Chapter 3

Results and Discussion

3.1) Inorganic phosphate fractions

3.1.1) DMT-HFO fraction

The role of the DMT-HFO is to act as a phosphate sink simulating a plant root in the soil. This will cause the equilibrium to shift to the phosphate sink, resulting in the mobilisation of solid phase phosphate to replenish the depleted phosphate in solution.

The origin of phosphate in solution can be de-adsorbed phosphate, phosphate from phosphate minerals and mineralised phosphate. It is however important to note that it is not possible to distinguish from which source the phosphate in solution originated. The fractional composition of the soil solution and the expected change in composition over time is at best an educated guess based on the facts that the release kinetics of some sources are slower or faster than that of others.

According to **Table 3.1** approximately the same amount of phosphate desorbed from the different phosphate treatments after 24 hours, indicating that the phosphate released from R, R75 and R150 after 24 hours, originated from the same solid phase with approximately the same release kinetics. Freese *et al*, (1995), found that for more or less 50 hours the phosphate release rate of 6 Belgium soils with a high phosphate saturation was higher than the transport rate through the DMT causing phosphate backup. In this study the amount of phosphate desorbed after 24 hours from all the treatments, were considerably less than that reported by Freese *et al.*, (1995), the amount of phosphate released from the different treatments in the first 24 hours varied form 2.06 –2.83 mg kg^{-1} compared to 27 to 130 mg kg^{-1} reported by Freese *et al.*, (1995). The rate constants of phosphate release of all the treatments were also lower that the rate constant of phosphate transport through the DMT reported. Therefore it is unlikely that phosphate "backup" occurred in this study.

Table 3.1. The effects of the different phosphate treatments on the amount of phosphate extracted with DMT-HFO over time.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	**x**[1]2.06 **ab**[2]	**x** 2.83 **b**	**x** 2.80 **b**
7	**x** 2.89 **b**	**y** 6.09 **a**	**z** 7.64 **a**
14	**x** 2.87 **b**	**x** 2.50 **bc**	**y** 8.20 **a**
21	**x** 2.05 **ab**	**xy** 1.31 **c**	**y** 0.85 **b**
28	**x** 1.00 **a**	**xy** 1.52 **bc**	**y** 1.52 **b**
35	**x** 1.07 **a**	**x** 1.66 **bc**	**x**1.21 **b**
42	**x** 1.26 **a**	**x** 1.47 **bc**	**x** 1.74 **b**
56	**x** 0.93 **a**	**y** 2.23 **bc**	**y** 2.81 **b**

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$).

2 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

Figure 3.1 describes the cumulative phosphate desorbed over the 56-day period for the different treatments. After 56 days, a total of 24, 16.8 and 12.1 mg P kg^{-1} were released from R150, R75 and R respectively.

Although about twice as much phosphate was released from R150 than from the control (R) treatment, it represented only 8 % of the phosphate applied to R150 and 6 % of R75.

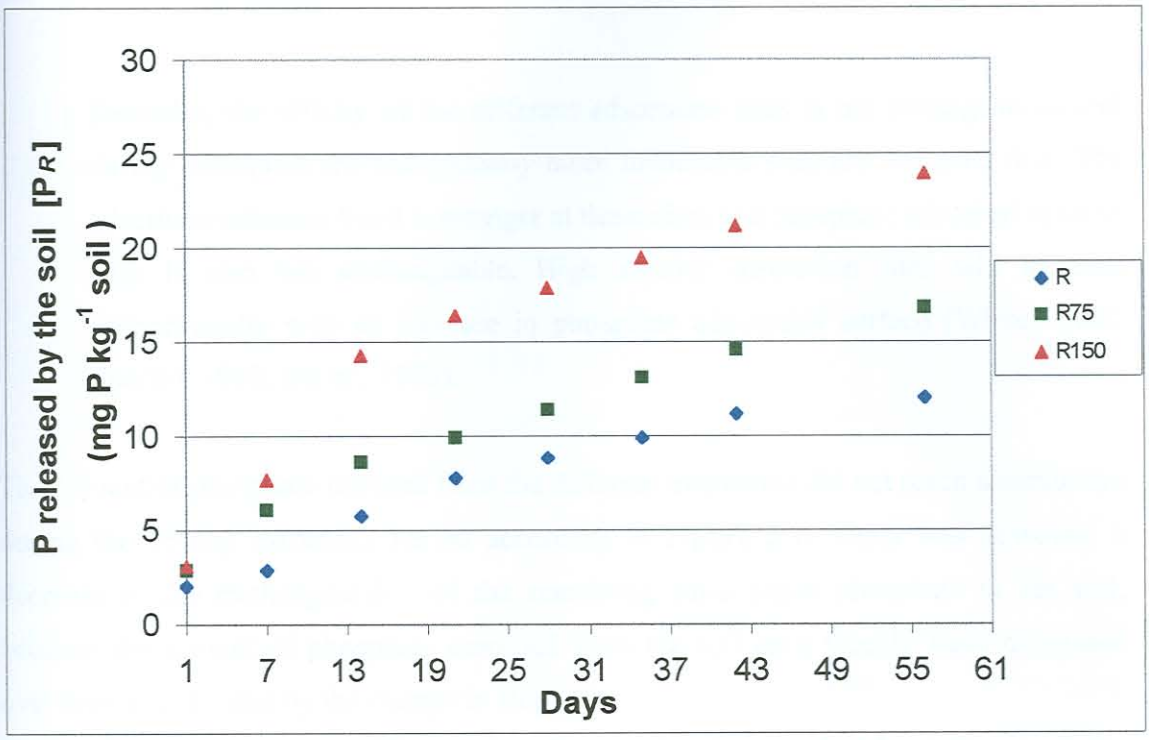

Figure 3.1. Cumulative P released from R, R75 and R150 over time.

The small amount of phosphate recovered from the treated soil is the result of the low exchangeability of sorbed phosphate from this soil.

1. Strengite and variscite are the predominant phosphate minerals present in acid soils and have a low solubility that decrease with a decrease in pH as illustrated in **Figure 1.5**. Because of the low solubility of strengite and variscite it will take a long time to replenish the phosphate removed from solution.
2. Phosphate adsorbed by aluminium and ferric oxy hydroxides also has a low exchangeability because of the strong bonded binuclear complexes that form with aluminium and ferric oxy hydroxides. The recovery of added phosphate also decreases with an increase in the adsorption surface of a soil. This is due to two factors; firstly, with an increase in saturation of the adsorption surface steric interference increases between neighbouring adsorbed species, which increase the exchangeability of these adsorbed species. Therefore the larger the adsorption surface the more phosphate can adsorbed before steric interference will occur.

Secondly, the affinity of the different adsorption sites is not homogeneous and during adsorption the energetically more favourable sites are occupied first. The adsorbate-substrate bond is stronger at these sites and phosphate adsorbed at these sites is also less exchangeable. High affinity adsorption sites will increase proportionally with an increase in phosphate adsorption surface (White, 1980, Sposito, 1989, Atkins, 1999).

The amount of phosphate released from the different treatments did not reach a maximum during the 56-day extraction period according to **Figure 3.1.** There was however, a decrease in the exchangeability of the remaining solid phase phosphate in the soil, because the amount of phosphate desorbed from the soil on a weekly basis decreased over time as indicated by the change in slope.

Figure 3.1 shows that two different phosphate pools, distinguished by different release kinetics participated in the desorption process of R75 and R150. A more labile phosphate pool (pool A), characterised by the higher phosphate release rate compared to the second phosphate pool (pool B), dominated the phosphate release kinetics during the first 14 days. The desorption rate of the control treatment decreased more gradually over time and did not show the distinct change in the phosphate release kinetics that R75 and R150 showed.

An explanation why the release kinetics of the control treatment did not show the same distinct change could be that the control treatment contained very little adsorbed phosphate. According to Hingston *et al.* (1974), and Lookman *et al.*, (1995), the faster release kinetics is usually associated with the desorption of adsorbed phosphate directly in contact with the soil solution.

The amount of phosphate released from R150 after 14 days was more than double (111%) the amount of phosphate released by the control treatment, while 49% more phosphate desorbed from R75. After ±14 days a less exchangeable phosphate pool (pool B) became more dominant, which lead to a decrease in the phosphate release rates of R R75 and R150. The slower release kinetics of Pool B is probably the result of slow dissolution kinetics of phosphate minerals (such as strengite and variscite) and/or the

slow diffusion kinetics of phosphate from interior adsorption sites inside sequi-oxides (Lookman *et al.* 1995).

3.1.1.1) Kinetics of phosphate desorption.

In this study, first order kinetics was used to describe the phosphate release kinetics of the soil. It is however important to note that, in a heterogeneous system like soil, the phosphate release kinetics would not necessarily follow first order kinetics exactly because of the different solid phases with different release kinetics which are responsible for the phosphate released in solution. It is therefore more correct to refer to it as pseudo first order kinetics.

As discussed earlier, **Figure 3.1** showed two different phosphate pools, distinguished by different release kinetics indicated by the change in the slopes of curves. The transition between the two phosphate pools were probably a more gradual process, with both phosphate pools participating from the start of the experiment. However because the release rate of pool A was higher than that of pool B, it dominated the release kinetics in the first 14 days and as the pool A became depleted Pool B became more dominant. To obtain one kinetic equation that describes simultaneous phosphate release from two phosphate pools with different release kinetics a two-component first order model was used.

