DISTRIBUTION, CHARACTERIZATION AND DYNAMICS OF SOIL ENZYMES IN SELECTED SOILS OF KERALA

by

B. APARNA

THESIS
submitted in partial fulfilment of the requirement
for the degree
DOCTOR OF PHILOSOPHY
Faculty of Agriculture
Kerala Agricultural University

Department of Soil Science & Agricultural Chemistry
College of Agriculture
Vellayani, Thiruvananthapuram

2000

DECLARATION

I hereby declare that this thesis entitled ***"Distribution, Characterization and Dynamics of Soil Enzymes in Selected Soils of Kerala"*** is a bonafied record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

Vellayani
21.06.2000

B. Aparna

B. Aparna

CERTIFICATE

Certified that this thesis entitled "***Distribution, Characterization and Dynamics of Soil Enzymes in Selected Soils of Kerala***" is a record of research work done independently by **Mrs. B. Aparna** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

Dr. P. RAJENDRAN
Chairman
Advisory Committee
Associate Professor
Department of Soil Science
& Agricultural Chemistry

Vellayani
21.06.2000

Approved by :

Chairman

Dr. P. Rajendran

Members

1. *Dr. V.K. Venugopal*
2. *Dr. P. Prabhakumari*
3. *Dr. P. Sivaprasad*
4. *Mrs. Brigit Joseph*

External Examiner :

19/07/2007

Dedicated to my dear daughter

Akshaya...

ACKNOWLEDGEMENT

I express my ineffable gratitude and heart felt thanks to Dr. P. Rajendran, Associate Professor, Department of Soil Science & Agricultural Chemistry, College of Agriculture, Vellayani Chairman of the Advisory Committee for the identification of the research project, adept and adroit guidance, constructive criticism and the encouragement evinced throughout the period of my investigation and in the preparation of the thesis. My telic pursuit of knowledge would have been futile but for the tireless, heuristic method offered by him whose wealth of information aided me in overcoming many obstacles I faced during the course of my research.

My fervent thanks to Dr. V.K. Venugopal, Head of the Department, Department of Soil Science & Agricultural Chemistry, for his encouragement, irrevocable and indefectible support rendered through out this research work.

I am fortunate to have Dr. P. Prabhakumari, Associate Professor, Department of Soil Science & Agricultural Chemistry, College of Agriculture, Vellayani as a member of my advisory committee and thankful for her succint ideas and guidance which were of timely help for my research.

I am grateful to Dr. P. Sivaprasad, Associate Professor, Department of Plant Pathology, College of Agriculture, Vellayani for his periodic suggestions at various stages as a member of my advisory committee.

My profound thanks to Mrs. Brijit Joseph, Assistant Professor, Department of Agricultural Statistics, College of Agriculture, Vellayani for her valuable help in the statistical interpretation of the data.

My sincere thanks to Mr. C.E. Ajith kumar, Programmer, Department of Agricultural Statistics, for his great help rendered during the statistical analysis of the data.

I take this opportunity to record my gratitude to Late Prof. Abdul Hameed, Department of Soil Science and Agricultural Chemistry, College of Agriculture, Vellayani for his timely help.

My acknowledgements to the Scientists and Staff of the Rice Research Station (RRS) - Kayamkulam, Regional Agricultural Research Station (RARS) - Pattambi, Central Experimental Station (CES) - Chethackal, Ranni and Coconut Research Station (CRS) - Balaramapuram for their immense help offered during the collection of soil samples.

I am beholden to all the Professors and Staff of the faculty of Department of Soil Science and Agricultural Chemistry for their support and encouragement.

I wish to declare my special thanks to Mr. Santhosh, Ms. Uma and Mr. Manu for their kind co-operation in the documentation work of the thesis.

I extend my heartfelt thanks to my friends Padma, Sreelatha

chechi, Anitha, Anju George, Lakshmi, Bindhu, Lakshmi and Byju Annan who unbiassed stood by me from the inception of my research to the terminus. I place my special thanks to Susan chechi for her moral support at times of need.

I sincerely thank the CSIR for granting me the scholarship for carrying out the research work.

Last but not the least, I am proud in mentioning my husband, parents-in-law, parents & brother whose ardent love, affection and magnanimous support sustained me from the start to the finish.

B. Aparna

B. APARNA

CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

INTRODUCTION

Nothing has underlined the common dilemma of all mankind so clearly as the environmental crisis now unfolding. Signs of stress on the ecosystem are everywhere today, reported daily in the press. The life support systems for man and other living things with which we share our earthly habitat are being threatened. This crisis affects everyone, rich and poor, old and young, Asian and American, Black and White, born and unborn (Lester Brown, 1972). One of the major life support systems of nature namely 'the soil' is the most affected natural resource in this process of ecological degeneration. Fuelled by the ever increasing pressure to produce more food and fibre to meet world demands from a limited rather shrinking land area, man's activity has resulted in soil degradation to an extent beyond restoration (Dick, 1994).

Understanding the underlying biological processes in tandem with the identification of indicators of the above ecosystem stress is inevitable to provide strategies and approaches to promote long term sustainability of the fragile eco-system. All members of the food chain are dependant on soil as a source of nutrients, support and sustenance besides their reliance on soil as a

sink for terrestial wastes. In this context, soil enzymes play an active role in the microbial ecology and transformations brought about by catalysing innumerable reactions in soils. The activity of soil enzymes thus hold potential as soil quality indicators though they are sensitive to temporal changes due to environmental and management factors.

The long term effects of soil management techniques such as crop rotation, liming, fertilization and pesticide applications on the biological regimes need detailed investigations. No systematic study has been hitherto attempted to assess the distribution and dynamics of enzymes in the soils of Kerala. Hence, work done in this direction is scanty and meagre. The present study will add to the theoretical corpus of knowledge of soil enzymology. The present day concept of organic agriculture leading to yield sustainability in the long run has to be theorised as the nutrient removal through economic produce always exceeds nutrient inputs. In this context, the role of microbially mediated enzyme systems to release plant nutrients from the soils needs elaboration.

The superiority of treatments under permanent manurial experiments receiving continuous application of organics over treatments receiving continuous application of chemical fertilizers are thought to be partially due to enhanced enzyme activity. The dynamics of major soil enzymes in permanent manurial experiments have not been studied in detail so far. Relationship between the electro chemical properties of soils and soil enzyme activity also needs further investigation.

The effect of agrochemicals and organic amendments on soil enzyme activity and microbial load has not been scientifically investigated in the soils of Kerala. As we are using a wide range of insecticides, fungicides, herbicides, organic manures and chemical fertilizers in soils, it is imperative that the effect of these chemicals on biological activity of the major soils need be studied.

In this context, the present study was undertaken to have a deeper insight into the various aspects of enzymology in selected soils of Kerala pertaining to different agro climatic regions and cropping systems. With the above theoretical background in view, the following major objectives were set for the thesis programme.

- To study in detail the activity of five major soil enzymes viz., urease, phosphatase, dehydrogenase, protease and cellulase.
- To study the dynamics of the above enzymes in permanent manurial experiments in selected agro climatic or cropping situations of Kerala.
- To study the effect of selected agrochemicals viz., insecticides, fungicides, herbicides and antibiotics and soil amendments on the activity of these enzymes.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Nutrient cycling in soils involve biochemical, chemical and physico chemical reactions with the biochemical processes mediated by plant roots, microorganisms and soil animals. It is well known that all biochemical reactions are catalysed by enzymes, which are proteins specialized to perform biological reactions. With the advent of plant and animal biochemistry, it was recognised that many reactions involving soil organic matter transformations may be catalysed by enzymes existing outside the microorganisms and plant root systems.

These enzymes are among the most remarkable biomolecules known because of their extraordinary specificity and catalytic power, which are far greater than the man made catalysts. With the increased extensive use of agricultural chemicals and the appearance of industrial pollutants including pesticides, considerable interest has been raised about research on the interactions of these chemicals in soil and with the soil enzymes. The current state of these studies on extracellular abiontic soil enzymes shows that this field of enquiry is on a sure scientific footing and as it approaches maturity, it will provide us with a more thorough understanding of the total biological and biochemical processes of soils.

Hence in this chapter, an attempt is made to review the work already done in the field of soil enzymology by consolidating the work carried out on five major soil enzymes with respect to their sources, activity, range and also the effect of soil amendments and agro chemicals on their performance to reveal the ecological significance.

2.1. Origin of enzymes in soil

It is accepted that the enzymes in soil originate from plant, animal and microbial sources. However, their presence in soils as derived directly and specifically from plant and animal sources, has yet to be demonstrated conclusively. An array of work have been undertaken to envisage the actual environment where the enzymes are synthesised or the sources of enzymes in soil and plant systems. The following section provides a brief review of the same.

Soil enzymes which catalyse reactions in soils that are important in the transformation of nutrients such as C, N, P and S are primarily of microbial origin but may also originate from plant and animal residues (Kiss, *et al.* 1975). Skujins (1976) introduced the term "abiontic" to describe all enzymes designated as accumulated enzymes plus those secreted by proliferating cells during normal growth. Soil enzymes also form a part of soil matrix as exoenzymes and endoenzymes in viable cells, whose activities are commonly correlated with microbial parameters as reported by Casida (1977), Hersman and Temple (1979).

Burns (1982) hypothesised the presence of ten different categories of enzymes according to their location in soil; enzymes associated with proliferating cells were subdivided into cytoplasmic, periplasmic and those attached to outer surface of cells; enzymes in non-proliferating cells; truly extracellular enzymes; enzymes attached to dead cells or cell debris; enzymes leaked from extant cells or lysed cells; enzymes associated temporarily in enzyme - substrate complexes and enzymes adsorbed to clay minerals or with humic colloids. Soils are also reported to contain a wide variety of active cell free enzymes which display remarkable stability (Stevenson, 1982).

Tabatabai (1982) reported that the soil enzymes mediate various biochemical processes which are derived from micro organisms, plant roots and soil animals. The most serious problem in the interpretation of measured enzyme activities of soil is to decide the contribution of enzymes according to their state of existence in soil (Ladd, 1985).

Soil enzymes play an important role in the mineralization process and also many other soil biological reactions which are truly microbial in origin (Tate, 1987). Van Huystee and Cairns (1992) reported that the enzymes occur in many isoforms and are involved in processes such as differentiation and growth. Nannipieri (1994) reported that enzymes have different locations in soil with various biotic and abiontic components.

All other evidence relating to the origin of soil enzymes in soil is indirect. Such evidence rests partly on enzymatic activities either of soil under fallow or different vegetations or homogenates of fresh plants, plant exudates or plant debris separated from soils or of specific micro organisms isolated from soils (Dick *et al.* 1994).

Nannipieri (1997) further concluded that a certain level of extra cellular enzyme activity exist in all soils and the enzymes secreted by living cells during normal cell activity, actively leaked from extant cells and/or released from lysed cells are short lived.

2.2. Soil enzyme activity and soil fertility

Historically, chemical and physical properties of soil have been used as crude measures of soil productivity. However there is growing evidence that soil biological parameters may hold potential as early and sensitive indicators of soil productivity or soil ecological stress or restoration.

The gross physical, chemical and biological characteristics of a soil give a severely distorted picture of the soil micro habitat and hence could not act as indicators of fertility.

It is also evident that the enzymes are substrate specific and individual measurements cannot reflect the total nutrient status of the soil (Howard, 1972). Several measurements have emerged as important parameters of the general biological activity in soil, especially dehydrogenase activity, respiration rates and ATP measurements.

Skujins (1978) reported that the activities of urease, phosphatase, protease, invertase and catalase can be used as fertility indices of soil to complement soil chemical analyses so as to assess nutrient availability and crop yield. Mc Gill *et al.* (1986) found that the soil microbial biomass is a source and sink for plant nutrients and an active participant in nutrient recycling thus serving as a fertility factor.

Powlson *et al.* (1987) suggested that changes in amounts of microbial biomass can be used as an early indicator of changes in total soil organic matter content. Verstraete and Voets (1987) reported a lack of relationship between enzyme activity, microbial activity and soil fertility. Similarly, Nannipieri *et al.* (1990) failed to find out any correlation between soil enzyme activity and crop productivity in a given soil.

The use of microbial biomass, dehydrogenase and alkaline phosphatase activity to obtain a more complete and precise definition of soil fertility was suggested by Beyer *et al.* (1992). Soil enzyme activities are sensitive indicators of management induced changes in soil properties, although in most instances their use has been limited to sites at which comparisons can be made between management practices or within a chrono or topo sequence (Dick 1992).

Other enzyme activities reported to be correlated with biotic factors in soil include : alkaline phosphatase, amidase and catalase which showed a close relationship with microbial respiration and biomass (Asmer *et al.* 1992). According to Dick (1994), the importance of soil enzyme activities as a potential indicator of soil quality mainly depends on soil sample pretreatment, assay procedures, units of measurements and the standardization of these factors to give the most accurate soil quality enzyme assay.

Jordan *et al.* (1994) suggested certain microbial methods as potential indicators of soil quality in the surface soils under short and long term cropping systems in Central Missouri which included soil microbial biomass C, N and fungal biomass. Acton (1994) suggested the use of enzyme activities for the validation of changes in quality predicted by simulation models from existing

data base or to verify the effectiveness of management practices in sustainable crop production systems.

Soil microbial carbon, phospholipid analysis, direct counts of fungal and bacterial biomass and soil enzymes (phosphatases) also serve as potential indicators of soil quality according to Jordan *et al.* (1995). These soil microbial biomass C and enzyme assays seemed to be better indicators of cropping histories than the other methods tested in the long term plots as suggested by Jordan *et al.* (1995).

Kennedy and Papendick (1995) also considered soil enzyme activities as one of the numerous measurements of soil microbial quality due to the easy measurement of activity with reaction rate indicating the amount of enzymes present. Park and Seaton (1996) also reported that enzyme activities can be used as indicators or key variables for soil assessments related to sustainability.

Monreal *et al.* (1998) reported that elevated enzyme activities appear to be associated with conditions promoting microbial synthesis of enzymes and such sensitivity would make soil enzyme activities as effective indicators of changes in soil quality. Further, he added that the soil quality could be defined in terms of unique ecosystem functions performed by soils, one of which is cycling of nutrients and C by assessing the enzyme activities.

2.3. Range of enzymes in the soil

A wide range of enzymes have been reported in both plant and soil systems. Soil enzymes most frequently studied are oxidoreductases and

hydrolases. Several investigations of transferase and lyase activities have been reported in soil but isomerase and lipase activities were not. Numerous work have been carried out to characterize and identify these bio catalysts in the past few decades contributing to the corpus of soil enzymology.

Soil enzymes most frequently studied are oxidoreductases and hydrolases. Thus studies on dehydrogenase, catalase, invertase, protease, phosphatase and urease activities account for most of the publications on soil enzymes (Sato, 1981). Dragan-Bularda and Kiss (1982) demonstrated dextran synthesis in a soil incubated with sucrose for several days in the presence of toluene.

The activity of the enzyme invertase increased with increased grazing activity and maximum variability for the enzyme occurred in the A_0 horizon of the matgrass - forbs pasture according to Mariskevich (1992).

Studies on nitrogenase enzyme by Roper *et al.* (1994) identified a reduction in nitrogenase activity by the addition of N fertilizer while cultivation encouraged the activity compared with zero tillage treatments. A strong relationship between aryl sulphatase activity and organic carbon content was observed for soils with differing cultivation histories as per the reports of Farrell *et al.* (1994).

Song *et al.* (1994) studied the enzyme status of the soil infected by *Tylenchulus semipenetrans* and identified that various enzymes such as invertase, amidase, urease, phosphatase (acid), and protease showed decreasing trend. Studies on the effect of effluents by Palaniswami and Ramulu (1995)

observed that the activity of a range of enzymes occurring in soils viz., invertase, catalase and peroxidase was declined to very low levels due to effect of continuous flow of effluents.

The soil aryl sulfatase exhibited a typical Michaelis - Menten Kinetics and variations in the kinetic parameters were considered to reflect changes in the enzyme activity brought about by the different land management schemes according to Farrell *et al.* (1994). Humic compounds with B-glucosidase activity were extracted from soils using tetra sodium pyrophosphate as extracting solution by Busto and Perezmateos (1995).

2.4. Persistence of soil enzymes

The persistence and stability of enzymes in soils are generally attributed to their association with clays and humus. Soil enzymes can be stabilized in many ways : absorbed on to internal or external clay surfaces; complexed with humic colloids by adsorption and cross linking, micro encapsulation, ion exchange entrapment and co-polymerization. (Mortland 1970).

Martin and Haider (1971) reported that many phenolic constituents of humus are able to auto oxidise and respond to enzyme or inorganic catalysts forming free radicals and quinones. Clay and complex organic hetero condensates are known to bind the soil enzymes thus immobilizing them according to the findings of Verma *et al.* (1975).

According to Perezmateos and Gonzales Cercedo (1985), the highest percentages of over all catalase, dehydrogenase and urease activities of soil

were observed in particles with diameter lower than 50 μm, characterized by the presence of clay and humic molecules.

The clay enzyme complexes are probably the most interesting class of clay organic complexes according to Cortez (1989). Khaziyev and Gulko (1990) reported that peroxidase enzyme extracted from horse radish was competitively inhibited by humic acids from a chernozem and grey forest soil.

Clay-enzyme interaction according to Boyd and Mortland (1990) is a very complex process which depends on clay content, moisture, nature of the exchangeable cations, nature and surface area of clay, pH of the bulk phase and pH of the clay-water interface. In some cases, the enzymes have been partly purified and shown to be complexed with soil humic material.

Martens *et al.* (1992) observed higher activity of soil enzymes due to enhanced protection and survival of abiontic enzymes in humic complexes of these soils. The enzyme adsorption by homoionic smectite generally decreased in the order monovalent > divalent > trivalent saturating or exchangeable cations (Gianfreda *et al.* 1992).

Tabatabai and Fu (1992) reviewed the types of the extracted free enzyme from enzyme-humus complexes, the procedures employed in their extraction and potential problems encountered in their separation and purification. Lahdesmaki and Piispanen (1992) reported the importance clay and humic colloids in the stability of abiontic enzymes to thermal stress such as heating and repeated freezing-thawing cycles.

The general acceptance that soil enzymes may be of plant, animal and

microbial origin was justified by Foster and Doormar (1993) who observed the occurrence of acid phosphatase in rhizodermal and cap cells of roots, soil fungi, bacteria, in mucilagine covering roots as well as in microbial membranes of soils. Ladd *et al.* (1993) suggested electron microscopic and cytochemical methods to study the stability of humus-enzyme complexes in soil.

The pH dependence of adsorption of some enzymes on montmorillonite, kaolinite, geothite and talc was studied by Quiquampoix *et al.* (1993). The stability and resistance of humus enzyme complex were also investigated by many authors.

Serban and Nissembaum (1996) observed that peroxidase and catalase-humic acid complexes were resistant to thermal and proteolytic denaturation than the free enzyme. The free enzymes in soils are short lived and the use of synthetic polyphenol complexes for stabilization was suggested by Nannipieri and Bollag (1997). Leonowiez and Bollag (1997) observed that extractants containing citrate ions and manganese formed complexes exhibiting laccase-like activity during the extraction.

2.5. Ecological significance of soil enzymes

Soil enzymes are produced by living organisms and hence it is obvious that any action altering the life functions of soil organisms could indirectly affect soil enzyme activities. The activities of various enzymes and micro organisms have been used as indices of soil pollution and degradation by the ecotoxicologists and environmental scientists. Thus the peripheral areas

of soil enzyme research involves exobiology and forensic science (Burns, 1982).

Babich and Stotzky (1983) made an attempt to quantify the effect of pollutants on the enzyme activity (Dehydrogenase) represented by Ecological Dose (EcD). A more comprehensive parameter, "Enzyme Activity Number" (EAN) was developed taking into consideration the activities of several enzymes by Beck (1984).

Schuster and Schroder (1990) reported that sequential application of chemicals or xenobiotics could exhibit stronger side effects testimonised by the behaviour of microbial biomass and dehyrogenase activity. Doelman and Haastra (1986) developed enzyme (urease) activity index to assess the short and long term effects of heavy metals.

Sinsabaugh *et al.* (1992) developed a single factor termed lignocellulase activity by cataloguing five cumulative enzymes, which can be used to assess the toxicity of chemicals in the ecosystem. Bardgett *et al.* (1994) developed an index which allows the quantification of the toxic effects of a chemical by integrating the values obtained from the measurements of parameters like dehydrogenase activity, microbial load and CO_2 evolution rate.

Doran and Parkin (1995) defined soil quality as the capacity of soil to function within ecosystem boundaries to sustain biological productivity, maintain environmental quality and promote plant and animal health.

Brookes (1995) reported that microbial biomass is a sensitive parameter and can be used as an early warning of changes in ecosystem before they are

detectable in other ways. Monreal *et al.* (1998) proposed that soil quality can be defined in terms of unique ecosystem functions performed by the soil, one of which is the cycling of nutrients and C.

2.6. Spatial variability of soil enzymes

Soil enzyme activities are sensitive indicators of management induced changes in soil quality. Soil enzyme assays have been used to monitor microbial activity related to specific macronutrient transformations.

In the assessment of soil quality and studies of macro nutrient cycling, the spatial variability of measured soil properties must be accounted for at a scale that adequately represents sites or treatments (Groffman and Tiedje, 1991).

Brameley and White (1991) suggested that depth of sampling serves as an important factor for the measurement of spatial dependence and observed that dehydrogenase and B-glucosidase activity decreases with depth.

For studying the spatial variability, geostatistical methods can be used to compare soil enzyme activities between heterogenous sites and to study relationship between soil enzyme activities and ecological processes at the land scape level as suggested by Janzen *et al.* (1992). Similarly to examine the spatial dependence of soil enzymes, geostatistical methods were found to be the best whereas conventional statistical approaches have yielded little information of this kind as opined by Burton & Mc Gill (1992).

Rossi *et al.* (1992) also developed certain geostatistical methods to assess spatial dependence of soil enzymes in ecology. Parkin (1993) also found

out geostatistical techniques as one of the several techniques for analyzing spatial and temporal heterogenity of field soils while semi variogram method was used by Cambardella *et al.* (1994) to compare range and degree of spatial variability.

Gonzalez and Zak (1994) measured the spatial auto correlation of soil properties in secondary tropical dry forests and compared to the spatial variability of soil enzyme activities. The studies on spatial patterns of enzyme activities and other soil properties along the slope to identify casual relationship at this scale was carried out by Pickett and Cadenasso (1995).

Several factors like water content, organic carbon content and the thickness of the Ap horizon were taken into consideration for the measurement of spatial dependence of enzymes as suggested by Zhang *et al.* (1995).

2.7. Urease

Urease, the enzyme that catalyzes the hydrolysis of urea to CO_2 and NH_3 is widely distributed in nature and has been detected in plants, animals and micro organisms. Variations in urease activities within and between soil groups have been related to soil properties, type of vegetation and cultural practices.

2.7.1. Sources of urease

Bremner and Mulvaney (1978) reported that urease is unique among soil enzymes and the enzyme extracted from the soil as an enzyme-humus conjugate represents 20-40% of total activity in soil.

The studies conducted by Rao and Ghai (1989) involving the isolation of urease from different systems revealed that the activity of urease was higher in the terrace and valley soils than on the dryland agriculture with a positive correlation to the soil organic C content (r = 0.92**).

The urease is found both in micro organisms and plants and also as an immobilized extra cellular enzyme, which is extracted from soil bound to organic and inorganic soil components (Mobley and Hausinger, 1989).

Urease isolated from plants or microorganisms was rapidly decomposed by microorganisms and by proteolytic enzymes as suggested by Ciurli *et al.* (1996) who extracted urease from soil bacterium *Bacillus pasteurrii* and estimated the activity in terms of Michaelis - Menten Kinetic parameters, V_{max} and K_m, the observed values being V_{max} = 1960 ± 250 units ml^{-1} and K_m = 235 ± 20 units ml^{-1} for free urease.

Thus *Bacillus pasteurri*, a widespread soil bacterium play a significant role in nutrient cycling since it synthesise a large amount of urease (Bennini *et al.* 1996). Studies on urease activity by Dinesh *et al.* (1998) also identified urease activity in forest soils and reported that the activity ranged in the order, 245.60 to 290.80 ppm of urea hydrolysed g^{-1} of soil 24 h^{-1}, in the mangroove forests of Andamans.

2.7.2. Assay methods

Numerous methods have been used for the assay of urease activity in soils. Most of these methods involved the estimation of ammonium released on incubation of toluene treated soil with buffered urea solution.

Skujins and Mc Laren (1969) suggested a method for activity assay involving estimation of the urea decomposed or the CO_2 released on incubating the soil with urea.

Douglas and Bremner (1971) suggested a method of urease activity assay by the colorimetric determination of urea hydrolysed on incubation of soil samples at 37^0 C for 5 hrs.

According to Tabatabai and Bremner (1972), the buffer method of urease activity assay involves the determination of ammonium released on incubation with THAM (Tris-H_2SO_4) buffer (pH 9.0), urea and toluene at 37^0 C for 2h, the ammonium released being determined by incubating the soil sample with 2.5 M KCl and by steam distilling an aliquot of the resulting soil suspension.

The possibility that radiation sterilization may be useful for assay of urease activity in soil and research on soil urease suggested by the observation that a californian soil exhibited urease activity after sterilization by an electron beam (Thente, 1970).

Douglas and Bremner (1980) suggested a colorimetric method of determination of urease involving the reaction of urea with p-dimethyl amino benzaldehyde or with diacetylmonoxime (DAM) or thiosemicarbazide (TSC).

O' Toole (1991) studied the ability of the TRIS buffer at 0.05 M and pH 9 to block NH_4 fixation and thus estimated the buffered urease activity in a soil with high ammonium fixing capacity.

2.7.3. Factors affecting urease activity

Numerous factors like pH, organic carbon, available nitrogen, depth of sampling, temperature and substrate concentration were found to influence the activity of urease. The activity may be either accelerated or retarded due to the influence of these factors. Studies relating to the effect of soil properties on the level of urease activity have indicated that the activity tends to increase with organic matter content and that sandy or calcareous soils tend to have a lower activity than heavy textured soils (Silva and Perera, 1971).

Kandeler and Gerber (1988) observed that the activity of urease increases with increase in moisture content of the soil and vice versa and it is difficult to account for concerning the positive effect of water level on urease activity.

The inoculation of Blue Green Algae (BGA) was found to increase activity of urease along with significant improvement in soil aggregation according to Rao and Burns (1990) through their studies on the effect of biofertilizers on urease activity.

The inactivation of urease is a consequence of the oxidation of the essential cysteine residue present in the active site of the enzyme according to Medina and Muller (1990). Tabatabai (1992) after studying the urease immobilized on surface-modified clays reported that there is an optimum pH for enzyme activity which does not shift upon enzyme immobilization.

The depth of sampling is considered to be an important factor affecting the urease activity. According to Fenn *et al.* (1992) a decrease in the urease

activity with increasing profile depth and greatest decrease could be noticed below the plough depth. Zelles *et al.* (1992) opined that activity of urease existing as extra cellular enzyme in part, may increase with organic carbon content because of the stabilization of the existing enzymes by adsorption on to soil organic matter.

Jha (1992) reported that in north-eastern India, the activities of urease was found to be higher in degraded forest soils depending upon the differences in the fungal and bacterial population between them.

One approach to reduce the problems associated with the urea as a fertilizer is to use compounds that will retard urea hydrolysis, there by decreasing the activity of urease. Joseph and Prasad (1993) found out that dicyandiamide and neemcake coated urea inhibited the activity of urease by reducing the hydrolysis of urea-derived NH_4^+.

The urease activity also depends on the quantum of the substrate available. Torello and Wehner (1993) found that higher amounts of NH_3 liberation from moist turf manured with urea resulted in lesser activity of urease.

Studies on the kinetic parameters of urease by Lai and Tabatabai (1994) revealed that the kinetic parameters of urease would not change upon binding of the urease enzyme to a clay-organic system.

Since soils contain urease producing microorganisms, it is not surprising to observe that the urease activity can be increased by the addition of organic amendments. Thus, greater urease activity in a young municipal solid

waste compost than with an older one was observed by Serra Wittling *et al.* (1995).

The activity of urease can also be correlated with the soil physical properties according to Sequi *et al.* (1995) who observed a significant correlation between urease activity and the soil pores in the 30 - 200 mm range in six non tilled and six conventionally tilled soils.

The activity of urease was greater in the surface than in the lower Ap horizon, rich in organic carbon as reported by Park and Seaton (1996).

The higher levels of urease activity found in forest and agricultural soils were due to the decomposition of lysed microbial cells following the winter freeze as reported by Vancleve *et al.* (1997).

2.8. Phosphatase

The element phosphorus is essential for plant growth and metabolism and they have, therefore received a great deal of study by agronomists and soil scientists throughout the world. The transformations of these elements in soil systems are not well understood, but it is generally accepted that they are taken up by plant roots as inorganic phosphate. Since a large proportion of P in many soils is organically bound, the mineralization of these organic fractions is of major agricultural and economic importance. Soil phosphatase enzymes have been accorded a major role in the mineralization process namely the catalysis of hydrolytic cleavage of ester phosphate bonds. The term "phosphatases" is used to describe a group of enzymes that hydrolyses the

esters of phosphates and anhydrides to orthophosphoric acid. Phosphatases thus perform an important function in soil by transforming organic P to inorganic phosphate (Skujins *et al.* 1962).

2.8.1. Assay methods

It is probably true that many contradictions have arisen from studies on soil phosphatases due to the differences in the treatment of soils before and during incubation and to the diversity of extraction and assay methods employed.

Skujins *et al.* (1962) suggested the use of Modified Universal Buffer (MUB) at pH 6.5 and pH 11.0 for acid and alkaline phosphatases respectively. The estimation of phosphatase activity using Na-B-nathyl phosphate was recommended by Ramirez Martinez and Mc Laren (1966).

The humus - phosphatase enzyme complex can be extracted following the procedure adopted by Danneberg (1973) and Danneberg and Schaffer (1974).

The determination of p-nitrophenyl phosphate released after incubation at 37^0 C can be used for the estimation of phospho monoesterase and phospho diesterase activity as suggested by Eivazi and Tabatabai (1977).

Water extraction of phosphatase at 22^0 C was carried out to investigate alkaline phosphatase present in aqueous phase as per the procedure suggested by Rhoades (1982). Malcom (1983) observed that soil phosphatase activities vary particularly with respect to pH, substrate concentration and pre treatment of the soil.

Schinner *et al.* (1993) suggested some modification on the procedures of phosphatase assay suggested by Eivazi and Tabatabai (1977) where by samples were centrifuged at 10,000 rpm for 10 min^{-}.

2.8.2. Sources of phosphatase

The sources of phosphatases may be due to the production of this enzyme by bacteria, fungi, yeasts, protozoa, mycorrhiza of fungi and plant roots as opined by Torrani (1968).

Phosphatase activity was first detected by Ischii and Hayano (1974) using a synthetic substrate. The role of phosphatase in living systems and in particular in plant nutrition and physiology was outlined by Halstead and Mc Kercher (1975).

Appiah and Thomson (1974) reported that this enzyme responsible for the mineralization of phosphatase, catalysed by root exocellular phosphatase, may be liberated as root exocellular enzyme or as "bound enzymes" in conjunction with root epidermal cells.

Brownman and Tabatabai (1978) reported the activity of phosphatase in various plants, soils and micro organisms. According to Parent and Mackenzie (1985), the phosphatase enzyme systems may be bound on to humus and/or clay mineral colloids, which may make its extraction more difficult.

Acid phosphatase was also observed in rhizodermal and root cap cells, in soil fungi and bacteria, mucilage covering roots, mucilage fragments and microbial membranes present in soil (Foster and Dormaar, 1991).

2.8.3. Factors influencing activity

Several factors like temperature, pH, fertilizers and organic matter were found to influence the activity of phosphatase.

Parks (1974) found that the bacteria E. *coli* proliferates at 55^0 C for 30 minutes of incubation, which is responsible for the higher synthesis of phosphatase.

Harrison (1983) suggested a positive relationship between phosphatase and organic matter content since the enzyme was seen bound to humic-protein complex. The environmental factors such as pH, water, surface charges and redox conditions at microsite level are of crucial importance for the activity of various forms of phosphatase (Nannipieri, 1984).

Correlation between phosphatase activity and soil texture was observed by many authors. Mc Laren (1985) observed a negative correlation with clay and silt content of soils and the phosphatase level.

Ladd (1985) reported high levels of phosphatase activity in the rhizosphere which can be attributed to high microbial activity promoted by plant residues. The exact pH optimum for phosphomonoesterase activity depends on the pH value of the soil horizon from which the samples have been according to opined by Trasar cepeda and Gilstores (1988).

Tarafdar and Junk (1988) observed that acid and alkaline phosphatases were influenced by corn plants with the enzyme activity in the rhizosphere being greatest near the root surface. This observation was also supported by Haussling and Marschnner (1989) who reported higher activity of phosphatase

in the rhizosphere than in the bulk soils. Partial inhibition of alkaline phospho monoesterase was due to the possible sorbtion of the enzyme on humic substances as reported by Kandeler (1990).

Speir and Cowling (1991) investigated the phosphatase activities of pasture plants and soils, its relationship with plant productivity, phosphorus fertility indices and reported the negative effect of P fertilization on the activity of phosphatase.

Studies on the spatial variability of phosphatase in a grass land soil was observed to have a value of CV = 36% which was close to the analytical variability (CV = 37%) according to Bonmati *et al.* (1991). Collins *et al.* (1992) reported greater activity of acid phosphatase with abandoned agricultural soils that had been enriched with P fertilizers.

Among several hydrolases present in soil acid phosphatase is one of the most ubiquitous, being produced by plant roots, bacteria and fungi. (Fox and Comerford, 1992).

The activity of phosphatase varied under different soils according to Adams (1992) and values as low as 3 mg of p-nitrophenyl phosphate in an alluvial soil was noticed.

Studies on kinetic parameters by Gianfreda and Bollag (1994) reported higher km values for acid phosphatases in several investigations with humic acids or other organic soil constituents.

A positive correlation between phosphatase activity and organic matter content was noticed by Chhonkar and Tarafdar (1994). The factors such as

plant age and soil moisture affected the usefulness of acid phosphatase in predicting grain yield and plant P status (Mc Lachlan, 1994).

Margesin and Schinner (1994) studied phosphomonoesterase, phosphodiesterase, phosphotriesterase and inorganic pyro phosphatase activities in forest soils and the effect of pH on enzyme activity and extractability.

Studies on amendment addition by Cooper and Warman (1997) revealed that the application of chicken manure compost significantly increased the phosphatase activity in a low organic matter silty clay soil but had no effect on a sandy loam soil.

Jose *et al.* (1997) found out that factors like organic matter and moisture served to be important in determining the distribution of phosphatase in moderately well and some what poorly drained soils. Increase in alkaline phosphatase activity with the addition of ammonium citrate was observed by Gangnon *et al.* (1997).

A highly significant and positive correlation between phosphatase activity and wheat yield was also observed by the same author. The high phosphatase activities in the surface horizons might induce a greater rate of hydrolysis of applied pyrophosphate (polyphosphate) fertilizers to orthophosphate and leads to undurable fixation of P according to Baligar *et al.* (1997).

Rogar *et al.* (1998) reported that the activity of the alkaline phosphatase enzyme increased due to the application of compost compared to the application

of ammonium nitrate alone or unfertilized. Baligar *et al.* (1998) observed a decline in phosphatase activity with increasing sampling depth which was attributed to the low organic matter content.

2.9. Protease

Protease represents a group of hydrolytic enzymes that hydrolyse the dipeptide derivatives of organic amino compounds and plays a significant role in the mineralization of N, thus making it available to the plants. Proteases are thus widely distributed among soils showing a wide range of activities and properties (Mayoudan *et al.*, 1975).

2.9.1. Assay methods

The best criterion for adopting a particular method of protease measurement and extraction is to find the degree of correlation obtained between the protease activity as they occur under natural conditions and laboratory conditions. The activity of proteases are usually evaluated in terms of micromoles of N hydrolysed from the soil per unit weight of soil.

Assays may be of short duration (1 - 2 hours) in the presence of toluene to minimize microbial growth and utilization of protein hydrolytic products (Voets and Dedeken, 1964). Voets and Dedeken (1965) suggested a method of protease activity assay by using toluene as sterilizing agent.

Ambroz (1966) suggested a method of protease activity assay using ovalbumin and casein as substrate for the hydrolysis and thus the initiation of protease activity.

Activities of proteases are assayed based on the release of amino compounds on the formation of colored products (Macura and Vagernova, 1969).

Ambroz (1970) suggested a method of protease activity assay which included the estimation of activity using benzoxy carbonyl phenyl alanine leucine. The same author also observed that air drying the soil resulted in considerable loss of protease activity.

The most widely adopted method of protease activity assay included the use of benzyloxy carbonyl phenyl alanine leucine as substrate (Ladd and Butler, 1972).

2.9.2. Factors influencing activity

Soil protease depends upon the various factors like pH, temperature, organic carbon and often correlated with the microbial population (Ladd and Butler, 1972).

The activity of protease was characterized and quantified in a rendzina under oak, acid brown forest soils under horn beam-beech and a leached brown forest soil under beech by Pantosh Derimova (1983).

Khaziyev and Khabirov (1983) observed that the activity of protease was highest in the steppe zone and decreased in soils of forest steppe and forest zones through a comparative analysis of the change in protease activity in soils of forest, forest-steppe and steppe zones of the cis - ural regions.

Hayano and Tubaki (1985) determined the sources of soil proteases by monitoring the decrease in enzyme activity after the selective inhibition of micro organisms. Thus soil protease activity have been correlated with the occurrence of a number of soil bacteria, but the specific organisms involved in protease synthesis were not identified according to Vardavakis (1989).

Sato and Omura (1989) reported that the properties of protease in uplands were different from those soils under water logged condition where *Bacillus* sp. served to be the major source of soil proteases. Protease activity was stimulated in a soil mixed with 25 g kg^{-1} of composted municipal waste according to Perucci and Guisquiani (1990).

Dilution plate analysis showed that the thermophilic and mesophilic bacteria producing protease enzyme included *Micrococcus, Streptococcus, Pseudomonas* and *Bacillus* (Abdel Monem and Aly, 1990). For the isolation of protease, several methods are available of which the most prominent one is suggested by Caplan and Fahey (1992), using an azocoll agar plate and gelatin liquefaction test for the isolation and selection of soil bacteria involved in the synthesis of soil protease.

The splitting sites and relative hydrolysis ratios of a polypeptides were used as an index for comparing the enzymatic properties of soil protease and microbial protease in samples of grey lowland soils from paddy fields under rotations of rice and wheat in Japan (Watanabe & Hayano, 1994)..

The bacterial groups selected by azocoll agar plate and gelation liquefaction test were characterized by high levels of extracellular protease (Z - Flase and Caesinase) activity as reported by Watanabe *et al.* (1994).

The activity of protease was greater in soils of higher fertility and thus could be used as a soil fertility index (Songyin *et al.*, 1994).

Under *in vitro* condition, Bending and Read (1995) studied the activities of nutrient mobilizing enzymes (Protease) in birch litter and concluded that the activity of protease increased in organic matter that was incubated for 28 - 30 days.

2.10. Dehydrogenase

Dehydrogenase is considered as one of the better indicators of microbial activity since it occurs only within living cells, unlike other enzymes which are mostly extra cellular. Active dehydrogenases are considered to exist in soil as integral parts of intact cells and are thought to reflect the total range of oxidative activities of the soil microflora.

2.10.1. Assay methods

Dehydrogenase activities of soils are determined from the rates of reduction of 2, 3, 5 Tri phenyl tetrazolium chloride to Tri phenyl formazon which is extracted and measured spectrophotometrically (Lenhard, 1966).

Thalman (1968) described a method of dehydrogenase activity assay by extracting TPF twice with acetone which was less toxic and efficient. Kiss *et al.* (1969) also recommended the estimation of dehydrogenase activity using Triphenyl tetra zolium chloride (TTC).

A method of dehydrogenase assay by using different extraction mixture (tetrachloroethylene : acetone) and a lower incubation temperature was described by Benefield *et al.* (1977).

Trevors *et al.* (1982) suggested a method using idonitro tetrazolium chloride INTC produced by microbes because of the easy extractability without interference from phenolic compounds normally present in soil. Trevors (1984) and Griffiths (1989) suggested modification over the existing method whereby the incubation of soil samples with the substrate was carried under optimal conditions.

An improved and accurate method for determining the dehydrogenase activity of soils with iodonitro tetrazolium chloride (INTC) was suggested by Von Mersi and Schinner (1991).

Spothelfer Magna *et al.* (1993) imposed modifications on the procedure described by Thalman (1968).

2.10.2. Factors influencing the enzyme activity

Dehydrogenase, being an indicator of soil microbial activity is highly sensitive to management practices such as fertilization, addition of amendments such as lime, manures and compost and also to the seasonal variations.

Dehydrogenase activity was found to decrease when the soil was stored air dried at room temperature (Ross, 1970). Activity was also reduced when the soil was stored moist at 4^0 C as reported by Pancholy and Rice (1972).

Bolton *et al.* (1985) compared the dehydrogenase activity with respect to conventional systems and organic systems and concluded that the dehydrogenase activity (DHA) was higher in the organic systems. The activity of dehydrogenase enzyme was also found to be greater in the earthworm cast than in the soil as per the reports of Mackay and Kladivko (1985).

The dehydrogenase activities of mesotropic and autotrophic brown forest soils according to Ohlinger (1986) were greatly reduced by simulated acid rains. The contamination by heavy metals arising from sludge applications did not affect the phosphomonoesterase activity of soil while an inhibitory effect on the dehydrogenase activity was observed by Reddy *et al.* (1987).

Thus dehydrogenase serve as a sensitive parameter and provides information on the activity of microbial communities and microbial biomass as suggested by Szegi (1988).

Doran (1990) observed that various tillage practices affect microbial biomass and consequently intracellular dehydrogenase activity was reduced. Wolters (1991) observed increased dehydrogenase activity by liming after 15 days of incubation at 15^0 C.

Studies on the dehydrogenase activity by Beyer *et al.* (1993) emphasised the importance of soil type and physico chemical properties to characterize the effect of crop management or cultivation on dehydrogenase activity.

Studies on the effect of compost addition by Martin and Marinissen (1993) revealed that the activity of dehydrogenase increased to a range of 2101 μg of TPF hydrolysed 24 h^{-1} with the application of vermicompost.

The various methods of tillage influenced the activity of dehydrogenase

and the activity was observed to increase in the conventionally tilled field than the no tilled field by Ross, *et al.* (1995) and Reicosky *et al.* (1995).

Soil dehydrogenase activity also declined with increasing soil pH, as observed in alkaline soils of pH 9.5 and 10.0 (Kumar and Kapoor, 1995). A significant correlation between soil dehydrogenase and pH values was observed by Nohrstedt (1995).

Dehydrogenase activities were either positively or negatively related to soil moisture, organic C, total N, soil texture, forms of P and S, CEC and ratio of Mg/Mg + Ca (Baligar *et al.* 1997). According to Cooper and Warman (1997) application of compost showed an increased activity of dehydrogenase in a silty clay soil than the application of manures or fertilizers.

Similarly Tateno (1998) observed increased activity of dehydrogenase due to the application of poultry manure in a clay loam soil.

Fraser *et al.* (1998) observed that the dehydrogenase activity was linked with the levels of available organic C substrates in the soil in a sandy loam soil.

A comparison of enzyme activity and microbial biomass showed that changes in dehydrogenase activity corresponds more closely to microbial biomass than to changes in either phosphatase or urease activity according to Sparrow and Cochran (1998).

2.11. Cellulase

Cellulase, a group of extracellular enzymes, hydrolyse insoluble

cellulose polymers into soluble sugars and act as primary agents of organic matter degradation. Process that influence cellulase activity in the soil could therefore influence cellulase degradation and hence the decomposition of litter and organic matter. As cellulose is insoluble, it cannot be directly assimilated by microorganisms and hence has to be hydrolysed extracellularly by cellulase. Consequently, the decomposition of carbohydrate polymers and the subsequent mineralization of the products have a special significance in the biological cycling of carbon and thus the perpetuation of life on our planet.

2.11.1. Assay methods

A variety of methods and substrates have been used for measuring cellulolytic activity in soils.

A cellulase activity assay can be done by using cellophane or cellulose powder by treating the soils with toluene, petroleum ether or merthiolate as suggested by Markus (1955).

Cellulase activity was reduced in presence of toluene and the gamma irradiation at minimum sterilizing dosage did not bring about any significant change in cellulase activity although at heavier doses some reduction occurred (Kozlov and Kislitsina, 1967).

Tomescu (1970) suggested a method of cellulase activity assay in which the minimum concentration of the substrate (carboxymethyl cellulose) should be at least 0.4% to maintain linearity in the increase of activity with time.

Cellulase activity can also be estimated by measuring the amount of

reducing sugar hydrolysed from cellulose or carboxy methyl cellulose powder as suggested by Pancholy and Rice (1973).

Prior incubation of soils with cellulose was found to enhance the assay of cellulase activity where by the process of hydrolysis was accelerated according to Ambroz (1973). Ibister *et al.* (1980) determined the activity of cellulose by the measurement of CO_2 evolved from labelled and unlabelled cellulose.

2.11.2. Factors influencing activity

Under natural soil conditions the cellulases, like other enzymes are continuously being synthesized and accumulated, inactivated and decomposed. Several work in this aspect have been undertaken.

Kanzawa and Miyashita (1987) found that the activity of cellulase decreased with increasing soil depth in a humus podzolic soil and Alphine Brown soil.

The effect of pH on the extractability of cellulase and protein in two horizons of forest soils was studied by Mc Claugherty and Linkins (1988) with the observation that the extractability of cellulase increased as the pH increased from 3.5 to 5.6. The studies on the activity, location and origin of cellulase revealed that the major portion of cellulase was bound to and protected by soil colloids (Hope and Burns, 1989).

Cellulase is believed to be synthesised by a group of microorganisms which includes bacteria, actinomycetes and fungi. Ishaque and Kluepfel (1990)

suggested that the actinomycetes possess a cellulase system more akin to that of fungi than bacteria and the cellulases are secreted to the surrounding medium.

Variations in the activity of cellulase was noticed due to the difference in the climatic regimes. Higher cellulase activity in the range of 30 - 80 ppm of glucose hydrolysed g^{-1} of soil hr^{-1} in the semi arid climate at Peru was observed by Brown (1991).

Sandor and Eash (1991) studied the activity of cellulase in temperate regions and found out that additions of farm yard manure or green manure stimulated the activity of cellulase. Studies on the cellulase activity by Schinner (1993) at the arctic region revealed that the activity increased after the thawing of frozen top soils.

Soil cellulase activity was considerably reduced due to crude oil application on vegetation which also contributed to a significant reduction in mycorrhizal root numbers and root respiration rates (Linkins and Fetcher, 1993).

Cellulase enzyme are supposed to bind the organic matter or humus, thereby they are immobilized. Mishra *et al.* (1993) also reported that the binding influenced the substrate either directly by restricting active sites or indirectly by decreasing the enzyme mobility. The adsorption affinity with respect to endocellulase was low compared to other exocellulase (Ryu *et al.* 1994).

Cellulase binding to non substrate organic matter considerably reduced the mobility of enzyme in soil and thus protect the enzyme from physical or biological degradation according to Engasser and Horvath (1994).

Dick *et al.* (1994) also observed that the long term cultivation could not affect the cellulase activity as the organic C level is maintained by sesquestering C. Often cellulase is correlated with many factors like organic C, water content and depth of sampling. But the findings of Pennock *et al.* (1994) did not reveal any strong correlation between moisture content and cellulase activity.

The correlation between cellulase activity and microbial biomass C (r = 0.45), was highly significant suggesting that it is more stable and tends to accumulate in complexed forms in soil matrix and less influenced by short term fluctuations in microbial biomass according to Hayano and Tubaki (1995).

For cellulase, the Michaelis - Menten constant, V_{max} was greater in the surface of the no-tilled soil compared to the conventionally tilled soil according to Sevinnen *et al.* (1995).

The addition of organic amendments had a significant positive effect on the cellulase activity according to Tateno (1998) and observed values as high as in the range of 345.23 ppm of glucose hydrolysed g^{-1} of soil 24 h^{-1} by straw mulch which was higher than with the addition of sewage sludge and poultry manure.

2.12. Effect of soil amendments

2.12.1. Organic amendments

Soils with long-term organic inputs typically have improved biological activity, which have implications for current interests in shifting from inorganic

to organic inputs and promoting efficient nutrient cycling in agro ecosystems. The organic amendments are supposed to be time tested materials for improving the fertility and productivity of soils. Soils managed with organic amendments generally have larger microbial populations than those managed with mineral fertilizers. Thus the incorporation of organic amendments in soil promotes microbial and enzyme activity according to the findings of Balasubramaniam *et al.* (1972).

Addition of fresh energy source to the soil also enhances the microbial growth and improved the fertility status of the soil by enhancing the mineralization of nutrients. Tate (1984) found that adding readily metabolizable sugarcane leaves (*Saccharum officinarum* L.) to a large pool of organic matter present in a histosol increased the activity of various enzymes.

The addition of N, P & K fertilizers + manures almost doubled the microbial biomass C compared to the generated values in soils treated with inorganic fertilizers alone as observed by Marumoto (1984).

According to Briton (1989), addition of farm manures annually for 18 years to a Swedish soil under a wheat-clover-grass-potato rotation increased both the dehydrogenase activity and soil respiration thus increasing the microbial population. The impact of residue management on soil enzyme activities studied by Cochran *et al.* (1989) revealed that application of straw to the crop fields resulted in increased activity of enzymes and biomass carbon.

Similarly, Anwarzay et al. (1990) found that soils managed with organic amendments generally have larger and more active microbial population that

those managed with mineral fertilizers alone. Sakamoto and Oba (1991) also observed that the application of organic manures like FYM usually increased the soil microbial biomass.

Haider (1991) observed an increased C : N ratio due to the application of cowdung along with oil cake, resulting in an increased microbial biomass C, activity of urease and dehydrogenase.

Gupta et al. (1992) suggested FYM as a good source of P and attributed increased levels of enzyme activities and microbial biomass due to the decomposition products of the manure.

The enzyme activities were observed to increase with the application of sugarcane bagasse along with an increase in the available N, P, K content and the bacterial population of the rhizosphere (Wang *et al.* 1992). Similarly, the findings of Martens *et al.* (1992) revealed an increased activity of aryl sulphatase, B-glucosidase, B-galactosidase, invertase, dehydrogenase, urease and phosphatase by the application of poultry manure, sewage sludge and plant residues.

Perucci (1992) also reported that addition of organic residues increased the activities of amylase, arylsulphatase, catalase, deaminase, dehydrogenase and phosphomonoesterase.

The farm yard manure, which is a composted mixture of cowdung and farmwaste is widely used as a nutrient source in the Indian subcontinent for sustaining the soil productivity by maintaining soil health. According to Goyal *et al.* (1993), the application of FYM increases the microbial biomass C and also the enzyme activities.

Singh and Singh (1993) observed that low input organic farming improved soil fertility and the responses of the microbial biomass in a tropical dryland agroecosystem.

Mary Fauci and Dick (1994) reported that recent organic inputs (peavine, beef manure, poultry manure and FYM) regardless of the long term management, had a positive effect on soil biological response, which was controlled by residue composition (lignin content). With repeated addition of organic residues, high levels of enzyme activities were inhibited by a feed back mechanism due to an adequate supply of energy (Caster, 1995).

Singaram and Kamalakumari (1995) in a long term field experiment in a Typic Ustropept observed enhanced activities of soil enzymes with higher rates of N, P and K fertilization with and without FYM. Study on the effect of organic amendments on soil biological properties by Ross *et al.* (1995) revealed an increase in soil organic matter content and soil enzyme activities as a result of rhizodeposition of winter wheat and barley (*Hordeum vulgare.* L)

2.12.2. Fertilizers

Fertilizers are the kingpin in the present day agriculture. The increase of agricultural productivity due to the application of inorganic fertilizers is quite obvious. These fertilizers supply nutrients essential for plant growth and also plays a significant role in nutrient cycling by stimulating the activity of various enzymes responsible for the nutrient transformations.

Vlasyuk and Lisoval (1964) found that the addition of inorganic

fertilizers increased the activity of both acid and alkaline phosphatase. Studies on phosphatase activity with the addition of inorganic N fertilizers by Eivazi and Tabatabai (1977) revealed increased activity of phosphatase which may be due to the changes in pH associated with inorganic inputs rather than to the direct effects of N. However, Bremner and Mulvaney (1978) observed that ammonium based fertilizers had no effect on soil urease activity.

A highly significant and positive correlation was observed between urease and amidase with inorganic N inputs according to Frankenberger and Tabatabai (1981). The phosphatase activity in the soil samples from several grassed and cropped lands showed a significant and positive correlation with inorganic sources (Sharpley, 1985).

The positive influence of mineral N, P and K on the enzyme activities (urease, invertase, catalase and phosphatase) of five soil types was noticed through a pot experiment by Peshakov *et al.* (1986).

Studies on the effects of systematic fertilizer application and manuring on biological processes in soil revealed that fertilization increased the microbial proliferation and also the enzyme activities (Ampova and Parishkova, 1988). The effects of chemical fertilizers with or without organic manures on the enzyme status was studied by He *et al.* (1992).

Though the inorganic fertilizers were observed to stimulate the activity, the stimulatory effect was found to be less compared to the addition of organic residues (Nannipieri, 1996).

The long term application of 30 - 70 kg each of nitrogen, phosphorus

and potassium was observed to increase the activity of invertase, urease and phosphatase according to Evdokimova and Tishchenko (1993).

2.12.3. Lime

During recent decades activities of man have increasingly lead to acidification of terrestial ecosystem, with a consequent negative impact on nutrient transformations and several attempts have been made to buffer the soil chemical alterations consequent upon acid input (Mc Lean et al. 1978).

Liming, as the term applies to agriculture is the addition to the soil of any calcium or calcium - and magnesium - containing compound that is capable of reducing acidity. However, data on the effects of liming on soil chemical, biochemical process and microbiological activity are often contradictory, probably due to spatial heterogenity and complexity of the soil environment and lack of standardization of the methods.

Liming, as it increases the available P due to phosphate solubility, plays a significant role in influencing the activity of phosphatase (Alexander, 1980). The microbiological properties as well as biochemical processes have been investigated in atmospherically acidified soils before and after liming by Haynes and Swift (1988).

Jenkinson (1988) suggested the determination of phosphatase activity to assess the effects of liming as this enzyme serves as an important component of the P cycle. The measurement of CO_2 evolution or O_2 uptake, ATP content and heat output of the microbial population cannot be taken as a standard

parameter for assessing the effect of liming as opined by Nannipieri *et al.* (1990).

Polland and Riha (1990) while studying the effects of liming on phosphatase activity observed a decrease in activity with the application of lime to an acid soil. Similar effects of liming showing a decrease in the activity of phosphatase was also reported by Haynes and Swift (1990).

Zelles et al. (1990) concluded that prokaryotes could replace fungi in long term dolomite amended soil by following changes in ratios of ergosterol or glucosamine to muramic acid.

Trasar Cepeda (1991) conducted studies on P mineralization and phosphatase activity in various limed soils and did not find any correlation between net P mineralization and phosphatase activity. The investigation on the effects of liming on chemical, biochemical and microbiological properties by Badalucco (1992) revealed a decrease in microbial biomass C : N ratio and acid phosphatase activity consequent to the application of lime.

Skogland et al. (1998) revealed that some micro organisms that inhabit acid soils were killed by liming and thus an increase in microbial biomass results in the release of nutrients from these dead microbes consequent to liming.

2.12.4. Biofertilizers and vermicompost

Earthworms constitute a major portion of soil faunal population and are known to accelerate plant residue decomposition, and the feeding activities

increases the microbial population while casting and excretion directly improves the enzyme activities according to Krishnamoorthy and Vajranabhaiah (1986).

Satchell *et al.* (1986) observed an increase in phosphatase activity in a culture medium consisting of cellulase pulp and phytin due to the effect of earthworm activity in increasing the microbial biomass in the culture medium.

The microflora of the intenstinal tract of the earthworms was responsible for the increased enzyme activities as opined by Gorbenko *et al.* (1986). The presence of earthworm casts in cultivated soil was observed to be responsible for the increased activity of amidase according to Sandor (1987).

Studies on the effects of vermicompost on soil fertility status carried out by Sharply and Syers (1987) revealed an increased availability of P due to the indirect microbial population and enzyme activities.

Tiwari et al. (1989) reported increased bacterial and fungal population by selective feeding by earthworms on organically rich substrates which breaks down during passage through the gut and ultimately resulting in greater enzyme activity in the casts.

The presence of earthworms *(Allolobophora callignosa)* resulted in an increased CO_2 uptake and activities of the enzymes like cellulase and sulphatase in the subsoil of judgeford silt loam as observed by Ross and Cairns (1992).

The presence of earthworms were found to alter the bioavailability of nutrients with subsequent increase in mineralization due to increased activity of the biomass (Jorgensen, 1992).

A positive influence of earthworms on N_2 fixing bacteria was also observed by Lee (1993). The same author reported that the increased microbial activity was responsible for the increased urease activity observed in soils amended with vermicompost.

Pedersen and Handriksen (1993) also observed changes in the biological properties of soils that come into contact with earthworms. Satchell and Martin (1994) reported increased phosphatase activity in the faeces of earthworm *(Allolobora caliginosa, Eisenia foetida* and *Lumbricus rubellus)* which was grown in the laboratory with a paper waste sludge/phytin mixture as substrate.

The possible interactions between the rhizosphere and earthworms with respect to pollutant degradation was investigated by Anderson *et al.* (1993).

The presence of earthworms was also found to affect soil exchange properties of a range of herbicides, increasing up to three fold the pesticide binding affinity of burrow linings compared to bulk soil according to the reports of Basker *et al.* (1994).

Changes in the physico-chemical properties of soils are also reported with the addition of vermicompost (Stehouwer et al. 1994). Andrew (1995) studied the bio availability of atrazine to soil microbes in the presence of earthworm *Lumbricus terrestrius*. L and concluded that the presence of earthworms had no effect on atrazine in soil solution.

2.12.5. Insecticides

Evidence for the direct influence of agrochemicals on soil enzymes is

most circumstantial, for example, due to inhibition of the active sites and the current literature about direct action of chemicals is scarce. Pesticides also found to impose indirect effects on enzyme activity due to their interactions with microbial populations. Adsorption of pesticides by soil colloids and perhaps their photo decomposition and volatalization, decreases pollutant concentrations and their inhibitory effects on soil enzyme activities (Cervelli *et al.* 1978).

The microbiological activity of soils is far too complex to be described adequately by a single parameter. Thus the assay of an enzyme activity alone is of little value for assessing the side effects of pesticides in soil and it should be considered as a supplementary tool to obtain functional information on specific aspects of the bioactivity of soil (Sikora *et al.* 1990).

Studies on the effect of malathion, ciodrin, dichlorvos and zinophos on the activities of soil enzymes like acid phosphatase, aryl esterase and aryl acylamidase revealed that the activity of aryl acylamidase was inhibited consequent to the application of these chemicals which could be restored with the degradation of pesticides (Nakamura *et al.* 1990).

Wu *et al.* (1990) reported that application of one or more pesticides to the same soil for many years lead to the build up of pesticide residues or metabolites where by the possibility of damaging effects upon soil microbial biomass or the enzyme activities was much greater.

Studies on the effects of pesticides on enzyme activities by Katayama and Kuwatsuka (1991) observed that the chlorothalonil interacts with thiol

functional groups of enzymes and inhibits certain steps in glycolysis responsible for the reduced cellulolytic activities.

The inhibitory effect of a pesticide on enzyme activity can last as long as the pesticide concentrations are sufficiently high to permit its interaction with the enzyme molecule (Nannipieri and Bollag, 1991).

The effects on dehydrogenase activity of formulated hexachloro cyclo hexane (HCH), carbaryl and benomyl in an alluvial soil were tested under flooded condition and found that at a dosage of 1 mg/kg, no inhibition occurred (Nannipieri and Bollag, 1991).

An increase in microbial biomass in a aldicarb treated sandy loam soils in the field even at lower doses were reported by Jones et al. (1991). The application of terbufos, triazophos, trichloronate, dichloropropane-dichloropropene and nitrapyrin caused a reduction in dehydrogenase activity to an extent of 60% (Tu, 1991).

The addition of phosphorothioates, fenitrothion, malathion and phorate at elevated doses between 50 - 100 mg/kg to a sandy clay loam (1.0% organic carbon, pH -5.8) and a silt loam (2.2% organic carbon, pH 5.4) strongly inhibited urease activity according to Lai and Tabatabai (1992).

Schaffer (1993) studied the interactions between insecticides and soil enzyme activities for oxidoreductases, esterases, glucoside hydrolases, proteases, amidohydrolases and found that the soil enzyme activities were affected by persistence, concentration, toxicity and bioavailability of the chemical and its mode of inhibition.

Studies on the effects of aldicarb on soil microbial biomass by Jenkinson and Coleman (1994) revealed that the consistent significant side effect caused by the addition of aldicarb was actually of beneficial nature and produced no long term harmful effects on soil microbial biomass or its activity.

Doses of profenphos (5.4 mg ai/g of soil) accelerated the urease activity for six weeks after treatment, but inhibited enzyme activity after longer periods as per the reports of Abdel Mallek et al. (1994). Hart and Brookes (1996) reported that the continuous use of the pesticides, either singly or in combination, therefore had no measurable long-term effects on the soil microbial biomass or its activity as assessed by C or N mineralization.

Experiments were also carried out at Rothamsted experimental station to study the effects of the long-term application of three pesticides on the yield of spring barley by Bromilow et al. (1996).

Wong *et al.* (1998) observed that the acid phosphatase activity was closely related to insecticide application history rather than to pH, CEC or type of soil and can thus be used for the prediction of enhanced biodegradation in soils prone to phosphorodithioate insecticide failure.

Landis (1991) reported that soil phosphatase activity is crucial in determining the terrestial environment polluted by organic phosphate pesticides.

2.12.6. Herbicides

The popular trend towards reduced tillage in agriculture has aroused increased interest in the application of herbicides in combination with urea

based fertilizers (Christenen and Magleby, 1982). This has emphasised the need for information concerning the effects of herbicides on transformations of various nutrients which results in their availability. Numerous studies on the effect of herbicides have been reported by various authors.

Use of herbicide Afalon (Linuron) in field over many years unfavourably affected the development of micro organisms and resulted in the decreased activity of cellulase and phosphatase activities according to Furczak and Gostkowska (1982).

The herbicide 2,4-D (2,4 - dichlorophenoxy acetate) has been used widely since 1962 to control dicotyledonous weeds and has been regarded as less toxic to the soil microorganisms and the enzyme synthesis mechanism (Stott *et al.*, 1983).

The applied herbicides may be rapidly degraded in soil and/or oxidatively coupled and incorporated in humic molecules according to Slott et al. (1983).

The application of the herbicide (Goltix) at 50 ppm, first reported to increase the urease activity followed by a decrease while the phosphatase activity remained similar (Gadkari, 1984).

Smith *et al.* (1989) suggested that repeated applications of 2, 4-D in the amine form can lead to faster degradation and result in soil microbial adaptation to herbicides.

Pototskaya and Tyul menkov (1990) reported that application of

herbicides to potatoes produced 20-30% variation in phosphatase, protease and dehydrogenase activity in soil compared to untreated plots.

Schuster and Schroder (1990) investigated the soil microbial adaption in response to long term herbicide application. Studies on the effects of butachlor by Olson and Lindwall (1991) revealed that under laboratory conditions application of butachlor reduced the nitrification and urease activity.

Gostkowska and Furczak (1991) reported a decrease in the number of heterotropic anaerobic bacteria, cellulolytic bacteria and enzyme activities (phosphatase) due to continual application of Gesatop 50 (A.S. Simazine).

Narain Rai (1992) reported that the fungal, bacterial and actinomycete population decreased while dehydrogenase activity and microbial respiration increased due to the application of 2,4-D at the rate of 0.95 kg ai ha^{-1}.

Martens and Bremner (1994) studied the effects of pre emergence herbicides (butylale, cyanazine and siduron) and post emergence herbicides (aciflourfen, diclofop, DPX - 6202, tridiphane) on the transformations of urea in a range of soils.

2.11.7. Pollutants and heavy metals

Pollutants in the form of heavy metals are added to the soil either in the form of municipal wastes (sewage sludge) or any other xenobiotic. The impact of these pollutants is drastic in such a way that it disrupts the biological cycle thus modifying the ecosystem.

The inhibitory effects of heavy metals like Cd, Cr, Cu, Ni, Pb and Zn

on enzymes such as urease, amidase, acid and alkaline phosphomonoesterase was studied by Juma and Tabatabai (1977).

Mitterer *et al.* (1981) found that the dehydrogenase and urease activity was not affected in soil by the fungicide quintozene whilst a pronounced and irreversible inhibition of xylanase resulted.

Measurement of enzyme activity can be used to study the anthropogenic effects of heavy metals and other inorganic and organic chemicals according to Doelman and Haastra (1986).

Dehydrogenase activity was found to be inhibited when terbufos, triazophos, tricholoronate, dichloropropionate, dichloropropene and nitrapyrin was applied to an organic soil with 26.8% organic carbon, according to Tu (1987).

Studies on the inhibitory effects of selenium by Wilke (1988) revealed that the application of Se at 50 mg/kg of soil reduced CO_2 production and dehydrogenase activity under controlled laboratory conditions.

The effect of heavy metals on urease, phosphatase and aryl sulphatase activity of soil has been quantified by determining the ED50 (Doelman and Haastra, 1989). The inhibitory effects of heavy metals such as Cd, Cr, Cu, Ni, Pb and Zn on urease, amidase, arylsulphatase and L-aspargenase was studied by Frankenberger and Tabatabai (1991).

