The decomposition of cocoa leaves and their effect on phosphorus dynamics in tropical soil

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Summary
Higher-yielding varieties of cocoa make heavier demands on phosphorus resources in soils and so it is important that the role of leaf litter in cycling P is understood. Fresh cocoa leaves and leaf litter were incubated moist with a soil inoculum for 80 days when between 16 and 33% of the mass was lost. Materials containing large amounts of P or incubated with added inorganic P initially decomposed more rapidly than those containing smaller amounts, indicating that decomposition was limited by lack of P. Fresh leaves had half of their P in an acid-soluble (0.1 M H₂SO₄) form, most of which was also water soluble, whereas in the litters about a third was acid-soluble. During incubation, P-rich materials showed an increase in the acid-soluble fraction and a decrease in water-soluble P. Litters with small concentrations of P simply lost P from the acid-soluble into the non-soluble organic fraction, and no water-soluble P remained after 80 days.

A soil from a cocoa-growing site fertilized with P contained almost four times as much biomass P as the non-fertilized control (30 and 8 mg kg⁻¹ soil, respectively), the amounts of bicarbonate-extractable P being 32 and 4 mg kg⁻¹. Soils from these and one other cocoa-growing site (8 mg kg⁻¹ biomass P, 7 mg kg⁻¹ bicarbonate-extractable P) were incubated either alone, with cocoa litter, or with cocoa litter plus inorganic P. In the soil that had the small amount of NaHCO₃-extractable P (4 mg kg⁻¹) addition of litter caused the biomass P to increase from 8 to 16 mg kg⁻¹ after 1 week’s incubation, the increase being larger than the amount of P added in the litter, but in the other two soils biomass P was not increased. Addition of inorganic P had no effect on biomass P in any of the soils.

Decomposing litter may compete with the crop for P, but addition of fertilizer P may increase the rate of mineralization of organic P in the litter. Suitable management of fertilizer P should allow the rate of release of P from the litter to be adjusted to suit crop demands.

Introduction
Cocoa is important in the economy of Ghana; its export accounts for about 60% of the country’s foreign earnings. The cocoa-growing soils generally contain little available phosphorus, which is the most limiting nutrient in the production of the crop (Smith & Acquaye, 1963). The bulk of the country’s cocoa is produced by peasant farmers who cannot afford to buy fertilizers, and who generally consider that, except during establishment, fertilization with phosphorus is not needed because once the canopy has formed, shaded cocoa in some way fertilizes itself. Thus it may be that the cycling of P within the soil–plant system is adequate for production. The phosphorus content of whole mature plants is about 48 kg ha⁻¹, and that removed in the beans and pods is about 5 kg ha⁻¹ year⁻¹ (Thong & Ng, 1980). The farmers’ assertion may therefore be correct, because Nye & Bertheux (1957) reported that in forest soils of Ghana with about 450 kg organic P ha⁻¹ in the topsoil the annual release of P by mineralization was about 14 kg P ha⁻¹. Nye & Greenland (1960) reported a decrease of 34 kg ha⁻¹ of soil organic P over 8 years following forest clearance in Ghana. This amount falls only a little short of the 48 kg ha⁻¹ present in the mature plants. Once the crop is established, between 4 and 5 kg P ha⁻¹ is returned each year in the cocoa litter, with about 1 kg ha⁻¹ being washed off the leaves into the soil and between 2 and 3 kg ha⁻¹ entering the system from rain (Ling, 1984).

Thus supplies of P might be adequate, even if only a small quantity of P is solubilized each year from the inorganic fraction. However, with the introduction of high-yielding varieties which make heavier demands on soil resources, the
present system of cropping without compensating for nutrient removal may be unsound farming practice. Fertilization may be needed, but the release of P returned to the soil in the litter will continue to provide an important component of the crop’s needs.

