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PHOSPHORUS NUTRIOPERIODISM IN RUBBER

JESSY, M.D.

**Thesis submitted in partial fulfilment of the requirement
for the degree of**

Doctor of Philosophy in Agriculture

**Faculty of Agriculture
Kerala Agricultural University, Thrissur**

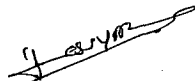
2004

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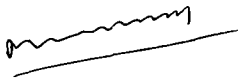
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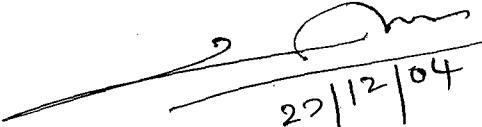
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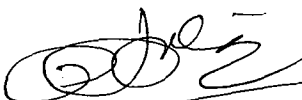

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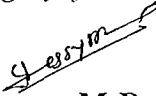
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LIST OF ABBREVIATIONS

%	Per cent.
°C	Degree Celsius
°E	Degree east
µg	Microgram
µm	Micromole
°N	Degree north
@	At the rate of
Al	Aluminium
AMF	Arbuscular mycorrhizal fungi
C	Carbon
Ca	Calcium
cc	Cubic centimetre
CD	Critical difference
CEC	Cation exchange capacity
cm	Centimetre
cmol	Centimol
CO ₂	Carbon dioxide
Cu	Copper
d	Day
DHA	Dehydrogenase activity
DRC	Dry rubber content
<i>et al.</i>	And others
Fe	Iron
Fig.	Figure
FW	Fresh weight
g	Gram
H ⁺	Hydrogen ions
h	Hour
ha	Hectare
<i>i.e.</i>	That is
K	Potassium

LIST OF ABBREVIATIONS CONTINUED

K ₂ O	Potash
kg	Kilogram
m	Metre
MDH	Malate dehydrogenase
meq	Milliequivalents
Mg	Magnesium
mg	Milligram
min	Minute
ml	Millilitre
mm	Millimetre
Mn	Manganese
N	Nitrogen
NADH	Nicotinamide adenine dinucleotide (reduced)
nmol	Nanomol
P	Phosphorus
P ₂ O ₅	Phosphate
PEPC	Phosphoenol pyruvate carboxylase
ppm	Parts per million
PSB	Phosphorus solubilizing bacteria
r	Correlation coefficient
S	Spiral
SE	Standard error
t	Tonnes
TPD	Tapping panel dryness
TPF	Triphenyl formazan
<i>viz.</i>	Namely
Zn	Zinc

Introduction

1. INTRODUCTION

Rubber (*Hevea brasiliensis* Muell. Arg.), a forest tree indigenous to the tropical rain forests of Central and South America is the only major commercial source of natural rubber. It is mainly cultivated in the South Asian countries like Indonesia, Thailand, Malaysia, China, India, Sri Lanka etc. The tree has an economic life span of about 30 years under cultivated conditions. Controlled wounding of the bark yields latex which contain 30-40 per cent rubber hydrocarbon.

Judicious nutrient management is of prime importance for sustaining soil productivity in any agricultural system. Rubber has an immaturity period of about six to seven years and proper nutrient management during this period will reduce this long unproductive period. During mature phase, it is of paramount importance to sustain productivity at high levels.

Nutrient requirement of rubber was generally considered to be modest mainly due to the fact that earlier plantations were raised mostly in newly cleared forest soils rich in plant nutrients (Krishnakumar and Potty, 1992). However, the scenario has changed with more and more marginal lands low or deficient in plant nutrients being brought under rubber cultivation. Moreover the existing rubber plantations are in the second or third cycle of replanting and nutrient management should be given adequate importance to sustain productivity at economic levels.

In plants, phosphate plays a pivotal structural and regulatory role at the nexus of photosynthesis, energy conservation and carbon metabolism. Its unique chemical properties place it at the centre of metabolism. However, it is frequently a major or even the prime limiting factor for plant growth. It is estimated that 5.7 billion hectares of soil worldwide

contain too little available phosphorus for sustaining optimal crop production (Batjes, 1997).

Among major nutrients, phosphorus is the least mobile and least available to plants in most soil conditions. The poor mobility of the soil inorganic P is due to the large reactivity of phosphate ions with the numerous soil constituents and their consequent strong retention on those constituents. Because of the unique interaction of P with other elements, up to 80 per cent of the applied phosphorus may get fixed in the soil. Therefore, only a marginal proportion of the soil phosphorus is present as P ions in the soil solution. At the current rate of usage, readily available sources of phosphate rocks will be depleted over the next 60 to 90 years (Raghothama, 1999). Although P ions can reach larger concentrations in highly fertilized soils, their concentration in the soil solution is in the micromolar range, ranging between 0.1 and 10 μm (Frossard *et al.*, 2000; Rae *et al.*, 2004). These are rather low compared with the adequate P concentration required for optimal plant growth, which can reach values of several millimoles for most demanding crops like bean, pea, cotton, potato, tomato etc.

Low soil P availability is of particular concern in tropical and subtropical regions of the world and for regions of Mediterranean basin that are largely dominated by calcareous and alkaline soils (Batjes, 1997). This is largely due to the peculiar mineralogy and ambient geochemistry of these soils that favour a strong retention of P ions on their solid constituents and maintain low levels of P ions in soil solution.

To adapt with low phosphate availability, which is a common situation in most natural and agricultural ecosystems, plants have evolved sophisticated metabolic and developmental strategies to enhance phosphate acquisition and remobilization. A higher P acquisition efficiency can be related to the development of a more extensive root system in association with or without mycorrhizal fungi, or specialized roots

like proteoid roots or root hairs and changes in root physiology allowing the uptake of P at lower concentrations in the soil solution and /or uptake of P from insoluble inorganic or organic forms (Gaume *et al.*, 2001).

In India, rubber is mainly cultivated in Kerala, parts of Tamil Nadu, Karnataka and north eastern states like Tripura, Assam and Meghalaya. Total area under rubber in India is 5.67 lakh ha, of which about 71 per cent is under mature rubber. More than nine lakh small holders depend on rubber for their livelihood. The price of rubber has been fluctuating in recent years and reducing the cost of cultivation of rubber is very important for sustaining rubber industry.

Growth management of cultivated plants on the basis of nutrioperiodism emphasizes the different nutritional requirements of crop plants at different growth stages. In the case of rubber, results from fertilizer experiments in India and other rubber growing countries have established that balanced application of nitrogen, phosphorus and potassium improve the growth during the early years of immaturity. During later phase of immaturity, nutrient release from ground covers and litter sets in and the response to fertilizer application was found to be variable. During mature stage, no consistent response to fertilizer application was noticed from various experiments in India and other countries. Lack of response to application of phosphatic fertilizers was reported from many experiments (Punnoose *et al.*, 1975; Sivanadyan *et al.*, 1995). Plants growing in soils with low available P status had leaf P status comparable with that of plants growing in P rich soil. Experiments initiated at the Rubber Research Institute of India in 1997 on mature rubber did not indicate any significant positive response to application of P fertilizers. In this context, the present investigation was taken up with the following objective.

To study the P dynamics in plant and soil and to investigate the possibility of reducing the present dose of P fertilizers for mature rubber by taking in to account the nutrient accumulation in dry matter, nutrient recycling through litter and microbial role in P availability to arrive at a cost effective fertilizer schedule for rubber.

Review of Literature

2. REVIEW OF LITERATURE

The investigation was carried out to explore the possibility of reducing the dose of phosphorus fertilizers to mature rubber, which is an important crop of Kerala, parts of Tamil Nadu and Karnataka.

Lack of consistent response of mature rubber to application of phosphatic fertilizers has been well documented in literature. Reports of lack of response even when the P status of the soil was very low are also available. However the mechanisms by which rubber plants acquire phosphorus under low/ deficient soil P status have not been studied so far. Very little information is available on other tree crops also. Hence the literature available on other crops pertaining to their mechanisms or adaptations to survive under low soil P status are reviewed, besides available literature on the response of rubber trees to applied P in India and other countries.

2.1 RESPONSE OF RUBBER TO APPLIED PHOSPHORUS

Widespread deficiency of available phosphorus and potassium in soils under *Hevea* in the traditional rubber growing areas was reported by George (1961) and Amma *et al.* (1976). A survey of rubber growing tracts of Kerala and Kanyakumari district of Tamil Nadu indicated that 75 per cent of the area have low available P, 15 per cent have medium and nine per cent have high available P status (NBSS and LUP, 1999).

2.1.1 Effect of Fertilizer Application on Growth

Beneficial effect of fertilizer application in improving the growth of rubber was reported by several workers (George, 1964; Punnoose *et al.*, 1975; Kalam *et al.*, 1979; Sivanadyan *et al.*, 1995). However in many experiments, the response was confined to the early years of immaturity. Ananth (1966) reported lack of response to nitrogen and phosphorus from

fifth year onwards and it was attributed to the large quantity of nutrients released from legume ground covers. In the red loam soils of South India, Punnoose *et al.* (1975) also reported lack of response to major nutrients applied from the fifth year onwards except for a marginal increase obtained by increasing K from 50 to 100 kg ha⁻¹, when leguminous ground covers were established with rubber. Nitrogen and phosphorus gave good response in the initial years of immaturity period, while potassium had no measurable effect. According to George *et al.* (1997) application of P @ 30 kg ha⁻¹ enhanced the growth of young rubber in a low P soil, but further increase in the dose to 60 kg ha⁻¹ did not indicate any additional beneficial effect.

2.1.2 Effect of P Application on Yield

Majority of the fertilizer experiments on mature rubber revealed the positive effect of N and K fertilizers in improving the yield (Pushparajah, 1969; Punnoose *et al.*, 1975; Pushparajah *et al.*, 1975). However phosphatic fertilizers had little or no effect. Pushparajah (1969) reported that application of adequate phosphatic fertilizers during immature period resulted in a build up of P in the soil and hence application of P did not increase yield during mature phase. Positive N and P interaction was also reported. Punnoose *et al.* (1975) observed an increasing trend in yield due to P application when soil P was low to medium. In an experiment conducted in Liberia, Guha (1975) noticed improvement in yield due to phosphorus application. Significant N and P interaction was also noticed. In another experiment P application was found to decrease yield. It was concluded that general manuring of mature rubber was not economically justifiable. Lim (1977) also suggested restriction of P application during mature phase. Taking in to account the residual effect of fertilizers applied during immaturity and likely levelling off of nutrient immobilization within the tree with age, Watson (1989) concluded that fertilizer application could be suspended for four years after the commencement of tapping and thereafter only

sufficient N should be applied to replace that lost through latex. After reviewing the fertilizer experiments conducted in rubber, Sivanadyan *et al.* (1995) concluded that fertilizer application does not necessarily increase yield in mature rubber plantations which were well managed in the immature phase. Application of phosphorus during immature period result in sufficient build up of P in the top soil of mature stand which can sustain the phosphorus requirement for some years of maturity against the phosphorus outflow through latex. It was suggested that *Hevea* stand act as a self-sustaining ecosystem with respect to phosphorus, provided adequate phosphorus fertilizers were applied in the immature phase. Experiments conducted on mature rubber revealed the lack of response to P in improving the yield of rubber even in low available P soils (RRII, 2000).

2.1.3 Effect of Nutrients on Latex Stability

Phosphorus and potassium are important nutrients that affect the stability of the latex. Philpot and Westgarth (1953) reported beneficial effects of P and K on stability of the latex. Balanced Mg/P ratio is essential for the stability of the latex. Phosphorus applied as rock phosphates increased the P and Ca content of latex and reduced Mg/P ratio. Mg/P ratio is an important factor influencing the pre-coagulation on tapping cut and hence would influence plugging (Beaufils, 1957).

2.1.4 Effect of P Application on Leaf Nutrient Status

Shorrocks (1960) observed that the effect of rock phosphate was neither rapid nor marked on leaf P concentration. Guha (1969) and Pushparajah and Teng (1972) reported the positive effect of rock phosphates in increasing the leaf P concentration. According to Kalam *et al.* (1979) application of P had no effect on the leaf nutrient status. Pushpadas *et al.* (1979) observed a deficiency of available P in soils which did not receive P fertilizers but no P deficiency in leaf samples. It

was suggested that trees could be meeting their P requirement from the organic forms resulting from cover crop residues and leaf litter of rubber. Joseph and Ranganathan (1996) derived optimum values of P in rubber leaves as 0.26 per cent through DRIS approach. In the experiments conducted in South India the leaf P status was generally maintained in the medium range even when the available P status of the soil was very low (RRII, 2000).

The critical levels of leaf nutrient concentration (%) in the medium range are 3.00-3.50, 0.20-0.25, 1.00-1.50 and 0.20-0.25 for N, P, K and Mg respectively, below which the concentrations are considered to be low and above which the concentrations are considered to be high (Krishnakumar and Potty, 1992).

2.1.5 General Fertilizer Recommendation

The general fertilizer recommendation by Rubber Research Institute of India for mature rubber is 30:30:30 kg NPK ha⁻¹ year⁻¹. Wherever possible, farmers are advised to adopt discriminatory fertilizer application based on soil and leaf analyses (Pushpadas and Ahmed 1980).

The literature cited above indicate lack of consistent response of mature rubber to fertilizer application. Majority of the experiments also revealed the lack of response to P application on growth and yield during the mature phase when adequate fertilizers were applied during immature phase.

2.2 INFLUENCE OF P NUTRITION ON ROOT DEVELOPMENT

Nutrients have profound effects on many aspects of root development. Many of these effects are specific to particular nutrients and are strongly dependent on the genotype of the plant. In most cases, the developmental responses are adaptive, they serve to increase the

efficiency of nutrient capture under situations of nutrient limitation (Forde and Lorenzo, 2001).

2.2.1 Root System of Rubber

In mature rubber plantations, the greatest root proliferation was observed in the top soil and proliferation decreased rapidly with depth. Greatest root proliferation was noticed in 0-7.5 cm soil layer and proliferation decreased rapidly with depth until at 30-45 cm soil layer, the amount of feeder roots was only about one tenth of the total amount present in the whole 0-45 cm. About 50 per cent of the feeder roots in the 0-45 cm layer were in the top 7.5 cm (Soong, 1976). In mature rubber plantations of seven to eight years, lateral roots extended over 9 m and tap roots were about 2.4 m deep (Webster and Paardekooper, 1989). Philip *et al.* (1996) observed greatest root concentration in fertilizer enriched area in a five year old plantation.

2.2.2 P Status and Root Development

Fohse and Jungk (1983) reported two to three fold increase in root surface area of tomato, rape and spinach when the phosphate concentration of the growing medium was decreased from 100 to 2 micromoles. This increase resulted from the increase in root hair length and decrease in the root hair density.

Phosphorus mobility in soil is governed by diffusion rather than mass flow and hence root phosphorus uptake is limited by localized phosphorus depletion around the root. The root architecture, growth and configuration of the root system in time and space is very important for the acquisition of P. Spatial variation in phosphorus availability results from its inherently low mobility, the spatial variation in factors controlling its availability, such as microbial activity, soil pH and colloid chemistry (Lynch and Brown, 2001). Lopez-Bucio *et al.* (2002) reported that at P limiting conditions, *Arabidopsis* root system underwent

major architectural changes in terms of lateral root number, lateral root density and primary root length.

Phosphorus deficiency elicited changes in branching patterns, total root length and root hair length. Species with greater root length, greater root hair length and specific branching patterns demonstrated increased P uptake (Bates and Lynch, 1996).

2.2.3 Root Hairs and P Uptake

Bates and Lynch (2001) reported that the length and density of root hairs are extremely plastic in response to soil P availability. After comparing the P acquisition of root hairless mutant and wild type of *Arabidopsis*, it was concluded that root hairs increased the competitiveness of plants under low P availability, but are neutral at high P conditions. It was suggested that the competitive advantage of root hairs is due to greater P uptake. Ma *et al.* (2001) demonstrated that low P availability increased root hair length and density, shortened tip to first root hair distance and increased number of epidermal cell files that bear root hairs in *Arabidopsis*.

2.2.4 Root Development in Crops under Low P Conditions

Gaume *et al.* (2001) observed increased root production as one of the mechanisms that allow maize genotypes to adapt to soils which are low in available P. Some of the other mechanisms observed were increased secretion of acid phosphatase and organic acids by roots.

Bosse and Koch (1998) reported that in tomato, under conditions of P deficiency, the above ground biomass decreased for the benefit of root growth, the root/ shoot ratio increased from 0.25 in P sufficient plants to 0.45 in P deficient plants. Steingrobe *et al.* (2001) observed that total root production of winter barley was two to three times higher under conditions of P deficiency.

According to Krasilnikoff *et al.* (2003) cowpea genotypes differed in root hair length, root length and intensity of root induced processes transforming non-labile soil P to labile soil P.

The literature cited indicated role of soil P status in influencing the root development of annual crops. These plants adapt to deficient soil P status by producing more roots and modifying the root architecture. However such studies are lacking in tree crops.

2.3 NUTRIENT REMOVAL THROUGH LATEX

According to Dijkman (1951), the quantity of nutrients removed through 2000 kg of dry rubber was 15.20, 6.00, 11.20 and 0.60 kg N, P₂O₅, K₂O, and CaO respectively. The average composition of nutrients in the latex was 0.23, 0.09, 0.17 and 0.01 per cent N, P, K, and Ca respectively. Pushparajah *et al.* (1971) reported that 1454 kg of dry rubber removed 7.60 kg N, 1.70 kg P, 5.10 kg K and 2.10 kg Mg. According to Krishnakumar and Potty (1992), the nutrient removal through latex can be considered negligible. Samarappuli and Yogaratnam (1997) observed that 9 kg N, 2 kg P, 8 kg K and 2 kg Mg were removed through 1400 kg of dry rubber. According to Gunatilleke (2002), 9.40 kg N, 2.30 kg P, 8.30 kg K and 1.70 kg Mg were lost through 1400 kg of dry rubber.

2.4 NUTRIENT ADDITION THROUGH LITTER

Dijkman (1951) reported that N equivalent to 700 kg of ammonium sulphate, P₂O₅ equivalent to 250 kg of double super phosphate and K₂ O equivalent to 120 kg of potassium sulphate are returned every year through litter in a hectare planted with 200 rubber trees. Huat and Pushparajah (1985) observed that the average content of nutrients in the litter was 1.36, 0.03, 0.34, 1.00, and 0.16 per cent N, P, K, Ca and Mg respectively. The C/N ratio of the litter was 33. According to

Krishnakumar and Potty (1992) approximately 6 tonnes of organic matter is added through annual leaf fall every year.

In a study conducted in the Rubber Research Institute of India, 95.60 kg N, 2.78 kg P, 47.77 kg K, 61.10 kg Ca 16.17 kg Mg 1.58 kg Mn, 0.38 kg Cu and 0.28 kg Zn were released from the litter in one hectare during the period from February to September. The quantity of litter disintegrated during this period was 4694.90 kg i.e., 94.12 per cent of the total litter. The average content of nutrients in the litter was 2.02, 0.06, 0.97, 1.31, and 0.34 per cent N, P, K Ca and Mg respectively (RRII, 1996). Verghese *et al.* (2001) reported that the total annual litter addition in a 14 year old RRIM 600 plantation with a density of 317 trees per ha ranged from 6.8 to 7.8 t per ha in Tripura. Nutrients ranging from 94 to 130 kg of N, 5 to 6 kg of P, 22 to 25 kg of K, 106 to 168 kg of Ca and 17 to 33 kg of Mg were returned to the soil through litter fall.

Sivanadyan *et al.* (1995) reported that rubber trees stringently retrieve the lost nutrients from the decomposing shed parts. A thin mat of freshly formed white roots invade the shed parts actively and feed directly on them.

2.5 P FRACTIONS

Chang and Jackson (1958) reported that phosphatic fertilizers added to the soil change in to three forms – Al-P, Fe-P, and Ca- P in acidic and neutral soils. According to Fiskell and Spencer (1964) as time elapses, Al-P shifts to Fe-P and with prolonged contact of added P with soil, such a shift takes place irrespective of soil pH. Anjaneyulu and Omanwar (1979) reported that total P increased significantly over control as the rate of P application increased in a slightly acidic soil. The increase in total P was mainly due to increase in the organic fraction.

Lee *et al.* (1990) reported that increased microbial activity reduced sorption of dissolved and inorganic P by soil, maintained

inorganic P in soluble and labile pools, increased microbial P, decreased mineral P and increased exchangeable aluminium. Vaz *et al.* (1993) studied the influence of soil acidity and P fertilization on P fractions and found that total dissolved phosphorus, dissolved reactive phosphorus, dissolved organic phosphorus and dissolved organic carbon increased as soil pH and P status increased.

According to Thomas *et al.* (1999), the labile P pools in soil were not affected by vegetation type. The resin extractable P was strongly correlated with soil carbon content suggesting that soil organic matter is the likely proximate source of plant available P. Nziguheba *et al.* (2002) reported that though P fertilization increased maize yield, soil labile P did not increase correspondingly.

Amma *et al.* (1991) observed saloid P in trace amounts in most of the rubber growing soils. Fe-P was the most predominant inorganic fraction in all the soils studied. Positive correlation between available P and Al-P was also reported. George *et al.* (2001) reported that Al-P and Fe-P are the major inorganic P fractions in rubber growing soils.

2.6 INFLUENCE OF P NUTRITION ON PLANT ENZYMES

2.6.1 Acid Phosphatases

Acid phosphatases are a general group of hydrolytic enzymes that have a wide range of functions in plant metabolism. Intracellular phosphatases accumulate in vacuoles and play an important role in the scavenging of P for metabolic redistribution. Increased intracellular and extracellular acid phosphatases could improve the acquisition and reutilization of P, thus helping plant growth under P- deficient condition (Yun and Kaeppler, 2001).

Phosphatases are capable of hydrolysing a widespread of organic phosphate esters. A possible direct involvement of phosphatases in

inorganic P uptake was also reported. It was suggested that phosphatases are very important to P nutrition of terrestrial plants (Antibus and Lesica, 1990).

Phosphatase is a dual function enzyme. In addition to catalysing the hydrolysis of organic P compounds, it can transfer a phosphate group from pyrophosphate to glucose to form glucose-6-phosphate which is the first intermediate in the translocation process. Consequently, root phosphatase activity may respond positively to the presence of inorganic P (Speir and Cowling, 1991).

2.6.1.1 Acid Phosphatase Activity and P Status

Using a leaf disc acid phosphatase assay for diagnosis of P starvation, Elliott and Lauchli (1986) found that acid phosphatase activity increased 2-3 times in leaves of P deficient maize plants compared with P sufficient leaves. McLachlan *et al.* (1987) observed increased intracellular acid phosphatase activity in wheat shoots under P-deficient conditions.

Production of acid phosphatases is considered to be an integral part of the plant response to P deficiency (Duff *et al.*, 1994). Induction of acid phosphatases in response to P starvation is a universal response in higher plants (Raghothama, 1999).

2.6.1.2 Acid Phosphatase Activity in Roots

Root phosphatase activity is a significant factor of nutritional efficiency under limited mineral phosphorus supply. Plant roots with high phosphatase activity had the potential to utilize soil organic phosphorus (Helal 1990). Antibus and Lesica (1990) reported that phosphatase activity of vascular epiphytes was positively correlated with plant P content in a Costa Rican forest. It was suggested that root surface phosphatase activity provided access to soil organic P which is the dominant form in organic soils. According to Tadano and Sakai (1991).

secretion of acid phosphatase from roots increases under P- deficient conditions, but levels of secretion differ remarkably among plant species.

McLachlan (1976) showed that root extracellular acid phosphatase activities of cereals and clover were related to productivity and that activities increased in all species as they become more P deficient. It was concluded that phosphatase activity could be used to distinguish species better adapted to use the existing P in low-fertility situations.

Speir and Cowling (1991) studied the effect of season on root phosphatase activity and reported that there was a sharp decline in root phosphatase activity in late summer autumn, and activities were highest during late winter/early spring.

Ascencio (1997) observed a twenty fold increase in root phosphatase activity under conditions of P- deficiency. Similar increases in the root phosphatase activity under P stress conditions were also reported by Adams and Pate (1992), Tadano *et al.* (1993) and Hayes *et al.* (1999).

Bosse and Koch (1998) observed that activities of P mobilizing enzymes (acid phosphatase, phytase and ribonuclease) in P deficient roots of 14 day old tomato seedlings were 32-fold higher than in the control plants. However the seedlings did not show any visible deficiency symptoms. Yun and Kaeppler (2001) observed that under P starvation, intracellular and secreted acid phosphatase activity increased substantially in leaves and roots of maize. Gaume *et al.* (2001) reported increased root development and exudation of acid phosphatase under P-deficient condition in maize. Effects on root formation and acid phosphatase were greater for the low- P tolerant genotypes than for the low- P susceptible varieties.

Jianzhong *et al.* (2001) suggested increasing the acid phosphatase secretion of plant roots through genetic engineering as a possible means for improving the adaptation of plants to P deficiency.

Coello (2002) reported that *Arabidopsis* roots responded to the absence of an exogenous phosphate source with an increase in the specific activities of secreted acid phosphatase. Increase in enzyme activity was noted two days after P- withdrawal and reached a maximum in six days. The activity was stimulated by calcium and inhibited by molybdate, phosphate, fluoride, vanadate and nitrate.

Ascencio (1996) reported lack of consistent response in root phosphatase activity to plant nutrition.

The literature reviewed clearly indicate that under conditions of P deficiency, acid phosphatase activity in the leaves and roots increase and roots secrete acid phosphatase to rhizosphere. Hence even though P deficiency symptoms are not exhibited by the plants, a hidden P stress is indicated by the increased acid phosphatase activity in plants.

2.6.2 Phosphoenol Pyruvate Carboxylase (PEP Carboxylase) and Malate Dehydrogenase (MDH) Activities in Roots

The cytosolic phosphoenol pyruvate carboxylase catalyses the irreversible β -carboxylation of phosphoenol pyruvate (PEP) using HCO_3^- as a substrate in a reaction that yields oxaloacetic acid and inorganic phosphate (Fontaine *et al.* 2003)

Gardner and Boundy (1983) explained the first evidence linking organic acid exudation from root to solubilization of poorly available soil P and enhanced P uptake. Specialised proteoid roots (dense bottle brush like clusters of roots) of white lupin (*Lupinus albus*) were shown to exude citrate, and it was proposed that citrate improved the P nutrition of the plant by releasing fixed forms of soil P.

Increased organic acid exudation in response to P deficiency has been reported by many scientists (Nagarajah *et al.*, 1970; Dinkelaker *et al.*, 1989; Bar-Yosef, 1991; Jones, 1998).

An increase in the exudation of citrate increased soil solution P by solubilizing Ca phosphate due to decrease in pH in the rhizosphere and by desorption reactions in acid soils (Hocking, 2001).

Johnson *et al.* (1996) demonstrated that increased citric and malic acid secretion from proteoid roots under P deficiency was correlated with the increased activity of several enzymes involved in organic acid synthesis, including phosphoenol pyruvate carboxylase. Neumann and Romheld (1999) observed an increase in the PEP carboxylase and aconitase activity in the roots of P-deficient wheat plants

A positive correlation was found between organic acid exudation from roots of rape seed and white lupin and enzymes involved in organic acid biosynthesis like PEP carboxylase and malate dehydrogenase (Hocking, 2001).

Gaume *et al.* (2001) observed that organic acid contents in the root tissues were increased under P deficiency and were related to increased PEP carboxylase activity.

Although a lot of literature is available on the exudation of organic acids by roots under P deficient condition, very little information is available on the PEP carboxylase and malate dehydrogenase activities in roots under conditions of P stress.

2.7 INFLUENCE OF P NUTRITION ON SOIL ENZYMES

Studies on enzyme activities in soil are important as they indicate the potential of soil to carry out the biochemical processes which are important to maintain soil fertility. The level of enzyme activity can be used as an indicator of soil fertility (Skujins, 1976). Ceccanti and Garcia (1994) suggested that the importance of soil enzyme activities lies in their participation in the evolution of organic matter and processes related to it. Because of their sensitivity, ease, and low cost of

measurement, soil enzyme activities may be key variables for soil assessments related to sustainability (Park and Seaton, 1996).

Kiss *et al.* (1975) suggested that enzymes accumulated in soil have biological significance as they participate in the cycling of elements and thus play a very important role in the initial phases of the decomposition of organic residues.

2.7.1 Soil Dehydrogenase

Beyer *et al.* (1992) identified dehydrogenase enzyme as an integral part of soil microorganisms and hence an indicator of soil microbial activity. According to Dormar *et al.* (1994), dehydrogenase activity in soils provides correlative information on the biological activity and microbial population. Garcia *et al.* (1997) also reported dehydrogenase as an enzyme which illustrated the microbial activity of the soil and the degree of soil degradation. According to Hayes *et al.* (1999) organic P in soil accounts for upto 80 per cent of total soil P. Hence soil dehydrogenase activity has an indirect role in the availability of phosphorus.

Bergstrom *et al.* (1998) suggested that dehydrogenase activity is a respiratory measurement and hence is more strongly representative of the size and activity of viable microbial community than the activity of other soil enzymes, which exist in viable cells and as enzymes stabilized in soil matrix. It was suggested that dehydrogenase activity may be more representative of the recent soil management practices, and the other enzymes more representative of historical management to the extent that part of their activities derive from stabilized enzymes.

Bolton *et al.* (1985) compared the dehydrogenase activity of conventional and organic systems and concluded that the activity was higher in the organic systems. Serra-Wittling *et al.* (1995) reported that highest level of dehydrogenase activity coincided with the mineralization flush at the beginning of an incubation period and both were related to intense activity of the microflora in degrading easily metabolizable compounds. As the easily biodegradable substances decreased,

dehydrogenase activity and microbial respiration decreased. Kumar and Kapoor (1995) noticed that dehydrogenase activity declined with increasing soil pH. Garcia *et al.* (1997) reported that there was no high correlation between soil dehydrogenase activity and organic matter content in degraded soils. It was concluded that organic matter was not as sensitive as dehydrogenase enzyme to some types of stress.

According to Fraser *et al.* (1988) dehydrogenase activity was related to the level of carbon substrates in the soil in a sandy loam soil. Cooper and Warman (1997) suggested that while studying enzyme activity, soil texture must be considered since microbial activities are lower in fine textured soils than coarse textured soils exposed to similar management conditions. Tateno (1998) observed increased activity of dehydrogenase due to application of poultry manure in a clay loam soil.

Doran (1990) observed that various tillage practices affect microbial biomass and hence reduced dehydrogenase activity. Bergstrom *et al.* (1998) reported higher dehydrogenase activity under no tilled situation.