Several attempts were made to quantify the toxic dose of the chemical applied. Thus Doelman and Haastra (1991) developed an equation for determining the ED50 value as $Y \rightarrow C\ (1 + e^{b\,(x\,-\,a)})^{-1} + E$ where $Y \rightarrow$ soil enzyme activity, $X \rightarrow$ natural log of heavy metal concentration, $C \rightarrow$ natural level of enzyme activity.

A negative correlation was observed between enzyme activities of soils and their contents of heavy metals especially of Cu^{2+}, Pb^{2+} and Zn^{2+} by Tyler (1991).

Martens et al. (1992) observed a concentration of 5 - 25 μ mol g^{-1} of heavy metals in compost which considerably decreased the activities of asparaginase, dehydrogenase and arylsulphatase.

Schaffer (1993) suggested that the indirect effects of pesticides on enzyme activities was due to their interaction with microbial population and the dehydrogenase activity can be used as an index for measuring side effects of pesticides.

Pier Lodovico (1994) studied the long term effects of heavy metals from composted municipal waste on soil enzyme activities in cultivated soil.

Decrease in enzyme activities with increasing chloride and sulphate levels due to specific effects on microbial growth and subsequent enzyme synthesis, osmotic desiccation and salting out effect modifying the ionic concentration of the active site of enzyme protein was also observed by Dinesh *et al.* (1995).

Doelman and Haastra (1996) found that zinc served to be an effective inhibitor of urease activity and ED50 was found to be varied from 100 to 300 mg kg^{-1} of soil.

Nannipieri (1997) studied the potential use of soil enzymes as indicators of productivity, sustainability, pollution and quantified the effect of heavy metals by Ecological Dose (ED50) which represents the heavy metal concentration at which the enzyme activity is half of the uninhibited value.

MATERIALS AND METHODS

MATERIALS AND METHODS

The present study on the distribution, characterization and dynamics of soil enzymes was carried out in the Department of Soil Science and Agricultural Chemistry, College of Agriculture, Vellayani during the year 1996-1999, using the facilities available at the post graduate laboratory of the Soil Science and Agricultural Chemistry and the Central Instrumentation Laboratory (CIL) of the NARP (Southern Region). The investigations pertaining to the study was grouped into six separate experiments. The materials used and methods employed for the study are discussed below.

3.1. Studies on Permanent manurial experiments

The first phase of the investigation involved the collection and analysis of soil samples from different Permanent Manurial Experiments (PME) carried out at different locations for various crops as described below.

A transect of land covering a cross section of the state from the coastal sandy tract to the forest loam of the hilly upland region through the midland was selected for the study. The coastal region represented a rice based cropping system from the Rice Research Station (RRS), Kayamkulam. The central midland region covered two important cropping systems viz.,

(i) a coconut based cropping system selected from the Coconut Research Station (CRS), Balaramapuram

(ii) a rice based cropping system located at the Regional Agricultural Research Station (RARS), Pattambi.

The hilly tract was represented by a rubber based cropping system located at the Central Experimental Station of the RRII, Chethackal, Ranni.

Thus the following locations were selected for the study representing the major crops of the area.

1. Rice Research Station (RRS), Kayamkulam - Rice
2. Regional Agricultural Research Station (RARS), Pattambi - Rice
3. Central Experimental Station (CES), Chethackal, Ranni - Rubber
4. Coconut research Station (CRS), Balaramapuram - Coconut

3.1.1. Description of the experimental sites

Site 1 : Rice Research Station (RRS), Kayamkulam

Kayamkulam represents the coastal sandy tract of the *Onattukara* region of Kerala, comprising Karthikapally, Mavelikkara and Karunagappally taluks of Kollam and Alappuzha districts. The details of permanent manurial experiment at the RRS, Kayamkulam is presented below :

Location	:	9° 30' N latitude and 76° 20' E longitude at an altitude of 3 m above MSL
Soil textural class	:	Loamy sand
Taxonomic classification	:	sandy, mixed, isohyperthermic, Ustic Quartzi Psamments.
Number of treatments	:	8
Replications	:	4
Design	:	Randomized Block Design (RBD)
Plot size	:	8 × 4 m^2
Spacing	:	20 × 15 cm
Variety	:	Bhagya

Treatment details :

T_1 : 80 kg N ha^{-1} as Cattle manure (CM)

T_2 : 80 kg N ha^{-1} as Ammonium sulphate (AS)

T_3 : 80 kg N ha^{-1} as Ammonium sulphate + 40 kg P_2O_5 ha^{-1} as Single super Phosphate (SSP)

T_4 : 80 kg N ha^{-1} as Ammonium sulphate + 40 kg K_2O ha^{-1} as Muriate of potash (MOP)

T_5 : 40 kg P_2O_5 ha^{-1} as Single super phosphate + 40 kg K_2O ha^{-1} as Muriate of potash

T_6 : 80 kg N ha^{-1} as Ammonium sulphate + 40 kg P_2O_5 ha^{-1} as Single super phosphate + 40 kg K_2O ha^{-1} as Muriate of potash

T_7 : 80 kg N ha^{-1} (60 kg N ha^{-1} as Ammonium sulphate + 20 kg N ha^{-1} as Farm yard manure (FYM) + 40 kg P_2O_5 ha^{-1} as Single super phosphate + 40 kg K_2O ha^{-1} as Muriate of potash

T_8 : Control

Site 2 : Regional Agricultural Research Station (RARS), Pattambi

A Permanent Manurial Experiment (old PME) was started in the year 1961 with two photosensitive tall indica varieties (Ptb-2 and Ptb-20) during the Viruppu (first crop season) and Mundakan (second crop) seasons respectively. A similar experiment was started during 1973 also (new PME) using dwarf indica variety (Jyothi). The details of these two experiments are presented below.

a. Permanent Manurial Experiment - I

Location	:	Latitude 10° 48' 16" Longitude 76° 11' 00"
Soil	:	Clay loam
Taxonomic classification	:	clayey mixed, Ustic Tropudalfs
Plot size	:	7.8 × 5.25 m
Number of treatments	:	8
Number of Replications	:	4

Design : Randomized block desigh (RBD)

Spacing : 20 × 15 cm

Variety : Jaya

Treatment structure :

T_1 : Cattle manure (20 kg)

T_2 : Green leaf manure (20 kg)

T_3 : Cattle manure (10 kg) + Green leaf manure (10 kg)

T_4 : N as Ammonium sulphate (488 g) or Urea (225 g)

T_5 : Cattle manure (10 kg) + N as Ammonium sulphate (244 g) or Urea (123 g) + P_2O_5 as single super phosphate (278 g) or Mussorie rock phosphate (225 g) + K_2O as Muriate of potash (83 g).

T_6 : Green leaf manure (10 kg) + N, P and K doses as in T_5

T_7 : Cattle Manure (5 kg) + Green Leaf Manure (5 kg) + N, P and K doses as in T_5

T_8 : N as Ammonium sulphate (488 g) or Urea (225 g) + P_2O_5 as Single super phosphate (278 g) or Mussorie rock phosphate (225 g) + K_2O as Muriate of potash (83 g)

b. Permanent Manurial Experiment - II

Location	:	Latitude 10° 48' 16"
		Longitude 76° 11' 00"
Soil	:	Clay loam
Taxonomic classification	:	clayey mixed, Ustic Tropudalfs
Plot size	:	4.85 × 4.55 m
Number of treatments	:	8
Number of Replications	:	4
Design	:	Randomized block design (RBD)
Spacing	:	20 × 10 cm
Variety	:	Jyothi

Treatment structure :

T_1 : Cattle manure (20 kg)

T_2 : Green leaf manure (20 kg)

T_3 : Cattle manure (10 kg) + Green leaf manure (10 kg)

T_4 : N as Ammonium sulphate (488 g) or Urea (225 g)

T_5 : Cattle manure (10 kg) + N as Ammonium sulphate (244 g) or Urea (123 g) + P_2O_5 as Single super phosphate (278 g) or Mussorie rock phosphate (225 g) + K_2O as Muriate of potash (83 g)

T_6 : Green leaf manure (10 kg) + N, P and K doses as in T_5

T_7 : Cattle manure (5 kg) + Green leaf manure (5 kg) + N, P and K doses as in T_5

T_8 : N as Ammonium sulphate (488 g) or Urea (225 g) + P_2O_5 as Single super phosphate (278 g) or Mussorie Rock Phosphate (225 g) + K_2O as Muriate of Potash

Site 3 : Central Experimental Station (CES), Chethackal

Location	:	Latitude 9° 24' 15" N Longitude 76° 49' 15" E
Soil Textural Class	:	Sandy clay loam
Taxonomic Classification	:	loamy-skeletal, kaolinitic, iso hyperthermic Ustic Humid troppepts
Number of experimental rows:		6
Number of replications	:	4

The spacing cum intercropping trial at the CES, Chethackal was started in the year 1983 in a rubber based cropping system. The various components of this system are discussed hereunder.

Treatments	*Details*
T_1	Rubber + Teak and fodder grass (*Panicum repens*)
T_2	Rubber (sole crop) - (5.11 m spacing between rows)
T_3	Rubber + Pineapple - spacing between rubber row and pineapple - 1.95 m, Between pineapple - 30 cm)
T_4	Rubber + Pepper and Cocoa - Spacing between pineapple and pepper - 1.95 m
T_5	Rubber + Banana - Cultivar. Nendran for 2 years followed by yam and colacassia spacing 2.25 m between pepper and banana.
T_6	Rubber + cover crop *(Calapagonium* sp*)* - Spacing 5.11 m from Banana

Site 4 : Coconut Research Station (CRS), Balaramapuram

Location : 8° 29' N latitude and 76° 57' E Longitude, 64 m above MSL.

Climate : Humid Tropical

Soil Textural class : Red Loam

Taxonomic Classification : fine loamy, kaolinitic, iso hyperthermic Kandic Haplustalf.

Treatments : N, P_2O_5 and K_2O at three levels each in all possible combinations applied as Ammonium sulphate (20.5% N) or urea (46% N), Single super phosphate (16% P_2O_5) and Muriate of potash (60% K_2O)

Number of replication : 2

Design : 3^3 Factorial experiment (Partially confounded interactions NPK^2 in RI; NP^2K^2 in R II)

Number of trees/treatment : 4

Number of total trees : 216

Number of border trees : 341

Number of blocks : 6 @ 3 trees/replication

Number of treatments/block : 9

Variety : West Coast Tall (WCT)

Spacing : 7.5 × 7.5 m.

Treatment doses (g/tree/year)

Level	N	P_2O_5	K_2O
0	0	0	0
1	340	225	450
2	680	450	900

Treatment combinations

$N_0P_0K_0$	$N_1P_0K_0$	$N_2P_0K_0$
$N_0P_0K_1$	$N_1P_0K_1$	$N_2P_0K_1$
$N_0P_0K_2$	$N_1P_0K_2$	$N_2P_0K_2$
$N_0P_1K_0$	$N_1P_1K_0$	$N_2P_1K_0$
$N_0P_1K_1$	$N_1P_1K_1$	$N_2P_1K_1$
$N_0P_1K_2$	$N_1P_1K_2$	$N_2P_1K_2$
$N_0P_2K_0$	$N_1P_2K_0$	$N_2P_2K_0$
$N_0P_2K_1$	$N_1P_2K_1$	$N_2P_2K_1$
$N_0P_2K_2$	$N_1P_2K_2$	$N_2P_2K_2$

3.1.2. Collection of soil samples

Soil samples were collected from the above experimental plots covering all the four locations. In the case of PME on rice, surface soil samples up to a depth of 0 - 15 cm was collected after the harvest of the crop using a soil tube specially fabricated for this purpose. The tubes were of 30 cm in length with a handle at the top, having an internal diameter of 5 cm. Soil tubes were hammered into the soil to a depth of 15 cm. The tubes were then taken out, the soil core inside was pushed out and packed in polythene bags.

Soil samples were collected upto a depth of 30 cm from the basins of the experimental plots of the CRS, Balaramapuram at a distance of 90 - 100

cm from the bole of the tree by reinforcing the soil tubes into the soil using a hammer. Soil samples were also collected from the experimental plots of the Central Experimental Station, Chethackal in the same manner using soil tubes upto a depth of 30 cm between rows. Collected samples were then transferred to thick polythene bags, labelled and brought to the laboratory for analysis.

3.1.3. Preparation of samples for analysis

The soil samples were then shade dried and clods were broken gently with a wooden mallet, sieved through a 2 mm nylon sieve and stored in plastic containers with screw cap. Part of the processed soil was retained for the analysis of the soil parameters while the remaining was transferred into another plastic container and stored under 50% field moisture capacity. These samples were then subjected to the analysis of soil enzymes within a week. Analysis of soil samples for various physical, electrochemical and biological properties were attempted after completing the enzyme assay.

3.1.4. Analysis of soil

Soil samples were subjected to three types of analysis :

i. Enzyme activity assay

ii. Physicochemical and electrochemical properties

iii. The respiratory activity

The analytical procedures followed and methods used are briefly outlined below.

3.1.4.1. Soil reaction (pH)

The pH of the air dried samples were determined as per the procedures described by Jackson (1973) with a soil water ratio of 1:2.5 using a pH meter (Model - ELICO-LI-612)

3.1.4.2. Electrical conductivity (EC)

The specific conductance was determined in the same soil-water suspension used for pH determination with the help of a conductivity meter (EC-TDS ANALYSER - ELICO - CM - 183) as per the procedure outlined by Jackson (1973).

3.1.4.3. Organic carbon (OC)

The organic carbon content of the soil samples were determined by the wet digestion method as defined by Walkley and Black (1934).

3.1.4.4. Cation exchange capacity (CEC)

The cation exchange capacity of the soil samples were determined by the method described by Jackson (1973) using normal ammonium acetate (pH 7).

3.1.4.5. Anion exchange capacity (AEC)

The anion exchange capacity was determined by the phosphate adsorption method as suggested by Hesse (1971).

3.1.4.6. Available nitrogen

Available nitrogen in soil was determined by the alkaline permanganage method (Subbiah and Asija, 1956).

3.1.4.7. Available phosphorus

Available P in soil was extracted using Bray No. 1 reagent and was determined colorimetrically by the chlorostannous-reduced molydophosphoric blue colour method in hydrochloric acid medium (Bray and Kurtz 1945).

3.1.4.8. Available potassium

The ammonium acetate leachate collected during the determination of CEC was directly used for the determination of potassium using a flame photometer (Model - Systronics) (Jackson, 1973).

3.1.4.9. Clay content

The clay content was determined by the hydrometer method as suggested by Gupta and Dakshinamoorthy (1980).

3.1.4.10. Sesquioxides

The sesquioxide content of the soil samples were estimated as per the standard analytical procedures as described by Jackson (1973).

3.1.4.11. Respiratory activity

The respiratory activity of the soil samples were determined according to a method outlined by Jenkinson and Powlson (1976), where the

CO_2 evolved from a fixed quantity of incubated soil was collected in an alkali and quantified.

3.1.4.12. Urease activity

The urease activity was determined by following the method described by Broadbent *et al.* (1964).

About 25 g of soil was weighed into an Erlen Meyer flask, to which 4 ml of urea substrate solution was added. Enough water was added to each flask to maintain a tension of 1/3 bar and incubated for 24 hours at 30° C. Then the flasks were removed, $CaSO_4$ solution was added to make up the volume to 100 ml. About 15 ml of the supernatant was taken and colour was developed by adding 10 ml of p-dimethyl amino benzaldehyde which was then read in a Spectro photometer (Bausch and Lomb: Spectronic 2000) at a wavelength of 420 nm. Standards were also prepared by using urea solutions of known concentrations. The results were expressed in terms of urea hydrolysed g^{-1} of soil hr^{-1} in ppm.

3.1.4.13. Phosphatase activity

The phosphatase activity was determined by following a procedure described by Eivazi and Tabatabai (1977).

To 1 g soil in a 50 ml Erlen Meyer flask, 0.2 ml toluene, 4 ml modified universal buffer (pH-6.5) and 1 ml p-nitrophenyl phosphate solution were added and incubated at 23° C for one hour. After incubation, 0.5 M $CaCl_2$

(1 ml) and 0.05 M NaOH (1 ml) were added. The contents were swirled and filtered through Whatman No. 2 filter paper and the intensity of the yellow colour developed was read in a Spectro photometer (Bausch and Lomb : Spectronic 2000) at a wavelength of 420 nm. One percent solution of p-nitrophenyl phosphate was used for the preparation of standards. The results were expressed in terms of p-nitrophenol hydrolysed g^{-1} of soil hr^{-1} in micrograms .

3.1.4.14. Protease activity

The protease activity was estimated as per the procedure defined by Nannipieri, *et al.* (1976).

About 0.5 g of the soil was weighed into a tissue culture tube to which, 1.8 ml of 0.1 M TRIS buffer and 2 ml 0.002 M phenyl alanine were added, and incubated for 60 minutes at 20° C. After the incubation period, the activity was arrested by adding 0.2 ml of 5 M HCl and centrifuged. Supernatant was taken and 2 ml of ninhydrin reagent was added. The violet colour developed was measured at 570 nm using a Spectro photometer (Bausch and Lomb : Spectronic 2000). A series of standards were prepared in the same manner. The results were expressed as micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}.

3.1.4.15. Dehydrogenase activity

The dehydrogenase activity was estimated as per the procedure described by Casida *et al.*, 1964.

About 6 g of the air dried soil was weighed to a 250 ml Erlen Meyer

flask. One ml of 3% triphenyl tetrazolium chloride was added and incubated for 24 hours at 27° C. After incubation, the soil was quantitatively transferred to a glass funnel and was given ethanol washings consecutively till the volume reached 100 ml. The colour intensity was then read in a Spectro photometer (Bausch and Lomb : Spectronic 2000) at 485 nm. A series of standards were used for preparing the calibration curve. The results were expressed in terms of Triphenyl Formazon hydrolysed g^{-1} of soil 24 hrs^{-1}, in micrograms.

3.1.4.16. Cellulase activity

The cellulase activity was estimated according to a method suggested by Pancholy and Rice (1973).

To 5 g of air dried soil taken in a 100 ml Erlen Meyer flask, 10 ml of acetate buffer and 10 ml 1% carboxy methyl cellulose was added. Then the flasks were incubated for 24 hours at 37° C and left undistributed. After the incubation, 50 ml of distilled water was added and contents were filtered. About 1 ml of the filtrate was taken and 4 ml of Anthrone reagent was added. The intensity of the green colour developed was read in a Spectro photometer (Bausch and Lomb : Spectronic 2000) at 620 nm. Glucose was used as standard at different concentrations to give a standard calibration graph. The results were then expressed as the amount of glucose hydrolysed g^{-1} of soil 24 hrs^{-1} in ppm.

3.1.4.17. Enzyme kinetics

The kinetics parameters K_m and V_{max} of the five soil enzymes were evaluated based on the Line Weaver - Burk plot. Varying substrate

concentrations were employed for this study using soil as the enzyme source. Enzyme activity rates (V) were determined at these concentrations. According to the Line weaver - Burk equation, when 1/V is plotted against 1/S a straight line graph is obtained. The slope is K_m / V_{max}, the intercept on the ordinate is $1/V_{max}$ and the intercept on the abscissa is $-1/K_m$. K_m and V_{max} of the enzymes were calculated from the Line Weaver - Burk plot prepared with 1/V against 1/S (Vaughan and Ord, 1991).

3.1.5. Statistical analysis

The data generated from these experiments were subjected to the analysis of variance as per the design and their significance was tested by the F test (Snedecor and Cochran, 1975). Correlation studies were also carried out between enzyme activities and soil parameters to find out the degree of association between them. Similarly multiple regression analysis was carried out to define the enzyme activity in terms of soil parameters. Path coefficient analysis was also attempted to study the interrelationship among soil parameters and enzyme activities (Panse and Sukhatme, 1967).

3.2. Pot culture experiments

This experiment was undertaken with an objective of assessing the effect of agrochemicals on the activities of the enzymes. The study was programmed through two pot culture experiments as given below.

a. To study the effect of fertilizers, manures, organic amendments and liming.

b. To study the effect of insecticides, fungicides, bactericides and herbicides.

3.2.1. Experimental site

The pot culture was carried out in the pot culture yard adjacent to the Department of Soil Science and at Agricultural Chemistry at the College of Agriculture, Vellayani in the garden maintained by the Department of Horticulture.

3.2.2. Soil

The soil used for the study was a clay loam taken from the paddy lands of the Instructional Farm of the College of Agriculture, Vellayani. The soil belonged to the taxonomic class Fine-loamy, kaolinitic, isohyperthermic, Typic Kandiustults. The soil was then shade dried, powdered and used as the potting medium @ 8 kg per pot.

3.2.3. Crop and variety

The study was conducted using rice as the test crop. The variety used was Kanchana, which is a short duration variety of about 105 - 110 days.

3.2.4. Season

The study was conducted during the third crop season from January to May, 1999.

3.2.5. Nursery

Sprouted seeds were sown in the pots under puddled condition and

seedlings were raised. Twenty one days old seedlings were transplanted to the main pots at the rate of 2 seedlings / hill.

3.2.6. Design of the experiment

The two pot culture experiments were laid out in a CRD which are detailed below :

a. Pot culture experiment - I

Number of treatments : 13

Number of replication : 3

Design : CRD

Treatment structure :

T_1 : Control	T_2 : Phorate	T_3 : Carbofuran
T_4 : Quinalphos	T_5 : Monocrotophos	T_6 : Chlorpyriphos
T_7 : Hinosan	T_8 : Bavistin	T_9 : Streptocycline
T_{10} : Benthiocarb	T_{11} : 2, 4-D	T_{12} : Butachlor
T_{13} : Oxyflourfen		

b. Pot culture experiment - II

Number of treatments : 16

Number of replication : 3

Design : CRD

Fertilizers : 2 levels

Lime : 2 levels

Manures : 4 levels

Treatment structure :

T_1 : $F_0L_0M_0$	T_2 : $F_0L_0M_1$	T_3 : $F_0L_0M_2$
T_4 : $F_0L_0M_3$	T_5 : $F_0L_1M_0$	T_6 : $F_0L_1M_1$
T_7 : $F_0L_1M_2$	T_8 : $F_0L_1M_3$	T_9 : $F_1L_0M_0$
T_{10} : $F_1L_0M_1$	T_{11} : $F_1L_0M_2$	T_{12} : $F_1L_0M_3$
T_{13} : $F_1L_1M_0$	T_{14} : $F_1L_1M_1$	T_{15} : $F_1L_1M_2$
T_{16} : $F_1L_1M_3$		

F_0 : Control	M_0 : Control
F_1 : Dose of N, P and K as per POP	M_1 : Farm yard manure
L_0 : Control	M_2 : Vermicompost
L_1 : Dose of lime as per POP	M_3 : Green leaf manure

The chemicals were applied to the crop as per the recommended dose in the package of practices. Staggered application of chemicals was done to simulate field conditions. The preemergent herbicide oxyflourfen was applied even before transplanting the crop. Remaining chemicals were applied on the same date so as to ensure uniformity in the time of reaction in soil and its interactions with soil components.

3.2.7. Treatment materials

The details of the experimental materials used are presented below in the following table :

Experimental Materials - Pot Experiment - II

Sl. No.	*Manures / fertilizers / amendments*	*Analysis*
1.	Urea	46% N
2.	Single super phosphate	15.7% P_2O_5
3.	Muriate of Potash	59% K_2O
4.	Farm yard manure	0.4% N, 0.3% P and 0.2% K
5.	Green leaf manure	0.8% N, 0.4% P_2O_5 and 1.01% K_2O
6.	Vermicompost	1.6% N, 0.76% P_2O_5 and 2.71% K_2O
7.	Lime ($CaCO_3$)	100 (Neutralizing value)

3.2.8. After cultivation and irrigation

Two hills per pots was maintained and gap filling was done in the case of pots where the seedlings failed to establish. The pots were kept weed free by hand weeding operation. The pots were kept under submergence by irrigating regularly.

3.2.9. Plant protection

Two spraying were given with Ekalux (@ 0.05 %) as a contingency spray by taking utmost care to avoid spray fluid falling on the soil in the pot.

3.2.10. Sample collection

Soil samples were collected from the pots at three critical stages of the

crop growth namely active tillering, panicle initiation and harvest, by following the funnel method of sampling. Collected samples were then taken in polythene bags, properly labelled and brought to the laboratory for further processing and analysis.

3.2.11. Harvest

The number of productive tillers were counted and then the crop was harvested. The straw and grains were separated, dried and weight of both were recorded.

3.2.12. Preparation of soil samples

The soil samples were dried in shade, powdered with a wooden mallet and sieved through a 2 mm sieve. Then the soils were stored in polythene containers with screw cap at 50% field moisture capacity for further analysis.

3.2.13. Analysis of soil

The initial soil samples without the application of treatments were subjected to the analysis of the different soil parameters and enzyme activities. The soil samples collected at different stages of the crop growth viz., active tillering, panicle initiation and harvest were analysed immediately for enzyme activities. The procedures adopted are detailed in the following paragraphs.

3.2.13.1. Urease activity

The urease activity was determined by following the method described by Broadbent *et al.* (1964) as in 3.1.4.12.

3.2.13.2. Phosphatase activity

The phosphatase activity was determined by following a procedure described by Eivazi and Tabatabai (1977) as in 3.1.4.13.

3.2.13.3. Protease activity

The protease activity was estimated as per the procedure defined by Nannipieri, *et al.* (1976) as in 3.1.4.14.

3.2.13.4. Dehydrogenase activity

The dehydrogenase activity was determined as per the procedure outlined by Casida *et al.* (1964) as in 3.1.4.15.

3.4.13.5. Cellulase activity

The cellulase activity was estimated according to a method suggested by Pancholy and Rice (1973) as in 3.1.4.16.

3.2.13.6. Microbial biomass

Microbial biomass was determined by the chloroform-fumigation-incubation method on a 12 g (fresh weight) soil sample (Jenkinson and Pawlson, 1976). Chloroform was added till saturation level and incubated for 24 hours at room temperature. About 100 mg of soil was added to the fumigated soil as inoculum. Simultaneously a blank was also maintained with only the fumigated soil to which no inoculum was added. The entire set up was left undisturbed

with CO_2 trap arrangements for a week at room temperature. The amount of CO_2 evolved during one week incubation following fumigation was determined by titration. The microbial biomass C (C_{mic}) was then determined using the formula.

$$C_{mic} = CO_2 \text{ fumigated} - CO_2 \text{ control} / 0.41$$

The results were then expressed as the amount of CO_2 evolved per gram of soil $hour^{-1}$ in micrograms.

3.2.13.7. Statistical analysis

The data generated from the pot experiments were subjected to the analysis of variance as per the design and their significance was tested by F test (Snedecor and Cochran, 1975).

Table : 1 *Details of Experimental Materials - Pot Culture Experiment - I*

Sl. No.	Chemical	Molecular Formula	Forumulation	Recommended dose
1.	Phorate	$C_7H_{17}O_2PS_2$	10% G	1.5 kg ai/ha
2.	Carbofuran	$C_{12}H_5NO_3$	3 G	0.5 to 9.75 kg ai/ha
3.	Quinalphos	$C_{12}H_{15}O_8N_2PS$	25 % EC	0.025 to 0.05 %
4.	Monocrotophos	$C_7H_{14}O_5NP$	40 % EC	0.05 %
5.	Chlorpyriphos	$C_9H_{11}Cl_3NO_3PS$	20 % EC	0.02 %
6.	Hinosan	$C_{10}H_{13}O_4PS$	50 % EC	0.1 %
7.	Bavistin	$C_{10}H_{17}N_3O_2$	50 % WP	500 g/ha
8.	Streptocycline	$C_{21}H_{39}N_7O_{12} + (9:1)$ $C_{22}H_{24}N_2O_8$	-	15 g/300/of H_2O
9.	Benthio carb	$C_6H_{28}ClNOS$	50 EC	2.0 kg ai/ha
10.	2,4-D	$C_5H_4O_3Cl_2$	80 %	1.0 kg ai/ha
11.	Butachlor	$C_{17}H_{27}N_2OCl$	5 %	1.0 kg ai/ha
12.	Oxyflourfen	$C_{10}H_5NO_3F_3Cl$	23.5 % EC	0.2 kg ai/ha

RESULTS

RESULTS

The present investigation on distribution, characterization and dynamics of soil enzymes involved the collection and analysis of soil samples from the four ongoing field experiments that are being conducted at the following locations.

a. Rice Research Station (RRS), Kayamkulam

b. Regional Agricultural Research Station (RARS), Pattambi

c. Central Experimental Station (CES), Chethäckal

d. Coconut Research Station (CRS), Balaramapuram

Besides, the effect of agrochemicals *viz.* insecticides, fungicides, herbicides, liming materials, fertilizers and manures on soil enzymes were also studied through pot culture experiments using rice as a test crop. Assay of the activity of selected soil enzymes and the analyses for different soil parameters like pH, EC, organic carbon, cation exchange capacity, anion exchange capacity, available nitrogen, available phosphorus, available potassium, clay content, sesquioxides and respiratory activity were carried

out in the collected samples. The data obtained were subjected to (1) the analysis of variance to study the treatment effects of the experiments on enzyme activities and soil parameters, (2) correlation analysis to study the association between soil parameters and enzyme activities and (3) regression analysis to study the functional relationship between the enzyme activities and soil characteristics. The results of the study conducted are thus presented in the following sections.

4.1. Permanent manurial experiment on rice at the R.R.S., Kayamkulam

4.1.1. Soil parameters

The results of the analysis of soil parameters are presented in Table 2.

4.1.1.1. Soil reaction

The treatments had no significant effect on soil pH. The mean values for pH ranged from 4.82 to 5.17 indicating an acidic soil reaction. The highest value for soil pH was noticed for T_7 (5.17) which did not differ significantly from other treatments.

4.1.1.2. Electrical conductivity

The results of the analysis for EC showed a significant difference with respect to treatment effects.

The mean values for EC ranged between 110.75 to 150.59 μSM^{-1}. The highest value was recorded for T_2 (150.59) which was on a par with T_6 (150.53) and were significantly different from T_1 (129.65), T_3 (110.75), T_4 (139.72), T_5 (141.48), T_7 (111.52) and T_8 (112.35) while the lowest value was recorded for T_3 (110.75).

4.1.1.3. Organic carbon

The results revealed that the applied treatments had significant effect on the organic carbon content of the soil samples. The mean values ranged between 0.79 to 1.13 per cent. The highest value was recorded for treatments T_1 (1.13) followed by T_6 (1.04) and T_7 (0.99) which were statistically on a par while the lowest value was recorded for T_4 (**0.79**) which was at par with T_8 (0.81), T_5 (0.88), T_3 (0.83) and T_2 (0.93).

4.1.1.4. Cation exchange capacity

From the results it was inferred that the treatments had produced significant difference in the cation exchange capacity of the soil. The mean values for the cation exchange capacity ranged from 5.40 to 6.35 c mol (p^+) kg^{-1}. The treatments T_2 (6.35) and T_6 (6.35) had the same effect on cation exchange capacity and these were on a par with treatments T_5 (6.30), T_7 (6.17), T_4 (5.87) and T_3 (5.95). The lowest value was recorded for T_1 (5.40) which was found to be at par with T_8 (5.50).

4.1.1.5. Anion exchange capacity

The results revealed that the treatments had no significant effect on the anion exchange capacity of the soil.

The range of mean values were from 1.76 to 3.03 c mol [e^-] kg^{-1} of soil. However the highest value was noticed for T_4 (3.03) which did not differ significantly from other treatments.

4.1.1.6. Available nitrogen

The results on available nitrogen status of the experimental site indicated that the treatments had a significant effect. The mean values for the available nitrogen ranged from 85.75 to 172.25 kg ha^{-1}. The highest value was noticed for T_4 (172.25) which was on a par with treatments T_3 (160.75), T_5 (156.75), T_7 (155.50), T_1 (154.25), T_2 (150.00) and T_6 (140.25). The available nitrogen content in all the treated plots were significantly higher than the control plot (85.75).

4.1.1.7. Available phosphorus

A highly significant difference in the available phosphorus status due to treatment effects was observed in this experiment. The mean values for the available phosphorus ranged from 12.84 to 59.03 kg ha^{-1}. The highest value was noticed for T_7 (59.03) which was significantly superior to all other treatments viz., T_1 (39.70), T_5 (34.69), T_3 (34.68), T_6 (32.49), T_2 (23.73), T_4 (18.07) and T_8 (12.84). The lowest value was recorded in the control plot (12.84) which was significantly lower than the treated plots.

4.1.1.8. Available potassium

From the results it was clear that the treatments had a significant effect

on the available potassium status of the site. The mean values ranged between 42.0 to 106.40 kg ha^{-1}. The highest value was recorded for T_6 (106.40) which was found to be on a par with the treatments T_7 (98.0), T_5 (89.6) and T_4 (84.0) while the treatments T_2 (61.60), T_8 (61.10), T_3 (53.2) and T_1 (42.0) were also at par statistically.

4.1.1.9. Clay content

The data on clay content revealed that the treatments did not impose any significant variation in the clay content of the soil. The mean values ranged between 5.0 to 7 per cent. The highest value was observed for T_4 (7.00) which did not differ significantly from other treatments T_2 (6.75), T_7 (6.5), T_1 (6.25), T_8 (6.00), T_3 (5.75), T_5 (5.50) and T_6 (5.00).

4.1.1.10. Sesquioxides

The results of the sesquioxide content of the samples recorded mean values ranging between 4.70 to 6.60 per cent. The highest value was recorded in T_4 (6.6) which was found to be on a par with T_3 (6.5), T_6 (6.4), T_5 (6.20) and T_2 (6.20). Similarly treatments T_8 (5.90), T_2 (6.20), T_5 (6.20) and T_6 (6.40) were also on a par statistically. The lowest value was recorded for T_1 (4.70) which was found to be at par with T_7 (5.10).

4.1.1.11. Grain yield

The data on the grain yield showed significant difference due to treatment effects. The mean values ranged between 631 to 2625 kg ha^{-1}. The

Table 2 : *Soil parameters and yield data - Permanent manurial experiment on rice, at the RRS, Kayamkulam*

Treat-ments	pH	EC (μ SM^{-1})	OC (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Avail. N (Kg ha^{-1})	Avail. P (Kg ha^{-1})	Avail. K (Kg ha^{-1})	Clay content (%)	R_2O_3 (%)	Yield (Kg ha^{-1})	
											Grain	Straw
T1	5.05	129.65	1.13	5.40	2.27	154.25	39.7	42.00	6.25	4.70	2625	2813
T2	4.90	150.59	0.93	6.35	2.27	150.00	23.73	61.60	6.75	6.20	900	1038
T3	4.97	110.75	0.83	5.95	2.52	160.75	34.68	53.20	5.75	6.50	1334	1513
T4	4.82	139.72	0.79	5.87	3.03	172.25	18.07	84.00	7.00	6.60	1069	1206
T5	4.92	141.48	0.88	6.30	2.27	156.75	34.69	89.60	5.50	6.20	1163	1281
T6	5.00	150.53	1.04	6.35	2.52	140.25	32.49	106.40	5.00	6.40	1938	2269
T7	5.17	111.52	0.99	6.17	2.02	155.50	59.03	98.00	6.50	5.10	2531	2738
T8	4.97	112.35	0.81	5.50	1.76	85.75	12.84	61.10	6.00	5.90	631	763
SEd	N.S	1.37	0.004	0.17	N.S	9.10	0.52	10.53	N.S	0.18	142.70	145.47
CD (.05)	N.S	4.03	0.14	0.51	N.S	26.76	1.53	31.00	N.S	0.54	420	428

highest grain yield was recorded for T_1 (2625) which was significantly different from all other treatments except T_7 (2531). Treatments T_5 (1163), T_3 (1334) and T_4 (1069) were found to be on a par while the lowest value for grain yield was noticed for T_8 (631) which did not differ significantly from T_2 (900). The experimental plot with the treatment T_1 which recorded the highest value for grain yield, registered comparatively higher values for soil enzyme activites viz., urease (199.90), phosphatase (47.90), protease (240.00), cellulase (10.25) and dehydrogenase (197.79). Treatments T_7 which recorded the highest values for urease, phosphatase and protease recorded a grain yield of 2531 kg ha^{-1} which was significantly superior than other treatments except T_1. Treatments T_4 and T_6 which registered the highest values for cellulase and dehydrogenase activity recorded a grain yield of 1069 kg ha^{-1} and 1938 kg ha^{-1} respectively which were comparatively lower than T_7. The treatment T_8, which recorded lowest value for grain yield was found to have very low activities of urease (139.92), phosphatase (35.39), protease (200.00), cellulase (5.50) and dehydrogenase (145.72) in the soil samples.

4.1.1.12. Straw yield

The data on straw yield revealed a strong statistically significant effect due to treatment influence. The mean values ranged between 763 to 2813 kg ha^{-1} with the highest value noticed for T_1 (2813) which was on a par with treatment T_7 (2738). Treatments T_3 (1513), T_5 (1281) and T_4 (1206) were found to be at par, while the lowest value was observed for T_8 (763), the control which was on a par with T_2 (1038). Enzyme activity values recorded

for soil samples collected from the treatment T_1 were 199.9, 47.9, 240.00, 10.25 and 197.79 for urease, phosphatase, protease, cellulase and dehydrogenase respectively. Treatment T_7 which recorded the highest value for urease, phosphatase and protease recorded a straw yield of 2738 kg ha^{-1} which was significantly superior than all other treatments except T_1. Treatments T_4 and T_6 which registered the highest values for cellulase and dehydrogenase recorded a straw yield of 1206 and 2269 kg ha^{-1} respectively which were comparatively lower than T_7. Treatment T_8 which recorded the lowest straw yield had given very low values for soil enzyme activities also.

4.1.2. Biological characters

It is obvious that the value of soil as a medium for plant growth is conditioned not only by the physicochemical nature, but also by the bio-chemical characteristics in terms of several kinds and amounts of enzymes that are produced and accumulated in soil. Soil samples from the permanent manurial experiments that have been differently fertilized over a period of time were collected and the enzyme activity and respiratory activity were determined for all the samples. The results of the same are presented below in the following pages and in Table 3.

4.1.2.1. Urease activity

The data on urease activity showed statistically significant results on analysis. The mean values for the urease activity ranged from 139.92 to 319.77 ppm of urea hydrolysed g^{-1} of soil hr^{-1}. The highest value was noticed for T_7

(319.77) which was significantly different from all other treatments while the lowest value was recorded for T_8 (139.92). Treatments T_4 (284.6), T_6 (274.85), T_3 (213.22) and T_2 (209.87) were found to be on a par which showed no significant difference on the activity of urease among these treatments. Treatment T_1 (199.9) was found to be at par with the control T_8 (139.92) and thus the addition of cattle manure alone could not produce any significant change in the urease activity.

4.1.2.2. Phosphatase activity

The activity of the enzyme phosphatase was expressed in terms of the quantity of p-nitrophenyl phosphate released g^{-1} of soil hr^{-1} in micrograms. The mean values for phosphatase activity ranged from 35.39 to 97.88 µg of p-nitrophenol released g^{-1} of soil hr^{-1}. The treatments imposed a significant effect with respect to the phosphatase activity. The highest value was recorded for T_7 (97.88) which was significantly superior to all other treatments. The treatments T_1 (47.90), T_2 (48.94) and T_4 (49.57) were on a par showing similar effects on phosphatase activity but was significantly superior to the control (T_8) which showed the lowest value.

4.1.2.3. Protease activity

The activity of protease was expressed in micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}. The data on statistical analysis indicated a highly significant effect due to treatments. The mean values ranged between 200 to 515 micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1} with the highest

Table 3 : *Biological characters - Permanent manurial experiment on rice at the RRS, Kayamkulam*

Treat-ments	Urease activity (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil $hour^{-1}$)	Dehydrogenase activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1})	Respiratory activity (µg of CO_2 evolved g^{-1} of soil hr^{-1})
T1	199.90	47.90	240.00	197.79	10.25	4.90
T2	209.87	48.94	235.00	163.76	16.25	4.60
T3	213.22	63.51	310.00	296.68	15.87	4.50
T4	284.60	49.57	315.00	364.35	19.50	3.80
T5	256.55	60.40	245.00	450.21	12.37	3.60
T6	274.85	70.81	360.00	585.56	16.25	4.30
T7	319.77	97.88	515.00	536.11	16.00	5.00
T8	139.92	35.39	200.00	145.72	5.50	3.50
SEd	21.03	3.74	11.44	25.04	N.S	0.20
CD	63.09	11.01	33.66	73.67	N.S	0.60

value recorded for T_7 (515) which was found to be significantly superior than all other treatments. T_6 (360) was found to be the second best treatment which was also significantly different from others. Treatments T_5 (245), T_1 (240) and T_2 (235) were found to be on a par. In general, the soil samples collected from the treated plots receiving different combinations of N, P and K were found to be significantly superior than the control plots (200) with respect to protease activity.

4.1.2.4. Dehydrogenase activity

The activity of dehydrogenase expressed in soil showed highly significant difference due to treatments on statistical analysis.

The mean values ranged from 145.72 to 585.56 µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was noticed for the treatment T_6 (585.56) which was at par with T_7 (536.1) and was significantly different from all other treatments. The second highest value was recorded for T_5 (450.21). The treatments T_4 (364.35) and T_3 (296.68) were at par but were significantly different from T_1 (197.79), T_2 (163.76) and T_8 (145.72). The lowest value was recorded in the control plot T_8 (145.72) thus indicating a positive effect of treatments with respect to dehydrogenase activity.

4.1.2.4. Cellulase activity

The cellulase activity was expressed in terms of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}. The results revealed no significant effect on the activity of the enzyme due to treatment variations. The mean values for the cellulase activity ranged from 5.5 to 19.5 with the highest value reported for T_4 (19.5).

4.1.2.6. Respiratory activity

The results showed highly significant effect due to treatments with respect to the respiratory activity of the samples collected from the experimental plots.

The mean values ranged from 3.50 to 5.0 μg of CO_2 evolved g^{-1} of soil hr^{-1}. The highest value was observed for T_7 (5.00) which was on a par with T_1 (4.90), T_2 (4.60) and T_3 (4.50). The lowest value was recorded for the control T_8 (3.50) which was at par with T_5 (3.60) and T_4 (3.80).

4.1.3. Correlation analysis

Correlation analysis was carried out between enzyme activities and soil parameters to the study functional relationship between them. The results of the analysis are presented in Table 4.

The results indicated highly significant positive correlations for urease activity with CEC (0.47), available nitrogen (0.54), available phosphorous (0.49) and available potassium (0.49). Positive but non significant correlations were observed for urease activity with pH (0.13), EC (0.15), organic carbon (0.03), AEC (0.09), clay content (0.08), sesquioxides (0.23) and respiratory activity (0.33). None of the soil parameters studied was found to be negatively correlated with the urease activity.

Significant positive correlations for phosphatase activity were also observed with CEC (0.35), available P (0.82), available K (0.39) and respiratory activity (0.42) while pH (0.34), OC (0.15), available N (0.28) and clay content

Table 4 : *Correlation matrix - (Soil Parameters Vs Enzyme activities) at the PME, RRS, Kayamkulam*

	pH	EC	Organic carbon	CEC	AEC	Available N	Available P	Available K	Clay content	Sesqui oxides	Respiratory activity
Urease activity	+0.13	+0.15	+0.03	+0.47**	+0.09	+0.54**	+0.49**	+0.49**	+0.08	+0.23	+0.33
Phosphatase activity	+0.34	-0.16	+0.15	+0.35*	-0.02	+0.28	+0.82**	+0.39*	+0.10	-0.11	+0.42*
Protease activity	+0.28	-0.24	+0.20	+0.25	+0.03	+0.28	+0.72**	+0.46**	-0.03	-0.10	+0.44*
Dehydrogenase activity	+0.12	+0.17	+0.17	+0.50**	+0.15	+0.30	+0.53**	+0.64**	-0.02	+0.15	+0.07
Cellulase activity	-0.17	+0.19	-0.02	+0.50**	+0.32	+0.42*	+0.13	+0.07	-0.01	+0.36*	+0.20

* Significant as 5% level
** Significant at 1% level

r at 5% = 0.35
r at 1% = 0.45

(0.10) showed only a positive and non significant relationship. The correlation observed was negative and non significant with respect to EC (-0.16), AEC (-0.02) and sesquioxides (-0.11) in this case.

Correlation analysis revealed positive and significant correlations for protease activity, with available phosphorous (0.72), available potassium (0.46) and respiratory activity (0.44) while significant negative correlations were observed with soil pH (0.28), organic carbon (0.20), cation exchange capacity (0.25), anion exchange capacity (0.03) and available nitrogen (0.28). The parameters like EC (-0.24), clay content (-0.03) and sesquioxides (-0.10) showed a negative but non significant correlation with the protease activity.

Significant and positive correlation coefficients were obtained in the case of dehydrogenase activity with cation exchange capacity (0.50), available phosphorus (0.53) and available potassium (0.64). Other parameters like pH (0.12), EC (0.17), organic carbon (0.17), anion exchange capacity (0.15), available nitrogen (0.30), sesquioxides (0.15) and respiratory activity (0.07) showed a positive but non significant correlation with dehydrogenase activity while it was non significant and negative with clay content (-0.02).

In the case of cellulase activity significant and positive correlations were obtained with cation exchange capacity (0.50), available nitrogen (0.42) and sesquioxides (0.36) while non significant positive relationship was observed with EC (0.19), anion exchange capacity (0.32), available phosphorus (0.13), available potassium (0.07) and respiratory activity (0.20).

The correlations were negative and non significant for soil pH (-0.17), organic carbon (-0.02) and clay content (-0.01).

4.1.4. Regression analysis

Regression analysis was carried out betwen soil enzyme activities and soil parameters to identify the important soil characteristics responsible for the activity and to delineate the functional relationship between these two factors. The results are presented in Table 5.

Out of the 11 characteristics studied, available nitrogen was found to give a highly significant regression coefficient (+2.40) with urease activity.

Regression analysis between phosphatase activity and soil parameters identified organic carbon (-2.23) and available phosphorus (+6.23) as the major factors related to phosphatase activity of the soils studied.

Protease activity was significantly influenced by available P and available K compared to other factors. The regression coefficients obtained with the enzyme activity were positive and highly significant (+2.71, +2.76) in both cases.

Highly significant regression coefficients were obtained for available N (+2.91), available P (+4.18) and respiratory activity (-2.10) in the case of dehydrogenase activity of the soils studied.

In the case of cellulase activity none of the 11 parameters studied was found to influence the enzyme activity significantly.

Table : 5 *Multiple regression analysis (Soil parameters Vs Enzyme activities) at the PME, RRS, Kayamkulam*

Sl. No.	Parameters	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase
1.	pH	+18.61 (+0.15)	+13.88 (+1.35)	+54.80 (+0.76)	+71.02 (+0.65)	- 6.21 (+0.87)
2.	EC	+0.37 (+0.21)	+0.17 (+1.14)	-0.78 -0.74)	+2.12 (+1.34)	-0.03 (-0.32)
3.	OC	-244.24 (-1.01)	-45.90 (-2.23*)	-136.31 (-0.94)	-20.60 (-0.09)	-1.10 (-0.07)
4.	Clay content	+8.98 (+1.02)	+0.75 (+1.00)	-3.12 (-0.59)	-8.35 (-1.05)	-0.31 (-0.59)
5.	CEC	+26.48 (+0.48)	+1.72 (+0.36)	-8.30 (-0.25)	+24.05 (+0.48)	+3.84 (+1.17)
6.	AEC	-1.25 (-0.04)	+1.05 (+0.41)	+24.24 (+1.36)	+52.79 (+1.97)	+1.67 (+0.95)
7.	Av. N	+1.33 (+2.40)*	-0.12 (-1.62)	-0.19 (-0.36)	-0.30 (+2.91)*	+0.05 (+1.03)
8.	Av. P	+3.95 (+1.50)	+1.43 (+6.23)*	+4.39 (+2.71)*	+10.19 (+4.18)*	+0.02 (+0.14)
9.	Av. K	+1.91 (+1.47)	0.11 (+1.64)	+1.32 (+2.76)*	+2.10 (-0.38)	-0.01 (-0.34)
10.	Respiratory activity	+9.19 (+1.70)	+2.95 (+0.64)	+4.14 (+1.27)	-1.03 (-2.10)*	+4.06 (+1.26)
11.	Sesquioxides	+60.54 (+1.65)	+4.00 (+1.27)	+19.47 (+0.88)	+28.44 (+0.85)	+3.22 (+1.47)
	R2	71.47	85.92	72.88	78.88	51.43

Values in paranthesis represents t - values

t_{20} at 5% - 2.08
at 1% - 2.84

4.2. Permanent manurial experiment on rice (Variety - Jaya) at the RARS, Pattambi

Soil samples were collected from the plots of Permanent manurial experiment at the Regional Agricultural Research Station, Pattambi. The soil samples were then subjected to the analysis of soil parameters and the assay of enzyme activities for the five major soil enzymes viz., urease, protease, phosphatase, cellulase and dehydrogenase. The results of the analysis are presented below (Tables 6 to 9).

4.2.1. Soil parameters

Soil parameters like pH, EC, OC, CEC, AEC, available N, P, K, clay content and sesquioxides were analysed and the results are presented here under.

4.2.1.1. Soil reaction

The results showed a significant effect for treatments on soil pH. The mean values for pH ranged from 4.32 to 5.36.

The highest value was recorded in the treatment T_6 (5.36) which was found to be on a par with treatments T_4 (5.23), T_5 (5.23), T_7 (5.19), T_3 (5.13) and T_8 (5.09) while the lowest value was recorded for T_1 (4.32) which was on a par with T_2 (4.69).

4.2.1.2. Electrical conductivity

The analysis of samples for EC did not give any significant effect on specific conductance of the experimetal site. The mean values for EC ranged from 99.91 to 133.62 μSM^{-1} with the highest value registered for T_4 (133.62) which did not differ significantly from other treatments.

4.2.1.3. Organic carbon

The treatments did not show any significant effect on the organic carbon status of the soil collected from the plots. The mean values for organic carbon ranged from 0.61 to 0.91 percent. The highest value for organic carbon was recorded for T_7 (0.91) which did not differ significantly from other treatments.

4.2.1.4. Cation exchange capacity

The treatments were found to impose significant effect on CEC and the mean values ranged from 9.27 to 14.72 c mol (p^+) kg^{-1}. The highest value was recorded for T_7 (14.72) which was found to be on a par with T_5 (13.55) and T_6 (13.20) while the treatments T_2 (11.77), T_3 (12.65), T_4 (12.42) and T_6 (13.2) were also at par. The lowest value was recorded for the treatment T_8 (9.27).

4.2.1.5. Anion exchange capacity

The results indicated significant effect due to treatments on the anion exchange capacity of the samples. The mean values for AEC ranged from 1.68 to 2.70 c mol (e^{-1}) kg^{-1} of soil. The highest value for AEC was noticed for

T_7 (2.70) which was found to be on a par with the treatments T_3 (2.54), T_1 (2.51) and T_2 (2.42). Treatments T_6 (2.20), T_4 (2.17) and T_5 (2.10) were also found to be at par while the lowest value was recorded for T_8 (1.68).

4.2.1.6. Available nitrogen

Significant variation in the available nitrogen status due to treatments were observed. The mean values for available nitrogen ranged from 128.92 to 161.06 kg ha^{-1}. The highest value was recorded for T_7 (161.06) which was significantly superior to all other treatments. Treatments T_8 (153.23) and T_6 (147.34) were found to be at par. Similarly treatments T_3 (142.97), T_4 (140.70), T_5 (140.30) and T_2 (138.73) were also at par. The lowest value for available N was recorded for T_1 (128.92) which was significantly lower than all other treatments.

4.2.1.7. Available phosphorus

The effect of the treatments were found to be significant with mean values for available phosphorus ranging from 57.34 to 105.72 kg ha^{-1}.

The highest value was recorded for T_7 (105.72) and was found to be significantly superior to all other treatments. Treatments T_8 (92.28) and T_6 (86.81) were found to be at par. Treatments T_4 (75.16), T_5 (74.27) and T_3 (67.19) were also on a par. The lowest value for available phosphorus was recorded for T_1 (57.34).

4.2.1.8. Available potassium

From the results, it was found that the treatments imposed a significant effect on available potassium content of the soil. The mean values for available potassium ranged between 61.62 to 201.60 kg ha^{-1}.

The highest value for available potassium was recorded for T_8 (201.6) which was significantly superior to all other treatments. Treatments T_7 (168) and T_6 (165.2) were found to be on a par. Similarly treatments T_5 (137.3), T_4 (134.4) and T_2 (134.4) were also at par. The lowest value was however noticed in treatment T_1 (61.62) which was significantly lower than other treatments.

4.2.1.9. Clay content

From the analysis of the soil samples, it was noticed that there was no significant influence of the treatments on the clay content. The mean values for the clay content ranged between 22.00 to 25.50 percent. The highest value for the clay content was noticed for the treatment T_8 (25.50) which did not differ significantly from other treatments viz., T_1 (24.00), T_3 (23.00), T_4 (22.00), T_5 (21.00), T_6 (22.00), T_7 (23.00) and T_8 (24.50).

4.2.1.10. Sesquioxides

The results revealed that the treatments vary significantly with respect to the sesquioxide content of the samples and the mean values ranged between 20.32 and 26.57 percent. The sesquioxide content was found to be maximum for T_8 (26.57) followed by T_4 (25.57), T_7 (24.55) and T_5 (24.10)

Table : 6 *Soil Parameters and yield data - P M E on rice (Var. Jaya), at the RARS, Pattambi*

Treat-ments	pH	Electrical conductivity (μSM^{-1})	Organic carbon (%)	Cation Exchange Capacity (c mol (p+) kg^{-1} of soil)	Anion Exchange Capacity (c mol(e^-) kg^{-1} of soil)	Available Nitrogen (Kg ha^{-1})	Available Phosphorus (Kg ha^{-1})	Available Potassium (Kg ha^{-1})	Clay content (%)	Sesqui-oxides (%)	Yield (Kg ha^{-1})	
											Grain	Straw
T1	4.32	102.93	0.61	11.15	2.51	128.92	57.34	61.62	24.0	21.17	3317	4015
T2	4.69	105.39	0.75	11.77	2.42	138.73	60.92	134.40	25.50	20.32	2827	3391
T3	5.13	108.53	0.64	12.65	2.54	142.97	67.19	100.80	23.00	22.12	2994	3992
T4	5.23	133.62	0.73	12.42	2.17	140.70	75.16	134.40	22.00	25.57	2554	3175
T5	5.23	100.4	0.74	13.55	2.10	140.30	74.27	137.30	25.00	24.10	3312	4068
T6	5.36	108.62	0.83	13.20	2.20	147.34	86.81	165.20	22.00	22.85	2718	3533
T7	5.19	99.91	0.91	14.72	2.70	161.06	105.72	168.0	23.00	24.55	3457	4494
T8	5.09	110.22	0.64	9.27	1.68	153.23	92.28	201.60	24.50	26.57	3020	3624
SEd	0.18	N.S	N.S	0.54	0.11	2.11	3.04	8.79	N.S	1.09	36.69	42.21
CD	0.55	N.S	N.S	1.59	0.33	6.22	8.95	25.87	N.S	3.27	110.07	126.63

which were found to be on a par. The lowest value was recorded for T_2 (20.32) which was statistically on a par with T_1 (21.17), T_3 (22.12) and T_6 (22.85).

4.2.1.11. Grain yield

The treatments had a significant effect on the grain yield with the mean values ranging between 2554 to 3457 kg ha^{-1}. The highest value was noticed for T_7 (3457) which was significantly superior than other treatments. Treatments T_1 (3317) and T_5 (3312) were found to be on a par while the lowest value was registered for T_4 (2554). Treatment T_7, which recorded the highest value for grain yield was reported to have higher activities of urease (359.10), phosphatase (40.00), protease (520.00), dehydrogenase (546.41) and cellulase (28.37) compared to other treatments. The treatment, T_4, which recorded the lowest value for grain yield had shown activities of urease, phosphatase, protease, dehydrogenase and cellulase in the order 269.32, 31.75, 455.00, 310.63 and 15.75 respectively.

4.2.1.12. Straw yield

From the results, it was inferred that there was significant difference between the treatments with respect to the straw yield. The mean values ranged between 3175 to 4494 kg ha^{-1}. The highest value was noticed for T_7 (4494) which was significantly superior than other treatments. Treatments T_1 (4015), T_5 (4068), T_8 (3624) and T_6 (3533) were found to be on a par while the treatment T_4 (3175) recorded the lowest value. The activities of urease, phosphatase, protease, dehydrogenase and cellulase have registered values in the order of

359.10, 40.00, 520.00, 546.41 and 28.37 respectively for T_7 which recorded the highest straw yield. Similarly T_4 recorded values 269.32, 31.75, 455.00, 310.63 and 15.75 for urease, phosphatase, protease, dehydrogenase and cellulase activity respectively.

4.2.2. Biological characters

The activity assay was carried out for the enzymes, viz., urease, phosphatase, protease, dehydrogenase and cellulase. Besides, the respiratory activity of the soil samples was also assessed and the results are presented in Table 7.

4.2.2.1. Urease activity

The activity assay of urease revealed a significant effect due to treatments. The mean values for the urease activity ranged from 143.05 to 359.1 ppm of urea hydrolysed g^{-1} of soil hr^{-1}.

The highest value for urease activity was noticed for T_7 (359.1) which was found to be on a par with treatment T_5 (339.15) and were significantly superior than other treatments. Similarly treatments T_6 (282.62), T_4 (269.32) and T_8 (269.32) were also found to be on a par and were significantly different from T_3 (222.77), T_2 (179.5) and T_1 (143.05).

4.2.2.2. Phosphatase activity

The results of the phosphatase activity did not show any significant

difference due to treatments. The mean values, for the phosphatase activity ranged from 26.75 to 40 µg of p-nitrophenyl phosphate released g^{-1} of soil hr^{-1}. The highest value was recorded for T_7 (40) which did not differ significantly from other treatments T_3 (35.5), T_8 (34), T_4 (31.75), T_6 (30.5), T_5 (30), T_2 (30) and T_1 (26.75).

4.2.2.3. Protease activity

It was observed from the results that the treatments imposed significant effect with respect to protease activity. The mean values for the enzyme ranged between 265 to 520 micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}. The highest value was recorded for T_7 (520) which was on a par with the treatments T_6 (475), T_4 (455) and T_5 (450) while the lowest value was recorded for T_1 (265) which was at par with the treatment T_2 (280).

4.2.2.4. Dehydrogenase activity

The treatments expressed a significant effect with respect to dehydrogenase activity. The mean values for the dehydrogenase enzyme ranged between 202.99 to 546.41 µg of TPF hydrolysed per gram of soil 24 hrs^{-1}.

The highest value was recorded for T_7 (546.41) which was significantly superior than other treatments. Treatments T_4 (310.63), T_1 (268.05), T_3 (257.65) and T_2 (255.14) were found to be on a par while the lowest value was recorded for T_8 (202.99).

4.2.2.5. Cellulase activity

Table 7 : *Biological characters - PME on rice (Var. Jaya), at the RARS, Pattambi*

Treat-ments	Urease activity (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenol released g^{-1} of hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase activity (µg of TPF hydro-lysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Respiratory activity (µg of CO_2 evolved g^{-1} of soil hr^{-1})
T1	143.05	26.75	265.00	268.05	11.12	3.90
T2	179.55	30.00	280.00	255.14	13.00	4.70
T3	222.77	35.50	335.00	257.65	15.75	4.50
T4	269.32	31.75	455.00	310.63	15.75	4.80
T5	339.15	30.00	450.00	455.43	27.00	4.60
T6	282.62	30.50	475.00	491.87	29.62	5.00
T7	359.10	40.00	520.00	546.41	28.37	5.10
T8	269.32	34.00	390.00	202.99	21.87	3.80
SEd	9.08	N.S	26.03	9.54	2.39	0.06
CD	26.71	N.S	76.59	28.06	7.04	0.19

The results of the cellulase activity assay indicated that the treatments vary significantly with respect to cellulase activity. The mean values ranged from 11.12 to 29.62 ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1}. The highest value for cellulase was observed for T_6 (29.62) which was found to be on a par with the treatments T_7 (28.37) and T_5 (27.0) and was superior to all other treatments. The lowest value was recorded for T_1 (11.12) and was found to be on a par with T_2 (13), T_4 (15.75) and T_3 (15.75).

4.2.2.6. Respiratory activity

The respiratory activity of the soil evaluated in terms of micrograms of CO_2 evolved g^{-1} of soil hr^{-1} indicated a significant effect due to treatments. The mean values ranged from 3.80 to 5.10 μg of CO_2 g^{-1} of soil hr^{-1}.

The highest value was recorded for T_7 (5.10) which was found to be statistically on a par with T_6 (5.0) and was significantly superior than all other treatments. Treatments T_2 (4.70), T_4 (4.80), and T_5 (4.60) were found to be at par while the lowest value was recorded in T_8 (3.80) which was found to be at par with T_1 (3.90).

4.2.3. Correlation analysis

The activities of urease, phosphatase, protease, dehydrogenase and cellulase were subjected to correlation analysis to assess the level of association and relationship with the soil parameters like pH, EC, organic carbon, clay content, CEC, AEC, available N, available P, available K, respiratory activity

Table 8 : *Correlation Matrix - (Soil parameters Vs Enzyme activities) - PME on rice (Var. Jaya) at the RARS, Pattambi*

Sl. No.	Enzyme activity	pH	EC	Organic Carbon	Clay Content	CEC	AEC	Available N	Available P	Available K	Respiratory activity	Sesqui oxides
1.	Urease activity	0.61**	-0.07	0.28	-0.19	0.49**	-0.19	0.69**	0.76**	0.59**	0.53**	0.57**
2.	Phosphatase activity	0.11	0.13	-0.09	-0.09	0.35*	0.12	0.46**	0.32	0.16	0.14	0.22
3.	Protease activity	0.60**	0.10	0.37*	-0.13	0.41**	-0.14	0.55**	0.71**	0.50**	0.57**	0.41**
4.	Dehydrogenase activity	0.41*	-0.22	0.43*	-0.50**	0.70**	0.23	0.42*	0.54**	0.25	0.75**	0.20
5.	Cellulase activity	0.43**	-0.16	0.24	-0.24	0.40**	-0.23	0.59**	0.61**	0.52**	0.43*	0.51**

t at 5% - 0.34 * Significant at 5% level
at 1% - 0.44 ** Significant at 1% level

and sesquioxide content. The results of the study are presented in Table 8.

Correlation studies revealed a highly significant and positive correlation between urease activity and various parameters like pH (0.61), CEC (0.49), available N (0.69) available P (0.76), available K (0.59), respiratory activity (0.53) and sequioxides (0.57), but the correlation was positive and non significant between urease activity and organic carbon (0.28). The correlation of urease activity with EC (-0.07), clay content (-0.19) and AEC (-0.19) were found to be negative and non significant.

Significant and positive correlations were observed in the case of phosphatase activity with the soil parameters like CEC (0.35) and available N (0.46). A positive and non significant correlation was observed for phosphatase activity with pH (0.11), EC (0.13), AEC (0.12), available P (0.32), available K (0.16), respiratory activity (0.14) and sesquioxides (0.22) while the correlations were negative and non significant in the case of OC (-0.09) and clay content (-0.09).

The study revealed significant and positive correlations for protease activity with pH (0.60), organic carbon (0.37), CEC (0.41), available N (0.55), available P (0.71), available K (0.50), respiratory activity (0.57) and sesquioxides (0.41). A positive but non significant correlation was noted between protease activity and EC (0.10) while correlations with clay content (-0.13) and AEC (-0.14) were found to be negative and non significant.

Positive and significant correlations, for dehydrogenase activity with pH (0.41), organic carbon (0.43), CEC (0.70), available N (0.42), available P

(0.54) and respiratory activity (0.75) were observed in the study. The correlation between dehydrogenase activity and clay content (-0.50) was found to be significant but negative while the correlations were positive and non significant with AEC (0.23), available K (0.25) and sesquioxides (0.20). Similarly the correlation was non significant and negative in the case of EC (-0.22).

The results of the correlation analysis between cellulase activity and soil parameters like pH (0.43), CEC (0.40), available N (0.59), available P (0.61) available K (0.52), respiratory activity (0.43) and sesquioxides (0.51) were found to be positive and significant. However, the correlations were positive but non significant with respect to organic carbon (0.24) and negative and non significant with EC (-0.16), clay content (-0.24) and AEC (-0.23).

4.2.4. Regression analysis

Multiple regression analysis was carried out to study the functional relationship between enzyme activity and soil parameters. The results of the analysis are presented in Table 9.

The results revealed the association of CEC (+2.37) and AEC (-2.32) as the two important factors significantly related to urease activity. The contributions by pH, EC, organic carbon, available N, P, K, clay content, sesquioxides and respiratory activity were non significant.

In the case of phosphatase activity, none of the soil factors studied were significantly associated with the activity though the contribution was positive for certain parameters.