There is good evidence that in tropical soils uptake of P by crops is significantly correlated with organic P content of the soil (Adepetu & Corey, 1976) and with amounts of mineralized P (Adepetu & Corey, 1977; Mueller-Harvey et al., 1985). Much of the mineralized P passes through the microbial biomass, and the annual flux has been estimated for British soils as between 5 kg P ha\(^{-1}\) year\(^{-1}\) for an unmanured arable soil and 23 kg P ha\(^{-1}\) year\(^{-1}\) for an unmanured grassland soil (Jenkinson & Ladd, 1981; Brookes et al., 1984). With much faster decomposition in the tropics than in temperate regions (Jenkinson & Ayanaba, 1977) the annual fluxes will also be larger. The role of the microbial biomass in soils under cocoa has not been studied, but it is likely that when cocoa leaves enter the soil much of the inorganic P (58% of the total P in cocoa leaves; Mueller-Harvey & Wild, 1986) can be rapidly assimilated by the biomass (Harrison, 1982), the organic P then being more slowly utilized during the decomposition of the leaves. It is not known what the effects will be if fertilizer P is added to the litter, but McLaughlin & Alston (1986) have shown that the biomass assimilated a proportion of the applied P equal to that taken up by plants in a pasture, and work by Guerra et al. (1985), Singh (1995), Goladi & Agbenin (1997) and He et al. (1997) has shown that additions of plant residues and fertilizer P tend to increase the amount of biomass P, although with marked seasonal fluctuations. Under cocoa, where there is generally little available P but where there is much litter, there is likely to be considerable potential for immobilization.

We have examined the changes in the forms of P in cocoa leaves and leaf litter when incubated alone and with soil to evaluate the significance of the microbial biomass in the transfer of P between the various pools in the soil.

**Materials and methods**

**Leaves and litter**

Samples were collected from two experimental sites of the Cocoa Research Institute in Ghana, namely Nankese and Bechem. At each site litter from P-fertilized and non-fertilized (control) plots was collected. Fresh leaves were picked from mature cocoa trees on the Nankese P-fertilized plot. Litter was also obtained from under cocoa trees grown in glasshouses at the University of Reading, where the trees had been well supplied with P. The materials were air-dried and cut into pieces approximately 1 cm square. The properties and the codes of the materials are listed in Table 1.

**Soil**

Samples of soil were from the A horizons on the Nankese control, Nankese P-fertilized and Bechem control plots. The fertilized plot had received 25 kg P ha\(^{-1}\) year\(^{-1}\) for 17 years before sampling. The soils are Forest Ochrosols according to the Ghanaian classification system (Brammer, 1962); they are immature reddish brown latosols formed on granodiorite under 1000–2000 mm rain year\(^{-1}\). The clay fraction is primarily kaolinite. The soils were air-dried and passed through a 2-mm sieve.

**Leaf and litter experiments**

**Incubation.** Water (15 ml) was added to leaf and litter samples equivalent to 5 g of air-dry material in a 150-ml beaker to produce moist but not saturated material. Six leaf and litter materials were used, together with two extra litter samples from the Nankese control and Nankese fertilized plots to which 25 mg P as Na\(_2\)HPO\(_4\) had been added per 5 g air-dry material, increasing their total P contents by 0.5%. These were coded NCPL and NPPL, respectively (Table 1). To each beaker was added 5 mg of soil (Nankese control) as an inoculant with thorough mixing. The beakers were placed in 1-l jars with 10 ml of water at the base to maintain humidity. Into each jar was placed a vial containing 20 ml of 1 M NaOH, and the closed jars were stored at 25°C in the dark. The CO\(_2\) produced by respiration was absorbed by the NaOH. Preliminary experiments had shown that the jars needed to be opened every three days during the first 15 days to maintain aeration, and subsequently at less frequent intervals, as assessed from the measured rate of CO\(_2\) production during the preceding interval. Thus, as required, the jars were opened for 10 min, the vials removed, the NaOH retained for titration, and fresh NaOH added before closing the jars. Water was added as required to the beakers to maintain the water content of the materials. After each interval the CO\(_2\) absorbed in the NaOH was measured by titrating with 0.5 M HCl using phenolphthalein as the indicator after adding 10 ml of 1 M BaCl\(_2\) to precipitate carbonate. Blanks incubated without leaf materials were included. Four replications of each material were sampled after 0, 20, 40 and 80 days of incubation.