2.7.2 Acid Phosphatases in Soil

Although organic P in soil accounts for up to 80 per cent of total soil P, it is considered to be unavailable to plants unless first mineralised by the action of phosphatases. In addition to the hydrolysis of soil organic P compounds, phosphatases partake in the hydrolysis and subsequent re-uptake of P esters leaked from plant roots (Tarafdar and Claassen, 1988; Hayes *et al.*, 1999).

2.7.2.1 Sources of Phosphatases in Soil

According to Torrani (1968), phosphatases originate in the soil from bacteria, fungi, yeasts, protozoa, mycorrhiza and plant roots. Harrison (1983) suggested a positive relationship between phosphatases and organic matter content since the enzyme was found bound to humic

protein complex. Ladd (1985) reported high level of phosphatase activity in the rhizosphere which was attributed to the high microbial activity promoted by plant residues.

2.7.2.2 Phosphatase Activity and P Status

Nielson and Eiland (1980) reported that phosphatase activity and ATP content in P depleted soil depended partly on the content of plant available P and partly on the root intensity in soils. Phosphatase enzymes are inducible under conditions of low P and this activity varies considerably among plant species and for mycorrhizal roots (Dodd *et al.*, 1987). Attiwill and Adams (1993) reported that the activity of phosphatase enzyme increased with a low concentration of soluble inorganic P in forest soils. Clarholm and Rosengren- Brinck (1995) observed a feedback mechanism for P, where low needle P concentrations in Norway spruce forest were associated with increased acid phosphatase activity in the humus layer. Production of extracellular phosphatase enzymes is also dependent on environmental conditions, the physiological state of the plant, root type, root age and position of the root (Antibus *et al.*, 1997).

Tarafdar and Jungk (1987) measured acid phosphatase activity in the rhizosphere of wheat and clover corresponding to a depletion of soil organic phosphates across this zone and suggested that rhizosphere phosphatases play an important role in the release of inorganic P from soil organic P for subsequent uptake by plants.

Production of extracellular and intracellular phosphatases increases during P starvation. Due to their low substrate specificity, they are involved in the non-specific hydrolysis of organic P resulting in the elevation of inorganic P (Duff *et al.*, 1994). Baldwin *et al.* (2001) suggested that extracellular phosphatases are likely to be involved in recapturing inorganic P from organic phosphorus compounds found in the extracellular matrix.

Increased secretion of acid phosphatases has been reported in many plants under P deficient conditions (Elliott and Lauchli, 1986; Tadano and Sakai, 1991; Trull *et al.*, 1997). The enzyme activity in temperate pasture legumes and grasses was significantly higher in extracts from roots grown in the absence of P relative to roots supplied with P (Hayes *et al.*, 1999). Yun and Kaeppler (2001) observed increased secretion of acid phosphatase from maize roots under P starvation. Differences in the quantity of enzyme secreted was also noticed among genotypes indicating differences in the P acquisition efficiency of different genotypes. Coello (2002) reported that *Arabidopsis* roots responded to the absence of an exogenous phosphate source with an increase in the specific activities of secreted acid phosphatases.

The preceding literature indicate that acid phosphatase activity in soil increases with the decrease in soluble P status of the soil. Increased secretion of phosphatases under P deficient conditions is an adaptive mechanism of many plants to survive under P such conditions.

2.8 RHIZOSPHERE pH AND P AVAILABILITY

Plant roots are responsible for considerable changes of rhizosphere pH which arise mostly from the release of H^+ or OH^-/HCO_3^- ions to counter balance the net excess of cations or anions entering the roots. When an excess of cations over anions are taken up, the plant roots compensate by releasing excess positive charges as protons, thereby resulting in acidification of rhizosphere. Conversely, when an excess of anions over cations are taken up the excess negative charges are released as hydroxyls or bicarbonate ions leading to alkalization of the rhizosphere (Nye, 1981 and Hinsinger, 1998, 2001).

2.8.1 Rhizosphere Acidification under P Deficiency

Enhanced H^+ release as a response to P deficiency was reported by many scientists (Imas *et al.*, 1997; Bertrand *et al.*, 1999; Neumann

and Romheld, 1999). Such a phenomenon was observed to be localized behind the root tips (Gregory and Hinsinger, 1999). Enhanced acidification of the rhizosphere might be related to an inhibition of NO_3^- uptake in response to P deficiency, and to a consequent increase in the excess of cation over anion uptake (Kirk and Le Van Du, 1997; Neumann and Romheld, 1999).

2.8.2 Rhizosphere pH and Availability of Soil P

Rhizosphere pH has a strong influence on the bioavailability of soil P. Riley and Barber (1971) obtained a wide range of pH values in the rhizosphere of pot grown soybean by supplying N as NH_4^+ and NO_3^- . Lower pH values were obtained with NH_4^+ than with NO_3^- . It was observed that the amount of P taken up by soybean increased linearly with decreasing rhizosphere pH. Root induced acidification of the rhizosphere which occurred for plants fed with NH_4^+ resulted in an enhanced bioavailability of soil P. Various studies with phosphate rocks indicated that H^+ release by plant roots could considerably increase the dissolution of phosphate rocks and bioavailability of P in the rhizosphere (Hinsinger, 1998). Some plants like buck wheat, oilseed rape and legumes were particularly efficient at using P from phosphate rocks due to their particular ability to release H^+ (Zoysa *et al.*, 1998).

2.8.3 Rhizosphere pH and Availability of Soil P in Acidic Soils

Root induced acidification of the rhizosphere dramatically increased the bioavailability of inorganic P whenever Ca phosphates were present *i.e.*, mostly in alkaline to mildly acidic soils. Its effect in acidic soils was not clear except when a source of Ca phosphates such as phosphate rock was added to soils (Hinsinger and Gilkes, 1996; Zoysa *et al.*, 1998). Fe and Al phosphates which are the dominant forms of P minerals in acid soils have a decreasing solubility with

decreasing pH. Besides, the positive surface charge of Al and Fe oxides and hence their P adsorption capacity increases with decreasing pH. Hence an increase in the rhizosphere pH might be more efficient in increasing bioavailability of inorganic P in acid soils (Hinsinger, 1998).

Gahoonia *et al.* (1992) noticed that rye grass fed with NH_4^+ took up less P from an acid soil than when fed with NO_3^- . Gahoonia and Nielson (1992) reported that rhizosphere alkalization resulted in better availability of inorganic P to oil seed rape grown in a comparatively more neutral soil. The $\text{OH}^-/\text{HCO}_3^-$ released by roots of plants fed with NO_3^- desorbed P ions from metal oxides *via* ligand exchange reactions.

Conversely, some experiments revealed that decreasing the pH of acidic soils could also lead to increasing solubility of soil P. Geelhoed *et al.* (1997) observed that decreasing the pH lead to less P adsorption on to goethite and hence in larger equilibrium concentration of P ions in soil solution. It was suggested that competitive adsorption of sulphate ions considerably increased with decreasing pH and thus resulted in larger equilibrium P concentration at acidic pH of about 4-5 than at pH 6-7. This indicated that H^+ release in the rhizosphere might increase the bioavailability of P sorbed on to metal oxides like goethite.

According to Youssef and Chino (1989) some plants could increase rhizosphere pH in acidic conditions and decrease in neutral or alkaline conditions, indicating the capacity of plants to adapt to adverse soil conditions. Marschner *et al.* (1991) reported that species growing naturally in very acidic soils like Norway spruce alkalized their rhizosphere.

Considering the large number of processes and reactions involved in soils that most often contain different forms of inorganic P with opposite geochemical behavior it is difficult to predict to what

extent and in which direction (positively or negatively) the bio-availability of soil P will respond to a change in soil pH.

2.9 ROLE OF MICROORGANISMS IN P NUTRITION

2.9.1 Role of Mycorrhizal Fungi (AMF) in the Uptake of Phosphorus by Plants

Mycorrhizae are widespread under natural conditions and occur in nearly all soils. They are found in association with most vascular plants except a few belonging mainly to the Chenopodiaceae, Cruciferae, Cyperaceae, Juncaceae and Proteaceae (Harley and Harley, 1987). The effect of mycorrhizae in increasing plant growth has been well documented and is mainly attributed to an enhancement of P nutrition of plants (Tinker, 1978; Xu *et al.*, 2001 ; Gyaneshwar *et al.*, 2002).

2.9.1.1 Environmental Conditions

The extent of mycorrhizal infection in root systems is influenced by environmental conditions. The most important are the age of the plant, the level of phosphate relative to the requirement of the plant in the soil and the capacity of the mycorrhizal propagules in the soil to form association. Percentage infection is frequently inversely related to soil P supply, though small additions of P to very deficient soils may increase infection (Smith *et al.*, 1992). Kahiluoto *et al.* (2001) reported that mycorrhization could be improved by omitting P application.

2.9.1.2 Mechanism of Increased Uptake of P by Mycorrhizal Plants

In most of the studies involving mycorrhizae, the mycorrhizal association increased the growth of plants solely by enhancing the uptake of nutrients especially P (Tinker, 1978; Fontela *et al.*, 2001). The rate of uptake of nutrients by mycorrhizal plants is faster than that by non mycorrhizal plants (Sanders and Tinker, 1973; Son and Smith, 1988). Sanders and Tinker (1973) observed that the rate of inflow of P in to

mycorrhizal roots was much higher (17×10^{-14} moles $\text{cm}^{-1} \text{s}^{-1}$) than that of non-mycorrhizal plants (3.6×10^{-14} moles $\text{cm}^{-1} \text{s}^{-1}$). Assuming that the difference in the rate of inflow was caused by mycorrhizal hyphae, the rate of inflow of P into the hyphae was calculated to be six times the rate in to the root hair.

Cress *et al.* (1979) opined that at very low concentration, mycorrhizal plants absorbed almost twice as fast as did non-mycorrhizal plants. It was observed that the increase in absorption rate was primarily due to increase in the affinity for absorption.

According to Mosse *et al.* (1973), in some soils only mycorrhizal plants could take up labelled phosphorus. Non-mycorrhizal plants could take up P only when the concentration of P in the soil solution was increased indicating that mycorrhizal roots had a lower threshold concentration for absorption than non-mycorrhizal roots.

Harley (1989) suggested that the production of phosphatases by mycorrhizal fungi is important in the solubilization of organic phytates, which constitute a large fraction of total phosphate in humic soils. Calculations of hyphal contribution to total phosphatase activity in the soil are in the range of two to four per cent (Joner *et al.*, 2000). Ectomycorrhizae produced large amounts of calcium oxalate, which might be involved in the chelation of iron and aluminium and thereby released P for plant uptake (Treeby *et al.*, 1989).

Differences in the uptake of cations and anions between mycorrhizal and non-mycorrhizal plants lead to differences in the rhizosphere pH and such changes affected the availability of adsorbed P to plants (Buwalda *et al.*, 1983). Increased uptake of calcium by mycorrhizal plants was suggested as a possible reason for the increase in P uptake from poorly soluble calcium phosphate by mycorrhizal plants (Bolan 1991). Wallander *et al.* (1997) reported that *Suillus variegates* improved

the P uptake of pine seedlings by reducing the pH of the soil. It was observed that mycorrhizal plants utilized ammoniacal nitrogen more efficiently than non-mycorrhizal plants. It was suggested that extrusion of hydrogen ions which accompany uptake of ammonium ion will take place from mycorrhizal hyphae and roots increasing the bio-availability of P.

2.9.1.3 Physical Exploration of the Soil

Mycorrhizal fungi in association with plant roots increase P uptake by more thorough exploration of soil volume thereby making positionally unavailable nutrients available. This is achieved by decreasing the distance for diffusion of phosphate ions and by increasing the surface area for absorption (Tinker, 1978).

Phosphorus in soil solution reach plant roots mainly by diffusion. Gerdemann (1968) estimated that mycorrhizae would increase P uptake 60-fold when diffusion limits the uptake whereas the increase would be 10 fold when diffusion is not limiting uptake. Sanders and Tinker (1973) concluded that the extensive hyphal growth of mycorrhizae effectively short circuits the distance for diffusion and thereby increases the uptake. Hattingh *et al.* (1973) found that mycorrhizal hyphae could intercept labelled P placed 27 cm from a mycorrhizal root, whereas it remained unavailable to non-mycorrhizal roots. Owusu-Bennoah and Wild (1980) showed that the radius of the depletion zone for P around mycorrhizal onion roots was twice that for non-mycorrhizal roots.

It was observed that greater responses to mycorrhizal infection occurred in coarse rooted plants than in fine rooted plants, in high P sorbing soils than in low P sorbing soils and in soils than in solution cultures (Bolan, 1991). Baon *et al.* (1994) reported that rye plants with short root hairs were more responsive to mycorrhizal infection.

Increase in the absorption of P by mycorrhizal plants has been attributed to increase in the surface area for absorption (Sanders and

Tinker, 1973). The mycorrhizal hyphae is very fine and it increases the surface area for greater absorption of nutrients and can enter in to pores in the soil and organic matter that can not be entered by root hairs.

2.9.1.4 Storage of Absorbed Phosphorus

Mycorrhizal hyphae can store larger amounts of absorbed P than plant roots, thus facilitating continued movement of P into hyphae. Inorganic phosphate absorbed by hyphae is stored in three forms; soluble orthophosphate, soluble polyphosphates and polyphosphate granules. In addition to storage in these three forms, there is a continued movement of phosphate during absorption into the host tissue which involves 10 per cent of that being absorbed. The bulk of the phosphate is stored as polyphosphates, mainly in the granular forms (up to 40 per cent of total P). Phosphate in solid form and located within the hyphal vacuoles, is suited to fulfil the role of a storage reserve in an osmotically inactive form (Bolan, 1991).

2.9.1.5 Sources of P for Mycorrhizal and Non-mycorrhizal Plants

In most of the studies comparing the effectiveness of P sources for mycorrhizal and non-mycorrhizal plants, it was observed that the increase in the plant growth due to AMF infection was larger with poorly soluble P than with soluble P (Bolan 1991). Several studies indicated greatest benefit from mycorrhizal inoculation with the least soluble source of iron phosphate (Yao *et al.*, 2001). Wallander (2000) observed that pine seedlings inoculated with mycorrhizal fungi were able to use calcium phosphates. The explanation for the increased uptake of P by the AMF plants suggested was that the mycorrhizae explored the soil volume more thoroughly and so found more of the point sources of P. Having found the sources they also increased the rate of uptake from the sources by increasing the diffusion gradient either by a closer approach to the source or by achieving a low concentration of phosphate at the surface. A

further possibility suggested was that mycorrhizal hyphae might be able to chemically modify the availability of iron phosphate by producing organic compounds with chelating properties, such as citrate.

2.9.1.6 Mycorrhiza Development as a Modifier of Nutrition

According to Smith *et al.* (1992) the important points to be considered when P efficiency of a crop is studied are whether the species forms mycorrhizal association. Crucifers including *Arabidopsis*, do not and hence their response is not complicated by mycorrhizal interactions. For a mycorrhizal species, the magnitude of dependency varies. Some species are strongly or obligately dependent, even in high P soils (e.g., *Stylosanthes*, *Cassava*, oil palm etc.) and in these plants, it may be very important to consider the potential of mycorrhizae. Others although infect, respond little and infection will not be a significant factor. The majority of plants are intermediate. They suggested that the infectivity of the soil, differences between trial sites and the effects of nutrient application on the rate and extent of mycorrhizal infection also should be considered.

2.9.1.7 Mycorrhizal Inoculation

Chulan and Regu (1986) reported that inoculation with mycorrhizal fungi enhanced the P content in the leaves of cacao plants in unsterile soil compared to plants growing in sterile soil. Improved growth and nutrient uptake with mycorrhizal infection was reported in many perennial crops like oil palm, *Citrus* sp., *Theobroma cacao*, etc and trees like *Acacia* sp., *Casuarina* sp., *Pinus* sp., etc. (Vogt *et al.*, 1997). Xu *et al.* (2001) observed that inoculation with mycorrhizal fungi enhanced the stand volume and growth of eucalyptus plants in P deficient soil.

Waidyanatha (1980) observed improved P uptake by *Hevea* plants with mycorrhizal infection. However, no effect was noticed on the growth of plants. A reduction in mycorrhizal root colonisation with P fertilization in

Hevea plants was reported by Ikram *et al.* (1992, 1996). They observed improved growth and nutrient uptake with inoculation when P was limiting.

The literature cited clearly indicate that mycorrhizal infection plays an important role in the P nutrition of plants. Response to applied P and the ability of plants to survive in low P soil are greatly influenced by the mycorrhizal association.

2.9.2 Role of P Solubilizers in P Uptake

Many bacterial, fungal, yeast and actinomycete species capable of solubilizing sparingly soluble P in pure culture have been isolated from soil and rhizosphere samples and they increase the availability of P to plants (Kucey *et al.*, 1989). In soil, P solubilizing bacteria constitute 1-50 per cent and fungi 0.5-1 per cent of the total respective population (Gyaneswar *et al.*, 2002). According to Zahir *et al.* (2004), mycorrhizosphere interaction between bacteria and fungi affect P-cycling, thus promoting a sustainable nutrient supply to plants.

2.9.2.1 Mechanism of P Solubilization

The ability to solubilize Ca-P complexes has been attributed to their ability to reduce the pH of their surroundings, either by the release of organic acids or protons (Bardiya and Gaur, 1972; Gaur, 1990). Oxalic acid has been reported as the major organic acid produced by most of the P solubilizing fungi. However, some species produced citric acid as the major organic anion (Whitelaw, 2000).

Whitelaw (2000) reported that N source affected P solubilization in liquid medium studies. Assimilation of ammonium often favoured higher acid production and greater solubilization of P in comparison to nitrate.

Inoculation with P-solubilizing fungi could enhance the utilization of soil phosphate by plants. Although the organic acid produced in the

rhizosphere would not remain untouched for long enough to affect the bulk P release from soil, it is possible that the short term reduction of the rhizosphere pH and the complexation of cations could produce an effective P solubilizing microenvironment, resulting in increased P uptake by the plant roots (Kucey *et al.*, 1989).

Majority of the P solubilizers solubilize Ca-P complexes and only a few can solubilize Fe-P and Al-P (Kucey *et al.*, 1989) Hence these P-solubilizers are effective in calcareous soils in which Ca-P complexes are present, but not in other soils in which phosphates are complexed with Fe and Al ions. However these could be effective even in these soils when supplemented with rock phosphate and P solubilization can be induced under P starvation (Gyaneswar *et al.*, 2002).

2.9.2.2 Crop Response to Inoculation

Tandon (1987) reported 10-15 per cent increase in crop yields in 10 out of 37 experiments due to inoculation with P solubilizers. Inoculation with P solubilizers enhanced growth and increased P contents in the tissues, but large variations were reported in the effectiveness (Kucey *et al.*, 1989).

Ralston and McBride (1976) reported that inoculation with P solubilizing bacteria enhanced seedling growth of red pine seedlings as well or better than soluble phosphate fertilizer. Increases in the yield of chick pea due to inoculation with P solubilizers was reported by Ahmad and Jha (1977) and of rice by Kundu and Gaur (1984). Chabot *et al.* (1996) reported that inoculation with P solubilizing *Rhizobium leguminosarum* biovar. *phaseoli* enhanced P uptake and growth of maize and lettuce. Whitelaw *et al.* (1997) found that treatment of wheat with *Penicillium radicum* increased grain yield by 14 per cent in the field and increased both P uptake (10 %) and yield (9%) in the greenhouse.

The ability of many rhizosphere and soil microorganisms to solubilize sparingly soluble P and increase P availability to plants has been well documented in the literature. However under field conditions, inoculation with these microorganisms did not give any consistent beneficial effect.

2.10 P BUDGETING

Pushparajah (1969) reported that application of adequate phosphatic fertilizers to rubber trees during immature period resulted in build up of P in the soil and hence application of P did not increase yield during mature phase. Lim (1977) suggested restriction of P application during mature phase taking in to account the residual effect of fertilizers applied during immaturity and likely levelling off of nutrient immobilization within the tree with age.

In a study of P budgeting, Smaling (1993) estimated annual losses of P through runoff and erosion to be 10 kg ha^{-1} when P was applied at 12 kg ha^{-1} . Buresh *et al.* (1997) recommended two strategies for building of soil P capital- to apply large quantities of P fertilizers, assuming that the applied P represented a long term investment with long lasting residual effects or seasonal small additions to replace crop uptake with gradual replenishment of P stocks. Contrary to this, Nziguheba *et al.* (2002) observed lack of long lasting residual effect of applied P. Cumulative addition of 750 kg P ha^{-1} failed to sustain maximum maize yield for more than two seasons after cessation of P addition, even though large amount of P could be measured in soil. It was suggested that continuous small additions of P fertilizers might be better in low P soils. Considering P losses due to run off and erosion, application of P @ 10 kg ha^{-1} was not found to be sufficient, but application of P @ 25 kg ha^{-1} resulted in gradual replenishment.

Considerable accumulation of residual P in the plough layer due to continuous P application was reported by Nambiar (1994) in long term manurial and intensive cropping trials in India.

The literature reviewed clearly indicate the lack of consistent response of mature rubber to application of P fertilizers and the several strategies evolved by plants to improve P acquisition under low P situations.

Materials and Methods

3. MATERIALS AND METHODS

The present investigation to explore the possibility of reducing the dose of P fertilizers for mature rubber was conducted in an ongoing fertilizer experiment initiated by the Rubber Research Institute of India, Kottayam in 1997 for studying the effect of P fertilizers on growth and yield of mature rubber. Detailed investigation was carried out during 2002-2004. The experiment area received the same treatments throughout. The details of the experimental site, weather conditions, materials used and methods adopted are presented.

3.1 EXPERIMENTAL SITE

3.1.1 Location

The experiment was conducted in Malankara estate, Thodupuzha, Idukki District, situated at $9^{\circ}50'$ N latitude and $76^{\circ}48'$ E longitude, at an altitude of 115 m above mean sea level.

3.1.2 Soil

The soil of the experimental site was sandy clay loam and belonged to the order Ultisol and suborder Humult. The soil type was Ustic Kandihumults. Before commencement of the experiment in 1997, the soil was medium in organic carbon, low in available P and available K, medium in exchangeable Mg and acidic in reaction. The physicochemical characteristics of the experimental site are presented in Table 1.

3.2 WEATHER

The experimental area experiences a typical warm humid tropical climate. The rainfall distribution of the experimental area is shown in Fig. 1 and Appendix I.

Table 1. Soil characteristics of the experimental site

Parameter	Unit	Content in soil	Method used
Mechanical composition			International pipette method (Piper, 1966)
Sand	%	57.51	
Silt	%	10.98	
Clay	%	31.51	
Texture		sandy clay loam	
Chemical properties			
Soil reaction (pH) 1: 2.5 water		4.69	pH meter with glass electrode (Jackson, 1958)
Organic carbon	%	1.06	Walkley and Black's method (Jackson, 1958)
Available P	kg ha ⁻¹	18.80	Bray No II extraction (Bray and Kurtz, 1945), Chlorostannous reduced molybdophosphoric blue colour in HCl system
Available K	kg ha ⁻¹	50.00	Ammonium acetate extraction, Flame photometric method
Exchangeable Mg	cmol kg ⁻¹	0.07	Ammonium acetate extraction
Total P	kg ha ⁻¹	740	Vanadomolybdophosphoric yellow colour in H ₂ SO ₄ system (Jackson, 1958)
Physical properties			
Bulk density	g cc ⁻¹	1.28	Core method (Gupta and Dakshinamoorthy, 1980)
Particle density	g cc ⁻¹	2.34	-do-
Porosity	%	45	-do-

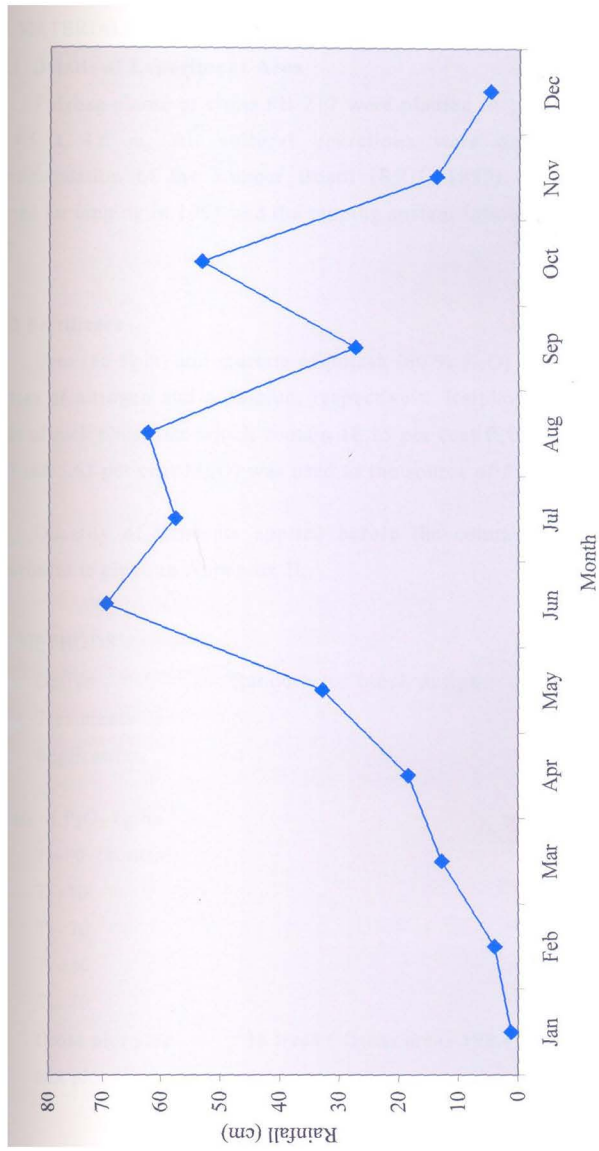


Fig. 1. Mean monthly rainfall (cm) during 1997-2004

3.3 MATERIALS

3.3.1 Details of Experiment Area

Polybag plants of clone PB 217 were planted in 1984 at a spacing of 4.6 x 4.6 m. All cultural operations were done as per the recommendation of the Rubber Board (RRII, 1980). The trees were opened for tapping in 1991 and the tapping system followed was 1/2S d/4 6d/7.

3.3.2 Fertilizers

Urea (46 % N) and muriate of potash (60 % K₂O) were used as the sources of nitrogen and potassium respectively. Rajphos (an indigenous grade of rock phosphate which contain 18.15 per cent P₂O₅, 38.30 per cent CaO and 5.63 per cent MgO) was used as the source of P.

Quantity of nutrients applied before the commencement of the experiment is given in Appendix II.

3.4 METHODS

Design : Randomised block design

Treatments : 5

Replications : 4

Levels of P₂O₅ kg ha⁻¹

T₁- 0 (control)

T₂- 10

T₃- 20

T₄- 30

T₅- 40

Gross plot size : 36 trees (Gross area- 598.48 m²)

Net plot size : 10 trees

Nitrogen and potassium were applied uniformly in all the treatments at the standard recommended rate ($30 \text{ kg ha}^{-1} \text{ year}^{-1}$) throughout the experimental period. Fertilizers were applied in two equal splits during April-May and September-October every year as broadcast in between the rows of rubber trees.

3.5 OBSERVATIONS

The experiment was commenced in 1997. Pretreatment observations were recorded during 1997-98 and treatment imposition was commenced in 1998. Detailed investigation was carried out during 2002 - 2004. Data before commencing the detailed investigation was collected from Rubber Research Institute of India.

3.5.1 Growth Characters

Observations were recorded from all the net trees and mean values were computed.

3.5.1.1 *Girth*

Girth of individual trees was measured at half yearly intervals at a height of 150 cm above the bud union during 2003-04 and expressed as cm.

3.5.1.2 *Length of Tapping Panel*

Length of tapping panel was measured during January 2004 and expressed as cm.

3.5.1.3 *Bark Thickness*

Bark thickness of individual plants was recorded during January 2004 using a bark measuring guage and expressed as mm.

3.5.2 Root Characters

3.5.2.1 *Root Spread*

For measuring the root spread, they were traced from the base of the tree and the distances covered by the roots were measured and area explored was expressed as m^2 .

3.5.2.2 Root Volume

Root volume was estimated by displacement method. A known volume of the soil (3.74 m³) was excavated and roots were separated. Roots were immersed in a container full of water and the volume of displaced water was measured and expressed as m³ per tree.

3.5.2.3 Vertical Distribution of Roots

The vertical distribution of fine roots was determined by separating roots from a known volume of soil up to a depth of 90 cm, washing and recording the dry weight. The percentage distribution of fine roots in various soil layers were computed.

3.5.2.4 Fine Root Production

Fine root production was measured by in growth core method as described by Steingrobe *et al.* (2001). Mesh bags (length 22 cm, diameter 6 cm and mesh width 3mm) were pulled over a plastic tube of the same size and inserted in to the soil at an angle of 45⁰. In each plot, four mesh bags were inserted, two between trees and two between rows (Appendix III). The mesh bags were inserted during January 2003. For filling the mesh bags, soil collected from the same plots were used after removing the roots through sieving. The root free soil was filled through the plastic tube into the mesh bag. The soil was compressed with a wooden stick to a density comparable to the average density of the bulk soil. The whole mesh bag was filled step by step with soil by repeating the procedure. The plastic tubes were pulled out after filling the net with soil. The bags were opened during August 2003 and roots were washed free of soil. Root length and root area in each mesh bag were measured using a root scanner (HP Scan Jet 6300C) and expressed as cm and cm² respectively.

3.5.2.5 Root density

Density of roots in the top 7.5 cm soil layer was measured using a core sampler. Two Samples were collected in two directions, between

trees and between rows (Appendix III). Four samples were collected per plot. Roots were separated by washing and sieving, and stored in 50 per cent alcohol until measurement. Root length and root area were measured using a root scanner (HP Scan Jet 6300C).

3.5.2.6 Root Hair Production

Root hair determination was made as described by Gahoonia *et al.* (1999) and Lopez-Bucio *et al.* (2002) with modifications. Soil cores with intact fresh roots were taken up to about 10 cm soil depth. The soil cores were immersed in water overnight in darkness at 5°C. The roots were then shaken gently for five minutes and washed free of soil carefully. Twenty root pieces of about 1 cm length were randomly chosen from each plot for root hair measurements. Root hair density and length were measured at random using a stereomicroscope at 10x magnification. Mycorrhizal strands associated with roots were excluded while counting.

3.5.3 Yield and Yield Attributes

3.5.3.1 Latex Yield

Yield of individual trees was measured once every month. Dry weight of a known volume of latex was determined and the mean annual yield was expressed as g tree⁻¹ tap⁻¹. Yield data during 1997-2002 was collected from the Rubber Research Institute of India

3.5.3.2 Dry Rubber Content of Latex (DRC)

Dry rubber content of latex was determined during 2003-2004. A known weight of latex was dried in a hot air oven at 80°C and the dry rubber content was determined and expressed as percentage.