Table : 9 *Multiple regression analysis (Soil parameters Vs Enzyme activities) - PME on rice (Var. Jaya) at the RARS, Pattambi*

Sl. No.	*Parameters*	*Urease*	*Phosphatase*	*Protease*	*Dehydro-genase*	*Cellulase*
1.	pH	+15.64 (+0.80)	-0.31 (-0.07)	+34.20 (+1.03)	-12.86 (-0.39)	-2.09 (-0.66)
2.	EC	-0.26 (-0.74)	+0.06 (+0.84)	+0.49 (+0.81)	-1.49 (-2.48*)	-0.05 (-0.95)
3.	OC	-12.27 (-0.28)	-12.13 (-1.22)	+67.02 (+0.90)	+31.97 (+0.44)	-2.21 (-0.31)
4.	Clay content	-0.31 (-0.04)	-0.13 (-0.19)	+2.70 (+0.52)	-4.95 (-0.98)	-0.54 (-1.11)
5.	CEC	+12.77 (+2.37*)	+1.74 (+1.44)	+5.15 (+0.56)	+25.19 (+2.82*)	+1.30 (+1.47)
6.	AEC	-62.08 (-2.32*)	+0.87 (+0.14)	-68.45 (-1.51)	-37.51 (-0.84)	-9.46 (-2.18*)
7.	Av. N	+0.11 (+0.08)	+0.58 (+2.00)	-1.62 (-0.74)	-4.40 (-2.05)	-0.01 (+0.002)
8.	Av. P	+1.60 (+1.95)	+0.05 (+0.31)	+3.45 (+2.47*)	+4.13 (+3.02**)	+0.09 (+0.70)
9.	Av. K	-0.09 (-0.30)	-0.01 (-0.08)	-0.70 (-1.36)	-0.21 (-0.42)	+0.02 (+0.51)
10.	Respiratory activity	+4.65 (+1.77)	-5.51 (-0.93)	+9.70 (+2.18*)	+1.20 (+2.76*)	+5.17 (+1.21)
11.	Sesquioxides	+6.98 (+1.42)	-1.03 (-0.93)	+10.67 (+1.28)	+6.61 (+0.75)	+0.80 (+1.01)
	R2	83.42	41.34	75.27	84.70	66.11

Values in paranthesis represents t - values

t_{20} at 5% - 2.08
at 1% - 2.84

The regression analysis between protease activity and soil parameters identified available P (+2.47) and respiratory activity (+2.18) as the two most important soil factors contributing significantly to the activity in the soil. The relationship between protease activity and other parameters were found to be non significant and ineffective.

In the case of dehydrogenase, significant and positive correlation were expressed through factors like EC (-2.48), CEC (+2.82), available P (+3.02) and respiratory activity (+2.76). Factors like pH, OC, AEC, available N, available K, clay content and sesquioxides did not contribute significantly towards the activity of this enzyme.

With respect to cellulase activity, out of the 11 soil parameters studied, only the anion exchange capacity (-2.18) was found to be significantly related to the activity. Though the effect of some other soil factors were found to be positive, their role and relative contribution towards the activity was non significant and meagre.

4.3. Permanent manurial experiment on rice (Variety : Jyothi) at the RARS, Pattambi

Soil samples collected from the old PME on rice were subjected to the analysis of soil parameters and enzyme activity. The results of the analysis are presented below and in Tables 10 to 13.

4.3.1. Soil parameters

4.3.1.1. Soil reaction

The results revealed variations in pH due to treatment effects. The mean values ranged between 5.10 to 5.72 while the highest value was recorded for T_2 (5.72) which was significantly superior compared to other treatments. The lowest value was noticed for treatment T_6 (5.10) which was at par with T_3 (5.13), T_1 (5.13) and T_4 (5.17).

4.3.1.2. Electrical conductivity

The results presented showed that the EC values vary significantly with variations in treatments. The mean values for electrical conductivity ranged between 92.94 to 110.47 μSM^{-1} with the highest value recorded for T_4 (110.47) which was on par with T_8 (108.39) and T_2 (107.53) while the lowest value was noticed for T_1 (92.94). Treatments T_6 (97.19) and T_7 (96.60) were found to be at par.

4.3.1.3. Organic carbon

The treatments did not vary significantly with respect to organic carbon status of the experimental sites. The mean values ranged between 0.38 to 0.57 per cent and the highest value was recorded for T_7 (0.57) which did not significantly differ from other treatments viz., T_6 (0.54), T_5 (0.48), T_2 (0.47), T_1 (0.43), T_3 (0.42), T_4 (0.41) and T_8 (0.38).

4.3.1.4. Cation exchange capacity

Significant variations with respect to treatments in the CEC were observed from the analysis of the samples. The mean values ranged between 10.55 to 12.9 c mol (p+) kg^{-1} with the highest value recorded for T_7 (12.9) which was significantly different from other treatments. Treatments T_6 (12.35), T_4 (12.27), T_3 (12.22), T_1 (11.92) and T_5 (11.92) were found to be on a par while the lowest value was recorded for T_8 (10.55).

4.3.1.5. Anion exchange capacity

The results revealed no significant difference with respect to the anion exchange capacity of the soil. The mean values ranged between 1.78 to 2.35 c mol (e^-) kg^{-1} with the highest value recorded for T_7 (2.33).

4.3.1.6. Available nitrogen

Analysis of the data revealed no significant difference among the treatments with respect to the available nitrogen status.

The mean values ranged between 121.02 to 144.14 kg ha^{-1} with the highest value recorded for T_7 (144.14) which did not differ significantly from the other treatments viz., T_1 (136.65), T_8 (135.56), T_6 (130.00), T_3 (124.14), T_4 (124.13), T_5 (122.58) and T_2 (121.02).

4.3.1.7. Available phosphorus

The results showed no significant difference between the treatments

with respect to the available phosphorus. The mean values ranged between 51.37 to 85.11 kg ha^{-1} with the highest value recorded for T_8 (85.11) which did not differ significantly from other treatments viz., T_6 (84.22), T_7 (76.78), T_4 (73.47), T_5 (72.57), T_3 (65.31), T_2 (59.11) and T_1 (51.37).

4.3.1.8. Available potassium

The results showed that the treatments varied significantly with respect to the available potassium status. The mean values ranged between 70 to 154 kg ha^{-1}. The highest value was recorded for T_8 (154) which was found to be on a par with T_7 (140). The treatments T_6 (134.4) and T_7 (140) were found to be at par while the lowest value was recorded for T_1 (70).

4.3.1.9. Clay content

The analytical results showed no significant effect for the treatments on clay content.

The mean values ranged between 15.25 to 17.75 per cent. However the highest value was recorded for T_1 (17.75) which did not differ significantly from other treatments viz., T_6 (17.25), T_7 (17.25), T_2 (17.00), T_8 (16.75), T_4 (16.00), T_5 (15.50) and T_3 (15.25).

4.3.1.10. Sesquioxides

The treatments imposed significant effect with respect to sesquioxide content of the samples. The mean values ranged between 19.72 to 25.72 per

Table 10 : *Soil parameters and yield data of the PME on rice (Var. Jyothi), at RARS, Pattambi*

Treat-ments	pH	EC (μ SM^{-1})	OC (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Avail. N (Kg ha^{-1})	Avail. P (Kg ha^{-1})	Avail. K (Kg ha^{-1})	Clay content (%)	R_2O_3 (%)	Yield (Kg ha^{-1})	
											Grain	Straw
T_1	5.13	92.94	0.43	11.92	2.25	136.65	51.37	70.00	17.75	20.07	3314	4013
T_2	5.72	107.53	0.47	11.60	1.88	121.02	59.11	112.00	17.00	19.72	2717	3260
T_3	5.13	106.75	0.42	12.22	2.02	124.14	65.31	89.60	15.25	21.31	2885	3490
T_4	5.17	110.47	0.41	12.27	1.83	124.13	73.47	123.20	16.00	24.37	2569	3490
T_5	5.35	101.00	0.48	11.92	1.91	122.58	72.57	106.40	15.50	23.60	3268	4019
T_6	5.10	97.19	0.54	12.35	2.29	130.00	84.22	134.40	17.25	21.73	2629	3128
T_7	5.67	96.60	0.57	12.90	2.33	144.14	76.78	140.00	17.25	23.45	3472	4161
T_8	5.35	108.39	0.38	10.55	1.78	135.56	85.11	154.00	16.75	25.72	3015	3827
SEd	0.04	1.03	N.S	0.16	N.S	N.S	N.S	5.82	N.S	1.03	35.19	41.27
CD	0.12	3.05	N.S	0.48	N.S	N.S	N.S	17.14	N.S	3.21	105.57	123.81

cent. The highest value was observed for T_8 (25.72) which was on a par with T_4 (24.37), T_5 (23.60) and T_7 (23.45) while the lowest value was recorded for T_2 (19.72) which was also at par with T_1 (20.07) and T_3 (21.31).

4.3.1.11. Grain yield

The treatments imparted a significant effect on the grain yield. The mean values ranged between 2569 to 3472 kg ha^{-1}. The highest value was recorded for T_7 (3472) which was significantly superior than other treatments. Treatments T_5 (3268) and T_1 (3314) were found to be on a par. T_4 (2569) recorded the lowest value for grain yield which was at par with T_6 (2629) and was significantly inferior to all other treatments. Treatment T_7 which recorded the highest value for grain yield was characterised by higher levels for activity for urease (405.65), phosphatase (35.25), protease (550.00), dehydrogenase (580.35) and cellulase (27.25). Treatment T_4 which recorded the lowest value for grain yield registered comparatively lower values for all enzymes in soil [urease (279.30), phosphatase (28.00), protease (400.00), dehydrogenase (333.49) and cellulase (15.75)].

4.3.1.12. Straw yield

The treatments varied significantly with respect to straw yield. The mean value ranged between 3057 to 4166 kg ha^{-1}. The highest value was recorded for T_7 (4166) which was significantly superior to all other treatments. Treatments T_1 (4013) and T_5 (4019) were observed to be on a par. Similarly treatments T_4 (3057) and T_6 (3128) were also at par though the former recorded

the lowest value for straw yield. Treatment T_7, which recorded the highest value also registered higher values for urease, phosphatase, protease, dehydrogenase and cellulase activity. (viz., 405.65, 35.25, 550.00, 580.35 and 27.25 respectively).

The treatment T_4 which recorded the lowest value for straw yield had shown lower values for enzyme activities in the order urease (279.30), phosphatase (28.00), protease (400.00), dehydrogenase (333.49) and cellulase (15.75).

4.3.2. Biological properties

Soil samples collected were then subjected to the enzyme activity assay and assessment of respiratory activity. The results of the assay are presented below in Table 11.

4.3.2.1. Urease activity

The results of the urease activity revealed significant variations in the activity with respect to treatment effects. The mean values ranged between 139.72 to 405.65 ppm of glucose hydrolysed g^{-1} of soil hr^{-1}. The highest value was recorded for T_7 (405.65) which was significantly superior than other treatments while the lowest value was observed for T_1 (139.72). Treatments T_8 (299.25) and T_4 (279.30) were found to be on a par.

4.3.2.2. Phosphatase activity

The results revealed that the activity vary significantly due to treatment

Table 11 : *Biological characters - PME on rice (Var. Jyothi) at the RARS, Pattambi*

Sl. No.	Treat-ments	Urease activity (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenol released g^{-1} of hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenaces activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Respiratory activity (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	T_1	139.72	26.20	245.00	257.72	10.37	4.00
2.	T_2	192.85	25.50	310.00	223.81	13.00	4.40
3.	T_3	246.05	34.30	345.00	239.43	15.37	5.30
4.	T_4	279.30	28.00	400.00	333.49	15.75	5.30
5.	T_5	359.10	18.95	450.00	476.11	27.00	5.10
6.	T_6	339.15	31.25	415.00	484.06	22.62	6.10
7.	T_7	405.65	35.25	550.00	580.35	27.25	5.40
8.	T_8	229.25	38.25	360.00	223.82	21.75	3.90
	SE	8.65	2.18	21.08	9.60	0.68	0.60
	CD	25.44	6.43	62.02	28.25	2.01	1.8

effects. The mean values ranged between 18.95 to 38.25 µg of p-nitrophenyl PO_4 released g^{-1} of soil hr^{-1}. The highest value was recorded for T_8 (38.25) which was found to be at par with treatments T_7 (35.25) and T_3 (34.30) while the lowest value was recorded for T_5 (18.95).

4.3.2.3. Protease activity

The analytical data revealed a highly significant difference in the protease activity. The mean values ranged between 245 to 550 micromoles of amino N hydrolysed g^{-1} of soil hr^{-1}. The highest value was recorded for T_7 (550) which was superior to all other treatments while the treatments T_5 (450), T_6 (415) and T_4 (400) were found to be on a par. Similarly treatments T_2 (310), T_3 (340) and T_8 (360) were also found to be at par and superior to T_1 (245) which recorded the lowest value.

4.3.2.4. Dehydrogenase activity

The results of the dehydrogenase activity showed significant variations due to treatment effects. The mean values for the treatments varied between 223.81 to 580.35 µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded for T_7 (580.35) which was significantly superior to all other treatments. Treatments T_6 (484.06) and T_5 (476.11), T_3 (239.43) and T_1 (257.72) were on a par while the lowest value was recorded for T_2 (223.81) which was statistically at par with T_8 (223.82) and T_3 (239.43).

4.3.2.5. Cellulase activity

The results of the cellulase activity also showed significant difference

due to treatment effects. The mean values ranged between 10.37 to 27.25 ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded for T_7 (27.25) which was on a par with T_5 (27) while the treatments T_4 (15.75) and T_3 (15.37) were also at par. Similarly treatments T_6 (22.62) and T_8 (21.75) were found to be on a par statistically though it was significantly superior to T_1 (10.37) which recorded the lowest value.

4.3.3. Respiratory activity

The results of the assessment of respiratory activity indicated significant difference due to treatment application.

The mean values ranged between 3.90 to 7.70 µg of CO_2 evolved g^{-1} of soil hr^{-1}. The highest value was noticed for T_3 (7.70) which was on a par with the treatment T_6 (6.10) while the lowest value was recorded for T_8 (3.90) which was statistically on par with T_1 (4.00), T_2 (4.40), T_5 (5.10), T_4 (5.30) and T_7 (5.40).

4.3.4. Correlation analysis

Correlation analysis was carried out between soil parameters and enzyme activities. The results of such analysis are presented below in Table 12.

Correlation studies conducted to explain the functional relationship indicated positive and significant correlations for urease activity with available P (0.83), available K (0.64) and sesquioxides (0.56). Similarly positive but

non significant correlations were observed with pH (0.22), organic carbon (0.34), CEC (0.30), AEC (0.10) available nitrogen (0.16) and respiratory activity (0.23). The remaining parameters, EC (-0.07) and clay content (-0.06) showed a non significant negative correlation with urease activity.

In the case of phosphatase activity, a positive and significant correlation was observed with available K (0.44). Similarly positive but non significant correlations were observed between EC (0.08), available N (0.27), available P (0.40), clay content (0.15), respiratory activity (0.09) and sesquioxides (0.11). The correlation with pH (-0.03), organic carbon (-0.02), CEC (-0.10) and AEC (-0.009) were found to be negative and non significant.

Positive significant correlations were observed in the case of protease activity with CEC (0.38), available P (0.75), available K (0.54) and sesquioxides (0.45) while the correlations were positive and non significant with pH (0.33), organic carbon (0.29), AEC (0.14), available N (0.15), clay content (0.29) and respiratory activity (0.25). A negative and non significant correlation was observed between protease activity and EC (-0.11).

Correlations observed were positive and significant for dehydrogenase activity with organic carbon (0.52), CEC (0.60) and available P (0.67). However, positive but non significant correlations were observed with pH (0.14), AEC (0.24) available N (0.22), available K (0.33), clay content (0.02), respiratory acitivity (0.16) and sesquioxides (0.26) in the study. A negative and significant correlation was observed between dehydrogenase activity and EC (-0.50).

Positive and significant correlations were obtained for cellulase activity

Table 12 : *Correlation Matrix - (Soil parameters Vs Enzyme activities) - PME on rice (Var. Jyothi) at the RARS, Pattambi*

Sl. No.	Enzyme activity	pH	EC	Organic Carbon	Clay Content	CEC	AEC	Available N	Available P	Available K	Respiratory activity	Sesqui oxides
1.	Urease activity	+0.22	-0.07	+0.34	-0.06	+0.30	+0.10	+0.16	+0.83**	+0.64**	+0.23	+0.56**
2.	Phosphatase activity	-0.03	+0.08	-0.02	+0.15	-0.10	-0.009	+0.27	+0.40	+0.44**	+0.09	+0.11
3.	Protease activity	+0.33	-0.11	+0.29	+0.29	+0.38*	+0.14	+0.15	+0.75**	+0.54**	+0.25	+0.45**
4.	Dehydrogenase activity	+0.14	-0.50*	+0.52**	+0.02	+0.60**	+0.24	+0.22	+0.67**	+0.33	+0.16	+0.26
5.	Cellulase activity	+0.25	-0.17	+0.30	-0.007	+0.19	+0.05	+0.18	+0.78**	+0.57**	+0.15	+0.49**

t_{30} at 5% - 0.34 * Significant at 5% level
at 1% - 0.44 ** Significant at 1% level

with available P (0.78), available K (0.57) and sesquioxides (0.49). Similarly the correlations observed were positive and non significant with soil pH (0.25), organic carbon (0.30), CEC (0.19), AEC (0.05), available N (0.18) and respiratory activity (0.15). Negative and non significant correlations were also obtained for cellulase activity with EC (-0.17) and clay content (-0.007).

4.3.4. Regression analysis

Regression analysis was carried out between enzyme activities and soil parameters to find out the relative importance of the soil characteristics in deciding the enzyme activities in soil. The results of the regression analysis are presented below in Table 13.

Of the various parameters studied, the content of sesquioxides (+2.21) was found to be the most significant single factor contributing maximum for the urease activity.

None of the eleven factors studied was found to be significantly associated with phosphatase activity in the experiment.

Of the factors analysed, pH (+2.44) and sesquioxides (+2.26) were found to be most significant factors related to the protease activity.

Soil parameters *viz*. EC (-3.78), CEC (+3.75) and available K (+2.29) were found to be the significant and dominant characteristics influencing the dehydrogenase activity in the soil studied. In the case of cellulase, none of the factors studied was found to influence significantly the activity as evident from the low coefficients obtained.

Table 13 : *Multiple regression analysis (Soil parameters Vs Enzyme activities) - PME on rice (Var. Jyothi) at the RARS, Pattambi*

Sl. No.	Parameters	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase
1.	pH	+50.17 (+1.16)	-11.44 (-1.79)	+132.06 (+2.44*)	+66.66 (+1.10)	+5.16 (+1.34)
2.	EC	-2.55 (-1.09)	+0.38 (+1.11)	-3.90 (-1.33)	-12.34 (-3.78**)	-0.33 (-1.60)
3.	OC	+69.96 (+0.66)	+7.47 (+0.47)	-31.87 (-0.24)	+103.23 (+0.69)	+1.72 (+0.18)
4.	Clay content	-0.59 (-0.10)	+1.02 (+1.17)	+0.82 (+0.11)	-10.78 (-1.31)	+0.12 (+0.23)
5.	CEC	+25.83 (+1.32)	-3.25 (-1.12)	+42.91 (+1.75)	+2.53 (+3.75**)	-0.47 (-0.27)
6.	AEC	+1.83 (+0.07)	-2.64 (-0.76)	+13.34 (+0.45)	-18.98 (-0.58)	-1.05 (-0.51)
7.	Av. N	-0.22 (-0.31)	+0.15 (+1.47)	-0.46 (-0.52)	-0.31 (-0.32)	-0.03 (-0.54)
8.	Av. P	+1.07 (+0.68)	+0.35 (+1.51)	+0.35 (+0.18)	-0.77 (-0.34)	+0.18 (+1.31)
9.	Av. K	+1.12 (+1.38)	-0.01 (-0.04)	+1.21 (+1.81)	+2.62 (+2.29*)	+0.02 (+0.32)
10.	Respiratory activity	+1.03 (+1.37)	+9.07 (+0.81)	+1.47 (+1.56)	+2.31 (+0.22)	+8.49 (+1.26)
11.	Sesquioxides	+14.00 (+2.21*)	-1.55 (-1.66)	+17.91 (+2.26*)	+14.84 (+1.68)	+0.77 (+1.38)
	R^2	79.54	44.07	74.37	84.21	69.52

Values in paranthesis represents t - Values

t_{20} at 5% - 2.08
at 1% - 2.84

4.4. Spacing cum intercropping experiment in rubber at the CES, Chethackal

Soil samples were collected from the ongoing spacing cum intercropping experiment on rubber at the CES, Chethackal and were subjected to the analysis of soil parameters and enzyme activities for five major soil enzymes viz., urease, protease, phosphatase, cellulase and dehydrogenase. The results are presented here under in Tables 14 to 17.

4.4.1. Soil parameters

4.4.1.1. Soil reaction

From the results it was observed that the treatments had a significant effect with respect to soil pH. The mean values for soil pH ranged between 4.21 to 4.53. The highest value was recorded for T_6 (4.53) which was on a par with T_4 (4.37) while the lowest value was observed for T_5 (4.21) which was found to be at par with T_3 (4.29), T_2 (4.29) and T_1 (4.30).

4.4.1.2. Electrical conductivity

The results of the analysis of specific conductance indicated a significant effect of treatments on the EC. The mean values ranged between 117.23 to 129.48 μSM^{-1}. The highest value was recorded for T_6 (129.48) which was on a par with T_1 (127.61) and were significantly superior to all other treatments while the lowest value was recorded for T_5 (117.23) which was at par with T_4 (117.33), T_3 (117.46) and T_2 (120.24).

4.4.1.3. Organic carbon

The results revealed that the treatments differ significantly with respect to organic carbon status. The mean values for organic carbon ranged between 1.97 to 2.51 per cent. The highest value was recorded for T_2 (2.51) and was found to be on a par with T_6 (2.47), T_3 (2.34) and T_1 (2.28) while the lowest value was recorded for T_5 (1.97) and was at par with T_4 (2.20).

4.4.1.4. Cation exchange capacity

The results indicated that the treatments imparted a significant effect with respect to the CEC of the soil. The mean values for the cation exchange capacity ranged between 10.8 to 13.17 c mol (p^+) kg^{-1}. The highest value was noticed for T_6 (13.17) which was on a par with T_2 (12.87), T_5 (12.77) and T_3 (12.37) while the lowest value was recorded for T_1 (10.8).

4.4.1.5. Anion exchange capacity

The results revealed that a significant effect due to treatments with respect to the anion exchange capacity. The mean values ranged between 2.24 to 2.81 c mol (e^-) kg^{-1}. The highest value was recorded for T_3 (2.81) which was on a par with other treatments T_2 (2.73), T_4 (2.70), T_5 (2.67) and T_6 (2.74) while the lowest value was recorded for T_1 (2.24).

4.4.1.6. Available nitrogen

The available nitrogen status of the soil was statistically significant

Table 14 : *Soil parameters of the inter cropping experiment (Rubber) at the CES, Chethackal*

Treat-ments	pH	Electical conductivity (μSM^{-1})	Organic carbon (per cent)	Cation Exchange Capacity (c mol (p+) kg^{-1} of soil)	Anion Exchange Capacity (c mol(e^-) kg^{-1} of soil)	Available Nitrogen (Kg ha^{-1})	Available Phosphorus (Kg ha^{-1})	Available Potassium (Kg ha^{-1})	Clay content (per cent)	Sesqui-oxides (per cent)
T1	4.30	127.61	2.28	10.80	2.24	258.70	55.01	94.02	13.28	25.90
T2	4.29	120.24	2.51	12.87	2.73	233.13	55.80	102.10	14.14	24.70
T3	4.29	117.46	2.34	12.37	2.81	231.79	55.12	93.55	16.42	23.20
T4	4.37	117.33	2.20	12.35	2.70	227.62	52.25	100.07	14.42	24.20
T5	4.21	117.23	1.97	12.77	2.67	219.37	51.69	89.51	13.57	24.10
T6	4.53	129.48	2.47	13.17	2.74	225.88	52.79	101.18	13.28	24.60
SEd	0.06	1.77	0.08	0.27	0.09	7.46	N.S	2.30	N.S	0.6
CD	0.19	5.09	0.23	0.83	0.27	21.44	N.S	6.52	N.S	1.8

under different inter crops with rubber as the major crop. The mean values for available nitrogen ranged between 219.37 to 258.70 kg ha^{-1}. The highest value was recorded for T_1 (258.70) which was significantly superior than all other treatments while T_5 (219.37) recorded the lowest value which was on a par with T_2 (233.13), T_3 (231.79), T_4 (227.62) and T_6 (225.88).

4.4.1.7. Available phosphorus

The results showed that the treatments did not impose any significant effect with respect to the available P status of the experimental site. The mean values ranged between 51.69 to 55.80 kg ha^{-1} with the highest value recorded for T_2 (55.80) which did not differ significantly from the other treatments.

4.4.1.8. Available potassium

The results indicated highly significant effect due to treatment difference with respect to the available K status of the soil. The mean values for available K ranged between 89.51 to 102.1 kg ha^{-1}. The highest value was noticed for T_2 (102.1) which was on a par with T_6 (101.18) and T_4 (100.07) while the lowest value was recorded for T_5 (89.51) which was at par with T_3 (93.55) and T_1 (94.02).

4.4.1.9. Clay content

The treatments did not impose any significant effect on clay content of the soil samples. The mean values for clay content ranged between 13.28 to 16.42 per cent. The highest value was recorded for the treatment T_3 (46.42).

4.4.1.10. Sesquioxides

Significant effect on sesquioxide content due to treatments was observed in this case. The mean values ranged between 23.20 to 25.90 per cent. The highest value for sesquioxide was noticed for T_1 (25.90) which was on a par with T_2 (24.70), T_6 (24.60) while T_3 (23.20) recorded the lowest value.

4.4.2. Biological characters

The enzyme activities of the samples collected from the CES, Chethackal under different inter crops were assessed and the results are presented below and in Table 15.

4.4.2.1. Urease activity

The urease activity assay of the soil samples showed highly significant effect due to treatments. The mean values ranged between 339.11 to 452.2 ppm of urea hydrolysed g^{-1} of soil hr^{-1}.

The highest value was recorded for T_4 (452.2) which was on a par with T_6 (446.50), T_2 (442.7) and T_5 (431.3) while the lowest value was recorded for T_3 (339.11) which was at par with T_1 (344.81).

4.4.2.2. Phosphatase activity

The results of phosphatase activity revealed no significant difference due to treatment effects. The mean values for phosphatase activity ranged between 95.83 to 113.85 μg of p-nitro phenyl phosphate released g^{-1} of soil hr

Table 15 : *Biological characters of the inter cropping experiment at the CES, Chethackal*

Sl. No.	Treat-ments	Urease activity (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Respiratory activity (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	T1	344.81	104.97	360.00	484.65	41.28	3.80
2.	T2	442.70	113.85	390.00	630.11	46.71	5.40
3.	T3	339.11	102.64	370.00	427.58	36.85	4.60
4.	T4	452.20	106.80	390.00	478.07	41.42	3.40
5.	T5	431.30	100.39	350.00	481.27	42.14	2.50
6.	T6	446.50	95.83	390.00	608.30	49.14	3.60
	SE	22.98	N.S	N.S	25.14	1.42	N.S
	CD	65.97	N.S	N.S	75.21	4.10	N.S

^{1}. The highest value was recorded for T_2 (113.85) which was not significantly superior compared to other treatments.

4.4.2.3. Protease activity

The results presented showed no significant effect due to treatments in the case of protease activity. The mean values for protease activity ranged between 350.00 to 390.00 micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}. The highest values were recorded for T_2 (390), T_4 (390) and T_6 (390) which were not significantly different from other treatments.

4.4.2.4. Dehydrogenase activity

From the results of the dehydrogenase activity it was inferred that the treatments had expressed highly significant effect. The mean values ranged between 427.58 to 630.11 µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded for T_2 (630.11) followed by T_6 (608.30), T_1 (484.65), T_5 (481.27) and T_4 (478.07) while the lowest value was recorded for T_3 (427.58).

4.4.2.5. Cellulase activity

The treatments showed a significant effect on the activity of the enzyme cellulase with the mean values ranging between 36.85 to 46.75 ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded in T_6 (49.14) under rubber + cover crop which was on a par with T_2 (46.71) while lowest value was recorded for T_3 (36.85).

4.4.2.6. Respiratory activity

The results of the assay of the respiratory activity revealed no significant effect for the treatments. The mean values for the respiratory activity varied form 2.50 to 5.40 mg of CO_2 evolved g^{-1} of soil hr^{-1}. The highest value was recorded for the treatment T_2 (5.4) which was observed to be on a par with other treatments.

4.4.3. Correlation analysis

Correlation between the soil enzyme activies and soil parameters were studied and the results are presented in Table 16.

The correlation analysis carried out between urease activity and soil parameters did not give any significant positive correlation for any of the soil characters studied. Significant negative correlation was observed between the urease acivity and respiratory activity (-0.37). A positive and non significant correlation was observed for urease activity with pH (0.30), clay content (0.04) and available K (0.03). While the factors CEC (-0.02), AEC (-0.21), sesquioxide (-0.13), available N (-0.12) and available P (-0.21) exhibited negative and non significant correlation.

The correlation analysis between soil phosphatase activity and the soil parameters like EC, pH, OC, CEC, AEC, available N, P, K, respiratory activity and sesquioxides showed no positive and significant correlation in this study. The soil parameters like OC (0.02), clay content (0.03), available N (0.02), available P (0.01), available K (0.14), respiratory activity (0.11) on the other

Table 16 : *Correlation matrix (Soil parameters Vs Enzyme activities), at the CES, Chethackal*

Sl. No.	Enzyme activity	pH	EC	Organic Carbon	Clay Content	CEC	AEC	Available N	Available P	Available K	Respiratory activity	Sesqui oxides
1.	Urease activity	0.30	-0.18	-0.14	0.04	-0.02	-0.12	-0.12	-0.21	0.03	-0.37*	-0.13
2.	Phosphatase activity	-0.32*	-0.04	0.02	0.03	-0.08	-0.15	0.02	0.01	0.10	0.11	-0.10
3.	Protease activity	0.28	0.05	0.33*	0.19	0.08	0.14	0.04	0.25	0.35*	0.34*	-0.08
4.	Dehydrogenase activity	0.20	0.17	0.25	-0.09	-0.12	-0.20	0.14	-0.01	0.23	0.24	0.28
5.	Cellulase activity	-0.05	-0.19	-0.13	0.03	0.09	-0.16	-0.09	-0.08	0.32*	0.00	0.27

t at 5% - 0.30
at 1% - 0.39

hand showed a positive non significant correlation with the urease activity. A negative but significant correlation was observed between phosphatase activity and soil pH (-0.32). Similarly a negative but non significant correlation was observed for soil phosphatase activity with EC (-0.04), CEC (-0.08), AEC (-0.15) and sesquioxides (-0.10).

The results of the correlation analysis showed a significant and positive correlation for protease activity with soil parameters like OC (0.33), available K (0.35) and respiratory activity (0.34). A positive and non significant correlation was observed for protease activity with pH (0.28), EC (0.05), clay content (0.19), CEC (0.08), AEC (0.14), available N (0.04) and available P (0.25) while a negative and non significant correlation was observed between protease activity and sesquioxides (-0.08).

None of the soil parameters viz., pH, EC, OC, CEC, AEC, available N, P, K, respirtory activity, clay content and sesquioxides correlated positively and significantly with the dehydrogenase activity. Positive non significant correlations were observed for dehydrogenase activity with pH (0.20), EC (0.17), available N (0.14), OC (0.25), available K (0.23), respiratory activities (0.24) and sesquioxides (0.28).

Correlations observed for dehydrogenase activity with clay content (-0.09), CEC (-0.12), AEC (-0.20) and available P (-0.01) were non significant and negative.

Significant and positive correlation was observed between cellulase activity and available K (0.32). The correlations for cellulase activity with

parameters like clay content (0.03), CEC (0.09), respiratory activity (0.01) and sesquioxides (0.27) were found to be positive but non significant.

Negative and non significant correlations were observed between cellulase activity with pH (-0.05), EC (-0.19), OC (-0.13), AEC (-0.16), available N (-0.09) and available P (-0.08).

4.4.4. Regression analysis

Multiple regression analysis carried out between the soil factors and the enzyme activities for all the five enzymes to study the functional relationship between these factors and the enzyme activities in soil. The results are presented in Table 17.

Out of the 11 parameters studied, factors like pH (+2.39), EC (+2.44) and respiratory activity (+3.02) were found to be related significantly with the urease activity.

The results of the multiple regression analysis showed that none of the 11 parameters were significantly related with the phosphatase activity.

In the case of protease activity no soil parameter studied was found to contribute significantly to the activity in soil.

The soil parameters like OC (+2.21), cation exchange capacity (+2.48) and available K (+3.35) established significant relation with the dehydrogenase activity as evident from the high regression coefficients.

In the case of cellulase activity, none of the soil parameters established a significant functional relationship with the activity of the enzyme.

Table 17 : *Multiple regression analysis (Soil parameters Vs Enzyme activities) at the CES, Chethackal*

Sl. No.	Parameters	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase
1.	pH	+1.03 (+2.39*)	-31.46 (-1.89)	+44.43 (+1.46)	+45.69 (+1.64)	-5.79 (+1.09)
2.	EC	-34.15 (+2.44*)	+0.31 (+0.59)	+0.81 (+0.82)	+1.27 (+0.007)	-0.01 (+0.94)
3.	OC	+968.84 (+1.76)	-27.76 (-1.31)	+12.81 (+0.33)	+11.70 (+2.21*)	-10.54 (+0.22)
4.	Clay content	+34.95 (+0.31)	-1.06 (-0.25)	+15.31 (+1.95)	+10.91 (-0.08)	-0.08 (+1.02)
5.	CEC	-408.97 (-1.30)	+5.42 (+0.44)	+8.54 (+0.38)	-22.78 (+2.48*)	+6.78 (-0.75)
6.	AEC	-23.05 (-0.74)	-1.51 (-1.27)	+1.89 (+0.87)	-1.90 (-0.08)	-0.11 (-0.64)
7.	Av. N	+1.95 (+0.44)	+9.09 (+0.54)	+0.04 (+0.15)	+0.02 (+0.14)	+0.01 (+0.07)
8.	Av. P	+1.95 (+0.08)	-0.14 (-0.16)	+1.56 (+0.93)	+0.24 (-0.76)	-0.15 (+0.10)
9.	Av. K	-6.91 (-0.50)	+0.68 (+1.29)	+0.99 (+1.03)	+0.04 (+3.35)**	+0.39 (+0.03)
10.	Respiratory activity	-2.18 (+3.02)*	2.30 (+0.83)	+4.28 (+0.84)	+9.42 (+1.57)	+9.90 (+1.35)
11.	Sexquioxides	+11.92 (+0.23)	-1.99 (-1.02)	+3.89 (+1.09)	+9.72 (+1.26)	+10.55 (+2.00)
	R^2	41.95	20.59	37.07	31.58	41.94

Values in paranthesis represents t - Values

t_{30} at 5% - 2.04

at 1% - 2.45

4.5. Permanent manurial experiment on coconut at the CRS, Balaramapuram

Soil samples were collected from the Coconut Research Station at Balaramapuram from the permanent manurial experiment. The samples were subjected to the analysis of soil parameters and enzyme activities. The results of the analysis on soil parameters and enzyme activities are presented below in Tables 18 to 25.

4.5.1. Soil parameters

The analyses of pH, EC, OC, CEC, AEC, available N, available P, available K, sesquioxides and clay content were carried out and the results after statistical analysis are summarised below in the following pages.

4.5.1.1. Soil reaction

The main effects of N and P were non significant while K imposed a significant effect on pH. For the main effects of N viz., N_0 (4.41) N_1 (4.68) and N_2 (4.81) were found to be on a par and there was no significant difference between these three levels. Similarly there was no significant difference due to P for all the three levels viz. P_0 (4.58), P_1 (4.65) and P_2 (4.66) with respect to their effects on soil pH. The main effects of K for the three levels K_0 (4.54), K_1 (4.67) and K_2 (4.69) on pH were found to be on a par and hence did not show any significant differences.

The interaction N × P was significant, while P × K and N × K were non significant. For N × P interaction, the highest value was recorded for N_0P_2 (5.05) which was on a par with treatments receiving N_1P_0 (4.96), N_1P_2 (4.82), N_2P_1 (4.81) N_2P_0 (4.57), N_2P_2 (4.57), N_0P_0 (4.51) and N_0P_1 (4.43). The lowest value was noted for N_1P_1 (3.97). Interaction between P × K revealed no significant difference among the various combinations viz., P_0K_0 (4.56), P_1K_0 (4.83), P_2K_0 (4.61), P_0K_1 (4.98), P_1K_1 (4.15), P_2K_1 (4.62), P_0K_2 (4.48), P_1K_2 (4.64) and P_2K_2 (4.82) with respect to soil pH. Similarly N × K interactions also did not impart any significant effect.

For the interaction among F, L and M significant difference due to treatments was observed with the highest value recorded for $N_1P_2K_2$ (5.11) and $N_2P_0K_1$ (5.11).

4.5.1.2. Electrical conductivity

The main effects of N, P and K were found to impose significant effect on EC. In the case of main effect of N, the highest value was noted for N_1 (209.93) which was on par with N_2 (208.00) and the lowest value was noted for N_0 (205.06) which was significantly lower than N_1 and N_2. Similarly for the main effect of P, the highest value was noticed for P_1 (213.69) which was significantly superior than P_2 (202.17) and P_0 (207.13). In the case of main effects of K, the highest value for EC was noticed for K_0 (212.86) which was significantly superior than K_1 (202.83) and K_2 (207.30) while the lowest value was recorded for K_1 (202.83).

The interactions, N × P, P × K and N × K imposed significant effect with respect to EC. For N × P interactions, the highest value was recorded for N_1P_1 (215.44) which was on a par with N_1P_0 (215.24) and N_0P_2 (214.56) while N_0P_0 (198.59) recorded the lowest value.

In the case of interactions between N and K, the highest value was noticed for N_0K_0 (217.80) which was found to be on a par with N_0K_2 (215.82) and N_2K_1 (213.36) while the lowest value was recorded for N_1K_0 (192.60) which was significantly lower than others.

Similarly for P × K interaction, the highest value was recorded for P_1K_1 (214.53), which was at par with P_0K_2 (213.54), P_0K_1 (213.31), P_2K_1 (213.24) and P_0K_0 (211.75) while P_1K_2 (192.60) recorded the lowest value which was significantly lower than other treatments.

The treatments imposed significant effect on EC of the samples. The highest value was recorded for T_{25} (222.35) which was on a par with T_7 (221.20) and T_{15} (218.72) while T_8 (180.90) recorded the lowest value which was statistically on a par with T_2 (181.83) and T_{26} (185.68).

4.5.1.3. Organic carbon

The main effects of N and P were found to be significant while the main effect of K was non significant. In the case of main effects of N, the highest value was recorded for N_1 (0.41) which was significantly superior than N_0 (0.31) and N_2 (0.20) while the lowest value was recorded for N_2 (0.20) which was significantly lower than the other two levels. For the main effect of

Table 18 : *The main effects of the factors on the soil properties, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	pH	EC (μ SM^{-1})	O C (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Available N (Kg ha^{-1})	Available P (Kg ha^{-1})	Available K (Kg ha^{-1})	Clay content (%)	Sesqui oxides (%)	Nut yield/ plot
A	N											
(i)	N_0	4.41	205.06	0.31	5.89	1.15	184.62	52.75	103.15	14.83	19.23	165
(ii)	N_1	4.68	209.93	0.41	6.45	1.08	194.71	61.04	102.76	14.38	20.41	148
(iii)	N_2	4.81	208.00	0.20	7.03	0.96	196.31	52.36	113.63	14.16	22.71	119
	CD	N.S	N.S	0.07	0.63	N.S	N.S	2.77	N.S	1.90	2.01	27
B	P											
(i)	P_0	4.58	207.13	0.33	6.38	0.97	203.78	55.54	99.11	14.16	19.30	158
(ii)	P_1	4.65	213.69	0.38	6.56	1.02	176.46	63.51	106.03	13.94	20.21	127
(iii)	P_2	4.66	202.17	0.22	6.43	1.20	195.40	47.10	114.40	15.27	21.47	136
	CD	N.S	N.S	0.07	N.S	0.19	24.79	2.77	10.98	1.90	2.01	27
C	K											
(i)	K_0	4.54	212.86	0.33	6.39	1.12	205.86	49.69	106.45	13.72	22.41	82
(ii)	K_1	4.67	202.83	0.27	6.20	1.01	177.17	57.53	97.23	14.16	19.43	110
(iii)	K_2	4.69	207.30	0.32	6.79	1.05	192.62	58.93	115.86	15.50	18.21	181
	CD	0.49	N.S	N.S	N.S	N.S	24.79	2.77	10.98	1.90	2.01	27

P, the highest value was recorded for P_1 (0.38) which was at par with P_0 (0.33) while the lowest value was recorded for P_2 (0.22) which was significantly lower than P_0 and P_1. In the case of main effects of K, the highest was recorded for K_0 (0.33) which was on a par with K_1 (0.27) and K_2 (0.32).

The interactions N × P, P × K and N × K were found to be significant. For N × P interaction, the highest value was recorded for N_1P_1 (0.55) which was at par with N_0P_1 (0.47). Similarly the lowest value was recorded for N_2P_2 (0.17) and N_0P_2 (0.17) which were on a par with N_2P_1 (0.23), N_1P_2 (0.27) and N_2P_0 (0.26).

For interactions between N and K, the highest value was recorded for N_2K_1 (0.45), which was on a par with N_0K_1 (0.41), N_1K_1 (0.39), N_0K_0 (0.36) and N_2K_0 (0.34) while the lowest value was recorded for N_2K_2 (0.18) which was at par with N_1K_2 (0.21), N_0K_2 (0.22) and N_1K_0 (0.23).

In the case of for P × K interactions, the highest value was recorded for P_0K_1 (0.43) which was on a par with P_2K_0 (0.40), P_2K_1 (0.37), P_1K_1 (0.34) and P_0K_0 (0.32) while P_2K_2 (0.20) recorded the lowest value.

The treatments imposed significant effects with the highest value noticed for T_{12} (0.60) and T_{15} (0.60) which were on a par with T_{13} (0.57), T_{10} (0.51), T_{14} (0.48), T_4 (0.48), and T_3 (0.45). The lowest value was recorded for T_{26} (0.12) which was statistically at par with T_{27} (0.15), T_{21} (0.15), T_8 (0.15), T_{16} (0.15), T_{18} (0.15), T_{20} (10.15), T_{19} (0.18), T_5 (0.21), T_{25} (0.24), T_{22} (0.24), T_{24} (0.24) and T_6 (0.27).

4.5.1.4. Cation exchange capacity

The main effects of P and K were non significant while the effect of N was significant. For the main effects of N, the highest value was noticed for N_2 (7.03) which was at par with N_1 (6.45) and the lowest value was recorded for N_0 (5.89) which was significantly lower than N_1 (6.45) and N_2 (7.03). In the case of main effects of P, the highest value was recorded for P_1 (6.56) which was on a par with P_0 (6.38) and P_2 (6.43) and did not impart any significant effect. Similarly for the main effect of K, the highest value was recorded for K_2 (6.79) which was on a par with K_0 (6.39) and K_1 (6.20).

The interactions N × P, P × K and N × K were found to be significant with respect to CEC. For the interaction between N and P, the highest value was recorded for N_2P_1 (7.13) which was on a par with N_0P_2 (7.11), N_2P_2 (7.1), N_1P_2 (6.9), N_1P_0 (6.41), N_1P_1 (6.36) and N_0P_0 (6.18) while the lowest value was observed for N_2P_0 (5.08) which was at par with N_0P_1 (5.86).

In the case of interaction between N and K, the highest value was recorded for N_2K_2 (7.13), which was found to be on a par N_1K_2 (7.06), N_0K_2 (6.91), N_2K_1 (6.71), N_2K_0 (6.53), N_1K_1 (6.41) and N_0K_1 (6.23) and the lowest value for N_1K_0 (5.11) which was on a par with N_0K_0 (6.03).

Similarly for P × K interaction, the highest value was recorded for P_2K_2 (7.23), which was on a par with other treatments except P_1K_2 (5.68) which was significantly different from the rest. The lowest value was noticed in P_1K_2 (5.68) which was at par with all other treatments except P_2K_1 (6.90) and P_2K_2 (7.23).

It was observed that the treatments imposed significant effect on CEC with the highest value noticed for T_{27} (7.5) which was on a par with T_{26} (7.25), T_{20} (7.15), T_{18} (7.15), T_{22} (7.15), T_{21} (7.10), T_{19} (7.05), T_{9} (7.05), T_{16} (7.05), T_{6} (7.00), T_{15} (6.90), T_{24} (6.80), T_{23} (6.75), T_{2} (6.65), T_{25} (6.55) and T_{1} (6.35). The lowest value was recorded for T_{8} (4.6) which was at par with T_{3} (5.55), T_{10} (5.55) and T_{7} (5.60).

4.5.1.5. Anion exchange capacity

The main effects of N and K were found to be non significant while the main effect P was found to be significant. The main effect of N was found to be at par for all the three levels. For the main effect of P on AEC, the highest value was noted at P_2 (1.2) which was significantly different from P_0 (0.97) and P_1 (1.02) and lowest value was recorded for P_0 (0.97) which was at par with P_1 (1.02). For the main effects of K on AEC, there was no significant difference though the highest value was recorded at K_0 (1.12).

The interactions N × P, N × K and P × K were found to impose significant effect with respect to AEC. For the N × P interaction, the highest value was recorded for N_0P_1 (1.66) which was on a par with N_2P_1 (1.11) while the lowest value was recorded for N_0P_2 (0.88) which was on a par with all other combinations except N_2P_1 (1.11) and N_0P_1 (1.66).

In the case of N × K interaction, the highest value was recorded for N_0K_0 (1.33), which was significantly superior than others while the lowest value was recorded for N_1K_2 (10.88) which was on a par with N_2K_2 (1.01), N_2K_0 (1.03), N_0K_1 (1.06) and N_1K_1 (1.06).

Similarly for P × K interaction, the highest value was recorded for P_0K_2 (1.36) which was at par with P_1K_2 (1.23) and P_0K_2 (1.36). The lowest value was observed for P_1K_1 (0.75) which was at par with P_0K_0 (0.83).

From the results of the analysis of AEC, it was observed that the treatments imposed significant effect with the highest value recorded for T_7 (1.75) followed by T_8 (1.70) which were on a par. The lowest value was recorded for T_5 (0.60) which was on a par with T_{14} (0.75), T_{10} (0.75), T_{19} (0.75), T_{26} (0.85), T_{23} (0.90), T_{20} (0.95), T_9 (0.90), T_{24} (0.90), T_{18} (0.95), T_3 (0.95), T_{21} (1.00), T_2 (1.00) and T_1 (1.00).

4.5.1.6. Available nitrogen

Except for N, the main effects of P and K were found to impose significant effect of available N. For the main effect of N, no difference between N_0 (184.62), N_1 (194.71) and N_2 (196.31) was observed. But for the main effect of P, the highest value for available N was recorded for P_0 (203.78) which was on a par with P_2 (195.40) and P_1 (176.46). For the main effect of K, the highest value was recorded for K_0 (205.86) which was superior than K_2 (192.62) and K_1 (177.17).

The interactions N × P, P × K and N × K were found to impose significant effects with respect to available nitrogen. In the case of N × P interactions, the highest value was recorded for N_0P_2 (211.28) which was on a par with N_0P_0 (202.46), N_2P_0 (199.68), N_2P_1 (187.23), N_0P_1 (197.60), N_2P_2 (199.31) and N_1P_1 (199.30) while the lowest value for available N was recorded for N_1P_0 (151.74) which was at par with N_1P_2 (178.34) and N_2P_1 (187.23).

Table 19 : *The interaction effects of the factors on the soil properties, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	pH	EC (μ SM^{-1})	O C (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Available N (Kg ha^{-1})	Available P (Kg ha^{-1})	Available K (Kg ha^{-1})	Clay content (%)	Sesqui oxides (%)	Nut yield/ plot
A	N x P											
(i)	N_0P_0	4.51	198.59	0.35	6.18	0.98	202.46	44.79	94.4	13.83	19.23	151
(ii)	N_1P_0	4.96	215.24	0.32	6.41	1.03	151.74	59.13	93.85	15.16	20.21	173
(iii)	N_2P_0	4.57	201.36	0.26	5.08	1.45	199.68	54.35	121.21	15.50	23.50	152
(iv)	N_0P_1	4.43	208.23	0.47	5.86	1.66	197.60	73.46	94.96	14.16	24.10	171
(v)	N_1P_1	3.97	215.44	0.55	6.36	1.06	199.30	62.71	109.05	13.83	19.80	130
(vi)	N_2P_1	4.81	206.12	0.23	7.13	1.11	187.23	46.95	104.26	15.16	18.90	114
(vii)	N_0P_2	5.05	214.56	0.17	7.11	0.88	211.28	48.37	107.98	14.50	19.20	176
(viii)	N_1P_2	4.82	210.40	0.27	6.90	0.96	178.34	68.68	115.21	12.83	19.40	142
(ix)	N_2P_2	4.57	199.04	0.17	7.10	1.03	199.31	40.01	117.71	15.16	22.71	93
	CD	0.86	4.09	0.12	1.09	0.34	42.94	4.81	19.02	3.29	4.02	32
B	P x K											
(i)	P_0K_0	4.56	211.75	0.32	6.31	0.83	213.28	52.55	96.86	13.50	18.73	105
(ii)	P_1K_0	4.83	201.34	0.27	6.60	1.06	201.93	56.74	84.78	13.83	19.63	72
(iii)	P_2K_0	4.61	208.29	0.40	6.25	1.03	196.12	57.34	115.70	15.16	20.22	70
(iv)	P_0K_1	4.98	213.31	0.43	6.46	1.18	197.21	56.14	110.33	12.83	21.47	172
(v)	P_1K_1	4.15	214.53	0.34	6.31	0.75	159.53	69.88	93.08	13.50	19.60	192

The interaction effects of the factors on the soil properties, at the PME, CRS, Balaramapuram (Contd....)

Sl. No.	Factor	pH	EC (μ SM^{-1})	O C (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Available N (Kg ha^{-1})	Available P (Kg ha^{-1})	Available K (Kg ha^{-1})	Clay content (%)	Sesqui oxides (%)	Nut yield/ plot
(vi)	P_2K_1	4.62	213.24	0.37	6.90	1.13	172.65	64.50	114.70	15.50	23.90	147
(vii)	P_0K_2	4.48	213.54	0.24	6.40	1.36	207.08	40.37	112.16	14.83	24.70	198
(viii)	P_1K_2	4.64	192.60	0.22	5.68	1.23	170.04	45.99	113.83	15.16	24.20	152
(ix)	P_2K_2	4.82	200.38	0.20	7.23	1.00	209.09	54.95	117.20	15.83	21.70	192
	CD	N.S	4.09	0.12	1.09	0.34	42.94	4.81	19.02	3.29	4.02	32
C	N x K											
(i)	N_0K_0	4.73	217.80	0.36	6.03	1.33	199.30	43.59	103.95	14.5	20.22	130
(ii)	N_1K_0	4.68	192.60	0.23	5.11	1.10	149.66	54.35	83.80	15.16	21.34	74
(iii)	N_2K_0	4.64	204.79	0.34	6.53	1.03	204.91	60.32	121.71	14.83	19.20	42
(iv)	N_0K_1	4.48	204.96	0.41	6.23	1.06	210.60	50.53	94.21	13.50	18.43	191
(v)	N_1K_1	4.10	211.46	0.39	6.41	1.06	177.88	59.13	105.26	13.83	21.22	178
(vi)	N_2K_1	4.64	213.36	0.45	6.71	1.11	195.65	73.46	108.80	15.83	22.47	175
(vii)	N_0K_2	4.81	215.82	0.22	6.91	0.98	207.67	54.95	121.20	13.16	23.10	170
(viii)	N_1K_2	4.84	204.41	0.21	7.06	0.88	203.95	59.13	102.63	13.50	24.50	191
(ix)	N_2K_2	4.78	203.76	0.18	7.13	1.01	177.31	43.00	117.08	15.83	22.10	93
	CD	N.S	4.09	0.12	1.09	0.34	42.94	4.81	19.02	3.29	4.02	32

For N × K interaction, the highest value was observed for N_0K_1 (210.60) which was on a par with N_0K_2 (207.67), N_2K_0 (204.91), N_1K_2 (203.95), N_0K_0 (199.30), N_2K_1 (195.65), N_1K_1 (177.88), N_2K_2 (177.31) and the lowest value was observed for N_1K_0 (149.66) which was at par with N_2K_2 (177.31) and N_1K_1 (177.88).

In the case of P × K interaction, the highest value was recorded for P_0K_0 (213.28) which was on a par with P_2K_2 (209.09), P_0K_2 (207.08), P_1K_0 (201.93), P_0K_1 (197.21), P_2K_0 (196.12), and P_2K_1 (172.65) and the lowest value was recorded for P_1K_1 (159.53) which was at par with P_1K_2 (170.04), P_2K_1 (172.65), P_2K_0 (196.12), P_0K_1 (197.21) and P_1K_0 (201.93).

The treatments imposed significant effects with respect to available nitrogen with the highest value recorded for T_{10} (225.55) and the lowest for T_5 (61.32).

4.5.1.7 Available phosphorus

The main effects of N, P and K were found to impose significant effects with respect to available phosphorus. For the main effect of N, the highest value was observed for N_1 (61.04) which was significantly superior to other levels while the lowest value was noticed for N_0 (52.75) which was on a par with N_2 (52.36). In the case of main effects of P, the highest value was recorded for P_1 (63.51) which was significantly superior compared to other two levels and the lowest value was registered for P_2 (47.10) which was significantly lower than P_1 (63.51) and P_0 (55.54).

For the main effect of K, the highest value was observed for K_2 (58.93) which was at par with K_1 (57.53) and the lowest value was noticed for K_0 (49.69) which was significantly lower than K_1 and K_2.

The interaction between N × K, P × K and N × P imposed significant effects with respect to available phosphorus. For the interaction between N and P, the highest value was recorded for N_0P_1 (73.46) which was on a par with N_1P_2 (68.68) while the lowest value was recorded for N_2P_2 (40.01) which was also at par with N_0P_0 (44.79).

In the case of N × K interaction, the highest value was noticed for N_2K_1 (73.46) which was significantly different from other combinations and the lowest value was noticed for N_2K_2 (43.00) which was on a par with N_0K_0 (43.59).

For P × K interaction, the highest value was noticed for P_1K_1 (69.88) which was significantly superior to other combinations and the lowest value was observed for P_0K_2 (40.37).

The treatments imposed significant effect with respect to the available P status. The highest value was noticed for T_{15} (84.22) which was on a par with T_{12} (80.63) and T_{23} (80.63). The treatment T_{27} (34.04) recorded the lowest value which was on a par with T_{16} (35.13) and T_1 (37.61).

4.5.1.8. Available potassium

Except for N, the main effects of P and K were found to impose significant effect with respect to available potassium. For the main effect of

Table 20 : *Effect of treatments on the soil properties, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	pH	EC (μ SM^{-1})	O C (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Available N (Kg ha^{-1})	Available P (Kg ha^{-1})	Available K (Kg ha^{-1})	Clay content (%)	Sesqui oxides (%)	Nuit yield/ plot
1.	$N_0P_0K_0$	4.46	216.26	0.27	6.35	1.00	196.17	37.61	98.55	13.50	17.00	140
2.	$N_0P_0K_1$	4.60	181.83	0.33	6.65	1.00	216.59	46.58	57.65	13.50	19.80	117
3.	$N_0P_0K_2$	4.49	197.69	0.45	5.55	0.95	194.61	50.17	127.00	14.50	16.65	196
4.	$N_0P_1K_0$	5.05	215.96	0.48	6.15	1.25	194.60	51.96	86.05	14.50	18.05	103
5.	$N_0P_1K_1$	5.06	215.09	0.21	6.10	0.60	61.32	69.88	93.90	15.50	18.70	253
6.	$N_0P_1K_2$	4.78	214.69	0.27	7.00	1.25	199.31	55.54	101.60	15.50	20.75	156
7.	$N_0P_2K_0$	4.68	221.20	0.33	5.60	1.75	207.14	41.21	127.25	15.50	22.00	146
8.	$N_0P_2K_1$	4.39	180.90	0.15	4.60	1.70	171.07	46.58	99.85	16.50	21.30	202
9.	$N_0P_2K_2$	4.64	201.99	0.30	7.05	0.90	220.82	75.26	136.55	14.50	22.70	178
10.	$N_1P_0K_0$	4.15	206.03	0.51	5.55	0.75	225.55	71.67	101.55	13.50	17.55	128
11.	$N_1P_0K_1$	4.78	207.36	0.30	5.95	1.30	164.81	68.09	100.5	14.50	20.65	243
12.	$N_1P_0K_2$	4.38	211.31	0.60	6.10	1.15	202.44	80.63	82.85	14.50	20.45	147
13.	$N_1P_1K_0$	4.89	211.80	0.57	6.10	1.20	211.85	44.79	104.45	13.50	17.50	59
14.	$N_1P_1K_1$	4.60	215.79	0.48	6.10	0.75	221.26	59.13	84.45	13.50	15.65	171
15.	$N_1P_1K_2$	4.44	218.72	0.60	6.90	1.25	164.81	84.22	138.25	15.50	18.90	161
16.	$N_1P_2K_0$	4.41	197.07	0.15	7.05	1.25	194.42	35.13	76.65	14.50	17.35	37
17.	$N_1P_2K_1$	4.93	211.24	0.39	7.20	1.15	147.59	50.17	130.85	13.50	17.85	122

Effect of treatments on the soil properties, at the PME, CRS, Balaramapuram (Contd...)

Sl. No.	Factor	pH	EC (μ SM^{-1})	O C (%)	CEC (c mol (p+) kg^{-1} of soil)	AEC (c mol(e$^-$) kg^{-1} of soil)	Available N (Kg ha^{-1})	Available P (Kg ha^{-1})	Available K (Kg ha^{-1})	Clay content (%)	Sesqui oxides (%)	Nut yield/ plot
18.	$N_1P_2K_2$	5.11	210.05	0.15	7.15	0.95	219.69	55.55	105.30	17.50	19.00	265
19.	$N_2P_0K_0$	5.06	212.96	0.18	7.05	0.75	218.12	48.38	90.50	13.50	20.70	48
20.	$N_2P_0K_1$	5.11	210.05	0.15	7.15	0.95	219.69	55.55	105.30	17.50	19.00	155
21.	$N_2P_0K_2$	4.97	215.88	0.15	7.10	1.00	191.33	41.21	137.25	16.50	23.10	252
22.	$N_2P_1K_0$	5.01	212.17	0.24	7.15	1.1	185.20	71.67	140.50	14.50	19.90	52
23.	$N_2P_1K_1$	4.80	212.72	0.33	6.75	0.90	196.00	80.63	100.90	14.50	16.25	150
24.	$N_2P_1K_2$	4.65	206.32	0.24	6.80	0.90	153.83	53.75	104.25	14.50	16.25	140
25.	$N_2P_2K_0$	4.37	222.35	0.24	6.55	1.10	219.69	44.79	132.60	14.50	23.65	26
26.	$N_2P_2K_1$	4.62	185.68	0.12	7.25	0.85	191.47	41.21	110.80	15.50	23.15	119
27.	$N_2P_2K_2$	4.72	189.10	0.15	7.50	1.15	186.76	34.04	109.75	15.50	20.15	132
	CD	1.05	5.01	0.15	1.33	0.42	52.60	5.89	23.29	4.03	7.42	48

N, the highest value was noticed for N_2 (113.63) which was on a par with N_0 (103.15) and N_1 (102.76). In the case of main effect of P, the highest value was recorded for P_2 (114.40) which was on a par with P_1 (106.03) and the lowest value was noticed for P_0 (99.11). For the main effect of K, the highest value was noticed for K_2 (115.86) which was on a par with K_1 (97.23) while the lowest value was noticed for K_0 (106.45).

The interactions N × P, P × K and N × K were found to cause significant effects with respect to available potassium. The interaction effect of N × P were found to be significantly superior with the highest value noticed for N_2P_0 (121.21) which was on a par with N_1P_2 (115.21), N_2P_2 (117.71), N_0P_2 (107.98), N_1P_1 (109.05), and N_2P_1 (104.26) while the lowest value was recorded for N_1P_0 (93.85) which was at par with N_0P_0 (94.40), N_0P_1 (94.96), N_0P_2 (107.98) and N_1P_1 (109.05). In the case of interaction between N × K, the highest value was recorded for N_2K_0 (121.71) which was on a par with N_0K_2 (121.20), N_2K_2 (117.08), N_2K_1 (108.80), N_1K_1 (105.26) and N_0K_0 (103.95) while the lowest value was recorded in N_1K_0 (83.80) which was at par with N_0K_1 (94.21) and N_1K_2 (102.63).

Similarly for interaction between P × K, the highest value was recorded for P_2K_2 (117.20) which was on a par with P_2K_0 (115.70), P_2K_1 (114.70), P_1K_2 (113.83), P_0K_2 (112.16) and P_0K_1 (110.33) and the lowest value was recorded for P_1K_0 (84.78) which was at par with P_1K_1 (93.08) and P_0K_0 (96.86). A significant effect was observed on available K due to treatment application. The highest value was registered for T_{22} (140.50) which was on a par with T_{15} (138.25), T_{21} (137.25), T_9 (136.55), T_{25} (132.60), T_{17} (130.85), T_7 (127.25)

and T_3 (127.00) while the lowest value was recorded for T_2 (57.65) which was significantly lower than other treatments.

4.5.1.9. Clay content

The main effects of N, P and K were not significant with respect to the clay content of the experimental site. The interactions N × P, N × K and P × K interactions also were non significant with respect to clay content.

4.5.1.10. Sesquioxides

The main effects of N, P and K were found to impose significant effect with respect to the sesquioxide content of the samples. In the case of main effect of N, the highest value was noticed at N_2 (22.71) followed by N_1 (20.41) and N_0 (19.23). In the case of main effects of P, the highest value was recorded for P_2 (21.47) which was on a par with P_1 (20.21) and the lowest value was observed for P_0 (19.3). The main effect of K, was highest for K_0 (22.41) which was significantly superior to K_1 (19.43) and K_2 (18.21) while the lowest value was observed at K_2 (18.21).

The interactions N × P, P × K and N × K were found to be significant with respect to sesquioxide content. For the interaction between N × P, the highest value was recorded for N_0P_1 (24.10) which was on a par with N_2P_0 (23.50), N_2P_2 (22.71) and N_1P_0 (20.21) while the lowest value was noted for N_2P_1 (18.9) which was at par with N_0P_2 (19.20), N_0P_0 (19.23), N_1P_1 (19.80), N_1P_2 (19.40), N_1P_0 (20.21) and N_2P_2 (22.71).

In the case of N × K interaction, the highest value was recorded for N_1K_2 (24.50) which was on a par with N_2K_1 (22.47), N_2K_2 (22.10), N_1K_0 (21.34) and N_1K_1 (21.22). Similarly the lowest value was noted for N_0K_1 (18.43) which was at par with N_0K_0 (20.22), N_1K_0 (21.34), N_2K_0 (19.20), N_1K_1 (21.22) and N_2K_2 (22.10). For P × K interaction, the highest value was noticed for P_0K_2 (24.7) which was on a par with P_1K_2 (24.20), P_2K_1 (23.90) and P_2K_2 (21.70) while the lowest value was noticed for P_0K_0 (18.73) which was on par with P_1K_0 (19.63), P_2K_0 (20.22), P_0K_1 (21.47), P_1K_1 (19.60) and P_2K_2 (21.70).

The treatments imposed significant difference with respect to sesquioxide content. The highest value was noticed for T_{25} (23.65) which was at par with all treatments except T_{14} (15.65) and T_{23} (16.25) while the lowest value was recorded for T_{14} (15.65).

4.5.1.11. Nut yield

The main effects of N, P and K were found to impose significant difference with respect to the nut yield. For the main effects of N, the highest value was noticed for N_0 (165) which was on par with N_1 (148) and was significantly different from N_2 (119). In the case of main effects of P, the highest value was noticed for P_0 (158) which was on par with P_2 (136) while P_1 (127) recorded the lowest value. In the case of main effects of K, the highest value was noticed for K_1 (170) while the lowest value was recorded for K_0 (82). The interaction between N x P, N x K and P x K were found to impose significant effect on the nut yield. For interaction between N x P, the highest

value was noticed for N_0P_2 (176) which was on par with N_0P_1 (171), N_1P_0 (173), N_2P_0 (152), N_0P_0 (151) and N_1P_2 (142) while N_2P_2 (93) recorded the lowest value.

For P x K interaction, the highest value was registered for P_0K_2 (198) which was significantly superior than other combinations except P_1K_1 (192), P_2K_2 (192) and P_0K_1 (172) while P_2K_0 (70) recorded the lowest value. Similarly for the interaction between N x K, the highest value was noticed for N_1K_1 (191) which was on par with N_0K_1 (178), N_2K_2 (175) and N_0K_2 (170) while N_2K_0 (42) recorded the lowest value for nut yield.

In the case of interaction N, P and K, the highest value was recorded for $N_1P_2K_2$ (265) which was on par with $N_2P_0K_2$ (252), $N_0P_1K_1$ (253) and $N_1P_0K_1$ (243) while the treatment $N_2P_2K_0$ (26) recorded the lowest value which was significantly lower.

4.5.2. Biological properties

The soil samples collected from the PME were subjected to the enzyme activity assay for urease, phosphatase, dehydrogenase, cellulase and protease. The results of the study are presented hereunder in Table 21 to 23.

4.5.2.1. Urease activity

The main effects of N, P and K were found to give significant variation with respect to urease activity. For the main effects of N, the highest value was recorded for N_1 (158.89) while the lowest value was recorded for N_0

(134.51). Similarly for the main effect of P, the highest value was recorded for P_0 (173.72) which was significantly different from P_1 (115.26) and P_2 (150.78), while the lowest value was recorded for P_2 (115.26). In the case of the main effects of K, the highest value was recorded for K_0 (168.58) followed by K_1 (136.71) which was at par with K_2 (134.47).

The interactions N × P, P × K and N × K showed significant difference with respect to urease activity for all combinations. For N × P interaction, the highest value was recorded for N_0P_2 (208.46) which was significantly superior than other treatments while the lowest value was noticed for N_1P_0 (82.01) and N_1P_2 (82.01) which were significantly lower compared to other treatments.

Similarly for N × K interactions, the highest value was recorded for N_0K_2 (204.13) which was significantly different from other combinations, while the lowest value was observed for N_2K_2 (99.75) which was at par with N_0K_0 (108.66).