The mass balance equation for the total exchangeable solid phase phosphate (SP_{total}) in the soil at time t = 0 is

$$SP_{total_0} = SP_{A_0} + SP_{B_0} \qquad \textbf{(3.1)}$$

where SP_{A_0} = Initial amount of phosphate in Pool A

SP_{B_0} = Initial amount of phosphate in Pool B

The mass balance equation at time *t* will therefore be:

$$SP_{total(t)} = SP_{A(t)} + SP_{B(t)} \qquad \textbf{(3.2)}$$

If the decrease in the different phosphate solid phases over time follow first order kinetics, the integrated rate laws for Pool A and B will be:

$$SP_{A(t)} = SP_{A_0}\, e^{-k_1 t} \qquad \textbf{(3.3)}$$

where k_1 = conditional first order rate constant (day^{-1}) for phosphate desorption from Pool A.

$$SP_{B(t)} = SP_{B_0}\, e^{-k_2 t} \qquad \textbf{(3.4)}$$

where k_2 = conditional first order rate constant (day^{-1}) for phosphate desorption form Pool B.

The total solid phase phosphate ($SP_{total(t)}$) left in the soil at time ***t*** will be given by:

$$SP_{total(t)} = SP_{A_0}\, e^{-k_1 t} + SP_{B_0}\, e^{-k_2 t} \qquad \textbf{(3.5)}$$

The amount of phosphate **released** at time ***t*** will be the difference between initial amount of phosphate in the respective pools and amount of phosphate in the pools at time ***t:***

$$P_{R(t)} = SP_0 - SP_0\, e^{-k\, t} \qquad \textbf{(3.6)}$$

The total amount of phosphate released at time *t* can therefore be expressed as:

$$\begin{aligned} P_{R(t)} &= SP_{A_0} - SP_{A_0}\, e^{-k_1 t} + SP_{B_0} - SP_{B_0}\, e^{-k_2 t} \\ &= SP_{A_0}\,(1 - e^{-k_1 t}) + SP_{B_0}\,(1 - e^{-k_2 t}) \end{aligned} \qquad \textbf{(3.7)}$$

(Snoeyink & Jenkins, 1980, Olson & Shuman, 1985, Freese *et al*, 1995, Lookman *et al*, 1995, Atkins, 1999).

Using mass balance principles the contribution of SP_A and SP_B to $P_{R(t)}$ as a function of time can be represented as follows:

Contribution of SP_B to $P_{R(t)}$ as a function of time:

$$\alpha_A = \frac{\left(1 - e^{k_1 t}\right) SP_{A_0}}{P_{R(t)}} \tag{3.8}$$

$$SP_{A(t)} = \alpha_A P_{R(t)} \tag{3.9}$$

Contribution of SP_B to $P_{R(t)}$ as a function of time:

$$\alpha_B = \frac{\left(1 - e^{k_2 t}\right) SP_{B_0}}{PR_{total(t)}}. \tag{3.10}$$

$$SP_{B(t)} = \alpha_B P_{R(t)} \tag{3.11}$$

The release kinetics of the soil under the influence of the DMT-HFO can be represented as follows:

$$SP \xrightarrow{k_R} P_{sol} \xrightarrow{k_T} P_{HFO} \tag{3.12}$$

Where SP = Solid phase phosphate

P_{sol} = phosphate in solution

P_{HFO} = phosphate adsorbed by HFO

k_T = rate constant of phosphate transport through the membrane

(0.09 ± 0.01 h^{-1}, Freese *et al*, 1995)

k_R = is the rate constant of phosphate release

In this study the rate constant of phosphate release (k_R) was lower than the rate constant of phosphate transport across the dialysis membrane (k_T), therefore phosphate release was the rate-determining step. It was therefore assumed that the rate constant of phosphate release from the soil was equal to the rate constant of phosphate adsorption (k_A) by the DMT-HFO.

The rate constant of phosphate adsorption (k_A) by the DMT-HFO was obtained from a plot of the natural logarithm (ln) of the phosphate adsorption by the DMT-HFO over time

(with the slope as k_A), this is illustrated in **Figures, 3.2, 3.3** and **3.4**. The desorption data was split into two parts to obtain the rate constants of the different pools. The desorption data of the control treatment was also divided at day 14 for the sake of comparison.

The SP_{A_0} of R, R75 and R150 were taken as the amount of P adsorbed (by the HFO) after 14 days. The SP_{B_0} of R, R75 and R150 were then obtained by solving **Equation 3.7** in terms of SP_{B_0} because it was the only unknown left in the equation.

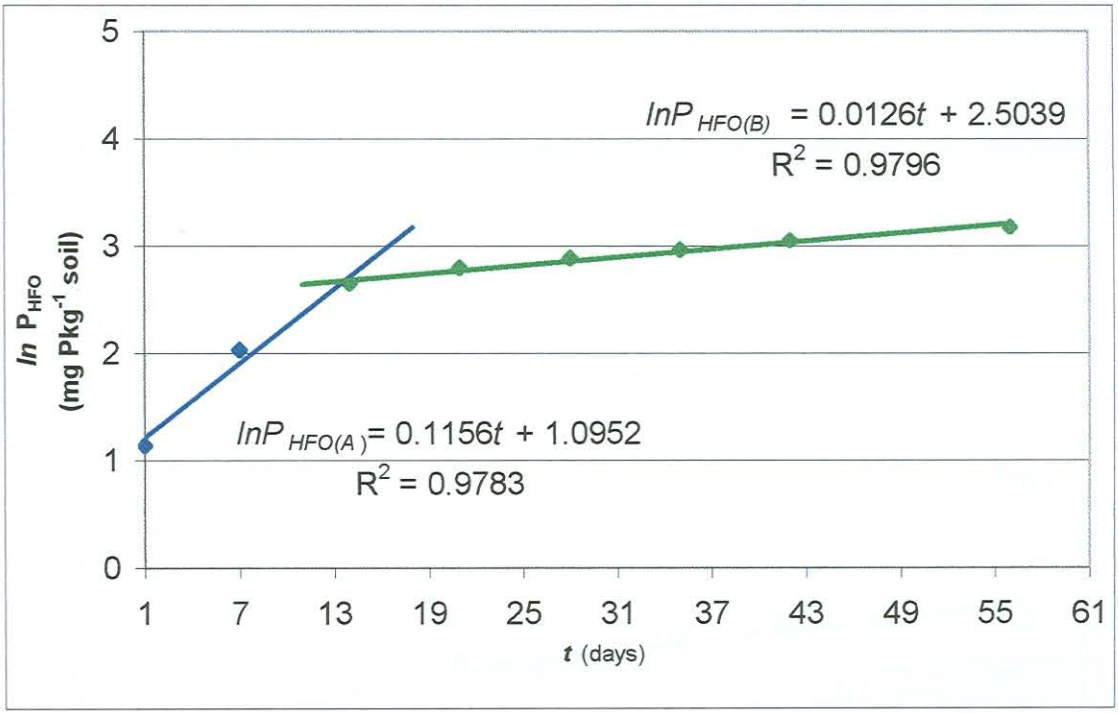

Figure 3.2. The natural logarithm (ln) of the phosphate released from R150 over time

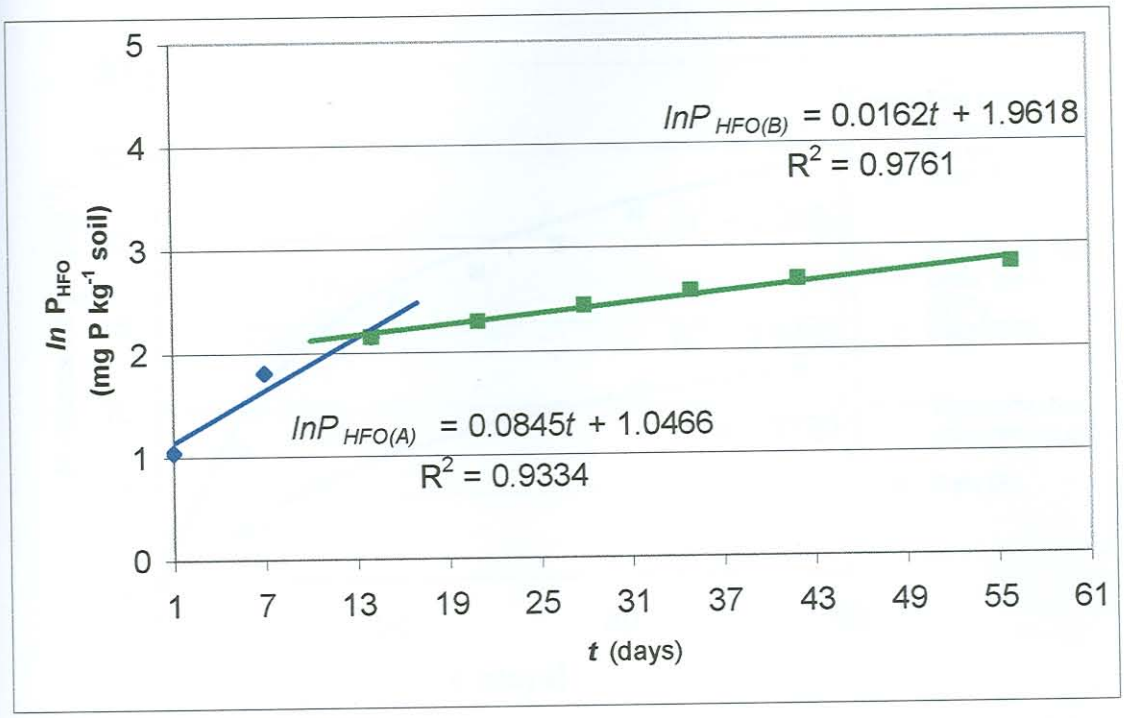

Figure 3.3. The natural logarithm (ln) of the phosphate released from R75 over time

