



Pergamon

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many undefined compartments (Fardeau *et al.*, 1988), making their calculated specific activities very difficult to interpret from a theoretical point of view. Second, they considered that the added ^{32}P equilibrated with the isotopically-exchangeable P within relatively short times, which is not correct since a slow, but significant, isotopic exchange can continue over much longer times (at least 3 months, Fardeau *et al.*, 1985; Morel and Plenchette, 1994; Frossard *et al.*, 1994). Therefore the decrease in specific activity assumed to be associated to mineralization of organic-P could also be partly attributed to the dilution of ^{32}P with increasing quantities of exchangeable P, that phenomenon being even more marked in high P sorbing soils (Fardeau, 1993).

Fardeau (1993) proposed that the data obtained from the exchange kinetic experiment may be analyzed with a multicompartamental model which considers five pools of P. The most important is the central pool containing free P which contains P ions immediately available to plants, i.e. solution P plus P present on the solid but which have the same mobility as solution P. These free ions can be exchanged with any other P present in the four other pools which are

this hypothesis, by measuring changes in the isotopic exchange kinetics basic parameters and in the quantity of free phosphate ions in an alfisol incubated in the presence of two organic phosphates, and by checking whether under such conditions the P content of other compartments could be reliably calculated. In addition, the quantity of resin-extractable P was measured in the same samples.

MATERIAL AND METHODS

The surface horizon (0–10 cm) of a cultivated alfisol developed in a loamy deposit was sampled for this study. The soil was sieved (2 mm) and kept moist prior to the incubation. Coarse plant debris were removed by hand before the experiment. The major physico-chemical characteristics of this soil are displayed in Table 1.

Two organic phosphates were added: a disodium adenosine 5'-triphosphate (ATP; MW 551.1) and β -nicotinamide adenine dinucleotide (NAD; MW 663.4). Both compounds were obtained from Sigma Chemical Co. (St Louis, MO, USA)

The following treatments were considered: control

Table 2. Basic parameters of isotopic exchange kinetics at different incubation times in an alfisol after the addition of two organic phosphates

Incubation time (days)	0.7	2	8	14	21	28
	Control					
$r(1)/R$	0.21 β^*	0.20 β	0.20 β	0.21 β	0.23 χ	0.23 χ
n	0.29 α	0.26 $\alpha\beta$	0.30 α	0.29 α	0.29 α	0.30 α
cp (mg P l ⁻¹)	0.16 χ/b^{**}	0.19 χ/b	0.16 β/b	0.25 χ/a	0.25 χ/a	0.24 β/a
	Irradiated					
$r(1)/R$	0.17 χ	0.16 χ	0.18 β	0.17 β	0.19 χ	0.20 χ
n	0.30 α	0.32 α	0.27 α	0.31 α	0.29 α	0.29 α
cp (mg P l ⁻¹)	0.18 χ/b	0.16 χ/b	0.25 β/a	0.19 χ/b	0.25 χ/a	0.25 β/a
	Soil + ATP					
$r(1)/R$	0.46 α/b	0.45 α/b	0.40 α/c	0.34 α/d	0.38 α/c	0.52 $\alpha\alpha$
n	0.11 β/b	0.19 β/a	0.19 β/a	0.18 β/a	0.18 β/a	0.19 β/a
cp (mg P l ⁻¹)	1.52 β/a	1.34 β/a	1.12 α/b	0.98 α/bc	0.80 α/c	0.83 α/c
	Soil + NAD					
$r(1)/R$	0.49 α/a	0.46 α/a	ND ^{***}	0.25 β/c	0.29 β/c	0.36 β/b
n	0.14 β/c	0.18 β/c	ND	0.21 β/b	0.25 $\alpha\beta/a$	0.21 β/b
cp (mg P l ⁻¹)	1.86 α/a	1.82 α/a	ND	0.59 β/b	0.64 β/b	0.79 $\alpha\beta$

* Data followed by the same Greek letter (α , β , χ , δ) in the same column are not significantly different at the level of 5% by the test of Newman-Keuls. This indicates the effect of treatment on a given parameter ($r(1)/R$, n or cp) for a given incubation time.

** Data followed by the same Roman letter (a, b, c, d) in the same line are not significantly different at the level of 5% by the test of Newman-Keuls. This indicates the effect of incubation time on the studied parameter for a given treatment.