For P × K interaction, the highest value was recorded for P_2K_2 (230.53) which was at par with P_1K_0 (212.86). Similarly the lowest value was observed for P_1K_2 (68.71) which was on a par with P_2K_1 (73.15).

The treatments imposed significant effects with respect to urease activity of samples. The highest value was noticed for T_9 (299.25) which was on a par with T_{20} (279.30) and T_{10} (272.95) while the lowest value was recorded for T_{26} (46.55) which was on par with T_{24} (66.50) and T_{16} (66.50).

4.5.2.2. Phosphatase activity

The main effects showed no significant difference except for K which

Table 21 : *The main effects of factors on the biological characters, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Respiratory (µg of CO_2 evolved g^{-1} of soil hr^{-1})
A	N						
(i)	N_0	134.51	45.83	151.11	183.63	26.63	2.70
(ii)	N_1	158.89	49.45	163.33	187.58	22.66	2.76
(iii)	N_2	146.36	49.46	165.55	189.47	25.97	2.60
	CD	11.03	N.S	N.S	N.S	N.S	N.S
B	P						
(i)	P_0	173.72	47.57	146.66	168.75	24.61	2.50
(ii)	P_1	115.26	49.89	165.55	176.33	26.02	2.58
(iii)	P_2	150.78	44.21	167.77	180.60	30.63	3.00
	CD	11.03	N.S	19.52	N.S	3.98	0.20
C	K						
(i)	K_0	168.58	50.81	176.66	187.40	22.27	2.72
(ii)	K_1	136.71	44.26	135.55	187.83	26.13	2.72
(iii)	K_2	134.47	46.66	167.77	190.45	26.86	2.70
	CD	11.03	5.32	19.52	N.S	3.98	N.S

imposed significant difference. For the main effect of N, the highest value was observed for N_1 (49.45) which was on a par with N_2 (49.46). Similarly for the main effect of P, the highest value was recorded for P_1 (49.89) which was on a par with P_0 (47.57) and P_2 (44.21). For the main effect of K, the highest value was noticed for K_0 (50.81) which was significantly superior than other levels while the lowest value was recorded for K_1 (44.26).

The interactions N × P, P × K and N × K showed significant effects with respect to phosphatase activity. In the case of N × P interaction, the highest value was recorded for N_0P_1 (54.9) which was on a par with N_1P_2, N_2P_0, N_1P_1, N_0P_2 and N_1P_0 while the lowest value was recorded for N_2P_2 (37.15) which was at par with N_0P_0 and N_2P_1.

Similarly for the interaction between N and K, the highest value was recorded for N_2K_0 (59.90) which was on a par with N_0K_2, N_1K_2, N_2K_1 and N_0K_1 while the lowest value was recorded for N_2K_2 (24.91).

For the P × K interaction, the highest value was noticed for P_0K_0 (65.65) which was on a par with P_2K_2 (60.18) while the lowest value was recorded for P_2K_0 (35.18) which was at par with P_1K_2, P_0K_2 and P_1K_0.

The results of the assay of the phosphatase revealed that the treatments imposed significant effect. Treatments T_9 (96.50) recorded the highest value which was on par with T_{10} (93.20) while T_{27} (18.30) recorded the lowest value which was on par with T_{16} (18.30), T_{21} (20.75), T_8 (28.30) and T_{11} (28.30).

4.5.2.3. Protease activity

Except for N, the main effects were significant with respect to protease

activity. For the main effects of N, the highest value was recorded for N_2 (165.55) which was on a par with N_0 (151.11) and N_1 (163.33) and was not superior.

In the case of main effect of P, the highest value for protease activity was recorded at P_2 (167.77) which was on a par with P_1 (165.55) and the lowest value was observed at P_0 (146.66).

Similarly for the main effect of K, the highest value was noticed for K_0 (176.66) which was at par with K_2 (167.77) while the lowest value was recorded for K_1 (135.55).

The interactions N × P, P × K and N × K was found to impose significant effect with respect to protease activity. For the N × P interaction, the highest value was recorded for N_2P_2 (210.00) which was on a par with N_2P_0 (190.00) and N_0P_1 (190.00) while the lowest value was recorded for N_0P_0 (103.33) which was at par with N_1P_0 (116.66) and N_1P_2 (126.66).

In the case of N × K interaction, the highest value was recorded for N_0K_1 (200.00) which was at par with N_2K_0 (193.33), N_0K_2 (183.33) and N_2K_2 (166.66) and the lowest value was recorded for N_1K_0 (113.33) which was on a par with N_2K_1 (143.33), N_1K_1 (146.66), N_0K_0 (146.66) and N_1K_2 (146.66).

Similarly for P × K interaction, the highest value was noticed for P_2K_2 (230) which was significantly different from other combinations while the lowest value was noticed for P_1K_2 (113.33) which was at par with P_1K_1 (126.66), P_2K_1 (126.66) and P_2K_0 (146.66).

Table 22 : *Interaction effects of the factors on the biological characters, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	**Urease** (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	**Phosphatase** (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	**Protease** (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	**Dehydrogenase** (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	**Cellulase** (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	**Respiratory** (µg of CO_2 evolved g^{-1} of soil hr^{-1})
A	N x P						
(i)	N_0P_0	150.80	38.23	103.33	180.61	26.25	2.30
(ii)	N_1P_0	82.01	46.85	116.66	170.20	21.00	2.60
(iii)	N_2P_0	170.73	52.41	190.00	200.10	32.66	3.10
(iv)	N_0P_1	161.91	54.90	190.00	194.86	25.08	3.00
(v)	N_1P_1	181.76	50.18	196.66	227.70	20.25	2.40
(vi)	N_2P_1	133.00	43.26	146.66	140.19	22.66	2.80
(vii)	N_0P_2	208.46	49.58	160.00	220.80	22.50	2.30
(viii)	N_1P_2	82.01	52.65	126.66	131.10	18.83	2.60
(ix)	N_2P_2	148.61	37.15	210.00	201.51	36.58	3.00
	CD	19.11	9.23	33.81	23.15	6.89	0.3
B	P x K						
(i)	P_0K_0	208.56	65.65	183.33	197.41	20.66	2.7
(ii)	P_1K_0	212.86	41.88	166.66	213.90	28.33	2.5
(iii)	P_2K_0	99.75	35.18	146.66	184.96	24.83	2.4
(iv)	P_0K_1	144.08	50.46	186.66	193.20	23.08	2.5
(v)	P_1K_1	128.56	54.60	126.66	167.90	21.75	2.5

Interaction effects of the factors on the biological characters, at the PME, CRS, Balaramapuram (Contd....)

Sl. No.	Factor	**Urease** (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	**Phosphatase** (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	**Protease** (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	**Dehydrogenase** (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	**Cellulase** (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	**Respiratory** (µg of CO_2 evolved g^{-1} of soil hr^{-1})
(vi)	P_2K_1	73.15	44.61	126.66	167.90	15.25	2.6
(vii)	P_0K_2	153.10	36.33	160.00	171.60	23.08	2.9
(viii)	P_1K_2	68.71	36.31	113.33	151.71	28.33	3.0
(ix)	P_2K_2	230.53	60.18	230.00	218.50	40.50	3.0
	CD	19.11	9.23	33.81	23.15	6.89	0.3
C	N x K						
(i)	N_0K_0	108.66	40.45	146.66	147.44	14.16	2.6
(ii)	N_1K_0	135.28	37.15	113.33	158.70	35.50	2.7
(iii)	N_2K_0	159.60	59.90	193.33	244.76	30.25	2.8
(iv)	N_0K_1	192.95	52.70	200.00	183.36	20.50	2.7
(v)	N_1K_1	139.65	40.48	146.66	181.59	21.41	2.9
(vi)	N_2K_1	144.08	55.16	143.33	197.80	26.08	2.5
(vii)	N_0K_2	204.13	59.30	183.33	231.40	32.16	2.7
(viii)	N_1K_2	135.21	55.16	146.66	193.21	21.50	2.5
(ix)	N_2K_2	99.75	24.91	166.66	128.80	24.25	2.7
	CD	19.11	9.23	33.81	23.15	6.89	0.3

ICT

The treatments imposed significant effect with respect to the protease activity. The highest value for protease activity was noted for T_9 (310.00) followed by T_{10} (270.00) which were superior compared to other treatments. The treatment T_{23} (60.00) recorded the lowest value which was on a par with T_{16} (70.00), T_5 (80.00), T_{17} (90.00), T_{15} (90.00) and T_8 (90.00).

4.5.2.4. Dehydrogenase activity

The main effects of N, P and K did not impose any significant effect with respect to dehydrogenase activity. In case of the main effect of N, the highest value was observed at N_2 (189.47) which was on a par with N_0 (183.63) and N_1 (187.58). In the case of the main effect of P, the highest value was noticed for P_2 (180.60) which was on par with P_0 (168.75) and P_1 (176.33). For the main effect of K, the highest value was observed at K_2 (190.45) which was on a par with K_0 (187.40) and K_1 (187.83).

The interaction between N × P, P × K and N × K were found to be significant. For the interaction between N and P, the highest value was recorded for N_1P_1 (227.7) which was on a par with N_0P_2 (220.80). Similarly the lowest value was noticed for N_1P_2 (131.10) which was at par with N_2P_1 (140.19).

For the interaction between N × K, the highest value was noticed for N_2K_0 (244.76) which was on a par with N_0K_2 (231.40). Similarly the lowest value was noticed for N_2K_2 (128.8) which was at par with N_0K_0 (147.44).

Similarly for interaction between P × K, the highest value was recorded for P_2K_2 (218.50) which was on a par with P_1K_0 (213.90) and P_0K_0 (197.41)

while the lowest value was observed for P_1K_2 (151.71) which was at par with P_1K_1 (167.90), P_2K_1 (167.90) and P_0K_2 (171.60).

From the results on dehydrogenease activity, it was observed that the treatments imposed significant effects. The highest value was noted for T_9 (296.70) which was on a par with T_{25} (273.50) while T_{16} (41.4) recorded the lowest value which was significantly different from other treatments.

4.5.2.5. Cellulase activity

Except for N, the main effects of P and K were found to be significantly different. For the main effect of N, the highest value was noticed for N_0 (26.63) which was on a par with N_1 (22.66) and N_2 (25.97). In the case of main effect of P, the highest value was noticed for P_2 (30.63) followed by P_1 (26.02) and P_0 (24.61). For the main effect of K, the highest value was noticed for K_2 (26.86) which was on par with K_1 (26.13) and the lowest value was recorded for K_0 (22.27).

The interactions N × P, P × K and N × K revealed significant effects for all combinations with respect to cellulase activity. For the interaction between N and P, the highest value was observed for N_2P_2 (36.58) which was on a par with N_2P_0 (32.66). Similarly the lowest value was observed for N_1P_2 (18.83) which was at par with N_1P_1 (20.25), N_1P_0 (21.00), N_0P_2 (22.50) N_2P_1 (22.66) and N_0P_1 (25.08).

For the interaction between N and K, the highest value was recorded for N_1K_0 (35.50) which was at par with N_0K_2 (32.16) and the lowest value was noticed for N_0K_0 (14.16) which was on a par with N_0K_1 (20.50).

Table 23 : *Effect of Treatment on the biological characters, at the PME, CRS, Balaramapuram*

Sl. No.	Factor	**Urease** (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	**Phosphatase** (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	**Protease** (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	**Dehydrogenase** (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	**Cellulase** (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	**Respiratory** (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	$N_0P_0K_0$	93.10	43.15	130.00	83.53	18.00	2.3
2.	$N_0P_0K_1$	259.55	29.95	170.00	241.50	32.00	2.2
3.	$N_0P_0K_2$	99.75	41.60	140.00	216.80	28.75	2.5
4.	$N_0P_1K_0$	93.10	45.75	140.00	158.70	10.75	2.6
5.	$N_0P_1K_1$	73.15	53.20	80.00	131.10	30.75	2.5
6.	$N_0P_1K_2$	79.80	41.60	130.00	220.80	21.50	2.8
7.	$N_0P_2K_0$	139.80	32.45	170.00	200.10	13.75	3.0
8.	$N_0P_2K_1$	73.15	28.30	90.00	103.50	43.75	3.3
9.	$N_0P_2K_2$	299.25	96.50	310.00	296.70	40.50	3.1
10.	$N_1P_0K_0$	272.95	93.20	270.00	260.30	13.00	3.1
11.	$N_1P_0K_1$	99.75	28.30	110.00	138.00	32.25	3.3
12.	$N_1P_0K_2$	113.05	43.20	190.00	186.30	30.00	2.6
13.	$N_1P_1K_0$	239.40	46.60	260.00	248.40	31.75	2.6
14.	$N_1P_1K_1$	232.75	47.40	240.00	255.30	17.75	2.7
15.	$N_1P_1K_2$	73.15	56.55	90.00	179.40	11.25	2.0
16.	$N_1P_2K_0$	66.50	18.30	70.00	41.40	16.75	2.6
17.	$N_1P_2K_1$	86.45	45.75	90.00	151.49	14.25	2.7

Effect of treatment on the biological characters, at the PME, CRS, Balaramapuram (Contd)

Sl. No.	Factor	**Urease** (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	**Phosphatase** (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	**Protease** (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	**Dehydrogenase** (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	**Cellulase** (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	**Respiratory** (µg of CO_2 evolved g^{-1} of soil hr^{-1})
18.	$N_1P_2K_2$	246.05	65.75	150.00	227.70	37.00	3.0
19.	$N_2P_0K_0$	259.65	60.60	150.00	248.40	31.00	2.8
20.	$N_2P_0K_1$	279.30	67.40	220.00	262.20	20.75	1.9
21.	$N_2P_0K_2$	86.45	20.75	110.00	151.80	15.75	2.2
22.	$N_2P_1K_0$	99.75	59.05	160.00	172.50	26.75	2.3
23.	$N_2P_1K_1$	79.80	63.20	60.00	117.30	16.75	2.4
24.	$N_2P_1K_2$	66.50	35.70	160.00	103.50	13.00	3.0
25.	$N_2P_2K_0$	253.00	58.25	240.00	273.30	38.75	3.1
26.	$N_2P_2K_1$	46.55	34.90	160.00	200.15	27.00	3.2
27.	$N_2P_2K_2$	146.30	18.30	230.00	131.10	44.00	2.8
	CD	23.40	11.30	41.41	28.36	8.44	0.4

Similarly for the P × K interaction, the highest value was noticed for P_2K_2 (40.5) which was significantly different from other combinations and the lowest value was noticed for P_2K_1 (15.25) which was on a par with P_0K_0 (20.66) and P_1K_1 (21.75).

The treatments imposed significant effect with respect to cellulase activity. The highest activity was noticed for T_{27} (44.00) which was on a par with T_8 (43.75), T_9 (40.50), T_{25} (38.75) and T_{18} (37.00) while the lowest value was recorded for T_4 (10.75) which was on par with T_{15} (11.25), T_{10} (13.00), T_{24} (13.00), T_7 (13.75), T_{17} (14.25), T_{21} (15.75), T_{16} (16.75), T_{23} (16.75), T_{14} (17.75) and T_1 (18.00).

4.5.2.6. Respiratory activity

Except for P, the main effects of N and K did not impose any significant difference. For the main effect of N, the highest value was noticed for N_0 and N_1 (2.7) which was on a par with N_2 (2.6). In the case of P, the highest value was noticed for P_2 (3.0) which was significantly different from P_0 (2.5) and P_1 (2.5), whereas the main effect was not significantly different at all the three levels of K.

The interactions N × P, P × K and N × K showed significant effect with respect to the respiratory activity. For the interaction between N and P, the highest value for respiratory activity was noticed for the treatment combination N_2P_0 (3.1) which was on a par with other combinations and the lowest value was noticed for N_0P_0 (2.3) which was on a par with N_0P_2 (2.30), N_1P_1 (2.40), N_1P_0 (2.60) and N_1P_2 (2.60).

For the interaction between N and K, the highest value was recorded for N_1K_1 (2.9) which was found to be on a par with all other combinations except N_2K_1 (2.5). Similarly the lowest value was noticed for N_2K_1 (2.5) and N_1K_2 (2.5) which were on a par with all other treatment combinations except N_1K_1 (2.9).

For the interaction between P and K, the highest value was recorded for P_1K_2 (3.0) and P_2K_2 (3.0) which was on a par with P_0K_2 (2.9) while the lowest value was recorded for P_2K_0 (2.4) which was at par with P_0K_0 (2.7), P_1K_0 (2.5), P_0K_1 (2.5) P_1K_1 (2.5) and P_2K_1 (2.6).

From the results it was inferred that the treatments imposed significant effect with respect to the repiratory activity. The highest value was noticed for T_8 (3.30) and T_{11} (3.30) which were on a par with T_{26} (3.20), T_{25} (3.10), T_9 (3.10), T_{10} (3.10), T_{18} (3.00) T_{24} (3.00) and T_7 (3.00) while the lowest value was noticed for T_{20} (1.90).

4.5.3. Correlation analysis

Correlation analysis was carried out between the enzyme activities and soil parameters to find out the functional relationship between them. The results of the correlation analysis are presented below in Table 24.

A positive and significant correlation was observed between urease activity with available N (0.40) while a negative and non significant correlation was observed with AEC (-0.29). The correlation was positive and non significant with soil parameters like pH (0.11), EC (0.03), OC (0.03), available

Table 24 : *Correlation matrix (Soil parameter Vs Enzyme activities) at the PME, CRS, Balaramapuram*

Enzyme activity	pH	EC	Organic carbon	CEC	AEC	Available N	Available P	Available K	Clay content	Sesqui oxides	Respiratory activity
Urease activity	0.11	0.03	0.03	-0.05	-0.29*	0.40**	0.01	0.02	-0.07	0.11	0.08
Phosphatase activity	0.03	0.30	0.12	0.03	-0.38*	0.16	0.56**	0.14	-0.13	-0.06	0.04
Protease activity	-0.08	0.01	0.16	-0.06	-0.24	0.33*	0.05	0.09	-0.09	0.18	0.17
Cellulase activity	0.05	-0.40	-0.32*	0.09	-0.01	0.12	-0.10	0.01	-0.08	0.33*	0.21
Dehydrogenase activity	0.12	0.15	0.20	-0.01	-0.21	0.28*	0.17	0.24	-0.07	0.16	0.03

t52 at 5% - 0.26
at 1% - 0.35

P (0.01), available K (0.02), sesquioxides (0.11) and repiratory activity (0.08). A negative and non significant correlation was observed for urease activity with CEC (-0.05) and clay content (-0.07).

A positive and significant correlation was observed between available P (0.56) and phosphatase activity. The correlation between phosphatase and AEC (-0.38) was negative and significant.

The correlations for phosphatase with parameters like pH (0.03), EC (0.30), OC (0.12), CEC (0.03), available N (0.16), available K (0.14) and respiratory activity (0.04) were positive but non significant. A negative and non significant correlation was observed with clay content (-0.13) and sesquioxides (-0.06).

A positive and significant correlation was observed between protease activity and available N (0.33). Similarly a positive but non significant correlation was observed for protease activity with EC (0.01), OC (0.16), available P (0.05), available K (0.09), sesquioxides (0.18) and respiratory activity (0.17).

The correlation for protease activity with pH (-0.08), CEC (-0.06), AEC (-0.24) and clay content (-0.09) were negative and non significant.

A positive and significant correlation was observed between dehydrogenase activity and available N (0.28). Similarly the correlations were positive but non significant with pH (0.12), EC (0.15), OC (0.20), available P (0.17) available K (0.24), sesquioxides (0.16) and respiratory acitivity (0.03).

Negative and non significant correlations were observed with CEC (-0.01), AEC (-0.21) and clay content (-0.07).

A positive and significant correlation was observed between cellulase activity and sesquioxides (0.33) while with organic carbon (-0.32), the correlation was negative and significant.

The correlations were positive and non significant with pH (0.05), CEC (0.09), available N (0.12), available K (0.01) and respiratory activity (0.21). Correlations obtained for cellulase with EC (-0.40), AEC (-0.01), available P (-0.10) and clay content (-0.08) were negative and non significant.

4.5.4. Regression analysis

Multiple regression analysis was carried out to study the functional relationship between soil parameters and enzyme activities and the results are presented in Table 25.

The results of the regression analysis showed that the urease activity was influenced to the maximum extent by the clay content (-2.18), AEC (-2.57) and available N (+3.15). The relationship with other parameters like pH, EC, OC, CEC, available P, available K, respiratory activity and sesquioxides were found to be non significant.

Out of the 11 parameters studied, it was observed that the phosphatase activity was associated significantly with clay content (-2.09), AEC (-2.55), and available P (+4.13) status of the samples.

In the case of protease activity, the parameters OC (+2.12), clay content

Table25 : *Multiple regression analysis (Soil parameters Vs Enzyme activities at the PME, CRS, Balaramapauram*

Sl. No.	Parameters	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase
1.	pH	+38.42 (+0.99)	-6.07 (-0.70)	-2.45 -0.07)	+50.54 (+1.50)	+8.32 (+1.49)
2.	EC	-0.52 (0.50)	+0.40 (+1.93)	-0.27 (-0.35)	-0.20 (-0.25)	-0.41 (-3.08)**
3.	OC	+99.22 (+1.09)	-33.53 (-1.82)	+143.99 (+2.12)*	+145.53 (+2.03)*	-10.61 (-0.90)
4.	Clay content	-9.89 (-2.18)*	-1.91 (-2.09)*	-8.78 (-2.60)*	-6.02 (-1.68)	-0.28 (-0.48)
5.	CEC	-6.92 (-0.37)	+1.62 (-0.44	-7.90 (-0.58)	-11.17 (-0.77)	-1.45 (-0.61)
6.	AEC	-91.72 (-2.57)*	-18.42 (-2.55)*	-81.08 (-3.05)**	-60.60 (-2.16)*	+0.87 (+0.18)
7.	Av. N	0.67 (+3.15)**	+0.07 (+1.76)	+0.46 (-2.93)**	+0.41 (-2.43)*	+0.04 (+0.18)
8.	Av. P	-0.68 (-0.79)	+0.71 (+4.13)*	-0.74 (-1.16)	-0.17 (-0.25)	+0.11 (+1.04)
9.	Av. K	+0.36 (+0.59)	+0.20 (+1.64)	+0.43 (+0.95)	+0.97 (+2.01)*	-0.01 (-0.01)
10.	Respiratory activity	+3.15 (+1.17)	+1.09 (+2.00)	+3.60 (+1.79)	+1.92 (+0.90)	+1.92 (+0.54)
11.	Sexquioxides	+7.35 (+1.42)	-0.78 (-0.75)	+8.85 (+2.30)*	+8.06 (+1.98)	+1.23 (+1.84)
	R2	34.96	55.47	42.21	37.81	34.85

Values within paranthesis represents t - Values

(-2.60), AEC (-3.05), available N (-2.93), sesquioxides (+2.30) were significantly related to the activity.

The dehydrogenase activity was found to be influenced significantly by the factors OC (+2.03), AEC (-2.16), available N (+2.43) and available K (+2.01) while with the other parameters the relationship was non significant.

Out of the 11 parameters studied EC (-3.08) was found to be the factor closely associated with cellulase activity.

4.6. Path coefficient analysis

Path coefficient analysis was carried out to assess the direct and indirect effects of the soil parameters viz., pH, EC, OC, clay content, CEC, AEC, available N, available P, available K, respiratory activity and sesquioxides on the activity of urease, phosphatase, protease, dehydrogenase and cellulase. The results of the analysis are presented below.

4.6.1. Permanent manurial experiment on rice at the RRS, Kayamkulam

The correlation matrix between soil parameters and the enzyme activity is presented in Table 4 and the path coefficient analysis showing the direct and indirect effects of these parameters on enzyme activity is given in Tables 26 and 27.

From the path coefficient analysis, it was observed that the maximum positive direct effect on urease activity with a significant positive correlation was noticed in the case of available N (+0.54). However, the negative indirect

effects of factors EC (-0.01), OC (-0.08) and AEC (-0.01) have substantially reduced the effect of this factor to (+0.32). The direct effects of sesquioxides (+0.37), available N (+0.32) and respiratory activity (+0.26) also was positive and influenced the urease activity substantially.

From the results, it is also clear that highly significant positive correlations of available P (+0.49), K (+0.49) and CEC (+0.47) were influenced negatively by the indirect effects of factors like OC (-0.02) and sesquioxides (-0.17) in the case of available P, pH (-0.001), clay content (-0.004) and AEC (-0.02) in the case of CEC. However the main effects were dominant as observed from the direct positive effects of these factors. The maximum indirect effects were exhibited by the factors, available P (+0.17) and respiratory activity (+0.14).

In the case of phosphatase activity, direct positive effects were expressed through CEC (+0.36), pH (+0.35), available K (+0.32) and respiratory activity (+0.19). However, highly significant positive correlations were observed in the case of available P (+0.82), available K (+0.39) and also respiratory activity (+0.42). These effects were reduced considerably by the indirect negative effects of the factors like OC (-0.03) and sesquioxides (-0.002) for available P, pH (-0.01), EC (-0.08), OC (-0.001) and clay content (-0.02) for available K and OC (-0.04), clay content (-0.003), AEC (-0.003) and sesquioxides (-0.003) for respiratory activity. Indirect effects on phosphatase activity was mainly through available K, in the case of its association with sesquioxides, available N and EC. In general, the cumulative indirect effects were less significant for phosphatase activity.

Table 26 : *Direct effects of parameters on enzyme activities - PME on rice at the RRS, Kayamkulam*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.01 (8)	+0.35 (2)	+0.25 (2)	+0.07 (8)	-0.06 (8)
2.	EC	-0.05 (9)	-0.42 (11)	-0.37 (11)	-0.06 (9)	-0.13 (10)
3.	OC	-0.05 (10)	-0.08 (10)	-0.00 (9)	+0.23 (4)	+0.01 (7)
4.	Clay content	+0.20 (6)	+0.17 (5)	-0.07 (10)	-0.07 (10)	-0.10 (9)
5.	CEC	+0.21 (5)	+0.36 (1)	+0.17 (5)	+0.29 (2)	+0.28 (3)
6.	AEC	-0.13 (11)	-0.03 (9)	+0.12 (7)	+0.13 (6)	+0.24 (5)
7.	Av. N	+0.32 (2)	+0.12 (6)	+0.08 (8)	+0.14 (5)	+0.27 (4)
8.	Av. P	+0.21 (4)	+0.04 (8)	+0.31 (1)	+0.35 (1)	-0.20 (1)
9.	Av. K	+0.11 (7)	+0.32 (3)	+0.14 (6)	+0.24 (3)	+0.07 (6)
10.	Respiratory activity	+0.26 (3)	+0.19 (4)	+0.17 (4)	-0.39 (11)	+0.38 (1)
11.	Sesquioxides	+0.37 (1)	+0.00 (7)	+0.21 (3)	+0.10 (7)	+0.32 (2)
12.	Residual R^2 =		41.02	42.52	42.13	50.92

Data on path coefficient analysis with respect to protease activity clearly indicate direct positive effects exerted by factors like available P (+0.31), pH (+0.25), sesquioxides (+0.21) and respiratory activity (+0.17) in the decreasing order. The maximum indirect effect of pH was brought out through available P (+0.13). In the case of sesquioxides though the correlation coefficient was negative, the direct effects were made positive through the additive direct positive effects of the factors such as OC (+0.04), CEC (+0.14), AEC (+0.5) and available N (+0.19). Similarly in the case of respiratory activity, the correlation coefficient was significant and positive (+0.44). This was considerably reduced by the negative indirect effects of factors like OC (-0.005) and sesquioxides (-0.11). The correlation coefficient in the case of protease activity and available P was highly significant and positive (+0.72). This positive correlation was reduced substantially through the indirect negative effects of factors OC (-0.003), clay content (-0.007), AEC (-0.0076) and sesquioxides (-0.09).

The maximum direct effects on dehydrogenase activity was brought about by factors like available P (+0.35), CEC (+0.29), available K (+0.24) and OC (+0.23). However from the correlation matrix highly significant positive correlation was obtained for CEC, available P and available K. The highly significant positive correlation observed for CEC (+0.50) was reduced through the negative indirect effects of factors like EC (-0.05) and available P (-0.03).

Similarly significant positive correlations observed for available P and K also was suppressed to some extent by the negative influence of factors like clay content (-0.001), AEC (-0.01) and sesquioxides (-0.15) in the case of

Table 27 : *Indirect effects of soil parameters on enzyme activities - PME on rice at the RRS, Kayamkulam*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	0.08 (x_8)	0.06 (x_{10})	0.13 (x_8)	0.15 (x_8)	0.12 (x_{10})
2.	EC	0.10 (x_{11})	0.13 (x_5)	0.06 (x_5)	0.10 (x_5)	0.10 (x_5)
3.	OC	0.16 (x_{10})	0.11 (x_1)	0.14 (x_8)	0.16 (x_8)	0.23 (x_{10})
4.	Clay content	0.00 (x_8)	0.03 (x_1)	0.07 (x_6)	0.08 (x_6)	0.14 (x_6)
5.	CEC	0.11 (x_{11})	0.10 (x_9)	0.05 (x_8)	0.07 (x_9)	0.11 (x_{11})
6.	AEC	0.11 (x_4)	0.10 (x_4)	0.03 (x_5)	0.06 (x_5)	0.06 (x_5)
7.	Av. N	0.09 (x_{10})	0.10 (x_5)	0.11 (x_8)	0.13 (x_8)	0.14 (x_{10})
8.	Av. P	0.17 (x_{10})	0.29 (x_9)	0.13 (x_9)	0.10 (x_3)	0.25 (x_{10})
9.	Av. K	0.12 (x_8)	0.11 (x_5)	0.28 (x_8)	0.32 (x_8)	0.08 (x_5)
10.	Respiratory activity	0.14 (x_8)	0.11 (x_1)	0.21 (x_8)	0.24 (x_8)	0.10 (x_7)
11.	Sesquioxides	0.07 (x_5)	0.12 (x_5)	0.21 (x_{11})	0.20 (x_{10})	0.10 (x_5)
12.	Residual R^2 =	40.06	41.02	42.52	42.13	50.92

available P and EC (-0.02), AEC (-0.03) and available P (-0.18) in the case of available K. Maximum indirect effects were contributed towards dehydrogenase activity through available K (+0.32) respiratory activity (+0.24) and sesquioxides (+0.20) which was effected through available P in both the cases of available K and respiratory activity while in the case of sesquioxide the indirect effects were through respiratory activity.

Correlation matrix between soil parameters and cellulase activity indicated significant positive values with respect to CEC (+0.50), available N (+0.42) and sesquioxides (+0.36). However path coefficient analysis revealed maximum direct effects contributed through respiratory activity (+0.38), sesquioxides (+0.32), CEC (+0.28) and available N (+0.27) in the decreasing order. Maximum indirect effects were observed by available P (+0.25) and OC (+0.23). Thus the indirect negative effects of factors EC (-0.07), AEC (-0.05) and available N (-0.04) have reduced the effect of CEC while an enhancement of the correlation coefficient was observed in the case of respiratory activity. The indirect negative effects of factors such as CEC (-0.01) have further reduced the positive effects of available N. Hence the correlation coefficient was reduced considerably in this case.

4.6.2. Permanent manurial experiment on rice (Variety - Jaya) at the RARS, Pattambi

The results of the path coefficient analysis carried out to study the direct and indirect effects of the factors on enzyme activities are presented in Tables 28 and 29.

In the case of urease, a significant positive correlation with pH (0.61), CEC (0.49), available N (0.69), available P (0.76), available K (0.59), respiratory activity (0.53) and sesquioxides (0.57) was noticed. The direct effects of these factors such as available P (+0.35), respiratory activity (+0.33), CEC (+0.32) and sesquioxides (+0.26) were found to be maximum with respect to urease activity. The negative indirect effects of the factors such as EC (-0.005), OC (-0.006), clay content (-0.01), AEC (-0.13) and available N (-0.0013) on CEC, factors such as available N (-0.003), available K (-0.03) and respiratory activity (-0.04) on sesquioxides, factors such as OC (-0.008), clay content (-0.0003), available N (-0.003) and available K (-0.05) on available P were found to nullify the positive effects thus reducing the values of correlation coefficient considerably.

In the case of indirect effects, the maximum values were expressed by the factors like available K (+0.26), respiratory activity (+0.20) and CEC (+0.20). The indirect effects were expressed through the factors such as available P, CEC and respiratory activity.

In the case of phosphatase activity, significant and positive correlation with CEC (+0.35) and available N (+0.46) was observed. The maximum direct effects of the factors was noticed with available N (+0.70), CEC (+0.36), EC (+0.20) and available P (+0.09). Not much difference with respect to the correlation coefficient of the CEC was noticed. In the case of available N, an enhancement of the value was noticed due to the additive effects of the factors such as clay content (+0.006), CEC (+0.09) and available P (+0.07).

Table 28 : *Direct effects of soil parameters on enzyme activities - PME - I at the RARS Pattambi*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.12 (5)	-0.04 (5)	+0.16 (4)	-0.01 (6)	-0.08 (8)
2.	EC	-0.11 (10)	+0.20 (3)	+0.01 (8)	-0.28 (10)	-0.15 (9)
3.	OC	-0.02 (8)	-0.22 (9)	+0.12 (6)	+0.04 (5)	-0.03 (7)
4.	Clay content	+0.03 (6)	-0.07 (8)	+0.10 (7)	-0.08 (7)	-0.16 (10)
5.	CEC	+0.32 (3)	+0.36 (2)	+0.13 (5)	+0.35 (3)	+0.27 (2)
6.	AEC	-0.30 (11)	+0.00 (5)	-0.25 (10)	-0.11 (8)	-0.40 (11)
7.	Av. N	-0.00 (7)	+0.70 (1)	-0.18 (9)	-0.34 (11)	+0.01 (6)
8.	Av. P	+0.35 (1)	+0.09 (4)	+0.60 (1)	+0.54 (1)	+0.17 (4)
9.	Av. K	-0.07 (9)	-0.05 (7)	-0.29 (11)	-0.11 (9)	+0.09 (5)
10.	Respiratory activity	+0.33 (2)	-0.32 (11)	+0.44 (2)	+0.50 (2)	+0.33 (1)
11.	Sesquioxides	+0.26 (4)	-0.27 (10)	+0.22 (3)	+0.11 (4)	+0.26 (3)
12.	Residual R^2 =	16.59	58.66	25.38	15.18	35.14

The maximum indirect effects of the factors were observed with available K (+0.46), available P (+0.56) and sesquioxides (+0.43) which expressed their indirect effects through the parameter available P.

In the case of protease activity, the maximum direct effects were expressed by the factors available P (+0.60), respiratory activity (+0.44), sesquioxides (+0.22) and pH (+0.16). A significant reduction in the value of the correlation coefficient of available P was due to the negative indirect effects of factors like EC (-0.002), clay content (-0.001), available N (-0.14) and available K (-0.22). Similarly a reduction in the correlation coefficient was observed in the case of respiratory activity due to the negative indirect effects of the factors, clay content (-0.04), AEC (-0.08), available N (-0.06), available K (-0.05) and sesquioxides (-0.03). Factors such as EC (-0.001), OC (-0.002), available N (-0.11), available K (-0.15) and respiratory activity (-0.6) imposed significant negative effect on sesquioxides, thus nullifying the positive direct effects of the factors. In the case of pH also, significant reduction in the correlation coefficient due to the factors such as clay content (-0.01), available N (-0.07) and available K (-0.15) was noticed.

The factors such as available N (+0.48), sesquioxides (+0.37) and available K (+0.44) imposed their maximum indirect effects through available P.

In the case of dehydrogenase, the direct effects were exerted by factors such as available P (+0.54), respiratory activity (+0.50) and CEC (+0.35). Significant positive correlation observed in the case of CEC (+0.70) have

Table 29 : *Indirect effects of soil parameters on enzyme activities - PME-I at the RARS, Pattambi*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.18 (x_8)	+0.30 (x_7)	+0.19 (x_{10})	0.27 (x_8)	0.14 (x_{10})
2.	EC	+0.03 (x_{10})	+0.02 (x_{11})	+0.04 (x_{10})	0.05 (x_{10})	0.03 (x_{10})
3.	OC	+0.14 (x_{10})	+0.18 (x_7)	+0.19 (x_8)	0.22 (x_{10})	0.14 (x_{10})
4.	Clay content	+0.15 (x_6)	+0.14 (x_{10})	+0.12 (x_6)	0.05 (x_6)	0.20 (x_6)
5.	CEC	+0.20 (x_{10})	+0.18 (x_7)	+0.27 (x_{10})	0.31 (x_{10})	0.20 (x_{10})
6.	AEC	+0.14 (x_5)	+0.16 (x_5)	+0.14 (x_{10})	0.16 (x_{10})	0.12 (x_5)
7.	Av. N	+0.16 (x_{11})	+0.09 (x_5)	+0.48 (x_8)	0.16 (x_{10})	0.16 (x_{11})
8.	Av. P	+0.16 (x_{11})	+0.56 (x_7)	+0.14 (x_{10})	0.16 (x_{10})	0.16 (x_{11})
9.	Av. K	+0.26 (x_8)	+0.46 (x_7)	+0.44 (x_8)	0.06 (x_{11})	0.17 (x_6)
10.	Respiratory activity	+0.20 (x_5)	+0.23 (x_7)	+0.19 (x_8)	0.22 (x_5)	0.07 (x_4)
11.	Sesquioxides	0.21 (x_8)	0.43 (x_7)	+0.37 (x_8)	0.33 (x_8)	0.12 (x_6)
12.	Residual R^2 =	16.59	58.66	25.38	15.18	35.14

been considerably reduced due to the impact of factors such as EC (-0.01), pH (-0.002), available N (-0.09) and through their negative indirect effects. With respect to respiratory activity, a reduction in the correlation coefficient was noticed due to the negative effects of the factors such as pH (-0.005), EC (-0.02), AEC (-0.03), available N (-0.11), available K (-0.02) and sesquioxides (-0.01).

The factor such as available K (-0.07), imposed indirect negative effects on the correlation coefficient of available N thus reducing the value to a negative figure.

The maximum indirect effects were exerted by the factors, sesquioxides (+0.33), CEC (+0.31) and pH (+0.27) through the factors available P, respiratory activity and available P respectively.

For cellulase activity, the direct effects were exerted by the factors such as respiratory activity (+0.33), CEC (+0.27) and sesquioxides (+0.26). A significant and positive correlation between cellulase activity and CEC (+0.40) was considerably reduced due to the negative indirect effects of the factors EC (-0.007), OC (-0.009), AEC (-0.18) and available K (-0.009). Even in the case of respiratory activity, the nullifying effects of the factors, pH (-0.03), EC (-0.01), OC (-0.01), AEC (-0.12) and sesquioxides (-0.03) was evident thus reducing the value of the correlation coefficient to a lower value. Factors such as pH (-0.03) and respiratory activity (-0.04) imposed significant negative indirect effects on sesquioxides.

The negative indirect effects of the factors such as EC and OC was

found to reduce the correlation coefficient with respect to pH. In the case of available N also, a similar reduction in the value of correlation coefficient was attributed to the negative indirect effects of factors OC and pH. The negative indirect effects of the factors, pH and OC was found to buffer the direct effects, thus tending the correlation coefficient to a lower value. Factors such as pH, OC, clay content and CEC were found to impose their negative indirect effects as evident from the lower values of correlation coefficient with respect to available K.

The indirect effects of the factors such as CEC (+0.20), clay content (+0.20) and available K (+0.17) was evident through the factors respiratory activity, organic carbon and AEC respectively.

4.6.3. Permanent manurial experiment on rice (Variety - Jyothi) at the PME, RARS, Pattambi

The results of the path coefficient analysis carried out to study the direct and indirect effects of the factors are presented in Tables 30 to 31.

In the case of urease activity, the maximum direct effects were imposed by the factors such as available P (+0.57), EC (+0.25) and sesquioxides (+0.23). The negative indirect effects of the factors such as EC (-0.02) was found to reduce the value of correlation coefficient with respect to available P. The factors such as CEC (-0.03) and respiratory activity (-0.11) were found to nullify the positive effects in the case of available P. An enhancement of the value due to the factors such as pH (+0.003), available K (+0.003), respiratory

activity (+0.01) and sesquioxides (+0.06) was observed with respect to EC. The factors such as pH (-0.004), OC (-0.007), clay content (-0.02), CEC (-0.01) and AEC (-0.02) was observed to reduce the value of correlation coefficient in the case of sesquioxides.

In the case of indirect effects, the maximum effects were exerted by the factors, available K (+0.44), sesquioxides (+0.32) and OC (+0.15). These effects were expressed through the factor available P for all these three parameters.

In the case of phosphatase, a significant positive correlation was observed with available K (+0.44). This value of the correlation coefficient was considerably reduced to a lower value due to the negative indirect effects of the factors such as pH (-0.09) and sesquioxides (-0.20). The factors such as available P (+0.69), EC (+0.30) and available N (+0.27) were found to influence through their maximum directs effects on phosphatase activity. An enhancement of the value due to factors such as OC (+0.02), clay content (+0.002), available N (+0.06), available K (+0.006) and respiratory activity (+0.01) on available P was observed. Similarly, an enhancement of correlation coefficient due to factors such as CEC (+0.09), AEC (+0.06), respiratory activity (+0.02) and available K (+0.002) on EC was noticed. The correlation coefficient between the phosphatase activity and available N was found to be unaltered due to the direct and indirect effects of the factors studied.

In the case of indirect effects, the maximum value was noticed due to the factors such as available K (+0.53), sesquioxides (+0.39) and OC (+0.18) which exerted their effects through available P.

Table 30 : *Direct effects of soil parameters on enzyme activities - PME - II at the RARS, Pattambi*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.03 (10)	-0.34 (10)	+0.32 (2)	+0.09 (4)	+0.09 (5)
2.	EC	+0.25 (2)	+0.30 (2)	-0.21 (11)	-0.53 (11)	-0.34 (11)
3.	OC	+0.19 (4)	+0.09 (6)	-0.03 (9)	+0.08 (5)	+0.02 (7)
4.	Clay content	+0.10 (8)	+0.25 (4)	-0.03 (8)	-0.13 (10)	+0.03 (6)
5.	CEC	+0.16 (5)	-0.27 (9)	+0.29 (3)	+0.49 (1)	-0.05 (8)
6.	AEC	+0.10 (7)	-0.15 (8)	+0.07 (7)	-0.05 (9)	-0.08 (10)
7.	Av. N	+0.05 (11)	+0.27 (3)	-0.06 (10)	-0.02 (7)	-0.07 (9)
8.	Av. P	+0.57 (1)	+0.69 (1)	+0.16 (6)	-0.03 (8)	+0.48 (1)
9.	Av. K	+0.01 (9)	+0.00 (7)	+0.27 (4)	+0.43 (2)	+0.10 (4)
10.	Respiratory activity	+0.12 (6)	+0.16 (5)	+0.22 (5)	+0.02 (6)	+0.20 (3)
11.	Sesquioxides	+0.23 (3)	-0.43 (11)	+0.38 (1)	+0.21 (3)	+0.27 (2)
12.	Residual R^2 =	19.79	58.15	26.55	18.26	30.96

In the case of protease activity, the maximum direct effects were exerted by the factors such as sesquioxides (+0.38), pH (+0.32) and CEC (+0.29).

A reduction in the value of correlation coefficient was noticed in the case of sesquioxides due to the factors pH (-0.03), EC (-0.06), clay content (-0.008), CEC (-0.03), AEC (-0.01) and available N (-0.003). Similarly, a reduction in the value of correlation coefficient observed was due to the negative effects of factors such as pH (-0.003), OC (-0.01), available K (-0.06) and sesquioxides (-0.04) with respect to CEC, while in the case of pH, not much difference in the correlation coefficient was noticed. The reduction in the value of correlation coefficient of available P was due to the negative indirect effects factors such as OC (-0.08) and available N (-0.01). Factors such as EC (-0.05), OC (-0.001), CEC (-0.06), available N (-0.01) and respiratory activity (-0.02) imposed significant negative effects thereby reducing the value of correlation coefficient of available K.

From the results, it was inferred that the maximum indirect effects were exerted by the factors available P (+0.21) and available K (+0.18). The aforesaid indirect effects were expressed through the factors such as available K and sesquioxides.

In the case of dehydrogenase activity, the maximum direct effects were expressed by the factors such as CEC (+0.49), available K (+0.43) and sesquioxides (+0.21). Significant reduction in the value of correlation coefficient was observed due to pH (-0.001), clay content (-0.008), AEC (-0.008), available K (-0.09) and sesquioxides (-0.05) on CEC. The nullifying

Table 31 : *Indirect effects of soil parameters on enzyme activities - PME-II at the RARS, Pattambi*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	0.13 (x_8)	+0.16 (x_8)	+0.07 (x_9)	+0.12 (x_9)	+0.11 (x_8)
2.	EC	0.06 (x_{11})	+0.09 (x_5)	+0.11 (x_{11})	+0.10 (x_9)	+0.08 (x_{11})
3.	OC	0.15 (x_8)	+0.18 (x_8)	+0.10 (x_5)	+0.20 (x_2)	+0.13 (x_2)
4.	Clay content	0.01 (x_6)	+0.11 (x_{11})	+0.05 (x_2)	+0.13 (x_2)	+0.08 (x_2)
5.	CEC	0.12 (x_8)	+0.14 (x_8)	+0.08 (x_{10})	+0.19 (x_2)	+0.12 (x_2)
6.	AEC	0.05 (x_8)	+0.10 (x_{11})	+0.09 (x_2)	+0.23 (x_2)	+0.15 (x_2)
7.	Av. N	0.14 (x_8)	+0.17 (x_8)	+0.07 (x_2)	+0.18 (x_2)	+0.12 (x_8)
8.	Av. P	0.13 (x_{11})	+0.06 (x_7)	+0.21 (x_9)	+0.33 (x_9)	+0.15 (x_{11})
9.	Av. K	0.44 (x_8)	+0.53 (x_8)	+0.18 (x_{11})	+0.10 (x_{11})	+0.37 (x_8)
10.	Respiratory activity	0.05 (x_5)	+0.06 (x_8)	+0.10 (x_5)	+0.18 (x_5)	+0.00 (x_7)
11.	Sesquioxides	0.32 (x_8)	+0.39 (x_8)	+0.13 (x_9)	+0.20 (x_9)	+0.27 (x_8)
12.	Residual R^2 =	19.79	58.15	26.55	18.26	30.96

effects of the factors such as AEC (-0.01), available N (-0.007) and sesquioxides (-0.008) were found to reduce the value of correlation coefficient of organic carbon. An enhancement of the value due to the positive effects of the factors such as pH (+0.02), OC (+0.003), available P (+0.02) and sesquioxides (+0.10) on available K was also evident.

Similarly the maximum indirect effects were exerted due to the factors such as available P (+0.33), followed by AEC (+0.23) and sesquioxides (+0.20). These factors were found to impose their indirect effects through available K, EC and available K respectively.

In the case of cellulase activity, the maximum direct effects were exerted by the factors such as available P (+0.48), sesquioxides (+0.27) and respiratory activity (+0.20). A reduction in the value due to the factors such as CEC (-0.011), AEC (-0.008) and available N (-0.01) was noticed in the case of available P. Similarly, in the case of sequioxides a reduction in the value of correlation was noticed due to pH (-0.02), EC (-0.09), OC (-0.00), clay content (-0.009) and available N (-0.003). Factors such as EC (-0.08), AEC (-0.007), available N (-0.01) and respiratory activity (-0.01) decreased the value of correlation coefficient in the case of available K.

The maximum indirect effects were noticed with the factors available K (+0.37), sesquioxides (+0.27) and available P (+0.15). These effects were expressed through the factors such as available P for both available K and sesquioxides. The indirect effect on available P (+0.15) was expressed by the factor, sesquioxides.

4.6.4. Spacing cum intercropping experiment in rubber at the CES, Chethackal

Path coefficient analysis was carried out to find out the direct and indirect effects of the factors on enzyme activities. The results of the analysis are presented in Tables 32 and 33.

In the case of urease, the maximum direct effects were exerted by the factors available N (+0.81), available K (+0.40), pH (+0.15) and OC (+0.12). The increase in the correlation coefficient of available N (-0.12) and available K (+0.03) can be attributed to the additive effects of the factors such as pH (+0.009), OC (+0.02) and CEC (+0.0012) for available N and pH (+0.02), EC (+0.006), OC (+0.05) and AEC (+0.02) for available K. A positive but non significant correlation observed between urease activity and pH (+0.30) was reduced due to the negative effects of the factors EC (-0.07), AEC (-0.002) and sesquioxides (-0.03).

The maximum indirect effects were exhibited by the factors sesquioxides (+0.39) and respiratory activity (+0.34). These indirect effects of the aforesaid factors were imposed through respiratory activity and clay content respectively.

The factors such as available N (+1.33), available K (+1.06), CEC (+0.65) and respiratory activity (-0.27) were found to impose maximum direct effects on phosphatase activity. The additive effect of the factor CEC (+0.006) on available N, factors such as EC (+0.003) and AEC (+0.04) on available K, factors such as EC (+0.19), AEC (+0.03) and available N (+0.01) on CEC were noticed from the study thus enhancing the correlation coefficient to a

higher value. In the case of respiratory activity the correlation coefficient was reduced from (+0.11) to (-0.27) due to the negative indirect effects of the factors OC (-0.64), clay content (-0.07), AEC (-0.01) and available P (-0.15).

The maximum indirect effects of the factors EC (+0.58), sesquioxides (+0.57) and clay content (+0.56) were exhibited thorough the factor clay content in both the case of EC and sesquioxides and available N in the case of clay content. Thus the prominence of the positive direct effects of factors is observed in the present study.

For the protease activity, factors such as available N (+24.23), sesquioxides (+23.20), available K (+16.47) and CEC (+10.84) were found to influence their positive direct effects.

A positive correlation coefficient observed in the case of available N (+0.04) has been increased to a very high value due to the positive effect of the factors such as clay content (+10.72) and CEC (+10.84). The increase in the value of correlation coefficient between phosphatase and available K (+0.35) was due to the positive indirect effects of the factor EC (+7.4) whereas in the case of respiratory activity (+0.34) the negative indirect effects of the factors such as OC (-8.15), clay content (-1.27), AEC (-0.16) and available P (-2.74) reduced the values of correlation coefficient. Similarly the negative indirect effects of the factors such as pH (-0.47), EC (-1.49), available P (-2.17) and respiratory activity (-7.22) on organic carbon was noticed.

In the case of protease, the maximum indirect effects were expressed by the factors clay content (+10.17), respiratory activity (+8.81) and OC

Table 32 : *Direct effects of soil parameters on enzyme activities - Intercropping experiment at the CES, Chethackal*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.15 (3)	-0.68 (8)	-5.99 (6)	-2.15 (5)	+0.86 (7)
2.	EC	-0.68 (9)	-0.32 (5)	-7.48 (7)	-2.69 (6)	+1.27 (6)
3.	OC	+0.12 (4)	-0.90 (9)	-11.48 (10)	-4.24 (9)	+1.39 (4)
4.	Clay content	-0.82 (10)	-1.39 (11)	-25.52 (11)	-9.57 (11)	+4.37 (1)
5.	CEC	+0.11 (5)	+0.65 (3)	+10.84 (4)	3.85 (3)	-1.51 (9)
6.	AEC	-0.23 (6)	-0.38 (6)	-4.20 (5)	-1.71 (4)	+0.63 (8)
7.	Av. N	+0.81 (1)	+1.33 (1)	+24.23 (1)	9.09 (1)	-4.11 (11)
8.	Av. P	-0.26 (7)	-0.44 (7)	-8.06 (8)	-3.23 (7)	+1.29 (5)
9.	Av. K	+0.40 (2)	+1.06 (2)	+16.47 (3)	+6.12 (2)	-2.18 (10)
10.	Respiratory activity	-1.03 (11)	-0.27 (4)	-10.17 (9)	-3.54 (8)	+2.10 (3)
11.	Sesquioxides	-0.66 (8)	-1.17 (10)	+23.20 (2)	-8.43 (10)	+4.13 (2)
12.	Residual R^2 =	61.20	85.38	11.48	219.54	96.05

(+7.42). The indirect effects were expressed through the factors available N, clay content and sesquioxides respectively.

In the case of dehydrogenase activity, the direct effect of the factors such as available N (+9.09), available K (+6.12) and CEC (+3.85) is evident from the present study. The additive effect of the factor CEC (+0.03) on available N was due to the positive effect of this factor. Similarly factors such as EC (-0.02) and AEC (+0.18) imposed their positive direct effects on available K while factors such as pH (-0.53), OC (-0.50), clay content (-0.28), available P (-0.42), available K (-1.53), respiratory activity (-0.07) and sesquioxides (-1.01) imposed their significant negative effects on CEC thus nullifying the positive effects.

The maximum indirect effects were expressed by EC (+4.02), sesquioxides (+3.92) and clay content (+3.82) which imposed their effects through clay content in the case of EC and sesquioxides. The indirect effect of clay content was expressed through available nitrogen.

With respect to cellulase, the direct effects of the factors such as clay content (+4.37), sesquioxides (+4.13) and respiratory activity (+2.10) were prominent. The highly significant and positive correlation observed between cellulase and available K (+0.32) has been considerably reduced to a very low value due to the nullifying effects of the factors EC (-0.01) and AEC (-0.06). An enhancement of the correlation coefficient was observed in the case of clay content and sesquioxides was evident. Factors such as AEC (+0.05) and respiratory activity (+0.10) in the case of clay content and factors such as pH

Table 33 : *Indirect effects of soil parameters on enzyme activities - Intercropping experiment at the CES, Chethackal*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.13 (x_4)	+0.22 (x_4)	+4.08 (x_4)	+1.53 (x_4)	+0.53 (x_{11})
2.	EC	+0.27 (x_7)	+0.58 (x_4)	+1.72 (x_4)	+4.02 (x_4)	+0.70 (x_{11})
3.	OC	+0.20 (x_{11})	+0.45 (x_9)	+7.42 (x_{11})	+2.69 (x_{11})	+1.49 (x_{10})
4.	Clay content	+0.34 (x_7)	+0.56 (x_7)	+10.17 (x_7)	+3.82 (x_7)	+0.10 (x_{10})
5.	CEC	+0.04 (x_2)	+0.03 (x_2)	+0.44 (x_2)	+0.53 (x_1)	+0.54 (x_9)
6.	AEC	+0.10 (x_7)	+0.32 (x_{11})	+6.49 (x_{11})	+2.36 (x_{11})	+0.34 (x_4)
7.	Av. N	+0.02 (x_3)	+0.00 (x_5)	+0.10 (x_5)	+0.12 (x_1)	+1.83 (x_4)
8.	Av. P	+0.14 (x_{11})	+0.25 (x_{11})	+5.10 (x_{11})	+1.85 (x_{11})	+0.71 (x_{10})
9.	Av. K	+0.05 (x_3)	+0.04 (x_6)	+0.46 (x_6)	+0.34 (x_1)	+0.60 (x_3)
10.	Respiratory activity	+0.34 (x_4)	+0.44 (x_{11})	+8.81 (x_{11})	+3.20 (x_{11})	+0.99 (x_3)
11.	Sesquioxides	+0.39 (x_{10})	+0.57 (x_4)	+3.86 (x_{10})	+3.92 (x_4)	+0.21 (x_2)
12.	Residual R^2 =	61.20	85.38	63.41	219.54	96.05

(+0.11) and EC (+0.21) on sesquioxides were found to impose positive effects on sesquioxides.

The maximum indirect effects of the factors were through available N (+1.83), OC (+1.49) and respiratory activity (+0.99). These effects were expressed through clay content, respiratory activity and organic carbon.

4.6.5. Permanent manurial experiment on coconut at the CRS, Balaramapuram

The results of the path coefficient analysis which revealed the direct and indirect effects of the factors are presented in Tables 34 and 35.

From the path coefficient analysis, it was noticed that the maximum direct effects were exerted by the factors available N (+0.42), sesquioxides (+0.21) and organic carbon (+0.18) with respect to urease activity. A highly significant and positive correlation between urease activity and available N (+0.40) was noticed from the Table 24. An additive effect by the positive effect of the factors OC (+0.01), clay content (+0.04), respiratory activity (+0.02) and available P (+0.02) have increased the value to (+0.42). Similarly from the data, it was inferred that the positive effect of the factors like available P (+0.1), available K (+0.2) and respiratory activity (+0.3) imposed an additive effect thereby increasing the coefficient from (+0.11) to (+0.21) in the case of sesquioxides.

In the case of indirect effects, the maximum effects were imposed by the factors such as clay content (+0.11), CEC (+0.07) and available P (+0.07).

Table 34 : *Direct effects of soil parameters on enzyme activities - PME at the CRS, Balaramapuram*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.11 (4)	-0.01 (9)	-0.00 (6)	+0.20 (5)	+0.20 (2)
2.	EC	+0.07 (6)	+0.19 (4)	-0.05 (7)	-0.03 (7)	-0.44 (11)
3.	OC	+0.18 (3)	-0.50 (11)	+0.34 (2)	+0.34 (1)	-0.15 (10)
4.	Clay content	-0.30 (10)	-0.05 (10)	-0.33 (10)	-0.22 (10)	-0.06 (8)
5.	CEC	-0.05 (8)	+0.00 (8)	-0.08 (8)	-0.11 (9)	-0.09 (9)
6.	AEC	-0.35 (11)	+0.65 (2)	-0.39 (11)	-0.28 (11)	+0.02 (6)
7.	Av. N	+0.42 (1)	+0.28 (3)	+0.38 (1)	+0.32 (2)	+0.20 (3)
8.	Av. P	-0.12 (9)	+0.87 (1)	-0.16 (9)	-0.03 (8)	+0.15 (4)
9.	Av. K	+0.06 (7)	+0.07 (6)	+0.12 (5)	+0.26 (4)	-0.01 (7)
10.	Respiratory activity	-0.15 (5)	+0.06 (7)	+0.23 (4)	+0.11 (6)	+0.07 (5)
11.	Sesquioxides	+0.21 (2)	+0.18 (5)	+0.32 (3)	+0.29 (3)	+0.29 (1)
12.	Residual R^2 =	66.66	19.48	59.28	63.90	66.25

These indirect effects were exerted through the factors available N, available N and AEC respectively. From the results, it was also observed that the direct effects of the factors were dominant with respect to urease activity.

In the case of phosphatase activity, the maximum direct effects were imposed by the factors available P (+0.87), AEC (+0.65), available N (+0.28) and EC (+0.19). An enhancement of values of the correlation coefficients due to the positive influence of the factors such as EC (+0.05), available K (+0.004) and sesquioxides (+0.02) on available P, pH (+0.02), clay content (+0.08), available K (+0.08) and respiratory activity (+0.09) on AEC and the factors like EC (+0.02), respiratory activity (+0.006) and sesquioxides (+0.001) on available N was noticed. In the case of indirect effects, the maximum effect was exerted through the factors OC (+0.41), sesquioxides (+0.20) and CEC (+0.20). The above indirect effects were exerted through the factors such as available P, organic carbon and organic carbon respectively.

In the case of protease, the maximum direct effects were observed through the factors available N (+0.38), OC (+0.34), sesquioxides (+0.32) followed by respiratory activity (+0.23). From the Table 24 it was observed that a highly significant and positive correlation existed between available N and protease activity (+0.33). The additive effects of the factors OC (+0.03), AEC (+0.6), available P (+0.005) and respiratory activity (+0.002) were found to increase the coefficient for available N. Similarly the direct effects factors clay content (+0.05), CEC (+0.33) and available N (+0.003) were found to increase the value of the correlation coefficient in the case of OC. The increase

in the correlation coefficient in the case of sesquioxides and respiratory activity also, can be attributed to the indirect positive effects of the factors like EC (+0.003), available P (+0.02), available K (+0.03) and respiratory activity (+0.05) for sesquioxides and EC (+0.01), CEC (+0.01), available N (+0.003) sesquioxides (+0.07) for the respiratory activity.

The maximum indirect effects were exerted by the factors available P (+0.16), clay content (+0.09) and EC (+0.09). These effects were expressed through the factors OC, available N and OC respectively.

For dehydrogenase, factors OC (+0.34), available N (+0.32), sesquioxides (+0.29) and available K (+0.26) were found to exert their maximum direct effects. A significant negative correlation (-0.32), in the case of OC was increased to a positive value due to the positive indirect effects of the factors such as clay content (+0.36), CEC (0.45) and available N (+0.003). The significant reduction in the correlation coefficient of sesquioxide from (0.33) as observed from Table 24 to (+0.29) can be attributed to the negative indirect effects of the factors like pH (-0.006), OC (-0.14), clay content (-0.05), CEC (-0.02), available N (-0.01) and available P (-0.003).

In the case of the indirect effects, the role of factors such as available P (+0.06), EC (+0.09) and available K (+0.09) was remarkable. These indirect effects were expressed through the factors OC, OC and sesquioxides respectively. The analysis of the data revealed the prominent role of the direct positive effects of these factors.

Table 35 : *Indirect effects of soil parameters on enzyme activities - PME at the CRS, Balaramapuram*

Sl. No.	Factor	Urease	Phosphatase	Protease	Dehydrogenase	Cellulase
1.	pH	+0.05 (x_6)	+0.08 (x_3)	+0.06 (x_6)	+0.04 (x_6)	+0.01 (x_7)
2.	EC	+0.05 (x_7)	+0.03 (x_7)	+0.09 (x_3)	+0.09 (x_3)	+0.06 (x_1)
3.	OC	+0.04 (x_4)	+0.41 (x_8)	+0.05 (x_4)	+0.04 (x_5)	+0.07 (x_8)
4.	Clay content	+0.11 (x_7)	+0.08 (x_3)	+0.09 (x_7)	+0.08 (x_7)	+0.07 (x_{11})
5.	CEC	+0.07 (x_7)	+0.20 (x_3)	+0.08 (x_{11})	+0.07 (x_{11})	+0.07 (x_{11})
6.	AEC	+0.04 (x_4)	+0.00 (x_{10})	+0.05 (x_4)	+0.01 (x_{11})	+0.02 (x_2)
7.	Av. N	+0.05 (x_6)	+0.02 (x_2)	+0.06 (x_6)	+0.04 (x_6)	+0.01 (x_1)
8.	Av. P	+0.07 (x_6)	+0.05 (x_2)	+0.16 (x_3)	+0.16 (x_3)	+0.00 (x_5)
9.	Av. K	+0.06 (x_9)	+0.07 (x_6)	+0.10 (x_{11})	+0.09 (x_{11})	+0.09 (x_{11})
10.	Respiratory activity	+0.04 (x_{11})	+0.09 (x_6)	+0.07 (x_{11})	+0.06 (x_{11})	+0.10 (x_2)
11.	Sesquioxides	0.03 (x_{10})	0.20 (x_3)	0.05 (x_{10})	0.08 (x_9)	0.06 (x_3)
12.	Residual R^2 =	66.61	19.48	59.28	63.90	66.25

For cellulase, the maximum direct effect of factors sesquioxides (+0.29), pH (+0.20), available N (+0.20) and available P (+0.15) was observed to influence the activity. The results revealed a significant reduction in the correlation coefficient available N from (+0.28) which might be attributed to the negative indirect effects of the factors EC (-0.05), OC (-0.001), clay content (-0.01), CEC (-0.01), AEC (-0.004), available P (-0.004) and sesquioxides (-0.003). The increase in the correlation coefficient of pH was noticed due to the positive indirect effects of the factors such as OC (+0.02), clay content (+0.002), available N (+0.01) and available K (+0.001). Similarly, in the case of sesquioxides also the positive indirect effects of the factors such as EC (+0.03), OC (+0.06), AEC (+0.001) and respiratory activity (+0.01) was observed. But in the case of available P, reduction in the correlation coefficient from (+0.17) to (+0.15) was noticed due to the nullifying effects due to the negative indirect effects of pH (-0.004), EC (-0.11), OC (-0.07), AEC (-0.005), available K (-0.001) and respiratory activity (-0.005).

The maximum indirect effects were observed for the factors respiratory activity (+0.10), available K (+0.09) and CEC (+0.07) which were expressed through the factors OC, sesquioxides and sesquioxides respectively.

4.6.6. Enzyme kinetics

Enzyme kinetic studies revealed variability in the kinetic parameters (V_{max} and K_m) for enzymes in the samples collected from different locations of

the permanent manurial experiments. Table 36 represents values of the kinetic parameters worked out for these soils. The data was treated assuming a Michaelis-Menten type of kinetic behaviour and the values were worked out based on Line Weaver - Burk plot.

The lowest value for Km in the case of urease was found in the case of Kayamkulam (2.4×10^{-3}) and Pattambi-I (2.4×10^{-3}) followed by Chethackal (2.7×10^{-3}), Balaramapuram (2.91×10^{-3}) and Pattambi-II (3.2×10^{-3}) in the increasing order. The values for phosphatase was high at Pattambi-II (4.5×10^{-3}) followed by Kayamkulam (0.8×10^{-3}), Pattambi-I (0.77×10^{-3}), Balaramapuram (0.75×10^{-3}) and Chethackal (0.75×10^{-3}).

The highest values for protease was registered in the case of Balaramapuram and Chethackal (1.9×10^{-3}), followed by Pattambi-I (1.5×10^{-3}), Pattambi-II (1.45×10^{-3}) and Kayamkulam (1.4×10^{-3}). The values observed in the case of dehydrogenase were 6.15×10^{-3}, 6×10^{-3}, 5.9×10^{-3}, 5.9×10^{-3}, 0.6×10^{-3} for Kayamkulam, Pattambi-I, Pattambi-II, Chethackal and Balaramapuram respectively. The highest values observed for phosphatase and cellulase were 3600 and 2800 respectively for Pattambi-I and Pattambi-II.

Values for V_{max} also varied widely with respect to different locations and is evident from Table 36.