3.5.4 Plant Characters

3.5.4.1 Leaf Nutrient Content

Leaf samples were collected at bimonthly intervals from May 2003 to November 2003 and analysed for phosphorus content (sampling periods I, II, III and IV). The samplings were done before and after fertilizer

application and analysed for various nutrients (sampling periods I, II, III and IV) as per the methods described by Jackson (1958). Samples were dried in a hot air oven at 80°C and powdered before analysis.

3.5.4.1.1 Nitrogen

Nitrogen content of leaves was determined by microkjeldahl method using a Nitrogen analyzer (Kjeltec 2300- Foss Tecator, Sweden)

3.5.4.1.2 Phosphorus

Phosphorus content of the leaves was determined by vanadomolybdate method using an autoanalyzer (AA 3-Brant Luebbe, Germany).

3.5.4.1.3 Potassium

Potassium content of the leaves was determined flame photometrically using an autoanalyzer.

3.5.4.1.4 Calcium

Calcium content was determined using atomic absorption spectrophotometer (Avanta-GBC Scientific Equipment Company Ltd., Australia).

3.5.4.1.5 Magnesium

Magnesium content was determined using atomic absorption spectrophotometer.

3.5.4.1.6 Micronutrients

Iron, manganese, zinc and copper content of the leaves was determined with the help of an atomic absorption spectrophotometer.

3.5.4.2 Root P and Micronutrient Status

Fine roots were collected, washed free of soil, dried in the oven at 80°C, powdered and analysed for phosphorus, iron, manganese, zinc and

copper at bimonthly intervals from May to November (sampling periods I, II, III and IV).

3.5.4.3 Dry Matter Accumulation

The total dry matter accumulation of trees was measured by destructive sampling of one sample tree in each treatment during 2003. The weight of leaves, branches, main stem and roots of the entire tree were recorded. Sub samples were dried in hot air oven at 80°C and dry weight was recorded. The total dry matter accumulation in leaves, branches, main stem and roots was estimated and expressed as kg per tree.

3.5.4.4 Nutrient Accumulation in Trees

Samples of leaves, twigs, branches, main stem and roots were collected from two trees in each treatment, dried in the oven at 80°C, powdered and analysed for nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc and copper following standard procedures.

3.5.4.5 Nutrient Removal Through Latex

Latex samples were collected at quarterly intervals, dried in the oven at 80°C and analysed for nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc and copper (RRIM 1973). Nutrient removal through latex was estimated from the total latex yield and nutrient content of the latex.

3.5.4.6 Cation Exchange Capacity (CEC) of Roots

The cation exchange capacity of roots was determined as per the method suggested by Crooke *et al.* (1964) and expressed as meq 100g dry root⁻¹.

3.5.4.7 Root Acid Phosphatase Activity

Root acid phosphatase activity was measured before and after fertilizer application. Fresh roots were collected from the field in ice buckets and stored at -80°C till enzyme assays were conducted (within one week of sample collection). Roots were washed gently under a stream of running water to remove soil and adherent organic materials

and enzyme assays were conducted as per the method described by Antibus and Lesica (1990) and expressed as μg nitrophenol mg^{-1} dry root.

3.5.4.8 *Phosphoenol Pyruvate Carboxylase (PEPC) and Malate Dehydrogenase (MDH) Activities in roots*

Fresh roots were collected from the field before and after second fertilizer application in ice buckets and stored at -80°C till enzyme assays were conducted. Roots were washed gently under tap water. Root PEPC and MDH activities were measured as per the methods described by Sadasivam and Manickam (1996) and expressed as $\text{nmol NADH oxidised ml enzyme extract}^{-1} \text{ min}^{-1} \text{ g FW}^{-1}$.

3.5.5 Nutrient Addition Through Litter Fall

Litter addition in each plot was quantified with the help of litter traps installed in the plots. Plastic nets of 1m^2 size were used as litter traps. The litter traps were set below the trees on wooden poles at a height of 30 cm from the ground. The quantity of litter collected in the litter traps was quantified at monthly intervals from February 2003 to January 2004 (from refoliation to wintering) and moisture content was determined by drying a sub sample at 80°C . The annual litter fall was calculated using the following formula

$$\text{Annual litter fall (kg ha}^{-1}\text{year}^{-1}) = \frac{\sum \text{Monthly litter collection in litter traps (kg)} \times 10000}{\text{Area of litter trap (m}^2\text{)}}$$

The litter collected from each plot was pooled and samples were dried in a hot air oven at 80°C and analysed for nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc and copper using standard procedures at quarterly intervals. From the total quantity of

litter added and the nutrient content of the litter, the nutrient addition through litter was calculated.

3.5.6 Soil Studies

3.5.6.1 Soil Chemical Properties

Soil samples (0-30 cm) were collected at bimonthly intervals from May 2003 to November 2003 and analysed for available phosphorus content (sampling periods I, II, III and IV). Soil samples were collected before and after fertilizer application and analysed for organic carbon, potassium and magnesium, iron, manganese, zinc and copper (sampling periods I, II, III and IV). Soil sample was mixed properly, dried in the shade and sieved through 2 mm sieve before analyses.

3.5.6.1.1 Organic carbon

Organic carbon content of soil was determined by Walkley and Black's method (Jackson, 1958).

3.5.6.1.2 Available Phosphorus

Available phosphorus was determined by chloromolybdic stannous chloride reduction method using Bray II extractant (Bray and Kurtz, 1945).

3.5.6.1.3 Available Potassium

Available potassium was determined flame photometrically.

3.5.6.1.4 Exchangeable Calcium

Exchangeable calcium content of the soil was determined using atomic absorption spectrophotometer.

3.5.6.1.5 Exchangeable Magnesium

Exchangeable magnesium content was determined using atomic absorption spectrophotometer.

3.5.6.1.6 Available Micronutrients

DTPA extractable iron, manganese, zinc and copper content of the soil were measured with the help of an atomic absorption spectrophotometer.

3.5.6.2 Phosphorus Fractionation

Soil samples (0-30 cm) were collected, dried in the shade, sieved through 2mm sieve and fractionated to saloid P, aluminium P (Al-P), iron P (Fe-P), calcium P (Ca-P), reductant soluble P and occluded P as per the method suggested by Peterson and Corey (1966) and expressed as ppm. Total phosphorus content of the soil (ppm) was determined by vanadomolybdophosphoric yellow color method (Jackson, 1958). The difference between total P and different P fractions was taken as the organic P.

3.5.6.3 Soil Enzymes

Soil enzymes were measured before and after fertilizer application (sampling periods I, II, III and IV) as per the methods described by Tabatabai (1994). Soil samples were dried in shade, sieved through 2 mm sieve and stored at 4°C till measurement.

3.5.6.3.1 Soil dehydrogenase

Soil dehydrogenase activity was measured as per the method described by Tabatabai (1994). Soil dehydrogenase was determined within five days of sample collection and expressed as $\mu\text{g TPF g}^{-1}\text{soil } 24 \text{ h}^{-1}$.

3.5.6.3.2 Soil acid phosphatase

Rhizosphere soil samples were collected for acid phosphatase assay. Acid phosphatase activity was measured within two days of sample collection as per the method described by Tabatabai (1994) and expressed as $\mu\text{g nitrophenol g}^{-1}\text{soil h}^{-1}$.

3.5.6.4 Rhizosphere and Non-rhizosphere Soil pH

Rhizosphere and non-rhizosphere soil pH was measured at bimonthly intervals from May to November (sampling periods I, II, III and IV). Rhizosphere samples were collected as per the method suggested by Bagayoko *et al.* (2000). The soil was dried in the shade, sieved and pH was measured at 1:2.5 soil water ratio using a pH meter with glass electrode.

3.5.7 Microbial Studies

Microbial studies were undertaken before and after fertilizer application (sampling periods I, II, III and IV).

3.5.7.1 Mycorrhizal Infection of Roots

Mycorrhizal infection of roots (%) was measured as per the method suggested by Phillips and Hayman (1970). The root segment was considered mycorrhizal if one of the three structures i.e., hyphae, arbuscules or vesicles were present.

Mycorrhizal colonisation was expressed using the following formula,

$$\text{Per cent colonisation} = \frac{\text{Number of root segments with AMF} \times 100}{\text{Total number of root segments examined}}$$

3.5.7.2 Enumeration of Microorganisms in the Rhizosphere

Feeder roots with adhering soil were collected from the field and stored at 4⁰C in polythene covers until they were processed. Rhizosphere microflora were enumerated following the serial dilution plate method (Timonin, 1940). Bacterial, phosphobacterial, actinomycetes and fungal populations were enumerated using soil extract agar, apatite agar, Kenknight's agar and Martin's rose bengal agar respectively. From appropriate dilutions, one ml was plated in agar plates in respective media. Counts of colony forming units were recorded after three days for bacteria, five days for fungi and seven days for actinomycetes and

expressed on dry weight basis. Percentage of phosphobacteria to total bacteria was computed.

3.5.8 Incidence of Tapping Panel Dryness (TPD)

Trees affected by TPD in each plot were enumerated and the percentage incidence of TPD was computed.

3.5.9 P Budgeting

P budgeting for mature rubber was attempted based on the data generated.

3.6 STATISTICAL ANALYSIS

The data collected from the experiment were analysed by applying the analysis of variance technique (Panse and Sukhatme, 1967). Wherever significant differences were observed between treatments, critical differences (CD at 5 per cent level) are provided for effective comparison of means. Correlation studies were also conducted to ascertain the relationship between various parameters.

Results

4. RESULTS

Detailed investigation was conducted to explore the possibility of reducing the dose of phosphorus fertilizers for mature rubber. The results obtained were statistically analysed and presented.

4.1 EFFECT OF PHOSPHORUS APPLICATION ON GROWTH CHARACTERS

Growth of rubber was measured in terms of girth, length of tapping panel and bark thickness.

4.1.1 Girth

Girth before commencement of the experiment (January 1998) and at half yearly intervals during the study period (January 2003, July 2003 and January 2004) and girth increment over pre-treatment (1998) are presented in Table 2. Girth was not significantly influenced by levels of phosphorus. Treatment which received phosphorus at the highest level (T_5) recorded 70.59, 71.48 and 72.28 cm girth and the treatment which did not receive any P fertilizer (T_1) recorded 70.44, 71.61 and 72.20 cm girth respectively during the first, second and third observations.

Table 2 Influence of P levels on girth and girth increment

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Girth (cm)				Girth increment (cm)		
	Jan 1998	Jan 2003	Jul 2003	Jan 2004	1998-Jan 2003	1998-Jul 2003	1998-Jan 2004
0 (T_1)	65.60	70.44	71.61	72.20	4.84	6.01	6.66
10 (T_2)	65.78	71.07	72.31	72.85	5.29	6.53	7.07
20 (T_3)	65.54	70.45	71.70	72.25	4.90	6.16	6.62
30 (T_4)	67.90	72.05	73.04	73.70	4.15	5.15	5.61
40 (T_5)	66.48	70.59	71.78	72.28	5.74	7.01	7.42
SE	0.85	1.93	2.04	2.07	1.16	0.30	1.32
CD	NS	NS	NS	NS	NS	NS	NS

Girth increment was also not significantly influenced by application of phosphorus. The treatment which received P at the highest level (40 kg P₂O₅ ha⁻¹ year⁻¹) recorded a girth increment of 5.74, 7.01 and 7.42 cm and T₄ which received the standard recommended dose of 30 kg P₂O₅ ha⁻¹ year⁻¹ recorded a girth increment of 4.15, 5.15 and 5.61 cm respectively during the first, second and third observations. The treatment which did not receive any P fertilizer (T₁) registered a girth increment of 4.84, 6.01 and 6.66 cm respectively during the first, second and third observations.

4.1.2 Length of Tapping Panel

Length of tapping panel recorded in January 2004 was not significantly influenced by application of phosphorus (Table 3). Length of tapping panel ranged from 49.13 in T₁ to 51.97 in T₄.

Table 3 Length of tapping panel and bark thickness as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Length of tapping panel, cm	Bark thickness, mm
0 (T ₁)	49.13	9.65
10 (T ₂)	50.92	9.48
20 (T ₃)	50.81	9.58
30 (T ₄)	51.97	9.90
40 (T ₅)	50.45	9.75
SE	0.80	0.02
CD	NS	NS

4.1.3 Bark Thickness

Bark thickness recorded in January 2004 was not significantly influenced by phosphorus application (Table 3).

4.2 INFLUENCE OF P LEVELS ON ROOT CHARACTERS

4.2.1 Root Spread and Root Volume

Rubber roots explored a remarkable soil area (Table 4). On an average, roots explored an area of 89.38 m². The root spread varied from 86.55 m² in T₃ which received P at 20 kg P₂O₅ ha⁻¹ year⁻¹ to 91.56 m² in T₅ which received the highest level of P. Root volume ranged from 0.043 m³ in T₃ to 0.054 m³ in T₅. The mean root volume measured was 0.047 m³ (Table 4).

Table 4 Influence of P levels on root spread and root volume

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Root spread, m ²	Root volume, m ³
0 (T ₁)	89.88	0.046
10 (T ₂)	88.20	0.054
20 (T ₃)	86.55	0.043
30 (T ₄)	90.72	0.046
40 (T ₅)	91.56	0.045
Mean	89.38	0.047

4.2.2 Vertical Distribution of Roots

The percentage of fine roots in different soil layers is presented in Table 5. The entire fine roots were considered to be confined to 0-90 cm layer. More than 60 per cent of the total fine roots explored top 10 cm layer and more than 20 per cent explored 10-30 cm layer.

Table 5 Effect of P on percentage of roots in different soil layers

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Depth of soil layer, cm			
	0-10	10-30	30-60	60-90
0 (T ₁)	63.82	29.34	5.70	1.14
10 (T ₂)	66.96	20.54	8.93	3.57
20 (T ₃)	69.98	20.24	7.16	2.62
30 (T ₄)	68.97	21.84	4.60	4.60
40 (T ₅)	60.00	27.44	10.70	7.42
Mean	65.95	23.88	7.42	2.75

4.2.3 Fine Root Production

Fine Root production between trees was more in T₁ which did not receive any P fertilizer (Table 6). Root area in T₁ (7.84 cm²) was significantly higher compared to all other treatments. All the P applied treatments had comparable root area and it was lowest in T₅, which received the highest level of P (1.31 cm²). Root length in T₁ was the highest (44.40 cm) and it was on par with T₂ and significantly greater than all other treatments. The treatments, which received P at 10, 20, and 30 kg P₂O₅ ha⁻¹ year⁻¹ (T₂, T₃ and T₄), were comparable. Root length was lowest in T₅ (4.87 cm) and it was comparable with T₃ and T₄.

Table 6 Effect of P levels on fine root production in 100 cm³ soil

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Root area, cm ²	Root length, cm	Root area, cm ²	Root length, cm
	Between trees		Between rows	
0 (T ₁)	7.84	44.40	1.95	13.64
10 (T ₂)	2.95	28.52	3.31	18.64
20 (T ₃)	2.21	17.39	4.95	21.73
30 (T ₄)	2.26	13.80	5.55	28.07
40 (T ₅)	1.31	4.87	4.97	16.57
SE	1.23	6.44	1.46	6.56
CD	3.79	19.85	NS	NS

Between rows, where fertilizers are applied, root production was more in the P applied treatments (Table 6). Root area and root length were lowest in T₁ (1.95 cm² and 13.64 cm respectively). No significant difference was noticed between treatments both in the case of root area and root length.

4.2.4 Root Density

Root density measured using a core sampler was higher in P applied treatments (Table 7). Root area and length were lowest in the

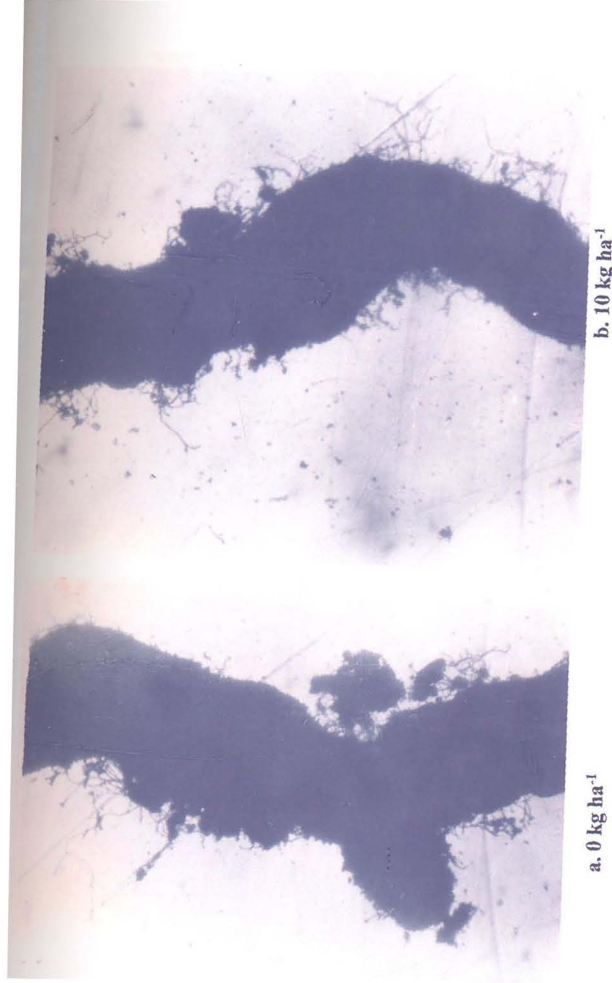
treatment which did not receive any P fertilizer (T_1) both between trees and between rows. No definite trend was observed among treatments which received P at various levels. However in most cases, root length and root area increased with increasing level of P. Root length between rows was significantly lower in T_1 . No significant difference was noticed between treatments which received phosphorus at 10, 20 and 30 kg ha⁻¹. The treatment which received the highest level of P (T_5) had a significantly higher root length compared to all other treatments (80.37 cm).

Table 7 Effect of P levels on root density in 100 cm³ soil

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Root area, cm ²	Root length, cm	Root area, cm ²	Root length, cm
	Between trees		Between rows	
0 (T_1)	3.69	34.88	3.49	29.64
10 (T_2)	7.68	44.71	5.93	42.97
20 (T_3)	5.68	46.76	6.35	53.28
30 (T_4)	9.53	52.36	5.66	48.64
40 (T_5)	8.81	54.83	8.56	80.37
SE	1.52	5.02	1.12	3.89
CD	NS	NS	NS	11.99

4.2.5 Length and Density of Root Hairs

In general, root hairs were shorter in the P applied treatments (Table 8 and Plate 1). Longest root hairs were observed in T_1 which did not receive any P fertilizer (0.63 mm). T_5 which received the highest level of P (40 kg P₂O₅ ha⁻¹ year⁻¹) had the shortest root hairs (0.27 mm) and it was significantly lower than that of other treatments. The root hair length of all the treatments except which received P at the highest level (T_5) was comparable.



a. 0 kg ha⁻¹

b. 10 kg ha⁻¹

Plate 1 Root hair production as influenced by levels of P ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$)



c. 20 kg ha⁻¹

d. 30 kg ha⁻¹

e. 40 kg ha⁻¹

Plate 1 Continued

Highest density of root hairs was observed in T₁ (67 mm⁻¹) and it was significantly higher compared to all other treatments (Table 8). Lowest density was observed in T₄ which received P at 30 kg P₂O₅ ha⁻¹ year⁻¹ (31mm⁻¹) and it was comparable with T₅. Treatments which received phosphorus at 10 and 20 kg ha⁻¹ were comparable, but significantly different from treatments which received P at higher levels.

Table 8 Influence of P levels on root hair length and density

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Length, mm	Density, mm ⁻¹
0 (T ₁)	0.63	67
10 (T ₂)	0.48	51
20 (T ₃)	0.53	48
30 (T ₄)	0.50	31
40 (T ₅)	0.27	36
SE	0.05	3.69
CD	0.15	11.36

4.3 INFLUENCE OF P LEVELS ON YIELD AND YIELD ATTRIBUTES

4.3.1 Yield

Yield of rubber from 1997-98 to 2003-04 is presented in Table 9. Yield was not significantly influenced by levels of phosphorus during the entire period. During 1997-98 (before treatment imposition), the yield ranged from 55.13 g tree⁻¹ tap⁻¹ in T₄ which was scheduled to receive P at 30 kg P₂O₅ ha⁻¹ year⁻¹ to 43.12 g tree⁻¹ tap⁻¹ in T₂ which was due to receive P at 10 kg P₂O₅ ha⁻¹ year⁻¹. During 1998-99, the yield ranged from 50.67 g tree⁻¹ tap⁻¹ in T₂ to 54.37 g tree⁻¹ tap⁻¹ in T₅ which received the highest level of P .

Table 9 Influence of P levels on yield (g tree⁻¹ tap⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	1997 – 98 (Pre-treatment)	1998-99	1999 - 00	2000-01	2001-02	2002-03	2003-04
0 (T ₁)	44.93	51.31	51.84	52.83	74.05	62.16	65.22
10 (T ₂)	43.12	50.67	48.84	59.94	72.60	66.53	70.81
20 (T ₃)	48.28	53.84	48.27	66.77	76.36	64.94	66.65
30 (T ₄)	55.13	52.56	53.86	64.13	79.28	69.69	66.25
40 (T ₅)	54.84	54.37	47.94	57.96	72.43	66.47	62.46
SE	4.51	3.72	3.72	4.34	2.05	4.08	4.99
CD	NS	NS	NS	NS	NS	NS	NS

During 1999-00, yield ranged from 47.94 g tree⁻¹ tap⁻¹ in T₅ to 53.86 g tree⁻¹ tap⁻¹ in T₄. The treatment which did not receive any P fertilizer (T₁), recorded 51.84 g tree⁻¹ tap⁻¹. The yield ranged from 66.77 g tree⁻¹ tap⁻¹ in T₃ which received P at 20 kg P₂O₅ ha⁻¹ year⁻¹ to 52.83 g tree⁻¹ tap⁻¹ in T₁ during 2000-01. During next year, there was an increase in yield in all the treatments and it ranged from 79.28 g tree⁻¹ tap⁻¹ in T₄ to 72.43 g tree⁻¹ tap⁻¹ in T₅. The treatment which did not receive any P fertilizer (T₁) recorded 74.05 g tree⁻¹ tap⁻¹. During 2002-03, there was a decline in yield in all the treatments and the values ranged from 62.16 g tree⁻¹ tap⁻¹ in T₁ to 69.69 g tree⁻¹ tap⁻¹ in T₄. During 2003-04, yield ranged from 70.81 g tree⁻¹ tap⁻¹ in T₂ to 62.46 g tree⁻¹ tap⁻¹ in T₅. The treatment which did not receive any P fertilizer during the entire period (T₁), recorded 65.22 g tree⁻¹ tap⁻¹.

4.3.2 Dry Rubber Content of Latex (DRC)

The DRC of latex measured at monthly intervals is presented in Table 10. There was no significant difference between treatments in the DRC of latex during the period under study (2003-04).

During April, DRC ranged from 38.38 per cent in T₄ to 36.30 per cent in T₁. There was a decline in DRC during May and it ranged from 34.71 per cent in T₂ to 30.21 per cent in T₃. During June, DRC ranged from 32.66 per cent in T₃ to 30.82 per cent in T₄ and in July from 31.14 per cent in T₄ to 33.96 per cent in T₂. There was an increase in DRC during August and September in most cases. During October, there was a decrease in DRC and it ranged from 33.04 per cent in T₁ to 36.45 per cent in T₃. Highest DRC was observed in T₅ (34.57 %) and lowest in T₂ (32.65 %) during November. During December there was an increase in DRC and it ranged from 39.20 per cent in T₅ to 36.53 in T₂. During January also, the same trend was observed. During February, the values ranged from 37.52 per cent in T₄ to 36.60 per cent in T₁ and during March from 36.10 per cent in T₄ to 37.36 per cent in T₁.

The mean DRC (2003-04) ranged from 34.83 per cent in T₁ to 35.63 in T₅.

4.4 EFFECT OF P APPLICATION ON LEAF NUTRIENT STATUS

Leaf P and other nutrients were determined four times during the period from May to November (I, II, III and IV)

The leaf P content was not significantly influenced by application of phosphorus (Table 11). Highest leaf P content was observed in the treatment which did not receive any P fertilizer (T₁) during the first sampling (0.23 %) and lowest in T₃ which received phosphorus at 20 kg ha⁻¹ (0.19 %). During the second sampling, highest leaf P content was observed in T₄ which received phosphorus at 30 kg ha⁻¹ (0.24 %) and lowest in T₅ which received P at the highest level (0.21 %). T₁, T₂ and T₃ contained 0.22 per cent P in their leaves. During the third sampling, highest leaf P content was observed in T₁ and T₅. During the last sampling, highest value was observed in T₄ (0.23 %) and lowest in T₂ and T₃ (0.20 %).

The content of nitrogen, potassium, calcium, iron, manganese, zinc and copper were also not influenced by application of phosphorus. Leaf N content varied from 3.18 to 3.34 per cent during first sampling, 2.85 to 3.03 per cent during second sampling, 3.25 to 3.36 per cent during third sampling and from 3.10 to 3.51 per cent during last sampling (Table 12).

Table 11 Influence of P levels on leaf phosphorus status (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.23	0.22	0.23	0.22
10 (T ₂)	0.20	0.22	0.21	0.20
20 (T ₃)	0.19	0.22	0.20	0.20
30 (T ₄)	0.20	0.24	0.20	0.23
40 (T ₅)	0.21	0.21	0.23	0.21
SE	0.01	0.01	0.01	0.01
CD	NS	NS	NS	NS

Table 12. Effect of P levels on leaf nitrogen status (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	3.34	2.85	3.29	3.10
10 (T ₂)	3.18	2.91	3.25	3.27
20 (T ₃)	3.23	3.02	3.30	3.21
30 (T ₄)	3.18	3.03	3.26	3.51
40 (T ₅)	3.22	2.92	3.36	3.25
SE	0.08	0.11	0.08	0.10
CD	NS	NS	NS	NS

In the case of leaf potassium content, no definite trend was observed during the different samplings (Table 13). Highest leaf K content was

observed in the T₁ during the first and third samplings (1.70 % and 1.66% respectively). During the second sampling, the leaf K content ranged from 1.64 per cent in T₁ to 1.84 per cent in T₂. During the last sampling, the leaf K status varied from 1.38 per cent in T₂ to 1.58 per cent in T₅.

Table 13 Leaf potassium content (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	1.70	1.64	1.66	1.57
10 (T ₂)	1.59	1.84	1.49	1.38
20 (T ₃)	1.61	1.76	1.39	1.42
30 (T ₄)	1.59	1.71	1.59	1.46
40 (T ₅)	1.66	1.73	1.58	1.58
SE	0.13	0.13	0.10	0.07
CD	NS	NS	NS	NS

Lowest calcium content was observed in T₁ during all the samplings (Table 14). All the treatments which received P at different levels had a higher leaf calcium status. However, there was no significant difference between treatments.

Table 14 Leaf calcium content (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.84	1.13	0.80	1.01
10 (T ₂)	0.92	1.19	0.98	1.28
20 (T ₃)	0.85	1.17	0.94	1.09
30 (T ₄)	1.05	1.28	0.87	1.12
40 (T ₅)	1.19	1.50	0.91	1.15
SE	0.11	0.10	0.02	0.13
CD	NS	NS	NS	NS

Magnesium content was higher in the treatments which received P after fertilizer application and the difference was significant during

fourth sampling (Table 15). All the P applied treatments had a significantly higher leaf Mg content compared to T₁ during this sampling.

Table 15 Effect of P levels on leaf magnesium content (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.39	0.34	0.41	0.28
10 (T ₂)	0.38	0.43	0.36	0.35
20 (T ₃)	0.39	0.39	0.36	0.36
30 (T ₄)	0.35	0.41	0.37	0.37
40 (T ₅)	0.40	0.42	0.36	0.36
SE	0.03	0.03	0.02	0.02
CD	NS	NS	NS	0.06

During the first sampling, leaf Fe content ranged from 286.42 ppm in T₂ to 402.30 ppm in T₃ (Table 16). During the second sampling, highest value was observed in T₃ (772.25 ppm) and the lowest in T₅ (387.75 ppm).

Table 16 Leaf iron content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	351.80	408.00	398.30	229.25
10 (T ₂)	286.42	549.00	465.15	346.45
20 (T ₃)	402.30	772.25	360.80	384.20
30 (T ₄)	348.27	428.50	396.60	308.75
40 (T ₅)	304.05	387.75	310.80	342.95
SE	44.41	199.70	66.76	59.04
CD	NS	NS	NS	NS

During the third sampling, the values ranged from 310.80 ppm in T₅ to 465.15 ppm in T₂. During the last sampling, the lowest value was observed in T₁ (229.25 ppm) and the highest in T₃ (384.20 ppm).

In the case of manganese also, no definite trend was observed between the levels of P and leaf content (Table 17). During the first sampling, leaf Mn content ranged from 140.45 ppm in T₂ to 201.47 ppm in T₄. During the second sampling, the values ranged from 181.25 ppm in T₂ to 223.50 ppm in T₄, from 130.15 ppm in T₂ to 171.55 ppm in T₃ during the third sampling and from 135.30 ppm in T₂ to 193.75 ppm in T₅ during the last sampling.

Table 17 Leaf manganese content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	150.60	204.00	142.35	172.15
10 (T ₂)	140.45	181.25	130.15	135.30
20 (T ₃)	182.05	207.00	171.55	152.10
30 (T ₄)	201.47	223.50	147.05	163.45
40 (T ₅)	189.35	217.50	141.65	193.75
SE	20.92	19.63	10.29	24.99
CD	NS	NS	NS	NS

In the case of leaf Zn content also, there was no significant difference between treatments (Table 18). However during the first and third sampling, the leaf Zn status was higher in T₁ which did not receive any P fertilizer. Leaf Zn status was maintained more or less in the same range during all the samplings. Among the various nutrients, the content of Zn was lowest in the leaves.

Table 18 Leaf zinc content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	47.60	42.50	42.45	46.65
10 (T ₂)	43.45	53.95	40.60	40.05
20 (T ₃)	43.60	54.30	40.15	41.20
30 (T ₄)	47.53	53.50	39.90	43.85
40 (T ₅)	40.55	47.55	40.45	47.95
SE	3.33	3.29	1.84	2.86
CD	NS	NS	NS	NS

Leaf copper content decreased from the first sampling to the fourth sampling (Table 19). During the first sampling, highest leaf Cu content was observed in T₁ (443.40 ppm) and lowest in T₄ (210.00 ppm). During the second sampling, highest value was observed in T₅ (119.5 ppm) and lowest in T₄ (81.75 ppm). During the third sampling, the values ranged from 70.95 ppm in T₄ to 95.10 ppm in T₂ and during the last sampling, from 37.80 ppm in T₄ to 60.40 ppm in T₃.