**Extractable inorganic P.** Following incubation, 2 g of moist material was shaken for 30 min at 20°C with 50 ml of distilled water. Similarly, on another 2-g subsample extraction with 0.1 M H\(_2\)SO\(_4\) was carried out. The extracts were filtered through a Whatman No 40 paper, and P was determined colorimetrically using an autoanalyser to give water-extractable and acid-extractable P.
Dry matter. At the end of each incubation period, the beaker with its remaining contents was weighed, and dried for 48 h at 80°C. After cooling in a desiccator it was weighed again to determine the mass of dry matter. Knowing the amounts removed for determination of extractable inorganic P, we calculated the decrease in dry matter during incubation.

Total carbon. A sample of dry matter (20 mg) was digested in acid dichromate solution and the carbon determined by titration with ferrous ammonium sulphate (Tinsley, 1950).

Total nitrogen. Nitrogen was determined in 200 mg of dry matter using a Kjeldahl digestion, distillation of NH₃ and titration (Bremner, 1965).

Total phosphorus. Phosphorus was determined in 200 mg of dry matter by digestion with concentrated sulphuric acid, hydrogen peroxide and selenium. The P was determined by the phospho-vanado-molybdate method (Cavell, 1955).

Soil experiments

Incubation. The soil samples were brought to 40% of their water-holding capacity (Jenkinson & Powlson, 1976) and preincubated at 25°C for 21 days. Moist samples equivalent to 20 g of oven-dry soil were treated as follows.

(a) Soil alone.
(b) Soil + litter. Milled Nankese control litter (0.1 g) was mixed with 20 g of soil, thus adding 3 mg P kg⁻¹ dry soil, equivalent to an annual litter fall per ha (2000 t soil) of 10 t of litter containing 6 kg of P.
(c) Soil + litter + P. Milled litter was added as above, but with 15 mg P kg⁻¹ soil added as KH₂PO₄ solution, a rate equivalent to 30 kg P ha⁻¹, which is typical of field applications for cocoa.

All the soil samples were then brought to 50% of their water-holding capacity and incubated in jars at 25°C. They were opened every three days to maintain aerobic conditions, and water was added as required to maintain the soil water content. Duplicate samples were taken for analysis after 0, 7, 14, 28 and 56 days of incubation.

Sodium bicarbonate-extractable P. A moist subsample from each jar (2 g oven-dry soil) was shaken with 50 ml of 0.5 M NaHCO₃ solution at pH 8.5 for 30 min at 20°C, filtered through a Whatman No 42 paper and the P determined by the ammonium molybdate–ascorbic acid method (Murphy & Riley, 1962).

Biomass P. We used a procedure based on the fumigation–extraction method of Jenkinson & Ladd (1981). From the remaining sample in each jar, moist subsamples were placed in a desiccator containing alcohol-free chloroform. The desiccator was evacuated until the chloroform had boiled for 5 min, and was then kept at 25°C for 24 h. The chloroform vapour was removed by repeated evacuation, and the samples were then extracted with NaHCO₃ solution as above. Similarly, non-fumigated subsamples were also extracted. The extracts were then digested in 70% HClO₄, and the P analysed as above. Biomass P was determined as the difference between the extracted P in the fumigated and non-fumigated soils, multiplied by 2.5 (Brookes et al., 1982) to account for the fraction of the biomass P that is assumed to be extracted.

Other soil properties. The soils were characterized by measuring particle size distribution, pH (1:2.5 in H₂O), percentage C (Tinsley, 1950) and percentage N (Bremner, 1965). Total P was measured by dry-ashing at 500°C and dissolving the ash in 6 M HCl before analysing by the phosphomolybdate method. Organic P was determined as the difference between the amounts of P extracted by 6 M HCl from soil samples before and after dry-ashing at 500°C (Olsen & Sommers, 1982).