Table 36 : *Kinetic parameters (V_{max} and K_m Values) of Soil enzymes*

	K_m Values (M)					V_{max}				
Location	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase*	Urease	Phosphatase	Protease	Dehydro-genase	Cellulase
KLM	2.4×10^{-3}	0.83×10^{-3}	1.4×10^{-3}	6.15×10^{-3}	2200	8.6×10^{-4}	0.59	1.56×10^{-5}	1.4	3.0
PTB-I	2.4×10^{-3}	0.77×10^{-3}	1.5×10^{-3}	6.00×10^{-3}	3600	7.75×10^{-4}	0.37	1.2×10^{-5}	1.03	3.65
PTB-II	3.2×10^{-3}	4.5×10^{-3}	1.45×10^{-3}	5.9×10^{-3}	2800	1.06×10^{-4}	0.36	1.18×10^{-5}	1.16	4.05
BLM	2.91×10^{-3}	0.75×10^{-3}	1.9×10^{-3}	0.6×10^{-3}	1500	9.6×10^{-4}	0.39	1.92×10^{-5}	0.96	2.7
CTL	2.7×10^{-3}	0.75×10^{-3}	1.9×10^{-3}	5.9×10^{-3}	2400	1.55×10^{-4}	0.46	2.64×10^{-5}	1.74	5.55

* Value in ppm

Units for V_{max}

Urease - Moles of urea hydrolysed g^{-1} of soil hr^{-1}

Phosphatase - µg of PNP released g^{-1} of soil hr^{-1}

Protease - µM of amino N hydrolysed g^{-1} of soil hr^{-1}

Dehydrogenase - µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}

Cellulase - ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}

4.7. Pot culture studies

Pot culture studies were carried out to assess the effect of liming, manures, fertilizers and agro chemicals viz., insecticides, fungicides and herbicides on the activity of five major soil enzymes. Soil samples were collected at the three critical stages of crop growth viz., active tillering, panicle initiation and harvest for the enzyme activity assay was done. The results of the analysis are presented below in Table 37 to 41. Analysis of the same for the initial sample before treatment application is also presented in Table 37 for comparison.

4.7.1. Effect of agro chemicals

4.7.1.1. Active tillering stage

4.7.1.1.1. Urease activity

The assay of urease activity at the active tillering revealed that the treatments had a significant effect on the activity. The mean values ranged between 35.5to 119.7 ppm urea hydrolysed g^{-1} of soil hr^{-1}. The highest value was recorded for the treatment T_1 (119.7) without the application of any chemical (control). This was followed by the treatments T_3 (86.56) and T_2 (84.29) and were found to be on a par. Similarly treatments T_4 (61.03), T_5 (60.96), T_6 (53.2), T_{12} (51.03), T_{13} (51.03), T_{10} (49.93) and T_{11} (48.80) were found to be at par while the lowest value was recorded in T_7 (35.50).

Table 37 : *Activities of enzymes in the pot experiment (I and II) - Zero sampling stage.*

Sl. No.	*Enzyme*	*Activity*
1.	Urease	141.23 (ppm of urea hydrolysed g^{-1} of soil hr^{-1})
2.	Phosphatase	88.04 (µ of p-nitrophenol released g^{-1} of soil hr^{-1})
3.	Protease	99.10 (Micromoles of amino N hydrolysed g^{-1} of soil hr^{-1})
4.	Dehydrogenase	275.81 (µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1})
5.	Cellulase	61.22 (ppm of glucose hydrolysed g^{-1} of soil hr^{-1})
6.	Microbial biomass	11.77 (µ of CO_2 evolved g^{-1} of soil hr^{-1})

4.7.1.1.2. Phosphatase activity

The treatments had a significant effect with respect to the phosphatase activity and the mean values for the phosphatase activity ranged between 24.4 to 68.75 µg of p-nitrophenol released g^{-1} of soil hr^{-1}. The highest value was recorded for T_1 (68.75) in the control which was significantly superior compared to other treatments. The treatments T_3 (39.95), T_5 (35.48), T_2 (35.31), T_4 (34.4), T_7 (31.08), T_6 (29.93) and T_{12} (28.88) were found to be on a par. The lowest value for phosphatase activity was recorded for T_9 (24.40) which was found be at par with the treatments T_8 (24.41), T_{13} (25.88), T_{10} (26.60) and T_{11} (27.70).

4.7.1.1.3. Protease activity

The results of the protease activity assay revealed that the treatments had a significant effect on the activity. The mean values for the enzyme ranged between 16.66 to 76.66 micromoles of amino nitrogen hydrolysed per g^{-1} of soil hr^{-1}. The highest value was recorded for T_1 (76.66) in the control which was significantly superior compared to other treatments. The treatments T_2 (43.33), T_5 (43.33), T_6 (40.00), T_{11} (33.33), T_{13} (33.33), T_{12} (30.00), T_3 (30.00), T_4 (30.00) and T_{10} (26.66) were found to be on a par, while the lowest activity was noticed for T_7 (16.66).

4.7.1.1.4. Dehydrogenase activity

From the results of the dehydrogenase activity, it was inferred that the treatments imposed highly significant effect on the activity. The mean values of the dehydrogenase activity ranged between 35.73 to 241.20 µg of TPF

Table 38 : *Activities of the enzymes - Pot culture experiment-I - Rice (Active Tillering)*

Sl. No.	Treat-ments	Urease activity (ppm of urea hydrolysed g^{1} of soil hr^{-1})	Phosphatase activity (mg of p-nitro phenol released g^{-1} of hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil $hour^{-1}$)	Dehydrogenases activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial Biomass (Microgram of CO_2 evolved g^{-1} of soil hr^{-1})
1.	T_1	119.70	68.75	76.66	241.2	38.33	10.33
2.	T_2	84.29	35.31	43.33	116.46	21.66	3.00
3.	T_3	86.56	39.95	30.00	80.40	15.00	2.00
4.	T_4	61.03	34.40	30.00	104.80	13.33	4.00
5.	T_5	60.96	35.48	43.33	116.13	21.66	4.00
6.	T_6	53.20	29.93	40.00	107.20	20.00	6.00
7.	T_7	35.50	31.08	16.66	78.00	8.33	3.00
8.	T_8	42.16	24.41	16.66	73.60	8.33	2.00
9.	T_9	39.93	24.40	20.00	53.60	10.00	2.66
10.	T_{10}	49.93	26.60	26.66	53.60	10.00	4.00
11.	T_{11}	48.80	27.70	33.33	71.46	11.66	4.66
12.	T_{12}	51.03	28.88	30.00	35.73	13.33	5.00
13.	T_{13}	51.03	25.88	33.33	53.60	11.66	3.00
	S E	5.60	3.88	6.91	17.70	3.66	1.08
	C D	16.28	11.30	20.11	51.47	10.66	3.16

IH1652 201

hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded for T_1 (241.2) the control which was significantly superior to all other treatments while treatments T_2 (116.46), T_5 (116.13), T_6 (107.2), T_4 (104.80), T_3 (80.4), T_7 (78.0), T_8 (73.6) and T_1 (71.46) were on a par. The lowest value was recorded for T_{12} (35.73) which was at par with the treatments T_{10} (53.6), T_{13} (53.6) and T_9 (53.60).

4.7.1.1.5. Cellulase activity

From the results, it was inferred that the treatments showed a significant effect on the cellulase activity and the mean values ranged between 8.33 to 38.33 ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}. The highest value was recorded for T_1 (38.33) which differed significantly from other treatments. Treatments T_2 (21.66), T_5 (21.66), T_6 (20.00), T_3 (15.0), T_4 (13.33), T_{12} (13.33), T_{11} (11.66) and T_{13} (11.66) were found to be on a par. The lowest value was recorded for T_8 (8.33) and T_7 (8.33) and was found to be at par with the treatments T_9 (10.00), T_{10} (10.00), T_{13} (11.66), T_{11} (11.66), T_{12} (13.33), T_4 (13.33) and T_3 (15.00).

4.7.1.1.6. Microbial bio-mass

The estimated microbial biomass indicated a highly significant effect due to treatment application. The mean values for the microbial load expressed in terms of µg CO_2 g^{-1} of soil hr^{-1} ranged between 2 to 10.33. The highest value was recorded for T_1 (10.33) in the control which was significantly superior than other treatments. Treatments T_6 (6.00), T_{12} (5.00), T_{11} (4.66), T_5 (4.00),

T_{10} (4.00), T_4 (4.00), T_7 (3.33), T_2 (3.00) and T_{13} (3.00) were found to be on a par, while the lowest values were recorded for T_8 (2.00) and T_3 (2.00).

4.7.1.2. Panicle initiation stage

4.7.1.2.1. Urease activity

The results of the analysis did not show any significant effect on the activity due to treatment application. The mean values for the urease activity ranged between 79.8 to 137.63 ppm urea hydrolysed g^{-1} of soil hr^{-1}. However the highest value was recorded for T_1 (137.63) in the control which did not differ significantly from other treatments.

4.7.1.2.2. Phosphatase activity

The results revealed that the treatments imposed significant effect with respect to phosphatase activity. The mean values ranged between 28.05 to 66.30 µg of p-nitrophenol released g^{-1} of soil hr^{-1}. The highest value was observed for the control T_1 (66.30) which was significantly superior compared to other treatments. Treatments T_5 (53.55), T_3 (51.00), T_2 (48.45), T_6 (48.45), T_7 (45.90), T_{10} (45.90), T_{13} (45.90), T_{12} (43.35) and T_{11} (40.80) were found to be on a par. The lowest value was recorded for T_9 (28.05) which was at par with T_4 (30.6) and T_8 (33.15).

4.7.1.2.3. Protease activity

The results of the protease activity assay showed that the treatment

Table 39 : *Activities of the enzymes - Pot culture experiment-I - Rice (Panicle Initation)*

Sl. No.	Treat-ments	Urease activity (ppm of urea hydrolysed g^{1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenol phosphate g^{-1} of soil hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil $hour^{-1}$)	Dehydrogenases activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial Biomass (Microgram of CO_2 evolved g^{-1} of soil hr^{-1})
1.	T_1	137.63	66.30	93.33	265.60	55.00	11.66
2.	T_2	97.53	48.45	61.66	119.60	33.33	6.33
3.	T_3	93.10	51.00	51.66	149.46	33.33	5.66
4.	T_4	97.53	30.60	56.66	134.53	48.33	6.66
5.	T_5	97.53	53.55	50.00	158.66	35.00	7.33
6.	T_6	79.80	48.45	43.33	169.46	53.33	8.33
7.	T_7	93.1	45.90	36.66	163.73	36.66	7.66
8.	T_8	93.10	33.15	43.33	169.46	51.66	11.33
9.	T_9	88.66	28.05	46.66	171.20	45.00	11.66
10.	T_{10}	106.40	45.90	60.00	160.30	51.66	11.00
11.	T_{11}	106.40	40.80	60.00	134.53	50.00	12.33
12.	T_{12}	101.96	43.35	43.33	169.4	43.33	10.00
13.	T_{13}	103.10	45.90	48.33	165.2	48.33	9.66
	S E	N.S	4.35	N.S	12.49	4.52	1.17
	C D	N.S	12.67	N.S	36.33	13.16	3.41

effects did not vary significantly with respect to the protease activity. The mean values ranged between 36.66 to 93.33 micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}. The highest value was recorded for the treatment T_1 (93.33) which was on a par with other treatments.

4.7.1.2.4. Dehydrogenase activity

The treatments imposed a significant effect on the dehydrogenase activity and the mean values ranged between 119.6 to 265.60 µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}.

The highest value was recorded for T_1 (265.60) in the control which was followed by the treatments T_6 (169.46), T_8 (169.46), T_{12} (169.4), T_{13} (165.20), T_7 (163.73), T_{10} (160.30), T_5 (158.66) and T_3 (149.46) in the descending order. The lowest value was recorded for T_2 (119.6) which was significantly lower than other treatments.

4.7.1.2.5. Cellulase activity

The results of the study revealed that the effects due to treatments vary significantly with respect to the cellulase activity. The mean values for the treatments ranged between 33.33 to 55 ppm of glucose hydrolysed g^{-1} of soil hr^{-1}. The highest value for cellulase activity was noticed in T_1 (55.00) followed by treatments T_6 (53.33) which was on a par with T_8 (51.66), T_{10} (51.66), T_{11} (50.00), T_4 (48.33), T_{13} (48.33), T_9 (45.00) and T_{12} (43.33). The treatments T_2 (33.33) and T_3 (33.33) recorded the same value which were the lowest.

4.7.1.2.6. Microbial bio-mass

The results of the microbial biomass showed that the treatments imposed a significant effect. The mean values ranged between 5.66 to 12.33 µg of CO_2 evolved g^{-1} of soil hr^{-1}. The highest value was noticed for T_{11} (12.33) which was on a par with the treatments T_9 (11.66), T_1 (11.66), T_8 (11.33), T_{10} (11.0), T_{12} (10) and T_{13} (9.66). The lowest value was registered for T_3 (5.66) and was found to be at par with T_2 (6.33), T_4 (6.66), T_5 (7.33), T_7 (7.66) and T_6 (8.33).

4.7.1.3. Harvest stage

4.7.1.3.1. Urease activity

The results of the urease activity assay revealed that the treatments had produced a significant effect on the urease activity. The mean values for the Urease activity ranged between 33.76 to 78.07 ppm of urea hydrolysed g^{-1} of soil hr^{-1}.

The highest value for urease activity noticed for T_1 (78.07) in the control which was on a par with T_{12} (73.15). Treatments T_{11} (56.26), T_8 (50.64), T_{13} (47.61), T_3 (42.2) and T_7 (40.02) were found to be at par while the lowest value was recorded for T_4 (33.76).

4.7.1.3.2. Phosphatase activity

From the results of the phosphatase activity, it was inferred that the treatments influenced this parameter significantly. The mean values for the phosphatase activity ranged between 17.03 to 55.96 µg of p-nitrophenol released g^{-1} of soil hr^{-1}.

The highest value was recorded for T_1 (55.96) in the control which was significantly different from other treatments. Treatments T_3 (36.5), T_6 (35.6), T_8 (34.06), T_5 (31.36), T_2 (28.66), T_4 (27.4), T_{10} (26.9), T_{11} (24.33) and T_7 (24.06) were found to be on a par while the lowest value was noticed for T_9 (17.03).

4.7.1.3.3. Protease activity

The results of the protease activity showed highly significant effect due to treatments. The mean values for the protease activity ranged between 35 to 70 micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1}.

The highest value for protease activity was noticed for the treatments T_1 (70.00) and T_{11} (70) while treatments T_3 (63.33), T_{10} (58.33), T_6 (56.66), T_2 (53.33), T_{12} (53.33), T_{13} (50), T_5 (46.66) and T_8 (46.66) were found to be on a par. The treatment T_4 (35.00) recorded the lowest value for protease activity.

4.7.1.3.4. Dehydrogenase activity

It is evident from the results that the dehydrogenase activity varied significantly due to treatment effects. The mean values ranged between 88.77 to 120.41 µg of TPF hydrolysed g^{-1} of soil 24 hrs^{-1}.

The highest value was recorded for T_1 (120.41) followed by T_5 (106.29) which was found to be on a par with the treatments T_3 (105.1), T_{12} (104.06), T_{10} (101.96), T_{11} (100.73), T_{13} (99.43), T_9 (98.81), T_6 (97.65), T_8 (93.76) and T_2 (91.26) while the lowest value was recorded for T_4 (88.77).

Table 40 : *Activities of the enzymes - Pot culture experiment-I - Rice (Harvest)*

Sl. No.	Treat-ments	Urease activity (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase activity (µg of p-nitro phenyl phosphate released g^{-1} of hr^{-1})	Protease activity (Micromoles of amino nitrogen hydrolysed g^{-1} of soil $hour^{-1}$)	Dehydrogenases activity (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase activity (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial Biomass (Microgram of CO_2 evolved g^{-1} of soil hr^{-1})
1.	T_1	78.07	55.96	70.00	120.41	38.33	10.00
2.	T_2	50.64	28.66	53.33	91.26	30.00	3.00
3.	T_3	42.20	36.50	63.33	105.10	33.33	1.66
4.	T_4	33.76	27.40	35.00	88.77	25.00	4.00
5.	T_5	36.57	31.36	46.66	106.29	30.00	3.33
6.	T_6	39.38	35.60	56.66	97.65	30.00	2.33
7.	T_7	40.02	24.06	43.33	89.40	20.00	1.66
8.	T_8	50.64	34.06	46.66	93.76	15.00	2.66
9.	T_9	39.38	17.03	38.33	98.81	16.66	2.66
10.	T_{10}	39.38	26.90	58.33	101.96	35.00	2.00
11.	T_{11}	56.26	24.33	70.00	100.73	36.66	3.33
12.	T_{12}	73.15	19.46	53.33	104.06	16.66	4.00
13.	T_{13}	47.61	19.46	50.00	99.43	8.33	3.66
	S E	5.91	4.40	6.04	5.65	3.58	0.59
	C D	17.19	12.81	17.51	16.44	10.41	1.72

4.7.1.3.5. Cellulase activity

From the results of the cellulase activity, it was inferred that the treatments imposed a highly significant effect on the activity. The mean values ranged between 8.33 to 38.33 ppm of glucose hydrolysed g^{-1} of soil 24 hrs^{-1}.

The highest value was recorded in the control T_1 (38.33) which significantly differed from other treatments. This was followed by T_{11} (36.66) which was on a par with T_{10} (35), T_3 (33.33), T_5 (30), T_6 (30) and T_2 (30) and the lowest value was observed in the case of T_{13} (8.33).

4.7.1.3.6. Microbial bio-mass

The microbial load estimated in terms of mg of CO_2 evolved g^{-1} of soil hr^{-1} showed significant differences due to treatment effects. The mean values ranged between 1.66 to 10 mg of CO_2 evolved g^{-1} of soil hr^{-1}.

The highest value was recorded for T_1 (10), the control which was followed by T_4 (4) and T_{12} (4) and was on a par with T_{13} (3.66), T_5 (3.33), T_{11} (3.33), T_2 (3), T_8 (2.66), T_9 (2.66) and T_6 (2.33). The lowest value was recorded for the treatments T_7 (1.66) and T_3 (1.66).

4.7.1.3.7. Number of productive tillers

The results of revealed significant effects of the treatments with respect to the number of productive tillers. The maximum number of productive tillers were observed for T_9 (13) and T_4 (13) which was on a par with the treatments T_{12} (11), T_2 (11), T_5 (11), T_6 (11), T_7 (11), T_{11} (11) and T_{13} (11). The control T_1 (7) recorded the lowest number of productive tillers.

Table 41 : *Yield and yield attributes - Pot culture experiment - I*

Sl. No.	*Treatments*	*No. of Productive tillers*	*Grain yield (g pot^{-1})*	*Straw yeidl (g pot^{-1})*
1.	T_1	7	25.10	40.99
2.	T_2	11	26.30	34.92
3.	T_3	10	28.53	40.47
4.	T_4	13	28.43	40.39
5.	T_5	11	27.83	40.00
6.	T_6	11	25.73	36.99
7.	T_7	11	27.36	38.07
8.	T_8	9	25.70	37.22
9.	T_9	13	27.23	38.13
10.	T_{10}	10	25.50	35.04
11.	T_{11}	11	27.86	37.22
12.	T_{12}	11	26.26	37.18
13.	T_{13}	11	27.00	39.90
	SEd	0.66	N.S	N.S
	CD	1.91	N.S	N.S

4.7.1.3.8. Grain yield

From the data on the grain yield it was observed that the treatments did not impose any significant effect. The mean values for grain yield ranged from 25.10 to 28.53 g/pot. The highest value was recorded for T_3 (28.53) which did not differ significantly from other treatments viz., T_1 (25.10), T_2 (26.30), T_4 (28.43), T_5 (27.83), T_6 (25.73), T_7 (27.36), T_8 (25.70), T_9 (27.23), T_{10} (25.50), T_{11} (27.86), T_{12} (26.26) and T_{13} (27.00)

4.7.1.3.9. Straw yield

The results on straw yield revealed no significant effects due to the application of treatments. The highest value was noticed for the control plot (40.99) which did not differ significantly from other treatments, viz., T_2 (34.92), T_3 (40.47), T_4 (40.39), T_5 (40.00), T_6 (36.99), T_7 (38.07), T_8 (37.22), T_9 (38.13), T_{10} (35.04), T_{11} (37.22), T_{12} (37.18) and T_{13} (39.90).

From the results it was clear that a spurt in the activities of the enzymes viz, urease, phosphatase, protease, dehydrogenase and cellulase exist in the soil from the seedling stage to the panicle initiation stage. The activities were highest at the panicle initiation stage compared to the other two stages. This was followed by a sharp decline in activity during the harvest stage.

4.7.2. Effect of fertilizers, manures and lime

Pot culture studies were conducted to assess the effect of fertilizers, manures (farm yard manure, green leaf manure and vermicompost) and lime

on the enzyme activities. Soil samples collected from the experimental pots at three stages viz., active tillering, panicle initiation and at harvest were subjected to the assay of enzyme activites for five enzymes viz., urease, phosphatase, protease, dehydrogenase and cellulase. The results of the study are presented here under in tables from 42 to 53.

4.7.2.1. Active Tillering Stage

Soil samples collected at this stage were subjected to the enzyme assay and the results are presented below and in Tables 42 to 44.

4.7.2.1.1. Urease activity

The main effects of F, L and M were found to be significant. F_1 (209.89) was significantly different and superior than F_0 (173.45). Similarly the main effect of lime L_1 (222.22) was significantly different from L_0 (161.12) with the highest value noticed for L_1 (222.22). In the case of manures, the highest value was recorded for M_2 (201.72) which was significantly superior than M_3 (200.30), M_1 (189.55) and M_0 (175.12).

The interaction effects of F × M and L × M were found to be significant while F × L was non significant. In the case of interaction between F × L, the highest value for urease activity was noted for F_1L_1 (240.51) which did not significantly differ from other combinations. The interaction between F × M was found to be significant with respect to urease activity. The highest value was noticed for F_0M_3 (221.67) which was significantly superior to other

combinations. The lowest value was noticed for F_0M_0 (150.73) which was significantly lower compared to other combinations. The interaction between L × M was found to be significant for all combinations and the highest value was recorded for L_0M_3 (234.97) which was significantly different from other combinations.

The three factor interactions among fertilizers, lime and manures were found to impose significant effect on the urease activity. The highest value was recorded for $F_1L_1M_2$ (261.57) which was significantly different from other combinations while the lowest value was noticed for $F_0L_0M_0$ (106.40) which represented the absolute control.

4.7.2.1.2. Phosphatase activity

The main effects of F, L and M did not produce any significant effect on phosphatase activity. F_0 (97.14) did not differ significantly from F_1 (139.94). Similarly L_0 (123.03) also did not vary significantly from L_1 (114.05). In the case of manures the highest value was noticed for M_2 (175.85) which did not vary significantly from M_3 (101.18), M_1 (104.75) and M_0 (92.38).

The interaction between the factors F × L, L × M and F × M were found to impose significant effect with respect to phosphatase activity with the highest values recorded for the combination F_0L_1 (162.37), L_0M_1 (232.54) and F_0M_3 (247.53). Similarly, the interaction effects of F, L and M also recorded significant effect with respect to phosphatase activity for many of the treatment combinations with the highest value recorded for $F_1L_1M_2$ (125.28).

Table 42 : *Means of the main effects of the treatments on the enzyme activity and microbial biomass of soil at active tillering stage*

Factor	*Urease* (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	*Phosphatase* (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	*Protease* (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	*Dehydrogenase* (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	*Cellulase* (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	*Microbial biomass* (µg of CO_2 evolved g^{-1} of soil hr^{-1})
a. Fertilizer						
i. F_0	173.45	97.14	121.04	350.80	58.75	14.92
ii. F_1	209.89	139.94	167.29	426.08	80.21	18.92
SE	2.12	N.S	3.21	6.82	1.20	0.31
CD	6.12	N.S	9.27	19.68	3.45	0.88
b. Lime						
i. L_0	161.12	123.03	108.75	325.83	55.63	14.46
ii. L_1	222.22	114.05	179.58	451.04	83.33	19.38
SE	2.12	N.S	3.21	6.82	1.20	0.31
CD	6.12	N.S	9.27	19.68	3.45	0.88
c. Manures						
i. M_0	175.12	92.38	135.42	351.27	66.25	15.17
ii. M_1	189.55	104.75	144.17	386.72	67.50	16.42
iii. M_2	201.72	175.85	154.17	413.13	75.00	18.67
iv. M_3	200.30	101.18	142.92	402.63	69.17	17.42
SE	3.00	N.S	N.S	N.S	1.69	0.43
CD	8.65	N.S	N.S	N.S	4.88	1.25

4.7.2.1.3. Protease activity

The main effects of F and L were significant while M did not impose any significant effect with respect to protease activity. F_0 (121.04) recorded a significantly lower value than F_1 (167.29) while L_1 (179.58) was found to register the highest value superior than L_0 (108.75). The effect of manures were not comparable with the control as the effect was the same for M_0 (135.42), M_1 (144.17), M_2 (154.17) and M_3 (142.92).

The interaction between F × L, L × M and F × M were found to impose significant effects with respect to protease activity. The highest value was registered for F_1L_1 (207.50) which was significantly superior than F_1L_0 (151.67), F_0L_1 (127.08) and F_0L_0 (90.42) indicating the superiority of liming and fertilizer application over the control. In the case of F × M interactions, the values recorded for treatment combinations were 115.00, 121.67, 130.83, 116.67, 155.83, 166.67, 177.50 and 169.17 for F_0M_0, F_1M_0, F_0M_1, F_1M_1, F_0M_2, F_1M_2, F_0M_3 and F_1M_3 respectively. However, the highest value was recorded for F_0M_3 (177.50) which was found to prove its superiority over the other combinations except F_1M_2, F_1M_3. In the case of L × M interactions, the highest value was registered for L_0M_3 (189.17) which was significantly superior than other combinations except L_0M_2 (175.00), L_1M_2 (180.83) and L_1M_3 (173.33).

Three factor interaction among fertilizers, lime and manures were found to impose significant effect on protease activity. However, the highest value was noticed for the treatment $F_1L_1M_2$ (215.00) which was significantly superior compared to all other treatments except $F_1L_1M_0$, $F_1L_1M_1$ and $F_1L_1M_3$.

Table 43 : *Interaction effects between two factors of the treatments on the enzyme activity and microbial biomass at active tillering stage*

Factors	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per $24\ hrs^{-1}$)	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
A. F x L						
F_0L_0	142.96	83.69	90.42	291.33	45.00	12.58
F_1L_0	203.93	110.60	151.67	410.27	72.50	17.25
F_0L_1	179.27	162.37	127.08	360.33	66.25	16.33
F_1L_1	240.51	117.51	207.50	491.82	94.17	21.50
SE	N.S	2.01	4.55	9.65	1.8	0.20
CD	N.S	7.2	13.11	27.83	6.48	0.72
B. L x M						
L_0M_0	139.65	80.35	95.83	271.87	50.00	12.17
L_1M_0	159.65	88.12	107.50	324.73	54.17	14.33
L_0M_1	168.47	232.54	119.17	336.67	60.00	16.33
L_1M_1	176.71	90.93	112.50	370.07	58.33	15.00
L_0M_2	210.58	104.23	175.00	430.67	82.50	18.17
L_1M_2	219.45	121.39	180.83	448.70	80.83	18.50
L_0M_3	234.97	119.17	189.17	489.60	90.00	21.00
L_1M_3	223.88	111.43	173.33	435.20	80.00	19.83
SE	4.24	2.10	4.6	9.9	2.1	0.60
CD	12.23	7.56	16.56	25.64	7.56	0.21
C. F x M						
F_0M_0	150.73	82.63	115.00	303.73	53.33	13.33
F_1M_0	172.95	100.30	121.67	351.93	55.83	14.67
F_0M_1	181.77	104.18	130.83	377.47	65.00	16.83
F_1M_1	188.34	101.45	116.67	170.07	60.83	14.83
F_0M_2	199.50	102.12	155.83	430.67	79.17	17.00
F_1M_2	206.15	109.21	166.67	448.70	79.17	18.17
F_0M_3	221.67	247.53	177.50	489.60	85.00	20.50
F_1M_3	212.25	100.92	169.17	435.20	77.50	20.00
SE	4.24	2.10	4.6	9.90	2.2	0.60
CD	12.23	7.56	16.56	25.64	7.92	0.21

4.7.2.1.4. Dehydrogenase activity

The main effects of F and L were found to impose significant difference on the dehydrogenase activity while the effect of M was non significant. F_1 (426.08) was found to be significantly superior than F_0 (350.80). Similarly L_1 (451.04) was found to be superior than L_0 (325.83). In the case of manures M_1 (386.72), M_2 (413.13) and M_3 (402.63) were found to be on a par and was no way superior to M_0 (351.27).

The interaction between F × L, L × M and F × M found to give a significant effect. F × L interaction was highly significant and the maximum value was observed in the case of F_1L_1 (491.82) which was superior over all other treatments while the control F_0L_0 recorded the lowest value (291.33). The interaction effects between F × M was significant and the highest value was recorded for F_0M_3 (489.60) which was significantly superior. Similarly the interaction between L × M was also significant with L_0M_0 (271.87) and L_0M_3 (489.60) registering the lowest and highest values respectively.

Three factor interaction among fertilizers, lime and manures was found to give significant effect on dehydrogenase activity. The highest value was noticed for $F_1L_1M_2$ (534.93) which differed significantly from other treatments.

4.7.2.1.5. Cellulase activity

The main effects of the factors F, L and M were significant with respect to the cellulase activity. F_1 (80.21) and L_1 (83.33) were significantly superior to their corresponding control treatments F_0 (58.75) and L_0 (55.63). In the

Table 44 : *Effect on treatments on enzyme activities and microbial biomass at active tillering stage*

Sl. No.	Treatments	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	$F_0L_0M_0$	106.40	65.47	80.00	208.53	36.67	9.00
2.	$F_0L_0M_1$	146.40	83.10	93.33	295.87	41.67	12.33
3.	$F_0L_0M_2$	155.17	93.07	98.33	310.67	50.00	15.00
4.	$F_0L_0M_3$	163.89	93.12	90.00	350.27	51.67	14.00
5.	$F_0L_1M_0$	195.07	99.80	150.00	398.93	70.00	17.67
6.	$F_0L_1M_1$	199.50	117.50	150.00	408.00	70.00	17.00
7.	$F_0L_1M_2$	208.37	115.30	163.33	444.27	80.00	18.67
8.	$F_0L_1M_3$	212.80	109.78	143.33	389.87	70.00	15.67
9.	$F_1L_0M_0$	172.90	95.58	111.67	335.20	63.33	15.33
10.	$F_1L_0M_1$	172.90	93.13	121.67	353.60	66.67	16.33
11.	$F_1L_0M_2$	181.77	127.05	140.00	362.67	70.00	17.67
12.	$F_1L_0M_3$	189.53	88.72	135.00	389.87	65.00	16.00
13.	$F_1L_1M_0$	226.10	108.65	200.00	462.40	95.00	18.67
14.	$F_1L_1M_1$	239.40	123.03	211.67	489.40	91.67	20.00
15.	$F_1L_1M_2$	261.57	125.28	215.00	534.93	100.00	23.33
16.	$F_1L_1M_3$	234.97	113.08	203.33	480.53	90.00	24.00
	SE	6.00	3.10	4.80	6.9	4.4	0.87
	CD	17.30	10.26	17.28	24.84	15.84	2.50

case of manures M_2 (75.00) recorded the highest value which was significantly superior than the control M_0 (66.25). However treatments M_0 (66.25), M_1 (67.50) and M_3 (69.17) were on a par with respect to cellulase activity.

The two factor interactions between F × L, L × M and F × M were found to impose significant effect with respect to cellulase activity with the highest values registered for the treatments F_1L_1 (94.17), F_0M_3 (85.00) and L_0M_3 (90.00) respectively.

The effect on cellulase activity was significant with respect to the three factor interaction among fertilizers, lime and manures. The highest value was recorded for the treatment $F_1L_1M_2$ (100.00) which differed significantly from other treatments except $F_1L_1M_0$ (95.00), $F_1L_1M_1$ (91.67) and $F_1L_1M_3$ (90.00).

4.7.2.1.6 Microbial bio-mass

The main effects of F, L and M were found to be significant. F_1 (18.92) and L_1 (19.38) were significantly superior to their corresponding controls F_0 (14.92) and L_0 (14.46). In the case of manures the highest value was recorded for M_2 (18.67) and the lowest value was registered for M_0 (15.17).

The interaction between F × L, L × M and F × M produced significant effect with respect to the microbial biomass. The highest values for these interactions were registered for F_0M_3 (20.50), L_0M_3 (21.00) and F_1L_1 (21.50) respectively.

The interaction among F, L and M was found to be significant with respect to the microbial biomass. The highest value was recorded for the

treatment $F_1L_1M_3$ (24.00) which was on a par with $F_1L_1M_2$ (23.33) while the lowest value was recorded for $F_0L_0M_0$ (9.00).

4.7.2.2. Panicle initiation stage

Soil samples collected at this stage were subjected to the enzyme activity assay and the results are presented in Tables 45 to 47.

4.7.2.2.1. Urease activity

The main effects of F, L and M were found to impose significant difference with respect to urease activity. F_1 (220.15) was found to be superior over F_0 (204.51). Similarly, the effect of liming was superior and prominent than without lime. Thus, L_1 (242.87) gave a significantly higher value than the L_0 (181.79). In the case of manures, M_2 (234.14) was found to record the highest value followed by M_1 (215.02) which was on a par with M_3 (213.91) while the lowest value was recorded for M_0 (186.25).

The interactions between F × L, L × M and F × M were found to be non significant with respect to urease activity. The highest values for these interactions were observed for F_1L_1 (247.44), F_0M_3 (246.62) and L_0M_3 (266.57) respectively.

The effect of three factor interaction among F, L and M was found to be significant with respect to urease activity. The highest value was noticed for $F_1L_1M_2$ (276.00) which was significantly superior compared to other treatments.

Table 45 : *Means of the main effects of the treatments on the enzyme activity and microbial biomass of soil at panicle initiation stage.*

Factor	*Urease* (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	*Phosphatase* (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	*Protease* (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	*Dehydrogenase* (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	*Cellulase* (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	*Microbial biomass* (µg of CO_2 evolved g^{-1} of soil hr^{-1})
a. Fertilizer						
i. F_0	204.51	94.25	179.38	366.47	86.88	19.58
ii. F_1	220.15	109.11	222.92	453.11	108.96	24.50
SE	3.21	1.48	4.21	7.85	1.67	0.38
CD	9.27	4.26	12.15	22.66	4.81	1.10
b. Lime						
i. L_0	181.79	88.83	171.88	327.83	83.13	18.42
ii. L_1	242.87	114.53	230.42	491.75	112.71	25.67
SE	3.21	1.48	4.21	7.85	1.67	0.38
CD	9.27	4.26	12.15	22.66	4.81	1.10
c. Manures						
i. M_0	186.25	94.35	180.00	391.06	90.83	19.33
ii. M_1	215.02	101.36	200.00	405.11	94.17	21.08
iii. M_2	234.14	110.97	213.75	435.55	107.08	25.00
iv. M_3	213.91	100.04	210.83	407.45	99.58	22.75
SE	4.55	2.09	5.95	N.S	2.36	0.54
CD	13.11	6.02	17.18	N.S	6.80	1.56

4.7.2.2.2. Phosphatase activity

The main effects of F, L and M were found to be significant with respect to the phosphatase activity. F_1 (109.11) was superior than F_0 (94.25). Similarly L_1 (114.53) was found to be superior than L_0 (88.83). In the case of manures, the highest value was noticed for M_2 (110.97) which was significantly superior over M_3 (100.04), M_1 (101.36) and M_0 (94.35).

The interaction between F × L and L × M were found to be significant while F × M was non significant. For F × L interactions, the highest value was noticed for F_1L_1 (119.41) followed by F_1L_0 (109.65) and F_0L_1 (98.81) while the lowest value for phosphatse activity was observed for F_0L_0 (78.85).

In the case of F × M interactions, the highest value was noticed for F_0M_3 (118.97) which did not differ significantly from other combinations. For L × M interaction, the highest value was noticed for L_1M_3 (168.38) which was on a par with L_1M_2 (116.03) while the lowest value was noticed for L_0M_0 (77.78).

Significant difference among various combinations of F, L and M with respect to the phosphatase activity was noticed. The highest value was recorded for $F_1L_1M_2$ (130.83) which differed significantly from other treatments.

4.7.2.2.3. Protease activity

The main effects of F, L and M were found to impose significant difference on the protease activity. F_1 (222.92) was found to be superior over the control F_0 (179.38). Similarly the effect of liming was evident in enhancing

Table 46 : *Interaction effects between two factors of the treatments on the enzyme activity and Microbial biomass at panicle initiation stage*

Factors	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
A. F x L						
F_0L_0	170.73	78.85	157.50	295.05	71.25	16.67
F_1L_0	238.29	109.65	210.25	437.89	102.50	22.50
F_0L_1	192.85	98.81	186.25	360.82	95.00	20.17
F_1L_1	247.44	119.41	259.58	545.61	122.92	28.83
SE	N.S	2.09	5.95	6.1	2.0	0.54
CD	N.S	6.02	17.18	21.91	7.2	1.56
B. L x M						
L_0M_0	148.62	77.78	145.00	304.42	76.67	14.17
L_1M_0	188.42	86.70	177.50	318.47	83.33	18.67
L_0M_1	201.72	99.15	180.00	346.57	88.33	21.67
L_1M_1	188.42	91.70	185.00	341.88	84.17	19.17
L_0M_2	223.88	110.93	215.00	477.70	105.00	24.50
L_1M_2	241.62	116.03	225.50	491.75	105.00	23.50
L_0M_3	266.57	122.79	247.50	524.53	125.83	28.33
L_1M_3	239.40	168.38	236.67	473.02	115.00	26.33
SE	N.S	2.95	4.8	7.04	2.4	0.76
CD	N.S	8.51	17.28	25.44	8.64	2.20
C. F x M						
F_0M_0	186.30	85.42	157.50	346.57	81.67	16.50
F_1M_0	206.15	95.63	180.00	360.62	80.00	19.50
F_0M_1	221.67	102.98	192.50	379.35	94.17	22.50
F_1M_1	203.93	92.98	187.50	379.35	91.67	19.83
F_0M_2	186.20	103.28	202.50	435.55	100.00	22.17
F_1M_2	223.88	107.10	220.00	449.60	108.33	22.67
F_0M_3	246.62	118.97	235.00	491.75	120.00	27.50
F_1M_3	223.88	107.10	234.17	435.55	107.50	25.67
SE	N.S	N.S	4.8	7.04	2.4	0.76
CD	N.S	N.S	17.28	25.44	8.64	2.20

protease activity, as the treatment L_1 (230.42) was significantly superior than L_0 (171.88). In the case of manures, the highest value was noticed for M_2 (213.75) which was on a par with M_1 (200.00) and M_3 (210.83) while M_0 (180.00) recorded the lowest value.

The interaction between F × L, L × M and F × M were significant with respect to protease activity. In the case of F × L interaction, the treatment F_1L_1 (259.58) recorded the highest value which was significantly different from F_0L_0 (157.50), F_1L_0 (201.25) and F_0L_1 (186.25). For F × M interaction, the effect was significant for all combinations with the highest value recorded for F_0M_3 (235.00) which was on a par with F_1M_2 (220.00) and F_1M_3 (234.17). For L × M interaction, the highest value was noticed for the combination L_0M_3 (247.50) which was on a par with L_1M_2 and L_1M_3.

From the results it was also observed that the various combinations of F, L and M differed significantly with respect to protease activity. The highest value was noticed for $F_1L_1M_2$ (275.00) which was on a par with $F_1L_1M_3$ (263.33) while the control $F_0L_0M_0$ (130.00) recorded the lowest value.

4.7.2.2.4. Dehydrogenase activity

The main effects of F and L were found to impart significant difference while M was found to be non significant. F_1 (453.11) was found to be superior over F_0 (366.47). Similarly L_1 (491.75) had indicated a significant effect compared to L_0 (327.83), the control. In the case of manures, the effect was non significant for all levels viz., M_0 (391.06), M_1 (405.11), M_2 (435.55) and

M_3 (407.45). The interaction between F × L, L × M and F × M were found to be significant with the highest values observed for F_1L_1 (545.61), L_0M_3 (524.53) and F_0M_3 (491.75) respectively.

The results revealed no significant difference between various combinations due to interaction among F, L and M. The highest value was recorded for $F_1L_1M_2$ (599.47) which differed significantly from other treatments.

4.7.2.2.5. Cellulase activity

The main effects of F, L and M were found to be significant with respect to cellulase activity. F_1 (108.96) was found to be superior than the control F_0 (86.88). Similarly L_1 (112.71) was found to register significantly superior values than L_0 (83.13). In the case of manures M_2 (107.08) was noticed to be superior over M_3 (99.58), M_1 (94.17) and M_0 (90.83) which recorded the lowest value.

The interaction between F × L, L × M and F × M was found to be significant with the highest values recorded for F_1L_1 (122.92), L_0M_3 (125.83) and F_0M_3 (120.00).

The interaction among F, L and M produced significant difference with respect to cellulase activity. The highest value was noticed for $F_1L_1M_2$ (141.67) which differed significantly from other treatments except $F_1L_1M_3$ (123.33).

4.7.2.2.6. Microbial bio-mass

The main effects of F, L and M were found to impart significant effect

Table 47 : *Effect of treatments on enzyme activities and microbial biomass at panicle initiation stage.*

Sl. No.	Treatments	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	$F_0L_0M_0$	137.63	63.75	130.00	271.63	63.33	11.00
2.	$F_0L_0M_1$	177.33	76.50	170.00	290.00	66.66	18.33
3.	$F_0L_0M_2$	186.20	91.20	165.00	309.10	78.33	20.00
4.	$F_0L_0M_3$	181.77	83.95	165.00	309.10	76.67	17.33
5.	$F_0L_1M_0$	234.97	107.10	185.00	421.50	100.00	22.00
6.	$F_0L_1M_1$	234.97	114.75	190.00	430.87	93.33	20.67
7.	$F_0L_1M_2$	257.13	114.75	220.00	449.60	110.00	25.00
8.	$F_0L_1M_3$	226.10	102.00	210.00	449.60	106.67	22.23
9.	$F_1L_0M_0$	159.60	91.80	160.00	337.20	90.00	17.33
10.	$F_1L_0M_1$	199.50	96.90	185.00	346.57	100.00	19.00
11.	$F_1L_0M_2$	217.23	107.10	195.00	384.03	98.33	23.33
12.	$F_1L_0M_3$	195.07	99.45	205.00	374.67	91.67	21.00
13.	$F_1L_1M_0$	212.80	114.75	245.00	533.90	110.00	27.00
14.	$F_1L_1M_1$	248.27	114.30	255.00	552.63	116.67	26.33
15.	$F_1L_1M_2$	276.00	130.83	275.00	599.47	141.67	31.67
16.	$F_1L_1M_3$	252.70	114.75	263.33	496.43	123.33	30.33
	SE	6.10	3.7	4.10	6.10	5.1	0.91
	CD	17.40	13.96	14.76	21.96	18.36	3.2

with respect to the microbial biomass. F_1 (24.50) was found to be superior than F_0 (19.58). Similarly L_1 (25.67) was found to be superior than L_0 (18.42). In the case of manures, the highest value was recorded for M_2 (25.00) followed by M_3 (22.75) and M_1 (21.08) which were significantly different from M_0 (19.33).

The interaction between F × L, L × M and F × M were found to be significant. In the case of F × L interaction, the highest value was noticed for F_1L_1 (28.83), which was significantly superior than F_0L_1 (20.17), F_1L_0 (22.50) and F_0L_0 (16.67) which recorded the lowest value.

In the case of F × M interaction, the highest value was registered for F_0M_3 (27.50) which was significantly superior than F_1M_3 (25.67), F_1M_2 (22.67), F_0M_2 (22.17), F_1M_1 (19.83), F_0M_1 (22.50), F_1M_0 (19.50) and F_0M_0 (16.50). For the L × M interaction, the highest value was noticed for L_0M_3 (28.33) which was significantly superior than L_1M_2 (23.50), L_0M_2 (24.50), L_1M_1 (19.17), L_0M_1 (21.67), L_1M_0 (18.67) and L_0M_0 (14.17) which registered values which were on a par.

The interaction among F, L and M was found to imposed significant difference with respect to the microbial biomass with the highest value was recorded for $F_1L_1M_2$ (31.67) which was on par with $F_1L_1M_3$ (30.33).

4.7.2.3. Harvest stage

Soil samples collected at this stage were subjected to the enzyme activity assay and the results are presented in Tables 48 to 50.

4.7.2.3.1. Urease activity

The main effects of F, L and M were found to impose significant effect on urease activity. F_1 (114.25) was found to be superior over control F_0 (119.38) and L_1 (150.84) was superior over the corresponding control L_0 (112.79). In the case of manures, the highest value was recorded for M_2 (140.74) which was superior over M_3 (137.88) and M_1 (131.59) while the lowest value was registered for M_0 (117.05).

The interaction effects of F × L and L × M were significant while F × M interaction was non significant. For F × L interaction, the highest value was recorded for F_1L_1 (162.13) which was significantly superior over F_0L_1 (126.36), F_1L_0 (139.54) and F_0L_0 (99.22) which recorded the lowest value. In the case of F × M interaction, the values were found to be 105.31, 119.08, 125.47, 127.66, 128.79, 144.09, 156.02 and 148.09 for F_0M_0, F_1M_0, F_0M_1, F_1M_1, F_0M_2, F_1M_2, F_0M_3 and F_1M_3 respectively.

For the interaction between L × M, the highest value was noticed for L_0M_3 (162.11) which was on a par with L_1M_3 (153.62), L_1M_2 (150.20) and superior than L_0M_2 (137.43), L_1M_1 (122.13), L_0M_1 (119.38), L_1M_0 (112.97) and L_0M_0 (96.67).

The interaction among F, L and M were found to be significant with respect to urease activity. The highest value was noticed for $F_1L_1M_2$ (180.15) which was significantly superior than other treatments while the lowest value was registered for $F_0L_0M_0$ (76.27), the absolute control.

Table 48 : *Means of the main effects of the treatments on the enzyme activity and microbial biomass at harvest stage.*

Factor	*Urease* (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	*Phosphatase* (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	*Protease* (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	*Dehydrogenase* (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	*Cellulase* (ppm of glucose hydrolysed g^{-1} of soil per $24\ hrs^{-1}$)	*Microbial biomass* (µg of CO_2 evolved g^{-1} of soil hr^{-1})
a. Fertilizer						
i. F_0	119.38	90.22	82.71	160.77	47.08	8.92
ii. F_1	114.25	102.60	110.83	202.70	60.42	9.88
SE	1.73	1.70	1.97	2.86	1.21	0.23
CD	5.00	4.92	5.69	8.24	3.50	0.65
b. Lime						
i. L_0	112.79	84.17	78.54	152.48	42.08	7.71
ii. L_1	150.84	108.65	115.00	210.99	65.42	11.08
SE	1.73	1.70	1.97	2.86	1.21	0.23
CD	5.00	4.92	5.69	8.24	3.50	0.65
c. Manures						
i. M_0	117.05	90.19	88.75	169.49	47.50	9.08
ii. M_1	131.59	95.68	102.92	179.94	50.83	8.58
iii. M_2	140.74	104.42	100.42	191.35	60.42	10.58
iv. M_3	137.88	95.36	95.00	186.15	56.25	9.33
SE	2.45	2.41	2.79	4.04	1.72	0.32
CD	7.07	6.95	8.04	11.66	4.96	0.92

4.7.2.3.2. Phosphatase activity

The main effects of F, L and M were found to impose significant difference with respect to phosphatase activity. F_1 (102.60) was observed to be superior than F_0 (90.22). Similarly L_1 (108.65) was found to be superior than L_0 (84.17). In the case of manures, the highest value was noticed for M_2 (104.42) which was significantly superior than M_3 (95.36) and M_1 (95.68) while the lowest value was registered for M_0 (90.19).

The interaction between F × L, F × M and L × M were found to be significant with respect to phosphatase activity. For F × L interaction viz., F_0L_0 (78.25), F_1L_0 (102.20), F_0L_1 (90.10) and F_1L_1 (115.10), the highest value was noticed for F_1L_1 (115.10) which differed significantly from other combinations. In the case of F × M interactions, the highest value was noticed for F_0M_3 (107.83) which was significantly superior than others with the lowest value recorded for F_0M_0 (76.68). In the case of L × M interaction viz., L_0M_0 (75.92), L_1M_0 (85.70), L_0M_1 (91.60), L_1M_1 (83.48), L_0M_2 (104.47), L_1M_2 (105.67), L_0M_3 (117.23) and L_1M_3 (107.23), the highest value was noticed for L_0M_3 (117.23) which differed significantly from other combinations.

The three factor interaction among F, L and M were found to impose significant effect on phosphatase activity. The highest value was registered for $F_1L_1M_2$ (122.33) which differed significantly from other treatments except $F_0L_1M_2$, $F_1L_1M_0$ and $F_1L_1M_1$.

4.7.2.3.3. Protease activity

The main effects of F, L and M were found to impose significant

Table 49 : *Interaction effects between two factors of the treatments on the enzyme activity and microbial biomass at harvest stage*

Factor	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro-phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per $24\ hrs^{-1}$)	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
A. F x L						
F_0L_0	99.22	78.25	65.42	136.39	37.08	6.67
F_1L_0	139.54	102.20	100.00	185.15	57.08	11.17
F_0L_1	126.36	90.10	91.67	168.57	47.08	8.75
F_1L_1	162.13	115.10	130.00	236.82	73.75	11.00
SE	3.96	2.10	4.55	4.04	1.4	0.32
CD	12.08	7.86	13.11	11.66	5.04	0.92
B. L x M						
L_0M_0	96.67	75.92	65.83	142.48	37.50	7.17
L_1M_0	112.97	85.70	79.17	148.92	40.83	7.00
L_0M_1	119.38	91.60	84.17	153.06	45.00	8.67
L_1M_1	122.13	83.48	85.00	165.47	45.00	8.00
L_0M_2	137.43	104.47	111.67	196.49	57.50	11.00
L_1M_2	150.20	105.67	126.67	210.97	60.83	10.17
L_0M_3	162.11	117.23	116.67	229.65	75.83	12.50
L_1M_3	153.62	107.23	105.00	206.83	67.50	10.67
SE	3.96	2.71	3.94	5.71	2.4	0.61
CD	12.08	9.75	11.37	16.49	8.04	0.96
C. F x M						
F_0M_0	105.31	76.68	75.83	148.69	42.50	8.83
F_1M_0	119.08	88.80	84.17	161.33	44.17	8.00
F_0M_1	125.47	101.00	90.00	167.60	52.50	9.67
F_1M_1	127.66	94.42	80.83	165.47	49.17	9.17
F_0M_2	128.79	103.70	101.67	190.29	52.50	9.33
F_1M_2	144.09	102.57	121.67	198.56	57.50	9.17
F_0M_3	156.02	107.83	110.83	215.11	68.33	11.50
F_1M_3	148.09	96.30	109.17	206.83	63.33	9.50
SE	N.S	2.74	3.94	N.S	2.4	0.61
CD	N.S	9.84	11.37	N.S	8.04	0.96

difference with respect to protease activity. F_1 (110.83) and L_1 (115.00) were superior than their respective controls F_0 (82.71) and L_0 (78.54). In the case of manures, the highest value was noticed for M_1 (102.92) which was on a par with M_2 (100.42) and M_3 (95.00) while the lowest value was noticed for M_0 (88.75).

The F × L, L × M and F × M interactions were found to give significant effect on protease activity. For F × L interaction, the highest value was noticed for F_1L_1 (130.00) which differed significantly form F_0L_1 (91.67), F_1L_0 (100.00) and F_0L_0 (65.42). Similarly for F × M interaction, F_1M_2 (121.67) recorded the highest value which differed significantly from F_1M_3 (109.17), F_0M_3 (110.83), F_0M_2 (101.67), F_1M_1 (80.83), F_0M_1 (90.00), F_1M_0 (84.17) and F_0M_0 (75.83). For the interaction between L × M, the highest value was noticed for L_1M_2 (126.67) while L_0M_0 (65.83) recorded the lowest value. Combinations, L_1M_0 (79.17), L_0M_1 (84.17) and L_1M_1 (85.00) were found to be on a par.

The effect of three factor interaction among F, L and M were found to be significant. The treatment $F_1L_1M_1$ (151.67) recorded the highest value which differed significantly from other treatments.

4.7.2.3.4. Dehydrogenase

The main effects of F, L and M were found to impose significant effect on the dehydrogenase activity. F_1 (202.70) was found to be superior than F_0 (160.77) while L_1 (210.99) was observed to be superior than L_0 (152.48). In the case of manures, the highest value was noticed for M_2 (191.35) which was

on a par with M_3 (186.15) while the lowest value was registered for M_0 (169.49) which was at par with M_1 (179.94).

The interaction effects between F × L and L × M were significant while F × M was non significant. In the case of interaction between F × L, the highest value was noticed for F_1L_1 (236.82) followed by F_1L_0 (185.15) and F_0L_1 (168.57) while the lowest value was recorded for F_0L_0 (136.39).

In the case of interaction between F × M, the highest value was noticed for F_0M_3 (215.11) which was not significantly different from other combinations, viz., F_1M_3 (206.83), F_1M_2 (198.56), F_0M_2 (190.29), F_1M_1 (165.47), F_0M_1 (167.60), F_1M_0 (161.33) and F_0M_0 (148.69).

In the case of L × M interaction, the highest value was noticed for L_0M_3 (229.65) which was significantly different from others while the lowest was noticed for L_0M_0 (142.48).

Significant difference in the dehydrogenase activity due to treatments were observed. The highest value was noticed for $F_1L_1M_2$ (264.75) which was significantly superior than other treatments while the lowest value was noticed for $F_0L_0M_0$ (115.36). Treatments $F_0L_0M_1$ (136.51) and $F_0L_0M_2$ (140.65) were found to be on a par.

4.7.2.3.5. Cellulase activity

The main effects of F, L and M were found to be significant with respect to cellulase activity. F_1 (60.42) was found to be superior than control, F_0 (47.08). Similarly liming imposed a significant effect on cellulase activity and thus L_1

Table 50 : *Effect on treatments on enzyme activities and microbial biomass at harvest Stage*

Sl. No.	Treatments	Urease (ppm of urea hydrolysed g^{-1} of soil hr^{-1})	Phosphatase (µg of p-nitro phenol released g^{-1} of soil hr^{-1})	Protease (Micromoles of amino nitrogen hydrolysed g^{-1} of soil hr^{-1})	Dehydrogenase (µg of TPF hydrolysed g^{-1} of soil $24hrs^{-1}$)	Cellulase (ppm of glucose hydrolysed g^{-1} of soil per 24 hrs^{-1})	Microbial biomass (µg of CO_2 evolved g^{-1} of soil hr^{-1})
1.	$F_0L_0M_0$	76.27	59.63	51.67	115.36	33.33	6.67
2.	$F_0L_0M_1$	100.76	80.80	66.67	136.51	35.00	6.00
3.	$F_0L_0M_2$	106.87	89.86	71.67	140.65	38.33	7.33
4.	$F_0L_0M_3$	112.97	82.70	71.67	153.06	41.67	6.67
5.	$F_0L_1M_0$	134.35	93.73	100.00	182.01	51.67	11.00
6.	$F_0L_1M_1$	137.40	96.80	101.67	186.15	53.33	10.00
7.	$F_0L_1M_2$	144.07	112.13	108.33	194.56	66.67	12.00
8.	$F_0L_1M_3$	142.35	106.13	90.00	177.88	56.67	11.67
9.	$F_1L_0M_0$	117.07	92.20	80.00	169.60	41.67	7.67
10.	$F_1L_0M_1$	125.19	90.60	91.67	161.33	46.67	8.00
11.	$F_1L_0M_2$	131.89	93.33	96.67	165.47	51.67	10.00
12.	$F_1L_0M_3$	131.29	84.27	98.33	177.88	48.33	9.33
13.	$F_1L_1M_0$	140.51	115.20	123.33	210.97	63.33	11.00
14.	$F_1L_1M_1$	162.99	114.53	151.67	235.79	68.33	10.33
15.	$F_1L_1M_2$	180.15	122.33	125.00	264.75	85.00	13.00
16.	$F_1L_1M_3$	164.88	108.33	120.00	235.79	78.33	9.57
	SE	4.90	3.2	3.9	4.8	N.S	0.62
	CD	14.15	11.5	14.04	17.27	N.S	2.23

(65.42) was superior than L_0 (42.08). The effect of manures was also significant with M_2 (60.42) and M_3 (56.25) showing significantly superior values over M_0 (47.50) and M_1 (50.83). The lowest value was recorded by the control, M_0.

The interaction effects between F × L, L × M and F × M were found to be significant. In the case of interaction between F × L, the highest value was noticed for F_1L_1 (73.75) which differed significantly from F_0L_1 (47.08), F_1L_0 (57.08) and F_0L_0 (37.08).

The effect of F × M interaction on cellulase activity was also found to be non significant with the highest value noticed for F_0M_3 (68.33) and F_1M_3 (63.33) while in the case of L × M interaction, the highest value was noticed for L_0M_3 (75.83).

The interactions among F, L and M were found to impart no significant effects with respect to cellulase activity. The highest value was recorded for $F_1L_1M_2$ (85.00) which did not differ significantly from other treatments.

4.7.2.3.6. Microbial bio-mass

The main effects of F, L and M were found to give significant difference with respect to the microbial biomass. F_1 (9.88) was found to be superior than F_0 (8.92). Similarly L_1 (11.08) was found to be superior than L_0 (7.71). In the case of manures, the highest value was noticed for M_2 (10.58) which was superior when compared to M_3 (9.33), M_1 (8.58) and M_0 (9.08).

The interaction between F × L, L × M and F × M was found to be significant. In the case of F × L interaction, the highest value was noticed for

F_1L_0 (11.17) which was on a par with F_1L_1 (11.00), while F_0L_0 (6.67) recorded the lowest value. For the interaction between F × M, the highest value was noticed for F_0M_3 (11.50) which differed significantly from other combinations.

Similarly the interaction between L × M was significant with the highest value recorded for L_0M_3 (12.50).

The interactions among F, L and M was found to impose significant difference with respect to the microbial biomass and the treatment $F_1L_1M_2$ (13.00) recorded the highest value.

4.7.2.3.7. Number of productive tillers

The main effects of F, L and M were found to impose significant difference. F_1 (11.00) was found to be superior than the F_0 (10.00) and L_1 (12.00) was found to be superior than L_0 (11.00). The effect of manures was found to impose significant difference with respect to the number of productive tillers at all levels.

The interaction between F × L, L × M and F × M were found to impose significant difference. The interaction effects of F × L was found to be significant and higher level combinations yielded superior results than the controls.

In the case of F × M interaction the highest value was noticed for F_1M_0 (12.00) and F_0M_3 (12.00) which was on a par with F_0M_2 (11.00), F_0M_1 (11.00), F_1M_2 (11.00) and F_1M_3 (11.00) while the lowest value was recorded for F_0M_0 (9.00).

Table 51 : *Means of the main effects of the treatments on the number of productive tillers, grain and straw yield*

Factor	*No. of Productive tillers*	*Grain yield (g pot^{-1})*	*Straw yield (g pot^{-1})*
a. Fertilizer			
i. F_0	10.00	24.78	33.91
ii. F_1	11.00	26.86	38.66
SE	0.38	0.9	1.02
CD	1.10	3.2	3.48
b. Lime			
i. L_0	11.00	24.71	32.86
ii. L_1	12.00	26.88	36.71
SE	0.38	0.9	1.2
CD	1.10	3.2	3.48
c. Manures			
i. M_0	10.00	24.13	37.11
ii. M_1	11.00	25.88	37.40
iii. M_2	11.00	27.36	36.77
iv. M_3	10.00	25.80	33.85
SE	0.38	0.9	N.S
CD	1.10	3.2	N.S

In the case of L × M interaction the highest value was recorded for L_0M_3 and L_1M_0 (12.00) which was on a par with L_0M_2 (11.00), L_1M_2 (11.00) and L_1M_3 (11.00) while the lowest value was recorded for L_0M_0 (9.00).

The interaction among F, L and M were found to impose no significant effect on the number of productive tillers. All the treatments imposed were found to be on a par.

4.7.2.3.8. Grain yield

The main effects of F, L and M were found to impose significant effect with respect to the grain field. F_1 (26.86) and L_1 (26.88) were found to be superior than F_0 (24.78) and L_0 (24.71) respectively. For the main effects of M, highest value was observed for M_2 (27.36) followed by M_1 (25.88) while the lowest value was registered for M_0 (24.13) which was at par with M_3 (25.80).

The interactions between F × L, L × M and F × M were found to be significant. The combination F_1L_1 (28.36), L_1M_3 (28.34) and F_1M_2 (27.97) were found to be superior than other combinations in the case of F × L, L × M and F × M interaction respectively while F_0L_0 (24.17), L_0M_0 (23.14) and F_0M_0 (23.08) recorded very low values for the grain yield.

The treatment $F_1L_1M_3$ (30.43) recorded the highest value for grain yield followed by $F_1L_1M_2$ (28.66) which were on a par . The lowest value was recorded for the control (21.30).

Table 52 : *Interaction of two factors of the treatment on number of productive tillers, grain yield and straw yield*

Factor	*No. of Productive tillers*	*Grain yield (g pot^{-1})*	*Straw yield (g pot^{-1})*
A. F x L			
F_0L_0	9.00	24.17	33.00
F_1L_0	11.00	25.55	34.81
F_0L_1	11.00	25.39	38.71
F_1L_1	11.00	28.36	38.60
SE	0.30	0.30	1.01
CD	0.87	0.87	3.06
B. L x M			
L_0M_0	9.00	23.14	38.88
L_1M_0	12.00	25.13	36.61
L_0M_1	11.00	25.15	35.72
L_1M_1	10.00	26.61	32.23
L_0M_2	11.00	27.20	35.34
L_1M_2	11.00	27.43	38.19
L_0M_3	12.00	23.26	37.84
L_1M_3	11.00	28.24	35.46
SE	0.42	0.30	1.41
CD	1.23	0.87	4.96
C. F x M			
F_0M_0	9.00	23.08	32.15
F_1M_0	12.00	25.19	35.04
F_0M_1	11.00	25.28	35.76
F_1M_1	10.00	26.49	32.68
F_0M_2	11.00	23.50	42.07
F_1M_2	11.00	27.97	39.76
F_0M_3	12.00	24.03	37.79
F_1M_3	11.00	27.58	35.01
SE	0.42	0.42	1.41
CD	1.23	1.23	4.96

4.7.2.3.9. Straw yield

The main effects of F and L were significant whereas M did not impose any significant difference with respect to straw yield. The effect of fertilizers and lime showed significant influence on straw yield with F_1 (38.66) and L_1 (36.71) recording significantly superior yield than the F_0 (33.91) and L_0 (32.86). The effect of manures were found to impart no significant difference on the straw yield.

The interactions between F × L, L × M and F × M was found to be significant with respect to the straw yield. Similarly the highest order interaction effect among F, L and M were also found to be significant with the highest value recorded for $F_1L_1M_1$ (41.55) which was on a par with all treatments involving the combination of lime and manures either with or without fertilizers.

The activity of the enzymes were observed to be maximum at the panicle initiation stage with a decline on either side for the active tillering and harvest stage.

Table 53 : *Effect of treatments on number of productive tillers, grain yield and straw yield.*

Sl. No.	*Treatments*	*Productive tillers*	*Grain yield (g pot^{-1})*	*Straw yield (g pot^{-1})*
1.	$F_0L_0M_0$	7.00	21.30	32.45
2.	$F_0L_0M_1$	11.00	24.23	35.25
3.	$F_0L_0M_2$	10.00	27.30	25.17
4.	$F_0L_0M_3$	9.00	21.80	29.13
5.	$F_0L_1M_0$	11.00	22.80	31.84
6.	$F_0L_1M_1$	11.00	26.33	34.84
7.	$F_0L_1M_2$	12.00	26.20	36.35
8.	$F_0L_1M_3$	11.00	26.26	36.23
9.	$F_1L_0M_0$	12.00	22.93	36.35
10.	$F_1L_0M_1$	12.00	26.08	36.23
11.	$F_1L_0M_2$	11.00	27.28	36.26
12.	$F_1L_0M_3$	11.00	24.73	35.32
13.	$F_1L_1M_0$	11.00	27.46	38.83
14.	$F_1L_1M_1$	12.00	26.90	41.55
15.	$F_1L_1M_2$	12.00	28.66	39.32
16.	$F_1L_1M_3$	10.00	30.43	34.70
	SE	N.S	0.91	1.41
	CD	N.S	3.27	4.96

DISCUSSION

DISCUSSION

Soils may be considered as a biological entity with complex biochemical reactions. Soil enzymes play a significant role in the microbial ecology by catalyzing innumerable reactions in soils. Enzymes are proteins that act as catalysts without undergoing permanent alteration and cause chemical reactions to proceed at faster rates (Quastel, 1946). However, soil enzymes activities' primary function may not be to measure biological activity a *per se*, but rather as an integrative indicator of changes in the biology and biochemistry of soil due to external management or environmental factors.

A major research effort is needed to develop a relative or universal index that would be interpretable without several measurements over time or comparisons among treatments. This approach makes it unlikely that a single absolute soil enzyme activity or any other biological measurement could be used to assess soil quality, because soils naturally vary widely in biological activity (Dick, 1994).

The activity measurement of selected soil enzymes hold potential as soil quality indicators because many enzyme procedures are relatively simple, rapid and could be done on routine basis; they are sensitive to temporal changes in soils due to environmental and management factors; and an abiontic component, which can provide information on long term effects of management of soils.

Systematic studies across ecosystem and long term soil management sites are needed to identify the most appropriate soil quality enzyme assays and to provide data for calibration and interpretation of these assays as independent soil quality indices or as a component of larger universal index that includes other soil properties.