Table 19 Effect of P levels on leaf copper content (ppm)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	433.40	111.25	90.30	55.90
10 (T ₂)	342.93	107.33	95.10	55.15
20 (T ₃)	358.00	108.75	71.10	60.40
30 (T ₄)	210.00	81.75	70.95	37.80
40 (T ₅)	325.45	119.50	72.65	59.35
SE	92.59	17.16	12.53	10.12
CD	NS	NS	NS	NS

4.5 EFFECT OF P LEVELS ON THE CONTENT OF PHOSPHORUS AND MICRONUTRIENTS IN FINE ROOTS

After fertilizer application (second and fourth sampling), root P content was higher in P applied treatments (Table 20). However, there was no significant difference between treatments. Significant difference between treatments was observed only during the third sampling, when the root P content in the treatments which received P at lower levels (@10 and 20 kg P₂O₅ ha⁻¹ year⁻¹) was significantly lower than that of T₄ which received phosphorus at 30 kg ha⁻¹. The control treatment (T₁) which did not receive any P fertilizer had the same root P content as that of T₅ which received the highest level of P.

Table 20 Phosphorus content of fine roots (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.07	0.11	0.12	0.11
10 (T ₂)	0.08	0.12	0.11	0.12
20 (T ₃)	0.07	0.13	0.11	0.14
30 (T ₄)	0.09	0.13	0.13	0.16
40 (T ₅)	0.08	0.12	0.12	0.16
SE	0.01	0.01	0.004	0.02
CD	NS	NS	0.013	NS

The content of micronutrients in the roots was not influenced by P application during any of the sampling (Table 21 to Table 24). Root iron content varied from 0.57 to 2.46 per cent, root manganese content from 140.55 to 432.65 ppm, root zinc content from 43.45 to 115.35 ppm and root copper content from 181.95 to 636.50 ppm.

Table 21 Iron content of fine roots (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	1.09	0.94	0.62	2.27
10 (T ₂)	1.23	1.09	0.91	2.18
20 (T ₃)	1.27	1.21	0.68	2.19
30 (T ₄)	1.63	1.05	0.58	2.18
40 (T ₅)	1.95	1.19	0.57	2.46
SE	0.30	0.13	0.12	0.32
CD	NS	NS	NS	NS

Table 22 Root manganese content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	283.70	230.05	177.13	359.75
10 (T ₂)	151.90	250.65	160.93	367.45
20 (T ₃)	165.11	271.10	151.95	432.65
30 (T ₄)	194.60	254.80	140.55	284.15
40 (T ₅)	274.60	226.90	157.65	358.90
SE	31.56	22.66	13.08	36.12
CD	NS	NS	NS	NS

Table 23 Zinc content of fine roots (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	86.85	59.65	46.88	122.30
10 (T ₂)	104.20	65.50	44.33	106.35
20 (T ₃)	84.60	57.90	46.95	114.18
30 (T ₄)	115.35	65.55	48.00	102.65
40 (T ₅)	97.27	56.80	43.45	97.05
SE	9.26	3.74	2.98	7.55
CD	NS	NS	NS	NS

Table 24 Copper content of fine roots (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	401.55	463.75	246.07	512.10
10 (T ₂)	427.90	448.80	232.60	601.60
20 (T ₃)	296.47	416.35	300.20	636.50
30 (T ₄)	554.15	461.40	239.90	416.55
40 (T ₅)	476.06	405.60	181.95	533.80
SE	58.13	40.55	30.13	48.03
CD	NS	NS	NS	NS

4.6 INFLUENCE OF P LEVELS ON NUTRIENT ACCUMULATION IN TREES

4.6.1 Dry Matter Accumulation in Trees

The dry matter accumulation in different parts of the tree is presented in Table 25. The maximum dry matter was accumulated in the branches followed by main trunk and roots. The lowest dry matter accumulation was observed in the twigs followed by leaves. Of the total dry matter, 39.43 per cent was accumulated in the branches, 32.72 per cent in the main trunk and 20.63 per cent in the roots. There was no appreciable difference between treatments in the dry matter accumulation or partitioning.

Table 25 Dry matter accumulation in different parts of the tree (kg) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Roots	Main trunk	Branches	Twigs	Leaves	Total
0 (T ₁)	218.05	342.28	417.60	28.94	43.05	1049.92
10 (T ₂)	220.00	360.46	405.20	42.62	50.50	1078.78
20 (T ₃)	232.14	330.86	418.40	22.96	40.20	1044.56
30 (T ₄)	212.50	370.55	422.50	48.05	48.44	1102.04
40 (T ₅)	216.20	340.44	436.86	20.21	41.20	1054.91

4.6.2 Nutrient Accumulation in Different Parts of the Tree

The nutrient accumulation in different parts of a tree is presented in Table 26a to Table 26i. Among the nutrients, calcium was accumulated in the highest quantity followed by nitrogen and potassium. Among micronutrients, iron was accumulated in the highest quantity followed by copper. Accumulation of nitrogen varied from 7655.52 g in T₅ to 9439.14 g in T₂. Highest nitrogen accumulation was noticed in the branches in most of the treatments and lowest in the twigs. In the case of phosphorus also, maximum accumulation was observed in the branches. Roots and main trunk also had a large share of P in their dry matter. Lowest P accumulation was observed in the twigs. The total P accumulation varied from 755.98 g in T₃ to 863.59 g in T₄. Accumulation of potassium varied from 5642.80 g in T₁ to 6884.04 g in T₂. Accumulation of calcium varied from 14192.47 g in T₃ to 17162.30 g in T₂. Highest accumulation was observed in the branches followed by main trunk. Magnesium accumulation varied from 2376.33 g in T₅ to 2744.87 g in T₄. Accumulation of iron varied from 191.19 g in T₅ to 327.68 in T₃. Unlike other nutrients, maximum iron accumulation was observed in the roots in all the treatments. Accumulation of manganese varied from 45.85 g in T₁ to 55.00 g in T₂. Like iron, manganese accumulation was highest in the roots in most of the cases. Zinc accumulation varied from 32.58 g in T₃ to 41.38 g in T₄ and maximum accumulation was observed in the branches. Accumulation of copper varied from 170.31 g in T₅ to 182.22 g in T₂. Highest copper accumulation was observed in the main trunk and lowest in the twigs.

Table 26a Influence of P levels on nitrogen accumulation in dry matter
(g tree⁻¹)[‡]

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	1412.00	248.89	2463.84	1711.40	2235.01	8071.14
10 (T ₂)	1636.20	392.10	2917.44	2667.40	1826.00	9439.14
20 (T ₃)	1330.60	183.68	2635.92	1985.16	2425.86	8561.22
30 (T ₄)	1584.00	470.89	2070.25	2260.36	1487.50	7873.00
40 (T ₅)	1412.00	248.89	2463.84	1711.40	2235.01	8071.14

Table 26b Influence of P levels on phosphorus accumulation in dry
matter (g tree⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	94.71	28.04	250.56	205.37	228.95	807.63
10 (T ₂)	106.50	42.62	243.12	216.28	242.00	850.52
20 (T ₃)	88.44	20.66	251.04	198.52	197.32	755.98
30 (T ₄)	111.41	48.05	295.75	185.28	223.10	863.59
40 (T ₅)	90.64	20.21	305.80	204.26	227.01	847.92

Table 26 c Influence of P levels on potassium accumulation in dry matter (g tree⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	572.56	272.04	1336.32	1848.31	1613.57	5642.80
10 (T ₂)	712.05	396.37	2107.04	2018.58	1650.00	6884.04
20 (T ₃)	490.44	215.80	2008.32	1852.82	1416.05	5983.43
30 (T ₄)	668.47	456.48	2197.00	1778.64	1338.75	6439.34
40 (T ₅)	601.52	185.93	2184.30	1702.20	1902.56	6576.51

Table 26d Influence of P levels on calcium accumulation in dry matter
(g tree⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	680.19	376.20	8352.00	4244.00	3248.95	16901.34
10 (T ₂)	984.75	575.37	6483.20	5731.31	3388.00	17162.63
20 (T ₃)	438.18	321.44	6945.44	4003.41	2484.00	14192.47
30 (T ₄)	721.76	648.68	6886.75	4150.16	2273.75	14681.10
40 (T ₅)	704.52	284.96	7688.74	4085.28	3329.48	16092.98

Table 26e Influence of P levels on magnesium accumulation in dry
matter (g tree⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	180.81	121.55	835.20	684.56	675.96	2498.08
10 (T ₂)	237.35	183.27	729.36	684.87	616.00	2450.79
20 (T ₃)	152.76	84.95	794.96	661.72	835.70	2530.09
30 (T ₄)	208.29	201.81	845.00	852.27	637.50	2744.87
40 (T ₅)	152.44	76.80	830.03	646.84	670.22	2376.33

Table 26f Influence of P levels on iron accumulation in dry matter (g tree⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T ₁)	9.51	1.32	4.09	7.75	170.59	193.26
10 (T ₂)	5.03	6.00	7.74	10.16	152.43	191.36
20 (T ₃)	12.24	1.97	7.61	11.38	294.49	327.68
30 (T ₄)	17.08	5.77	5.20	7.71	197.61	233.37
40 (T ₅)	15.27	3.23	5.85	5.86	160.98	191.19

Table 26g. Influence of P levels on manganese accumulation in dry matter (g tree^{-1})

P_2O_5 levels, $\text{kg ha}^{-1} \text{ year}^{-1}$	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T_1)	6.97	1.43	7.14	13.14	17.17	45.85
10 (T_2)	5.84	2.22	8.91	17.37	20.66	55.00
20 (T_3)	6.15	1.46	10.95	13.10	20.52	52.18
30 (T_4)	8.74	2.34	9.21	19.31	14.56	54.16
40 (T_5)	6.39	1.47	8.91	14.67	17.03	48.47

Table 26h Influence of P levels on zinc accumulation in dry matter (g tree^{-1})

P_2O_5 levels, $\text{kg ha}^{-1} \text{ year}^{-1}$	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T_1)	1.93	1.22	14.20	11.08	7.65	36.08
10 (T_2)	1.95	1.60	14.76	10.60	9.11	38.02
20 (T_3)	1.45	0.74	12.33	8.87	9.19	32.58
30 (T_4)	2.47	2.33	16.14	11.56	8.88	41.38
40 (T_5)	2.07	1.00	14.90	11.23	10.57	39.77

Table 26i Influence of P levels on copper accumulation in dry matter (g tree^{-1})

P_2O_5 levels, $\text{kg ha}^{-1} \text{ year}^{-1}$	Leaves	Twigs	Branches	Main trunk	Roots	Total
0 (T_1)	6.29	4.05	40.53	89.03	36.52	176.42
10 (T_2)	4.80	4.19	35.90	86.55	50.78	182.22
20 (T_3)	4.20	2.28	38.53	82.98	51.77	179.76
30 (T_4)	5.77	5.30	40.48	86.00	36.04	173.59
40 (T_5)	4.91	1.88	36.74	88.92	37.86	170.31

4.7 INFLUENCE OF P LEVELS ON PLANT ENZYMES

4.7.1 Root Acid Phosphatase Activity

Trees which did not receive P fertilizers were found to secrete more acid phosphatase from roots than trees which were supplied with P (Table 27). During the first sampling, secretion of acid phosphatase from roots was highest from the control treatment (T₁) which did not receive any P fertilizer (2.30 μg nitrophenol mg^{-1} dry root), and it was comparable with plants which received phosphorus at 10 kg ha^{-1} (T₂), but significantly higher compared to other treatments which received higher levels of P. Secretion of acid phosphatase from trees which received phosphorus at 10 kg ha^{-1} was comparable with that from trees which received phosphorus at 20 and 30 kg ha^{-1} (T₃ and T₄), but significantly higher than that from T₅ which received P at the highest level.

Table 27 Influence of P levels on the secretion of acid phosphatase from roots (μg nitrophenol mg^{-1} dry root)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	2.30	1.78	1.80	2.05
10 (T ₂)	2.05	1.48	1.57	1.70
20 (T ₃)	1.70	1.48	1.45	1.46
30 (T ₄)	1.75	1.45	1.35	1.48
40 (T ₅)	1.58	1.40	1.45	1.49
SE	0.12	0.04	0.09	0.27
CD	0.37	0.13	0.27	NS

During the next sampling, a decrease was noticed in the secretion of acid phosphatase from roots. The quantity of secretion from the roots

of trees which received P at various levels were comparable. The trees which did not receive P fertilizers (T_1) had the highest acid phosphatase secretion ($1.78 \mu\text{g}$ nitrophenol mg^{-1} dry root) and it was significantly higher compared to the P applied treatments. During the third sampling also, the highest secretion of acid phosphatase was observed from T_1 ($1.80 \mu\text{g}$ nitrophenol mg^{-1} dry root) and it was comparable with that from trees which received phosphorus at 10 kg ha^{-1} ($1.57 \mu\text{g}$ nitrophenol mg^{-1} dry root). All the trees which were supplied with P at various levels had comparable levels of acid phosphatase secretion. During the next sampling also, the same trend was observed, but there was no significant difference between treatments. The acid phosphatase secretion from roots showed an increase during this period.

4.7.2 Phosphoenol Pyruvate Carboxylase (PEPC) and Malate Dehydrogenase (MDH) Activities in Roots

Phosphoenol pyruvate carboxylase (PEPC) and malate dehydrogenase (MDH) activities in roots were measured before and after second fertilizer application and the data are presented in Table 28.

Higher activities of PEPC and MDH were found in the root tissues of trees which did not receive P fertilizers. During the first sampling, no significant difference was noticed between treatments in the PEPC activity. However the activity was highest in T_1 which did not receive any P fertilizer ($9.35 \text{ nmol NADH oxidised ml}^{-1} \text{ enzyme extract min}^{-1} \text{ g}^{-1} \text{ FW}$). During the next sampling also, highest PEPC activity was noticed in T_1 ($19.6 \text{ nmol NADH oxidised ml}^{-1} \text{ enzyme extract min}^{-1} \text{ g}^{-1} \text{ FW}$) and the activity was significantly higher compared to P applied treatments. There was no significant difference between treatments which received P at different levels.

Table 28 PEPC and MDH activities in the roots (nmol NADH oxidised ml⁻¹ enzyme extract min⁻¹ g⁻¹ FW) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	PEPC		MDH	
	I	II	I	II
0(T ₁)	9.35	19.6	5.95	7.26
10(T ₂)	8.78	5.21	3.90	4.24
20(T ₃)	8.93	2.18	5.80	4.03
30(T ₄)	8.91	2.76	5.42	1.45
40(T ₅)	6.23	3.20	3.30	2.18
SE	1.31	1.44	0.64	0.78
CD	NS	4.43	1.96	2.41

In the case of MDH also, highest activity was observed T₁ during both samplings. During the first sampling, the MDH activity in T₁ was 5.95 nmol NADH oxidised ml⁻¹ enzyme extract min⁻¹ g⁻¹ FW and it was comparable with treatments which received phosphorus at 20 and 30 kg ha⁻¹. The activity was lowest in T₅ which received P at the highest level (3.30 nmol NADH oxidised ml⁻¹ enzyme extract min⁻¹ g⁻¹ FW). During the next sampling also, highest MDH activity was noticed in control treatment (7.26 nmol NADH oxidised ml⁻¹ enzyme extract min⁻¹ g⁻¹ FW) and it was significantly higher compared to the P applied treatments.

4.8 EFFECT OF P ON CATION EXCHANGE CAPACITY (CEC) OF ROOTS

No significant difference was noticed between treatments with respect to root CEC (Table 29). The CEC was lowest in T₁ (31.06 meq/100g dry root). All the P fertilized treatments had a higher CEC.

Table 29 Effect of P levels on root CEC

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	meq/100g dry root
0 (T ₁)	31.06
10 (T ₂)	31.50
20 (T ₃)	33.55
30 (T ₄)	33.08
40 (T ₅)	32.56
SE	1.26
CD	NS

4.9 INFLUENCE OF P LEVELS ON NUTRIENT RECYCLING THROUGH LITTER

4.9.1 Annual Litter Addition

The annual litter addition in different treatments were comparable (Table 30). The treatment which did not receive P fertilizers (T₁) had the lowest annual litter addition (7.12 t ha⁻¹) and the treatment which received P at the highest level (T₅) had the highest (7.85 t ha⁻¹).

Table 30 Litter addition (t ha⁻¹) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Annual litter addition	Litter addition during December-February
0 (T ₁)	7.12	3.72
10 (T ₂)	7.49	4.44
20 (T ₃)	7.80	3.50
30 (T ₄)	7.79	4.37
40 (T ₅)	7.85	4.52
SE	0.64	0.55
CD	NS	NS

4.9.2 Nutrient Content of Litter

Litter samples were analysed at quarterly intervals for major and micronutrients and the data are presented in Table 31 to Table 39. Among the nutrients, calcium was present in the highest quantity followed by nitrogen, potassium, magnesium, iron, phosphorus, manganese, copper and zinc. However, there was no significant difference between treatments with respect to the content of nutrients during any of the sampling periods. Litter samples collected during January (fourth sampling) had the lowest content of nitrogen, phosphorus, iron and zinc. Litter P content decreased from first sampling (April) to the fourth sampling (January). In the case of potassium, litter samples collected during January had the highest content and that collected in July (second sampling) had the lowest content. Highest calcium content was observed in the litter samples collected during October (third sampling) in most cases, and lowest in April (first sampling). Highest magnesium content was observed in April. In the case of manganese, highest content was observed in January and in the case of copper, highest content was observed in July.

Table 31 Nitrogen content of litter (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	1.72	1.98	1.86	1.38
10 (T ₂)	1.98	1.78	2.07	1.46
20 (T ₃)	1.57	1.74	1.96	1.45
30 (T ₄)	1.92	1.78	1.86	1.63
40 (T ₅)	1.51	2.16	1.73	1.44
SE	0.17	0.11	0.16	0.12
CD	NS	NS	NS	NS

Table 32 Effect of P levels on phosphorus content of litter (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.14	0.09	0.08	0.04
10 (T ₂)	0.14	0.09	0.09	0.04
20 (T ₃)	0.11	0.09	0.09	0.05
30 (T ₄)	0.14	0.09	0.11	0.05
40 (T ₅)	0.13	0.10	0.10	0.04
SE	0.01	0.004	0.01	0.01
CD	NS	NS	NS	NS

Table 33 Potassium content of litter (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.80	0.34	0.52	0.93
10 (T ₂)	1.00	0.37	0.42	0.95
20 (T ₃)	0.59	0.29	0.41	0.88
30 (T ₄)	0.77	0.29	0.51	0.87
40 (T ₅)	0.78	0.37	0.57	0.90
SE	0.09	0.05	0.05	0.04
CD	NS	NS	NS	NS

Table 34 Calcium content of litter (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	1.37	1.79	1.96	1.88
10 (T ₂)	1.35	1.82	2.15	1.84
20 (T ₃)	1.49	2.12	1.93	1.85
30 (T ₄)	1.53	1.78	2.12	1.77
40 (T ₅)	1.39	2.00	2.30	1.51
SE	0.12	0.12	0.13	0.13
CD	NS	NS	NS	NS

Table 35 Effect of P levels on magnesium content of litter (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.45	0.27	0.27	0.31
10 (T ₂)	0.49	0.33	0.30	0.30
20 (T ₃)	0.42	0.25	0.28	0.32
30 (T ₄)	0.51	0.27	0.31	0.29
40 (T ₅)	0.42	0.28	0.28	0.31
SE	0.03	0.02	0.03	0.01
CD	NS	NS	NS	NS

Table 36 Iron content of litter (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.20	0.45	0.54	0.06
10 (T ₂)	0.16	0.43	0.68	0.07
20 (T ₃)	0.14	0.24	0.58	0.07
30 (T ₄)	0.17	0.48	0.71	0.06
40 (T ₅)	0.22	0.51	0.61	0.06
SE	0.70	0.11	0.07	0.01
CD	NS	NS	NS	NS

Table 37 Effect of P levels on manganese content of litter (ppm)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	236.00	271.15	298.55	557.85
10 (T ₂)	248.00	277.95	281.90	555.60
20 (T ₃)	256.00	239.20	317.95	518.05
30 (T ₄)	281.50	276.13	297.15	497.30
40 (T ₅)	217.50	257.75	319.00	492.45
SE	18.14	14.80	25.04	36.33
CD	NS	NS	NS	NS

Table 38 Zinc content of litter (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	54.20	42.25	44.20	34.80
10 (T ₂)	58.40	46.65	45.00	36.75
20 (T ₃)	46.90	36.20	43.20	34.15
30 (T ₄)	57.60	41.60	41.60	34.35
40 (T ₅)	48.40	45.90	41.85	36.45
SE	3.32	2.63	3.64	1.68
CD	NS	NS	NS	NS

Table 39 Copper content of litter (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	34.95	255.80	121.90	87.35
10 (T ₂)	33.73	399.40	169.25	90.75
20 (T ₃)	45.55	225.07	156.45	81.35
30 (T ₄)	39.13	347.20	151.60	81.05
40 (T ₅)	33.93	424.00	180.10	105.65
SE	4.82	58.30	24.21	8.76
CD	NS	NS	NS	NS

4.9.3 Annual Nutrient Addition Through Litter

No significant difference was observed between treatments with respect to nitrogen addition through litter fall (Table 40). The nitrogen addition was lowest in the T₁ (114.17 kg ha⁻¹ year⁻¹) and highest in T₄ (134.70 kg ha⁻¹ year⁻¹).

Table 40 Nutrient addition through litter ($\text{kg ha}^{-1} \text{ year}^{-1}$) as influenced by levels of P

P_2O_5 levels, kg ha^{-1} year^{-1}	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu
0 (T_1)	114.17	4.84	53.59	127.83	12.64	15.59	2.91	0.29	0.77
10 (T_2)	124.29	5.10	61.26	133.68	17.16	15.51	3.29	0.32	0.89
20 (T_3)	124.72	5.13	54.29	145.86	15.43	17.29	3.22	0.28	0.85
30 (T_4)	134.70	5.90	56.39	135.11	13.93	17.98	3.14	0.31	0.74
40 (T_5)	124.31	5.86	60.12	128.36	15.09	18.05	3.12	0.31	0.77
SE	9.95	0.25	5.33	11.94	1.78	1.84	0.31	0.02	0.05
CD	NS	0.77	NS	NS	NS	NS	NS	NS	NS

Significant differences were observed between treatments in the total quantity of P recycled through litter. The phosphorus recycling in T_1 was the lowest ($4.84 \text{ kg ha}^{-1} \text{ year}^{-1}$) and significantly lower compared to T_4 and T_5 .

No significant difference was observed between treatments with respect to recycling of other nutrients through litter. Potassium addition was lowest in T_1 ($53.59 \text{ kg ha}^{-1} \text{ year}^{-1}$) and highest in T_2 ($61.26 \text{ kg ha}^{-1} \text{ year}^{-1}$). The recycling of calcium through litter fall ranged from $127.83 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_1 to $145.86 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_3 . The recycling of magnesium ranged from $12.64 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_1 to $17.16 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_2 . In the case of iron, the lowest addition was observed in T_2 ($15.5 \text{ kg ha}^{-1} \text{ year}^{-1}$) and the highest in T_5 ($18.05 \text{ kg ha}^{-1} \text{ year}^{-1}$). The addition of manganese ranged from $2.91 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_1 to $3.29 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_2 . Among the various nutrients, recycling of zinc was the lowest and ranged from $0.28 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_3 to $0.32 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_2 . Recycling of copper ranged from $0.74 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_4 to $0.89 \text{ kg ha}^{-1} \text{ year}^{-1}$ in T_2 .

4.10 INFLUENCE OF P LEVELS ON NUTRIENT REMOVAL THROUGH LATEX

4.10.1 Nutrient Content of Latex

Nutrient content of latex observed at quarterly intervals is presented in Table 41 to Table 49. The content of different nutrients in the latex was comparable in different treatments during all the samplings.

During the first sampling (April), nitrogen content varied from 0.50 per cent in T₄ to 0.55 per cent in T₂ (Table 41). The highest N content was observed in T₂ (0.62 %) during the second sampling in July. During the third sampling (October), highest N content was observed in T₁ (0.62%) and lowest in T₃ and T₄ (0.51 %). During the last sampling in January, there was not much variation in the N content in the latex in different treatments.

Table 41 Nitrogen content of latex (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.53	0.60	0.62	0.66
10 (T ₂)	0.55	0.62	0.52	0.65
20 (T ₃)	0.52	0.60	0.60	0.66
30 (T ₄)	0.50	0.58	0.51	0.65
40 (T ₅)	0.53	0.58	0.51	0.64
SE	0.04	0.04	0.04	0.04
CD	NS	NS	NS	NS

Phosphorus content of latex was also not influenced by the application of phosphorus (Table 42). During the first sampling, phosphorus content varied from 0.12 per cent in T₄ to 0.09 per cent in T₂. There was an increase in the latex P content during the next sampling and highest content was observed in T₃ and T₄ (0.35 %) and lowest in T₂ (0.31 %).

Table 42 Phosphorus content of latex (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.10	0.32	0.14	0.17
10 (T ₂)	0.09	0.31	0.13	0.16
20 (T ₃)	0.10	0.35	0.13	0.17
30 (T ₄)	0.12	0.35	0.14	0.21
40 (T ₅)	0.10	0.33	0.14	0.20
SE	0.01	0.02	0.01	0.02
CD	NS	NS	NS	NS

During the third sampling, there was a decline in the latex P content and the highest value (0.14%) was observed in T₁, T₄ and T₅. During the last sampling in January, highest P content was observed in T₄ (0.21 %) and lowest in T₂ (0.16%).

During the first sampling, potassium content varied from 0.52 per cent in T₁ to 0.45 per cent in T₅ (Table 43). There was a general increase in the latex K content during the next two samplings and a decrease during the last sampling. There was no significant difference in the latex K content in different treatments.

Table 43 Effect of P levels on potassium content of latex (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.52	0.60	0.61	0.51
10 (T ₂)	0.49	0.60	0.60	0.51
20 (T ₃)	0.48	0.59	0.59	0.56
30 (T ₄)	0.48	0.60	0.61	0.52
40 (T ₅)	0.45	0.62	0.60	0.50
SE	0.04	0.03	0.04	0.04
CD	NS	NS	NS	NS

There was not much variation in the latex magnesium content between treatments and also during different samplings and the values ranged from 0.12 to 0.17 per cent (Table 44).

Latex calcium content varied from 8.08 ppm in T₅ to 9.84 ppm in T₄ during first sampling. During the next sampling, there was a slight increase in the latex calcium content and it varied from 14.13 ppm in T₅ to 11.41 ppm in T₄. There was a decrease in the calcium content during the third sampling and the highest value was observed in T₂ (8.33 ppm). During the last sampling, the latex calcium content increased and it varied from 33.44 ppm in T₂ to 22.63 ppm in T₄.

Table 44 Effect of P levels on magnesium content of latex (%)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.13	0.12	0.16	0.15
10 (T ₂)	0.12	0.17	0.17	0.13
20 (T ₃)	0.13	0.16	0.12	0.13
30 (T ₄)	0.12	0.15	0.16	0.15
40 (T ₅)	0.13	0.13	0.14	0.14
SE	0.01	0.02	0.02	0.01
CD	NS	NS	NS	NS

Table 45 Calcium content of latex (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	8.88	11.76	6.32	32.00
10 (T ₂)	9.22	12.82	8.33	33.44
20 (T ₃)	8.43	12.88	6.90	25.44
30 (T ₄)	9.84	11.41	7.29	22.63
40 (T ₅)	8.08	14.13	6.27	24.23
SE	0.82	0.98	1.18	4.93
CD	NS	NS	NS	NS

Latex iron content varied from 6.74 ppm in T₅ to 9.62 ppm in T₁ during the first sampling, from 6.10 ppm in T₂ to 7.46 ppm in T₁ during the second sampling, from 7.22 ppm in T₃ to 12.85 ppm in T₅ during the third sampling and from 7.93 ppm T₅ to in 11.91 ppm in T₁ during the last sampling (Table 46).

Table 46 Iron content of latex (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	9.62	7.46	7.98	11.91
10 (T ₂)	7.82	6.10	9.42	10.44
20 (T ₃)	7.86	6.55	7.22	11.05
30 (T ₄)	8.12	6.43	11.01	10.51
40 (T ₅)	6.74	6.92	12.85	7.93
SE	1.35	0.48	2.73	1.30
CD	NS	NS	NS	NS

Among the different nutrients, manganese was present in the lowest quantity in the latex (Table 47). Manganese content varied from 0.59 ppm (T₅) to 0.79 ppm (T₄) during the first sampling, 0.49 ppm (T₄) to 0.75 ppm (T₁) during the second sampling, from 0.46 ppm (T₄) to 0.56 ppm (T₅) during the third sampling and from 0.59 ppm (T₄) to 0.68 ppm (T₁ and T₃) during the last sampling.

Table 47 Manganese content of latex (ppm) as influenced P levels

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.68	0.75	0.54	0.68
10 (T ₂)	0.62	0.71	0.50	0.63
20 (T ₃)	0.74	0.57	0.49	0.68
30 (T ₄)	0.79	0.49	0.46	0.59
40 (T ₅)	0.59	0.63	0.56	0.65
SE	0.05	0.12	0.07	0.06
CD	NS	NS	NS	NS

Zinc content in the latex varied from 3.26 ppm^f (T₁) to 3.83 ppm (T₅) during the first sampling, from 3.53 ppm (T₃) to 4.80 ppm (T₁) during the second sampling, 4.45 ppm (T₅) to 6.16 ppm (T₄) during the third sampling and from 4.47 ppm (T₃) to 4.74 ppm (T₁) during the last sampling (Table 48).

Table 48 Zinc content of latex (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	3.26	4.80	5.49	4.74
10 (T ₂)	3.48	4.22	5.15	4.71
20 (T ₃)	3.61	3.53	5.49	4.47
30 (T ₄)	3.77	4.45	6.16	4.52
40 (T ₅)	3.83	4.27	4.45	4.71
SE	0.21	0.47	0.41	0.39
CD	NS	NS	NS	NS

Latex copper content varied from 3.73 ppm (T₁) to 4.56 ppm (T₄) during the first sampling, 3.79 ppm (T₁) to 4.82 ppm (T₅) during the second sampling, from 4.76 ppm (T₃) to 7.11 ppm (T₁) during the third sampling and from 3.25 ppm (T₂) to 5.02 ppm (T₁) during the last sampling (Table 49).