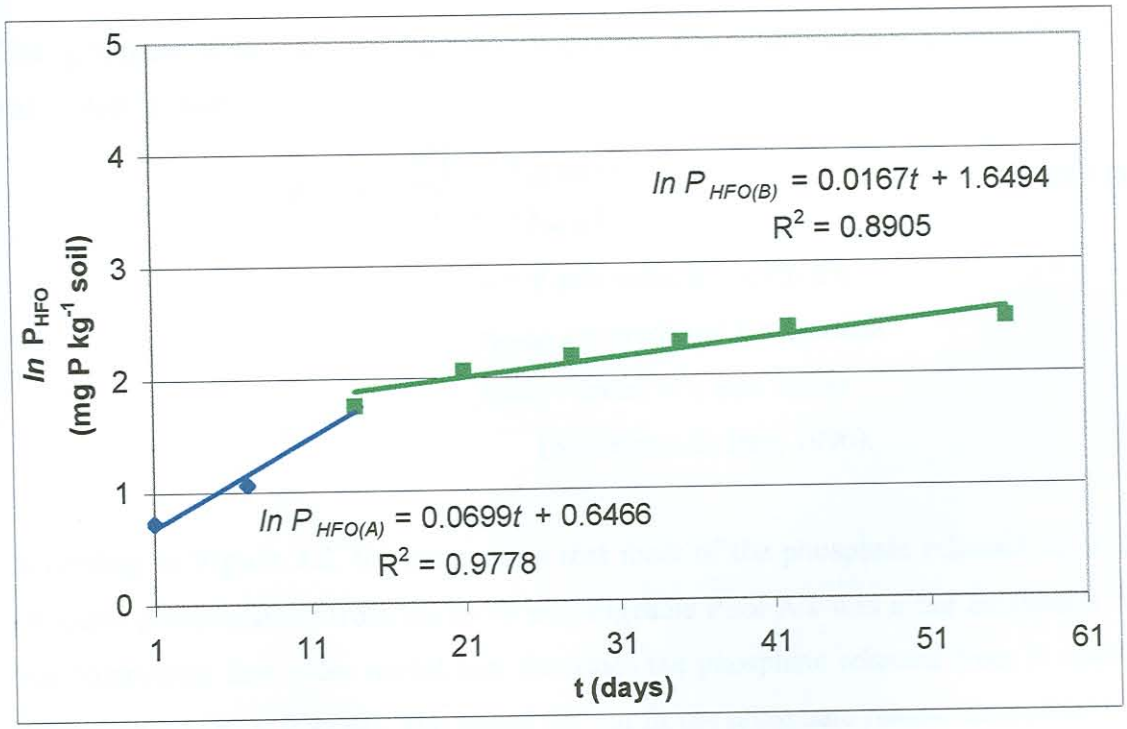

Figure 3.4. The natural logarithm (ln) of the phosphate released from R over time

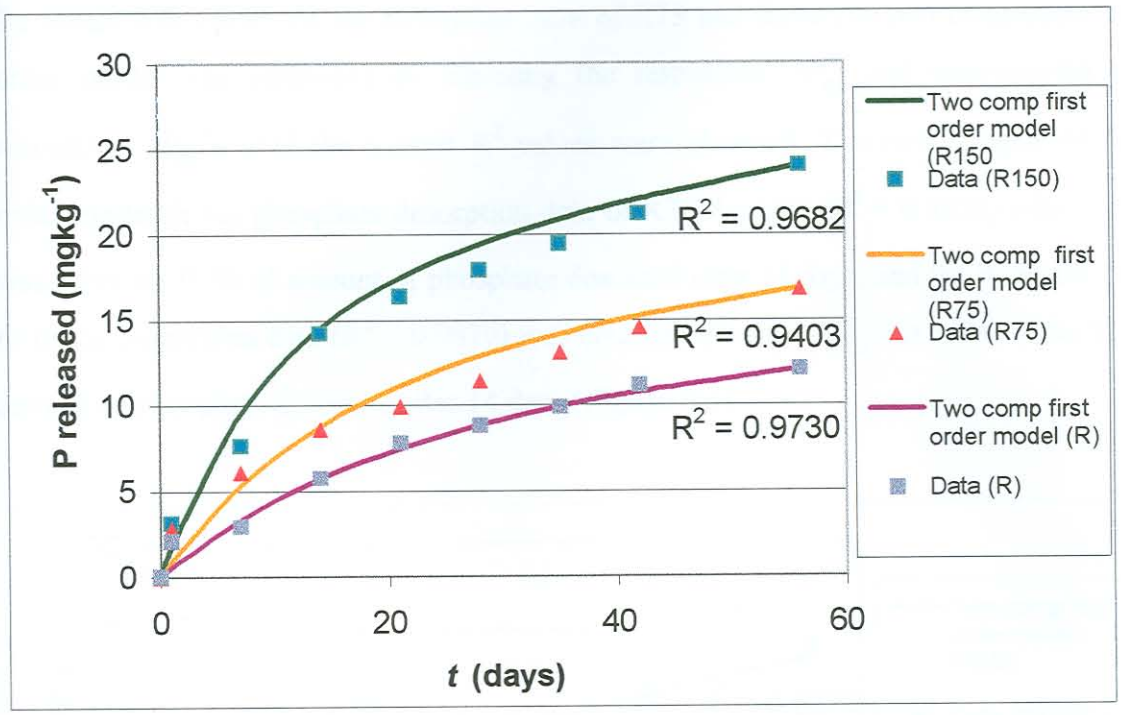

Figure 3.5. Comparison of the desorption data of the different treatments over time with the two-component first model

The goodness-of-fit (R^2) of the two-component first order model to the data, was calculated as follows:

$$R^2 = 1 - \frac{\sum (y_i - y_{predicted})^2}{\sum (y_i - y_{mean})^2} \qquad \textbf{(3.13)}$$

y_i = y axis value at the ith day

$y_{predicted}$ = predicted y axis value

y_{mean} = mean of y axis values

(Schulthess & Dey, 1996).

According to **Figure 3.5**, the assumption that most of the phosphate released up to day 14, came predominantly from the more exchangeable Pool A's was a fair estimation. The two-component first order model best describes the phosphate released from R over the 56-day period (R^2 = 0.9730). The model did not fit the phosphate release data of R75 and R150 that well (R^2 = 0.9403 and R^2 = 0.9682 for R75 and R150 respectively).

To obtain a fit better for the desorption data of R75 and R150 the two component first order model was optimised by adjusting the respective SP_{A_0} and recalculating the respective SP_{B_0}'s until the highest R^2 values were obtained. The two component first order model fit the phosphate desorption data of R75 the best (R^2 = 0.9528) when SP_{A_0} was taken as 78 % of amount of phosphate desorbed after 14 days, and for R150 the best fit to the desorption data (R^2 = 0.9870) was obtained of when SP_{A_0} was taken as 86 % of amount of phosphate desorbed after 14 days (**Figure 3.6**).

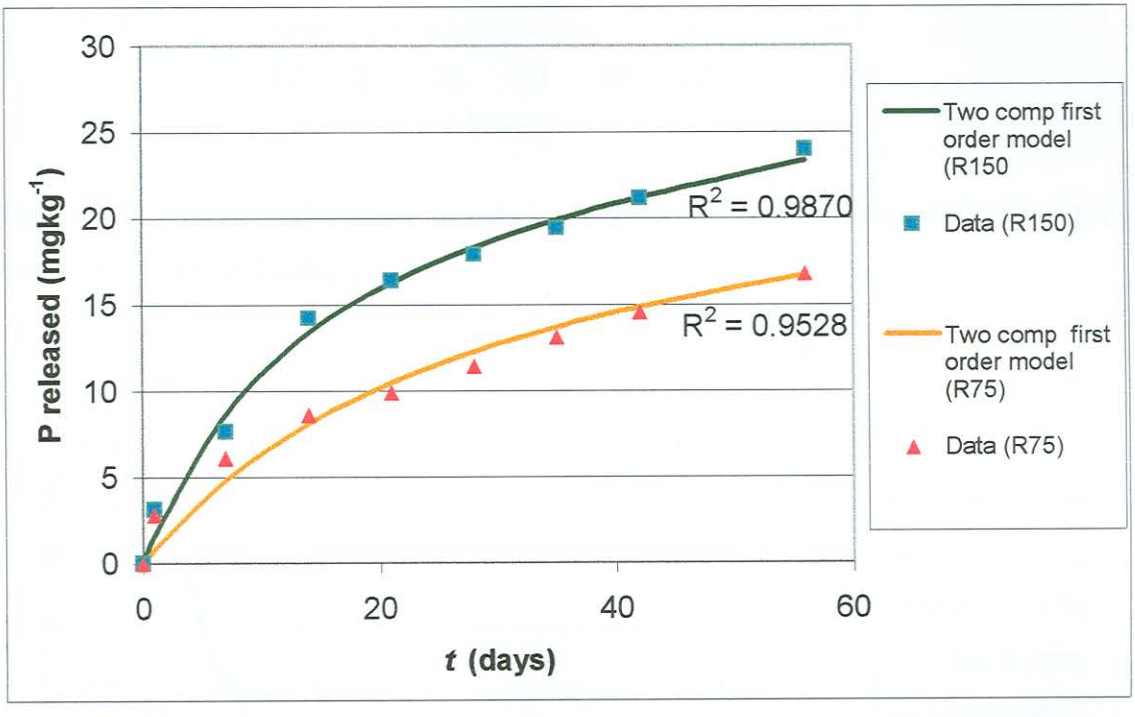

Figure 3.6. Comparison of the desorption data of R75 and R150 over time with the two-component first model using the second estimation of the respective pool A's.