Agricultural soil management techniques, such as monoculture versus crop rotation, liming and fertilization; the methodology and the type of pesticide applications; mechanical treatments such as mulching, ploughing and water logging; and climatic variations especially precipitation and temperature all influence the microbial and enzyme activities in the field. With the overriding objective to project the effect of long term fertilization and the effect of amendments (fertilizer, lime and manures) and agrochemicals (pesticides, fungicides, herbicides and antibiotics) on the enzyme activities which ultimately reflects on soil fertility, this investigation was carried out.

The soil samples collected from the permanent manurial experiments on rice (Kayamkulam and Pattambi), coconut (Balaramapuram), rubber

(Chethackal) and from the pot culture studies were subjected to the enzyme activity assay to generate data for calibration and interpretation of the results as independent soil quality indices in relation to soil properties. A brief interpretation of the results pertaining to the above study are presented in this chapter.

5.1. Studies on the permanent manurial experiments

5.1.1. Soil Physico-chemical properties

Much of the attention has been paid to the yield and yield attributes of the crops grown in permanent manurial experiments, during the past few decades. The integrated use of organic manures and chemical fertilizers in PME, provides favourable physical and ecological conditions thereby provides stability in crop production. Relatively little work has been carried out with respect to the effect of continuous application of chemical fertilizers in combination with manures on soil enzymes.

Soil biochemical and microbiological properties are dependant on the soil chemical characters, hence they act as "indices" of soil fertility. The activity of enzymes in a particular locality is dependent upon either directly or indirectly the soil properties. Thus this investigation was designed to study the effect of long term application of chemical fertilizers and manures over a long period with crops like coconut, rice and rubber on the soil chemical characters as well as their relationship with the biological characters such as enzyme activities

and respiratory activity. The salient findings emanated from this study are discussed below.

5.1.1.1. Soil reaction

The results of the pH of the samples from the PME on rice at Kayamkulam lies in an acidic range. From the data in Table 2, it was inferred that the application of chemical fertilizers alone in the treatments T_2, T_3, T_4, T_5 and T_6 or the application of manures alone as in T_1 or a combination of both as in T_7 were observed to be on a par with the control. Moreover, wide fluctuations in pH was not observed with the application of treatments due to the buffering action of manures (Raychaudhury, 1977). The acidic nature can also be attributed to the low native organic matter status in a typical sandy soil coupled with contribution from the continuous use of chemical fertilizers. Further, the dynamics of pH change was not pronounced due to the poor turn over of carbon under submerged (rice) condition.

In the case of the PME at Pattambi (Variety - Tall indica), treatments involving the combined application of chemical fertilizers like Ammonium sulphate, Single super phosphate and Muriate of potash imposed a significant difference and an increase of 33% over the control was noticed (Table 6). Application of FYM in greater proportion or in equal proportion with chemical fertilizers enhanced the soil pH which might be due to the buffering action

associated with organics by a slow and steady increase of bases as suggested by Brady (1996).

The moderating effect of FYM on soil reaction was also reported by Yifong and Chingwan (1993). Similarly in the PME II (Variety Dwarf indica), application of chemical fertilizers in combination with green leaf manure registered more or less the same effect (Table 10). The acidity developed under this treatment could be attributed to the preferential utilization of NH_4^+ ions, by the rice crop thus resulting in the disappearance of basic cations from the ionic stream (Brady, 1996).

The spacing cum intercropping experiment at the CES, Chethackal, indicated significant effect of treatments on pH (Table 14). All the samples collected from this experiment registered pH in the acidic range. Significantly lower pH values were recorded for treatments involving intercropping of rubber with teak and fodder, rubber (sole crop) and rubber + pine apple. Slow decomposition of organic matter, leading to an acid leachate coupled with cool micro climate accounts to the persistence of organic acids for a longer period. Thus the acidity development through deprotonation of the organic acids is more stronger in this case. Further, the acidic root excudates produced in the root zone by these crops might have also contributed to the development of acidity. Though the other treatments also have these effects, the values are comparatively higher than the above three experiments. In the case of treatments involving cover crop carrying legumes, the decomposition of organic matter

could be extremely faster leading to the release of appreciable quantities of bases preferentially over the protons generated from the organic acids.

In the case of the PME on coconut at CRS, Balaramapuram significant effect due to treatments was noticed (Table 20). The main effect of K was alone significant while N and P were found to be non significant. But the interaction effects of N and P were found to be significant with respect to soil pH. The soil in this locality is characterized by low activity clay (LAC) which can be attributed to the acidic pH of this location. However, application of chemical fertilizers have also influenced the soil reaction. Thus imposing significant difference among treatments.

5.1.1.2. Specific conductance

Significant difference with respect to the specific conductance within the permanent manurial experiments under different crops like rice, rubber and coconut was observed which can be generally attributed to the variations in soil type, organic matter content hydrological situations and treatment effect.

At the RRS, Kayamkulam, the highest value recorded for T_2 (150.59) as observed from Table 2 was due to the application of Ammonium sulphate followed by T_6 which involved the application of Ammonium sulphate, Single super phosphate and Muriate of potash. An increase of about 34.21% of the value over the control was observed which could be due to an increase in the ionic suite contributed from the treatment materials applied. Further the effect

of submergence also have contributed an enhancement of ions consequent to soil reduction.

In the case of PME-II at Pattambi (Variety - Dwarf indica), an increase of 18.88% over the control was noticed due to the application of ammonium sulphate (Table 10). It is noteworthy to point out that application of chemical fertilizers recorded the highest value among the treatments and the lowest value was recorded for cattle manure (92.94 μSM^{-1}). In contrast to the above findings, the effects of green manure, cattle manure chemical fertilizers or their combinations imposed similar effects on the conductance in PME-I (Variety - Tall indica).

EC values obtained in the case of the spacing cum intercropping experiment at the CES, Chethackal registered highest value for treatment involving rubber + cover crop (T_6) followed by rubber + Teak and Fodder (T_1) while other treatments were all on a par (Table 14). The enhancement of EC in T_6 could be attributed to the release of bases and organic acids consequent to a faster decomposition of the organic matter containing a legume crop. Further, legumes have got a high root CEC and a high recycling capacity with respect to bases especially, poly valent cations. Thus it is logical to believe that T_6 (rubber + cover crop) could impart a significant effect on EC.

From the table 18 to 20, it was inferred that the PME, under coconut at CRS, Balaramapuram showed highly significant results with respect to main effects and interactions. This enhancement of EC observed in treatment T_{25}

could be attributed to the release of soluble salts accumulated through chemical fertilizers added over a period of 33 years since the inception of the experiment. Thus the ionic mobility observed per unit volume of the saturation extract could be exceedingly high for soils receiving fertilizer treatments. This is in agreement with the findings of Sree Latha (1993).

5.1.1.3. Organic carbon

A close scrutiny of the data generated from RRS, Kayamkulam, revealed significant effect of treatments on organic carbon. From the Table 2, it was inferred that the highest value was recorded for T_1 (1.13) due to the application of cattle manure. The organic carbon content of the soil tend to increase with the addition of organic residues in soil. Similar results have been reported by Tisdale *et al*, 1995. Application of FYM or cattle manure in higher proportion was reported to increase the soil organic carbon content compared to the application of chemical fertilizers without organics. Such increase in organic carbon content due to manuring was also reported by Mathan *et al* (1978) and Biswas (1982). Treatments receiving the chemical fertilizers alone recorded invariably low values in all plots. The increase in organic carbon content in the manured plots might be due to the cumulative effects of continuous application of organics as treatments. The above findings goes on hand with the reports of Maurya and Ghosh (1972).

In both the PME-I (Variety - Tall indica) and PME-II (Variety - Dwarf

indica) at Pattambi, not much difference between manured and fertilized plots was observed (Table 6 & 8) with respect to organic carbon. This might be due to the higher rate of mineralization of soil organic matter observed leading to a faster disappearance of organic C (Prasad *et al*, 1971). Moreover the status of available nutrients and microbial activity in general was observed to be higher for the soils of Pattambi.

In the intercropping experiment on rubber at the CES, Chethackal, comparatively higher values for organic carbon than the permanent manurial experiment for rice and coconut was registered due to higher native organic matter status accounted through litter fall and slower decomposition. The highest value for OC was reported under rubber (sole crop) followed by rubber with cover crop while the lowest value was registered for rubber + banana intercrop (Table 14). This difference could be explained due to a high turn over and accumulation of organic matter through the litter fall in rubber as pure crop and with cover crop combinations. This observation is in conformity with the reports of Patel *et al.* (1963) and Shinde and Ghosh (1971). Introduction of banana as intercrop considerably reduced the biomass contributed through litter fall owing to an increase in spacing and concommitant decrease in canopy coverage.

In the case of PME at Balaramapuram, significant variation in the organic carbon status observed could be attributed to the differences in the rate of mineralization of organic matter. Application of 340 g N and 900 g

K_2O recorded the highest value for organic carbon (Table 20). Thus, the variations imposed through fertilizer levels and combination are believed to influence the microbial activity and C mineralization considerably causing plot wise difference in the organic carbon status. Favourable influence of higher quantity of inorganic source on organic carbon content of the soil was also reported by Yifong and Chingwan (1993).

Further treatment variations via fertilizers influences the general growth and rhizosphere activity and the nature and quantity of root exudates. This is also in conformity with the findings of Martin (1977).

5.1.1.4. Cation exchange capacity

A close scrutiny of data on CEC values of the plots in the PME at Kayamkulam, clearly indicate significant differences due to treatment effects (Table 2). The treatment T_2, receiving Ammonium sulphate registered highest value followed by T_6, with the application of Ammonium sulphate, Single super phosphate and Muriate of potash. As the charge contributing factors like OC, clay content, pH and nature and content of minerals all are unfavourable for this observation, the observed increase in CEC could not be explained at this juncture and needs further elaboration.

On the other hand, in the case of both the PME at Pattambi, treatment T_7 receiving the combined application of organic manures and chemical fertilizers have recorded the highest value (Table 6). This increase in CEC

might be attributed to an increase in the organic colloids, microbial biomass and root excretions accrued through a high biological activity. Further, an increased crop growth in these plots might have incorporated comparatively higher amounts of plant residues in to the soil over many years. In addition to this, clay organic complexes formed also might have enhanced the CEC. Similar increase in CEC consequent to the application of organic amendments and fertilizers have been reported by Tisdale (1995).

The spacing cum intercropping experiment at the CES, Chethackal revealed significant difference due to treatment effects (Table 14). The highest value was recorded for the treatment involving rubber + cover crop followed by rubber (sole crop). This might be due to the high organic colloids accumulated in the soil system by the decomposition of the leaf litter in the plantation floor. Further, the organo mineral complexes formed also must have modified the exchange behaviour of the soil to the advantage of cation retention.

The lowest value of CEC observed in the case of T_1 could be attributed to the low organic matter content contributed from the intercropping of rubber with teak and fodder. Further the decomposition of the leaf litter in this case is very slow thus, resulting in a slow process of active organic colloid formation.

The highly significant result observed in the case of the PME at Balaramapuram, consequent to treatment effect could be explained by an increase in the organic carbon content, microbial activity and an increase in the root secretions induced by mineral fertilizers. Thus, the treatment receiving

the highest doses of fertilizers T_{27} ($N_2P_2K_2$) recorded the highest value (Table 20). The individual effects of N was found to impose a significant effect on the CEC. All the interactions, at all levels were found to influence CEC positively. Thus, it is evident that the increase in CEC in a locality like Balarampauram with low activity clay soils, might be due to a general enhancement of the soil biological activity.

5.1.1.5. Anion exchange capacity

A critical review of the analytical data on AEC on the PME at the RRS, Kayamkulam revealed no significant difference due to treatment effects (Table 2). This is probably due to the comparatively low and uniform sesquioxide content of the locality coupled with low organic matter status. As the negative charge contributing sites are restricted to variable charge components of the clay and sesquioxides, the AEC observed was very low. Similar behaviour of low activity clay soils low in sesquioxides had been reported by Rajendran (1992).

The Permanent Manurial Experiment - I (Var. Tall indica) at Pattambi, showed significant effect due to treatments on AEC (Table 6). The highest and significant value obtained in the treatment, T_7 involving the combination of organics with chemical fertilizers might be due to an increase in the positive charges contributed from organic matter, sesquioxides and pH effect. The particular organo mineral complex formed with sesquioxides and organic matter

at low pH would have modified the exchange complex in a way as to derive a relatively higher percentage of positive charges. Hence, the observed increase in AEC is a combined effect of pH and sesquioxides linked through the organic colloids.

However, no significant effect could be observed in the case of PME-II (Var. Dwarf indica) though slight increase in the AEC values was seen for T_7 (Table 10). This observed non significant effect could be attributed to the uniform charge behaviour of the soil even after treatment application. The exact reason for this effect needs further elaboration.

In the case of the intercropping experiment at the CES, Chethackal, significant effect on AEC was observed consequent to treatment T_3 involving pineapple as the intercrop, which recorded the highest value as expected, since the biomass contribution towards soil organic matter was the minimum. Thus, a relative increase in the an ion sites could be expected owing to a decrease in CEC, subsequent to low organic matter addition. Hence the variable charge behaviour of the oxide rich material will be able to express its positive charge behaviour which otherwise would have subdued in an organic matter rich environment.

The PME at the CRS, Balaramapuram, indicated significant effects on AEC due to treatments and the main effects of P was found to be highly significant (Table 18 to 20). All the interaction effects of N, P and K at all combinations were found to be significant. The observed main effect of

phosphates on AEC might be due to a relative increase in the positive charge sites of the clay minerals and sesquioxide consequent to the adsorption of the phosphate ligands at low pH. Similar reactions leading to an increase in AEC has been reported by Russel (1973). This is further evident from the observation that the plots receiving the highest dose of P ($N_0P_2K_0$), recorded the highest value for AEC. The effect of organic matter and its interaction with the oxidic materials are not pronounced in this case owing to the very poor organic matter status of the soil.

5.1.1.6. Available nitrogen

The data from the PME, at the RRS, kayamkulam revealed a marked influence of treatments on available N (Table 2). From the results it is interesting to note that both the plots receiving treatments of organic manures and fertilizers showed higher nitrogen status than the control plots. An increase in the available N status was noticed in the treatment (T_4) involving the application of Ammonium sulphate + Muriate of potash (Table 2). This might be due to an increase of the microbial activity leading to an enhancement of the mineralization of native and added organic matter resulting in an increase of available nitrogen status. Further fertilizer N added also must have contributed partially to the observed increase in available N. These results were supported by the findings of Sanyasi Raju (1952), Mandal and Pani (1985).

In the PME (Variety - Tall indica) at Pattambi, the highest value was

recorded for T_7 receiving the application of cattle manure and green leaf manure in combination with N, P and K fertilizers (Table 6). In this case treatment receiving combinations of both organic and inorganic N sources maintain a higher level of available N consequent to a steady mineralization of organic manures coupled with a supplementary source from inorganic N fertilizers. Thus even after plant uptake this treatment could sustain a higher level of available N. In this context, it is believed that the narrow C : N ratio achieved in the soil system through the above combination can also trigger an increase in population of microorganisms leading to the synthesis of higher microbial biomass protein which releases N upon decomposition. This is in agreement with the findings of Bitzer and Sims, 1988.

On the other hand, the PME-II (Variety - Dwarf indica), receiving the same treatment combination did not give any significant results (Table 10). This observed non significance could be attributed to the varietal effects and the higher requirement of the dwarf indica varieties leading to a faster depletion of the mineralized and applied N. The greater biomass produced within a short span of time served as a sink for the depletion of the N pool at a faster rate (Yadav *et al.*, 1991).

The observed enhancement in available N in the experiment at Chethackal might be due to a faster decomposition and mineralization of the added litter leading to the attainment of a narrow C : N ratio, owing to a faster canopy coverage. On the other hand, the treatments with rubber as the major

crop stand having a higher canopy coverage, substantially reduced the mineralization rate though, the litter contribution was high. Thus the quantity of mineralized N was higher in the treatment involving rubber + teak and fodder, even though the total N reserve of the system was higher in other treatments.

In the case of PME at Balaramapuram, the individual effects and interactions of N, P and K were found to be highly significant (Table 18 and 19). Treatments receiving higher doses of N at N_1 and N_2 level over the uniform level of organic manures considerably increased the available N status. This might be attributed to the residual effect of manures and fertilizers added over a long period. Further, the quantity of mineralized N is likely to be higher in treatments receiving fertilizer N than the control owing to a higher microbial activity.

5.1.1.7. Available phosphorus

In the PME at the RRS Kayamkulam, the treatment T_7 involving the application of FYM along with the chemical fertilizers like Ammonium sulphate, Single super phosphate and Muriate of potash resulted in the highest value for available P (Table 2). An increase in available P was was observed consequent to the application of treatment (T_7) over the control. This enhancement of available P as expected is due to soluble P contributed from the mineral fertilizers and the mineralized P released from the organic sources.

On the other hand, treatments without mineral P fertilizers, had registered invariably low values consequent to the very poor mineralization of P from the added organic sources.

The Permanent Manurial Experiment at Pattambi (Variety - Tall indica) showed same trend as in the case of RRS, Kayamkulam for available P (Table 6). The highest value was recorded for the combination of chemical fertilizers with manures and could be explained by the direct contribution from chemical fertilizers and mineralized P from organic sources. Other treatments without chemical fertilizers were not able to maintain such a higher level of available P due to the low mineralization rate, high C : P ratio of the organic manures and plant uptake. Further, interaction of available P released from chemical fertilizers with organic anions might have reduced the loss by fixation through Fe and Al complex formation. The above results are supported by the findings of Tisdale *et al* 1995.

Organic matter increased the P availability and uptake through formation of organic PO_4 complexes that are more easily assimilated by plants, an ion replacement of $H_2PO_4^-$ on adsorption sites and reducing P fixation by providing a protective cover on Fe^{2+} and Al^{3+} oxides by the humus. Application of higher quantities of FYM promoting the available P was observed earlier by Subbiah *et al* (1983) and Bagavathi ammal and Muthiah (1995). Similarly the increase in available P could be attributed to mineralization of fixed native P to available

P. The results of the PME-II (Var. Dwarf indica), further confirmed the beneficial effects of added P availablity.

Since a part of the added P alone was absorbed by the crop and the rest converted to other forms of P, which later undergoes transformations later releases available P in soils. The above results are supported by the findings of Muthuvel *et al.* (1977). Moreover, higher levels of FYM application promoting the actual P balance of the soil was also reported by Selvi and Ramaswami (1995).

At the spacing cum intercropping experiment under rubber the available P was observed to be low compared to the PME at Pattambi, Kayamkulam and Balaramapuram. With reference to the various treatments, no significant difference was observed with respect to available P status. A significant point remains in the fact that the addition of biomass through litter fall resulted in the production of organic acids, which chelates Fe and Al, greatly reduced P fixation thus increasing the P availability to plants. High nutrient uptake and intensive cropping can be attributed to the low P status under rubber. Similar results under high density cropping was also observed by Bajwa and Paul (1978).

The humic substances produced by the root residues, the accelerated mineralization and P solublization effect of soil microflora brought about by the component crops of the system though favoured the level of soluble P in

the soil, was drastically reduced by a heavy demand for available P by the total cropping system.

In the case of coconut based cropping system, individual and interaction effects of treatments were found to be highly significant (Tables 18 to 20). This observed effect could be attributed to an enhanced microbial proliferation including P solublizing bacteria owing to treatment effects, besides the direct contribution of soluble P through fertilizers. The observed decline in the available P status with respect to a higher dose (P_2) initially might be due to microbial immobilization following a spurt in population. From the interaction effects it is seen that the associated effect of N and K in combination also have substantially contributed to the release of available P, probably by enhancing the specific microorganisms involved in P solublization.

5.1.1.8. Available potassium

The available K was markedly influenced by different treatments at the RRS, Kayamkulam. The data on the available K cited in Table 2 indicated that the application of K fertilizers profoundly increased the available K content. This might be due to the direct contribution by fertilizer K to the pool of available K as reported by Mc Lean and Doyle (1963). More over, the application of ammoniacal N to the soil in the form of $(NH_4)_2SO_4$ also replaces the K^+ ion from the exchangeable complex, there by contributing to the pool of available K.

Further, under submerged situations the anoxic conditions and soil reduction might have increased the solubility of Fe and Mn by soil reduction. This increase in concentration coupled with a high NH_4^+ build up leads to an increase in available K through cation exchange. Application of organic matter in conjunction with chemical fertilizers also have resulted in an increased fertilizer use efficiency, thus reducing the losses pertaining to K^+.

In the PME at Pattambi, the available K status was higher than the sandy Onatukkara (Table 6). The highest values recorded for T_8 with values 201.6 and 154.00 kg ha^{-1} in both the experiments were due to the application of N as Ammonium Sulphate, P_2O_5 as Single super phosphate and K_2O as Muriate of potash. The lowest values were recorded with the application of cattle manures in both the experiments. It is also noteworthy to suggest that the long continued addition of FYM might have transformed the soil in to one with a higher K availability. Similar increase in available K status with the application of FYM was reported by Muthuvel *et al.* 1977.

The intercropping experiment on rubber at Chethackal, indicated higher levels of K mainly due to the organic form of K derived from the soil organic matter. The highest value recorded for available K under rubber (sole crop) and rubber + cover crop might be due to the higher amounts of litter added (Table 14). The effect of organic matter in increasing the K availability is manifold. Similar results were reported by Sahu and Nayak (1971). As opined by Tisdale *et al* (1995) organic matter added through litter fall enhanced the

buffering capacity of soil which represents the ability of soil to supply nutrients to the soil. Under the high buffering capacity with moderately high CEC, the total K^+ ion in the soil solution and the exchange complex resulting from a high K^+ saturation showed obviously higher K^+ concentration. Thus the K flux in this ecosystem always favours a high exchangeable and soil solution K^+ resulting in higher available K^+. Similar results have been reported by Sahu and Nayak (1971).

Under the coconut based cropping system at Balaramapuram, the main effects of nutrients except N and all the interaction effects were found to be significant (Tables 18 to 20). This observed increase in available K^+ might be attributed due to a direct contribution from fertilizer sources derived from treatments. As the native K^+ of experimental site is low, higher available K^+ could be derived only through fertilizer sources. Further the 1:1 type of clay minerals dominated by Kaolinite in the clay fractions of the locality has very poor retention capacity with respect to K^+ ion. The experimental site is dominated by oxidic materials of low activity clays which contributes very low K^+ by weathering. Similar response to K^+ application in red loam soils have been reported by Sree Latha (1993).

5.1.1.9. Clay content

The clay content in all the four experimental site were found to be non significant with respect to treatment effects. It is clear from the inference that

the textural make up especially clay content of the soils are not affected by treatments and hence it is independent of treatment effects. As the clay content appears to be an intrinsic property of the soil of any locality, this observed non significance needs no elaboration.

5.1.1.10. Sesquioxides

At the RRS, Kayamkulam significant increase in sesquioxide content was observed in plots receiving chemical fertilizers (Ammonium sulphate, single super phosphate and muriate of potash) either alone or in combination could be explained by the higher solubility of Fe and Al oxides, consequent to the higher acidity created in these plots. The absence of organic manures in these treatments favours the formation of sesquioxides. In the case of treatments receiving continuous application of organic manures independently or in conjuction with chemical fertilizers resulted in the chelation and immobilization of Fe and Al which prevents their precipitation and subsequent formation of sesquioxides.

The wetting and drying process associated with continuous submergence in rice culture gradually mobilizes the inert Fe oxides present in the soil into an active form and reprecipitates in to sesquioxides. This process of cumulative enrichment of sesquioxides is substantially disrupted by the organic ligands derived from the applied manures.

Similarly in both the experiments at Pattambi, significant difference with respect to sesquioxides was observed (Tables 6 and 10). The highest value recorded for T_8 can be attributed to the acidity created by the chemical fertilizers applied in the form of Ammonium Sulphate, Single Super Phosphate and Muriate of Potash. It is also noteworthy to point out the role of green leaf manure and cattle manure either alone or in combination with chemical fertilizers resulting in lower values for sesquioxide content due to their chelating properties. Thus, the present observation reemphasises the role of organic fractions derived from the manures applied in controlling the excess Fe and Al concentration.

Under the rubber based cropping system at the CES, Chethackal, significant difference with respect to treatments was noticed (Table 14). This significance could be attributed to the difference in the quantum of biomass added to the soil through litter fall. Thus the highest value for sesquioxides registered under rubber + teak and fodder combination might have been due to the solubility of Fe and Al oxides brought about by the decomposition products and reprecipated as sesquioxides. Generally, the low values reported for rubber with legume cover crop can be attributed to the chelating properties of the organic residues formed through the decomposition of litter.

In the case of the PME on coconut at the CRS, Balaramapuram significant effects due to treatments was observed (Table 20). A close scrutiny of the data also indicates a higher value of sesquioxide compared to other

experiments carried out at Kayamkulam and Pattambi. A decrease in the sesquioxide content with the increased level of N and P application was noticed. This might be due to the higher amount of native soluble sesquioxides in a red loam soil where the effect of N and P on the solubility was noticed while in the case of K, a positive effect on solubility was noticed.

5.1.1.11. Grain Yield

The data on the grain yield revealed the superiority of manured plots over the fertilized plots. The addition of cattle manure was found to be superior than other treatments and ultimately resulted in a 3-fold increase in grain yield over the control at the RRS, Kayamkulam (Table 2). The usefulness of FYM in increasing crop yields was more pronounced when used in conjunction with chemical fertilizers. Thus the treatments T_1 and T_7 receiving cattle manure alone and cattle manure + chemical fertilizers respectively were found to be on a par.

Sustained release of nutrients and increase in the efficiency of utilization could be accounted for the superiority of manure treated plots over other. Further, the improvement of physico-chemical characteristics of soil also must have contributed to a higher utilization efficiency of plant nutrients. Use of manures in combination with chemical fertilizers could support the immediate requirement of the crop from the mineral sources and a sustained availability could be achieved through mineralization from the organics. Further, the loss

of nutrients from the soil system also is likely to be reduced considerably in plots receiving organics. Improvement in enzyme activities and enhancement of microbial load achieved through higher nutrient status and energy sources obtained from organics could account to a higher grain yield. Similar results have been reported by Albanell *et al.* 1988.

More or less, the same trend could be observed in the case of both the PME at the RARS, Pattambi, though the treatments receiving manures and fertilizers in combination (T_7) was significantly superior over other treatments (Tables 6 and 10). In this context, treatments represented by the application of ammonium sulphate alone in T_4 was found to record the lowest yield. Superiority of the combined application of chemical fertilizers and manures was evident here also. Values recorded with respect to enzyme activities, microbial biomass and plant nutrients were all found to be significantly higher for these treatments compared to T_4. Thus, it is clear from the data, that the observed increase in grain yield achieved is attributed to an increase in the nutrient use efficiency, microbial activity and improvement in the physico chemical characteristics of the soil. Further, nutrient losses from the system is also likely to be lowest from this treatment compared to others. The role of enzymes in the mineralization of N, P, K and other nutrients in different cropping systems have been reported by Rai et al (1982) and Reddy *et al.* (1987).

In the coconut based manurial experiments, at the CRS, Balaramapuram highest nut yield was recorded in treatment $N_1P_2K_2$ which registered higher values for the enzyme activity also (Table 20). This superiority could be attributed to the enhanced nutrient availability from the chemical fertilizers, improvement in the microbial load and consequent enzyme activity. By and large, it is observed that the nut yield of coconut in the PME is controlled mainly by the nutritional role of fertilizers rather than its collateral effects on the physico-chemical properties and microbiological characters. Thus it is evident from the data that the N, P and K nutrition virtually controlled the nut yield of palms.

5.1.1.12. Straw Yield

Data on straw yield on analysis shows significant effect due to treatments. Thus the treatments T_1 and T_7 receiving cattle manure alone and cattle manure + chemical fertilizers respectively recorded the highest values in the case of the RRS, Kayamkulam (Table 2). Yield of straw followed the same trend as the yield of grains with respect to treatment effects. High nutrient status obtained through chemical fertilizers and the sustained release of plant nutrients by the mineralization of the applied organics accounted for the higher straw yield registered. Further, the enhancement of microbial activity, associated enzyme activity and improvement of the soil environment might also have contributed to the increased straw yield. Similar trend in the yield of straw

recorded in the PME experiments of the RARS, Pattambi could also be explained in the same way. As these two soils are comparatively lower in organic matter, the effect of added manures and fertilizers were fully expressed in terms of yield of grain and straw.

5.1.2. Biological properties

Soil is a living system in which biological activities take place with the help of enzymatic processes. Quantitative measurements of soil enzyme activities can contribute to our understanding of these biological transformations by allowing us to evaluate the activity present in the soil. Measurement of the activity of intra cellular enzymes provides information on the biological activities of soil microorganisms. Enzyme activity is therefore a useful indicator of the microbial metabolism and hence is a parameter for the measurement of the activity of microbes in soil. The biological properties investigated in the present study are discussed here under.

5.1.2.1. Urease activity

Urease is unique among soil enzymes because it affects the fate and performance of fertilizer urea. Urea added to soil as fertilizer is rapidly hydrolysed to ammonium carbonate in most soils through the activity of soil urease; and is responsible for the rapid release of ammonia when urea is

applied. Urease is a constitutive enzyme found in a large number of microorganisms, especially in ureolytic bacteria and fungi (Bremner and Mulvaney, 1978).

The Permanent Manurial Experiment at the RRS Kayamkulam revealed that the treatment involving the combinations of inorganic fertilizers with FYM (T_7) was significantly superior over other treatments, recording the highest value for urease activity (Table 3; Fig. 1). Similar results were observed for both the Permanent Manurial Experiments at the RARS, Pattambi where the absolute values for the activity was higher than at Kayamkulam (Table 7 and 11; Fig. 2). This increase in the urease activity at Pattambi, may be attributed to the desirable soil characteristics and the associated beneficial effects.

The increased urease activity in manured plots in combination with fetilizers might be due to the high amounts of urease in the viable microbial population and to the elevated levels of accumulated urease from dead biomass. The same results were reported by Dick et al., 1988. A significant and positive correlation of urease with CEC, available N, available P and available K at Kayamkulam and Pattambi, also indicated the role of organic manures in the availability of nutrients which ultimately resulted in higher activity of urease. This was evidently due to the higher availability of substrate nitrogen (Urea) and other nutrients, which promoted the urease activity.

The regression analysis also showed the importance of these parameters in defining the urease activity [Kayamkulam (available N), Pattambi (CEC,

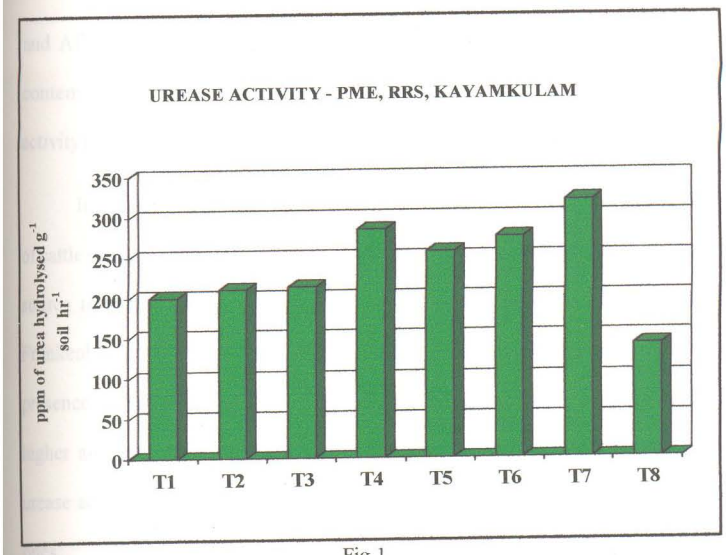
UREASE ACTIVITY - PME, RRS, KAYAMKULAM
ppm of urea hydrolysed g-1 soil hr-1
350
300
250
200
150
100
50
0
T1
T2
T3
T4
T5
T6
T7
T8

Fig 1

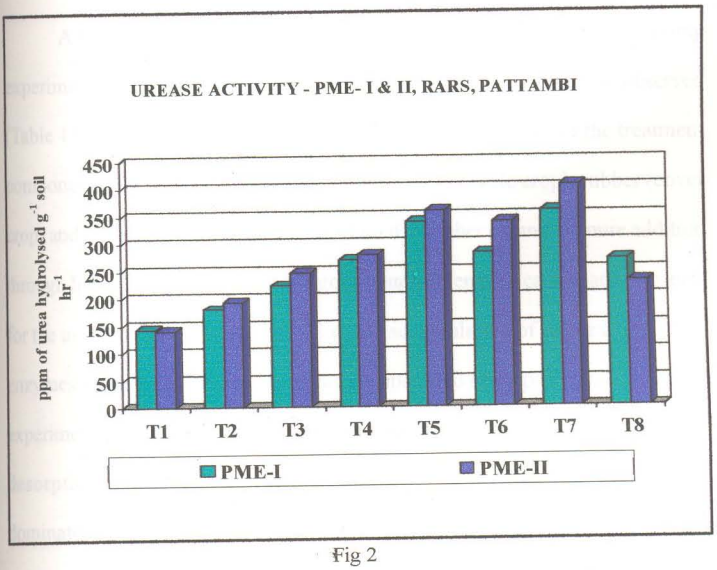
UREASE ACTIVITY - PME- I & II, RARS, PATTAMBI
ppm of urea hydrolysed g-1 soil hr-1
450
400
350
300
250
200
150
100
50
0
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 2

and AEC for PME-I and sesquioxides for PME-II), Balaramapuram (Clay content, AEC and available N) and Chethackal (pH, EC and respiratory activity)].

It is also noteworthy to point out that treatments involving application of cattle manure alone reported low values probably due to the need of inorganic source to stimulate the activity. This is in agreement with the findings of Frankenberger and Dick (1983). Therefore it is obvious that factors like the presence of extracellular urease, higher population of ureolytic bacteria and higher amounts of organic carbon might have contributed to the increased urease activity observed in the measured plots compared to the unmeasured ones.

A tremendous increase in the activity of urease under the intercropping experiment at the CES, Chethackal compared to rice and coconut was observed (Table 15; Fig. 3). The higher values registered for urease under the treatment components like rubber + pepper and cocoa, rubber (sole crop), rubber (cover crop) and rubber + banana could be attributed to higher organic manure addition through litter fall, which served as a good source of energy, carbon and nutrients for the ureolytic microorganisms. Thus the accumulation of both exo and endo enzymes in the soil system was higher compared to other permanent manurial experiments studied. Further, the loss of urease by denaturation, decomposition, desorption and leaching was also minimum in a plantation eco system dominated by rubber with higher canopy coverage. Similar studies highlighting

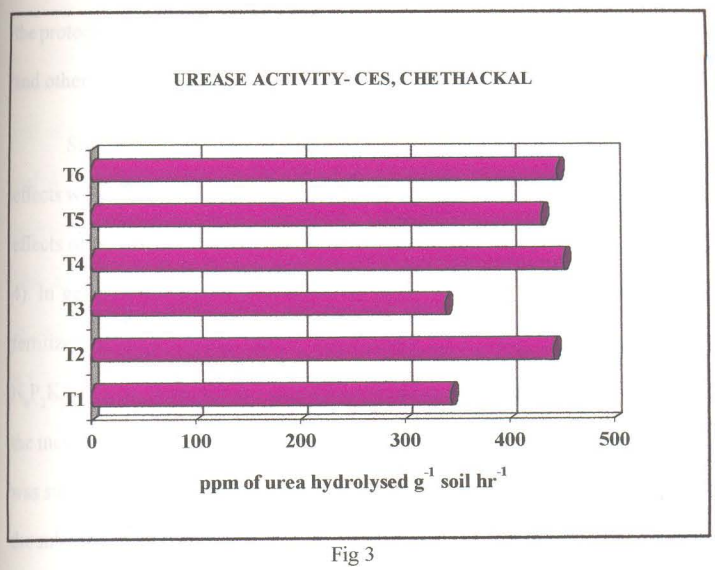
UREASE ACTIVITY- CES, CHETHACKAL
T6
T5
T4
T3
T2
T1
0
100
200
300
400
500
ppm of urea hydrolysed g-1 soil hr-1

Fig 3

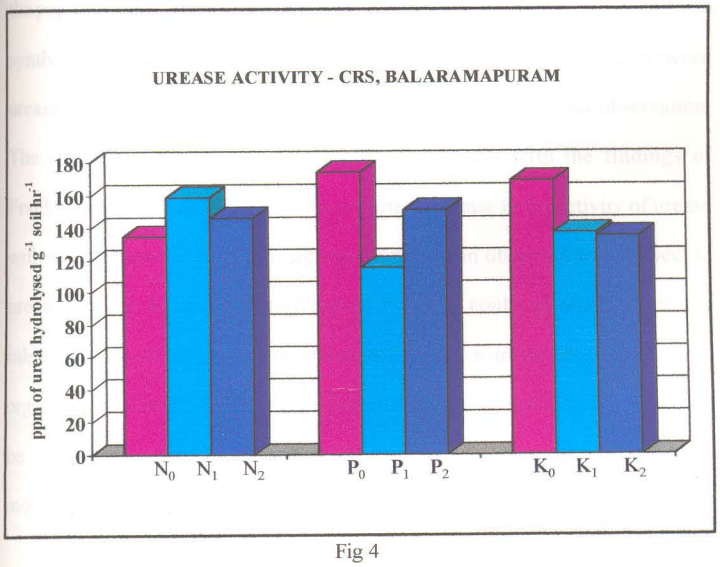
UREASE ACTIVITY - CRS, BALARAMAPURAM
ppm of urea hydrolysed g-1 soil hr-1
180
160
140
120
100
80
60
40
20
0
N0
N1
N2
P0
P1
P2
K0
K1
K2

Fig 4

the protection of enzymes by humic complexes, clay organic matter complexes and other inorganic colloids have been reported by Lann and Doran (1984).

Significant differences with respect to urease activity due to treatment effects were observed in the PME at Balaramapuram. Individual and interaction effects of N, P and K were found to be highly significant (Table 21 to 23; Fig. 4). In general, there was a decrease in urease activity with higher doses of fertilizer addition except for N. Treatments T_9, T_{10} and T_{20} represented by $N_0P_2K_2$, $N_1P_0K_0$ and $N_2P_0K_1$ respectively recorded the highest values, where the increasing doses of N (N_0, N_1 and N_2) could give a positive effect. Since N was supplied as urea, an increase in urease activity was generally expected, as the substrate concentration was high. Further addition of N might have increased the population of ureolytic organisms initially which resulted in a higher urease synthesis. The highly significant correlation coefficient obtained between urease activity and available nitrogen (+0.40) also support this observation. The results of the present study are in agreement with the findings of Frankenberger and Dick (1983) who reported increase in the activity of urease with the addition of urea. The significant reduction observed with respect to urease activity for the direct effects of P and K could be attributed to the inhibitory effects of higher concentration of P and K to the specific ureolytic organisms involved. The adverse inhibitory effects of chemical fertilizers would be further magnified in an upland situation like Balaramapuram where the moisture and organic carbon content are low.

In general the values recorded for urease activity at the CRS Balaramapuram was comparatively lower than other locations. The red loam soil of this location is highly impoverished, depleted of bases with high Fe and Al content, poor in organic matter and low in microbial activity. This infertile nature might have resulted in a general decline of enzyme activities. This is in accordance with the findings of Bruckert *et al.* (1978) who also reported low activity of urease in a soil low in base status.

5.1.2.2. Phosphatase activity

Inorganic phosphorus is nearly universally deficient in soils. A large portion of total soil phosphorus is bound in complex organic molecules. Labile organic P compounds are mineralized in soils, by enzymes, collectively called "Phosphatases" that catalyzes the hydrolysis of esters and anhydrides of phosphoric acid. The activity of phosphatase was evaluated in terms of p-nitro phenyl phosphate released g^{-1} of soil hr^{-1}.

Under rice culture, both at Kayamkulam and Pattambi (Fig 5 and 6) the treatments involving the application of FYM along with N, P, K fertilizers (T_7) continued to record the highest activity for phosphatase indicating the beneficial effects of FYM. The unmanured plots recorded very low activity of phosphatase both cases. This observed positive effects of N, P, K fertilizers and organic manures on phosphatase activity might be due to the capacity of this treatment to supply mineral nutrients, C and energy source for the fast multiplying microorganisms producing phosphatases, thus an enhancement

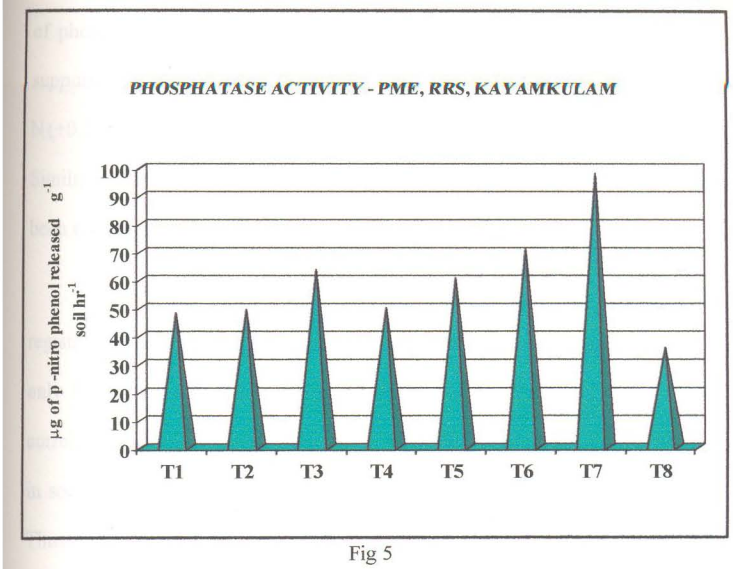
PHOSPHATASE ACTIVITY - PME, RRS, KAYAMKULAM
µg of p -nitro phenol released g-1 soil hr-1
100
90
80
70
60
50
40
30
20
10
0
T1
T2
T3
T4
T5
T6
T7
T8

Fig 5

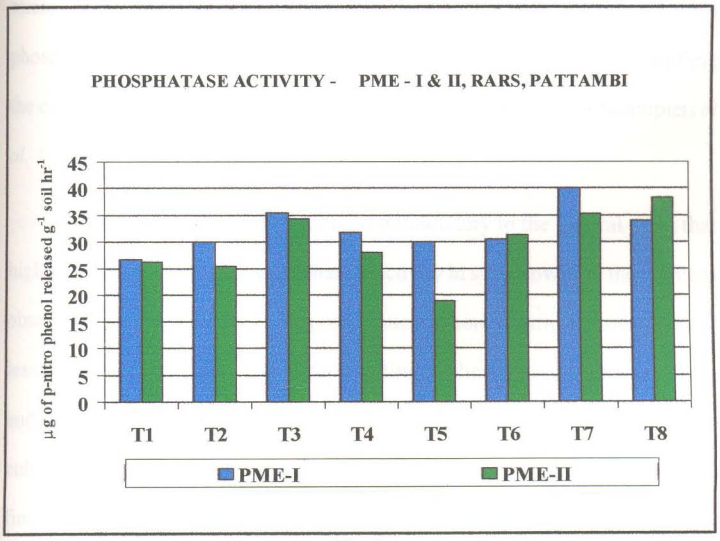
PHOSPHATASE ACTIVITY - PME - I & II, RARS, PATTAMBI
µg of p-nitro phenol released g-1 soil hr-1
45
40
35
30
25
20
15
10
5
0
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 6

of phosphatase activity could be achieved. This observation was further supported by the highly positive correlation coefficients obtained for available N (+0.28), available P (+82) and available K (+0.39) with phosphatase activity. Similar positive relations with phosphatase activity and soil parameters have been reported by Trasar Cepeda and Gil stores (1987).

However, in the PME at Pattambi, (var. Tall indica) though T_7 had registered the highest value, significant and positive correlation was observed only for the CEC (+0.35) and available N (+0.46) (Table 8). This shift in correlation with soil parameters might be attributed to the inherent variation in soil fertility, textural make up and exchange behaviour of the two soils. Thus it is evident that in the soils of Pattambi, role of N in the build up of microbial biomass protein and the exchange behaviour of the soil to retain phosphtase from losses and denaturation are the dominant aspects controlling the enzyme dynamics. This is in accordance with the findings of Nannipieri *et al.* 1990.

Thus, the present observation is contradictory to the general trend that high available P suppresses phosphatase activity in soil. However, the increase observed here might be due to the proliferation of non specific microorganisms, less sensitive to high available P contributing to phosphatase both from living and dead cells. Hence, phosphatase derived through these sources might have enhanced the total phosphatase activity. The results are in agreement with the findings of Haynes and Swift (1988). Further, the combined use of organics

with mineral fertilizers might have annulled the suppressive effect of the inorganic phosphates by the associated organic phase, stimulating the microflora involved in the mineralization of organic phosphates. This is in agreement with the findings of many workers [Dalal (1975), Halstead and Sowden (1968) , Chhonkar and Tarafdar (1994)].

Under the rubber ecosystem, the activity was reported to be higher than under rice and coconut (Table 15; Fig. 7). Not much difference between various crop components like rubber + teak and fodder, rubber (sole crop), rubber + pine apple, rubber + pepper and cocoa, rubber + cover crop and rubber + banana was observed with respect to their effect on phosphatase activity. In this case the variation in chemical composition of the litter accumulated did not differ significantly in relation to the specificity of the organisms involved in phosphatase synthesis. Hence the effect of treatments were more or less the same.

The higher activity observed in this locality representative of a hilly plantation ecosystem might be due to the protection of the phosphatase produced, by the adsorption and stabilization mechanism brought about by a higher level of organic colloids than the other ecosystems studied. Similar mechanisms of enzyme protection in soil systems by organic fractions have been reported by Hayano (1977).

Thus it is noted that under a forest ecosystem, where there is lack of a fertilizer programme, a higher activity of phosphatase was noticed. This could

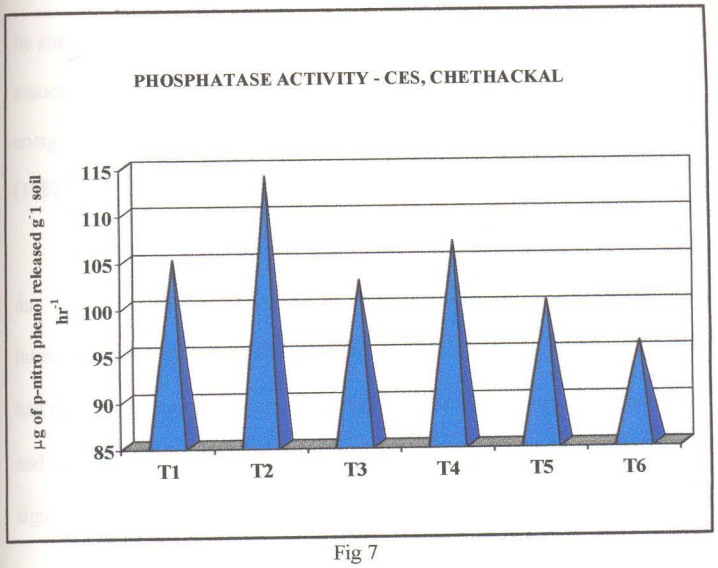
PHOSPHATASE ACTIVITY - CES, CHETHACKAL
µg of p-nitro phenol released g-1 soil hr-1
115
110
105
100
95
90
85
T1
T2
T3
T4
T5
T6

Fig 7

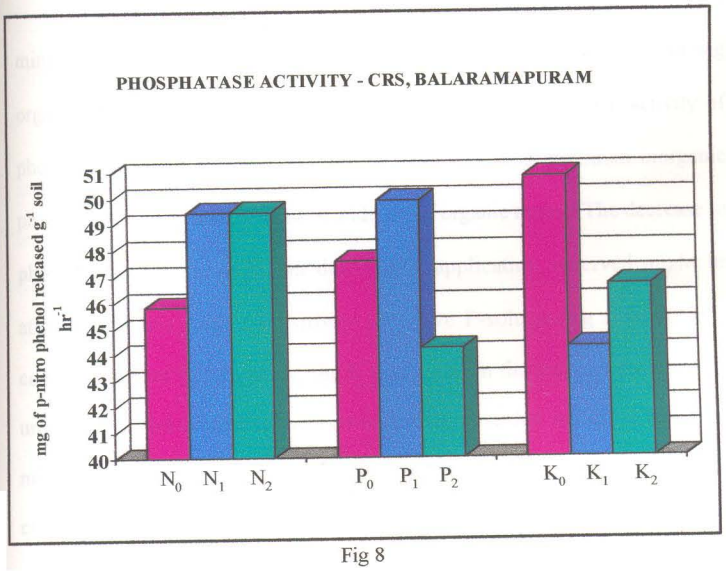
PHOSPHATASE ACTIVITY - CRS, BALARAMAPURAM
mg of p-nitro phenol released g-1 soil hr-1
51
50
49
48
47
46
45
44
43
42
41
40
N0 N1 N2
P0 P1 P2
K0 K1 K2

Fig 8

be attributed to the release of P from organic matter by the acid phosphatase associated with dead and living cells through internal cell metabolism and energy transformation reactions. This in accordance with the findings of Burns (1982).

In the PME, at Balaramapuram, significant effect on phosphatase activity due to treatments was observed in the case of K alone (Fig. 8). However interaction effects were all significant at all levels. Application of nitrogen had shown beneficial effects on phosphatase activity though not significant. P and K showed a negative effect on phosphatase activity, with the latter giving significant reduction.

The decrease in phosphatase activity consequent to higher doses of mineral P could be attributed to the reduced activity of phosphorus solubilizing organisms in response to a high available P. Thus, the maximal activity of phosphatase could be observed only in plots receiving minimum inorganic phosphates unless there is a heavy addition of organic matter. The decrease in phosphatase activity at higher doses of K application observed might be attributed to the higher sensitivity of active P-solublizing flora to high concentration of K. Moreover, the K derived from the soil and the organics uniformly added to all treatments would have provided sufficient K for the microbial multiplication. Thus, higher levels of K, though beneficial to the crop, seems to be toxic to the P-solublizing bacteria. Similar suppressive effect

on phosphatase activity due to higher levels of mineral nutrients have been reported by Feder, 1973.

5.1.2.3. Protease activity

Protease include a group of hydrolytic enzymes that hydrolyze the peptide bonds of the proteins of the organic substance followed by mineralization to amino-nitrogen. The results of the protease activity assay pertaining to the PME on rice at Kayamkulam and Pattambi (Fig. 9 and 10), revealed the superiority of the treatment T_7, representing the combined application of chemical fertilizers and organic manures over other treatments. This observed increase might be due to the availability of mineral nutrients, C and energy sources in larger quantities for the faster multiplication of proteolytic organisms, thus releasing both exo and endo enzymes.

Haynes (1986) also reported the positive effect of accumulation of mineral N as a result of operation of mineralization and immobilization cycle, leading to a higher level of substrate N for the proteolytic bacteria derived from organic sources. The beneficial effects accrued from organic sources in this context was further supplemented through the nutritional effects of mineral fertilizers leading to a fabulous increase in the proteolytic organisms.

In addition to the release of nutrients, and the role as energy and carbon sources, organics colloids derived from the added manures might have involved in the stabilization and protection of enzymes thus contributing to a higher

activity of protease. Similar effects of organic manures on soil enzyme activity have been reported by Miller and Dick (1995). Thus it is evident from the data that the integrated application of fertilizers and manures increased the microbial load there by contributing to an increased synthesis of protease and its activity in the soil. It is relevant in this context to believe that the combined application of manures and fertilizers will have a profound influence in the multiplication of many microorganisms, other than the specific proteolytic organisms. This amounting biomass protein derived from the lysis of the active cells contributes a reserve of substrate protein in the soil for the proteolytic microflora to proliferate thus expressing a higher protease activity. Hence the effect of organics with respect to protease activity is complimentary through the activity of other non specific and non proteolytic organisms. This is in accordance with the findings of Hayano (1993).

The low values for activity reported in control plots of both the locations could be attributed to the poor microbial activity and nutrient status. Control plots of the PME at Kayamkulam, without chemical fertilizers and organic manures registered the lowest value which is obviously due to the low organic matter status, nutrient content and low cation exchange capacity while in the case of control plots at Pattambi, receiving cattle manure alone had registered values higher than the control plots of Kayamkulam but the lowest compared to other treatments involved. Thus consequent to the application of organics, microbial activity was considerable even in the control plot. However,

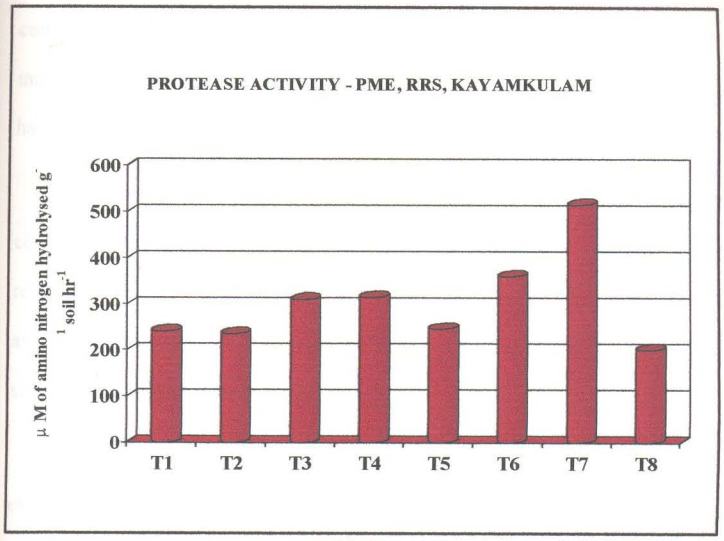
PROTEASE ACTIVITY - PME, RRS, KAYAMKULAM
μ M of amino nitrogen hydrolysed g soil hr-1
600
500
400
300
200
100
0
T1
T2
T3
T4
T5
T6
T7
T8

Fig 9

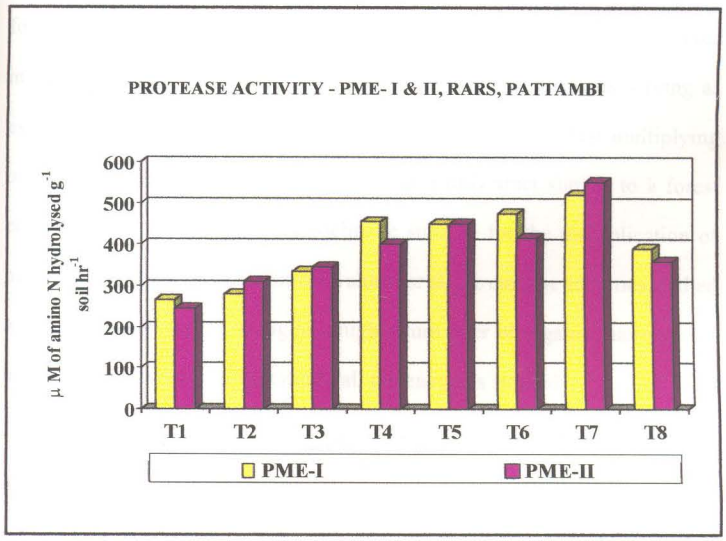
PROTEASE ACTIVITY - PME- I & II, RARS, PATTAMBI
μ M of amino N hydrolysed g-1 soil hr-1
600
500
400
300
200
100
0
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 10

combined application with chemical fertilizers had further increased the multiplication of micro flora involved in protease synthesis. Similar findings have been reported by Lebedeva and Gommonova (1972).

The above observation is indirectly supported by the strong positive correlations obtained for protease activity with available P, available K and respiratory activity in all the three experiments. Correlation obtained for available nitrogen was also strongly positive in the above cases but non significant except the PME-I at Pattambi.

A close scrutiny of the data on protease activity, of the intercropping experiment at the CES, Chethackal revealed non significant results due to treatment effects (Fig. 11). However, the values were comparatively higher for this location than the other experiments studied. The higher values observed in this case could be attributed to a high organic matter status serving as excellent source for carbon, energy and nutrients for the fast multiplying microorganisms. This location representing a hilly tract similar to a forest ecosystem has a favourable microclimate suitable for the multiplication of microorganisms. In this experiment, values for all the enzyme activities studied were reasonably higher owing to a higher turn over of organic matter and a higher microbial activity specific to all the enzymes studied.

The non significant results obtained from the various components of this cropping system could be attributed to the uniformity in soil conditions observed in this area with respect to the physico-chemical characteristics,

organic matter status and exchange behaviour. Though, minor variations could be observed in the chemical composition of the organic matter formed from the litter accumulation in different treatments, it has not affected the total microbial biomass and the enzyme activity considerably. This might be due to the compensating effect on the total protease pool by the multiplication of a group of non specific proteolytic organisms, when specific proteolytic species were suppressed by variations in litter composition. Thus the contribution of the protease activity in toto from various groups of microorganisms involved will be more or less the same, irrespective of the minor variations in the individual contribution from each species. Similar results were observed by Sato and Omura (1989) in a study on microbial count of paddy soils in relation to enzyme activity.

The protease activity of the PME at Balaramapuram, registered significant variations due to treatment effects. The individual and interaction effects of P and K were found to be significant while the effect of N was significant only for interaction effects at all levels (Fig. 12). The treatments $N_1P_0K_0$ and $N_0P_1K_1$ recorded the highest and lowest values respectively.

As the dose of organic manures applied was uniform for all the treatments, the beneficial effects accrued could be attributed to the contributions from the chemical fertilizers added. It is seen from the data, that application of nitrogenous fertilizers is beneficial though not significantly superior. The N-mineralized from the native and added organic matter thus is sufficient to

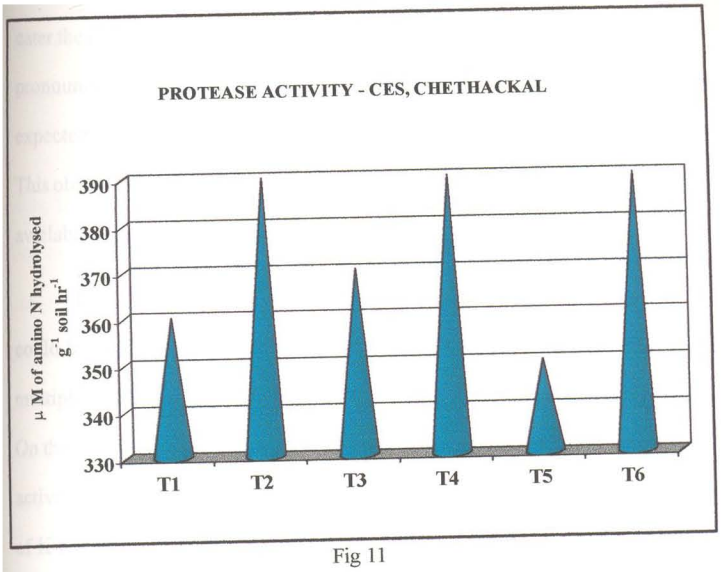
PROTEASE ACTIVITY - CES, CHETHACKAL
μ M of amino N hydrolysed g⁻¹ soil hr⁻¹
390
380
370
360
350
340
330
T1
T2
T3
T4
T5
T6

Fig 11

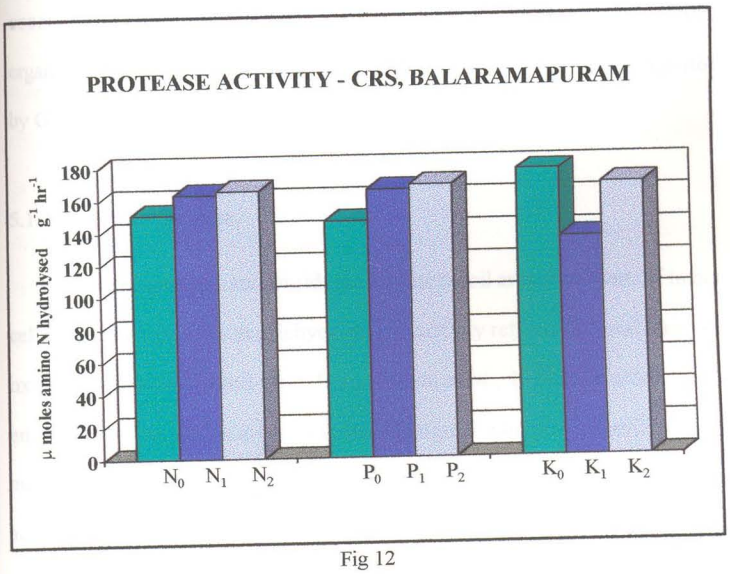
PROTEASE ACTIVITY - CRS, BALARAMAPURAM
μ moles amino N hydrolysed g⁻¹ hr⁻¹
180
160
140
120
100
80
60
40
20
0
N0 N1 N2
P0 P1 P2
K0 K1 K2

Fig 12

cater the requirement of the proteolytic organisms. However, the effect is more pronounced and highly significant for higher addition rates of phosphates as expected, since the soil is inherently poor in P with high P fixation capacity. This observation is also supported by the positive correlation observed between available P and protease activity.

Thus the increase in protease activity observed with higher levels of P could be explained by the nutritional role of phosphorus with respect to the multiplication of the specific microorganisms involved in proteolytic activity. On the other hand, the effect of K is found to be negative in relation to protease activity, probably due to the inhibitory effects imparted by the external addition of K fertilizer. Though, the soil is found to be deficient for K with respect to coconut, the K status is assumed to be sufficient for the specific proteolytic organisms. Similar effects of mineral nutrients on protease activity was reported by Gonzales Cercedo (1978).

5.1.2.4. Dehydrogenase

Dehydrogenases are considered to exist in soil as integral parts of intact cells of microorganisms and dehydrogenase activity reflects the total range of oxidative activities of soil microflora (Casida *et al.*, 1964). Dehydrogenase enzymes play a significant role in the biological oxidation of soil organic matter, by transferring H^+ from substrates to acceptors. Dehydrogenase activities have been commonly used as indicators of biological activity in

soils because of its occurrence only within living cells, unlike other enzymes which can occur in an extracellular state also.

Studies from the long term experimental sites have shown that the soil dehydrogenase activity are sensitive in discriminating the treatment effects due to various soil management practices. It was observed that the effect of integrated use of chemical fertilizers with organic manures was to accelerate the microbial multiplication, ultimately resulting in enhanced dehydrogenase activity under wet land rice culture irrespective of the soil type. Hence, higher values were reported for treatment T_7, involving cattle manure and green manure in combination with inorganic fertilizers irrespective of soil type at the PME I and II at Pattambi and Kayamkulam.

In the case of PME at the RRS Kayamkulam, the treatments T_6 or T_7 was found to register higher values, both receiving organic manures in combination with fertilizers (Table 3; fig. 13). It is also worthwhile to mention in this context that the addition of cattle manure and green leaf manure manifested maximum dehydrogenase activity. Similar observations have been reported by many workers (Chendrayan *et al.*, 1980; Pedrazzeni and Mc Kee, 1984 and Kalidurai, 1988). Organic systems, which used crop residues and green manures in combination with chemical fertilizers was found to have higher dehydrogenase activity than a conventional system according to the reports of Bolton *et al.* (1985).

The dehydrogenase activity has been often linked with the levels of

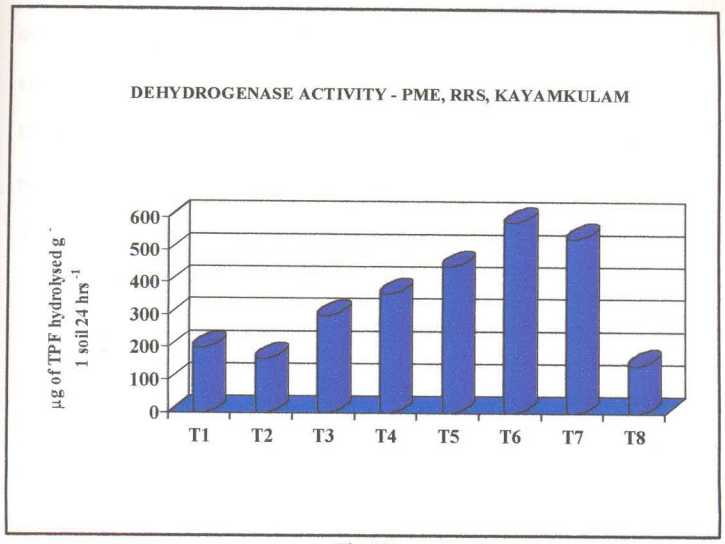
DEHYDROGENASE ACTIVITY - PME, RRS, KAYAMKULAM
µg of TPF hydrolysed g -1 soil 24 hrs -1
600
500
400
300
200
100
0
T1
T2
T3
T4
T5
T6
T7
T8

Fig 13

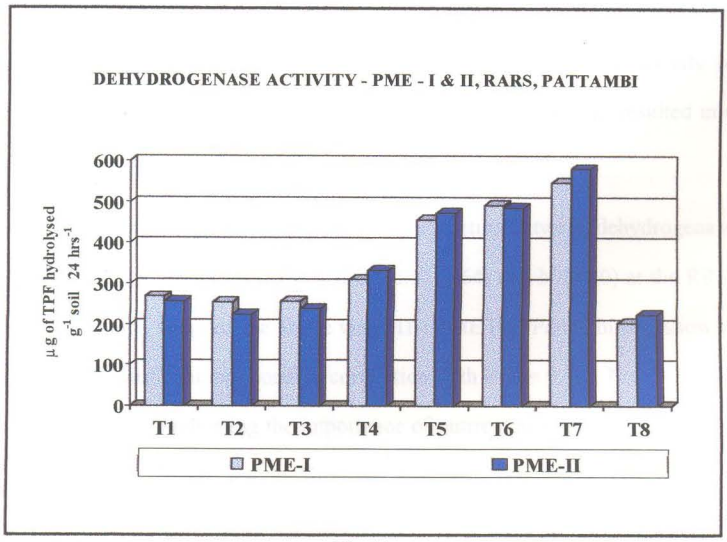
DEHYDROGENASE ACTIVITY - PME - I & II, RARS, PATTAMBI
µ g of TPF hydrolysed g -1 soil 24 hrs -1
600
500
400
300
200
100
0
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 14

available organic C substrates in the soil, as they serves as the source of electrons and H^+ for accomplishing reduction reactions. The higher levels of DHA observed under rice at the RRS, Kayamkulam in the manured plots, might be due to the higher organic C and nutrient status. It was also inferred that the dehydrogenase activity increased with increasing microbial population following the application of organic amendments with mineral nutrients as observed from the results of the PME at Kayamkulam (Table), which goes on hand with the findings of Ladd and Paul (1973).