Table 49 Influence of P levels on copper content of latex (ppm)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	3.73	3.79	7.11	5.02
10 (T ₂)	3.95	4.36	5.28	3.25
20 (T ₃)	4.15	4.81	4.76	3.98
30 (T ₄)	4.56	4.54	6.92	4.85
40 (T ₅)	3.84	4.82	5.73	3.83
SE	0.47	0.36	0.89	0.62
CD	NS	NS	NS	NS

Table 50 Influence of P levels on nutrient removal through latex

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	N		P		K		Mg		Ca		Fe		Mn		Zn		Cu	
	kg ha ⁻¹ year ⁻¹		kg ha ⁻¹ year ⁻¹		kg ha ⁻¹ year ⁻¹		kg ha ⁻¹ year ⁻¹		kg ha ⁻¹ year ⁻¹		g ha ⁻¹ year ⁻¹		g ha ⁻¹ year ⁻¹		g ha ⁻¹ year ⁻¹		g ha ⁻¹ year ⁻¹	
0 (T ₁)	13.90	4.30	13.12	3.29	28.24	20.42	1.54	10.79	11.25									
10 (T ₂)	14.97	4.40	13.80	3.78	29.74	20.63	1.55	10.95	10.58									
20 (T ₃)	14.13	4.56	13.30	3.22	27.80	18.72	1.44	10.47	10.11									
30 (T ₄)	15.24	4.81	13.91	3.62	29.11	24.13	1.56	11.38	13.20									
40 (T ₅)	12.91	4.26	12.22	2.99	23.61	19.92	1.33	9.67	12.21									
SE	0.60	0.33	0.89	0.24	1.85	1.22	0.08	0.76	0.85									
CD	NS	NS	NS	NS	NS	NS	NS	NS	NS									

4.10.2 Nutrient Removal through Latex

The nutrient removal through latex is presented in Table 50. There was no significant difference between treatments in the quantity of different nutrients removed through latex. Among the nutrients, nitrogen was removed in the largest quantity followed by potassium. Removal of nitrogen was highest from T₄ (15.24 kg ha⁻¹year⁻¹) and lowest from T₅ (12.91 kg ha⁻¹year⁻¹). Removal of phosphorus was also highest from T₄ (4.81 kg ha⁻¹year⁻¹) and lowest from T₅ (4.26 kg ha⁻¹ year⁻¹). Removal of P from T₁ which did not receive any P fertilizer was 4.30 kg ha⁻¹year⁻¹. Removal of potassium varied from the 12.22 to 13.91 kg ha⁻¹year⁻¹. The removal of magnesium varied from 2.99 kg ha⁻¹year⁻¹ in T₅ to 3.78 kg ha⁻¹ year⁻¹ in T₂. The calcium removal varied from 23.61 g ha⁻¹ year⁻¹ in T₅ to 29.74 g ha⁻¹ year⁻¹ in T₂. Removal of iron was also lowest from T₅ (19.92 g ha⁻¹ year⁻¹). The removal of manganese ranged from 1.33 g ha⁻¹ year⁻¹ in T₅ to 1.56 g ha⁻¹ year⁻¹ in T₄. Removal of zinc also followed the same trend, highest quantity from T₄ and lowest from T₅. The copper removal ranged from 10.11 g ha⁻¹ year⁻¹ in T₃ to 13.20 g ha⁻¹ year⁻¹ in T₄.

4.11 Mg/P RATIO IN LATEX AS INFLUENCED BY LEVELS OF P

Mg/P ratio in latex is presented in Table 51. The data indicate that, this parameter is not affected by the levels of phosphorus applied.

Table 51 Mg/P ratio of latex as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	1.28	0.63	1.16	0.90
10 (T ₂)	1.39	0.56	1.08	0.83
20 (T ₃)	1.31	0.54	0.95	0.84
30 (T ₄)	1.08	0.52	1.14	0.75
40 (T ₅)	1.25	0.55	1.04	0.79
SE	0.13	0.06	0.10	0.11
CD	NS	NS	NS	NS

During the first sampling, the Mg/P ratio in the latex was highest in T₂ (1.39) and lowest in T₄ (1.08). During the next sampling, there was a decrease in the ratio and highest value was observed in T₁ (0.63) and lowest in T₄ (0.52). During the third sampling, the Mg/P ratio was higher in all the treatments. During the last sampling, there was again a decrease in the ratio; highest value was observed in T₁ (0.90) and lowest in T₄ (0.75).

4.12 INFLUENCE OF PHOSPHORUS APPLICATION ON SOIL NUTRIENT STATUS

Soil nutrient status in different treatments is presented in Table 52 to Table 60.

During the first sampling, all the phosphorus applied treatments had a significantly higher soil P status compared to the control treatment (Table 52). During the second sampling also, lowest P status was observed in T₁ which did not receive any P fertilizer (21.51 kg ha⁻¹) and highest in T₅ which received the highest level of P (145.60 kg ha⁻¹). Soil P status of treatments which received phosphorus at 10 and 20 kg ha⁻¹ (T₂ and T₃) were comparable with the control treatment and significantly lower than the treatments which received phosphorus at higher levels *i.e.*, 30 and 40 kg ha⁻¹. During the third sampling also, same trend was observed. Lowest soil P status was observed in T₁ (14.11 kg ha⁻¹) and highest in T₅ (69.55 kg ha⁻¹). Soil P status in T₁, T₂ and T₃ were comparable, and T₄ was comparable with T₅. During the last sampling also, similar trend was observed.

Table 52 Influence of P levels on soil available phosphorus status (kg ha⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	14.00	21.51	14.11	14.56
10 (T ₂)	27.46	44.80	24.19	28.63
20 (T ₃)	42.56	46.48	28.45	42.11
30 (T ₄)	43.68	120.98	52.75	110.95
40 (T ₅)	59.92	145.60	69.55	133.79
SE	4.32	12.80	5.59	9.63
CD	13.32	39.46	17.21	29.66

The organic carbon content was in the medium range and comparable in different treatments (Table 53). The organic carbon content varied from 1.23 per cent in T₁ to 1.33 per cent in T₄ during the first sampling, from 1.36 per cent in T₅ to 1.55 per cent in T₁ during the second sampling, from 1.07 per cent in T₄ to 1.21 per cent in T₂ during the third sampling and from 1.04 per cent in T₅ to 1.30 per cent in T₁ during the last sampling.

Table 53 Soil organic carbon status (%) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0(T ₁)	1.23	1.55	1.16	1.30
10(T ₂)	1.30	1.45	1.21	1.14
20(T ₃)	1.30	1.44	1.18	1.12
30(T ₄)	1.33	1.54	1.07	1.23
40(T ₅)	1.28	1.36	1.08	1.04
SE	0.08	0.10	0.10	0.09
CD	NS	NS	NS	NS

The potassium status of the soil was also comparable in different treatments during all the samplings (Table 54). The potassium content varied from 149.81 kg ha⁻¹ in T₁ to 193.20 kg ha⁻¹ in T₃ during the first sampling. The second sampling was done after the first fertilizer application and there was an increase in the potassium content in all the treatments and it varied from 207.20 kg ha⁻¹ in T₂ to 239.40 kg ha⁻¹ in T₁.

Table 54 Soil available potassium status (kg ha⁻¹) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0(T ₁)	149.81	239.40	173.60	178.92
10(T ₂)	169.40	207.20	130.20	189.00
20(T ₃)	193.20	210.00	217.00	172.20
30(T ₄)	177.82	217.00	145.60	201.60
40(T ₅)	184.80	228.20	154.93	199.73
SE	0.04	27.75	25.34	22.87
CD	NS	NS	NS	NS

During the third sampling, soil K content varied from 130.20 kg ha⁻¹ in T₂ to 217.00 kg ha⁻¹ in T₃ and during the last sampling from 172.2 kg ha⁻¹ in T₃ to 201.60 kg ha⁻¹ in T₄.

There was no significant difference between treatments in the soil calcium content during the first sampling (Table 55). It was lowest in T₁ (0.22 cmol kg⁻¹) and highest in T₅ (0.42 cmol kg⁻¹). During the second sampling, significant differences were observed between treatments. Soil calcium content in T₁ was significantly lower than those in T₄ and T₅ and comparable with the other treatments which received lower levels of phosphorus. During the third and fourth sampling also, the same trend was observed.

Table 55 Soil exchangeable calcium content (cmol kg⁻¹) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0(T ₁)	0.22	0.35	0.20	0.29
10(T ₂)	0.28	0.39	0.22	0.30
20(T ₃)	0.38	0.40	0.27	0.37
30(T ₄)	0.33	0.61	0.29	0.57
40(T ₅)	0.42	0.58	0.27	0.47
SE	0.05	0.05	0.02	0.04
CD	NS	0.17	0.06	0.14

During the first sampling, there was no significant difference between treatments in the soil magnesium content (Table 56). However, magnesium content was higher in P applied treatments compared to the treatment which did not receive P fertilizer. Lowest magnesium content was observed in T₁ (0.12 cmol kg⁻¹) and highest in T₅ (0.19 cmol kg⁻¹). During the second and fourth sampling (after fertilizer application), soil Mg status was comparable in T₁, T₂ and T₃ and was significantly lower than those of T₄ and T₅. During the third sampling also, the same trend was observed. However, there was no significant difference between treatments.

Table 56 Effect of P levels on soil exchangeable magnesium content (cmol kg⁻¹)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0(T ₁)	0.12	0.14	0.08	0.12
10(T ₂)	0.16	0.14	0.08	0.12
20(T ₃)	0.16	0.14	0.09	0.15
30(T ₄)	0.16	0.23	0.11	0.26
40(T ₅)	0.19	0.22	0.11	0.20
SE	0.02	0.01	0.01	0.02
CD	NS	0.03	NS	0.05

During the first sampling, soil iron status in all the treatments were comparable (Table 57). During the second sampling also, there was no significant difference between treatments. During the third sampling, soil iron status in T₁ was the highest (23.62 ppm) and it was significantly higher compared to the P applied treatments. The lowest iron content was observed in T₅ which received the highest level of P (13.75 ppm) and it was significantly lower than that of all the other treatments. During the last sampling also, the same trend was observed. However, there was no significant difference between treatments.

Soil manganese status during the first sampling was highest in the control treatment (9.70 ppm) and it was comparable with T₂ and T₃ and significantly greater than T₄ and T₅ (Table 58). Soil Mn status was lowest in T₅ (5.25 ppm) and it was comparable with T₃ and T₄. During the second sampling also, highest manganese status was observed in T₁ (4.04 ppm) and lowest in T₅ (2.26 ppm). There was no significant difference between treatments. During the third sampling also, the same trend was observed. Soil Mn status was highest in T₁ (5.61 ppm), and was comparable with T₂ and T₃. Soil Mn status was lowest in T₅ (2.97 ppm) and it was comparable with T₄. During the last sampling (after second fertilizer application), no definite trend was observed in the soil Mn status and there was no significant difference between treatments.

Table 57 Soil available iron content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	4.70	4.41	23.62	25.13
10 (T ₂)	4.79	4.44	18.37	20.74
20 (T ₃)	4.71	4.46	17.37	21.79
30 (T ₄)	4.63	4.46	17.16	21.12
40 (T ₅)	4.81	4.57	13.75	21.10
SE	0.07	0.05	1.10	2.43
CD	.NS	NS	3.40	NS

Table 58 Effect of P levels on soil available manganese content (ppm)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0(T ₁)	9.70	4.04	5.61	9.87
10(T ₂)	8.38	3.93	5.56	11.15
20(T ₃)	8.04	2.72	4.86	9.47
30(T ₄)	6.10	2.70	3.30	8.54
40(T ₅)	5.25	2.26	2.97	8.37
SE	0.98	0.38	0.45	1.05
CD	3.01	1.18	1.39	NS

There was no significant difference between treatments in the soil zinc content during any of the samplings (Table 59).

In the case of soil copper content, no significant difference was observed between treatments during any of the samplings (Table 60). Highest content was observed in T₁ during the first and second sampling (20.18 ppm and 26.23 ppm respectively) and lowest in T₅ (8.91 ppm and 15.42 ppm respectively). During the third sampling, no definite trend was observed. During the last sampling, highest copper status was observed in T₁ and lowest in T₄.

Table 59 Soil available zinc content (ppm) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	0.70	0.89	0.44	0.59
10 (T ₂)	0.74	0.66	0.37	0.55
20 (T ₃)	0.65	0.61	0.43	0.67
30 (T ₄)	0.70	0.65	0.34	0.65
40 (T ₅)	0.70	0.58	0.34	0.59
SE	0.04	0.07	0.06	0.04
CD	NS	NS	NS	NS

The Fe-P content varied from 67.00 ppm in T₄ (P @ 30 kg P₂O₅ ha⁻¹ year⁻¹) to 103.25 ppm in T₁ which did not receive any P fertilizer. The Ca-P content varied from 32.25 ppm in T₃ to 62.00 ppm in T₄. Among different fractions, Al-P was present in the lowest quantity and it varied from 7.02 ppm in T₅ to 11.75 ppm in T₃. The reductant P content varied from 12.91 ppm in T₁ to 25.27 ppm in T₄. Occluded P content was lowest in T₂ (P@ 10 kg P₂O₅ ha⁻¹ year⁻¹) and highest in T₄. Organic P content was lowest in T₁ (172.24 ppm) and highest in T₄ (269.67 ppm). The total P content showed significant difference between treatments. Highest total P content (437.50 ppm) was observed in T₄ and it was comparable with T₃ and T₅. Lowest content was observed in T₁ (336.85 ppm) and it was comparable with T₂ and T₃.

4.14 INFLUENCE OF P LEVELS ON SOIL ENZYMES

4.14.1 Soil Dehydrogenase Activity (DHA)

Soil dehydrogenase activity measured before and after fertilizer application is presented in Table 62. During the first sampling highest dehydrogenase activity was observed in the treatment which received phosphorus at 10 kg ha⁻¹ (168.75 µg TPF g⁻¹ 24 h⁻¹). There was no significant difference between treatments. During the next sampling, there was a general increase in the dehydrogenase activity, but there was no significant difference between treatments. During the third sampling, there was a further increase in the dehydrogenase activity. All the treatments were comparable. Dehydrogenase activity was comparable between treatments during the last sampling also. A decline in dehydrogenase activity was observed in three treatments, and the activity was maintained more or less at the same level in the other two treatments during this period.

Table 62 Soil dehydrogenase activity ($\mu\text{g TPF g}^{-1}$ soil 24 h^{-1}) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	120.83	191.67	218.75	183.33
10 (T ₂)	168.75	183.33	197.92	161.11
20 (T ₃)	150.00	188.89	193.75	181.95
30 (T ₄)	158.33	197.22	208.36	210.42
40 (T ₅)	120.83	183.33	229.17	208.33
SE	15.52	25.30	18.18	23.15
CD	NS	NS	NS	NS

4.14.2 Soil Acid Phosphatase Activity

Soil phosphatase activity measured before and after fertilizer application was higher in the treatment which did not receive any P fertilizer (T₁) during most of the samplings (Table 63).

Table 63 Soil phosphatase activity ($\mu\text{g nitrophenol g}^{-1}$ soil h^{-1}) as influenced by levels of P

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	58.00	73.00	71.67	84.25
10 (T ₂)	59.25	71.50	56.25	63.25
20 (T ₃)	56.00	69.50	59.00	75.50
30 (T ₄)	54.50	64.00	58.50	55.75
40 (T ₅)	57.00	65.50	57.80	66.25
SE	2.52	2.93	2.74	8.18
CD	NS	NS	8.45	NS

During first sampling, phosphatase activity was highest in the treatment which received phosphorus at 10 kg ha^{-1} ($59.25 \mu\text{g nitrophenol g}^{-1}\text{ soil h}^{-1}$) followed by T_1 which did not receive any P fertilizer. No significant difference was observed between treatments. During the next sampling, the highest phosphatase activity was observed in T_1 ($73.00 \mu\text{g nitrophenol g}^{-1}\text{ soil h}^{-1}$), followed by T_2 . No significant difference was observed between treatments. During the third sampling phosphatase activity in T_1 ($71.67 \mu\text{g nitrophenol g}^{-1}\text{ soil h}^{-1}$) was significantly higher compared to P applied treatments. During the fourth sampling, an increase in the phosphatase activity was observed in all the treatments except that which received phosphorus at 30 kg ha^{-1} . Highest activity was noticed in T_1 . However, there was no significant difference between treatments.

4.15 INFLUENCE OF P LEVELS ON NON-RHIZOSPHERE AND RHIZOSPHERE PH

Soil pH measured at bimonthly intervals did not indicate any significant difference between treatments (Table 64). Though not significant non-rhizosphere soil pH was higher in the P applied treatments in most of the cases.

Table 64 Influence of P levels on non-rhizosphere soil pH

P_2O_5 levels, $\text{kg ha}^{-1}\text{ year}^{-1}$	I	II	III	IV
0 (T_1)	4.64	4.41	4.80	4.84
10 (T_2)	4.79	4.44	4.93	4.88
20 (T_3)	4.71	4.46	4.82	4.79
30 (T_4)	4.71	4.46	4.83	4.80
40 (T_5)	4.81	4.57	4.86	4.78
SE	0.04	0.05	0.03	0.04
CD	NS	NS	NS	NS

Rhizosphere pH measured at bimonthly intervals is presented in Table 65. In general, rhizosphere pH was lower in T₁ which did not receive any P fertilizer. During the first sampling, rhizosphere pH was lowest in T₁ (4.53) and it was significantly lower compared to T₂ (P@ 10 kg P₂O₅ ha⁻¹ year⁻¹) and T₅ (P@ 40 kg P₂O₅ ha⁻¹ year⁻¹). During the next sampling, T₂ recorded the lowest rhizosphere pH (4.72) and T₅ recorded the highest (4.85), but there was no significant difference between the treatments. During the third sampling also, lowest rhizosphere pH was measured in T₁ which did not receive any P fertilizer (4.80) and highest in T₅ which received P at the highest level (5.01). Rhizosphere pH in T₁ was significantly lower compared to all other treatments. No significant difference was observed between the different levels of P application. During the next sampling also, same trend was noticed. Lowest rhizosphere pH was measured in T₁ (4.70) and highest in T₅. However there was no significant difference between treatments.

Table 65 Influence of P levels on rhizosphere pH

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	4.53	4.73	4.80	4.70
10 (T ₂)	4.72	4.72	4.98	4.80
20 (T ₃)	4.58	4.69	4.94	4.80
30 (T ₄)	4.66	4.73	4.97	4.80
40 (T ₅)	4.68	4.85	5.01	4.96
SE	0.04	0.05	0.03	0.06
CD	0.14	NS	0.10	NS

4.16 INFLUENCE OF P LEVELS ON RHIZOSPHERE MICROORGANISMS

4.16.1 Root Colonization by Arbuscular Mycorrhizal Fungi (AMF)

AMF infection of roots before and after fertilizer application was not influenced by phosphorus application during any of the samplings (Table 66). Trees in all the treatments had a high percentage of infection during all the samplings. During the first sampling, AMF infection in T₁ which did not receive any P fertilizer was 90.63 per cent. During the second sampling, AMF infection in T₁ was 86.75 per cent, and all other treatments except T₃ had a higher percentage of infection. During third sampling, the infection percentage varied from 86.88 per cent in T₁ to 88.75 per cent in T₃, T₄ and T₅ and during the last sampling, from 79.38 per cent in T₃ to 88.13 per cent in T₄ and T₅.

Table 66 Influence of P levels on AMF infection of roots (%).

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	I	II	III	IV
0 (T ₁)	90.63	86.75	86.88	85.00
10 (T ₂)	87.50	88.38	87.50	87.75
20 (T ₃)	90.00	79.25	88.75	79.38
30 (T ₄)	86.88	88.75	88.75	88.13
40 (T ₅)	86.88	88.75	88.75	88.13
SE	2.32	3.42	3.08	3.08
CD	NS	NS	NS	NS

4.16.2 Total Microbial Count and Phosphorus Solubilizers in the Rhizosphere

Total microbial count before and after fertilizer application are presented in Table 67a to Table 67d.

The population of P solubilizing bacteria (PSB) and the percentage of PSB to total bacteria was significantly influenced by

Table 67a Influence of P levels on soil microbial population during first sampling

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	PSB (10 ⁷)	% of PSB to total bacteria	Phosphorus solubilizing fungi (10 ⁵)	Total bacteria (10 ⁷)	Total fungi (10 ⁵)	Actinom- ycetes (10 ⁶)
0 (T ₁)	4.23 (2.04)	7.54 (2.70)	10.60	67.05	36.68	2.73
10 (T ₂)	5.89 (2.39)	11.12 (3.26)	9.67	57.02	54.33	2.20
20 (T ₃)	6.18(2.44)	10.48 (3.18)	8.75	58.61	51.25	3.33
30 (T ₄)	3.93(1.96)	6.73 (2.53)	10.53	63.47	44.75	3.35
40 (T ₅)	6.19 (2.45)	8.97(2.93)	8.96	72.33	56.50	1.98
SE	0.25	0.36	1.87	5.37	7.67	0.63
CD	NS	NS	NS	NS	NS	NS

Values in parentheses are transformed values (square root)

Table 67b Influence of P levels on soil microbial population during second sampling

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	PSB (10 ⁷)	% of PSB to total bacteria	Phosphorus solubilizing fungi (10 ⁵)	Total bacteria (10 ⁷)	Total fungi (10 ⁵)	Actinomycetes (10 ⁶)
0 (T ₁)	27.25 (5.19)	38.80 (6.22)	5.47	64.76	14.90	5.83
10 (T ₂)	12.21(3.39)	21.99 (4.68)	7.71	54.86	24.50	5.35
20 (T ₃)	18.56 (4.13)	29.26 (5.35)	7.95	62.92	19.78	4.60
30 (T ₄)	13.34 (3.46)	18.72 (4.14)	8.93	68.10	25.31	6.00
40 (T ₅)	5.84 (2.05)	8.16 (2.55)	5.30	47.95	22.08	5.50
SE	0.59	0.57	1.59	6.53	3.86	1.21
CD	1.81	1.75	NS	NS	NS	NS

Values in parentheses are transformed values (square root)

Table 67c Influence of P levels on soil microbial population during third sampling period

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	PSB (10 ⁷)	% of PSB to total bacteria	Phosphorus solubilizing fungi (10 ⁵)	Total bacteria (10 ⁷)	Total fungi (10 ⁵)	Actinomycetes (10 ⁶)
0 (T ₁)	15.02 (3.77)	25.28 (4.89)	4.55	61.67	60.75	2.08
10(T ₂)	15.50(3.63)	25.92 (4.71)	3.48	61.07	49.39	2.60
20(T ₃)	18.89 (4.32)	24.07 (4.88)	5.85	68.02	60.64	2.85
30(T ₄)	11.88 (3.25)	26.19 (4.93)	3.29	42.83	70.07	1.57
40(T ₅)	8.71 (2.49)	12.58 (3.07)	4.25	62.17	55.85	2.55
SE	0.74	0.84	0.74	6.76	9.95	0.36
CD	NS	NS	NS	NS	NS	NS

Values in parentheses are transformed values (square root)

Table 67d Influence of P levels on soil microbial population during fourth sampling

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	PSB (10 ⁷)	% of PSB to total bacteria	Phosphorus solubilizing fungi (10 ⁵)	Total bacteria (10 ⁷)	Total fungi (10 ⁵)	Actinomycetes (10 ⁶)
0 (T ₁)	11.67 (3.25)	22.13 (4.70)	15.92	53.51	124.82	8.72
10 (T ₂)	2.96 (1.71)	12.62 (3.51)	12.78	51.83	90.78	9.28
20 (T ₃)	7.04 (2.53)	13.57 (3.68)	7.49	61.19	127.43	8.27
30 (T ₄)	6.85 (2.44)	11.12 (3.33)	10.21	60.34	128.10	7.51
40 (T ₅)	5.66 (2.35)	12.36 (3.50)	12.92	45.11	133.73	9.74
SE	0.40	0.18	2.45	6.03	12.91	2.40
CD	NS	0.57	NS	NS	NS	NS

Values in parentheses are transformed values (square root)

application of phosphorus. During first sampling, there was no significant difference between treatments. During second sampling (after fertilizer application), T₁ which did not receive any P fertilizer had the highest population of PSB (27.25×10^7) and it was significantly higher than that of T₅ which received P at the highest level (@40 kg P₂O₅ ha⁻¹ year⁻¹). During third and fourth sampling, there was no significant difference between treatments. The percentage of PSB to total bacteria also showed a similar trend. But during the fourth sampling also, T₁ had a significantly higher percentage of PSB to total bacteria. All the treatments which received P were comparable.

The population of P solubilizing fungi was not influenced by application of phosphorus. During the first sampling, population of P solubilizing fungi ranged from 10.6×10^5 in T₁ to 8.75×10^5 in T₃. During the second sampling, the population ranged from 5.3×10^5 in T₅ to 8.93×10^5 in T₄. During the next sampling, there was a decline in the population of phosphorus solubilizing fungi and it ranged from 3.29×10^5 in T₄ to 5.85×10^5 in T₃. During the last sampling, there was an increase in the population and it ranged from 7.49×10^5 in T₃ to 15.92×10^5 in T₁.

The population of total bacteria, actinomycetes and total fungi were not influenced by application of phosphorus.

Total bacterial count ranged from 57.02×10^7 in T₂ to 72.33×10^7 in T₅ during first sampling, from 47.95×10^7 in T₅ to 68.10×10^7 in T₄ during the second sampling and from 42.83×10^7 in T₄ to 68.02×10^7 in T₃ during third sampling. During the last sampling, there was a decline in the population in most cases.

The population of total fungi ranged from 36.68×10^5 in T₁ to 56.50×10^5 in T₅ during the first sampling. During the second sampling, there was a general decline in the population of total fungi in the rhizosphere and it ranged from 14.90×10^5 in T₁ to 25.31×10^5 in T₄. During the next sampling, there was an increase in the population of total fungi and it ranged from 49.39×10^5 in T₂ to 70.07×10^5 in T₄. During

the last sampling, there was a further increase in the population and it ranged from 90.78×10^5 in T_2 to 133.73×10^5 in T_5 .

The population of actinomycetes showed an increase after fertilizer application. During first sampling (before first fertilizer application), the population ranged from 1.98×10^6 in T_5 to 3.35×10^6 in T_4 . During the second sampling (after first fertilizer application), there was a general increase in the population of actinomycetes in the rhizosphere. During the next sampling (before second fertilizer application), there was a decline in the population of actinomycetes and it ranged from 1.57×10^6 in T_4 to 2.85×10^6 in T_3 . During the last sampling (after second fertilizer application), there was an general increase in the population and highest population was observed in T_5 (9.74×10^5) and lowest in T_4 .

4.17 INCIDENCE OF TAPPING PANEL DRYNESS (TPD)

Tapping panel dryness is a major problem encountered in rubber plantations and is characterized by a gradual or sudden drying up of the latex vessels resulting in abnormally low yield or complete stoppage of latex production. In the present study, Incidence of tapping panel dryness was not influenced by application of phosphorus (Table 68).

Table 68 Incidence of tapping panel dryness (%) as influenced by levels of P

P_2O_5 levels, $kg\ ha^{-1}\ year^{-1}$	TPD incidence
0 (T_1)	10.87 (3.20)
10 (T_2)	14.88 (3.64)
20 (T_3)	23.82 (4.94)
30 (T_4)	16.73 (3.77)
40 (T_5)	16.87 (3.87)
SE	(0.89)
CD	NS

Values in parentheses are transformed values (square root)

4.18 INFLUENCE OF P LEVELS ON CORRELATION BETWEEN SOIL AND PLANT CHARACTERS

The correlation between different variables are presented in Table 69 and Table 70.

Table 69 Correlation coefficients between soil P and other parameters

Parameter	Correlation coefficient
Leaf P	0.115
Root acid phosphatase secretion	-0.366*
Soil phosphatase	-0.167
Root PEP carboxylase activity	-0.519 *
Root Malate dehydrogenase activity	-0.513 *

*Significant at 5 per cent level

Table 70 Correlation coefficients between leaf P and other parameters

Parameter	Correlation coefficient
Root acid phosphatase secretion	0.049
Soil phosphatase	0.095
PEP carboxylase activity	0.089
Malate dehydrogenase activity	0.066

No significant correlation was observed between soil available P status and leaf P content (Table 69). Significant negative correlation was observed between soil P status and root acid phosphatase secretion ($r = -0.366$). Root PEP carboxylase activity and malate dehydrogenase activity were negatively correlated with soil P (-0.519 and -0.513). No significant correlation was observed between soil P and soil acid

phosphatase activity. Leaf P was not significantly correlated with any of the parameter studied *i.e.*, root acid phosphatase secretion, soil acid phosphatase activity, root PEP carboxylase activity and malate dehydrogenase activity (Table 70).

4.19 P BUDGETING

P dynamics in the rubber plantation is presented in Table 71 and annual P budget for 2003-04 is presented in Table 72 and Fig.12. Available P status of the soil at planting of trees was not available. The total P accrued in dry matter per hectare during the entire period (1984-2003) was computed based on the P accumulation per tree and number of trees per hectare. Cumulative addition of P through litter and removal through latex could not be computed with the available data.

Table 71 P dynamics in the plantation (kg ha^{-1}) as influenced by levels of P

P_2O_5 levels, $\text{kg ha}^{-1} \text{ year}^{-1}$	Soil available P (1997)	Total soil P (1997)	Applied P (1984-2003)	P accumulation in dry matter (1984-2003)	Available P in soil (2003)	Total P in soil (2003)
0 (T_1)	18.80	740.00	193.50	302.86	16.05	754.54
10 (T_2)	18.80	740.00	219.30	318.95	31.27	762.94
20 (T_3)	18.80	740.00	245.10	283.49	39.90	901.60
30 (T_4)	18.80	740.00	270.90	323.85	82.09	1039.90
40 (T_5)	18.80	740.00	296.70	317.97	102.22	980.00

Total P in the soil ranged from $754.54 \text{ kg ha}^{-1}$ in the treatment which did not receive any P fertilizer (T_1) to $1039.90 \text{ kg ha}^{-1}$ in the treatment which received P at 30 kg ha^{-1} (T_4). Available P content in the soil was lowest in T_1 (16.05 kg ha^{-1}) and highest in T_5 which received P at 40 kg ha^{-1} ($102.22 \text{ kg ha}^{-1}$). Available P status at the commencement of

the experiment (1997) was 18.80 kg ha^{-1} and the decline was less than 15 per cent in T_1 after six years. Quantity of P added through fertilizer ranged from $193.50 \text{ kg ha}^{-1}$ in T_1 to $296.70 \text{ kg ha}^{-1}$ in T_5 . Cumulative trunk storage of P was not influenced by the levels of P applied and was highest in T_4 ($323.85 \text{ kg ha}^{-1}$) and lowest in T_3 ($283.49 \text{ kg ha}^{-1}$).

Removal of P through latex was not influenced by P application (Table 72). Highest removal was observed from T_4 (4.81 kg ha^{-1}) and lowest from T_5 (4.26 kg ha^{-1}). Addition of P through litter was lowest in T_1 (4.84 kg ha^{-1}) and highest in T_4 (5.90). There was a net addition of $0.54 \text{ kg P ha}^{-1} \text{ year}^{-1}$ even in T_1 which did not receive any P fertilizer.