*** ND: not determined.

Changes in the P content of the pool of free ions ($E_{1\text{min}}$) and in the pool of P exchangeable between 1 min and 1 d ($E_{1\text{min-24h}}$)

Control and irradiated treatments. The P content of $E_{1\text{min}}$ varied between 7.6 and 12.2 mg P kg⁻¹ during the incubation in the control soil, and between 10.1 to 13.5 mg P kg⁻¹ in the irradiated soil (Table 3). The P content of the $E_{1\text{min-24h}}$ pool ranged between 51.0 and 73.0 mg P kg⁻¹ in the control soil and between 77.6 and 90.1 mg P kg⁻¹ in the irradiated treatment. $E_{1\text{min}}$ values were higher at the end of the incubation in both treatments and were slightly higher in the irradiated soil compared to the control. The higher $E_{1\text{min}}$ values observed in the irradiated sample may be attributed to the release of microbial P to the solution following the biocidal treatment (Seeling and

Zasoski, 1993; Zou et al., 1992). No particular trends were observed in the $E_{1\text{min-24h}}$ pool.

ATP and NAD treatments. The addition of organic phosphates resulted in large changes in the P content of the various pools (Table 3). The quantity of P present in the pool of free ions ($E_{1\text{min}}$) increased strongly just after the addition of both substrates to the soil reaching 33.0 mg P kg⁻¹ with ATP and 37.9 mg P kg⁻¹ with NAD, and then decreased with time. The quantity of P exchangeable between 1 min and 1 d ($E_{1\text{min-24h}}$) increased from 33.7 mg P kg⁻¹ and 59.0 mg P kg⁻¹ at the beginning of the experiment to 56.2 mg P kg⁻¹ and 70.4 mg P kg⁻¹ after 28 days of incubation in soils amended with ATP and NAD, respectively.

$E_{1\text{min}}$ values were significantly higher in the soils amended with NAD and ATP, showing that these

Table 3. Changes in the quantities of isotopically exchangeable P (mg P kg⁻¹ soil) after different incubation times in an alfisol following the addition of two organic phosphates

Incubation time (days)	0.7	2	8	14	21	28
P pool	Control					

labelled phosphate ions. When a quantity R of $(E_{1000-900})$ was calculated as the difference between the