This observation was further substantiated by Eiland (1980) who obtained significantly higher activity of dehydrogenase in fertilized plots compared to unfertilized plots in a Danish long term experiment. From this investigation, it could be seen that a nutrient rich environment coupled with high C and energy source was mandatory for the enhanced activity of dehydrogenase and the elimination of any one of the nutrients resulted in a lower activity.

A highly significant and positive correlation between dehydrogenase activity and available P (0.53), available K (0.64) and N (0.30) at the RRS, Kayamkulam, supports the above view. The PME II at Pattambi also showed a highly significant and positive correlation with P ($r = 0.67$), N ($r = +0.22$) and K (+0.33), indicating the importance of nutrient amendments in relation to the dehydrogenase activity. Similarly a significant and positive correlation for the PME I, at Pattambi between dehydrogenase activity and available N

(+0.42) and available P (0.54) again confirms the above findings. Thus the change in dehydrogenase activity corresponds more closely to microbial biomass generated through an enhanced microbial activity rather than the direct nutritional or amendment effect. Hence, dehydrogenase activity will be a more representative index of the recent soil management practices especially manuring. This view is in accordance with the findings of Burns (1982).

The higher values reported for the PME at Chethackal, under the different treatment components of the rubber eco-system (Table 15; Fig. 15) than the rice and coconut eco system would be attributed to the higher litter accumulation and the biomass added. Thus, these soils could invariably supply higher amounts of C, energy sources and mineral nutrients for the faster multiplication of the microbial population contributing to an enhanced activity of dehydrogenases.

As the soils of Chethackal represents a forest hilly tract with an abundant supply of biodegradable organic substrate and an inherently higher dehydrogenase activity, the treatment effects were not much pronounced except for T_2 and T_6 with respect to the activity of dehydrogenase. This might be due to the free availability of organic residues in all the treatments uniformly to support the microbial biomass. Thus dehydrogenase activity in any soil could be used as a useful parameter to evaluate the overall microbial activity. This observation is in agreement with the findings of Nannipieri *et al.*, 1990.

Dehydrogenase activity was also seen influenced by the mineral nutrient

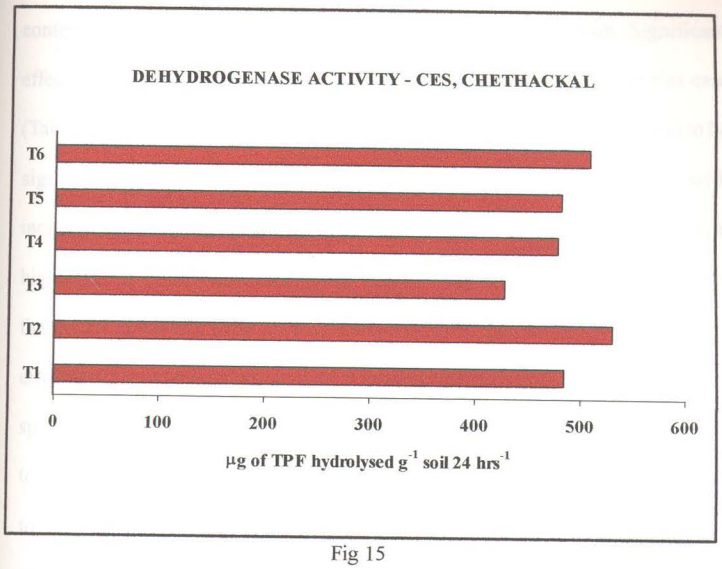
DEHYDROGENASE ACTIVITY - CES, CHETHACKAL
T6
T5
T4
T3
T2
T1
0
100
200
300
400
500
600
µg of TPF hydrolysed g-1 soil 24 hrs-1

Fig 15

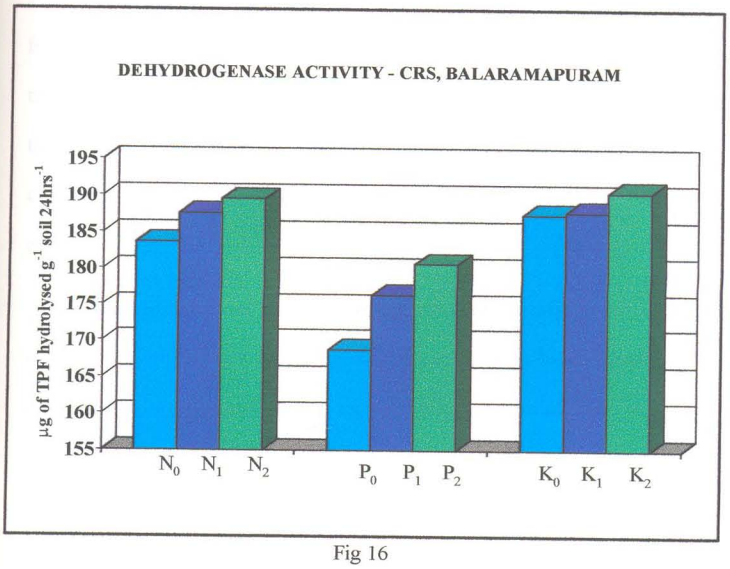
DEHYDROGENASE ACTIVITY - CRS, BALARAMAPURAM
µg of TPF hydrolysed g-1 soil 24hrs-1
195
190
185
180
175
170
165
160
155
N0
N1
N2
P0
P1
P2
K0
K1
K2

Fig 16

content of the soils in the case of the PME at Balaramapuram. Significant effects of the treatments on dehydrogenase activity was observed in this case (Table 21 to 23; Fig. 16). The individual effects of N, P and K was found to be significant and enhanced activity of dehydrogenase was noticed with incremental additions. This observed increase in dehydrogenase activity at higher levels of nutrients could be attributed to the increase in population of the microorganisms in general consequent to a favourable mineral nutrient environment created since the dehydrogenase enzymes are produced by a broad spectrum of microorganisms. The interaction effects of N, P and K were found to be significant showing the role of the association of nutrients in combination to decide the enzyme dynamics. Thus combined application expressed a situation entirely different from individual effects and is always advantageous for enzyme activity in this case. The highest activity was recorded for the treatment $N_0P_2K_2$ which indicated the role of P and K nutrition in influencing the dehydrogenase activity. The lowest value recorded for T_1, the absolute control ($N_0P_0K_0$) clearly established the effect of mineral nutrients on enzyme activity.

Multiplication of microbes irrespective of specificity could enhance the total activity. Hence suppression of any species by higher doses could be compensated immediately by an excess multiplication of other non specific groups. To support this view, positive correlations were obtained with available N, available P and available K though not significant except for N. Though a

general increase in activity was noticed at higher doses, some of the treatments had shown lower values which might be due to the direct inhibitory effects of mineral nutrients brought about through the raised osmotic potential of the soil solution by salt effect. This effect, though temporary has resulted in a partial sterilization effect leading to a reduced enzyme activity. This observation is in agreement with the findings of Cooper and Warman (1996).

5.1.2.5. Cellulase activity

Cellulase catalyses the conversion of insoluble cellulose into simple, water soluble mono or di saccharides, a reaction characteristic of the entire cellulolytic flora and consist of three distinct classes of hydrolytic enzymes including B-glucosidase, which at the end of cellulose degradation hydrolyses cellulose to glucose (Alexander, 1977).

The mechanism where by microorganisms and enzymes decompose the complex polymers into simple sugars and subsequent mineralization products have a special significance in nature because of their inevitable role in carbon cycle and soil aggregation, mediated mainly through cellulases.

The treatments imposed no significant effect with respect to cellulase activity in the case of the permanent manurial experiment at the RRS, Kayamkulam (Table 3; Fig. 17). In this case, the treatment represented by the application cattle manure alone has given comparatively higher values owing to a higher reserve of organic carbon, energy and nutrient sources than other

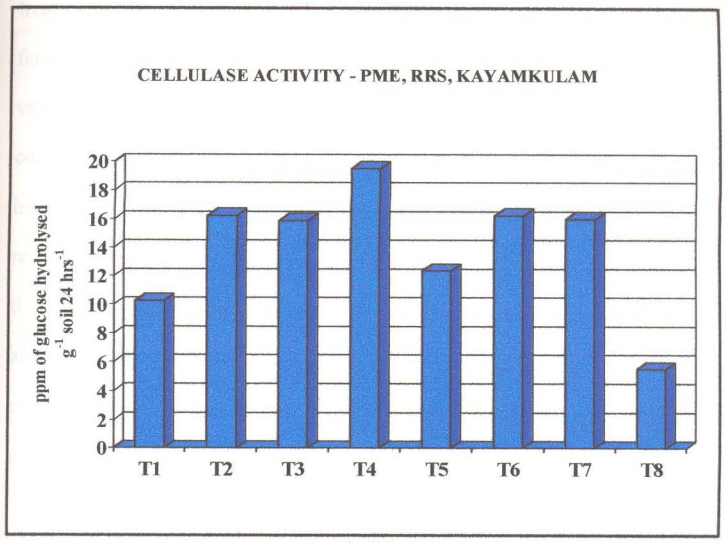
CELLULASE ACTIVITY - PME, RRS, KAYAMKULAM
ppm of glucose hydrolysed g-1 soil 24 hrs-1
0
2
4
6
8
10
12
14
16
18
20
T1
T2
T3
T4
T5
T6
T7
T8

Fig 17

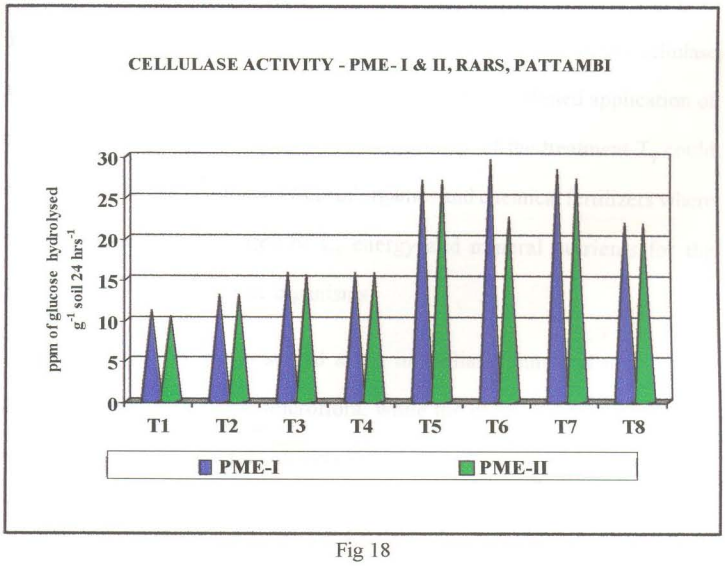
CELLULASE ACTIVITY - PME-I & II, RARS, PATTAMBI
ppm of glucose hydrolysed g-1 soil 24 hrs-1
0
5
10
15
20
25
30
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 18

treatments. The treatments represented by various combinations of chemical fertilizers and combinations of chemical fertilizers with organics had given values on a par with the control. Thus, though chemical fertilizers and organic combinations were able to enhance the cellulolytic activity, a substantially higher activity expressed by a high reserve of cellulase obtained from the plot receiving the application of cattle manure alone, had smothered the effect of those treatments. Further, the soil is of sandy nature, with low organic matter and clay content with very poor cellulase activity and the addition of organics expressed an increased cellulase activity. This is in agreement with the findings of Frankenberger and Dick (1983).

In the case of the PME (Var. Tall indica) and PME II (Var. Dwarf indica) at RARS Pattambi, highly significant results were obtained with respect to treatment applications (Tables 7 and 11; Fig. 18). The highest value for cellulase activity was noticed for the treatment (T_7) receiving combined application of chemical fertilizers with organics. The superiority of the treatment T_7 could be explained by the combined effect of organics and chemical fertilizers where it served as a steady source of C, energy and mineral nutrients for the multiplication of cellulolytic organisms.

Chemical fertilizers served as an immediate source of nutrients to stimulate the growth of the microflora, while the organic sources assured a sustained release of mineral nutrients and supplied energy and carbon for new cell formations. Though, there were three treatments receiving cattle manures

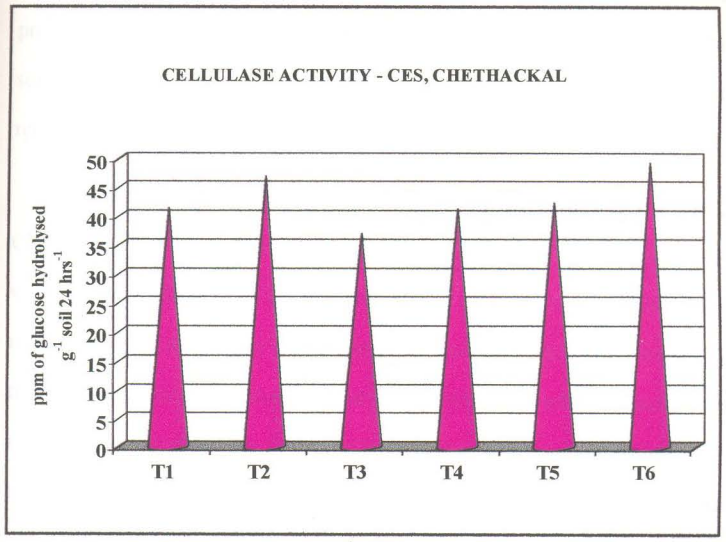
CELLULASE ACTIVITY - CES, CHETHACKAL
ppm of glucose hydrolysed g -1 soil 24 hrs-1
50
45
40
35
30
25
20
15
10
5
0
T1
T2
T3
T4
T5
T6

Fig 19

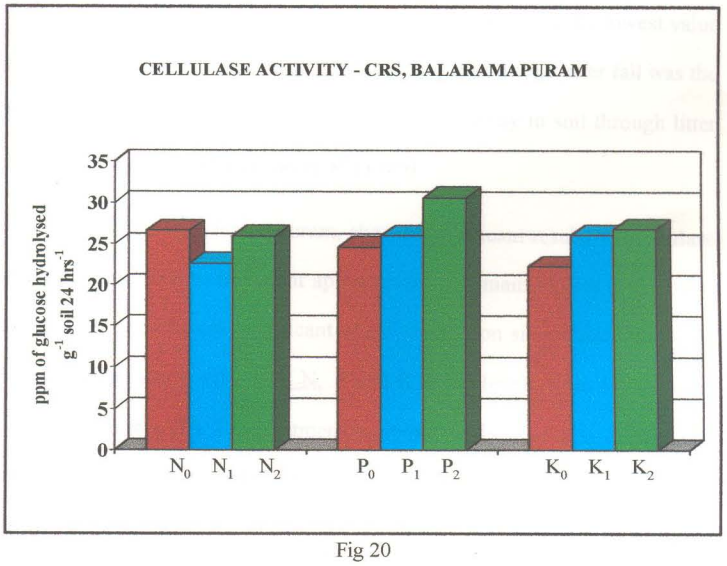
CELLULASE ACTIVITY - CRS, BALARAMAPURAM
ppm of glucose hydrolysed g -1 soil 24 hrs-1
35
30
25
20
15
10
5
0
N0 N1 N2
P0 P1 P2
K0 K1 K2

Fig 20

and green manures alone, T_7 was significantly superior over these treatments probably due to the nature of the soil, which is medium in fertility. The superiority of chemical fertilizers in combination with manures have been reported by Vilasyuk and Lisoval (1964) and Speir and Ross (1978).

Under the spacing cum intercropping experiment at the CES, Chethackal, in rubber plantations, significant differences due to treatments were observed (Table 15; Fig. 19). The highest values were registered under rubber sole crop and rubber + cover crop. This could be attributed directly to the large quantities of biomass added through litter fall which served as C, energy and nutrient sources for the cellulolytic microorganisms and thus their multiplication had resulted in high cellulolytic activity. The role of biomass accumulation in the soil on cellulase activity is indirectly evident from the treatment involving pine apple as intercrop which recorded the lowest value for cellulase as the total quantity of biomass added through litter fall was the minimum. Similar enhancement of cellulolytic activity in soil through litter fall has been reported by Wood *et al.* (1980).

The PME at Balaramapuram, showed significant results for cellulase activity consequent to treatment applications. The main effects of fertilizer nutrients, P and K were significant while N was non significant (Table 21; Fig. 20). Interaction effects of N, P and K at all levels were found to be significant (Table 22). The treatment receiving the highest dose of fertilizer levels (T_{27}), $N_2P_2K_2$ recorded the maximum values. As all the plots received

organic manures uniformly with other treatments the observed significant result could be attributed to the effect of chemical fertilizers. Thus, in a low activity clay soil of this locality, stimulatory effect on the microbial multiplication could be observed due to the nutritional effects of N, P and K resulting in higher cellulase activity.

5.1.2.6. Respiratory activity

The size of the microbial biomass and its activity are the two important factors in the recycling of plant nutrients and serve two functions for the microflora; providing energy for growth and supplying cell carbon for new cell formation. Apart from this, the mineralization and release of CO_2 in to the atmosphere accomplishes the task of bridging the carbon cycle. A variety of micro-organisms are involved in the process and their proliferation and sensitivity is governed by a pleothora of soil and other environmental factors.

A critical examination of the data obtained from the PME of the RRS, Kayamkulam revealed significant effect due to treatments on the respiratory activity (Table 3; Fig. 21). The highest value was recorded for the treatment T_7 receiving combined application of chemical fertilizers with organics.

A faster multiplication of the microflora in response to an abundant source of energy rich compounds, carbon and nutrients derived from the added organics and chemical fertilizers could be attributed to this observation. This substrate induces respiratory effect was significantly higher in T_7

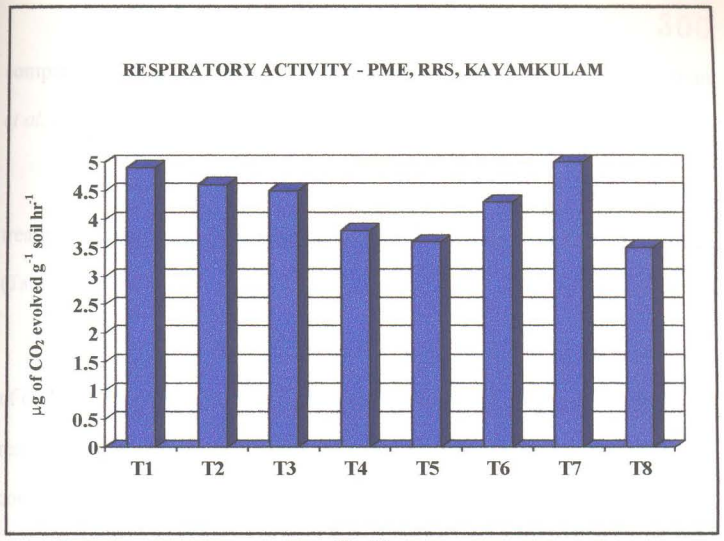
RESPIRATORY ACTIVITY - PME, RRS, KAYAMKULAM
µg of CO2 evolved g-1 soil hr-1
5
4.5
4
3.5
3
2.5
2
1.5
1
0.5
0
T1
T2
T3
T4
T5
T6
T7
T8

Fig 21

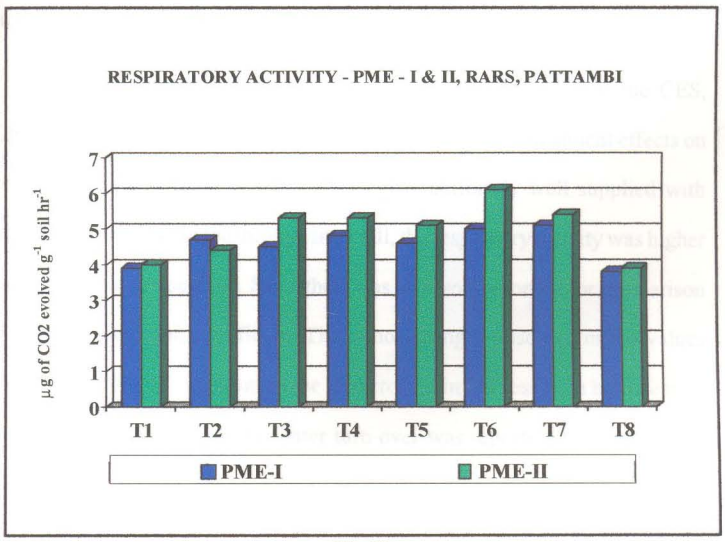
RESPIRATORY ACTIVITY - PME - I & II, RARS, PATTAMBI
µg of CO2 evolved g-1 soil hr-1
7
6
5
4
3
2
1
0
T1
T2
T3
T4
T5
T6
T7
T8
PME-I
PME-II

Fig 22

compared to other treatments. Similar results have been reported by Cochran *et al.* (1988).

A similar result had been observed in the PME-I at Pattambi, for the treatment T_7 receiving the same combination of fertilizers and organic manures (Table 7; Fig. 22).

In the PME II at Pattambi, treatments T_3 and T_6 involving the application of cattle manure and green manure with combinations of inorganic fertilizers respectively registered higher values (Table 11). In all the above cases, the combined application of manures and fertilizers provided a balanced supply of carbon, energy source and mineral nutrients which increased the microbial activity and substrate induced respiratory activity.

The intercropping cum spacing experiment in rubber at the CES, Chethackal registered no significant result with respect to treatment effects on respiratory activity (Fig. 23). As the field is uniformly well supplied with organic matter contributed through litter fall, the respiratory activity was higher compared to other locations. Since there was no absolute control for comparison the results were non significant. The minor changes observed in the values were merely due to variations in the intercrops. Similar results in high density cropping systems with greater litter turn over was reported by Asmer *et al.* 1992.

Data obtained for the PME at Balaramapuram, showed significant effect

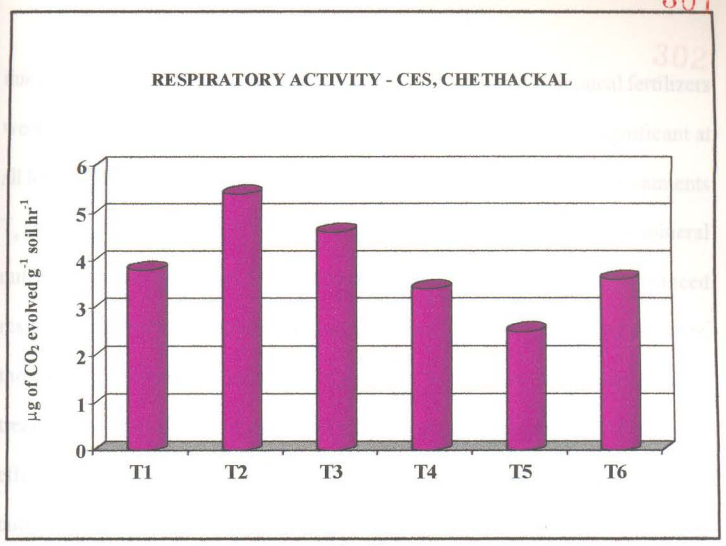

Fig 23

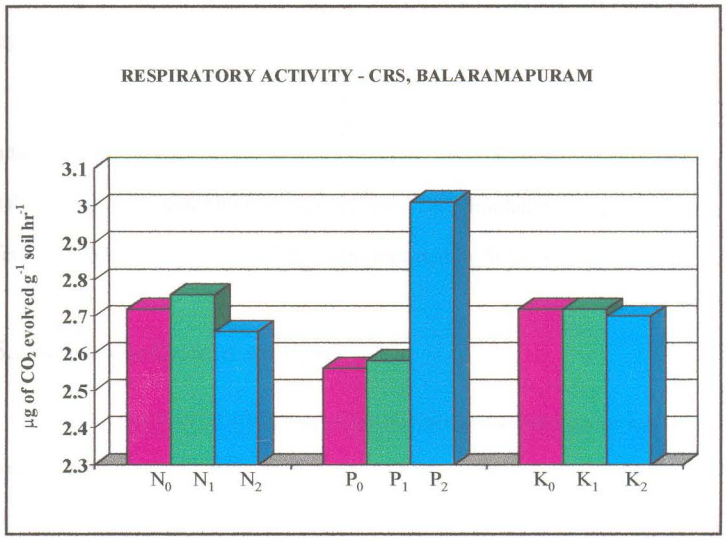

Fig 24

due to treatments on respiratory activity. The main effects of chemical fertilizers were significant except for K while the interaction effects were significant at all levels (Table 21 to 23; Fig. 24). Higher values were registered for treatments T_8 and T_{11} involving $N_0P_2K_1$ and $N_1P_0K_1$. The benificial effects of mineral nutrition on the proliferation of microflora might be reason for this enhanced respiratory activity. As the treatments carried uniform dose of organic manures, the general increase observed in microbial respiration is common for all treatments. Thus the enhanced respiratory activity could be attributed to the effect of chemical fertilizers alone. Further, the inherent status of these mineral nutrients in this soil also is low which give a quick responses to added fertilizers. Similar effects on respiratory activity due to mineral nutrition had been reported by Anwarzay *et al.* (1990).

Thus even with a moderately high organic sources, supplemental addition of chemical fertilizers could improve the activity of soil microflora substantially to produce high enzyme activity. The stimulatory role of chemical fertilizers on enzyme activity have been reported by Wheatley *et al.* 1990.

5.2. Path coefficient analysis

Direct and indirect effects of soil parameters studied on enzyme activities were brought about by path coefficient analysis. A close scrutiny of the data generated from the PME at the RRS, Kayamkulam, presented in Tables 26 to 27, indicated maximum positive direct effects on urease activity expressed

through sesquioxides (+0.37) followed by available nitrogen, respiratory activity and available P.

The direct effect of sesquioxide as expected was low in a quartzi psaments of the coastal Onatukara region, since the clay fraction was extremely low. The higher surface area manifested through the amorphous and crystalline sesquioxides compared to the quartz grained coarser fractions, could be able to sequester and protect the enzyme from denaturation and loss. Further, the population of microorganisms and the retention of exoenzymes are also likely to be higher in sesquioxide rich environment. This is again explained with its positive relationship with CEC through the indirect effects since appreciable increase in CEC is likely only through an increase in the finer fractions like sesquioxides. Similar results were reported by Crooke *et al.* 1982.

The direct effects of available N (+0.32) and respiratory activity (+0.26) is well explained by a higher microbial load associated with these two factors and hence the build up of urease as both exo and endo enzymes.

This is further evident through the indirect effects of respiratory activity (+0.14) and available P content (+0.17). As these soils are low in available P, P availability to microorganism is a limiting factor for its proliferation thus the content of both exo and endo enzymes. As P forms an essential nutrient, for the synthesis of high energy compounds in microbial cells, the energy turn over and all enzymatic reactions are controlled to a greater extent by its availability (Zantua and Bermer, 1975).

In the case of phosphatase activity, the maximum direct effect was expressed by CEC (+0.36), followed by pH (+0.35) and available K (+0.32) (Table 26). This is quite obvious from the relationship between the enzyme retention and CEC contributed through higher surface charge of the exchange complex. Further, the exchange complex, may be modified in many cases to the advantage of the enzyme molecule as the solubility and ionization is highly charge dependent owing to the iso electric nature of the protein part of the enzyme. The positive direct effect of pH on phosphatase activity as expected is due to the low ionic concentration of soluble PO_4 at higher pH and the specific protonation - de protonation mechanisms responsible for the enzyme stability and activity. Owing to the Zwitter ionic nature, the enzyme protein is also highly pH dependent, thus the creation of an optimum pH range for the maximal activity of this enzyme is essential. Similar effects of pH dependance of phosphatase has been reported by Feder (1973).

The indirect effect is exhibited through the available K (+0.18) and is well explained by the low K status of this soil since the maintenance of optimum population of phosphatase producing microorganisms requires a higher K flux in the soil solution.

With respect to protease activity, maximum direct effect was contributed by available P (+0.31) followed by pH (+0.25), sesquioxides (+0.21) and respiratory activity (+0.17) (Table 26). The nutritional effect of available P controlling the multiplication of microorganisms producing proteolytic enzymes

can be attributed to the direct effect of available P. Besides these, low N/P ratio accounted by high available P also impart a specific role in the multiplication of proteolytic bacteria contributing to higher protease activity (Rowell *et al*, 1973).

The optimum pH responsible for the activity of the proteolytic bacteria decides the quantity of protease released through the mineralization of organic matter. The increased surface area by the presence of sesquioxides in a sandy tract can be attributed to the adsorption of organic matter, and humic particles which ultimately contributed to the increased protease activity. Besides these, sesquioxide particles due to their increased surface area adsorb the extracellular protease thus accounting for higher protease activity in the sandy Onatukkara soils.

In the case of dehydrogenase, the direct effects are exhibited through available P (+0.35), CEC (+0.29), available K (+0.24) and organic carbon (+0.23) (Table 26). As in the case of protease, the available P plays a significant role by increasing the activity of dehydrogenase, which is an indicator of the overall microbial activity. The role of P as a constituent of high energy phosphate of the microbial biomass contributes to the acceleration of microbial population and thus results in increased dehyrogenase activity. Similar findings were also reported by Ladd and Paul (1973).

With the increase in CEC, the total quantity of organic colloids and the rate of adsorption of nutrient ions to the charged sites increases, which serves

as source of nutrients and energy for the proliferating microorganisms. This ultimately resulted in higher dehydrogenase activity, which indirectly represents the microbial load. This goes on hand with the findings of Naogar *et al.* (1997). Similarly the nutritional role of K, is inevitable as it influences the NH_4^+/K^+ ratio, which decides the availability of N. An acceleration of the microbial population was noticed which might be due to the immobilization of N resulting ultimately in the higher dehydrogenase activity.

The indirect effect was maximum with available K which was expressed through available P (0.32) (Table 27). Similarly the indirect effect of respiratory activity was mainly through the available P (0.24). In the sandy Onatukkara tract the indirect role of respiratory activity on dehydrogenase activity might be due to the significant effect of P, which is a major constituent of the energy rich compounds of the microbial cells. The indirect effect of sesquioxides was manifested through the respiratory activity (0.20). This clearly indicates that the role of sesquioxides similar to finer clay particles, thus serving as binding sites of organic matter and humic particles resulting in increased microbial load and respiratory activity.

The direct effects on cellulase activity was manifested by the factors respiratory activity (+0.38), sesquioxides (+0.32) and cation exchange capacity (+0.28) (Table 26). The higher the respiratory activity of the sample, the more was the cellulase activity. The respiratory activity is supposed to be a measure of the microbial population which includes all types of bacteria, fungi and

actinomycetes producing both extra cellular and intracellular enzymes including cellulase. This goes on hand with the finding of Pancholy and Rice (1973). Sesquioxides, at this tract serve as microsites like clay particles, whereby it binds these enzymes to their surface ultimately contributing to a higher activity.

As already mentioned, CEC imposes significant effect on the adsorption and release of nutrient ions which meet the nutritional requirement of the microbes, thereby increasing the activity of these enzymes which are either secreted by the living cells or released by the lysis of these cells.

The maximum indirect effect in the case of cellulase was by the factors available P (+0.25) and organic carbon (+0.23) through the respiratory activity (Table 27). As already mentioned, the available P content of the soil has a direct relationship with the respiratory activity of the microbes. It is also observed that the respiratory activity is a direct measure of the organic matter content and thus the increase in the organic matter will accelerate the respiratory activity. Similar results were noticed by Lehlinger, (1981).

Thus, under the sandy Onatukkara soils, the activity of various enzymes viz., urease, phosphatase, protease, dehydrogenase and cellulase were limited by the availability of P, sesquioxide content and the cation exchange capacity. The activity of these enzymes are thus contributed chiefly by the above factors.

In the case of the PME (Vty - Tall indica) at Pattambi, urease activity was seen influenced to the maximum extent by available P (+0.35) followed

by respiratory activity (+0.33) and CEC (+0.32). From the soil data presented in Tables 28 and 29 it is evident that the available P status is extremely low and hence a high urease activity with available P increase is obviously due to an increase in the microbial population. This nutritional effect of available P is also evident from the high respiratory activity observed due to a sudden spurt in the microbial population. Thus both the exogenous and endogenous urease activity was believed to be higher in this soil.

High cation exchange capacity directly influences the adsorpiton and retention of exoenzymes in the soil matrix thus protecting the enzyme molecules being lost from the system. Hence the activity of urease is likely to increase with increase in the cation exchange capacity of the soil. The reported positive direct effects of sesquioxides is possibly due to its indirect contribution to the surface area of the soil colloids and exchange sites which increased the P retention for microbial proliferation (Fauci and Dick, 1994).

The effect of CEC (+0.20), available K (+0.26), sesquioxides (+0.21) and respiratory activity (+0.20) on urease was indirectly influenced to the maximum extent by respiratory activity, available P and CEC. This inter dependence of CEC and respiratory activity is well documented through the effect of organic matter which serves as a source of energy for microorganisms and the chief source of organic colloidal fractions to contribute negative adsorpiton sites. The positive indirect effect of sesquioxides on available P under wet land paddy situations is primarily due to the reduction and subsequent

transformations of Fe and Al phosphates to soluble forms. Thus the phosphorus economy of the soil solution is always influenced positively by the sesquioxides under continuous submergence which in turn might have increased the P source for microbial proliferation (Fauci and Dick, 1994). The factors studied with respect to urease in this context has accounted for 83.4% of the total variability.

In the case of phosphatase activity, maximum positive direct effects was contributed by available N (+0.70) and CEC (+0.36) while respiratory activity expressed a negative effect (Tables 28 and 29). The positive influence of available N and CEC as expected is due to the multiplication of microorganisms in an N rich environment and the retention of higher levels of enzymes at higher CEC. As the soil is deficient in phosphate, the higher respiratory activity leads to faster immobilization of P from the soil by a wide variety of microorganisms which is believed to smother the activity of phosphate sensitive microorganisms. This competition between P sensitive bacteria and other microorganisms considerably reduces the multiplication of *Bacillus Cereus* and *Bacillus megaterium* which reduces the phosphatase activity. The positive influence of other factors are not relevant and significant in this context and hence are not discussed.

The indirect effects are chiefly centered around available P (0.56), available K (0.46), pH (+0.30), CEC (+0.18), sesquioxides (+0.43) and organic carbon (+0.18) which are expressed mainly thorough the effect of available N. Thus it is quite clear from the observation, that the mineraliztion of organic

sources and release of nitrogen, phosphorus and potassium virtually controlled the factors contributing to the main indirect effects. However only 41.4% of the total variability could be accounted.

With respect to protease activity, the main effects are due to available P (+0.60), respiratory activity (+0.44) and sesquioxides (+0.22). Besides the nutritional effect of available P, it serves as a constituent of the high energy compounds which control the microbial metabolism of proteolytic organism thereby influencing the activity. An increase in the respiratory activity indicated an increased microbial population which in turn had contributed to a higher protease activity. Similar results were reported by Asmer (1992).

Sesquioxides play an important role in forming organo mineral complexes and its solubility under submerged condition, influences the availability of P. Then these organo mineral complexes are subjected to microbial attack by the proliferating microbial colonies, thereby contributing to an increased protease activity. This is in conformity with the findings of Zantua and Bremner, 1977.

In the case of indirect effects of the factors the maximum values were noticed due to available N (+0.48) which was expressed through available P and the indirect effect of available K (+0.44) was due to available N (Table 29). It is universally accepted that N serves as an substrate to the proliferating microorganisms derived mainly through organic sources. This mineralization of organic matter results in the concomitant release of available P and available

K also. Hence the nutritional role of all the three elements are interlinked. This interdependence of these nutrients which are released from organic sources ultimately increases the microbial population which includes specific proteolytic bacteria resulting in increased protease activity.

In the case of dehydrogenase activity, the maximum direct effects were due to available P (+0.54), respiratory activity (+0.50), and CEC (+0.35) (Table 28). It is inferred from the data, that the multiplication of microorganisms in this soil is restricted to the availability of phosphorus. The vital nutritional role of P in the microbial proliferation is thus evident. Respiratory activity as expected is an indirect measure of the microbial biomass and hence its effect on dehydrogenase activity stands substantiated. This observation is in agreement with the findings of Martens *et al* (1992). The role of high CEC in protecting exo enzymes from losses, could be attributed to the direct positive effect.

The indirect effects on dehydrogenase activity was mainly exhibited by sesquioxides (+0.33) and pH (+0.31) (Table 29). As the sesquioxide content determines the total adsorptive surface for microorganisms per unit mass of soil, the total microbial biomass and the dehydrogenase activity to a greater extent depends on this parameter. Role of soil reaction in this context is very important owing to the protein nature of the enzyme with optimum activity at a particular pH. This observation is in conformity with the findings of Kumar and Kapoor (1995).

The respiratory activity (+0.33), CEC (+0.27) and sesquioxides (+0.26) served to be the major factors for cellulase activity (Table 28). The dependence of cellulease activity on CEC and respiratory activity is well documented through the effect of organic matter which serves as a substrate and source of energy for microorganisms and the chief source of organic colloidal fractions to contribute negative adsorption sites. Similarly sesquioxides also serve as colloidal fractions that bind the humic materials there by providing ample substrate for the cellulolytic bacteria to act upon. Similar effects of sesquioxides on cellulase activity was reported by Szegi (1988).

In the case indirect effects, the maximum values were reported by the factors CEC (+0.20) and clay content (+0.20) through respiratory activity and organic carbon (Table 29). It is already stated that, high CEC is associated with the organic matter in the soil, which serves as a source of carbon, energy and mineral nutrients for the multiplication of microorganisms contributing to higher cellulase activity. A direct relationship between clay content and OC is evident as an increase in clay content is also accompanied by increase in organic carbon status which serve as an energy source as well as substrate to stimulate the activity. The above views are supported by the findings of Tabatabai (1982).

A close scrutiny of the data generated from the PME-II revealed that the urease activity is influenced by the direct effect of factors available P (+0.57) EC (+0.25) and sesquioxides (+0.23) as observed from Table 30.

In the case of sesquioxides, the direct positive effect observed could be explained by its capacity to contribute adsorption sites for the free enzyme as well as for the enzyme organic complex. The positive relation with EC might be due to the nutritional role of mineral elements in the multiplication of microorganisms. The indirect effect exhibited by organic carbon is well explained by its role as a source of carbon and energy for the fast multiplying microorganisms contributing urease. Potassium on the other hand is a very important essential element in the mineral nutrition of microorganisms especially in a soil low in available potassium.

Phosphatase activity in this experiment was directly influenced through the parameters available P (+0.69), EC (+0.30) and available N (+0.27) (Table 30). The observed effect of available N and P on phosphatase activity could be attributed mainly to its nutritional role, since both are required for the synthesis of microbial protein and energy rich compounds. Thus, the enhanced multiplication of microorganisms both specific and non specific to phosphatase contributes to a higher enzyme activity. The influence of electrical conductivity on phosphatase activity might be due to the nutritional role of mineral elements present in the soil solution. The indirect effect of available K (+0.53) could also be explained through its nutritional role enhancing the microbial population thus resulting in a higher build up of urease in the soil. The effect of sesquioxide (+0.39) is indirect through its role as the site of adsorption for the released enzyme, thus minimizing the losses.

Maximum direct effects on protease activity was expressed by R_2O_3(+0.38), pH (+0.32) and CEC (+0.29) (Table 30). Thus it is clear from the data that the proleolytic organisms are highly pH dependent and are abundant in soils of high CEC. This is probably due to the higher fertility of the soil contributing higher levels of mineral nutrients, carbon and energy sources which results in a faster multiplication of the proteolytic organisms. The high sesquioxide content probably may add to high adsorptive surfaces for the free enzyme. Hence the losses that are likely for protease is minimised considerably.

The indirect effects exhibited by available P (+0.21) and available K (+0.18) in this context could be explained only through its nutritional effect in the multiplication of microorganisms producing protease.

In the case of dehydrogenase activity, CEC (+0.49), available K (+0.43) and sesquioxide content (+0.21) were found to impart direct positive effects (Table 30). The direct relationship between CEC and dehyrogenase activity as expected is attributed to a high nutrient status, carbon and energy source leading to a faster multiplication of the microorganisms resulting in a high dehydrogenase activity. Since the dehydrogenase represents the internal redox reactions of the microbial electron transport system, a high reserve of organic colloids associated with the soil is mandatory to serve as a source of electrons. The nutritional effect of K on the multiplication of general micro flora needs no explanation since it is vital in many metabolic activities. The role of

sesquioxides is mostly through its contribution to a higher surface area and adsorption site per unit weight of soil material for the microorganisms. This eventually results in a high dehydrogenase activity.

The indirect effects exhibited was mainly through the factors available P and AEC (Table 31). Both these parameters are closely related to P nutrition as the former deciding the concentration of immediately available P and the later related closely to the reserve of fixed P. Further, the relationship between AEC and sesquioxides also could impart indirect positive effects.

In the case of cellulase activity, maximum direct effects were noticed due to available P, R_2O_3 content and respiratory activity (Table 31). The nutritional role of P and its influence on the population of the microflora could be the main reason for this effect. Sesquioxides owing to its capacity to adsorb and complex the extracellular enzymes modify the activity and protect the cellulase group of enzymes from the possible losses. Further, the organo mineral complexes that are formed from sesquioxides will be able to harbour a variety of cellulolytic organisms per unit mass of soil than the constituent components separately. In all these cases the total load of cellulolytic organisms and thus the enzyme concentration will be higher.

Total respiratory activity being an indirect measure of the microbial proliferation is directly influencing the cellulase activity in the present study. An increase in respiratory activity in general increases the microbial biomass and the release of extracellular enzymes by cell lysis. Hence a higher cellulase

activity is theoretically sound at higher levels of respiratory activity (Alexander, 1978).

Maximum indirect effects were, manifested through the nutritional role of K and N derived from mineral and organic sources probably by their simulatory role in cell multiplication and biomass production. Thus the dynamics of cellulases in this location is mainly controlled by factors like available N, available P, available K, R_2O_3 and respiratory activity status.

Under the components of rubber based cropping system at the CES, Chethackal, the direct and indirect effects of the factors on enzyme activities were assessed and the results are discussed below.

With regard to urease activity, significant direct effects were noticed due to available N (+0.81), available K (+0.40) and pH (+0.15) (Table 32). The relationship between available N and the microbial biomass is well explained due to the role of N in the synthesis of microbial biomass protein. The availability of N increases the population of the urea hydrolysing organism thus contributing to high urease activity. This observation is in conformity with the findings of Gupta and Germida (1988).

The nutritional value of K is considered important in this context as large amounts of microbial protein consequent to a higher population could be able to contribute more nitrogen upon hydrolysis which could have stimulated the ureolytic bacteria. Thus, the effect is secondary through the

stimulation of the population of broad spectrum, non specific microorganisms in the soil. This is also in agreement with the reports of Gupta and Germida (1988).

In the case of indirect effect, the maximum values were noticed due to sesquioxides (+0.39) and respiratory activity (+0.34) which was expressed through available N (Table 33). The positive indirect effect expressed by the respiratory activity could be explained by the large microbial biomass formed contributing to a substantially higher level of N and other nutrients. Upon cell lysis the enzymes thus released might have been protected by adsorption on large sites of the sesquioxides in combination with organic colloids. This is in agreement with the findings of Martens *et al.* (1992) who described similar mechanism of enzyme protection.

It is observed from the Table 32 that the phosphatase activity at Chethackal is directly controlled by available N (+1.33), available K (+1.06) and CEC (+0.65). The direct effects of N and K, in the multiplication of microorganisms contributing to phosphatase activity is logical through the nutritional effects and effect of CEC is well explained by its role in protecting the enzyme being lost from the system. Indirect positive effects were also indicated by CEC (+0.58), sesquioxides (+0.57) and clay (+0.56) all the three contributing ultimately to an enhancement of the exchange behaviour, base status and the colloidal make up of the soil. Thus the sum total of direct and indirect effects is always favourable for the build up and protection of microbial

phosphatase in soils. This is in agreement with the findings of Kudzin *et al.* (1980).

In the case of protease activity, direct effects were expressed through available N, (+24.23) and sesquioxides (+10.84) while the indirect effects were contributed through clay content (+10.17) and respiratory activity (+8.81) as evident from Table 33.

It is clear from the observation that the nutritional role of N and K is dominant in this case, leading to a higher accumulation of protease enzymes from a variety of microorganisms. Their persistence and activity is seem stabilized by sesquioxides probably by adsorption and complex formation. The indirect positive effects of clay content as expected is consequent to its role in increasing the adsorption capacity for the enzymes. Higher respiratory activity obviously is related to a higher enzyme build up thus increasing the total protease activity. This is in agreement with the findings of Ladd and Butler (1972).

In the case of dehydrogenase also, direct effects were chiefly contributed through available N (+9.09) available K (+6.12) and CEC (+3.85) thus highlighting further the nutritional role of N and K and the exchange behaviour of the soil in maintaining a high enzyme activity. Indirect effects contributed through EC (+4.02) and sesquioxids (+3.92) could also be attributed to an increase in adsorptive surface and base status of the soil. Hence the combined action of direct and indirect effects contributed through five important soil

parameters namely available N, available K, CEC, EC and sesquioxides virtually control the activity of dehydrogenase.

Cellulase activity in this location is guided by the main effects of clay (+4.37), sesquioxides (+4.13) and respiratory activity (+2.10) while indirect effects are manifested mainly through available N (+1.83) and organic carbon (+1.49) (Table 33). All the above five parameters are closely related to either an increase in the microbial populations leading to a high enzyme build up of factors that contribute to the adsorption of enzyme, minimizing the loss. Thus a general increase in microbial activity coupled with a clean protection of the exo and endo enzymes in the soil system is the ultimate result.

Further, an increase in the organic matter provides easily degradable substrate for the cellulolytic organisms to utilize leading to an increased rate of cellulolysis and mineralization of organic matter. The observation is in agreement with the reports of Pennock *et al.* (1994).

Under the coconut based cropping system, at Balaramapuram the major effects on urease were due to available N (+0.42), sesquioxides (+0.21) and organic carbon (+0.18) (Table 34). As higher available N, stimulates initially the activity of microbes producing urea hydrolyzing enzyme, it has a significant direct effect on urease activity. The direct effect of sesquioxides on urease activity might be attributed to its role in combination with the organic phase of the soil to alter the exchange behaviour of the colloidal phase. Similar results have been reported by Rajendran (1992). The exchange properties of

the soil influences the adsorption and protection of urease secreted extracellularly. Thus the direct effects of these factors on urease activity stands substantiated.

Similarly the maximum indirect effects was due to clay content (+0.34) which was expressed through available N (Table 35). Clay content in a highly weathered soil of the study area serve as binding sites for the humic colloids which function as source of carbon and energy rich compounds for urease specific microorganisms in soils.

With respect to phosphatase activity, the maximum direct effect was exhibited by available P (+0.87), followed by AEC (+0.65) and available N (+0.28) (Table 34). The highly weathered oxidic soils of the PME at Balaramapuram was found to be deficient in P. Application of P through fertilizer sources had increased the P availability, which might have stimulated the proliferation of specific phosphatase producing organisms through its nutritional role. Further these organisms were able to utilize the unavailable fixed P and organic P as substrates, thus exhibiting a high phosphatase activity.

The positive effect observed in the case of AEC, could be attributed to its effect by providing insoluble phosphatase accumulated through P fixation. This positive indirect effect exhibited by organic carbon might be due to its role as source of carbon and energy for phosphate producing organisms to multiply. This is in accordance with the findings of Halm *et al* (1971).

With respect to protease activity, available N (+0.38), OC (+0.34) and sesquioxides (+0.32) were found to impose direct effects (Table 34). The positive direct effect of available N on protease activity is attributed to the nutritional and stimulatory role of nitrogen in the multiplication of microorganisms in general with higher build up of biomass protein. Subsequent cell lysis leads to the release of proteases into the soil to become a part of the pool of exo enzymes. With the increased availability of easily degradable organic sources, the proliferation of the proteolytic bacteria might have increased considerably enhancing the activity of protease. Adsorption of the enzyme on the sesquioxides might have protected enzyme protease from various losses (Rowel *et al*, 1973).

The maximum indirect effect by available P (+0.16) imparted was through organic carbon and both exhibit their nutritional role in relation to the multiplication of proteolytic organisms.

While in the case of dehydrogenase, a close scrutiny of the data indicates the direct effects of the factors, organic carbon (+0.34), available N (+0.32) and sesquioxides (+0.29) (Table 34). The increase in the microbial biomass is dependent on the availability of organic C sources and ultimately an increase in the dehyrogenase activity as they reflect the total range of oxidative activities of soil microflora. Similarly N serve as a stimulator of enzymatic activities through enhanced microbial activity thus contributing to higher dehydrogenase activity (Ladd and Paul, 1973). As mentioned earlier the role of

sesquioxides as binding agents of enzymes cannot be ignored in the PME at Balaramapuram.

The indirect effect of available P expressed trough organic carbon could be attributed to the association of phosphorus in the organic form (organic P) serving as a substrate, mainly for the multiplication of P-solublizing organisms. Thus the high microbial activity observed could be able to contribute for a higher dehydrogenase activity.

In the case of cellulase activity, the positive direct effects expressed through sesquioxides, (+0.29), pH (+0.20) and available N (+0.20) could be explained by the highly weathered nature of the soil with acidic soil reaction. The positive direct effects of available N might be attributed to the nutritional role in the multiplication of microorganisms involved in cellulolytic activity. A pH optimum for cellulase activity also is expected, because of the protein nature of the enzyme. A higher sesquioxide content in such a highly weathered soil could be able to sustain and protect a high reserve of the enzyme by adsorption and complex formation. This observation is in conformity with the findings of Kong and Dommergues, 1972.

The indirect positive effects expressed by respiratory activity was through organic carbon which is well explained by the role of both in maintaining a higher microbial activity. Thus, cellulase activity in this soil is mainly controlled by the R_2O_3 content, pH, available N, respiratory activity and organic carbon.

5.2.1. Enzyme kinetics

The Michaelis constant (K_m) is a measure of the affinity of an enzyme for its substrate, the lower the K_m the higher the affinity (Vaughan and Ord, 1991). It is observed from the data on Table 36 that there exists a positive relationship between V_{max} and K_m values of all five enzymes for the five locations included in the study. Since V_{max} is proportional to enzyme concentration, the data suggests that the soils may contain different amounts of free and bound enzymes (Perucci and Scarponi, 1983). As the enzymes bound to soil components generally have higher values of K_m than free enzyme (Nannipieri *et al.* 1978), the comparatively higher K_m values observed for samples from Chethackal, Pattambi and Balaramapuram might be attributed to the immobilization of liberated enzymes by the active colloidal fractions, both organic and inorganic (Cervelli *et al* 1978). It is relevant in this context to state that the affinity of the enzymes in general is more in soils low in colloidal fractions like Kayamkulam at lower concentrations of the substrate as evident from the low values of K_m, primarily due to the occurrence of the lion's part of the enzymes in the free form. This observation is in conformity with the findings of Speir and Cowling (1991).

Thus it is clear from the study that the affinity of urease for substrates in soil decreases in the order Kayamkulam = Pattambi-I > Chethackal > Balaramapuram > Pattambi-II, while that of phosphatase follows the sequence Balaramapuram = Chethackal > Pattambi-I > Kayamkulam > Pattambi-II.

Highest affinity for substrate in the case of protease was observed for Kayamkulam followed by Pattambi-II, Pattambi-I, Balaramapuram and Chethackal. On the other hand the affinity constants showed the lowest values for dehydrogenase and cellulase in the case of Balaramapuram and the highest values were reported for Kayamkulam and Pattambi-I respectively.

The highest total enzyme activity observed for samples collected from the CES, Chethackal, even with a low K_m value for all the enzymes may be attributed to a higher total pool of the enzymes, both free and bound forms released over a period of incubation. Thus even though the affinity at lower concentration is poor for this soil, the higher reserve of total enzymes could achieve a comparatively higher turn over within a period of time.

As the V_{max} values are well correlated positively with the total enzyme concentration, a higher V_{max} is always indicative of a higher total activity of the enzymes. It is quite evident from the data presented in Table 36. This observation is in conformity with the findings of Vaughan and Ord (1991).

5.3. Pot culture studies - Effect of agrochemicals

The results obtained from the pot culture experiment to study the effect of agrochemicals on five major soil enzymes viz., urease, phosphatase, protease, dehydrogenase and cellulase are discussed below.

5.3.1. Urease activity

The assay of urease activity in samples collected at the three critical stages of the crop clearly indicated highly significant effect due to treatment variations. As these agrochemicals tested are reported to suppress microbial activity, significant reduction in the urease activity is expected since it is of microbial origin. In all the cases, urease activity decreased drastically compared to control towards the active tillering stage. At the active tillering stage, the maximum reduction for treatments involving insecticides has been observed in the case of monocrotophos followed by quinalphos, carbofuran and phorate (Table 38; Fig. 25). It is observed that irrespective of the stage of the crop, values for urease activity was maximum for the control. However, the inhibitory effects were more pronounced in the case of herbicides than insecticides exhibiting maximum effect by 2, 4-D followed by benthiocarb, oxyflourfen and butachlor. The treatment wise inhibitory effect on urease activity was maximum for hinosan when the whole result was examined. The observed reduction in urease activity consequent to the application of fungicide is believed to be due to the interference in the turn over rate of urea-enzyme substrate complex by these chemicals. Hence, they act as competitive inhibitors for the reversible binding of the actual substrate, thus reducing the active enzyme substrate complex molecules temporarily immobilized leading to a decreased product formation. This observation is in conformity with the findings of Lethbridge *et al* (1981) who reported suppressing of urease activity

at elevated doses of phosphorothioates such as fenetrothion, malathion and phorate. It was further reported by Sarkar *et al* (1989) that the same effect could be achieved by herbicides in soil.

Thus it is clear that degradation of these chemicals in soil chemically or biochemically could restore the activity with the advancement of time. This is well explained by the observed increase in the activity towards the panicle initiation stage irrespective of the nature of chemicals applied. The exact mechanism by which these pesticides impart suppression of urease activity needs further elaboration.

The observed increase in activity during panicle initiation stage could be attributed to the rhizosphere effect leading to a faster multiplication of the active flora involved in urease production triggered by the excretions and secretions of the roots. Further, degradation of the pesticide applied, chemically and biochemically, its inactivation by absorption processes with clay organic complexes and plant uptake had reduced the concentration of these chemicals with time (Burns, 1982). A close scrutiny of the data reveals that the relative persistence of the active chemical is more in the case of insecticides, fungicides and antibiotics compared to other agro chemicals (Table 39; Fig. 25). Though the treatments did not produce any significant effect on urease activity, the maximum suppression at this stage was exhibited by chlorpyriphos (79.80) compared to the control (137.63). The best inhibitory effect at this stage was expressed by fungicides and antibiotics though the effect of the treatments

were non significant. Hence the inhibitory activity on urease follows the sequence herbicides < insecticides < fungicides < antibiotics.

The suggested mode of action of these chemicals on enzyme activity includes direct inhibition of intra and extra cellular enzymes, by a reversible binding of the pesticide to the enzyme protein competitively with the substrate, thus mimicing the transition stage analogs (Brownman and Tabatabai, 1978) in many cases. However herbicides like butachlor, oxyflourfen 2, 4-D and benthiocarb are reported as irreversible enzyme inhibitors of urease after disappearance of a major portion of the actual substrate. The observed decline in the activity of urease by streptomycin was reported to be due to the substitution of amide groups of the enzyme protein by this chemical which changes the protein conformation and sequence of catalytic events specific for the hydrolysis of the natural substrate (Andrews *et al.* 1984).

During the harvest stage where the physiological maturity of the crop is attained, biological processes in the rhizosphere is deteriorated due to a decline in the microbial activity. This effect was clearly noticed with respect to urease activity at this stage. Low moisture content and nutrient status also influenced the urease activity in this context and ultimately resulted in a decrease. At this stage maximum suppression of the activity was observed in the case of insecticides, followed by antibiotics, fungicides and herbicides (Table 40; Fig. 25). Thus it is evident that under an undesirable soil condition

like the harvest stage, insecticides (quinalphos) impart higher inhibitory activity than the other chemicals tested.

5.3.2. Phosphatase activity

Phosphatase represent a broad range of intra cellular as well as soil accumulated enzymes that catalyzes the hydrolysis of both esters and anhydrides of phosphoric acid (Speir and Ross, 1978).

With respect to phosphatase activity, a similar trend as observed for urease activity, with a drastic decline from the zero sampling stage to the active tillering stage followed by an increase upto the panicle initiation stage and thereafter with a steep decline at harvest stage was observed. At the active tillering stage (Fig 26.) it was noticed that the effect of insecticides like phorate, carbofuran, quinalphos, monocrotophos and fungicides like hinosan were less pronounced when compared to herbicides which included benthiocarb, 2-4-D, butachlor and oxyflourfen. However streptocycline imparted the maximum inhibitory effect.

Thus it is clear from the data that maximum inhibitory effect is accrued in the case of antibiotics followed by fungicide, herbicide and insecticides. Since bacterial population is the most affected by antibiotics, it is evident that phosphatase activity is restricted to a greater extent in bacterial cells. As herbicides, insecticides and fungicides are more prone to chemical and bio-chemical degradation, their persistent toxic effects were less compared to

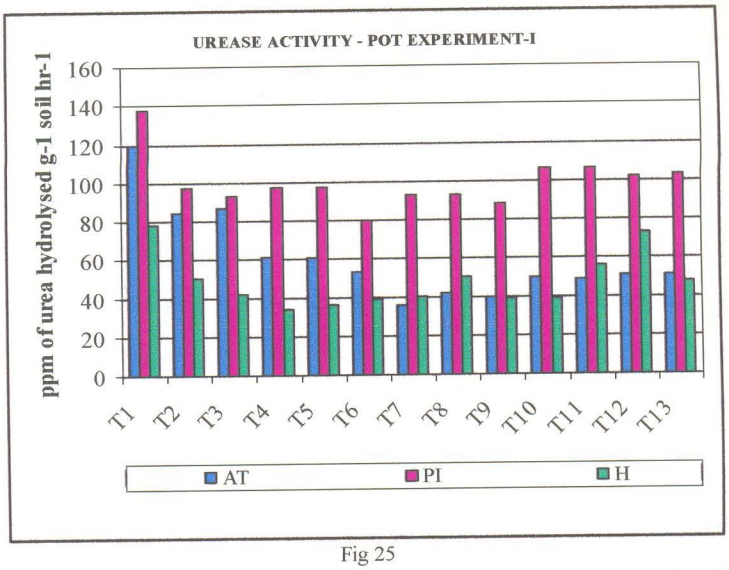
UREASE ACTIVITY - POT EXPERIMENT-I
ppm of urea hydrolysed g-1 soil hr-1
160
140
120
100
80
60
40
20
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 25

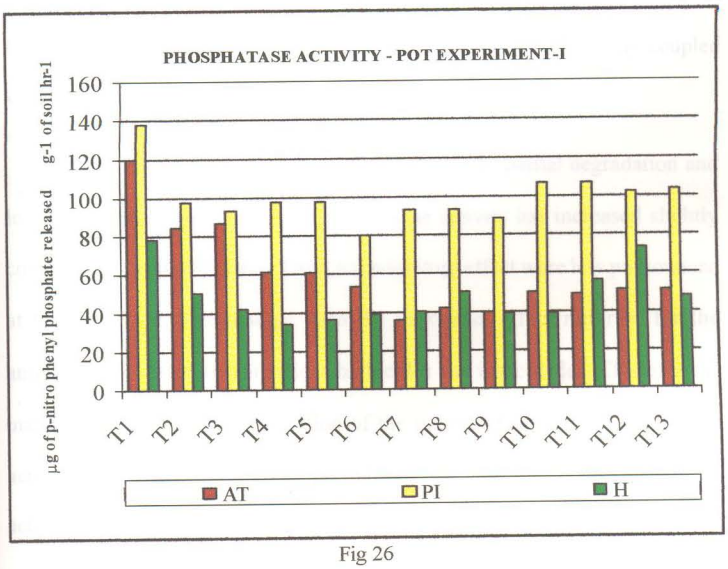
PHOSPHATASE ACTIVITY - POT EXPERIMENT-I
µg of p-nitro phenyl phosphate released g-1 of soil hr-1
160
140
120
100
80
60
40
20
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 26

antibiotics. Further the interaction of insecticides, herbicides and fungicides with soil components also might have diluted the inhibitory effects of these chemical on enzyme activity.

Thus, the observed inhibitory effect of agrochemicals on phosphatase activity reported was due to a drastic decline in the population of phosphatase producing bacteria and interference of these chemicals in the mechanism of enzyme action; both irreversible and reversible mechanisms of inhibition have been suggested to affect free enzyme and enzyme substrate complex formed.

With the advancement of time it is seen that the activity gradually increases due to the inactivation of these chemicals in soil by different mechanisms. The above observations are in agreement with the findings of Quilt *et al.* (1979) who observed a decrease in phosphatase activity coupled with the application of herbicides and antibiotics.

With the advancement of time, consequent to partial degradation and loss of activity of the chemicals, the enzyme activity has increased slightly compared to the initial status. Thus, the inhibitory effect were less pronounced at the panicle initiation stage with the maximum effect reported for the antibiotics, followed by fungicides herbicides and insecticides. This effect is mainly due to a gradual restoration of the microbial population which was active in the transformation of phosphates. Further, enhanced rhizosphere activity, availability of C-source and mineral nutrients at this particular stage

of the crop might have contributed to the production of both exo and endo enzymes.

At the harvest stage, a drastic decline in the phosphatase activity was noticed as observed from Fig 26. The activity recorded for the control plot was comparatively higher than other treatments involving the application of agrochemicals. The maximum inhibitory effect was noticed with the application of antibiotic (streptocycline) while the effect of herbicides, insecticides and fungicides were on a par with each other. Similar results were reported by Greaves *et al.* 1981 who observed reduction in phosphatase activity due to the application of Dalapon.

Owing to the significant results obtained for treatments at the harvest stage (Table 40), the persistence of these chemicals and their continued inhibitory effect in the soil is evident. Besides persistence, the formation of bound residues with soil humic matter may result in continued activity till harvest. The highest value recorded for the control indicated the normal non-inhibitory metabolic reactions, where by the activity was increased. At this stage a general decrease in activity observed compared to the other two stages could be attributed to the lack of optimum conditions for the availability of moisture, C-substrates and nutrients.

5.3.3. Protease activity

Protease activity in soil samples collected arises out of protein substrates

of plant and animal origin. The activities are based on the release of amino compounds determined quantitatively by colorimetric methods.

Significant difference in the protease activity due to treatment effects was observed in the study. At the active tillering stage, maximum suppression of protease activity was observed in the case of treatments receiving fungicides, hinosan and bavistin followed by the treatment receiving the antibiotic streptocycline (Table 38; Fig. 27). The inhibition was maximum during the active tillering phase compared to the initial values at zero sampling stage. The inhibitory effects of insecticides viz., phorate, monocrotophos and chlorpyriphos were low compared to fungicides and antibiotic. The effects of herbicides were statistically on a par with insecticides.

The decrease in activity of protease observed during this stage consequent to the application of chemicals is correlated to the decline in the microbial population and hence the decrease in the production of these enzymes. Further, the sensitivity of the bacterial species to these chemicals also contribute to this effect since the mode of action varies considerably with the chemical nature of the material used. Thus, it is inferred that the species of microbes which produces protease in relatively larger amounts are affected mostly by fungicides, followed by herbicides and the least by insecticides. Hence, it is logical to conclude that fungi is the dominant species involved in protease production in the soil used for the study. Further, bonding of these enzymes to colloidal fractions may protect the enzyme proteins unless free

active enzymes are continually synthesised depending upon the availability of energy sources. As the population of the species involved in the synthesis of this enzyme is affected adversely by these chemicals, even under a high energy source, the activity is expected to be very low. The observed results are in conformity with the findings of Rowell *et al.* (1973).

An increase observed at the panicle initiation stage (Fig. 27) in protease activity might be partly attributed to a sudden spurt in the population of the proteolytic flora leading to an increase in the mineralization rate of organic matter accumulated during the active tillering stage and the associated lysis of the microbial cells contributing to extracellular proteases. In addition to this, the degradation of the chemicals undergone by chemical and microbial processes leading to a substantial reduction in the pesticidal activity also might have contributed to a fabulous increase in the microbial population involved in protease activity. At this stage, the effect of treatments were not significant while the lowest value was recorded for fungicides (hinosan and bavistin). The non significant results were attributed to the observed decrease in the population of proteolytic bacteria and fungi which ultimately resulted in a reduced proteolytic activity. Similar effects on protease activity have been reported by Nowak and Michalwicz (1988) consequent to the application of phenmediphan at 10mg/kg. The relatively non significant decrease in activity due to herbicide treatment may be due to the highly specific nature of the chemicals and the selectivity relationship with the species involved in protease

synthesis. Such selectivity of agro chemicals on protease activity have been reported by Wantanabe and Hayano (1994) in upland soils.

At the harvest stage, significant effect due to treatments was evident (Table 40). The effect of quinalphos was prominent which showed high persistence in soils as indicated by the lowest activity for proteases. It is also indirectly evident that the accumulation of these chemicals in soil is non uniform consequent to differential utilization by plants and its disappearance from the soil. Thus, the individual effects of these chemicals are governed by a group of broad environmental and plant factors rather than its direct toxic effects on microorganisms producing this enzyme (Burns, 1982). The observed increase in activity of protease towards the harvest stage was more or less same for all the treatments except for quinalphos compared to the initial value at zero sampling. Thus the reversible increase in the activity may be attributed to the restoration of the activity of proteolytic microorganisms. An enhancement in C-mineralization rate at the harvest stage leading to a higher energy source also might have contributed to a faster multiplication of these organisms.

