.4	43.4	85.1	12.2	18.1	21.7
	Dystric Arenosols (Humic)			**Rendzic Phaeozem Hyperhumic)**			**Calcic Gleysols (Arenic, Humic)**		
	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks	1 week	3 weeks	13 weeks
40 mg/kg	34.3	33.8	28.9	71.9	68.9	90.6	73.6	71.8	75.1
80 mg/kg	17.1	34.2	23.5	75.0	64.3	75.2	67.3	62.4	71.0
160 mg/kg	2.1	11.8	25.7	64.4	68.0	74.4	63.1	58.2	64.4
320 mg/kg	14.9	16.7	25.8	54.9	52.7	58.0	55.2	55.2	62.5

3.4.2. *Without plant*

The P_w without plant values ranged between 6.55 and 102.37 mg/kg Calcic Gleysol (Arenic, Humic) (Fig. 8, Table 5).

The P_{IE} values without plant ranged between 4.84 and 93.58 mg/kg. The P_w and P_{IE} value show small changes with incubation time and increases with P rates, similar to changes with plant. The values are higher than those obtained with plant.

The $P_{tightly}$ values ranged between 1027.02 and 1219.41 mg/kg on Calcic Gleysol (Arenic, Humic) (Fig. 8, Table 5). The values are almost the same for 1, 3 and 13 weeks incubation time. An increase was observed with increasing P doses. Values with plant growing are slightly higher than values without plant.

The P_{AL} values without plant ranged between 114.31 and 398.77 mg/kg (Fig. 8, Table 5). The values did not change compared to soil with plant and increased with P doses.

3.5. *Comparison of soils studied*

In this work, four soil types were studied (Dystric Arenosols (Humic), Calcic Vertisols (Gleyic), Rendzic Phaeozem (Hyperhumic), Calcic Gleysols (Arenic, Humic)). In a previous paper (Balla et al., 2021), similar data for two other soils (Calcaric Arenosol (Humic) and Chernozem) were presented. Here, the results of all six soil types are evaluated together.

The most obvious effect is that the phosphorous taken up by plant increases when the phosphate rate increases. The highest increase (4–5 times compared to the untreated soils) is observed for the soils with high humus content (Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic), while 1.5–2 times increase is for Chernozem and Arenosols. However, a significant portion of added phosphate strongly sorbs on the soil, becomes tightly sorbed species, as fast as one week incubation before sowing, thus cannot be utilized by plant (Table 6). The transformation is mainly determined by the soil type: its degree is rather high for the soils with great humus content (Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic)) and Chernozem, and the smallest for Arenosols with lowest humus content. The incubation time (Nagy et al., 2018) and P rate have smaller effects. This transformation also happens during plant growing (Table 6). This means that plant slightly influences the transformation of P into tightly sorbed form.

Table 7
The correlations between P fractions and P uptake of the plant in the case of 4 soils examined (Results of Chernozem and Calcaric Arenosol (Humic) are from our previous work (Balla et al., 2021).

	Dystric Arenosol (Humic)		**Calcic Vertisol (Gleyic)**	
	n = 30	R^2	n = 30	R^2
$P_w - P_{AL}$	$P(P_{AL}) = 1.330\ P_w + 3.596$	0.950	$P(P_{AL}) = 1.651\ P_w + 41.855$	0.974
$(P_w + P_{IE}) - P_{AL}$	$P(P_{AL}) = 1.14(P_w + P_{IE}) - 4.138$	0.944	$P(P_{AL}) = 1.237\ (P_w + P_{IE}) + 24.205$	0.958
$P_w - P_{uptake}$	$P(P_{uptake}) = 2.562\ \ln(P_W) - 4.694$	0.866		
$(P_w + P_{IE}) - P_{uptake}$	$P(P_{uptake}) = 2.696(P_w + P_{IE}) - 5.899$	0.878		
	Rendzic Phaeozem (Hyperhumic)		**Calcic Gleysol(Arenic, Humic)**	
	n = 30	R^2	n = 30	R^2
$P_w - P_{AL}$	$P(P_{AL}) = 1.565\ P_w + 13.41$	0.965	$P(P_{AL}) = 2.75\ P_w + 94.17$	0.981
$(P_w + P_{IE}) - P_{AL}$	$P(P_{AL}) = 1.211\ (P_w + P_{IE}) + 6.1$	0.965	$P(P_{AL}) = 1.967(P_w + P_{IE}) + 88.629$	0.979
$P_w - P_{uptake}$	$P(P_{uptake}) = 49.43\ \ln(P_w) - 79.89$	0.884	$P(P_{uptake}) = 31.120\ \ln(P_w) - 23.579$	0.845
$(P_w + P_{IE}) - P_{uptake}$	$P(P_{uptake}) = 54.05\ln(P_w + P_{IE}) - 118.59$	0.855	$P(P_{uptake}) = 33.65\ \ln(P_w + P_{IE}) - 46.46$	0.855
	Chernozem		**Calcaric Arenosol (Humic)**	
	n = 30		n = 30	R^2
$P_w - P_{AL}$	$P(P_{AL}) = 1.206\ P_w + 27.5$	0.981	$P(P_{AL}) = 0.942\ P_w + 183.24$	0.942
$(P_w+P_{IE}) - P_{AL}$	$P(P_{AL}) = 0.866\ (P_w+P_{IE}) + 22.8$	0.956	$P(P_{AL}) = 0.863\ (P_w + P_{IE}) + 177.1$	0.950
$P_{IE} - P_{uptake}$	$P(P_{uptake}) = 6.05\ \ln(P_{IE}) + 1.58$	0.608	$P(P_{uptake}) = 0.720\ \ln(P_{IE}) + 1.63$	0.502
$(P_w + P_{IE}) - P_{uptake}$	$P(P_{uptake}) = 5.66\ \ln(P_w + P_{IE}) - 3.49$	0.848	$P(P_{uptake}) = 0.545\ \ln(P_w + P_{IE}) - 0.924$	0.604