In **Figure 3.7,** using **equations 3.9** and **3.11,** the release kinetics of the two phosphate pools of the different treatments were plotted separately to show the different release kinetics of each pool over the 56-day period. In **Figure 3.8** the desorption rates of the two phosphate pools of the different treatments were plotted as a function of time.

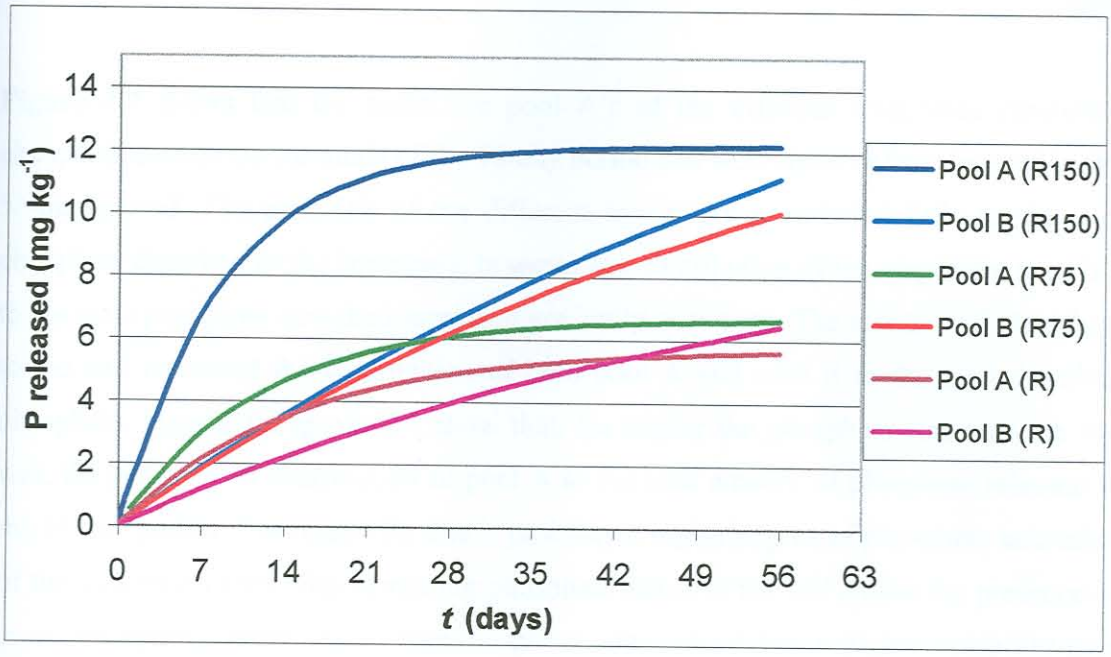

Figure 3.7. Simulated phosphate release from respective pool A and pool B of the different treatments over the 56-day period.

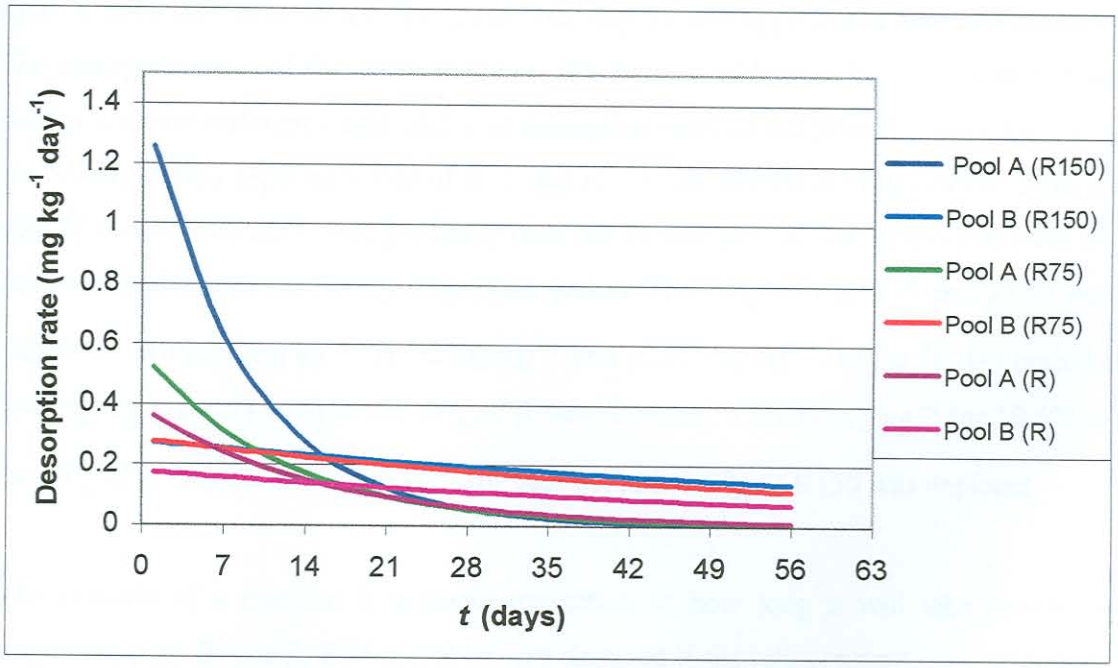

Figure 3.8. Desorption rates of the respective pool A's and pool B's of the different treatments over the 56-day period.

Figure 3.7 shows that the respective pool A's of the different treatments controlled phosphate desorption for much of the 56-day period and were more or less depleted in the 56-day period. The pool B's of the different treatments contributed little to the total phosphate desorbed in the beginning, however the contribution of the respective pool B's to the total phosphate desorbed increased gradually over time. The addition of phosphate to the soil increased the contribution of both pool A and pool B to the total desorbed phosphate. However **Figures 3.7** show that, the higher the phosphate status of the soil was, the greater was contribution of pool A to the total amount of phosphate released in the 56-day period. The reason for this is probably a higher degree of phosphate saturation of the adsorption sites with increasing phosphate status of the soil and/or the presence of more amorphous ferric and aluminium phosphates, which has a higher solubility than crystalline ferric and aluminium phosphates. **Figure 3.8** shows that the desorption rate of the pool A of R150 was initially higher than the desorption rates of the pool A's of the other treatments. The desorption rates of the respective pool A's decreased rapidly in the first 14 days and were almost the same after day 21 and approaches zero after day 35. The desorption rates of the respective pool B's were considerably lower than that of the pool A's of the different treatments The desorption rates of the pool B's were similar in the 56-day period especially that of R75 and R150, the desorption rates of the different pool B's decreased also more gradually over the 56-day period. The respective Pool B's were not depleted in the 56-day desorption period. The SP_{B_0} 's of R, R75 and R150 were calculated at 10.60 mg kg $^{-1}$ 16.92 mg kg $^{-1}$ and 22.07 mg kg $^{-1}$. In the 56-day period ± 6.44 mg kg $^{-1}$ (or 53.1 %) of the SP_{B_0} of R was depleted, ± 10.09 mg kg $^{-1}$ (or 59.6%) of the SP_{B_0} of R75 and ± 11.17 mg kg $^{-1}$ (or 50.6 %) of the SP_{B_0} of R150 was depleted.

The half-life of a reaction is a useful indication of how long it will take before the respective pool B's of R, R75 and R150 are depleted if the rate constants do not change. The half-life for a reaction, following first order kinetics is derived as follows.

At $t_{1/2}$ of the respective pool B's:

$$SP_{B_{(t)}} = \frac{1}{2} SP_{B_0} \qquad \textbf{(3. 14)}$$

Using the integrated rate law of a first order reaction (**Equation 1.21**), **Equation 3.14** can be rewritten as follows:

$$\ln\left(\frac{\frac{1}{2}SP_{B_0}}{SP_{B_0}}\right) = -k_2\, t_{1/2}$$

$$\ln\left(\frac{1}{2}\right) = -k_2\, t_{1/2}$$

$$t_{1/2} = \frac{ln2}{k_2} \qquad \textbf{(3. 15)}$$

(Snoeyink & Jenkins, 1980, Atkins, 1999).

Using **equation 3. 15** the half-lives of the pool B's were calculated at ± 41.5 days for R, ± 43 days for R75 and that of R150 at ± 55 days. The addition of 75 mg kg^{-1} prolonged the half-life of Pool B of the studied soil only slightly. The 150 mg kg^{-1} treatment however prolonged the half-life of Pool B of the studied soil with ± 13.5 days. Although it will take only 55 days for pool B of R150 to halve it would take ± 1500 days or 4 year before the pool B of R150 is depleted at the current rate constant of phosphate release for the pool B of R150!

Figure 3.9 shows the release kinetics of the two phosphate pools extrapolated over a 600-day period, the critical assumptions here were that the rate constants do not change over time and no phosphate is added to the soil.