Table 72 P budget in the plantation as influenced by levels of P (2003-04)

P ₂ O ₅ levels, kg ha ⁻¹ year ⁻¹	Quantity of P (kg) added through			Latex removal (kg) (2003-04)	Net addition (kg)
	Fertilizers (2003-04)	Litter (2003-04)	Total		
0 (T ₁)	0	4.84	4.84	4.30	0.54
10 (T ₂)	4.37	5.10	9.47	4.40	5.07
20 (T ₃)	8.74	5.13	13.84	4.56	9.30
30 (T ₄)	13.11	5.90	19.01	4.81	14.19
40 (T ₅)	17.48	5.86	23.34	4.26	19.07

Discussion

5. DISCUSSION

The investigation entitled 'Phosphorus nutrioperiodism in rubber' was carried out to explore the possibility of reducing the dose of phosphorus for mature rubber. The experiment was initiated during 1997 and detailed observations were recorded during 2002-2004. The response of mature rubber to phosphorus, P dynamics in soil and plant, role of microorganisms in P nutrition and the processes by which trees improve their P acquisition are discussed below.

5.1 EFFECT OF PHOSPHORUS APPLICATION ON GROWTH CHARACTERS

5.1.1 Girth

Girth and girth increment of rubber were not significantly influenced by the application of phosphorus (Table 2). This result is in conformity with the findings of Ananth (1966) and Punnoose *et al.* (1975, 1994). It was reported that nitrogen and phosphorus improved the growth of rubber up to the fifth year of planting and thereafter, there was no response when ground covers were established with rubber. Nutrient release from ground cover and litter sets in during the late immature phase and the plants might be able to meet part of their nutrient requirement from this nutrient release. Rate of growth decreases during late immature and mature phase and the plants might be able to maintain growth with less nutrients. Moreover, the roots explore a large volume of soil giving the trees access to phosphorus in this large soil volume (Table 4). Being an immobile nutrient, the exploration of large volume of soil is particularly important for the uptake of phosphorus. The leaf P content also indicates that plants are able to maintain the P status without external application of P (Table 11) which might be the reason for the lack of response of trees in terms of growth to application of phosphorus.

5.1.2 Length of Tapping Panel

Length of tapping panel was not influenced by levels of phosphorus (Table 3). Length of tapping panel is directly dependent on the girth of trees and the lack of influence of P application on length of tapping panel might be due to its lack of influence on girth.

5.1.3 Bark Thickness

Bark thickness was not influenced by application of phosphorus (Table 3). This also indicates the ability of trees to maintain their growth and growth attributes without external application of phosphorus.

5.2 EFFECT OF PHOSPHORUS LEVELS ON YIELD AND YIELD ATTRIBUTES

5.2.1 Yield

Yield of rubber was not significantly influenced by levels of phosphorus during the period under study (Table 9). No definite trend was also observed between the levels of phosphorus and yield. During the first year after imposing the treatments, the highest yield was recorded in the treatment which received P at the highest level (@ 40 kg P₂O₅ ha⁻¹year⁻¹) and lowest yield in the treatment which received P at the lowest level (@ 10 kg P₂O₅ ha⁻¹year⁻¹). During the second year of treatment imposition, yield in the control treatment (T₁) was higher than that in the treatments which received P at 10, 20 and 40 kg P₂O₅ ha⁻¹year⁻¹ (T₂, T₃ and T₅). There was no decline in yield in the control treatment compared to the previous year. During 2000-01 (third year of treatment imposition) also, there was no decline in yield in the control treatment or in the treatments which received lower levels of P. In fact the yield in the treatment which received P at the highest level (@ 40 kg P₂O₅ ha⁻¹year⁻¹) was lower than that in the treatment which received P at the lowest level (@ 10 kg ha⁻¹year⁻¹). During next year, there was an increase in yield in all the treatments. Yield in

the control treatment was higher than that in T₅ (@ 40 kg P₂O₅ ha⁻¹ year⁻¹) and T₂ (@ 10 kg P₂O₅ ha⁻¹ year⁻¹). During 2002-03, yield was lowest in T₁ which did not receive any P fertilizer. However yield in T₅, which received the highest level of P (@ 40 kg P₂O₅ ha⁻¹ year⁻¹) was lower compared to T₂ and T₃. During 2003-04, highest yield was recorded in T₂ (@ 10 kg P₂O₅ ha⁻¹ year⁻¹) and lowest in T₅.

The data indicated that in general, the levels of phosphorus application during the period under study did not significantly influence yield. This result is in conformity with the observations of Pushparajah (1969) that when adequate phosphatic fertilizers are applied during immature phase, application of P did not increase yield during the early years of maturity. Lim (1977) also suggested restricting P application during mature phase. Taking into account the residual effect of fertilizer applied during immaturity and likely levelling off of nutrient immobilization within the trees with age, Watson (1989) suggested that fertilizer application could be suspended for four years after commencement of tapping and thereafter only sufficient N should be applied to replace that lost through latex.

Pushparajah and Tan (1972) observed that trees will respond to external application of phosphorus only if the leaf nutrient content is less than 0.21 per cent. In the present experiment, the leaf nutrient content even in the treatment which did not receive P fertilizers was always maintained above this level (Table 11). Once the plants reach the mature phase, the rate of growth decreases and a large quantity of nutrients will be immobilized within the trees (Watson, 1989). The nutrient requirement for further growth decreases. The only removal of nutrients from the tree is through latex. At the same time rubber is a deciduous tree and a large quantity of nutrients are recycled back through litter fall. After reviewing the fertilizer experiments conducted in rubber, Sivanadyan *et al.* (1995) concluded that fertilizer application does not necessarily increase yield in

mature rubber plantations which was well managed in the immature phase. Application of P during immature period results in sufficient build up of P in the top soil of mature stand and this can sustain the P requirement for some years of maturity against the P outflow through latex. Lack of response to phosphorus indicates that trees are able to meet their P requirement even when the soil available P status is low without external application of P. The leaf P status which was in the medium range also indicates that the plants are able to acquire sufficient P even in the control treatment (Table 11).

5.2.2 Dry Rubber Content of Latex (DRC)

The DRC values presented in Table 10 indicated that the different treatments did not affect the DRC of latex significantly. Definite trend was also not observed between the DRC values and phosphorus levels indicating that this yield component was not influenced by the phosphorus levels.

5.3 EFFECT OF APPLIED PHOSPHORUS ON NUTRIENT CONTENT IN DIFFERENT PARTS OF THE TREE

5.3.1 Leaf Nutrient Content

The leaf P content during different samplings is presented in Table 11 and Fig. 2. The different levels of phosphorus (0, 10, 20, 30 and 40 kg P₂O₅ ha⁻¹ year⁻¹) did not influence the leaf P status. In fact, the leaf P content in the control treatment (T₁) was higher than most of the P applied treatments during the first and third samplings. In many earlier experiments also, application of P did not influence leaf P status. The leaf P content was maintained in the medium range even when the available P status of the soil was low (Kalam *et al.*, 1979; Pushpadas *et al.*, 1979). It was suggested that trees could be meeting their P requirement from organic forms resulting from the cover crop residues and leaf litter of

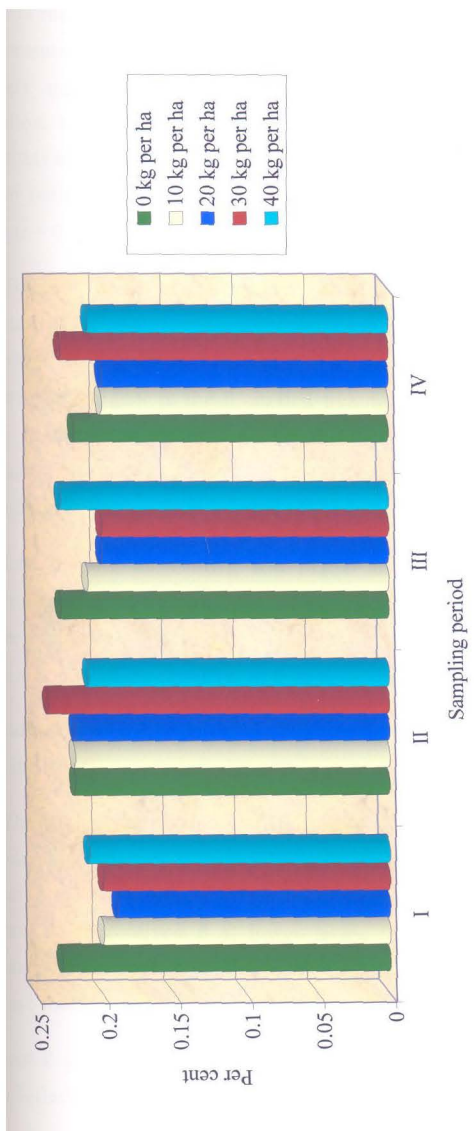


Fig. 2 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on leaf phosphorus status during different sampling periods

rubber. In the present experiment also, cover crop was maintained during the immature phase. Trees in the treatment which did not receive P fertilizers might be able to acquire sufficient P without external application. Lack of influence of P application on growth and yield also support this observation (Table 2 and Table 9). Correlation studies (Table 69) also indicated lack of correlation between leaf P status and soil P content ($r = 0.115$).

Different levels of P did not significantly influence the content of other nutrients in the leaves except magnesium. In the case of calcium, even though no significant difference was observed between treatments, the content was higher in the P applied treatments (Table 14). This might be due to the addition of calcium through rock phosphate in these treatments. Pushparajah and Teng (1972) and Punnoose (1993) also observed higher leaf calcium content in P applied plots.

Leaf magnesium content was higher in the P applied treatments after fertilizer application and the difference was significant after second fertilizer application (Table 15). This might be due to the supply of Mg through rock phosphate (5.63 %) in P applied treatments.

5.3.2 Influence of P Levels on the content of P and Micronutrients in the Roots

The content of phosphorus in fine roots during first, second and fourth sampling was not significantly influenced by P application (Table 20). During the third sampling (before second fertilizer application), the treatments which received P at lower levels (@10 and 20 kg ha⁻¹) had significantly lower root P content compared to other treatments. During second and fourth sampling (after first and second fertilizer application),

root P content was higher in P applied treatments. However, this was not reflected in the leaf P content (Table 11). The content of iron,

manganese, zinc and copper in the root was not influenced by application of P (Table 21 to Table 24). The decrease in the soil micronutrient content observed in the treatments which received higher levels of P (Table 57 to Table 60) was not reflected in their respective root contents.

5.4 EFFECT OF PHOSPHORUS ON ROOT CATION EXCHANGE CAPACITY (CEC)

The application of phosphorus did not influence the CEC of roots significantly (Table 29). However CEC was lowest in the control treatment which did not receive any P fertilizer. Fox and Kakar (1964) reported that uptake of P in legumes was positively correlated with the CEC of roots. Significant positive correlation between the CEC of roots and P uptake was reported by Ram (1980).

5.5 INFLUENCE OF P LEVELS ON DRY MATTER ACCUMULATION

The dry matter accumulation in different parts of the tree are presented in Table 25 and Fig. 3. The total dry matter accumulation observed in the present experiment was slightly less than that reported by Amma (1995) possibly due to the difference in the size of the trees. The total dry matter accumulation in different parts of the tree showed a variation of less than 6 per cent. This might be due to the lack of influence of P on the growth of trees. Similar results were also reported earlier by Ananth (1966) and Punnoose *et al.* (1975). None of the growth parameters studied (girth, length of tapping panel and bark thickness) in the present experiment were influenced by levels of P applied (Table 2 and Table 3). Application of phosphorus brought about a change in some of the root parameters (Table 6 to Table 8), but this failed to bring a change in the total dry matter accumulation. Maximum dry matter accumulation was observed in the branches followed by main trunk. These results are in conformity with those reported by Gunatilleke (2002).

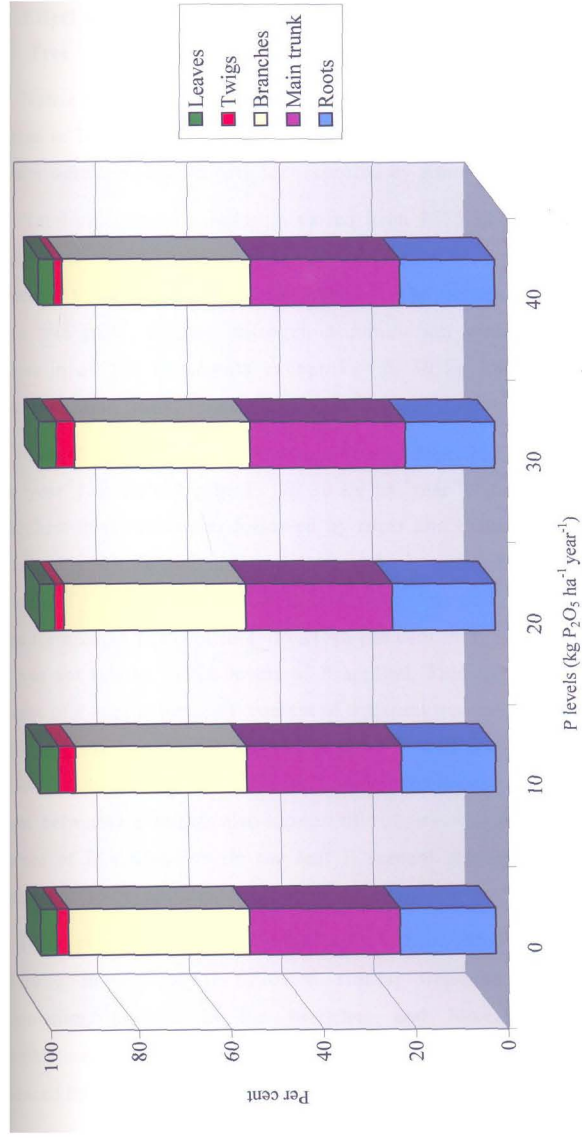


Fig. 3 Dry matter accumulation in different parts of the tree (%) as influenced by levels of P

5.5.1 Effect of P on Nutrient Accumulation in Different Parts of the Tree

Nutrient accumulation in the different parts of the tree are presented in Table 26a to Table 26i. The nutrient accumulation in different tree parts were comparable with that reported by Amma (1995).

Total nitrogen accumulation varied from 7655.52 g in T₅ (@ 40 kg ha⁻¹year⁻¹) to 9439.14 g in T₂ (@ 10 kg ha⁻¹year⁻¹). Between the treatments, there was no definite trend in the content of nitrogen in various tree parts. Highest nitrogen accumulation was observed in the branches in all the treatments except T₄ (@ 30 kg ha⁻¹year⁻¹) and was followed by main trunk. Lowest accumulation was observed in the twigs.

Total P accumulation in the trees ranged from 755.98 g in T₃ (@ 20 kg ha⁻¹year⁻¹) to 863.59 g in T₄ (@ 30 kg ha⁻¹year⁻¹). Accumulation of P was highest in the branches followed by roots and main trunk in most of the cases (Fig. 4). Total P accumulation in the branches ranged from 28.58 per cent in T₂ to 36.07 per cent in T₅. Accumulation of P in the main trunk ranged from 26.26 per cent in T₃ to 21.46 per cent in T₄. The accumulation of P was not related to the levels of P applied. This indicates the lack of influence of P application on P content of different tree parts. Trees appear to be able to maintain the P content without P addition even though the available P status of the soil was low. Lack of significant variation in leaf P content between treatments also support this observation (Table 11). Lack of influence of P application on the leaf P content was earlier reported by Kalam *et al.* (1979) and Pushpadas *et al.* (1979).

Total potassium accumulation varied from 5642.80 g in T₁ to 6884.04 g in T₂ and it followed similar trend as the dry matter accumulation, highest in the branches and lowest in the twigs. Accumulation of potassium in different tree parts did not appear to be influenced by the levels of P applied.

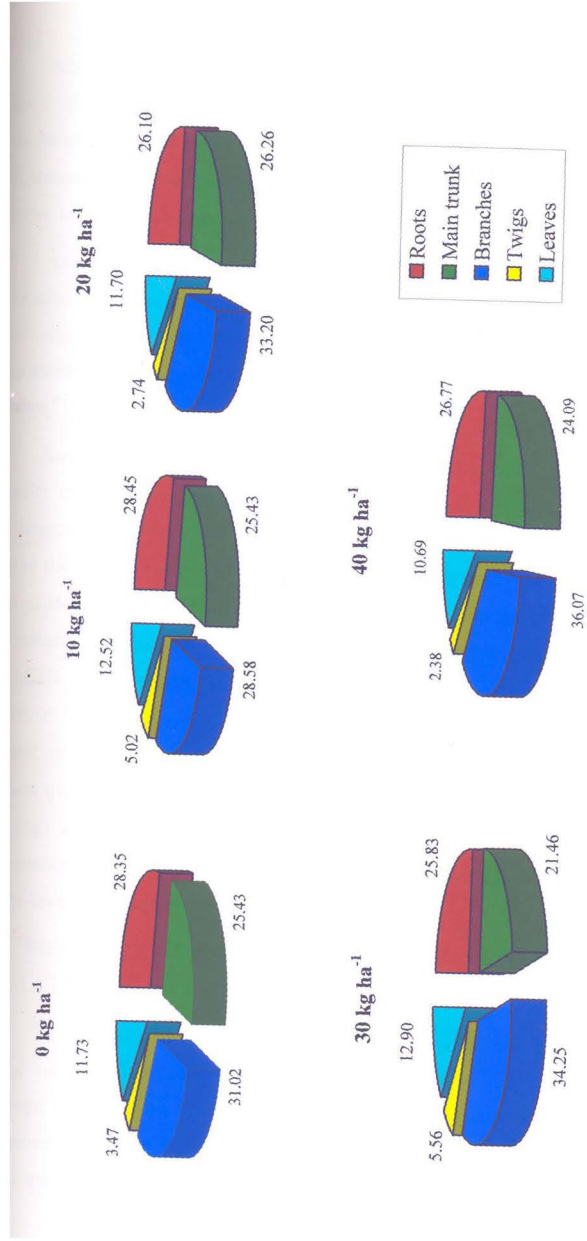


Fig. 4 Distribution of P in the dry matter (%) as influenced by levels of P ($kg\ P_2O_5\ ha^{-1}\ year^{-1}$)

Among the different nutrients, accumulation of calcium was the highest and it varied from 17162.63 g in T₂ to 14192.47 g in T₃. More than 70 per cent of the total calcium was accumulated in the main trunk and branches. The content and distribution of calcium in different parts of the tree was not influenced by the application of P.

Accumulation of magnesium also followed the same trend as the dry matter accumulation. Branches, main trunk and roots accumulated the major share of the total magnesium in the dry matter. The distribution of magnesium in various tree parts did not appear to be influenced by the levels of P applied.

Among the micronutrients, iron was accumulated in the highest quantity in the dry matter. Unlike other nutrients, iron was accumulated in the largest quantity in the roots. This might be due to the deposition of iron in the roots in soils with high sesquioxide content. More than 80 per cent of the total iron in the tree was accumulated in the roots. However, the accumulation of iron in the various parts of the tree was not influenced by levels of P application. Highest accumulation of manganese was observed in the main trunk followed by branches. Zinc accumulation was highest in the branches. Next to iron, copper was accumulated in the highest quantity. Copper content was highest in the main trunk (232.1 – 261.2 g) and lowest in the branches (84.1-97.2 g). The content or the distribution of the various micronutrients did not appear to be influenced by the levels of P applied.

5.6 INFLUENCE OF P LEVELS ON NUTRIENT CONTENT IN THE LATEX

The content of different nutrients in the latex was not influenced by the treatments (Table 41 to Table 49). Definite trend was also not observed between the levels of phosphorus and nutrient content of latex. Compared to other nutrients, nitrogen and potassium were present in higher quantity in the latex. Manganese was present in the lowest quantity.

The data clearly indicated that application of phosphorus did not influence the latex P content. This result is in agreement with other results obtained in the present experiment, *i.e.*, leaf P content and P accumulation in the trees were not influenced by application of phosphorus, indicating that trees are able to maintain sufficient P content without external application of P.

5.7 INFLUENCE OF P ON NUTRIENT REMOVAL THROUGH LATEX

Perusal of the data indicated that nutrient removal through latex was not influenced by levels of P applied (Table 50). Nutrient removal through latex is directly related to the nutrient content in the latex (Table 42 to Table 49) and yield (Table 9). Both these parameters were not influenced by the different treatments and hence the nutrient removal through latex was also not affected. The removal of nitrogen varied from 12.91 kg ha⁻¹year⁻¹ in T₅ (@ 40 kg P₂O₅ ha⁻¹year⁻¹) to 15.24 kg ha⁻¹year⁻¹ in T₄ (@ 30 kg P₂O₅ ha⁻¹year⁻¹). The removal of P varied from 4.26 kg ha⁻¹year⁻¹ in T₅ to 4.81 kg ha⁻¹year⁻¹ in T₄ (Fig.5). In fact the removal of P was lowest from the treatment which received the highest level of P. This again indicated the lack of influence of P on the removal of P through latex. The removal of potassium varied from 12.22 kg ha⁻¹year⁻¹ in T₅ to 13.91 kg ha⁻¹year⁻¹ in T₄. The quantities of nutrients removed through latex in the present experiment are comparable with those reported by Dijkman (1951) *i.e.*, 15.20, 6.00, 11.20, and 0.60 kg ha⁻¹ year⁻¹ of N, P, K, and Ca respectively.

When the loss of nutrients through run off, leaching etc are not considered, the only loss from a mature rubber plantation is through latex on an annual basis. The data indicated that in the case of rubber, removal through latex is less when compared to other crops where a part or whole of the plant is removed as the economic produce. The P addition through litter annually is slightly more than removal through latex and this probably led to non-response to external P addition through fertilizers.

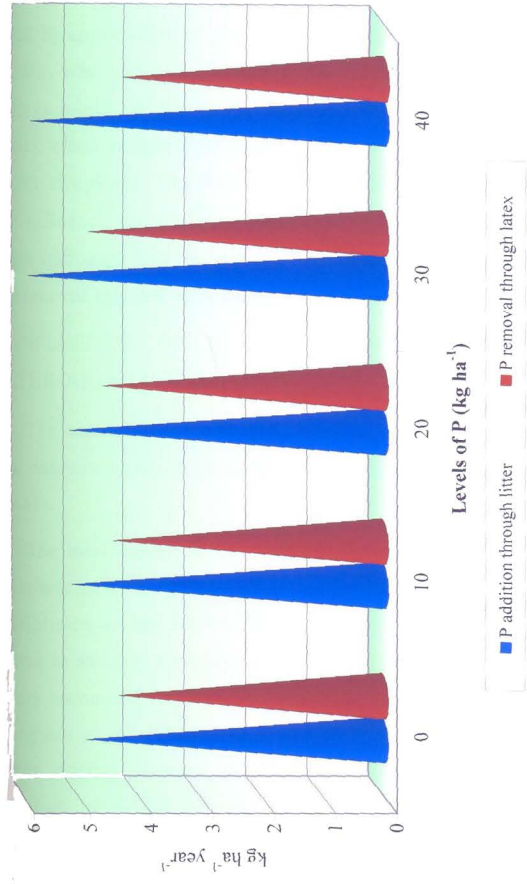


Fig. 5 P addition through litter and removal through latex as influenced by levels of P (kg P₂O₅ ha⁻¹ year⁻¹)

5.7.1 Effect of P on Mg/P Ratio in Latex

The data presented clearly indicated that Mg/P ratio in the latex was not influenced by the application of phosphorus (Table 51). The ratio between magnesium and phosphorus in the latex is directly related to their respective contents in the latex. Latex Mg and P contents were not affected by application of phosphorus (Table 42 and Table 45) and hence their ratio was also not affected. The variation in the ratio between the samplings might be due to the variation in the content of phosphorus in the latex since magnesium content did not show much variation during different samplings. Mg/P is one of the stability parameters of the latex which influence the pre coagulation on tapping cut (Beaufils, 1957) and the data indicated that the stability of the latex is not influenced by levels of phosphorus applied with respect to this parameter.

5.8 INFLUENCE OF P LEVELS ON NUTRIENT RECYCLING THROUGH LITTER

The annual litter addition, nutrient content in the litter and the annual nutrient addition through litter are presented in Table (Table 30 to Table 40).

The total annual litter addition was comparable with that reported by Vergheese *et al.* (2001) *i.e.*, 6.8 to 7.8 t ha⁻¹. Rubber is a deciduous tree and defoliates during December-January. Maximum quantity of litter was collected in January followed by February. Litter fall during December to February accounted for 54.4 per cent of the total litter addition. According to Vergheese *et al.* (2001), 72.7 and 76.7 per cent of the total litter fall occurred during January to March in consecutive years in Tripura. Application of P did not have any significant influence on the litter production in the present experiment. Contrary to this, Punnoose (1993) observed that application of phosphorus at 30 kg ha⁻¹ significantly enhanced litter production when compared with the treatment which did not receive any P fertilizer.

The content of different nutrients studied (nitrogen, phosphorus, potassium, calcium, magnesium, iron, manganese, zinc and copper) were comparable in different treatments during all the samplings. Lowest contents of N, P, Fe and Zn in the litter during the defoliation period indicate efficient re-translocation of these nutrients from the senescing leaves for storage in the trees and subsequent re-mobilisation in the emerging leaves. In the case of P, lowest concentration in the litter was observed in the treatment which did not receive any P fertilizer during October (Table 32). During January also, the P concentration was low in the control treatment. This might be the adaptive mechanism of plants in low P soil for improving P utilization within plants. According to Rao *et al.* (1999), about 50 to 75 per cent of the P contents are re-translocated from a leaf before it is shed and the ability of a plant to remobilize P from senescing to growing points forms an important mechanism to improve utilization of acquired P.

Potassium content was highest in the litter collected in January and then showed a decreasing trend. Calcium content was lowest in the litter collected during April and then showed an increasing trend till October and then decreased during January. Magnesium content was highest during April and then decreased during subsequent samplings. Zinc content was highest during April whereas copper content was highest during July.

The total quantity of nitrogen added through litter varied from 114.17 kg ha⁻¹ year⁻¹ in the T₁ (@0 kg P₂O₅ ha⁻¹ year⁻¹) to 134.70 kg ha⁻¹ year⁻¹ in T₄ (@30 kg P₂O₅ ha⁻¹ year⁻¹). No significant difference was observed between treatments with respect to nitrogen addition.

Application of phosphorus significantly influenced the total annual P addition through litter fall (Table 40). The lowest addition was observed in T₁ which did not receive any P fertilizer (4.84 kg ha⁻¹ year⁻¹) and highest in T₄ (5.90 kg ha⁻¹ year⁻¹). The P addition in T₁ was comparable with T₂ and T₃ which received P₂O₅ @ 10 and 20 kg P₂O₅ ha⁻¹ year⁻¹. The

treatments which received higher levels of P (@ 30 and 40 kg P₂O₅ ha⁻¹ year⁻¹) had a significantly higher P addition compared with T₁. This might be due to the lower litter addition and lower litter P content in T₁.

The total addition of other nutrients through litter was not influenced by application of P. The addition of potassium varied from 53.59 kg ha⁻¹ year⁻¹ in T₁ to 61.26 kg ha⁻¹ year⁻¹ in T₂ (@10 kg P₂O₅ ha⁻¹ year⁻¹). Addition of calcium varied from 127.83 kg ha⁻¹ year⁻¹ in control treatment to 145.86 kg ha⁻¹ year⁻¹ in T₃ (@ 20 kg P₂O₅ ha⁻¹ year⁻¹), magnesium from 12.64 in T₁ to 17.16 kg ha⁻¹ year⁻¹ in T₂, iron from 15.51 to 18.05 kg ha⁻¹ year⁻¹, manganese from 2.91 to 3.29 kg ha⁻¹ year⁻¹, zinc from 0.28 to 0.32 kg ha⁻¹ year⁻¹ and copper from 0.74 to 0.89 kg ha⁻¹ year⁻¹. The nutrient addition through litter was comparable with that reported by Verghese *et al.* (2001) except for potassium. The addition of potassium through litter in the present experiment was higher and this might be due to the higher content of potassium in the litter.

The addition of N, K, Ca, Mg, Fe, Mn, Zn and Cu through litter is much more than their removal through latex. The addition of P through litter, 4.84 kg ha⁻¹ year⁻¹ is slightly more than the removal through latex, 4.30 kg ha⁻¹ year⁻¹ (Fig.5). The loss of P through run off will be low in a rubber plantation where adequate soil conservation measures are adopted. Hence if run off and leaching losses are not considered, the only loss of P from a mature rubber plantation is through latex, and the data indicate that the rubber ecosystem can sustain its P level without external application during the mature phase through nutrient recycling from litter.

5.9 INFLUENCE OF P LEVELS ON P FRACTIONS IN THE SOIL

Application of P significantly influenced the total P content of the soil, but did not influence the content of different fractions (Table 61). The distribution of added phosphates into various forms depends on the added fertilizer, its solubility and nature, soil characteristics like

pH, minerology, amount of free iron and aluminium oxides etc. (Anjaneyulu and Omanwar, 1979). Very low saloid P content of the soil observed might be due to the very high fixation capacity of the soil as reported by Kothandaraman and Krishnamoorthy (1979). Amma *et al.* (1991) also observed saloid bound P in trace amounts in soil of the same area. Nziguheba *et al.* (2002) reported that continuous P application for five seasons at the rate of 150 kg ha⁻¹ did not result in an increase in the labile P content of the soil in a P-fixing Acrisol in Kenya.

Scrutiny of the data revealed that Fe- P was the most predominant inorganic P fraction and the mean percentage of Fe- P to total P was 20. This might be due to the gradual change of different P fractions to Fe-P in acidic soils with high sesquioxide content. Chang and Jackson (1958) reported that phosphatic fertilizers added to the soil change in to three forms, Al-P, Fe-P, and Ca-P in acidic and neutral soils. It was reported that as time elapses, Al-P shifts to Fe-P (Hsu and Jackson, 1960; Fiskell and Spencer, 1964). With prolonged contact of added P with soil, such a shift takes place irrespective of soil pH. Amma *et al.* (1991) reported Fe-P fraction and George *et al.* (2001) reported Fe-P and Al-P as the major inorganic P fractions in rubber growing soils.

Next to Fe-P, the major inorganic fraction was Ca-P, the mean values accounting for 10.72 per cent of total P. Reductant P accounted for 5.52 per cent of total P. Application of different levels of P failed to bring any significant difference in the reductant P content in different treatments. This result is in conformity with the findings of Chang and Jackson (1958). The organic fraction was the most predominant fraction and it varied from 51.13 per cent in the treatment which did not receive any P fertilizer (T₁) to 52.23 per cent in the treatment which received the highest level of P (T₅) with a mean of 55.65 per cent.