Table 1 Composition of the cocoa leaves and litter

<table>
<thead>
<tr>
<th>Code</th>
<th>Total C (%)</th>
<th>Total N (%)</th>
<th>Total P (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nankese control litter</td>
<td>NCL</td>
<td>36.4</td>
<td>0.97</td>
</tr>
<tr>
<td>Nankese control litter + P</td>
<td>NCPL</td>
<td>36.4</td>
<td>0.97</td>
</tr>
<tr>
<td>Nankese P-fertilized litter</td>
<td>NPL</td>
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<td>1.16</td>
</tr>
<tr>
<td>Bechem control litter</td>
<td>BCL</td>
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<td>1.25</td>
</tr>
<tr>
<td>Bechem P-fertilized litter</td>
<td>BPL</td>
<td>37.7</td>
<td>1.29</td>
</tr>
<tr>
<td>Nankese P-fertilized leaves</td>
<td>NPF</td>
<td>38.3</td>
<td>2.08</td>
</tr>
<tr>
<td>Nankese P-fertilized leaves + P</td>
<td>NPPL</td>
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<td>2.08</td>
</tr>
<tr>
<td>Reading litter</td>
<td>RL</td>
<td>40.0</td>
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</table>
Results and discussion

The decomposition of leaf material

The C, N and P contents of the materials are listed in Table 1, which provides the codes for the materials and treatments also. The C:P ratios range from 143:1 (RL) to 615:1 (BCL). The data indicate that before use in these experiments P had already been released from the litter over a few months between leaf fall and sampling. Fertilized plots had produced litter with only slightly larger concentrations of P.

The losses of dry matter during incubation are shown in Figure 1. After 80 days between 16 and 33% of the initial mass had been lost. Similar values were obtained for the measured losses of organic carbon, the carbon lost as CO$_2$ in respiration, and the loss of carbon calculated from the mass loss and the C contents shown in Table 1. The results show that the fresh leaf, the Reading litter and the litters with added P (NPF, RL, NCPL, NPPL) initially decomposed more rapidly than the other litters, especially in the early part of the incubation (Figure 1). The other litters (BCL, NCL, BPL, NPL) were less well supplied with P (Table 1), suggesting that decomposition rates might have been limited by the lack of P. The curvature of the graphs for the Reading litter and the fresh leaves (NPF) is similar to that for the decay of fresh plant residues in soils (Jenkinson & Ayanaba, 1977). This suggests that two fractions were present which decayed according to first-order kinetics but with different rates. The graphs for the remaining materials show much less curvature, which suggests either that these litters were already partly decomposed and so were decaying as though only one fraction was present, or that the shortage of P had a pronounced effect on the fraction that would otherwise have decayed quickly. We had too few data to fit kinetic models precisely. Measurements of total P and total N showed that there were no losses of these elements during incubation.

Figure 2 shows changes in the fractions of inorganic P. We consider that water-soluble P is present as H$_2$PO$_4^-$, and that the extra P which dissolves in 0.1 M H$_2$SO$_4$ (Figure 2c), although called inorganic, is partly inorganic soluble at low pH and partly organic, for example glucose-1-phosphate (Anderson, 1960) which is soluble in the dilute acid. We consider that the P not extracted by acid is organic.

The materials fall into three groups, as follows.

1. The Ghanaian litters without added P (NCL, BPL, NPL, BCL) initially had about 30% of their total P in the acid-soluble fraction (Figure 2b). It was also mostly water soluble (Figure 2a). With time, the amounts in the acid-soluble fraction fell to about 10%, mostly as a result of a decrease in water-soluble P, and there were slight increases in the acid-minus water-soluble P fraction. The organically bound P not extractable in acid (100 minus the values in Figure 2b) must have increased from about 70 to 90% of the totals.

2. The Nankese leaves (NPF) and Reading litter (RL) initially had 52 and 27% of their P in the acid-soluble fraction, respectively, again mostly water soluble. During incubation, the acid-minus water-soluble fraction increased in both materials to about 30% of the total during the first 20 days.