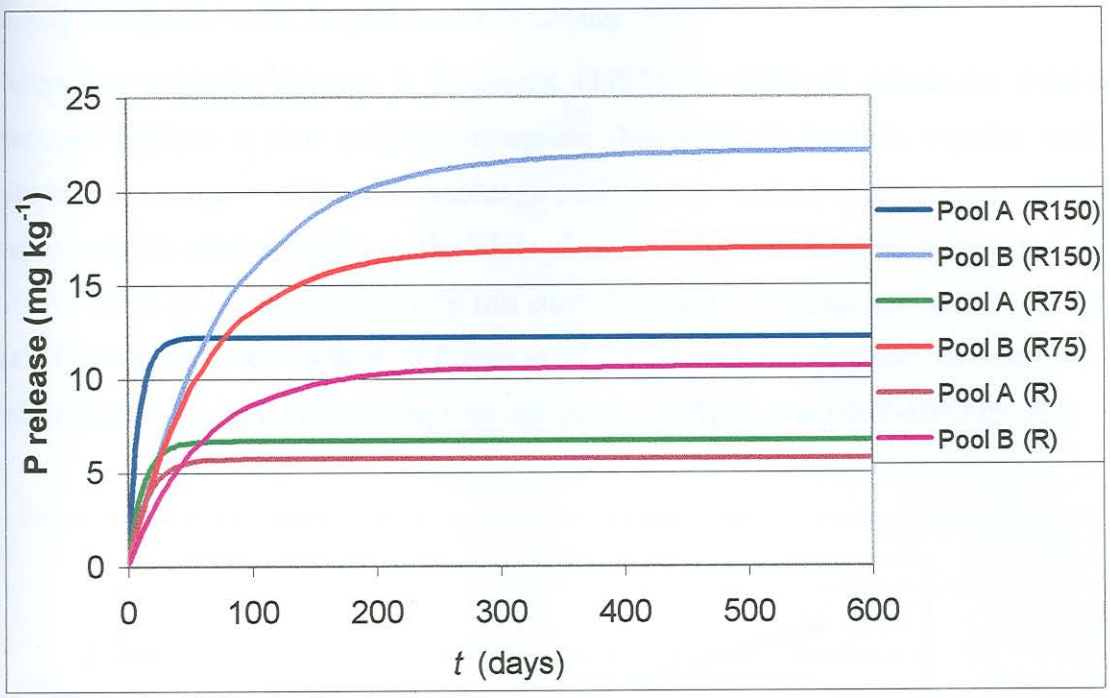

Figure 3.9. Simulated phosphate release from respective pool A and pool B of the different treatments extrapolated over a 600-day period.

Figure 3.9 shows the importance of the less exchangeable pool B in the long term (>56 days) phosphate desorption kinetics of the studied soil, eventually the less exchangeable pool B will control phosphate desorption kinetics. In the 56-day window, the effect of the addition of phosphate on the desorption kinetics of the less exchangeable pool B were not that evident, extrapolating the desorption kinetics to 600 days gave an indication of the long term effect of applied phosphate on the desorption kinetics of the pool B of the studied soil.

If the assumption is true that pool A represents predominantly adsorbed phosphate and pool B represents predominantly mineral phosphate and phosphate that diffused into occluded adsorption sites, then this study also gives an insight into the rates at which added phosphate diffuses into occluded adsorption sites and transforms into strengite and variscite in this particular soil. During the five-month incubation 73.9 mg kg^{-1} P (or 98.6%) of the 75 mg kg^{-1} phosphate applied and 143.4 mg kg^{-1} (or 95.6%) of the 150 mg kg^{-1} phosphate was transformed to mineral P or diffused into occluded adsorption sites

3.1.2) Inorganic $NaHCO_3$ (HCO_3-Pi) fraction

According to Hedley Stewart & Chaunhan, (1982), the HCO_3-Pi reflects the short-term seasonal changes in plant available phosphate. The HCO_3-Pi fraction, together with the phosphate extracted with anion exchange resin in the method of Hedley *et al* (1982), represents the plant available or the labile phosphate forms in the soil (Bowman & Cole, 1978 , Olsen & Watanabe 1957). In this study the resin extraction was substituted by the DMT-HFO extraction method of Freese *et al* (1995), and it is therefore assumed that the labile phosphate pool would be represented by the HCO_3-Pi plus DMT-HFO fraction.

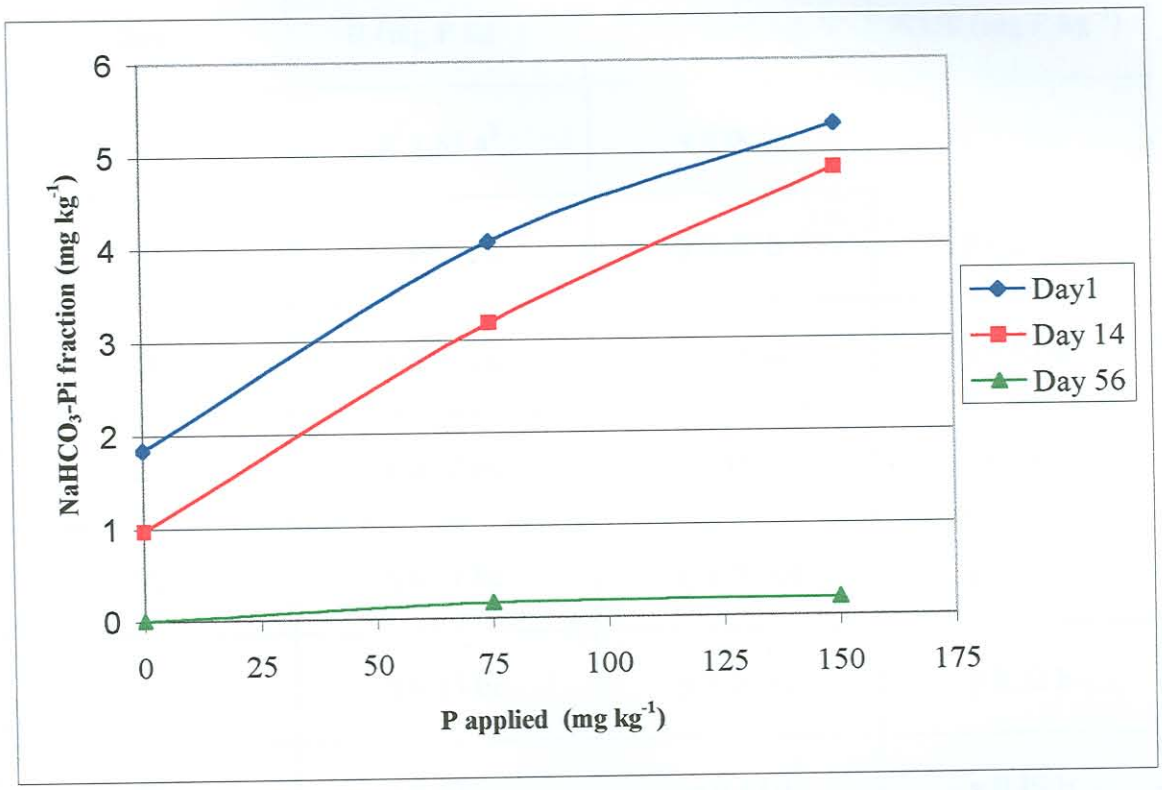

Figure 3.10. The change in the HCO_3-Pi fraction as influenced by phosphate application and DMT-HFO extraction as a function of time.

There was an increase in the HCO_3-Pi fraction of the soil with an increase in the amount of phosphate applied. According to **Figure 3.10,** the 75 mg P kg^{-1} and 150 mg P kg^{-1} treatments increased the HCO_3-Pi fraction of this soil by 120 % and 187% respectively (day 1 values).

The difference between the control and the different treatments was significant according to **Table 3.2**, however, the difference between the HCO_3-Pi fractions of R75 and R150 was never significant over the 56-day period. Under the influence of the DMT-HFO the HCO_3-Pi fractions of the different treatments were virtually depleted after 56 days.

Table 3.2. The influence of the DMT-HFO extraction on the HCO_3-Pi fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	**x**[1] 1.84 **a**[2]	**y** 4.06 **a**	**y** 5.30 **a**
7	**x** 1.56 **ad**	**y** 2.49 **b**	**y** 4.25 **a**
14	**x** 0.97 **ad**	**y** 3.18 **ab**	**y** 4.83 **a**
21	**x** 0.42 **bc**	**y** 0 **c**	**y** 0 **b**
28	**x** 0.21 **bc**	**xy** 0.83 **cd**	**y** 0.97 **b**
42	**x** 0.13 **bc**	**y** 0.26 **cd**	**y** 0.32 **b**
56	**x** 0.0006**c**	**y** 0.17 **d**	**y** 0.19 **b**

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

2 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

The HCO_3-Pi fractions of R, R75 and R150 were very small compared to the other fractions, which are characteristic of highly weathered soils. Du Preez (1997), Cross & Schlesinger (1995) and Tiessen & Moir (1993), also found that the HCO_3-Pi fraction of highly weathered soils seldom exceed 10 mg P kg^{-1}.

This finding is surprising because it is expected that the increase of the soil suspension pH to 8.5 or even to 7 would significantly increase the solubility of strengite and variscite in highly weathered soils. Despite this little phosphate was extracted with 0.5M $NaHCO_3$. A possible explanation for the small amounts of phosphate usually extracted in highly weather soils is that the Point of Zero Charge (P.Z.C.) of ferric oxy hydroxide like goethite can be as high as 8-8.5 (the reason for this is that the surface groups of oxy hydroxide are weak Brønsted acids). It is also known that phosphate adsorption can take place even beyond the P.Z.C. of the adsorption surfaces because phosphate adsorption is predominantly a chemisorption process and not electrostatically adsorbed. It is therefore possible that phosphate that are extracted with HCO_3 because of the dissolution of strengite and variscite will be re-adsorbed by goethite and gibbsite or any other oxy hydroxide with high P.Z.C..