Total P in the treatments which received higher levels of P (T₄ and T₅) was significantly greater than that of the control treatment.

Anjaneyulu and Omanwar (1979) also reported similar results in a slightly acidic soil.

5.10 INFLUENCE OF P LEVELS ON SOIL NUTRIENT STATUS

Soil phosphorus status at bimonthly intervals from May to November is presented in Table 52 and Fig 6. The soil P status in the control treatment (T₁) was in the low range during all the samplings. During the first sampling, all the P applied treatments had significantly higher P status compared to T₁. The samples were collected before fertilizer application and the higher P status in the P applied treatments might be due to the residual effect of the previously applied phosphorus. Treatments which received P at 20 and 30 kg P₂O₅ ha⁻¹ year⁻¹ (T₃ and T₄) had a significantly higher soil P status compared with T₂ which received P at 10 kg/ha. Soil P status was highest in the treatment which received the highest level of P (T₅) and it was significantly higher compared to other treatments which received P at lower levels. During other samplings, soil P status in the control treatment was comparable with T₂ and T₃ which received P at 10 and 20 kg P₂O₅ ha⁻¹ year⁻¹. Soil P status in T₄ and T₅ were comparable and were significantly higher compared to other treatments. This is in conformity with the reports of Pushparajah (1969) and Lim (1977) in rubber. Compared to nitrogen and potassium, loss of phosphorus from the soil is limited and a part of the applied phosphorus gets accumulated in the soil leading to higher soil P levels. However, P status of treatments which received P at lower levels (0, 10 and 20 kg P₂O₅ ha⁻¹ year⁻¹) were comparable with the control treatment during three samplings. According to Nziguheba *et al.* (2002), considering P losses due to run off and erosion, application of P at 10 kg ha⁻¹ was not sufficient, but application at 25 kg ha⁻¹ resulted in gradual replenishment.

Soil organic carbon content and potassium status were not influenced by the levels of P applied during any of the samplings (Table 53 and Table 54).

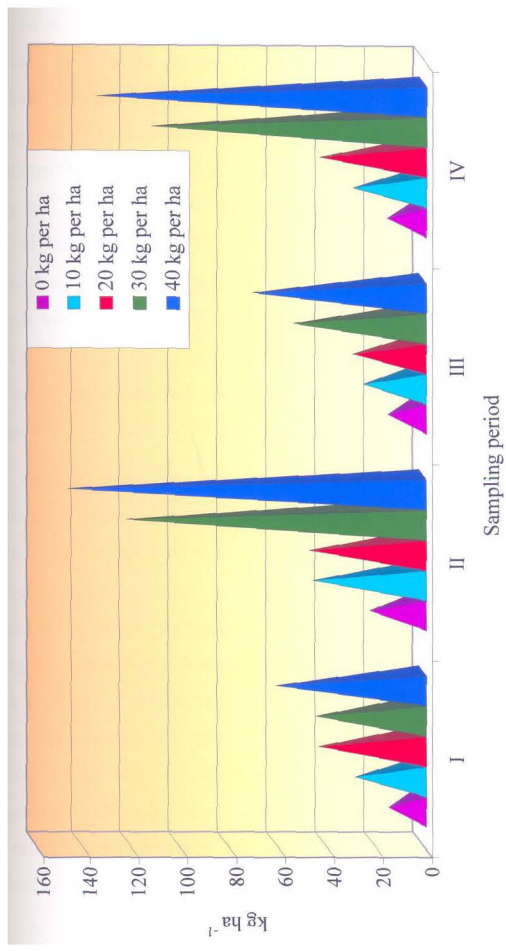


Fig. 6 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on soil phosphorus status during different sampling periods

Soil calcium content was higher in the phosphorus applied plots (Table 55). During the first sampling, there was no significant difference between treatments. Second sampling was done after first fertilizer application and significant differences were observed between treatments. Soil calcium content in the treatments which received P at higher levels (T_4 and T_5) were significantly higher compared to other treatments. During the third sampling also, lowest calcium content was observed in the control treatment and it was comparable with T_2 which received P at 10 kg ha^{-1} , and significantly lower than that of other treatments which received higher levels of P. During the last sampling (after second fertilizer application) also, the same trend was observed. The higher calcium content in the P applied plots might be due to the addition of calcium through Rajphos (CaO-38.30 %). However, higher calcium content in the P applied treatments was not reflected in the leaf calcium content.

Soil magnesium status was higher in the phosphorus applied treatments (Table 56). With increasing levels of P, the soil Mg content increased in majority of the situations. During the first and third sampling (before first and second fertilizer application), there was no significant difference between treatments, however, the same trend was noticed. During the second and fourth sampling, soil Mg content in the treatments which received P at higher levels (30 and 40 kg ha^{-1}) were significantly higher. This higher Mg status in the P applied treatments might be due to the addition of magnesium through Rajphos which contain 5.63 per cent magnesium. During the fourth sampling (after second fertilizer application), leaf nutrient content also reflected the same trend (Table 15), leaf Mg content was significantly higher in P applied treatments.

In the case of soil iron content, there was no significant difference between treatments during the first two samplings and no definite trend was observed (Table 57). During the third sampling, iron content was

highest in the control treatment (T₁) and the treatments which received P had a significantly lower iron content. During the last sampling also, iron content was highest in the control treatment. This might be due to the formation of iron phosphates with less solubility under higher levels of P. Antagonism between iron and phosphorus was earlier reported by Lucas and Knezek (1972).

In the case of manganese also, highest content was observed in the control treatment (Table 58). In general as the level of P increased, the soil Mn status decreased and the difference was significant during the first and third samplings. This might be due to the formation of phosphorus manganese complex under higher levels of phosphorus as reported by Rao *et al.* (2002) in rubber growing soils. Lucas and Knezek (1972) also reported antagonism between phosphorus and manganese. However, no significant difference was observed in the leaf Mn content (Table 17).

In the case of soil zinc and copper status, no significant difference was observed between treatments (Table 59 and Table 60). No definite trend was observed in the soil zinc status during the first and fourth samplings. However, during the second and third samplings, highest zinc content was observed in the control (T₁) treatment and lowest in the treatment which received P at the highest level (T₅). Soil copper content was highest in the T₁ during all the samplings except during the third sampling. The antagonism of phosphorus with zinc and copper might be the reason for their higher content in the control treatment (Lucas and Knezek, 1972).

5.10.1 Influence of P on Soil Dehydrogenase Activity (DHA)

Dehydrogenase activity is an indication of soil biological activity and microbial population, and is affected by many factors like pH, soil type, organic matter content, fertilizer application, soil moisture content etc. An appraisal of the data indicated that the dehydrogenase activity was not affected by any of the treatments during all the samplings (Table 62).

This might be due to the lack of influence of P levels on microbial population of the soil. Among the different microorganisms only the population of PSB was influenced by the various treatments, which was suppressed after first and second fertilizer application. The population of total bacteria, total fungi phosphofungi and actinomycetes was not influenced by the application of P (Table 67a to Table 67d). However seasonal variations in dehydrogenase activity was observed. The DHA measured during second sampling was higher than that during the first sampling and this level was maintained during the third sampling also. Decomposing rubber litter during the favourable moisture conditions during second and third sampling (July and September) might have resulted in intense microbial activity leading to higher DHA during this period. Serra-Wittling *et al.* (1995) observed higher levels of DHA during the mineralisation flush due to the activity of the microflora in degrading easily metabolizable compounds. As the easily biodegradable substances decreased, DHA also decreased. In the present experiment also, a decrease in DHA was noticed during the last sampling in most plots indicating the near completion of the degradation of rubber litter by this time. The favourable moisture conditions during the second and third sampling also might have contributed to the higher DHA activity during this period. Results in conformity with this were reported by Tiwari *et al.* (1989). They observed lower DHA under lower soil moisture conditions.

5.11 INFLUENCE OF P LEVELS ON THE POPULATION OF RHIZOSPHERE MICROORGANISMS

5.11.1 Root Colonization by Arbuscular Mycorrhizal Fungi (AMF)

An appraisal of data on AMF infection of rubber roots revealed high percentage of infection, but it was not significantly influenced by the different P levels (Table 66). The mean AMF infection was more than 85 per cent in all cases except in the treatment which received P at 20 kg ha⁻¹ during second and fourth sampling. This indicates that rubber roots have a

high percentage of AMF infection. Joseph (1997) observed mycorrhizal spores in all rubber growing soils. However, AMF infection was not influenced by the different treatments. The levels of P tested in this experiments did not have an inhibitory effect on AMF infection. Role of AMF in improving P acquisition by plants is well documented. Enhanced surface area for absorption, production of phosphatases, increased uptake of calcium by mycorrhizal plants and increased affinity for phosphorus absorption are some of the mechanisms by which AMF enhance P uptake (Bolan, 1991). However, there exists variation in the efficiency in mycorrhizal uptake of phosphorus. Baon *et al.* (1994) observed that plants with short root hairs which were agronomically less efficient were more responsive to either mycorrhizal infection or P addition than long root hair plants. Schweiger *et al.* (1995) observed that variation in the degree of VAM colonization between 27 wheat lines and 10 barley cultivars was not significant though they differed in root hair length and density under low P situations. It was suggested that even though the occurrence of mycorrhizae is quite universal, its variation and beneficial effect for P acquisition may be less for crops with extensive root systems with long root hairs. Mature rubber trees have an extensive root system with root hairs (Table 4 and Table 8) and high percentage of AMF colonization, which did not vary between treatments. Hence the extent to which AMF infection improve P acquisition when soil P is low needs to be studied further. In young rubber, Waidyanatha (1980) and Ikram *et al.* (1996) observed better P uptake in plants with mycorrhizal infection. Young *Hevea* plants have limited root system and need more nutrients for active growth. Unlike mature rubber, young rubber responds to phosphorus application (Punnoose *et al.*, 1994). Hence AMF infection might be beneficial for young rubber and its dependency may decrease during mature stage due to extensive root system, likely levelling off of nutrients within the trees, limited crop removal and nutrient recycling through litter.

The treatment which did not receive P fertilizer had several adaptations to improve P acquisition like longer and denser root hairs (Table 8), higher exudation of acid phosphatase from roots (Table 27) etc., indicating that these are more important traits for improving P acquisition in rubber than mycorrhizal uptake.

5.11.2 Influence of P Levels on Total Microbial Count and Phosphorus Solubilizers (PSB) in the Rhizosphere

The data clearly depicts that after fertilizer application, there was a decline in the population of PSB in P applied treatments (Table 67a to Table 67d). The percentage of PSB to total bacteria also showed similar trend. This might be due to the suppression of PSB by added phosphates (Kucey *et al.*, 1989). Gyaneswar *et al.* (2002) reported that substrate availability in the soils affect the population of PSB and availability of utilizable substrates limit the success of inoculation. Before fertilizer application, the population of PSB was comparable in all the treatments. The suppression of PSB after fertilizer application is temporary and might not affect the P nutrition of trees on a long term basis. Results of many inoculation experiments with PSB did not indicate any consistent beneficial effect under field condition (Tandon, 1987).

The population of total bacteria, total fungi, phosphate solubilizing fungi and actinomycetes were not influenced by the levels of phosphorus (Table 67a to Table 67d).

5.12 INFLUENCE OF P LEVELS ON THE INCIDENCE OF TAPPING PANEL DRYNESS

Tapping panel dryness is a major problem encountered in rubber plantations and is characterized by a gradual or sudden drying up of the latex vessels resulting in abnormally low yield or complete stoppage of latex production. The cause of this syndrome is still unknown. It is generally considered to be a physiological disorder caused by excessive

exploitation (Paranjothy *et al.*, 1975). Pushpadas *et al.* (1975) suggested that unbalanced nutrition may lead to the incidence of tapping panel dryness. In the present study, incidence of tapping panel dryness was not influenced by application of phosphorus (Table 68).

5.13 PROCESSES BY WHICH PLANTS IMPROVE THEIR P ACQUISITION UNDER LOW SOIL P SITUATIONS

5.13.1 Root Acid Phosphatase Activity

Secretion of acid phosphatase from roots was highest from trees which did not receive any P fertilizer during all the samplings (Table 27 and Fig. 7).

Phosphatases are enzymes which are capable of hydrolyzing organic phosphate esters and are very important for the P nutrition of plants (Antibus and Lesica, 1990). Enhanced intracellular and extracellular acid phosphatase activity could improve the acquisition and re-utilization of P, thus helping plant growth under P deficient conditions (Yun and Kaepler, 2001). Several scientists have reported many fold increase in root phosphatase activity under P deficient conditions (Ascencio, 1997; Adams and Pate, 1992; Hayes *et al.*, 1999). In the present experiment also, highest acid phosphatase secretion was observed from trees which were not supplied with P fertilizers (T₁) during all the samplings. Acid phosphatase secretion from T₁ was significantly higher in all the samplings except during the fourth sampling. This might be an adaptive mechanism of trees to utilize the organically bound P in soil which constituted 55.65 per cent of total P (Table 61) Pushpadas *et al.* (1979) also suggested that trees could be meeting their P requirement from the organic P in soil and the lack of response of rubber trees to application of phosphorus was attributed to this ability. Trees which received P at various levels had comparable levels of acid phosphatase secretion during most of the samplings. Significant differences between P applied treatments were observed only during the first sampling when the

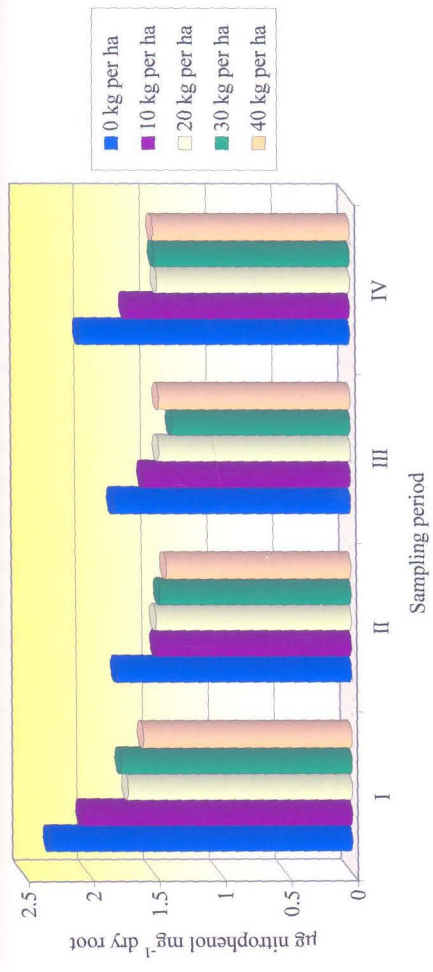


Fig. 7 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on the secretion of acid phosphatase from roots during different sampling periods

secretion from trees supplied with P at 10 kg ha⁻¹ (T₂) was significantly higher than that from trees which received P at the highest level (@ 40 kg ha⁻¹). Significant negative correlation was also observed between soil P status and acid phosphatase secretion from roots ($r = -0.366$).

In general, acid phosphatase secretion was highest during the first sampling. There was a reduction in acid phosphatase secretion during the next two samplings, but showed a slight increase during the fourth sampling. Rubber is a deciduous tree and adds about 5-6 tonnes of organic matter annually through litter fall (Krishnakumar and Potty, 1992). The trees shed their leaves during December-January and refoliate during January- February. Pre-monsoon showers are received during April and plants resume active growth during this period after the summer and refoitation. During this period, release of P from the litter also will be less and the trees will not be able to meet the increased demand for P caused by increased growth and hence secrete more acid phosphatase to increase P availability. This is in conformity with the report of Clarholm and Rosengren-Brinck (1995).

During the subsequent period P from the litter will become gradually available, and acid phosphatase secretion was also comparatively less. Through mineralization, N and P will usually be released from the organic matter in consistent proportion (Berg and Mc Claugherty, 1989). In deciduous forests, most of the P taken up by trees was found to be mineralized from organic matter in the forest floor (Yanai, 1992). It was suggested that feed back mechanism exist between the dead and live components of the ecosystem to favour plant growth (Van-Breemen, 1993). Roots and microorganisms produce phosphatases when P is limiting. This enzyme selectively release P from organic matter without concomitant release of carbon (McGill and Cole, 1981). There was an increase in the secretion of acid phosphatase during the last sampling. Most of the P in the litter will be mineralized before this period

and plants might secrete more enzyme to hydrolyze organic P to meet the P requirement.

The data indicate that trees improve their P acquisition under low soil P situations by increased secretion of acid phosphatase which will hydrolyze organic P which account for more than 50 per cent of the total soil P. This might be one of the reason for the lack of response of rubber trees in terms of growth and yield to application of phosphorus (Table 2 and Table 9). Leaf nutrient status which was comparable in all the treatments also support this observation (Table 11). This indicate the possibility of skipping application of P fertilizers to mature rubber.

5.13.2 Soil Acid Phosphatase Activity

In general, soil phosphatase activity was lower in the treatments which received P at higher levels (Table 63 and Fig. 8).

Plants can utilize only inorganic P and since a large proportion of the total soil P is organically bound, the mineralization of organic fraction is an important factor in P nutrition. During the first sampling, highest soil phosphatase activity was noticed in T₂ which received P at the lowest level (@10 kg ha⁻¹). The stimulation of soil phosphatase activity by low levels of fertilizer phosphorus was reported by Speir and Ross (1978). In forest soils, the activity of phosphatase enzyme increased with a low concentration of soluble inorganic P (Attiwill and Adams, 1993). It was attributed to the increased microbial numbers and plant growth which over a period of time would cause a build up of soil organic matter and enzyme levels. The activity declined at higher levels of P application. However this stimulatory effect was noticed only during the first sampling which was before the first fertilizer application. In the treatments which received P at higher levels, the higher soil P status might have caused a decline in phosphatase activity. Result in conformity with this was earlier reported by Spiers and McGill (1979). During next sampling, there was no significant difference between treatments, but activity was higher in T₁

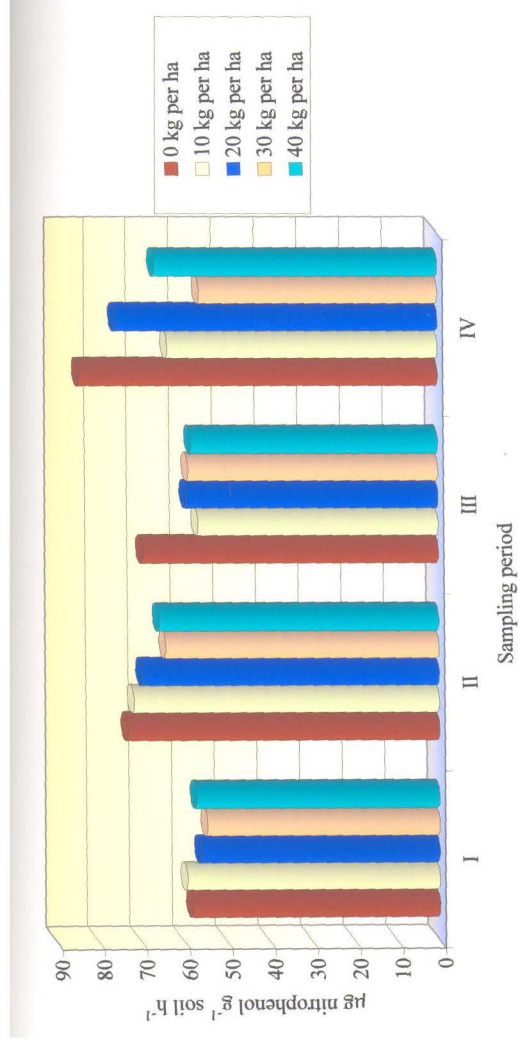


Fig. 8 Soil phosphatase activity as influenced by levels of P ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) during different sampling periods

which did not receive any P fertilizer. T₁ received nitrogen and potassium fertilizers and Speir and Ross (1978) observed that application of fertilizers which did not contain phosphorus generally caused an increase in soil phosphatase activity. The secretion of acid phosphatase in larger amounts in T₁ also might have contributed to the higher soil phosphatase activity in this treatment. During the third sampling, the phosphatase activity in T₁ was significantly higher than that of the P applied treatments. During the last sampling also, the same trend was noticed, but there was no significant difference between treatments. The higher soil P status in the P applied treatments might have declined the activity of phosphatase as suggested by several scientists (Attiwill and Adams, 1993; Hayes *et al.*, 1999).

Phosphatase activity showed variation during the sampling periods. The activity was lowest during the first sampling and then increased during subsequent samplings. Being a deciduous tree, rubber defoliates during December-January. The higher activity observed during the second sampling might be due to the higher microbial activity associated with the decomposing rubber litter during this period and favourable moisture conditions. During the next samplings, the enzyme activity was higher, but no definite trend was observed. Enzyme activity decreased during the third sampling and showed an increase in most of the cases during the fourth sampling. The fluctuations in soil moisture content and microbial population might have resulted in this variation. Similar results were reported by Breakwell and Turco (1989), who observed greatest phosphatase activity under field conditions at the beginning of the decomposition period. The activity then decreased approximately for 75 days and increased near the end of the decomposition period which was attributed to the rupturing of cells and release of cell constituents.

Decomposition by microorganisms is the dominant process for P release from organic matter. Some P is also actively released from organic

matter through biochemical mineralization when roots and microorganisms selectively release P from the organic matter through production of phosphatases (McGill and Cole, 1981). The induced phosphatases hydrolyze the ester bonds binding P to carbon in organic matter releasing P without concomitant release of carbon. Phosphatases are produced when P is the most growth limiting nutrient and an increase in phosphatase activity in soil reflect an increased need for P. In the present experiment, the higher soil phosphatase activity in T₁ which was not supplied with P fertilizers might be to increase the solubilization of organic P.

Secretion of acid phosphatase by plant roots in to the rhizosphere form a source of phosphatase in soil. In general, secretion of acid phosphatase by roots and soil phosphatase activity was higher in T₁ which did not receive any P fertilizer and lower in P applied treatments (Fig.7 and Fig. 8). However, no definite trend was observed in the seasonal variations in enzyme activity. The acid phosphatase secretion by roots was highest during the first sampling, then showed a declining trend during the next two samplings whereas the soil phosphatase activity was lowest during this period and then showed an increasing trend. This indicates that the major source of phosphatases in soil in mature rubber plantations is microorganisms, and not the root secretion.

5.13.3 Root Phosphoenol Pyruvate Carboxylase (PEPC) and Malate Dehydrogenase (MDH) Activities

In general, PEPC and MDH activities in the roots were higher in the trees which did not receive any P fertilizer (Table 28, Fig. 9 and Fig.10). Several scientists have observed increased exudation of organic acids from roots of many plants under conditions of P stress (Bar-Yosef, 1991 and Jones, 1998). The most frequently reported ones are those from the Kreb's cycle and associated biochemical pathways that are important metabolites within plant cells *i.e.*, di and tricarboxylic acids like oxalic acid, oxaloacetic acid, malic acid, fumaric acid, succinic acid, isocitric

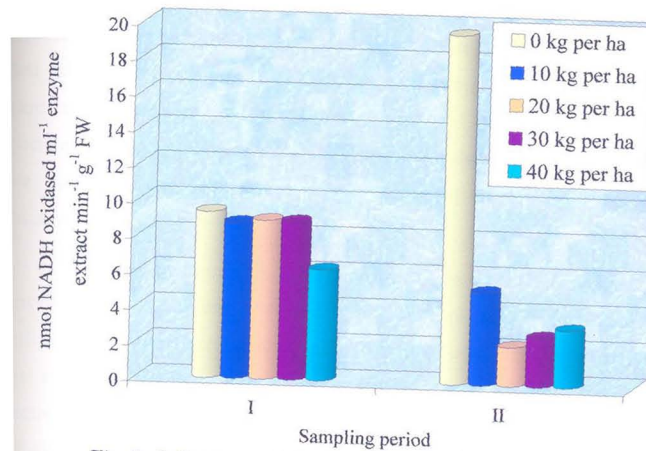


Fig. 9 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on PEP carboxylase activity

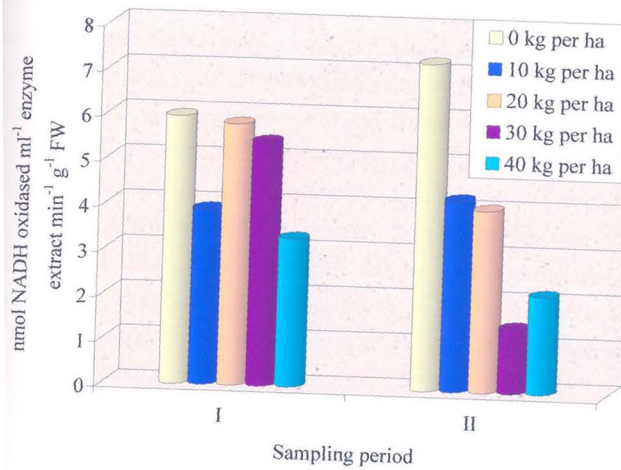


Fig. 10 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on malate dehydrogenase activity

and citric acid. Among these citric acid and oxalic acid have the highest affinity for soil adsorbents and hence most effective in solubilizing soil inorganic P by displacing P ions sorbed on to soil constituents through ligand exchange (Hocking, 2001). Because of the low pK values of many organic acids compared with the neutral cytosol pH, organic acids are dissociated in the cytosol of root cells and are released as organic anions along with H^+ ions leading to some acidification of the rhizosphere (Hinsinger, 2001)

In the present experiment, higher PEPC and MDH activities were observed in the treatment which did not receive any P fertilizer (T_1). Significant negative correlation (Table 69) was observed between soil P status and root PEPC and MDH activities ($r = -0.519$ and -0.513 respectively). This might be the biochemical response of plants to limited soil P availability. The P deficiency induced exudation of carboxylic acids depends on the ability of plants to accumulate carboxylic acids in the root tissue, which in turn is determined by the biosynthesis, degradation and partitioning of carboxylic acids or related precursors between root and shoot system. Johnson *et al.* (1996) suggested transcriptional regulation of PEPC in response to P limitation. Enhanced expression and activity of PEPC was linked with P deficiency induced biosynthesis and root exudation of carboxylic acids under P limitation. PEP carboxylase reaction which liberates oxaloacetate and P_i have a function for P_i recycling in PEP catabolism as a bypass for the ADP and P_i dependent pyruvate kinase (Theodoru and Plaxton, 1993). In P deficient white lupin, dark CO_2 fixation by PEPC provided up to 34 per cent of the carbon which was exuded as citrate in to the rhizosphere (Johnson *et al.*, 1996). The accumulation and subsequent oxidation of large amount of citric acid by roots of P deficient plants depend on the differential regulation of both biosynthesis and breakdown of citric acid in the root tissue.

According to Bar-Yosef (1991), under P stress, organic acid synthesis rate, particularly citric acid is enhanced. It was found that PEPC in roots was involved in the extra citric acid production, probably due to malate conversion to citrate. Neumann and Romheld (1999) observed increased PEPC activities in the roots of P deficient white lupin, chickpea and tomato. Hocking (2001) also reported positive correlation between organic acid exudation from roots of rape seed and white lupin and PEPC and MDH activity.

In most higher plants, an energy mediated co-transport process, driven by the protons generated by a plasma membrane H^+ ATP ase, is mostly responsible for P_i uptake. Phosphate absorption is accompanied by H^+ influx with a stoichiometry of 2 to 4 H^+ / H_2PO_4 transported. The number of protons increased as the concentration of P_i in the media decreased, the required protons being supplied by the media or activated proton pumps in the membrane (Raghothama, 1999). In the present experiment, the higher PEPC and MDH activity in the control treatment indicate a possibility for exudation of organic anions and H^+ ions in to the rhizosphere.

Except in some extreme cases such as that reported for proteoid roots of white lupin, the organic anions exuded by plant roots are rather small fluxes in comparison with fluxes of H^+ or OH^-/HCO_3^- released by roots. In addition, they are rapidly metabolized by rhizosphere microflora, the measured half life ranging from 2 to 12 h. Hence the efficiency of organic anions exuded by the roots to significantly increase the bio-availability of inorganic P is a question for debate. However, increased activity of enzymes involved in the organic acid synthesis might be playing a role in P uptake along with other adaptations of plants under low P situations (Hinsinger, 2001).

The data indicate that the higher levels of PEPC and MDH in the roots of trees which were not supplied with P fertilizers might be an

adaptive mechanism to improve their P acquisition. Lack of response of rubber trees to application of P and leaf P status which was maintained in the medium range in the control treatment (Table 11) also support this observation. This again indicate the possibility of skipping application of P fertilizers to mature rubber without any adverse effect.

5.13.4 Rhizosphere pH

Rhizosphere pH was lower in the treatment which did not receive any P fertilizer (T₁) compared to the treatments which received phosphorus fertilizer, and the same trend was noticed during most of the samplings (Table 65 and Fig.11).

Rhizosphere pH has a strong influence on the availability of P to plants. Riley and Barber (1971) observed that the amount of P taken up by soybean increased linearly with decreasing pH. Enhanced H⁺ release by plants under conditions of P deficiency was reported by many scientists (Imas *et al.*, 1997; Bertrand *et al.*, 1999). Under conditions of P deficiency, inhibition of NO₃ uptake led to rhizosphere acidification due to increase in the uptake of cations over anions (Neumann and Romheld, 1999). Studies with phosphate rocks indicated that H⁺ release by plant roots considerably increased the dissolution of phosphate rocks and availability of P to plants.

Though rhizosphere pH was lower in the control treatment which did not receive any P fertilizer, it was higher than non-rhizosphere soil pH in many cases (Table 64). According to Hinsinger and Gilkes (1996) root induced acidification of the rhizosphere increased the bio-availability of P whenever calcium phosphates were present *i.e.*, in alkaline to mildly acidic soil, but its effect was not clear in acidic soils. The dominant forms of P minerals in acidic soil are Fe and Al phosphates and their solubility decreases with decreasing pH. Moreover the positive surface charge of Fe and Al oxides and hence their P adsorption capacity increases with decreasing pH. Hence

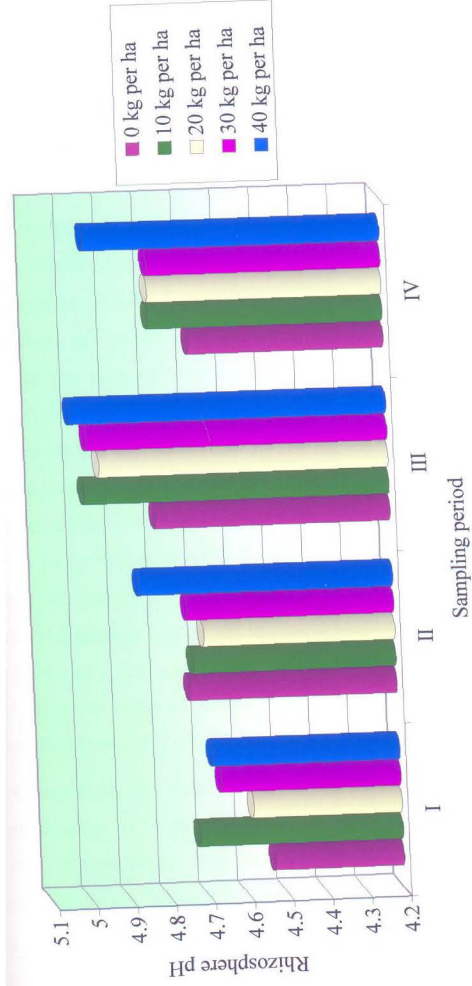


Fig. 11 Influence of P levels ($\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) on rhizosphere pH during different sampling periods

increasing the rhizosphere pH might be more effective in increasing the availability of P in acidic soils (Hinsinger, 1998). Contrary to this, Geelhoed *et al.* (1997) observed that decreasing the pH led to lesser P adsorption on to goethite and increased availability of P to plants. It was noticed that competitive adsorption of sulphate ions considerably increased with decreasing pH resulting in larger equilibrium concentration of P in soil solution at acidic pH range of 4-5 than at a pH of 6-7.