3. The Ghanaian litters with added P (NCPL, NPPL) had about 65% of their P in the acid-soluble fraction at time zero (Figure 2b). Calculation shows that the quantity of P added should have initially increased the acid-soluble fraction to nearly 90%, and so in the short time between treatment and the zero time extraction there must have been a rapid conversion of added P to a form that was not extractable by acid. With time the acid-soluble P increased slowly and water-soluble P decreased rapidly with a corresponding large increase in acid-minus water-soluble P, so that after 80 days more than 60% of the added P was in this fraction (Figure 2c).

The P content of the materials again appears to be important in determining their behaviour. Those well supplied with P showed an increase in acid-soluble P and a decrease in water-soluble P. The Ghanaian litters with small concentrations of P simply lost P from the inorganic fractions into the organic fraction, and after 80 days they had little water-soluble P remaining, presumably because of immobilization during decomposition.
Soil±leaf incubations

Table 2 reports the results of soil analysis. The soils have most of their P in the organic fraction, the Nankese control having twice the P content of the Bechem control. The fertilized Nankese soil had accumulated P in the inorganic fraction and bicarbonate-extractable P had increased from 4 mg kg ±1 in the control to 32 mg kg±1.

Changes in the amounts of bicarbonate-extractable and biomass P are shown in Figure 3, where the biomass P is displayed as the shaded interval. Even after preincubation for 21 days, the soils still showed small changes in the forms of P during the experiment. The Nankese P-fertilized soil had about four times the biomass-P content of the control. This is a large increase when considered in the context of the carbon content which had been increased only from 2.2 to 2.6% by 17 years of fertilization and the organic P:C ratio which had been only slightly decreased. If yields were increased by fertilization then there has been an increase in annual litter input and in the rate of turnover of organic matter, and so we expect an increase in biomass P. Brookes et al. (1984) found that fertilizer P increased biomass P in the Rothamsted Park Grass plots by 47%. Guerra et al. (1995) found that biomass P was doubled by applications of P to plots growing Brachiaria decumbens resulting from an increase in the concentration of P in the biomass. A similar increase was found by Jonasson et al. (1996) in organic soils of an arctic-alpine heath, again resulting from an increase in the concentration of P in the biomass rather than an increase in the biomass itself. They concluded that the microbial biomass is a strong sink for nutrients and that it can withdraw substantial amounts of nutrients from the plant-available pool at least from time to

<table>
<thead>
<tr>
<th>Clay /%</th>
<th>Nankese control</th>
<th>Nankese P fertilized</th>
<th>Bechem control</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH 1:2.5 in water</td>
<td>6.0</td>
<td>6.3</td>
<td>5.4</td>
</tr>
<tr>
<td>Organic C /%</td>
<td>2.2</td>
<td>2.6</td>
<td>2.0</td>
</tr>
<tr>
<td>NaHCO3-extractable P /mg kg±1</td>
<td>4.0</td>
<td>32.1</td>
<td>5.6</td>
</tr>
<tr>
<td>Organic P /mg kg±1</td>
<td>262</td>
<td>283</td>
<td>132</td>
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<tr>
<td>Total P /mg kg±1</td>
<td>297</td>
<td>523</td>
<td>155</td>
</tr>
<tr>
<td>Total N /g kg±1</td>
<td>1.8</td>
<td>2.2</td>
<td>1.2</td>
</tr>
</tbody>
</table>

Figure 2 Changes in the amounts of water-soluble inorganic P (3.8), acid-soluble P (3.0) and acid-minus water-soluble P during the decomposition of leaf and litter. The values above in () are the standard errors for the data expressed as a percentage of the mean. The total P in each material is shown in Table 1.
time. The composition of the biomass was also found to change seasonally in pastures (He et al., 1997) with farmyard manure and P fertilizer resulting in the largest amounts of biomass C and P.