3.1.3) Inorganic NaOH (NaOH-Pi) fraction

A 0.1 *M* NaOH solution (pH = 13) is more basic than a 0.5 *M* $NaHCO_3$ and therefore a stronger extractant of solid phase phosphate for the following reasons:

1. The higher solubility of variscite and strengite at a pH of 13,
2. The excess OH^- in solution will displace all the remaining adsorbed phosphate in the soil after the 0.5 *M* $NaHCO_3$.
3. The increased dissolution of gibbsite and goethite (see **Figure 1.1)** exposes phosphate adsorbed in occluded adsorption sites. The subsequent decrease in adsorption surfaces further reduces the risk of possible re-adsorption of phosphate that comes in solution because of the dissolution of variscite and strengite.

At a pH of 4.1 according to **Figure 1.5,** strengite is less soluble than variscite and if variscite did form in this soil it would transform to strengite. Whether strengite or variscite is the predominate phosphate mineral depends on how long the pH of the soil was under 5 and the transformation rate of variscite to strengite.

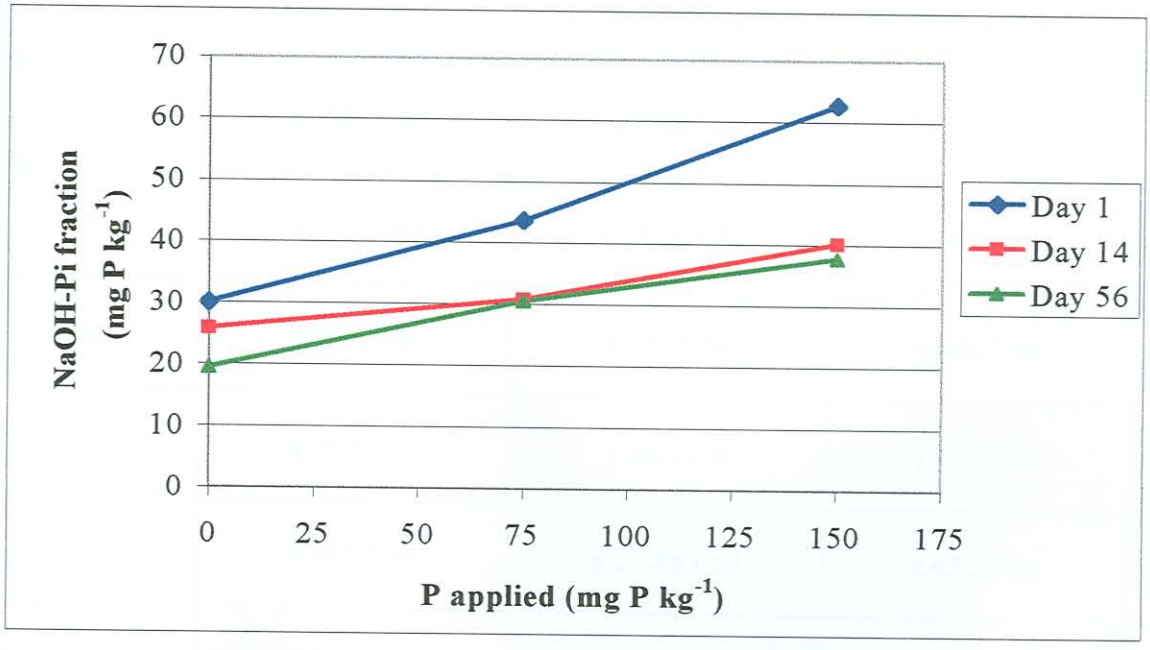

Figure 3.11: The change in the OH-Pi fraction as influenced by phosphate application and DMT-HFO extraction as a function of time.

According to **Figure 3.11,** the 75 mg P kg^{-1} and 150 mg P kg^{-1} treatments increased the OH-Pi fraction of this soil by 45% and 109% respectively (day 1 values) the difference between the different treatments were significant according to **Table 3.3.** From this data it can e concluded that more phosphate was transformed into less soluble NaOH extractable phosphate than to the more soluble $NaHCO_3$ extractable phosphate forms. The difference between the OH-Pi fraction of the control and the OH-Pi fraction of the treatments was greater than the difference between the HCO_3-Pi fraction of control treatment and the HCO_3-Pi fraction of the treatments. For every 1 mg P kg^{-1} the HCO_3-Pi fraction of R75 increased, the OH-Pi fraction increased by 6.16 mg P kg^{-1}. This ratio increased to 1:9.5 for R150.

Table 3.3. The influence of the DMT-HFO extraction on the OH-Pi fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	**x**[1] 30.00 **a**[2]	**y** 43.63 **a**	**z** 62.74 **a**
7	**x** 24.54 **bd**	**y** 34.26 **bde**	**z** 49.82 **b**
14	**x** 25.75 **b**	**y** 30.75 **bc**	**z** 40.07 **ce**
21	**x** 26.54 **ab**	**y** 36.23 **d**	**y** 40.58 **ce**
28	**x** 23.96 **bd**	**y** 31.88 **bcd**	**z** 45.24 **bc**
42	**x** 21.53 **cd**	**y** 26.64 **c**	**y** 36.59 **de**
56	**x** 19.45 **c**	**y** 30.42 **ce**	**z** 37.72 **cd**

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

3 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

There was a general tendency for the OH-Pi fractions of R, R75 and R150 to decrease with increasing DMT-HFO extraction time and the OH-Pi fractions of the different treatments were significantly lower than the respective day 1 levels. The OH-Pi fraction of the different treatments showed the biggest decrease (18 – 20%) in the first 7 days. The sharp decrease in the first week cannot solely be contributed to the influence of the phosphate sink, because the phosphate solid phase in the soil was also trying to equilibrate with the volume of solution added.

3.1.4) Comparison between phosphate released and HCO-Pi and OH-Pi fractions of the different treatments.

Discreet phosphate pools do not exist in the soil and the phosphate extracted with different extractants represents a continuum of easily to less exchangeable phosphate rather than distinct phosphate pools. However, correlations have been found between $NaHCO_3$ extractable phosphate and plant available phosphate and, to a lesser extent between NaOH extractable phosphate and plant available phosphate in tropical soils. (Olsen & Watanabe 1957, Bowman & Cole, 1978 , Ball-Coelho, Salcedo, Tiessen & Stewart, (1993).In this section the total phosphate desorbed in the 56-day period from the different treatments were compared to the decrease in HCO_3-Pi and OH-Pi fractions to determine how important role these fractions played in the desorption process

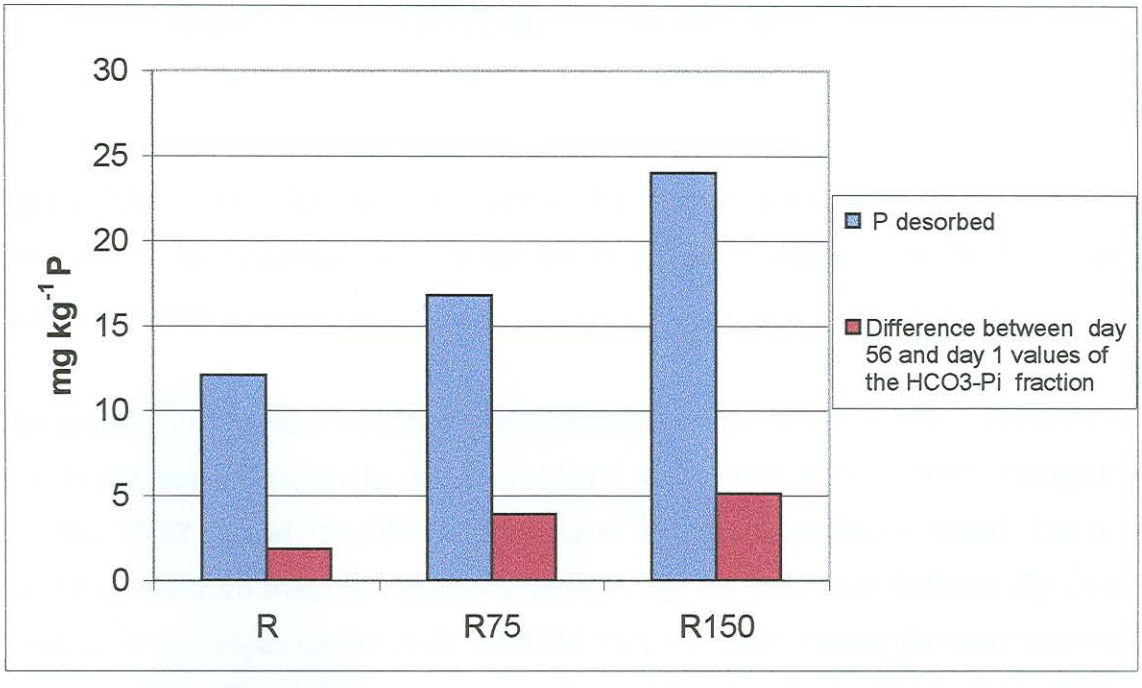

Figure 3.12. Total phosphate desorbed in the 56-day period from R, R75 and R150 compared to the differences between the day 56 and day 1 values of the HCO-Pi fraction of R, R75, and R150.

According to **Figure 3.12** very little of the desorbed phosphate originated from the HCO_3-Pi fractions of the different treatments. The decrease in the HCO_3-Pi fractions of

R, R75 and R150 represented only 15.27 %, 23.18 % and 21.31 % respectively of the total phosphate released from the soil.