It is important to note that the total phosphorus content of the original soils (Table 1) increases as the humus content increases. At the same time, as seen previously when discussing the soils separately, the P_w and P_{IE} values are the smallest in the soils with the highest humus

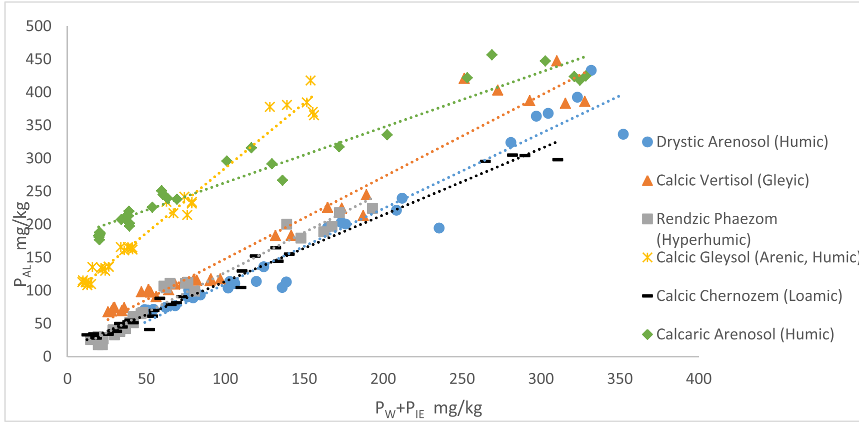

Fig. 9. Relationship between ammonium lactate soluble phosphorus (P_{AL}) and dissolved + weakly sorbed phosphate ($P_w + P_{IE}$).

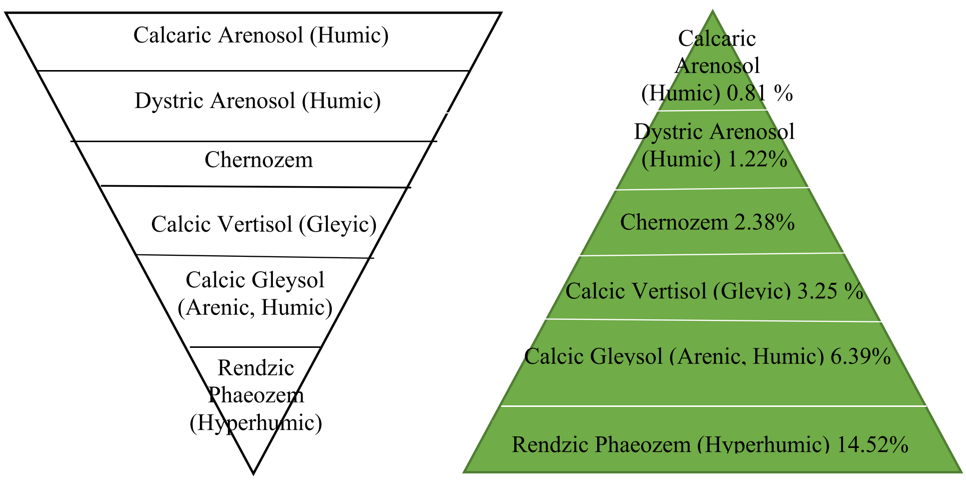

Fig. 10. Relationship between humus content (right) and dissolved phosphate (P_w) and dissolved + weakly sorbed phosphate ($P_w + P_{IE}$) (left).

content. As seen previously also that as a result of plant growing the P_w and P_{IE} values decrease, that is, plant takes up phosphorous from the soil solution and weakly sorbed phosphate. The addition of phosphate increases these values in the highest degree in the soils with high humus content, enhancing the available phosphorous quantity for plant.