The increase in biomass P in the Nankese soil is thus larger than those recorded elsewhere. Questions have been raised by Brookes et al. (1982) regarding the reliability of the method of measurement. They discussed the effects of fixation of inorganic P released from the biomass by fumigation and developed a method in which a ‘spike’ of inorganic P is added to a subsample of the non-fumigated soil in the NaHCO₃ solution, thereby allowing the proportion of the added P, which is not extractable, to be calculated. The inorganic P released from the biomass is regarded as fixed in the same proportion. Although we did not include this modification, we did calculate for each soil a proportion of the added inorganic P which was fixed at time zero based on the results displayed in Figure 3. This shows that 57% was fixed in the Nankese control soil plus litter, 65% in the Nankese fertilized soil plus litter and 53% in the Bechem soil plus litter. Following

Figure 3 The amounts of bicarbonate-extractable and biomass P during incubation of soils with Nankese control cocoa litter (see Table 1): (a) soil alone, (b) soil + litter and (c) soil + litter + inorganic P. The bars show the amounts of P in the litter (3 mg kg⁻¹) and added as the treatment (15 mg kg⁻¹). The standard errors for the data expressed as a percentage of the mean are 4.7 for bicarbonate P and 10.4 for biomass P.
Brookes et al. (1982), these values can be used to ‘correct’ the biomass-P values, which become 1.75, 1.54 and 1.89 times the values shown for the three soils. The effect of differential fixation of P in the method therefore does not seem to be the reason for the large increase in biomass P in the fertilized Nankese soil, and we conclude that fertilization over the years contributed substantially to the amount of P in the biomass.

The addition of litter (Figure 3) caused a doubling of biomass P in the Nankese control after 1 week, and we presume that it is the result of the increased availability of substrate for microbial growth. The added litter contained only 3 mg P kg⁻¹, and yet biomass-P increased by about 9 mg kg⁻¹, so that P must have been taken up from the inorganic fraction to supply the demand of the biomass. Bicarbonate-extractable P was not changed even though the litter had only a small concentration of P, and the leaf incubation experiments indicate that immobilization was occurring. However, the amounts of P involved indicate that the potential for immobilization is small: the 20 g of soil contained 0.08 mg of extractable P, the 0.1 g of litter contained 0.06 mg P, and during leaf incubation 0.01 mg P was immobilized. In the Nankese fertilized and Bechem control soils litter had little effect on the distribution of P, although there was some indication that biomass P was increasing towards the end of the incubation.

The addition of P with the litter (Figure 3) had the expected effect on bicarbonate-extractable P; about 60% of that added was extractable at time zero. After incubation for 60 days, between 6 and 27% of the P added remained extractable, the smallest amount being in the Nankese control, which also had the smallest amount of extractable P (Table 2). The addition of P had little effect on the amount of biomass P in any of the soils. Although the literature reports that addition of crop residues and fertilizer P increases biomass-P contents of soils (Singh, 1995), our results show less consistency; only the Nankese control soil showed a marked effect of litter, and no soil showed an effect of added fertilizer P. The Nankese control soil had the smallest amounts of biomass P and NaHCO₃-extractable P before treatment, which may have contributed to the observed effects of the litter, but we were surprised that the effect of fertilizer P, which increased the amount of NaHCO₃-extractable P, was not seen in an increase in biomass P.

Conclusions

Fresh cocoa leaves are a source of readily available P, and following leaf fall the amounts of P released will depend on the extent to which mineralization and immobilization occur. In our experiments addition of inorganic P to the litter stimulated mineralization, and so fertilization in the field might increase the rate of release of P from litter. When no P was added, that already present was immobilized during the decomposition of cocoa litter, even where this came from fertilized crops. At least for part of the year the decomposing litter may compete with the crop for P. The phosphorus in the microbial biomass was increased by annual fertilization of the Nankese soil, but the short-term effects of additions of P in the laboratory were not clear. Addition of litter increased biomass P only in the Nankese control soil where the increase was larger than the amount of P added in the litter. Better understanding of the role of the microbial biomass in the dynamics of P in these soils will require measurement of both P and C in the biomass and also estimates of the rate at which P passes through the biomass into an available form.

References