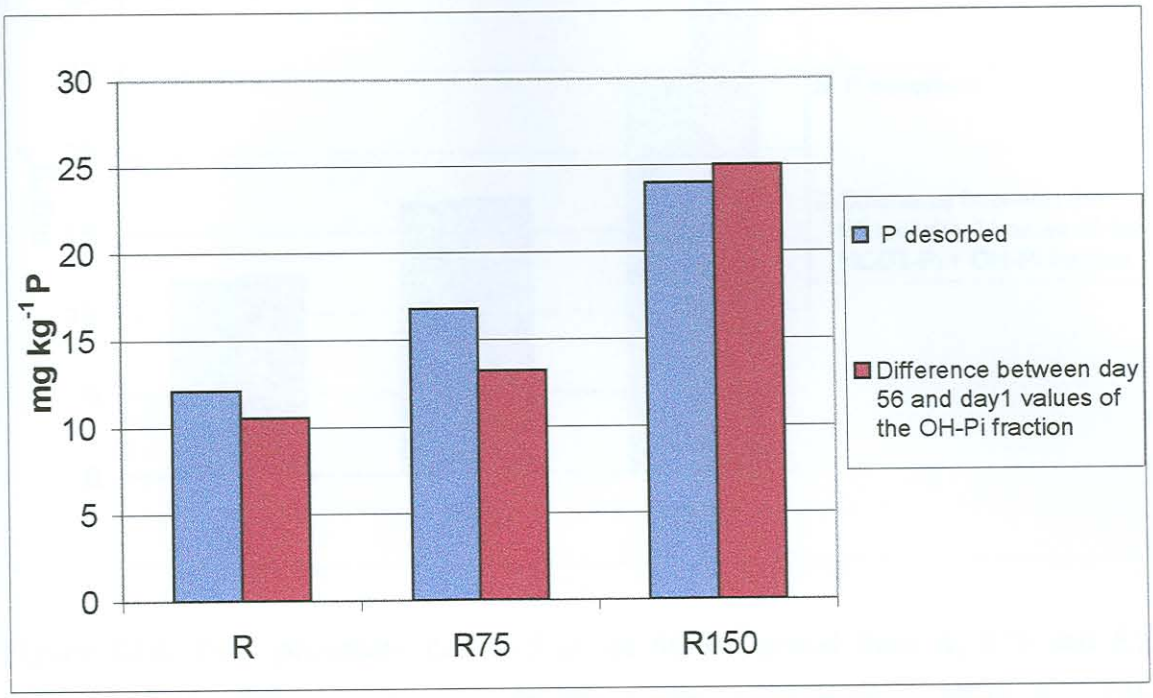

Figure 3.13. Total phosphate desorbed in the 56-day period from R, R75 and R150 compared to the differences between the day 56 and day 1 values of the OH-Pi fraction of R, R75, and R150.

The majority of the desorbed phosphate originated from the OH-Pi fraction (**Figure 3.13**) and the difference between the day 56 and day 1 values of R, R75 and R150 represented 87.38%, 78.67 % and 104.30% respectively of the total phosphate released. The total phosphate desorbed from the different treatments and the differences between day 56 and 1 values of the respective HCO_3-Pi + OH-Pi fractions were virtually the same according to **Figure 3.14.** The differences between the day 56 and day 1 values of the different treatments represented 102.66 %, 101.85 % and 125.61 % of the total phosphate released from R, R75 and R150 respectively.

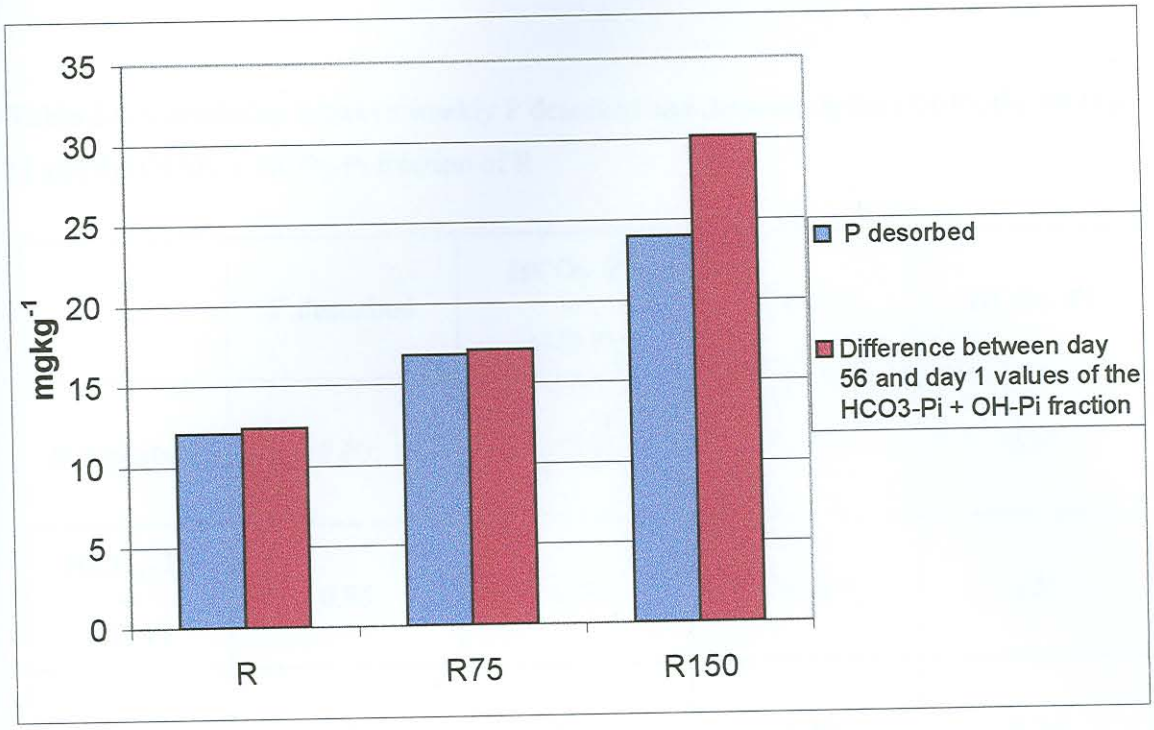

Figure 3.14. Total phosphate desorbed in the 56-day period from R, R75 and R150 compared to the differences between day 56 and day 1 values of the HCO_3-Pi + OH-Pi fraction of R, R75, and R150.

Determining the origin of the desorbed phosphate by comparing phosphate desorption data and decrease in the different fractions, was complicated by the fact that some phosphate that entered the solution, because the of the dissolution of phosphate minerals is probably re-adsorbed again by adsorption surfaces. This fact and experimental error was probably the reason why the phosphate released could not precisely be accounted for in the decrease of the HCO_3-Pi and OH-Pi fractions.

A correlation analysis was done with SAS to determine how strong the linear relationship was between the weekly phosphate desorbed from the different treatments and the weekly decrease in the OH-Pi, HCO3-Pi and OH-Pi + HCO3-Pi fractions of R, R75 and R150.

Table 3.4. Correlation between weekly P desorbed and decrease in the OH-Pi, the HCO_3-Pi and the OH-Pi + HCO_3-Pi fraction of R.

	P desorbed	**HCO_3- Pi + OH-Pi**	**OH-Pi**	**HCO_3- Pi**
P desorbed	1.00	0.85	0.80	0.57
HCO_3- Pi + OH-Pi	0.85	1.00	0.99	0.51
OH-Pi	0.80	0.99	1.00	0.34
HCO_3- Pi	0.57	0.51	0.34	1.00

There was a low correlation between the weekly decrease in the HCO_3-Pi fraction and the phosphate released from the different treatments according to **Tables 3.4, 3.5** and **3.6.** The correlation between the weekly decrease in the HCO_3-Pi fraction and the phosphate released from this soil, decreased from r = 0.567 (**Table 3.4)** for the control to r = 0.376 for R150 **(Tables 3.6)**. There was a higher correlation between the weekly change in the OH-Pi fractions and the phosphate desorbed from R, R75 and R150. The correlation between the weekly change in the OH-Pi fraction and phosphate desorbed increased from r =0.79 for the control to r = 0.915 for R150.

Table 3.5. Correlation between weekly P desorbed and the decrease in the OH-Pi, the HCO_3-Pi and the OH-Pi + HCO_3-Pi fraction of R75.

	P desorbed	HCO_3- Pi + OH-Pi	OH-Pi	HCO_3- Pi
P desorbed	1.00	0.86	0.82	0.51
HCO_3- Pi + OH-Pi	0.86	1.00	0.98	0.54
OH-Pi	0.82	0.98	1.00	0.35
HCO_3- Pi	0.51	0.54	0.34	1.00

According to **Tables 3.4 and 3.5,** the weekly phosphate desorbed from R and R75 showed the highest correlation of r = 0.85 and r =0.86 respectively with the weekly change in the OH-Pi + HCO_3-Pi fraction. In the case of R150, the correlation between the weekly change in the OH-Pi fraction and the weekly phosphate desorbed, and the correlation between the weekly change in the OH-Pi + HCO_3-Pi fraction and the weekly phosphate desorbed were the same

Table 3.6. Correlation between weekly P desorbed and the decrease in the OH-Pi, the HCO_3-Pi and the OH-Pi + HCO_3-Pi fraction of R150.