In the present experiment, the soil of the experiment area was acidic, but still a reduction in rhizosphere pH in the control treatment was observed, when compared with the P applied treatments in most of the cases. Reduction in rhizosphere pH might increase the solubilization of calcium P fractions in the soil. In the present experimental site, Ca-P was the second highest P fraction after Fe-P (Table 61). Higher levels of PEPC and MDH in the roots of trees which did not receive any P fertilizer indicate exudation of H^+ ions along with organic anions from the roots and this might have contributed to this reduction in rhizosphere pH. Release of H^+ ions by P solubilizers (Kucey *et al.*, 1989) and AMF fungi associated with the roots (Buwalda *et al.*, 1983) also might have contributed to this reduction in rhizosphere pH. However, the slightly higher pH in the rhizosphere when compared with non-rhizosphere soil pH in most cases indicate a general rhizosphere alkalization by trees to increase the availability of phosphorus by anion exchange from iron and aluminium phosphates.

Comparing between the sampling dates, significant differences between treatments were observed during first and third sampling (before fertilizer application). Samples collected after fertilizer application (second and fourth sampling) did not indicate any significant difference among treatments. Rock phosphate was applied as the source of P and rhizosphere acidification might have occurred in the fertilizer applied plots for the solubilization of calcium phosphate. This might be the reason

for the lack of significant difference among treatments after fertilizer application.

The lower rhizosphere pH in the control treatment (Table 65) and higher PEPC and MDH activities in roots (Table 28) indicate that trees improve their P acquisition when the soil P is low by exuding organic anions and H^+ ions. However the general alkalization of the rhizosphere indicate that the trees enhance the solubility of iron and aluminium phosphates by increasing pH. This might be one of the reason for the lack of response of rubber trees to P application both in terms of growth and yield. The leaf P status which was comparable in different treatments also support this finding.

5.13.5 Root Characters

5.13.5.1 Root Spread and Root Volume

Rubber roots explored a large area of soil (89.38 m^2) when they were 19 years old (Table 4). This will give the plants access to soil nutrients and water in this large area. According to Webster and Paardekooper (1989), lateral roots of mature rubber of 7-8 years extended over 9 m. Roots of adjacent plants were found to overlap indicating a possibility of competition for nutrients and water among adjacent plants.

The roots also had a large volume (0.047 m^3) which help the plants to explore large volume of soil (Table 4). The different treatments did not cause much variation in the root area and volume indicating that this is a genetic character of the tree and is not influenced by the different levels of P applied.

5.13.5.2 Vertical Distribution of Roots

Vertical distribution of roots is shown in Table 5. More than 60 per cent of the roots explored the top 0-10 cm layer. This result is in conformity with that of Soong (1976). About 23.88 per cent roots explored 10-30 cm, 7.42 per cent 30-60 cm layer and 2.75 per cent in the 60-90 cm

soil layer. The different treatments did not show much variation in the depth wise root distribution indicating that it is not influenced by application of P fertilizers.

Most of the feeder roots were found to be concentrated in the surface layer (more than 60 %). This will help the plants to exploit the surface soil rich in organic matter through litter addition more efficiently. In most cases a mat of fine roots was observed on and just beneath the litter layer, above the soil surface also. Similar observations were made by Sivanadyan *et al.* (1995). The minimum tillage practiced in rubber plantations ensures maximum fine root development in surface layer, since root disturbance is kept to a minimum. The concentration of roots in the surface layer will enhance uptake of P. Since P availability is usually highest near the soil surface, the production of shallower root system under P stress is an important contributor to the efficiency of P acquisition (Ge *et al.*, 2000). Forde and Lorenzo (2001) reported that under conditions of low P availability, the growth angle of basal roots were reduced and they became more horizontal. Lynch and Brown (2001) explained the biochemical regulation of root growth under low P situations. It was suggested that P could regulate ethylene synthesis, which would alter ethylene levels and therefore ethylene responses in tissues. By reducing ethylene responsiveness under low P availability, root elongation continue under conditions which reduce overall growth, permitting the plant to explore additional soil domains. In the case of mature rubber, roots explore a large area of soil giving the plants access to phosphorus in this area. Minimum tillage and the litter layer on the surface will promote surface root development which will enhance P uptake. The data indicate that mature rubber tree has an inherent high P acquisition efficiency.

5.13.5.3 Fine Root Production

A spatial variation in fine root production was observed in different treatments (Table 6). Fine root production was more in the treatment

which did not receive P fertilizers (T_1) between trees. However, more fine roots were produced in the P applied treatments between rows.

One of the most commonly observed effects of P deficiency on plant growth is increased root production (Forde and Lorenzo, 2001). Under conditions P deficiency shoot: root ratio decreased in many annual crops (Bosse and Koch 1998; Steingrobe *et al.*, 2001). In the present experiment also, more roots were produced in T_1 between trees, where fertilizers are not applied. Root area and root length were significantly higher in this treatment. This might be the adaptive mechanism of plants to enhance P uptake. However between rows, where fertilizers are applied, although there was no significant difference between treatments, more roots were produced in the P applied treatments. Root area was higher in between rows compared to that between trees in all the P applied treatments. Root length was also higher between rows in all the P applied treatments except T_2 which received the lowest level of P (@ $10 \text{ kg}^{-1} \text{ ha}^{-1}$). Similar results were reported in bean plants by Snapp *et al.* (1995). Roots of P stressed bean plants presented with a P-enriched soil branched more extensively within the soil patch than did roots of P-enriched plants. More root production in the P applied plots in between rows might be the plant adaptation to enhance uptake of fertilizers. Similar results were reported by Philip *et al.* (1996) in a five year old rubber plantation.

5.13.5.4 Root Density

Root length and area measured using a core sampler between trees and between rows were higher in P applied treatments, though not significant always (Table7). Between trees, all treatments were comparable both in terms of root area and root length. Root area between rows was also comparable between treatments, but root length was significantly lower in the control treatment. Results in conformity with this has been reported by Steingrobe *et al.* (2001) in winter barley. The

absolute increase in the standing root length due to P shortage observed in solution culture was not observed under field condition. Root mortality also influence root density measured at a time and it was observed that total root mortality of P deficient plants was higher (39 km m^{-2}) than that of the better-supplied plants (29 km m^{-2}). Fresh root production and root mortality was intensified at phosphate deficiency. During the period of highest root growth, the calculated advantage in P uptake by root renewal was highest and it increased at P shortage. This higher renewal and mortality might be the reason for the higher fine root production in T_1 between trees (Table 6) and no such difference in root density when measured with core sampler.

5.13.5.5 Length and density of root hairs

Root hairs were longer and denser in the treatment which was not supplied with P than in the P applied treatments (Table 8 and Plate 1a to 1e). P mobility in soil is governed by diffusion and hence its acquisition is dependent on the temporal and spatial exploration of the soil by the root system (Ma *et al.*, 2001).

The length and density of root hairs are extremely plastic in response to soil P availability. The plasticity of root hairs in response to P availability is local, occurring at the level of individual root hairs regardless of the P availability to the rest of the system (Bates and Lynch, 1996). According to Bates and Lynch, (2001), root hairs conferred competitive advantage to *Arabidopsis* plants in low P environments. Since P mobility is through diffusion, P uptake is limited by localized P depletion around the root. Root hairs extend the P depletion zone from the root epidermis thereby increasing the rate of P uptake and the total amount of P accessible by the root. The longer and denser root hairs observed in the present study might be an adaptation of trees to enhance the P uptake under low P conditions.

Phosphorus availability regulate root hair extension by changing the proton flux across the plasma membrane, which alters the intracellular pH gradients needed for tip growth and root hair density by changing the number of epidermal cell files that differentiate in to trichoblasts (Ma *et al.*, 2001). The regulation of root hair development by P availability is part of a suite of characteristics regulated by P that are related to P acquisition and the data indicate that in rubber also soil P content regulate root hair length and density. According to Rao *et al.* (1999), root hairs increase P acquisition by enlargement of root surface area and because the root hairs penetrate the soil perpendicular to the root axis giving access to a larger volume of soil per unit root length. Fohse *et al.* (1991) calculated that in soils low in P, contribution to P uptake by root hairs was up to 90 per cent of the total uptake.

In the present experiment, no significant difference was noticed between the length of root hairs between treatments which received P at 10, 20 and 30 kg ha⁻¹, indicating that the level of this adaptation is comparable in these treatments. Plants might be able to meet the P requirement at comparable levels at these different P levels. In the case of root density also, treatments which received P at 10 and 20 kg ha⁻¹ were comparable.

In mature rubber, root density was higher in P applied treatments, whereas fine root production was more between trees in the control treatment. The length and density of root hairs were also higher in control treatment. According to Forde and Lorenzo, (2001) primary root growth is much less sensitive to nutritional effects than growth of secondary or higher order roots. Hence though the effect of deficiency of P on root hair length and density are clear, its effect on standing root system often varies. Simple measurement of the relative biomass of the shoot and root fail to reveal the many subtleness of the root response to change in nutrient supply. Rubber roots explored a mean area of 89.38 m² during the

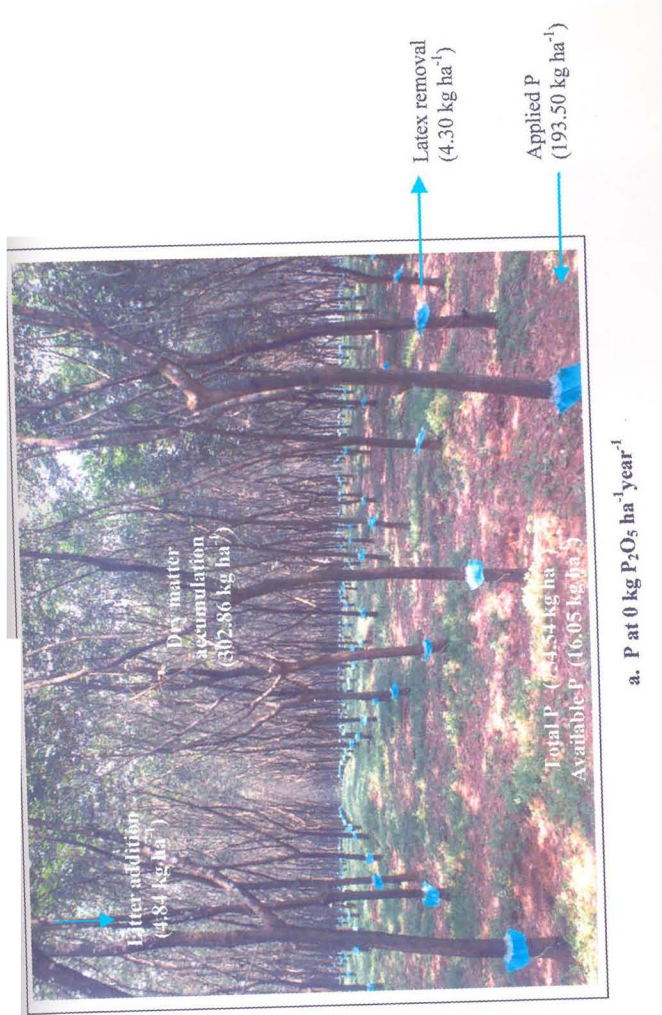
mature phase. Since the volume of soil explored by root for nutrients is high, a deficiency of P may affect the growth of higher order roots rather than the lower order roots.

Rubber tree has extensive root system and produce longer and denser root hairs under low P conditions. However, though extensive mycorrhizal infection was noticed, there was no significant difference between the percentage of infection in different treatments (Table 66). Hence root hairs might be playing more important role in P acquisition than mycorrhizal infection.

The extensive root system and the concentration of feeder roots in the surface layer indicate the inherent high P acquisition efficiency of mature rubber. Higher fine root production or root renewal and longer and denser root hairs observed in the treatment which did not receive any P fertilizer indicate the ability of trees to enhance their P acquisition through these adaptations when the soil P is low. The observations in this study, leaf P status (Table 11), latex P content (Table 42) and accumulation of P in the dry matter (Table 26a to Table 26e) also clearly indicate that trees are able to acquire sufficient P without external application of P.

5.14 INFLUENCE OF P LEVELS ON THE P BUDGET OF THE PLANTATION

P dynamics of the plantation and annual P budget for 2003-04 is presented in Table 71 and 72 and Fig. 12. The data on available P status of the soil at planting, dry matter accumulation during each year and cumulative recycling of P through litter and removal through latex were not available. When compared with the pre-treatment data, the decline in soil available P status in the control treatment which did not receive any P fertilizer was less than 15 per cent after six years. The total P content showed an increase T_1 also during 2003, and this might be due to the addition of P through litter. The total P and available P were higher in the



a. P at 0 kg P₂O₅ ha⁻¹ year⁻¹

Fig. 12 P budget of a mature rubber plantation

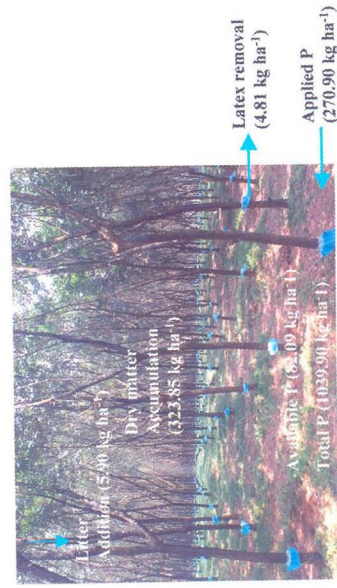
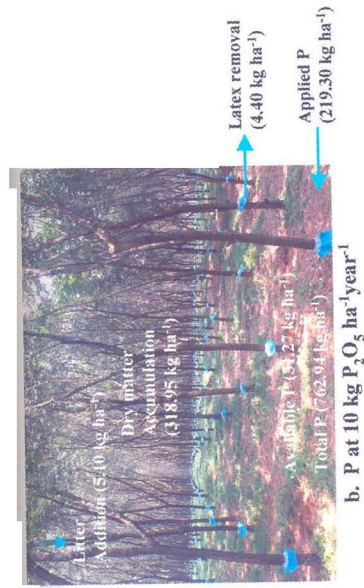
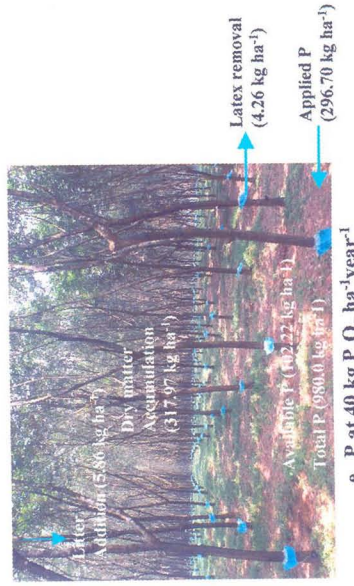
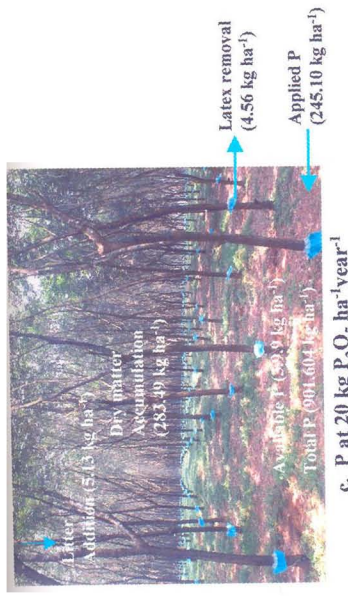


Fig. 12 Continued

P applied treatments. However the uptake of P was not influenced by the application of P fertilizers as indicated by the cumulative P accumulation in the dry matter which showed a variation of less than 13 per cent between treatments. The leaf P status which was comparable in different treatments also support this observation.

The data on annual P recycling through litter, addition through fertilizers and removal through latex was only considered for budgeting. The latex P content and latex yield was not influenced by application of P and hence P removal through latex was also comparable in different treatments. There was a net addition of $0.54 \text{ kg P ha}^{-1} \text{ year}^{-1}$ even in T_1 which did not receive any P fertilizer throughout the experimental period. This might be the reason for the lack of response of mature rubber to applied phosphorus.

Requirement of P is maximum during the active growing period due to the higher rate of growth. During this period, the soil area exploited by roots is also less and plants respond to application of P fertilizers. As the trees become older, the rate of growth decreases and there will be a levelling off of nutrients in the dry matter. The P from the decaying legume cover and leaf litter becomes available to plants. Several scientists have reported lack of response to P application during later immature and early mature phase (Watson, 1989; Sivanadyan *et al.*, 1995). In this experiment, plants in the T_1 ($@ 0 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$) were supplied with P fertilizers till the commencement of the experiment *i.e.*, during the immature and early tapping phase and application was avoided during the middle yielding phase. During this period also, trees are able to acquire sufficient P through its several adaptations as evidenced by leaf P, latex P and P content in the dry matter yield. On an annual basis, the P recycling through litter is slightly greater than the P removal through latex indicating the sustainability of the system in terms of P. Hence the data indicate the possibility of avoiding P application during this period. This

situation is likely to continue during the later phase also. However if a need is indicated through leaf status, P application can be resumed.

Information regarding the loss of P from the system through run off and leaching is lacking in the present experiment. The only loss considered was latex removal and hence further experimentation for detailed elaboration of the P-dynamics in a rubber ecosystem will be beneficial. However, the results of the present study emphasize the concept of a self-sustainable phosphorus cycle suggested by Sivanadyan *et al.* (1995) in a well managed rubber plantation which was fertilized during the active growing phase of the plant when the P requirement is maximum.

The price of rubber has been fluctuating in recent years. Reducing the cost of cultivation of rubber is of paramount importance for economic sustainability of the rubber plantation industry which is dominated by small holders with an average holding size of 0.49 ha. Of the total area of 5.67 lakh hectares in India, 71 per cent is under mature rubber and avoiding P application in 50 per cent area even for one year will lead to a saving of around Rs.90.23 million at the current price of rock phosphate.

As a non-renewable resource with relatively low concentrations in the biosphere, the use of fertilizer P inputs in any agricultural system must be carefully rationalised. Reducing the use of phosphatic fertilizers in the mature rubber sector will lead to considerable saving of P fertilizers which can be diverted for application at more appropriate growth stages and for more P demanding crops.

Most of the rubber plantations are raised on sloping or undulating terrains and even with proper soil conservation measures, traces of fertilizers are transported to water ways leading to eutrophication and ground water pollution. In the changing scenario of integrated nutrient management to need based application, avoiding excess application of any nutrient is a global norm and is ecologically sound and socially sustainable.

Summary

6. SUMMARY

A detailed investigation was carried out in an ongoing experiment of the Rubber Research Institute of India, Kottayam during 2002-2004 to explore the possibility of reducing the dose of phosphorus for mature rubber. The response of mature rubber to phosphorus, P dynamics in soil and plant, nutrient removal through latex, nutrient recycling through litter, microbial role in P nutrition and the processes by which trees improve their P acquisition were studied and the salient results are summarized below.

- The different growth characters studied, *viz.*, girth, girth increment, length of tapping panel and bark thickness were not influenced by application of phosphorus.
- The yield and dry rubber content of latex was not significantly influenced by application of phosphorus during the entire period of the study.
- Application of P did not influence the leaf P content significantly. The contents of nitrogen, potassium, calcium, iron, manganese, zinc and copper in the leaves were also not influenced.
- The leaf magnesium content was higher in the P applied treatments after fertilizer application and the difference was significant after the second fertilizer application.
- The content of P in fine roots was not influenced by application of P. The root micronutrient content was also not influenced by the different P levels.
- The root cation exchange capacity (CEC) was not influenced by application of P.

- The accumulation of different nutrients in the dry matter was not influenced by P levels. The partitioning of nutrients in different parts of the tree was also not affected. The total accumulation of P in the dry matter ranged from 755.98 g to 863.59 g.
- The content of different nutrients in the latex was not influenced by application of P. Mg/P ratio in the latex was also not influenced.
- Nutrient removal through latex was not influenced by the different treatments. The removal of P through latex varied from 4.26 to 4.81 kg ha⁻¹ year⁻¹.
- The litter addition was not appreciably influenced by P levels. The content of different nutrients in the litter was also not influenced by application of P. The addition of P through litter (4.84 kg ha⁻¹) was lower in the treatment which did not receive any P fertilizer. The recycling of other nutrients was not appreciably influenced by application of P.
- Application of P did not influence the saloid P, Fe-P, Al-P, Ca-P, Reductant P, occluded P and organic P content in the soil. Organic P constituted more than 50 per cent of the total P in the soil. The total P content in the treatment which did not receive any P fertilizer was comparable with those of treatments which received lower levels of P (@10 and 20 kg P₂O₅ ha⁻¹). The total P content in the treatments which received the higher levels of P (@30 and 40 kg P₂O₅ ha⁻¹) was higher compared to the control treatment. The soil available P content in all the P applied treatments was higher than that of the control treatment.
- Soil dehydrogenase activity (DHA) was not influenced by P application. Soil phosphatase activity was generally higher in the rhizosphere of control, and the difference was significant during the third sampling (before second fertilizer application).

- The root colonization by AMF was more than 85 per cent in most cases. However application of P did not inhibit or skipping P application did not promote root colonization.
- After fertilizer application, there was a decline in the population of phosphorus solubilizing bacteria (PSB) in the rhizosphere of P applied treatments. The percentage of PSB to total bacteria also showed a similar trend. The population of phosphorus solubilizing fungi, total fungi, total bacteria and actinomycetes in the rhizosphere was not influenced by application of P.
- Secretion of acid phosphatase from roots was highest from the control treatment which did not receive any P fertilizer. During the first and third sampling (before fertilizer application), the acid phosphatase secretion from the control was comparable with the treatment which received P at $10 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$ and greater compared to other treatments which received higher levels of P. During the second sampling (after first fertilizer application), the acid phosphatase secretion from all the P applied treatments was lower compared to the control treatment. During the fourth sampling (after second fertilizer application), the acid phosphatase secretion from all the treatments were at comparable levels.
- Phosphoenol pyruvate carboxylase (PEPC) activity was higher in the control treatment which did not receive any P fertilizer and the difference was significant during the second sampling (after second fertilizer application). Malate dehydrogenase activity (MDH) was higher in the control treatment during both samplings (before and after second fertilizer application).

- Rhizosphere pH was lower in the treatment which did not receive any P fertilizer and the difference was significant before fertilizer application. However, compared to non-rhizosphere soil pH, rhizosphere pH was slightly higher in most cases.
- Roots of mature rubber tree explored a large area of soil (mean-89.38 m²) and had a large volume (mean-0.047 m³). More than 60 per cent of fine roots of a mature rubber tree explored top 0-10 cm soil layer.
- Difference in spatial distribution in fine root production and distribution was observed between control and other treatments. Trees which were not supplied with P produced more fine roots than trees supplied with P. In the control, fine root production was more between trees whereas in the treatments which received P, more fine roots were produced between rows.
- Root length and root area were more in the P applied treatments. Longest root hairs (0.63 mm) were observed in the control which did not receive any P fertilizer. Density of root hairs was also highest in the control (67 mm⁻¹).
- The annual removal of P through latex is less than the annual addition of P through litter.
- No correlation was observed between soil available P content and leaf P status.
- The data indicate the possibility of skipping P application and thereby considerable saving of P fertilizers for mature rubber.

Future line of work

1. Multilocational experiments will give more information on the influence of pedogenic factors, soil nutrient status, terrain aspects and climatic conditions on the response of rubber to phosphorus application.
2. Clones may differ in their P acquisition ability and response to P and this may be studied.
3. The experiment may be continued to study the long term effect of skipping P application on growth and yield of trees and the data generated will be useful for refining the results of the present study.

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Appendices

APPENDIX - 1

Monthly rainfall (cm) during 1997-2004

Month	1997	1998	1999	2000	2001	2002	2003	2004	Mean
Jan	-	5.78	-	2.25	-	-	-	1.33	1.17
Feb	-	1.55	1.35	10.85	9.13	2.15	2.75	0.25	3.50
Mar	7.48	8.13	7.83	6.20	10.88	13.48	35.60	12.65	12.78
Apr	7.83	16.90	21.45	16.38	31.33	25.78	9.50	-	18.45
May	30.20	23.23	78.50	13.18	22.30	43.38	19.18	-	32.85
Jun	32.95	88.33	29.95	87.03	134.95	57.70	54.83	-	69.39
Jul	45.90	61.10	57.08	35.03	87.83	49.93	65.70	-	57.51
Aug	26.63	69.13	83.20	103.05	29.58	63.93	59.98	-	62.21
Sep	15.73	78.90	24.70	20.98	27.60	10.93	12.65	-	27.36
Oct	18.95	70.53	97.38	3.72	42.80	58.40	80.00	-	53.11
Nov	15.00	22.40	7.03	2.80	25.70	17.13	6.85	-	13.84
Dec	2.25	21.83	2.28	3.35	3.15	-	1.70	-	4.94

APPENDIX - II

Nutrient addition through fertilizers (kg ha⁻¹)

Year of planting	N	P ₂ O ₅	K ₂ O	MgO
1	20	20	8	3
2	40	40	16	6
3	50	50	20	7.5
4	40	40	16	6
5 and above	30	30	30	-

APPENDIX - III

 Between rows

 Between trees



Fertilizer application zone



Sites of root sample collection

PHOSPHORUS NUTRIOPERIODISM IN RUBBER

JESSY, M.D.

**Abstract of the
thesis submitted in partial fulfilment of the requirement
for the degree of**

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Kerala Agricultural University, Thrissur**

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ABSTRACT

Mature rubber tree does not respond to application of phosphorus fertilizers consistently. In majority of the earlier fertilizer experiments on mature rubber, there was no response to application of phosphorus fertilizers, in terms of growth and yield. Trees continued to maintain leaf P status without application of P even when the soil P status was low. An experiment was initiated at the Rubber Research Institute of India, Kottayam in 1997 in an area planted in 1984 to study the response of mature rubber to phosphorus application. From planting to till the commencement of the experiment all trees were given uniform quantities of nutrients. Treatments were imposed from 1998 onwards. Detailed investigation was carried out in this experiment during 2002-2004 to explore the possibility of reducing the present dose of P fertilizer ($30 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$) for mature rubber.

Phosphorus was applied at five levels (0,10,20,30 and 40 $\text{kg P}_2\text{O}_5 \text{ ha}^{-1} \text{ year}^{-1}$). Nitrogen and potassium were applied @ $30 \text{ kg ha}^{-1} \text{ year}^{-1}$ at uniform rates. None of the growth characters studied, viz., girth, girth increment, length of tapping panel and bark thickness were influenced by the application of P. The yield and dry rubber content were also not influenced by levels of applied P.

Trees which did not receive any P fertilizer continued to maintain leaf P status at comparable levels with that of the P applied treatments. Magnesium content of the leaves was generally higher in the P applied treatments after fertilizer application. Content of other nutrients (N, K, Ca, Fe, Mn, Zn and Cu) in the leaves was not affected by the different treatments. The accumulation of P and other nutrients in the dry matter was not influenced by application of P.

The content of P and other nutrients (N, K, Ca, Mg, Fe, Mn, Zn and Cu) in the latex and their annual removal were not influenced by the levels of applied P. The quantity of litter added and the litter nutrient content were also not affected by the different treatments. Removal of P through latex was slightly less than addition through litter. Before defoliation the litter P content was comparatively less in the treatment which did not receive any P fertilizer indicating high P utilization efficiency of trees.

Application of P did not influence the content of different P fractions (saloid P, Fe-P, Al-P, Ca-P, reductant P, occluded P and organic P) in soil. Organic P constituted more than 50 per cent of the total P in the soil. Total P content was significantly higher in the treatments which received higher levels of P. Soil available P status was higher in P applied treatments. In most cases, a decline in micronutrient content was observed in the treatments which received higher levels of P.

More than 85 per cent of the roots were colonised by arbuscular mycorrhizal fungi (AMF), but no relation was observed between the levels of applied P and root colonization. Population of phosphorus solubilizing bacteria (PSB) in the rhizosphere was suppressed after application of P, whereas the population of other microorganisms (total bacteria, phosphofungi, total fungi and actinomycetes) was not influenced.

The trees which were not supplied with P improved their P acquisition by several processes. Secretion of acid phosphatase, an enzyme which hydrolyzes organic P, was found to be higher from the roots of trees which did not receive P fertilizers. Soil phosphatase activity was also higher in the rhizosphere of trees which did not receive any P fertilizer.

Activities of phosphoenol pyruvate carboxylase (PEPC) and malate dehydrogenase (MDH), enzymes involved in the biosynthesis of organic acids were higher in the roots of trees which were not supplied with P fertilizers indicating the possibility of more exudation of organic anions and H^+ from these trees. Rhizosphere pH was generally lower in the

treatment which did not receive P fertilizers. Compared with non-rhizosphere soil pH, rhizosphere pH was comparatively higher in most cases.

Roots of mature rubber trees explored a large area of soil (89.38 m²). More than 60 per cent of the fine roots explored the surface 10 cm layer indicating that the trees have a high P acquisition efficiency. A spatial difference in fine root production and distribution was observed between treatments. Trees which were not supplied with P produced more fine roots than trees supplied with P. Root hair production was also higher in the control trees. All these adaptations contributed to a high P acquisition efficiency of rubber trees which were supplied with P during active growing phase.

The results of the present study indicate the self sustainability of phosphorus cycle in a well managed mature rubber plantation which was fertilized during the active growing phase of the plant suggesting the possibility of skipping P application in trees of that age group. This situation is likely to continue during the later phase also, however if a need is indicated through leaf P status, application of P can be resumed.