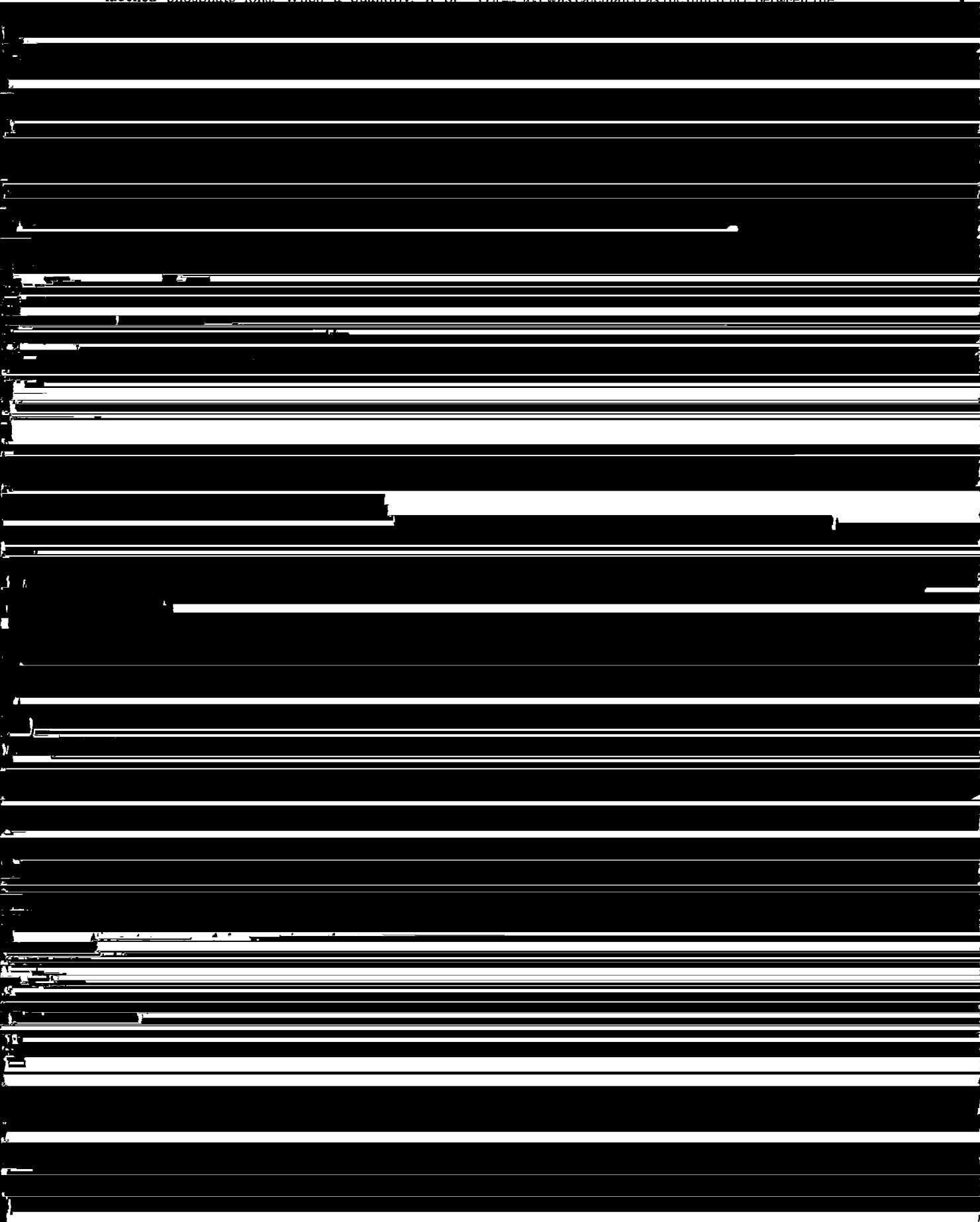


Table 4. Changes in resin-extractable P (mg P kg⁻¹ soil) after different incubation times in an alfisol following the addition of two organic phosphates

Incubation time (days)	0.7	2	8	14	21	28
Control	42.8 χ a	40.2 β a	30.7 χ b	42.4 β a	43.6 χ a	38.1 χ a
Soil + ATP	85.6 α a	85.1 α a	74.2 α a	75.1 α ab	67.8 α b	81.4 α ab
Soil + NAD	89.5 α a	84.8 α ab	76.2 α bc	71.6 α c	71.3 α c	77.1 α bc
Irr [*] . control	43.0 χ b	39.4 β b	50.0 β a	38.8 β b	38.2 χ b	31.0 χ c
Irr. + ATP	68.7 β bc	93.2 α a	79.9 α b	70.0 α bc	59.1 β c	59.8 β c
Irr. + NAD	73.9 β ab	88.3 α a	ND ^{***}	69.1 α b	56.2 β b	56.7 β b

* See Table 2 for the key for the Greek and Roman letters.

** Irr.: irradiated soil.

*** ND: not determined.

organic P compounds were mineralized. However no significant differences were observed between $E_{1 \text{ min-24 h}}$ values suggesting that either no mineralized P reached this pool, or more probably, that the $E_{1 \text{ min-24 h}}$ determinations were not reliable enough.

Changes in the resin-extractable P (P_{resin})

Control and irradiated treatments. P_{resin} remained constant during the incubation in the control soil, except for a low value observed after 8 days (Table 4). Similarly P_{resin} was constant in the irradiated soil without applied P except for a higher value noted after 8 days and a lower value observed after 28 days

mineralization was microbiologically-mediated and could not solely be attributed to phosphatase activities. The mineralized P could be of two origins: either this was the remains of the initially introduced but not yet degraded ATP or NAD, or it resulted from the mineralization of newly-synthesized microbial P built up during the early phases of the incubation.

Calculation of the mineralization rate of added organic P

The main hypothesis is that the mineralization

Table 5. Net quantities of P mineralized in an incubated alfisol amended with organic P (results are expressed in mg P kg⁻¹).