Table 7 shows that a significant correlation is found between P_w and P_{uptake} as well as the sum of $P_{IE} + P_w$ and P_{uptake} of plant, except for Calcic Vertisol (Gleyic) where the mass of plant was extremely low. P_w and P_{IE} are in equilibrium with each other and both are utilized by plant. Thus, the sum of $P_{IE} + P_w$ is a better indicator of plant available P fraction than P_w or P_{IE}. P_w and P_{IE} can be also measured accurately by using heterogeneous isotope exchange examination with ^{32}P tracer.

Table 7 shows that there is a significant correlation between the P_w and P_{AL} as well as $P_w + P_{IE}$ and P_{AL} values (Fig. 9). This means that the traditionally applied quantity (P_{AL}) used for the characterization of phosphorous supply is proportional to the values obtained by heterogeneous isotope exchange (P_w and $P_w + P_{IE}$). The relations between P_{AL} and P_w and $P_w + P_{IE}$ can quantitatively be characterized by a straight line, however, the slope of the P_{AL} vs P_w and P_{AL} vs $P_w + P_{IE}$ plots strongly depends on the soil type. If the slope is equal to 1, the quantities are equal. When the slopes are higher than 1, the ammonium lactate extraction shows higher values of phosphate supply than heterogeneous isotope exchange. For the different soil types, the slope of P_{AL} vs $P_w + P_{IE}$ ranges from 0.863 to 1.967. The sum of $P_w + P_{IE}$ values is fairly close to the P_{AL} fraction, that is the slope on Arenosols and Chernozem, while for the other soils the sum of $P_w + P_{IE}$ (Calcic Vertisol (Gleyic), Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic) is significantly lower than the P_{AL}. This shows that the P_{AL} can well be applied to characterize the P supply only some soil types. The heterogeneous isotope exchange, however, can provide more general, quantitative information on different soil types.

The right side of Fig. 10 shows the humus content of the soils examined. As the green triangle narrows, the humus content in the soils decreases. The dissolved + weakly sorbed phosphate ($P_w + P_{IE}$) are on the left side triangle. As the white triangle narrows $P_w + P_{IE}$ decreases. The ($P_w + P_{IE}$) values are inversely proportional to the humus content of different soils (Fig. 10).

4. Conclusion

The studies with six different soil types prove that the radioactive method (Kónya and Nagy, 2015) is suitable for the determination of phosphate available for plants which is the sum of the dissolved (P_w) and weakly sorbed (P_{IE}) phosphate. These two quantities are in equilibrium with each other. The method can follow the effect of soil types, P rates, incubation times and plant growing. A significant correlation is between the sum of P_w and P_{IE} and P_{uptake} of plant. During plant culture, the P_w and P_{IE} values always decrease showing that plant directly utilizes these phosphate forms.

The advantage of radioactive method compared to the traditional ammonium lactate extraction method is that the radioactive method quantitatively provides the phosphate amount available for plant, while the ammonium lactate extraction gives values proportional to the phosphate available for plant, depending on the soil type (Fig. 9).

The P addition increases the P_{uptake}, however, the transformation of added phosphate to tightly sorbed phosphate species is significant and plant cultivation does not inhibit this process (Table 6). The tightly sorbed phosphate is not directly utilized by plant. The degree of this transformation is the highest for soils with great humus content (Rendzic Phaeozem (Hyperhumic) and Calcic Gleysol (Arenic, Humic). The degree of transformation to tightly sorbed phosphate is low in soils with low humus content (e.g., Arenosols). In this case, the concentration of phosphate in the soil solution (P_w) becomes great, but the P_{uptake} remains low (at most 2 times increase). This means that the dissolved phosphate is not utilized and can get into the surface water leading to eutrophication.

Our studies clearly reveal that the soils are very unique; each soil type is a complex system. The type of soil is the basic factor determining the P uptake by plants. The tightly sorbed phosphate of original soils (if its quantity is suitable) should be transformed to weakly sorbed phosphate before or during the vegetation period, e.g., by microbiological treatments.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

No data was used for the research described in the article.

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