	P desorbed	**HCO_3- Pi + OH-Pi**	**OH-Pi**	**HCO_3- Pi**
P desorbed	1.00	0.91	0.92	0.38
HCO_3- Pi + OH-Pi	0.91	1.00	0.98	0.51
OH-Pi	0.92	0.98	1.00	0.34
HCO_3- Pi	0.38	0.51	0.34	1.00

The good correlation between the decrease in the OH-Pi fraction of the different treatments and phosphate desorbed shows that, although the OH-Pi fraction is less exchangeable than the HCO_3-Pi fraction, it could still be considered part of the labile phosphate pool of the studied soil. If the DMT-HFO simulated plant phosphate uptake, then the OH-Pi fraction is a better indicator of the plant-available phosphate over the longer term (± 2 months) than the HCO_3-Pi fraction. The decrease in the correlation between the weekly change in the HCO_3-Pi fraction and the phosphate desorbed, and the increase in the correlation between the change in the OH-Pi fraction and phosphate desorbed, also illustrated that applied phosphate transforms to less exchangeable NaOH extractable phosphate rather than $NaHCO_3$ extractable phosphate.

3.1.5) Inorganic 1 *M* HCl (1M HCl-Pi) fraction

According to Tiessen & Moir, (1993), 1 *M* HCl is very effective in extracting phosphate associated with calcium phosphates. Considering the pH of the studied soil, it is expected that the precipitation of aluminium and ferric phosphate would be favoured over that of calcium phosphate. It is therefore more likely that the phosphate extracted with 1 *M* HCl was aluminium and ferric phosphate that was left in the soil after the NaOH extraction.

Table 3.7. The influence of the DMT-HFO extraction on the 1M HCl-Pi fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	x[1]1.10 a[2]	xy 1.49 bc	y 1.90 cd
7	x 1.20 a	xy 1.58 bc	y 1.95 cd
14	x 1.08 a	x 1.17 ac	x 1.38 cd
21	x 1.35 a	x 1.46 bc	x 1.71 cd
28	x 1.40 a	x 1.60 bc	x 1.98 cd
42	x 1.15 a	x 0.94 c	x 1.048 d
56	x 1.20 a	x 2.13 bd	x 2.20 c

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

2. Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

There was no significant change in the 1 *M* HCl-Pi fraction of the different treatments over time (**Table 3.7**) and except for day 7 and 14, there also was no significant difference between R, R75 and R150.

3.1.5) Inorganic Concentrated HCl (con HCl-Pi) fraction

The concentrated HCl-Pi fraction represents the stable residual phosphate pool of the soil. This stable residual phosphate probably consists of: occluded phosphate; ferric and aluminium phosphates covered by a ferric oxy-hydroxides coating which protected the inner phosphate nucleus from solvent action, and maybe also stable apatite type minerals that could not be extracted with 1*M* HCl (Chang & Jackson, 1957, Lindsay, 1979). Most of the total phosphate extracted came from this fraction and it seems that the concentrated HCl-Pi fraction was the fate of most of the phosphate originally added to the soil.

Table 3.8. The influence of the DMT-HFO extraction on the concentrated HCl-Pi fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
7	x[1] 104.62 ac[2]	y 116.04 a	z 136.11 a
14	x 116.07 a	xy 125.89 a	y 133.82 a
21	x 116.36 a	x 112.75 a	x 116.67 a
28	x 92.31 c	xy120.50 a	y 124.67 a
42	x 104.14 ac	xy 116.82 a	y 120.91 a
56	x 98.59 ac	xy 117.41 a	y 119.20 a

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

2 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

There was no significant change over time in the concentrated HCl-Pi fraction of the different treatments (**Table 3.8**). There was however a significant difference between concentrated HCl-Pi fraction of R150 and that of the control.

3.2) Organic phosphate fractions

Because of the low organic content of South African soils (especially soils under cultivation) organic P is not as an important source of phosphate than inorganic phosphate forms. It is important to note that the DMT-HFO cannot simulate the biochemical reactions involved in the mineralisation of organic P. Organic P mineralisation in the soil is usually faster in the presence of plant roots than in the absence of plant roots, the reason for this is the absence of the enzyme phosphatase which is vital for P mineralisation. This enzyme enhances the hydrolysis of phosphorus from the organic compounds, and the absence of phosphatase greatly reduces organic P mineralisation.
(Thompson & Black, 1970a, Islam & Ahmed, 1973, Campbell & Racz, 1975, Blair & Boland, 1978, Racz, 1979).

Phosphatase enzymes originate from two sources:

- Rhizosphere microorganisms. The highest concentration of phosphatase producing enzymes are found in the rhizosphere because of the higher carbon concentration in the rhizosphere due to root debris and also carbon-rich root exudates like glucose (Alexander, 1961 Greaves & Webley, 1965);
- Plant roots. Plant roots also produce and exudes phosphatase enzymes. (Szember, 1960b, Esterman & McLaren, 1961).

Because of these factors it is not expected that these experimental conditions accurately simulated the organic P dynamics that normally take place in the soil.

3.2.1) Organic $NaHCO_3$ (HCO_3-Po) fraction

The HCO_3-Po fraction consists of the portion of organic P forms (such as RNA nucleotides and glycerophosphates), which can easily be mineralised by biological processes (Bowman & Cole, 1978, McKercher & Tollefson, 1978, Tarafdar & Claassen, 1988).

Table 3.9: The influence of the DMT-HFO extraction on the $NaHCO_3$-Po fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	**x**[1] 1.14 **b**[2]	**y** 4.00 **b**	**y** 5.43 **b**
7	**x** 6.62 **a**	**y** 8.57 **c**	**z** 10.52 **a**
14	**x** 2.14 **b**	**y** 11.11 **d**	**y** 11.43 **a**
21	**x** 1.39 **b**	**x** 0.0 **a**	**x** 0.71 **c**
28	**x** 3.08 **ab**	**y** 9.64 **dc**	**y** 9.11 **a**
42	**x** 2.78 **ab**	**x** 4.52 **b**	**x** 5.95 **b**
56	**x** 1.98 **b**	**x** 2.73 **b**	**x** 2.92 **bc**

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

2 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

According to **Table 3.9.** the HCO_3-Po fraction of the different treatments *increased* during the first two weeks. A possible explanation for this is that some of the phosphate that desorbed was immobilised again by microorganisms. There were a significant differences between the HCO_3-Po fraction of the different treatments up to day 28,

indicating that some of the initially applied phosphate was immobilised by microorganisms. After day 28 there was no significant difference between the different treatments.

In this study the $NaHCO_3$ fraction was the only fraction where there was more $NaHCO_3$-Po than HCO_3-Pi. Armstrong & Helyar (1992) and Du Preez (1997), also found that the majority of the total HCO_3 extractable phosphate to consists of organic P. Cross & Schlesinger (1995), reported a general trend that the HCO_3-Po fraction increases with an increase in the degree of weathering of the soil.

3.2.2) Organic NaOH (OH-Po) fraction

The OH-Po fraction extracts the less labile alkali soluble organic P in the soil. Because the nature of a large part of organic P is still unknown, it is not possible to determine exactly which organic P compound was extracted (Anderson, 1975). According to Tiessen & Moir (1993), the organic P extracted with $NaHCO_3$ (HCO_3-Po) and the organic P extracted with NaOH probably represents the same organic P pool namely the alkali soluble organic P.

Table 3.10. The influence of the DMT-HFO extraction on the NaOH-Po fractions of R, R75 and R150.

Day	R (mg P kg^{-1})	R75 (mg P kg^{-1})	R150 (mg P kg^{-1})
1	x[1]22.73 a[2]	x 22.43 a	x 21.84 a
7	x 27.83 a	x 24.59 a	x 19.54 a
14	x 26.85 b	x 26.75 a	x 22.51 a
21	x 24.02 a	x 25.33 a	x 24.26 a
28	x 27.70 a	x 23.28 a	x 20.54 a
42	x 26.81 a	x 25.41 a	x 23.86 a
56	x 24.09 a	x 24.90 a	x 25.46 a

1 Mean values in rows with different letters x y z are significantly different ($\alpha = 0.05$)

2 Mean values in columns with different letters a, b, c, d, e and f are significantly different ($\alpha = 0.05$)

There was no significant change in the OH-Po fraction over the DMT-HFO extraction time (**Table 3.10**). The different phosphate levels the soil was incubated with caused no significant increase in the OH-Po fraction of this particular soil. It seems that no significant amount of P was mineralised from this fraction. This could probably be attributed to the absence of rhizosphere microorganisms responsible for the mineralisation of organic P.

3.2.3) Organic Concentrated HCl (con HCl-Po) fraction

The organic P extracted with concentrated HCl represents a very stable organic P pool or labile organic P that is not alkali soluble (Tiessen & Moir, 1993). The soil had a low organic material content and after the previous two extractions it seemed that virtually no organic P was left in the soil. There was little difference between the total P extracted with concentrated HCl ($HCl\text{-}P_{tot}$) and inorganic P extracted with concentrated HCl. In most instances there was no difference. Organic P extraction with concentrated HCl involved the most steps resulting in the end in the largest extrapolation factor to relate the determined values to mg P kg soil $^{-1}$. The small difference between the con HCl-Pi fraction and $HCl\text{-}P_{tot}$ and the big difference between the extrapolation factors of concentrated HCl-Pi and concentrated $HCl\text{-}P_{tot}$ meant that the slightest error in one of the four replications of the $HCl\text{-}P_{tot}$ determination, magnified by the high extrapolation factor, could have resulted in the average of the con $HCl\text{-}P_{tot}$ being dragged lower than that of the con HCl-Pi