Days of incubation	0.7	2	8	14	21	28
$\delta 10cp$						
ATP	13.6	11.8	9.6	8.2	6.4	6.7
NAD	17.0	16.6	ND	4.3	4.8	6.3
δE_{1min}						
ATP	25.4	22.2	20.7	21.5	13.3	8.3
NAD	30.3	30.6	ND	15.7	14.2	14.6
δE_{24h}						
ATP	8.1	49.4	40	41.5	14.9	13.5
NAD	38.3	71.6	ND	38.2	57.6	34
δP_{resin}						
ATP	42.8	42.3	31.3	32.2	25.0	38.5
NAD	46.7	42.0	33.4	28.7	28.5	34.2

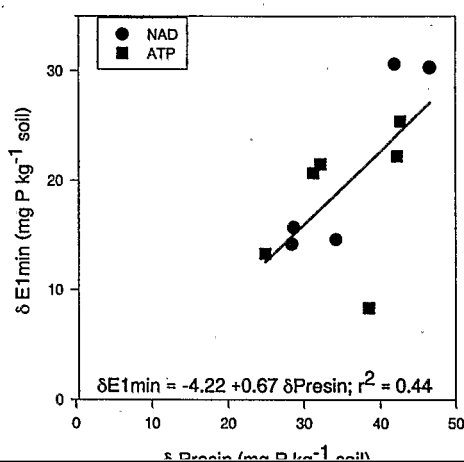
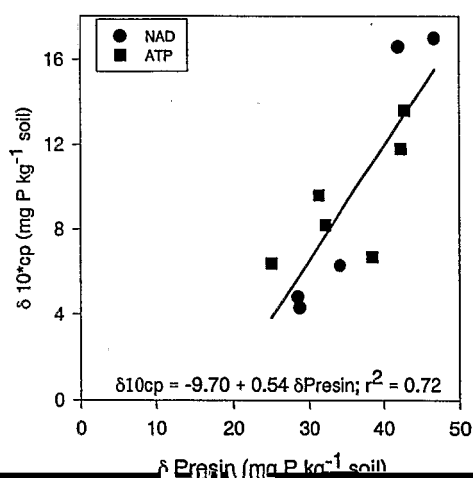
$\delta 10cp$: quantity of mineralized P remaining water soluble; δE_{1min} : quantity of mineralized P remaining in the pool of free ions; δE_{24h} : quantity of mineralized P remaining in the pool of P isotopically-exchangeable within 24h; δP_{resin} : quantity of mineralized P remaining resin-extractable.

ND: not determined.

3 weeks and increased again during the last week of the experiment to reach values close to 34 to 41% of the added P. By contrast widely-fluctuating values showing no particular trends were observed when E_{24h} was used to calculate the amount of mineralized P.

A highly significant linear correlation was observed between the quantities of mineralized P calculated with water-extractable P ($\delta 10cp$) and resin-extractable P (P_{resin}) (Fig. 1). Similarly a significant positive relation was noted between the quantities of mineralized P calculated with the quantity of P isotopically-exchangeable within 1 min (δE_{1min}) and resin-extractable P (δP_{resin}) (Fig. 2). However on this graph one point lay outside of the correlation with too low a δE_{1min} value. This point originated from the analysis of the last sampling date of the experiment in the presence of ATP where the

resin extraction still showed that high quantities of P were mineralized, while the E_{1min} value was at its lowest. The correlation became highly significant when this outlier was taken out. No relationship was observed between δE_{24h} and δP_{resin} . The relations observed between $\delta 10cp$ and δP_{resin} and between δE_{1min} and δP_{resin} suggest that they give information on the same process, whereas the absence of relation existing between δE_{24h} and δP_{resin} suggests that δE_{24h} can not be used to assess the quantity of mineralized P. Nevertheless, our results give some indications on the fate of P following its mineralization. For instance just after the addition (0.7 day of incubation) of NAD 17.0% of the added P was water soluble, 13.3% was sorbed but still had the same mobility as water-soluble ions, 16.4% was sorbed more tightly on soil surfaces but was still resin-extractable and 53.3% was not recovered. By the end



of the incubation these figures had changed as follows: 6.3% of the added P was water soluble, 8.3% was sorbed but still had the same mobility as water-soluble ions, 19.6% was sorbed more tightly on

also been reported in high P-sorbing soils where the determination of very low concentrations of P in the solution is delicate and where some of the basic assumptions underlying this approach, such as the

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