5.3.4. Dehydrogenase activity

Dehydrogenase conduct a broad range of oxidative activities that are responsible for degradation of organic matter. In a cascade of events involving specific carriers, electrons and hydrogen are transferred from substrates to oxygen as the final acceptor. Hence, the dehydrogenase activity is considered

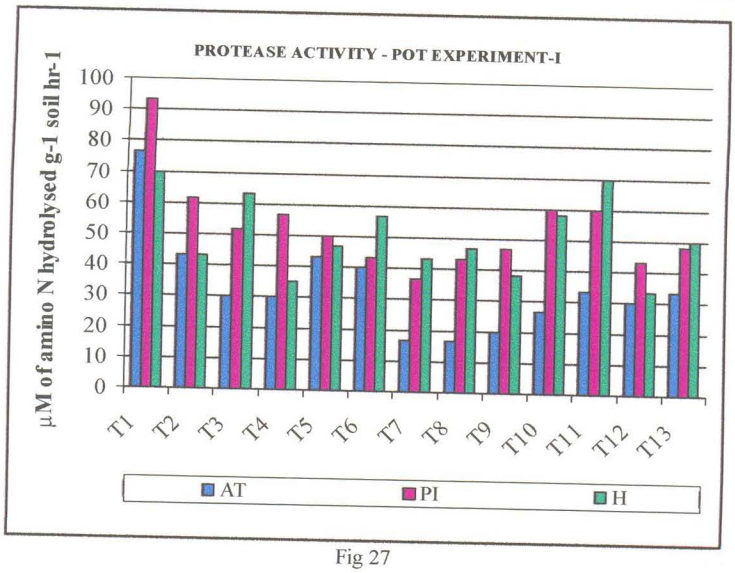
PROTEASE ACTIVITY - POT EXPERIMENT-I
µM of amino N hydrolysed g-1 soil hr-1
100
90
80
70
60
50
40
30
20
10
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 27

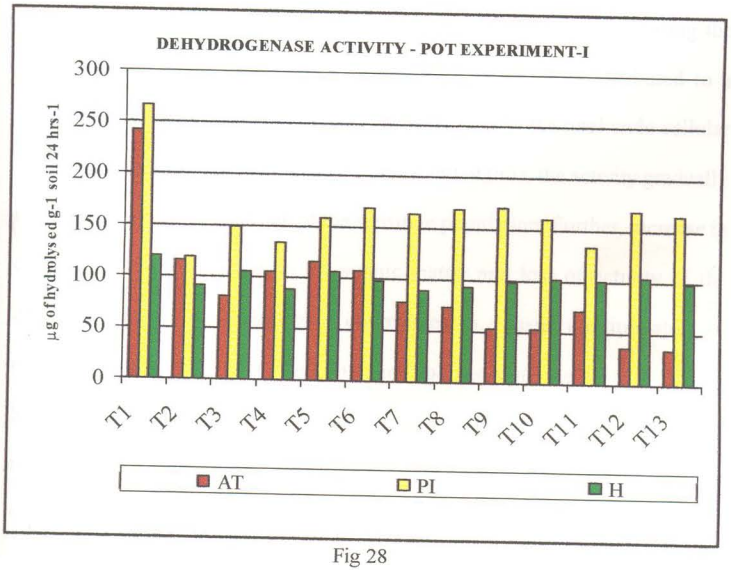
DEHYDROGENASE ACTIVITY - POT EXPERIMENT-I
µg of hydrolysed g-1 soil 24 hrs-1
300
250
200
150
100
50
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 28

as one of the better indicators of microbial activity since this enzyme occur within living cells while others can occur in an extra cellular state. They represent a class of enzymes that has been most intensely studied, to gain information about the influence of xenobiotics on activities of microorganisms in soils. The effect of agro chemicals studied in the present experiment on dehydrogenase activities are discussed below.

From the results, it is well understood that the treatments imposed significant effect on the dehydrogenase activity (Table 38). The effect of streptocycline (antibiotic) was drastic resulting in decreased activity. Similarly the effects of benthiocarb and oxyflourfen were also very prominent and on a par with streptocycline. As the dehydrogenases can be regarded as indicators of microbial activities of soil, this observed decrease in activity during the active tillering stage compared to initial zero sampling is attributed to a suppression of microbial population and the decline in the total endo cellular dehydrogenase in the system. With advancement of time, the activity gradually resumed owing to the build up of the sensitive population. Further, increase in carbon source, mineralization of organic matter and loss of activity of the chemical applied also might have contributed to a faster multiplication of microorganisms producing more enzymes and restoring the activity in soil. Similar effects of pesticides on dehydrogenase activity in soil have been reported by Chander and Brookes (1991) and in all cases the total endo cellular enzyme pool determines the activity.

A decrease in the microbial population was observed with the application of herbicides (benthiocarb, butachlor and oxyflourfen) and antibiotic streptocycline. This might due to the impairment of one segment of the microbial community, which is compensated by increased activity of other genotypes, with a resulting increase in overall activity at the next stage (Chander and Brookes, 1991).

The behavioural pattern was also similar with an increase from active tillering to panicle initiation stage following a decline at the harvest stage (Fig. 28). At the panicle initiation stage, the effect of phorate and 2, 4-D was more pronounced resulting in decreased activity probably due to selectively higher persistence of these chemicals in soils. An increase observed at this stage can be attributed to the mineralization of native organic matter which ultimately resulted in increased microbial activity. At the harvest stage the effect of quinalphos and hinosan was found to be dominant and resulted in the decreased activity of dehydrogenase (Table 40; Fig. 28). The observed results are in conformity with the findings of Dick (1994). A general decrease in the activity of dehydrogenase at this stage could also be attributed to the low moisture content, organic carbon status and nutrient availability.

5.3.5. Cellulase activity

Cellulase catalyses the conversion of insoluble sugars into water soluble mono or di saccharides and consist of three distinct classes of hydrolytic

enzymes including β-Glucosidase, which at the end of cellulase degradation hydrolyses cellulose to glucose.

At the active tillering stage, the effects of the fungicides like hinosan and bavistin were drastic which resulted in lower activity for cellulase (Table 38; Fig. 29). The application of pesticides like phorate, monocrotophos and chlorpyriphos imposed lesser toxic effects compared to other chemicals. Similar effects on cellulolytic activity of soil was reported by many workers (Ross *et al.* 1984; Katayama and Kuwatsuka, 1991) consequent to the application of herbicides, fungicides and insecticides Schaffer (1993) reported complete inhibition of cellulolytic activity with the addition of fungicide (Trichlamide) at higher doses whereas at field rate the activity was about 50% inhibited.

In the case of flooded soils since anaerobic cellulolytic bacteria and cellulolytic fungi are predominant, the inhibitory effects exhibited by fungicides and antibiotics on these are expected. Thus insecticides as a class, has lower inhibitory effects on cellulase activity under flooded conditions indicating that various cellulolytic microorganisms are less affected comparatively at the recommended dose.

On the contrary, the effect of insecticides seems to persist till the panicle initiation stage and give lower values for enzymes compared to other chemicals (Table 39; Fig. 29). Hence, phorate, carbofuran and monocrotophos had given significantly lower values for cellulase activity. This inhibitory effect observed by insecticides over other chemicals might be attributed to the relatively higher

persistence in the flooded conditions. The above inhibitory effects of these insecticides are reported to be due to the interference of these chemicals and some of their metabolites in specific steps in the glycolytic sequence of the cellulolytic bacteria suppressing their activity in the soil. This is in conformity with the findings of Katayama and Kuwatsuka (1991).

At the harvest stage the effect of herbicides (butachlor and oxyflourfen) were found to be much pronounced while the effect of insecticides was less pronounced due to the restoration of the activity of the cellulolytic bacteria (Table 40). The order of sequence is given as herbicides > antibiotics = fungicides > insecticides. This also suggests possible effects of reduction in biosynthesis of proteins and nucleotides and suppression of cell division of the cellulolytic bacteria by these chemicals. This goes on hand with the findings of Ross *et al.* 1984.

5.3.6. Microbial biomass

Measurements of soil microbial biomass have been shown to give early indications of long term changes in soil organic matter content (Powlson *et al.* 1987). This experiment therefore affords a unique opportunity to assess the effects of application of insecticides, fungicides, herbicides and antibiotics on soil microorganisms in a statistically balanced experiment.

At the active tillering stage microbial biomass remained relatively unaffected by the effect of treatments involving the application of herbicides

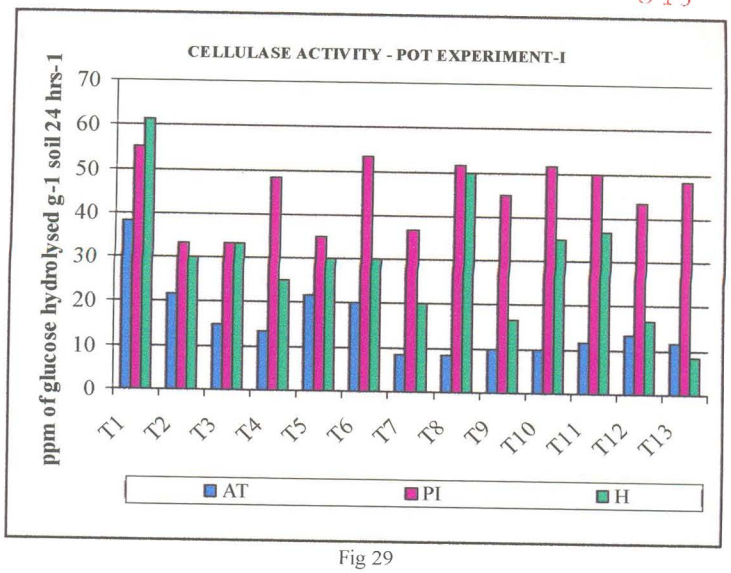
CELLULASE ACTIVITY - POT EXPERIMENT-I
ppm of glucose hydrolysed g-1 soil 24 hrs-1
70
60
50
40
30
20
10
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 29

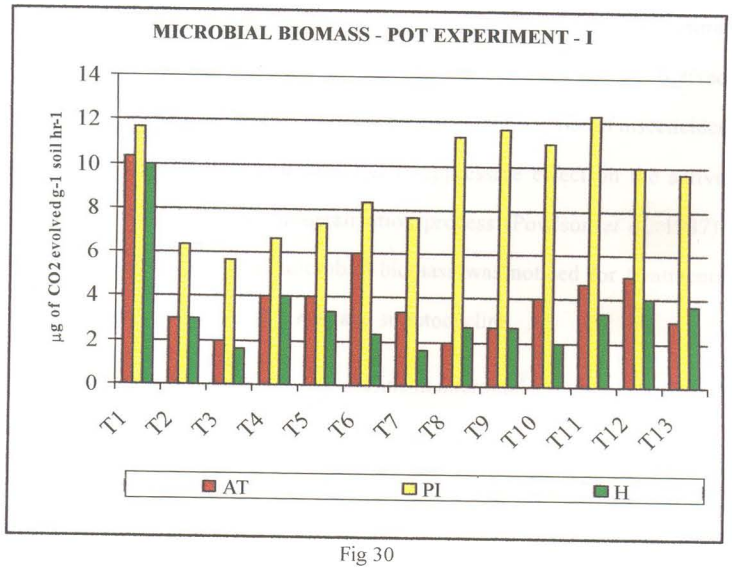
MICROBIAL BIOMASS - POT EXPERIMENT - I
µg of CO2 evolved g-1 soil hr-1
14
12
10
8
6
4
2
0
T1
T2
T3
T4
T5
T6
T7
T8
T9
T10
T11
T12
T13
AT
PI
H

Fig 30

(2, 4-D, benthiocarb, oxyflourfen and butachlor) and insecticides (monocrotophos, chlorpyriphos and quinalphos) (Table 38). On the other hand, a drastic decrease in the microbial load was observed due to the application of carbofuran, phorate, streptocycline hinosan and bavistin. This observed decrease in microbial biomass could be attributed to the decrease in the population of microorganisms involved in various transformations. The effect is significant compared to control in all cases. The observed decrease in biomass C is supposedly due to direct toxic effects of these chemicals on microorganisms as well as due to the indirect effects caused through plant and soil factors.

As in the case of enzymes an increasing trend was noticed with the advancement of growth (Fig. 30). At the panicle initiation stage significant suppressive effect on the microbial biomass was observed with the application of phorate, carbofuran, quinalphos, monocrotophos and hinosan. The observed decrease could be attributed to the relatively higher persistence of insecticides and fungicides through their prolonged suppressive effect on the active microflora involved in the mineralization process (Powlson *et al.* 1987). However higher values for microbial biomass was noticed for treatments involving the application of 2, 4-D and streptocycline.

At the harvest stage a drastic decline in microbial biomass was noticed which might be due to the addition of low amounts of organic matter which ultimately resulted in lower microbial biomass. However significant effects due to treatments was also observed (Table 40; Fig. 30). The treatments

involving the application carbofuran and hinosan have caused a drastic decline compared to other chemicals which might be due to their selective action on the active bacterial and fungal population involved in mineralization processes. Similar selective action of agrochemicals on soil microbial biomass was reported by Jones *et al.* (1991).

5.3.7. Yield and yield attributes

The effect of treatments on yield and yield attributes showed significant difference (Table 41). Number of productive tillers were more in the case of treatments involving the application of quinalphos and streptocycline. However, when the final yield of grain and straw was computed, no significant difference could be noticed due to treatment effects. Thus, the application of treatments alone could not make any improvement in the final yield though it had influenced the soil quality considerably by affecting the activity of major enzymes. The impact of this observed suppressive effect is multi dimensional and is not visible through a pot experiment in a single season or year. Continued effect of agrochemicals on microbial transformations of soil organic matter and its environmental consequences are to be assessed further through repeated pot experiments and field experiments. Thus the study needs further elaboration to make any generalization on yield and yield attributes. The present study is intended to assess the effect of agrochemicals on enzyme activities and hence the above aspects are beyond the scope of this study.

5.4. Pot culture studies - Effect of fertilizers, lime and manures

Results of the pot culture experiment to study the effect of fertilizers, lime and manures on soil enzyme activities are discussed below.

5.4.1. Urease activity

A close scrutiny of the data at the active tillering stage presented in Table 42; Fig. 31 revealed significant effects due to the application of fertilizers, lime and manures. Except for the interaction between F x L, other interactions were found to be significant. The highest value recorded for $F_1L_1M_2$ (261.57) indicated the significant effect of fertilizer, liming and vermicompost application compared to the control. Thus it is evident that soil enzyme (urease) activities increased with the application of inorganic fertilizers (Nannipieri, 1983). This suggests that these inorganic fertilizers stimulate the activity of the urease hydrolysing organisms leading to a higher level of urease.

At the panicle initiation stage, the effects of fertilizers, lime and manures were found to be significant. Among the interactions, the L x M interaction alone was significant (Table 44; Fig. 31). As observed in the active tillering stage, the highest value was noticed for the treatment $F_1L_1M_2$ involving the application of fertilizer, lime and manures (vermicompost) (276.00). A gradual increase in the urease activity from the active tillering stage to the panicle initiation stage was noticed (Fig 31). This general increase can be attributed

to the increased microbial biomass, due to the supply of nutrients and carbon source from the amendments besides changing the soil reaction. This spurt in microbial population resulted in an enhanced synthesis of urease both extracellulary and internally.

A sudden decline in the activity was noticed at the harvest stage which might be due to the general decrease in microbial population (Fig 31). At the harvest stage, the observed decline was attributed to the lack of C substrates or the exhaustion of C sources thus leading to a decrease in microbial flora resulting in reduced activity. Even at this stage, the main effect and the interaction effects of treatment was highly significant. The beneficial effects of fertilizers, lime and vermicompost were noticed even at the harvest stage. Similarly microorganisms associated with organic residues (vermicompost) may also contribute to the urease pool in the soil. This increase in urease activity with the addition of vermicompost along with fertilizers and lime may be due to the incorporation and promotion of urease enzyme fraction upon increasing the soil nutrient status, carbon source energy sources and humus. The low values recorded for $F_0L_0M_0$ are due to the limited supply of carbon substrates, nutrients and energy sources.

The application of vermicompost also suggests the release of growth promoting trigger molecules or the release of a growth promoter by the decay of the organic amendments that stimulate soil organisms to secrete high levels of urease enzymes. This promotion of soil urease activity was suggested by

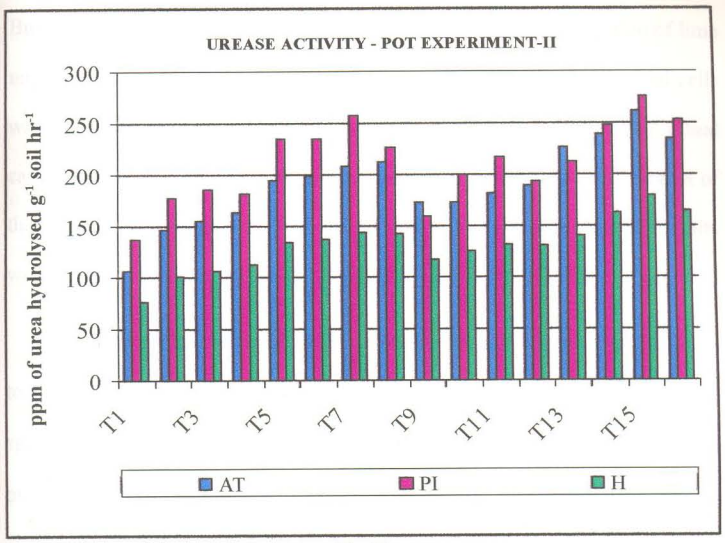
UREASE ACTIVITY - POT EXPERIMENT-II
ppm of urea hydrolysed g-1 soil hr-1
300
250
200
150
100
50
0
T1
T3
T5
T7
T9
T11
T13
T15
AT
PI
H

Fig 31

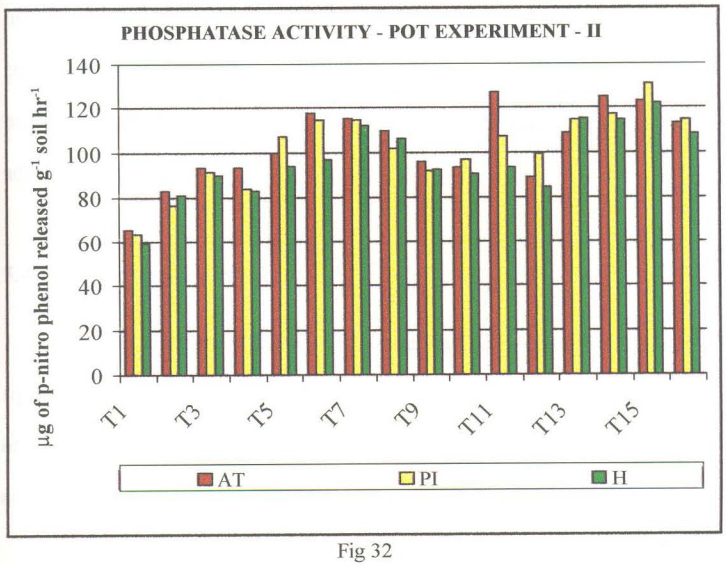
PHOSPHATASE ACTIVITY - POT EXPERIMENT - II
µg of p-nitro phenol released g-1 soil hr-1
140
120
100
80
60
40
20
0
T1
T3
T5
T7
T9
T11
T13
T15
AT
PI
H

Fig 32

Burns (1982). The increase in the urease activity with the application of lime might be due to the leakage of these enzymes from the dead microbial cells which are killed by liming. Moreover, the increased availability of microbial carbon source as a result of liming might have improved the proliferation of the microflora responsible for the synthesis of this enzyme. Similar results were reported by Skogland *et al* (1998).

Thus a general increase of the urease activity from that active tillering to panicle initiation stage followed by a decline in the activity at the harvest stage was observed (Fig). From this study, it is also evident that the application of fertilizers, lime and manures (vermicompost) in combination impart significant positive effect on the microbial population ultimately resulting in increased urease activity. Similar studies conducted revealed the long term enhancement of soil urease activities consequent to the formation of stable humic enzyme complexes derived from the organic amendments routinely added (Sandor, 1987).

5.4.2. Phosphatase activity

A close scrutinity of the data presented in Tables 42 to 50 revealed the significant effect due to treatments on phosphatase activity. The addition of organic amendments were found to maintain high levels of acid phosphatase activity compared to the control. The phosphatase activity showed a steady

increase from zero sampling through active tillering upto the panicle initiation stage and then registered a decrease at the harvest stage.

At the active tillering stage as shown in Table 42; Fig. 32 both the individual and interaction effects of F, L and M were non significant. It is inferred that the application of fertilizers, lime or manures can not alter the rate of phosphatase synthesis immediately which show the non availability of substrate immobilized in the form of organic P from the applied manures. Further P fertilizers temporarily increased the soluble phosphate pool of the soil and reduced the utilization of insoluble phosphates by phosphatase. Similar reports were been reported by many workers (Cunderova and Zubets, 1969; Nannipieri *et al.* 1978).

No relationship between net P mineralization and phosphatase activity was observed by Traser Cepeda *et al* (1991) who reported that the increase in microbial biomass after liming might have obscured the decrease in phosphatase activity due to high available P derived from fertilizers.

But at the panicle initiation stage, the response to fertilizers, lime and manures and their interaction effects except F × M interaction, were found to be significant (Tables 45 to 47; Fig. 32). The highest activity noticed for $F_1L_1M_2$ (130.83) with the application of fertilizers, lime and manures (vermicompost) was found to be superior compared to other treatments. This might be due to the P contributed by the chiefly organic manures (vermicompost) which would have stimulated the microflora producing hydrolytic enzymes such as

phosphatase to release inorganic P for microbial growth. There is a possibility that the observed high levels of phosphatase activity in the rhizosphere is not due to plant enzymes, but due to microbial growth promoted by plant residues (Ladd, 1985). The number of bacteria and fungi increased after passing through the earthworm gut and this vermicompost when applied to the soil increased the phosphatase activity. This is in conformity with the findings of sharpley and Syers, (1976).

Similarly, as observed in the case of urease, a decrease in the activity of phosphatase also have been observed at the harvest stage. This decrease in phosphatase activity as observed from Tables 48 to 50; Fig. 32 could be attributed to the low rhizosphere activity coupled with a general decline in the nutrient and energy source for microbial multiplication. Even at the harvest, the effect of fertilizers, lime and manures were found to influence their effects on phosphatase activity. Among the manures used for the study, vermicompost was found to be comparatively superior than the others. The increase in phosphatase activity after liming can be attributed to release of phosphates from the immobilized forms such as Al PO_4 and $FePO_4$ resulting in higher microbial activity and thus contributing to higher phosphatase activity. This is in conformity with the findings of Haynes and Swift 1988.

Thus, it is obvious that the addition of lime to acid soils had beneficial effects on microbial biomass and microbial activity which influence the phosphatase activity of the soil both directly and indirectly. These results are

indicative of the beneficial effects of liming in a soil characterized by very low metabolic activity.

5.4.3. Protease activity

A close scrutiny of the data presented in Tables 42 to 44; Fig. 33 clearly indicated highly significant effects of treatments on protease activity. The main effects of F, L and M and as well as the interaction between F × L, L × M and F × M was found to be significant with respect to the protease activity. The combined effects of fertilizers, lime and manures (vermicompost) were evident due to the highest values registered for $F_1L_1M_2$.

Application of fertilizers especially nitrogenous fertilizers stimulate the activity of microorganisms which includes the proteolytic group responsible for the synthesis of protease enzyme. A major reason for this observation is the accumulation of mineral N as the net result of the positive mineralization - immobilization turn over balance sheet which finally stimulated the synthesis of protease by the protolytic bacteria.

An increasing trend of protease towards the panicle initiation stage was attributed to a sudden spurt in the microbial population of the rhizosphere at this stage where the availability of energy and nutrient sources for growth and multiplication was assured. The individual as well the interaction effects of fertilizers, lime and manures were significant with respect to protease activity which was evident from the statistical analysis at this stage. The highest value

was recorded for treatments involving fertilizer, lime and manures (vemicompost) - $F_1L_1M_2$ (275.00). This might be due to the favourable positive effects of fertilizers, lime and manures on the microbial community whereby the synthesis of the enzymes was increased. Similar results were observed by Tateno (1988) with the application of fertilizers, lime and manures.

It is also evident that the application of lime might have improved the soil reaction which was conducive for the multiplication of microorganisms responsible for the synthesis of proteases. This is in agreement with a report by Jenkinson and Johnson (1977). Thus it is evident that the maximum activity of protease at this stage could be attributed to the residual effects of these amendments.

The drastic decline in activity followed by an increase at the panicle initiation stage can be attributed to the stage of the crop where all the metabolic processes tends to cease. At this stage, availability of moisture, nutrients, energy and substrates is limited thus limiting the proliferation of microbes. Even the poor activity noticed might be due to the plant root exudates which have contributed protease to the soil enzyme pool. At this stage, the effects of fertilizers, lime and manures were significant and the residual effects of these amendments resulted in increased activity (Fig. 33). The effect of FYM was more pronounced at this stage in contrast to the other two stages where the effect of vermicompost was significant.

The prolonged supply of N from FYM is quite obvious from the present

study which might have served as a substrate to the growing microbes resulting in the synthesis of protease intracellularly as well as extracellularly during the lysis of these cells contributing to higher values for treatments involving FYM (151.67).

5.3.4. Dehydrogenase activity

In the present study, it was observed from Tables 42 to 44; Fig. 34 that the effect of F and L was more significant than manures at the active tillering stage. The interaction effects of F × L, L × M and F × M were also found to be significant. This might be due to the prolonged mobilization time required to release nutrients like N, P and K from the organic amendments (viz., FYM, GLM and vermicompost) added.

The dehydrogenase activity increased upon incorporation of the organic amendments compared to the unamended pots. This goes in conformity with the findings of Martens *et al.* 1992. The increase in dehydrogenase activity with addition of lime could be attributed to the fast multiplication of bacteria over fungi in the composition of soil micro flora. Moreover, increased levels of soil acidity depresses fungal spore germination, hyphal growth and plant infection (Robson and Abbot, 1989). This increased levels of microflora dominated by bacteria might have contribute to the enhanced dehydrogenase activity.

Of the various treatments imposed, the treatment involving the

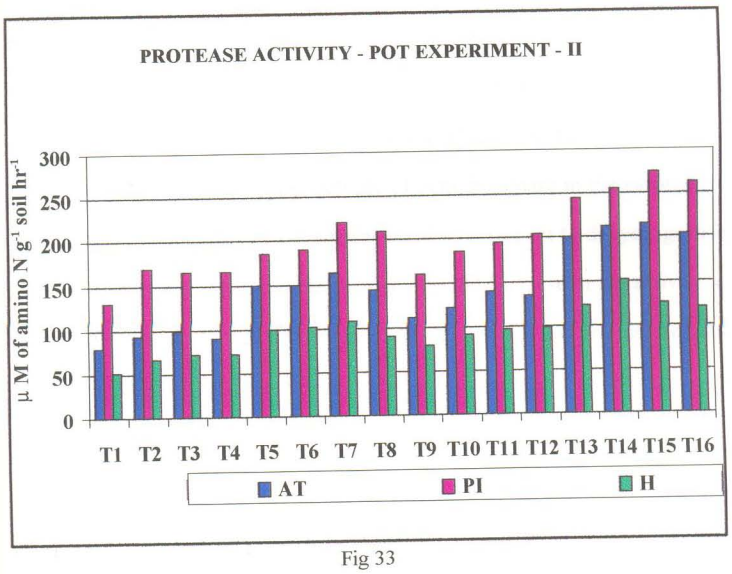
PROTEASE ACTIVITY - POT EXPERIMENT - II
μ M of amino N g-1 soil hr-1
300
250
200
150
100
50
0
T1 T2 T3 T4 T5 T6 T7 T8 T9 T10 T11 T12 T13 T14 T15 T16
AT
PI
H

Fig 33

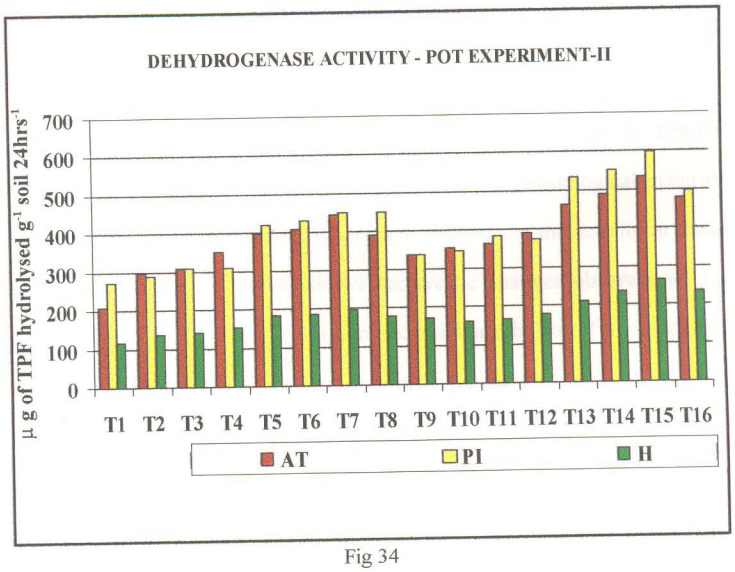
DEHYDROGENASE ACTIVITY - POT EXPERIMENT-II
μ g of TPF hydrolysed g-1 soil 24hrs-1
700
600
500
400
300
200
100
0
T1 T2 T3 T4 T5 T6 T7 T8 T9 T10 T11 T12 T13 T14 T15 T16
AT
PI
H

Fig 34

application of fertilizer lime and manures (vermicompost) recorded the highest value for dehydrogenase activity. The superiority of vermicompost over the other manures (GLM and FYM) is clearly evident from the present study. This might be due to the higher activity of dehydrogenase in the earthworm casts than the other manures. A similar trend with respect to dehydrogenase activity was observed from Table but with a slight increase in the activity at the panicle initiation stage. The main effects of F and L and the interactions F × L, L × M and F × M were found to be significant at this stage (Tables 45 to 47; Fig. 34).

This increased activity might be attributed to the enhanced microbial load, increase in the rate of microbial metabolism triggered by an ample supply of carbon, energy and nutrients through fertilization, favourable pH created by liming and the buffering action of the organic amendments added.

Following an increase in the dehydrogenase activity at the panicle initiation stage a decline in the activity towards the physiological maturity was noticed. This increase and subsequent decrease in the activity at these two critical stages of growth shows the drastic changes observed in the rhizosphere of the crops with respect to microbial metabolism. The superiority of vermicompost over other manures used in the present study is clearly evident at this stage with the highest value recorded for $F_1L_1M_2$ (264.75) (Fig. 34). In many cases, the activity of vermicompost which evoke responses similar to those of auxins or giberllins in plants even though the same did not give any

proof for the presence of such chemicals on analysis (Vaguhan *et al.* 1985). This nutrient enrichment of vermicompost may be ascribed to the digestion and mineralization of partially decomposed matter which contains higher amounts of nutrients besides the enhancement by specific enzyme activities triggered by a variety of gut microorganisms.

Thus, it is concluded that the application of fertilizers, lime and manures (vermicompost) resulted in an increased activity of microbes ultimately reflecting on the dehydrogenase activity, an index of active microbial dynamics and metabolism.

5.3.5. Cellulase activity

From the data presented in Tables 42 to 44, it is observed that the application of fertilizers, lime and manures imposed significant different with respect to cellulase activity at the active tillering stage. The main effects of F, L, M and interaction effects were all found to be significant. The highest value was noticed with the combination of fertilizers, lime and vermicompost (100.00) which revealed the beneficial effect of amending the soil with vermicompost.

The application of N, P and K fertilizers stimulated the microbial proliferation and degradation of organic matter contributing to soil, the nutrients required for microbial growth and multiplication while the application of lime reduced the acidity enhancing a favourable environment for the mass

multiplication of cellulolytic bacteria. This is in conformity with the findings of Szegi (1988) who reported that a high pH which stimulates cellulolytic microorganisms and straw decomposition in soil.

Thus the incorporation of organic residues in soil promotes microbial and soil enzyme activity. It is also reported that cellulase activity in soil depends on the level of extracellular enzyme present, the amount of active enzyme with in dead cells and associated cell fragments, and the level of activity associated with living cells (Skujins, 1976).

A similar trend of cellulase activity at panicle initiation stage, but for an increase in the activity was noticed (Fig. 35). The increase may be due to the active microbial dynamics at this stage due to the supply of nutrients through the fertilizers as well as the organic amendments added. The superiority of vermicompost over other two manures (viz., GLM and FYM) added was clearly evident from this study. An appreciable quantity of cellulase activity reported in vermicompost can be attributed to the same effect. Thus it is observed that the nutrient rich organic matter support greater microbial activity, because of greater supplies of energy and nutrients. Additionally, greater humic matter could facilitate the incorporation of soil enzymes in to the soil matrix thus allowing stabilization of higher levels of exoenzymes in soils as humic compounds are extremely important in soil enzyme complexation (Paul and Mc Laren, 1975).

As observed with the activities of urease, phosphatase, protease and

dehydrogenase, a decline in the activity of cellulase at the harvest stage was observed here also (Fig. 35). This behaviour could be due to the decreased level of energy substrate, decreased microbial population and change in the microclimate of the rhizosphere which finally resulted in a drastic decline in the synthesis of this enzyme.

Moreover at the harvest stage, the quantity of root exudates formed also decreases substantially leading to a decrease in the enzyme activity. This view is supported by the findings of Dick (1994). Even at this stage, the main and interaction effects of fertilizer, lime and manures were significant. This might be due to the treatment effect lingering in the soil in a proportionate manner though at a reduced intensity.

Soil enzymes thus show significant differences in their activity consequent to management practices thus it is inferred that management practises which minimise the addition of organic amendments to soils diminish the potential for enzymic activity. It is also evident that the earthworms play a vital role in the contribution of active microflora involved in the mineralization of plant nutrients and biomass carbon mediated through enhanced enzyme activities (Krishnamoorthy & Vajranabhaiah, 1986).

Thus it is evident from the present study that the activities of enzyme are greatly influenced by the application of organic residues especially vermicompost. This might be due to the direct contribution of enzymes by the residues or by the formation of recalcitrant organic molecules that contributes

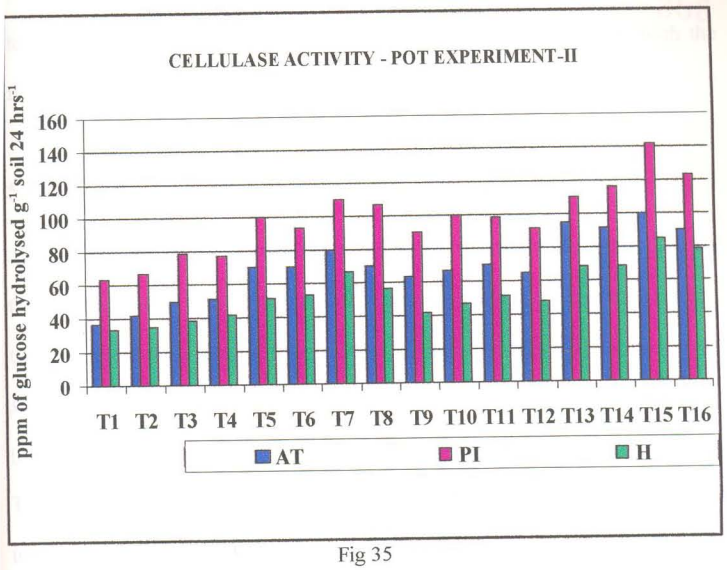
CELLULASE ACTIVITY - POT EXPERIMENT-II
ppm of glucose hydrolysed g-1 soil 24 hrs-1
160
140
120
100
80
60
40
20
0
T1 T2 T3 T4 T5 T6 T7 T8 T9 T10 T11 T12 T13 T14 T15 T16
AT
PI
H

Fig 35

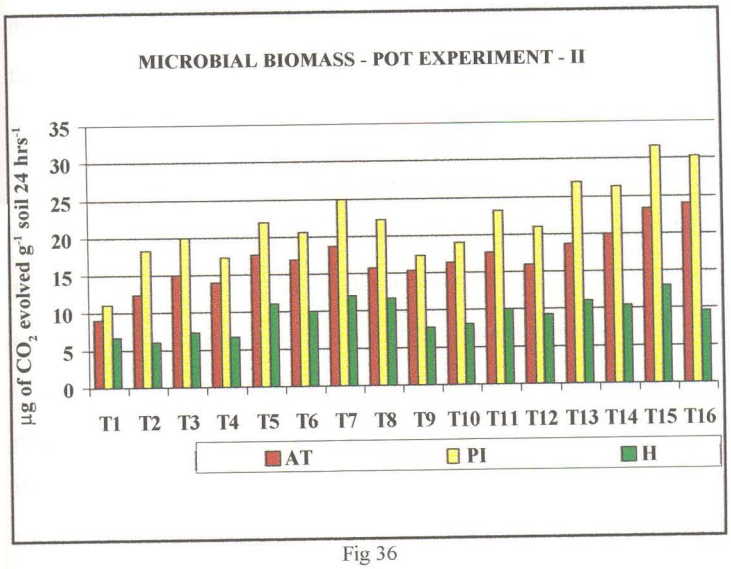
MICROBIAL BIOMASS - POT EXPERIMENT - II
µg of CO2 evolved g-1 soil 24 hrs-1
35
30
25
20
15
10
5
0
T1 T2 T3 T4 T5 T6 T7 T8 T9 T10 T11 T12 T13 T14 T15 T16
AT
PI
H

Fig 36

to the chemical stability of soil ecosystem. This is in conformity with the findings of Speir, (1977).

5.3.6. Microbial biomass

A close scrutiny of the data presented in Tables 42 to 44; Fig. 36 shows that the fertilizers, lime and manures imposed significant effect at the active tillering stage with respect to microbial biomass. This might be due to the stimulation of microbial activity resulting in a high microbial respiration (q CO_2). This is in conformity with the findings of Fauci and Dick (1994). The values for the microbial load at the active tillering stage was low compared to the panicle initiation stage. This might be attributed to a drastic increase in the uptake of plant nutrients by the crop at this stage thus substantially reducing the share of mineral nutrients available for microbial immobilization and the associated cell multiplication.

An increase in the microbial biomass as observed from Tables 45 to 47; Fig. 36 indicates the rapid proliferation and multiplication of microbial flora at the panicle initiation stage. At this stage, the effect of fertilizer, lime and manures were found to be significant. The interaction between F × L, L × M and F × M were also significant. This is attributed to the increased availability of nutrients like N, P and K whereby the carbon metabolism is increased through which the microbes scavenge for these nutrients thus reporting higher values for microbial biomass. The above mentioned effect was more

pronounced in the case of vermicompost and hence resulted in highly significant difference.

An asymptotic behaviour of the soil enzymes were reflected in relation to the microbial biomass with an increase towards the panicle initiation stage, followed by a decline at the harvest stage. At the harvest stage main effects as well as interaction effects were found to be significant (Fig 36). A decrease in the microbial load at harvest stage might be due to the exhaustion of nutrients, carbon substrates, moisture and other conditions needed for the microbial proliferation. The predominance of vermicompost in enhancing the enzyme activities and other related microbiological processes is evident from the present study. Thus it is inferred that application of organic residues in the form of vermicompost, in combination with fertilizers and lime resulted in an increased enzyme activity consequent to an increase in the microbial biomass which not only act as an agent of organic matter decomposition but also act as a source of nutrients.

5.3.7. Yield and yield attributes

Significant difference in the number of productive tillers, grain yield and straw yield was noticed (Tables 51 to 53). The higher values recorded for the combination for fertilizers, lime and manures especially vermicompost indicated a positive effect on the grain and straw yield. This might be due to the enhancement of microbial population in these treatments as a result of

application of these amendments thereby contributing to increased enzyme activity. The subsequent increase in mineralisation of organic residues might have assured an adequate supply of nutrients for the plants. Similar results with the application of manures and fertilizers were reported by Singaram and Kamalakumari (1993).

SUMMARY AND CONCLUSIONS

SUMMARY AND CONCLUSIONS

The present study was undertaken to investigate in detail the activities of five major soil enzymes viz., urease, phosphatase, protease, dehydrogenase and cellulase and the dynamics of these enzymes in permanent manurial experiments in selected cropping situations of Kerala. Two pot culture experiments were also conducted to study the effect of selected agro chemicals and soil amendments on the activities of above enzyme. The selected chemicals included insecticides, fungicides, herbicides and antibiotics. Soil amendments used were lime, chemical fertilizers and organic manures. The salient findings obtained and conclusions drawn from the studies conducted are summarized below.

- Analysis of samples collected from experimental plots of the permanent manurial trials revealed drastic differences in the activities of the soil enzymes. Enzyme activities observed were the highest in soils collected from the rubber based cropping system at Chethackal, followed by the rice based experiments at Pattambi and Kayamkulam while the lowest activity was noticed at Balaramapuram in the PME on coconut.

- Permanent manurial experiments on rice in both the locations revealed more or less comparable values for urease, phosphatase, protease, dehydrogenase and cellulase in the case of Pattambi (PME-I and PME-II) while the activities were slightly lower for urease and cellulase at Kayamkulam. On the other hand, the activity of phosphatase and dehydrogenase showed significantly superior values for Kayamkulam in comparison to PME-I and II at Pattambi.

- Experiments conducted in the uplands registered very low values for protease and dehydrogenase, moderatily high values for urease and phosphatase in the case of Balaramapuram while the values registered were the highest at Chethackal for all the five enzymes.

- In all the permanent manurial experiments activities of enzymes were found to be higher in plots receiving treatments involving combinations of organic manures and chemical fertilizers. However the highest values for cellulase and dehydrogenase was observed in the case of treatments receiving chemical fertilizers for Pattambi (I and II) and Kayamkulam respectively.

- It is evident from the study of the permanent manurial experiments, that the total quantity of organic amendments added to the soil *per se* could not sustain a higher enzyme activity but required supplemental addition of mineral nutrients through chemical fertilizers also.

- In the intercropping cum spacing trial at the CES, Chethackal highest values for all the enzymes have been recorded in T_2 involving rubber as sole crop.

- Of the physico chemical characteristics of the soils studied througpath coefficient analysis and regression analysis, organic carbon, cation exchange capacity, sesquioxide content, available N, available P, available K and respiratory activity were found to influence the enzyme activities to the maximum extent.

- Pot culture experiments to study the effect of amendments and agrochemicals revealed significant effects due to treatments. In all the cases, the maximum inhibitory effects on enzyme activities were observed consequent to the application of treatments involving antibiotics followed by fungicides, insecticides and herbicides.

- The decline in the enzymatic activity after the application of the agro chemicals was drastic during the active tillering stage compared to the control, the activity gradually resumed during the panicle initiation stage and then decreased asymptotically towards the harvest stage.

- Yield and yield attributes of the crop were not affected significantly consequent to the observed decrease in enzyme activity. However the long term effect of this practice need to be investigated in detail.

- Among the various amendments used in the present study, treatments involving the application of organic amendments as vermicompost in combination with lime and fertilizers were found to be significantly superior. All the five enzymes thus registered the highest values in this treatment irrespective of the stage of the crop.

Though the present study has been conducted over a vast stretch of land covering the major agroclimatic and soil variations, the effect of seasonal changes in the enzyme activity need further detailed investigations. This aspect is particularly important since the farming situations of Kerala accounts for two types of transformations in soils, aerobic and anaerobic where the agents of transformations, substrates, products and the relative efficiency are totally different.

Integration of soil parameters, enzyme assay and fertility rating in a single index or a few indices to develop a scale for assessing soil quality and sustainability will be a promising attempt of immense practical significance in future soil research in this context.

Soil enzyme activities have not been consistently correlated with crop yields, which is partially related to external input factors such as fertilizer and water, which can have a major impact on plant productivity but not on soil biological activity over a short span. This points to the need for conducting long

term experiments in different agroclimatic zones to develop a universal index compositing soil biological, chemical and physical parameters.

Further the study of the environmental risks of agro chemicals to soil microbial activity and related biological factors need be carried out with analyses on microbial diversity since the species specificity and relative toxicity varies considerably with the nature of the chemical used.

REFERENCES

REFERENCES

Abdel Mallek, A.Y., Mohararam, A.M., Abder Kader, M.I.A. and Omar, S.A. 1994. Effect of soil treatment with OP insecticides profenfos on fungal flora and some microbial activities. *Microbiological Research* 149 : 2, 167-171

Abdel Monem, M.H.A.A. and Aly, O.K.A. 1990. Existence of soil microflora producing amylase and protease in eastern region of Saudi Arabia. *Arab Gulf Journal of Scientific Research* 8 (3) : 121-135

Acton, D.F. 1994. A programme to assess and monitor soil quality in Canada. Soil Quality Evaluation Programme Summary report. Centre for Land and Biological Resources Research. *Agriculture* and *Agri-Food*, Canada

Adams, M. 1992. Phosphatase activity and phosphorus fractions in Karri forest soils. *Biol. Fertil. Soils* 14 : 200-204

Albanell, E., Plaixate, J. and Cabrero, T. 1988. Chemical changes during vermicomposting (*Eisenia foetida*) of sheep manure mixed with cotton industrial wastes. *Biol. Fert. Soils* 6 : 266-269

Alexander, M. 1977. Introduction of the rhizosphere. p. 423-43. In : *Introduction to Soil Microbiology*. 2nd Edn. John Wiley & Sons, New York

Alexander, M. 1980. *Microbial ecology*, Wiley, New York

Ambroz, Z. 1966. Some reasons for differences in the occurrence of proteases in soils. *Rostt. Vyroba* 12, 1203-1210

Ambroz, Z. 1970. Effect of drying wetting on enzymatic activity of soils. *Rostt. Vyroba* 16, 869-876

Ambroz, Z. 1973. Study of the cellulase complex in soil. *Rostt. Vyroba* 19, 207-212

Ampova, G. and Parishkova, A. 1983. Effects of systematic fertilizing and manuring on biological processes in soil and yields of tobacco. *Pochvoznanie -i- Agrokhiiya* 18 : 6, 69-75

Anderson, T.A., Guthrie, E.A. and Walton, B.T. 1993. Bioremeditation in the rhizosphere. *Environmental Science and Technology* 27 : 2630-3636

Andrew, A.M. 1995. Bioavailability of atrazine to soil microbes in the presence of earth worm *Lumbricus terrestrius*. *Soil Biology and Biochemistry* 28 : 555-559

Andrews, R.K., Blakley, R.L. and Ierner, B. 1984. Urea and Urease. 6 : 245-283. In. L. Eichorn and L.G. Merzilli (Eds.). *Advances in Organic Biochemistry*, Elsevier Science Publishing, New York

Anwarzay, M.O., Blum, W.E.H., Strauss, P. and Kandezer, E. 1990. Biological activity in soil in an 80 year long term field experiment. *Foerderungs dienst.* 38 : 12-22

Appiah, M.R. and Thomson, E.J. 1974. The successive cropping on soil organic phosphorus. *Ghana Fl. Agric. Sci.* 7: 25-30

Asmer, F., Eiland, F. and Nielson, N.E. 1992. Interrelationship between extra cellular enzyme activity, ATP content, total counts of bacteria and CO_2 evolution. *Soil Biology and Fertility* 14 : 288-292

Babich, H. and Stotzky, G. 1983. Developing standards for environmental toxicants : the need to consider abiotic environmental factors and microbe-mediated ecological processes. *Environ. Health Persp.* 45 : 247-260

Badalucco, L., Grego, S., Dell'oreo, S. and Nannipieri, P. 1992. Effect of liming on some chemical, biochemical and microbiological properties of acid soils under spruce (*Picea abies* L.) *Biol. Fertil. Soils* 14 : 76-83

Bagavathiammal and Muthiah, N.D. 1995. Studies on the application of potassium and organic manures on the uptake of major nutrients in red soil. *Madras Agric. J.* 82 (1) 55-57

Bajwa, M.S. and Paul, J. 1978. Effect of continuous application of N, P, K and Zn on yield and nutrient uptake of irrigated wheat and maize and on available nutrients in a tropical brown soil. *J. Indian Soc. Soil Sci.* 26 : 160-165

Balasubramaniam, A., Siddaramappa, R. and Rangaswami, G. 1972. Effect of organic manuring on the activities of enzymes hydrolyzing sucrose, urease and on soil aggregation. *Plant Soil* 327-328

Baligar, V.C., Wright, R.J. and Smedley, M.D. 1997. Enzyme activieis in Appalachian soils : Dehydrogenase. *Communication in Soil Science and Plant analysis* 22 : 1797-1804

Baligar, V.C., Wright, R.J. and Smedley, M.D. 1998. Acid phosphatase activity in soils of the Appalachian region. *Soil Science Society of America Journal* 52 : 1612-1616

Bardgett, R.D., Speir, T.W., Ross, D.J., Yeates, G.W. and Kettles, H.A. 1994. Impact of pasture contaminated by copper, chromium, and arsenic timber preservative on soil microbial properties and nematodes. *Biol. Fertil. Soils* 18 : 17-19

Basker, A., Kirkman, J.H. and Mac gregor, A.N. 1994. Changes in potassium availability and other soil properties due to soil ingestion by earthworms. *Biol. Fertil. Soils* 14 : 300-303

Beck, T. 1984. Bacterial and enzymatic bioassays for toxicity testing in the environment. *Rev. Environ. Contamin. Toxicol* 125 : 1-21

Bending, G.D. and Read, D.J. 1995. The structure and function of vegetative mycelium of ectomycorrhizal plants. *New phytologist* 130 : 3, 411-417

Benefield, C.B., Howard, P.A., Howard, D.M. 1977. The estimation of dehydrogenase activity in soil. *Soil Biology and Biochemistry* 9 : 67-70

Bennini, S., Gersa, C. and Ciurl, S. 1996. Urease from soil bacterium *Bacillus Pasteurii*. A heteropolymeric enzyme with a binuclear active site. *Soil Biol. Biochem.* 28 : 6, 811-817

Beyer, L., Wachendorf, C., Balzer, F.M. and Balzer graf. 1992. The use of biological methods to determine the microbiological activity of soils under cultivation. *Biol. Fertil. Soils* 13 : 242-247

Beyer, L., Wachendorf, C., Elsner, D.C. and Knabe, R. 1993. Suitability of dehydrogenase activity assay as on index of soil biological activity. 16 : 52-56

Biswas, T.D. 1982. *Journal of Indian Society of Soil Science* 30 : 427

Bitzer, C.C. and Sims, T.J. 1988. Estimating the availability of nitrogen in poultry manure through laboraty and field studies. *J. Environ. Qual.* 17 (1) : 47-54

Bolton, H., Elliot, L.F. Papendick, R.I. and Bezdick, D.F. 1985. Soil microbial biomass and selected soil enzyme activities : Effect of fertilization and cropping practises. *Soil Biol. Biochem.* 17 : 297-302

Bonmati, M. Ceccanti, B. and Nannipieri, P. 1991. Spatial variability of phosphatase, urease, protease, organic carbon and total nitrogen in soil. *Soil Biol. Biochem.* 23 : 391-396

Boyd, S.A. and Mortland, M.M. 1990. Enzyme interactions with clays and clay organic matter complexes. In : Soil Biochemistry (Eds.) Bollag, J.M. and Stotzky, G. Vol 6 : pp 1-28

Brady, N.C. 1996. *The Nature and Properties of Soils*. Prentice Hall of India Pvt. Ltd., New Delhi

Bramiley, R.G.V., and White, R.E. 1991. An analyses of variability in the activity of nitrifiers in a soil under pasture I. Spatially dependent variability and optimum sampling strategy. *Aust. J. of Soil Res.* 39 : 95-108

Bray, R.H. and L.t. Kurtz. 1945. Determination of total, organic and available forms of phosphorus in sols. *Soil Sci.* 59 : 39-45

Bremner, J.M. and Mulvaney, R.L. 1978. Urease activity in soils. In : *Soil Enzymes* (Ed.). Burns, R.G. Academic Press, New York, USA. p. 149-196

Briton, K. 1989. Enzyme activity of soils. *Geoderma* 12 : 43-48

Broadbent, F.E., Hill, G.N. and Tyler, K.B. 1964. Transformation and movement of urea in soils. *Soil Sci. Soc. Am. Proc.* 22, 303-307

Bromilow, R.H., Evans, A.A., Nicholls, P.H., Todd, A.D. and Birggs, G.G. 1996. The effect on soil fertility of repeated application of pesticides over 20 years. *Pesticide Science* 48, 63-72

Brookes, P.C. 1995. The use of microbial parameters in monitoring soil pollution by heavy metals. *Biol. Fertil. Soils* 19 : 269-279

Brown, J.R. 1991. Long term field research. *Agron. J.* 83 : 1-85

Brownman, M.G. and Tabatabai, M.A. 1978. Phosphodiesterase activity of soils. *Soil Sci. Soc. Am. J.* 42 : 289-290

Brownman, M.G. and Tabatabai, M.A. 1978. Phosphodiesterase activity of soils. *Soil Sci. Soc. Am. J.* 42 : 284-290

Bruckerts, S., Andreux, F., Correa, A., Ambouta, K.J.M., Sourcheir, B. 1978. Fractionnnement cles agregats applique al' analyse des complexes organo mineraux de sols burrs et de chernozems. *Plant and Soil* 57 : 274-295

Burns, R.G. 1982. Enzyme activity in soil. Location and possible role in microbial ecology. *Soil Biology and Biochemistry* 14 : 423-427

Burton, D.L. and W.B. Mc Gill. 1992. Spatial and temporal fluctuation in biomass, nitrogen and mineralizing reactions and mineral nitrogen in a soil cropped to barley. *Canadian Journal of Soil Science* 72 : 31-42

Busto, M.D. and Perezmateos, M. 1995. Extraction of humic-beta glucosidase fractions from soils. *Biology and Fertility of Soils* 20 : 1, 77-82

Cambardella, C.A., Moorman, T.B., Novak, J.M., Parkin, T.B., Karlen, D.L., Turco, R.F. and Konopoka, A.E. 1994. Field scale variability of soil properties in Central Iowa soils. *Soil Sci. Soc. Am. J.* 58 : 1501-1511

Caplan, J.A. and Fahey, J.W. 1992. Plate clearing technique to screen mixed microbial populations for protein degraders. *Soil Biology and Biochemistry* 14 : 373-375

Casida, L.E. Jr. 1977. Microbial metabolic activity in soil as measured by dehydrogenase determinations. *Applications of Environmental Microbiology* 34 : 630-636

Casida, L.E. Jr., Klein, D.A. and Santoro, T. 1964. Soil dehydrogenase activity. *Soil Science* 98 : 371-376

Caster, M.R. 1995. The influence of tillage on the proportion of organic carbon and nitrogen in the microbial biomass of medium-textured soils in a humid climate. *Biology and Fertility of Soils* 11, 135-139

Cervelli, S., Nannipieri, P. and Sequi, P. 1978. Interactions between agrochemicals and soil enzymes. In : Soil enzymes. (Ed.) Burns, R.G. pp 251-293, Academic Press, New York

Chander, K. and Brookes, P.C. 1991. Is the dehydrogenase activity invalid as a method to estimate microbial activity in Cu contaminated soils. *Biol. Fert. Soils* 32 : 112-113

Chendrayan, K., Adhya, T.K. and Sethunathan, N. 1980. Dehydrogenase and invertase activity of flooded soils. *Soil Biology and Biochemistry* 12 : 271-273

Chhonkar, P.K. and Tarafdar, J.C. 1994. Accumulation of phosphates in soils. *Journal of Indian Soc. Soil Sci.* 32 : 260-272

Christenen, L.A. and Magleby, G. 1982. Conservation tillage use. J. *Soil Water Cons.* 38 : 159-157

Chunderova, A.I. and Zubets, T., 1969. Phosphatase activity in chernopodzolic soils. *Pochvovedenie* 11 : 47-53

Ciurli, S., Marzadori,C., Benini, S., Deiana, S. and Gessa, C. 1996. Urease from the soil bacterium *Bacillus pasteurii* Immobilization on ca-polygalact uronate. *Soil Biology and Biochemistry* 28 (6) ; 811-817

Cochran, V.L., Elliot, L.F. and Lewis, C.E. 1989. Soil microbial biomass and enzyme activity in subastic agricultural and forest soils. *Biology and Fertility of soils* 7 : 283-288

Cochran, V.L., Hortan, H.A. and Cole, C.V. 1988. An estimation of microbial growth rate during wheat straw decomposition. *Soil Biology and Biochemistry* 20 : 293-298

Collins, H.P., Rasmussen, P.E. and Douglas, C.L. 1992. Crop rotation and residue management effects on soil C and microbial dynamics. *Soil Sci. Soc. Am. J.* 56 : 783-788

Cooper, J.M. and Warman, P.R. 1997. Effects of the fertility amendments on phosphatase activity, organic C and pH. *Canadian Journal of Soil Science* 77 : 281-283

Cooper, J.M. and Warman, P.R. 1997. Monitoring soil organic matter following application of fertility amendments to two soil series. Report to Canada Agreement on soil conservation. *Soil Conservation Monitoring programme*. Queen's partner, Truro, N.S. pp 73

Coorke, W.M. and Knight, A.H. 1982. *Soil Science* 93 : 365-392

Cortez, J. 1989. Effect of drying and rewetting on mineralization and distribution of bacterial constituents in soil fractions. *Biology and Fertility of Soils* 7 : 142-151

Dalal, R.C. 1975. Urease activity in some Trinidad soils. *Soil Biology and Biochemistry* 7 : 5-8

Danneberg, O.H. 1973. Uber die Extraction Von Huminstoffer Schwarzorde. *Bodenkuttur* 24 : 111-119

Danneberg, O.H. and Schaffer, K. 1974. Eine einfache Kolorimetrische se des Huminstoff systems. *Bodenkultur* 25 : 360-368

Dick, R.P. 1992. Long term effects of agricultural systems on soil biochemical and microbial parameters. *Soil Biology and Biochemistry* 40 : 25-36

Dick, R.P. 1994. Soil enzyme activities as indicators of soil quality. Defining soil quality for sustainable environment : Proceedings of Symposium, Soil Science Society of America Inc. pp 107-124

Dick, R.P. Sandor, J.A. and Eash, N.S. 1994. Soil enzyme activities after 1500years of terrace agriculture in Colca Valley. Agriculture, Ecosystems and Environment. 50 : 123-131

Dick, R.P., Rasmussen, P.E. and Kerle, E.A. 1988. Influence of long term residue management on soil enzyme activities in retention to soil chemical properties of a wheat fallow system. Biology and Fertility of soils. 66 : 159-167

Dinesh, R., Ramanathan, G. and Harjit Singh. 1995. Influence of chloride and sulphate ions on soil enzymes. *Journal of Agronomy and crop sciences* 175 (2) : 129-133

Dinesh, R., Shome, B.R. and Bandyopadhyay. 1998. Soil enzymes in the mangroves : Activities and their relation to relevant soil properties. *Current Science* 75 (5) : 510-512

Doelman, P. and Haastra, L. 1986. Short and long term effects of heavy metals on urease activity in soils. *Biology and Fertility of Soils* 2 : 213-218

Doelman, P. and Haastra, L. 1989. Short and long term effects of heavy metals on phosphatase activity in soils. *Biology and fertility of Soils* 8 : 235-242

Doelman, P. and Haastra, L. 1991. An ecological dose response model approach to short and long term effects of heavy metals on aryl sulphatase activity in soil. Biology and Fertility of Soils 11, 18-23

Doelman, P. and Haastra, L. 1996. Urease activity in chernopodzolic soils. *Pochvovedenic* 11 : 47-53

Domescui, K.H. 1970. Status and perspectives of side-effect testing, Toxicol. Environ. Chem. 11 : 147-152

Doran, J.W. 1990. Soil microbial and biochemical changes associated with reduced tillage. *Journal of the American Soil Science Society* 44 : 765-774

Doran, J.W. and Parkin, T.B. 1995. Defining and assessing soil quality. In : Defining soil quality for a sustainable Environment, (Eds.) Doran, J.W. Coleman, D.C., Berzclicek, D.F. Soil Science Society. Madison, W1, pp 3-21

Douglas, L.A. and Bremner, J.M. 1971. A rapid method of evaluating different compounds as inhibitors of urease activity in soils. *Soil Biology and Biochemistry* 3, 309-315

Dragan-Buralda, M. and S. Kiss. 1982. Soil enzymes and its significance in nutrient transformations. *Soil Biology and Biochemistry* 17 : 424 - 425

Eiland, F. 1980. The effect of manure and NPK fertilizers on the soil in a Danish long term field experiment. *Tidsskrift for Planteavl* 84 (5) : 447-454

Eivazi. P and Tabatabai. M.A. 1977. Phosphatases in soil. *Soil Biology and Biochemistry* 9 : 167-172

Endo, T., Kusaka, T., Tan, W. and Sakai, M. 1982. Effect of insecticide Cartap hydrochloride on soil enzyme activities, respiration and nitrification. *Journal of Pesticide Science* 7 : 101-110

Engasser, J.M. and Horvath, C. 1994. Inhibition of bound enzymes : I. Antienergistic interaction of chemical and diffusional inhibition. *Biochemistry* 23 : 3845-3848

Evdokimova, N.V. and Tishchenko, A.T. 1993. Enzyme activity in genetically heterogenous soils in long term fertilizer trials. Izvestiya Timirya Zeiskoi sel Skozozyai stvennoi-Akademii 1 : 103-107

Farrell, R.S., Gupta, V.V.S.R. and George, T. 1994. Effect of activity and kinetics of phosphatase in Saskwan soils. *Soil Biology and Biochemistry* 26 : 1033-1040

Fauci, F.M. and Dick, R.P. 1994. Soil Microbial dynamics : Short and long term effects of inorganic and organic N. *Soil Sci. Soc. Am. J.* 58 : 801-806

Feder, J. 1973. The phosphatases. In : Environmental Phosphorus Hand book (Ed.) Griffith, E.J., Beeton, A., Spencer, J.M. and Mitchell, D.T. 475-508. John Wiley and Sons, New York

Fenn, L.B., Tipton, J.I. and Tatum, G. 1992. Urease activity in two cultivated and non cultivated arid soils. *Biol. Fertil. soils* 26 : 151-154

Foster, R.C. and Doormar, J.F. 1991. Bacteria grazing amoeba *insitu* in the rhisosphere. *Biol. Fertil. soils* 11 : 83-87

Foster, R.C. and Doormar, J.I. 1993. Phosphatases in the rhizosphere location and sites of secretion in young roots of *Lolium perenne*. *Biol. Fertil. soil* 6 : 189-203

Fox, T.R. and Comerford, N.B. 1992. Rhizosphere phosphatase activity and phosphatase hydrolyzable organic phosphorus in two forested spodosoil. *Soil Biol. Biochemistry* 24 : 579-583

Frankenberger, W.T. Jr., and Tabatabai, M.A. 1991. L-Glutaminase activity of soils. Soil Biol. Biochem. 23 : 869-874

Frankenberger, W.T. and Dick, W.A. 1983. Relationship between enzyme activities and microbial growth and activity indices in soils. *Soil Sci. Soc. Am. J.* 42 : 945-951

Frankenberger, W.T. and Tabatabai, M.A. 1981. Enzyme activity of soils. *Biology and Fertility of soils* 11 : 6-12

Fraser, D.G., Doran, W.J., Sahs, W.W. and Leosing, G.W. 1998. Soil microbial population and activities under conventional and organic management. J. *Environ. Quality* 17 : 585-590

Furczak, J. and Gostkowska, K. 1982. Effects of long term application of Afalon in soil under potato monoculture. *Polish journal of soil science* 15 : 1, 45-52

Gadkari, D. 1984. Influence of the herbicide Goltex on extra cellular urease and phosphatase in suspended soil. *Biol. Fertil. soils* 139 : 6, 415-424

Gangnon, B., Simard, R.R., Robitaille, R., Goulit, M. and Rioux, R. 1997. Effect of compost and inorganic fertilizers on spring wheat growth and N uptake. *Canadian journal of Soil Science* 77 : 487-495

Gianfreda, L. and Bollag, J.M. 1994. Effect of soils on the behaviour of immobilized enzymes. *Soil Sci. Soc. America J.* 58 : 1672-1681

Gianfreda, L., Rao, M.A. and Violante, A. 1992. Adsorption, activity and kinetic properties of urease on montmorillinite, aluminium hydroxide and (AlOH) x-montmorillinite complexes. S*oil Biology and Biochemistry* 24 : 51-58

Gonzale Z Carcedo, S. 1978. Yestudio de un ranker de la provincia de Burgos. *Ann. Edaf. Agrobiol.* 37 : 1055-1066

Gonzales, O.J. and Zak, D.R. 1994. Geo statistical analysis of soil properties in a secondary tropical dry forest. *Plant Soil* 163 : 43-54

Gorbenko, A.Y., Panikov, N.S. and Zvyaginsten, D.V. 1986. The effect of invertebrates on the growth of soil micro organisms. *Mikrobiologia* 55 : 515-521

Gostkowska, K. and Furczak, J. 1991. Effects of long term application of Gesatop 50 in the soil under maize monoculture. *Polish Journal of soil science* 14 : 2, 151-158

Goyal, S., Mishra, M.M., Dhankar, S.S., Kapoor, K.K. and Batra, R. 1993. Microbial biomass turn over and enzyme activities following the application of FYM to field soils with and without previous long term applications. *Biology and Fertility of soils* 15 : 60-64

Greaves, M.P., Davies, H.A., Marsh, J.A.P. and Wingfield, G.I. 1981. Effect of pesticides on soil microflora using Dalapon as an example *Arch. Environ. Contam. Toxicol.* 10 : 437-499

Griffiths, B.S. 1989. Improved extraction of iodonitroformazon from soil with dimethyl formamide. *Soil Biology and Biochemistry* 21 : 179-180

Groffman, P.M. and Tiedje, J.M. 1991. Relationship between denitrification, CO2 production and air filled porosity in soils of different texture and drainage. *Soil Biology and Biochemistry* 23 : 299-302

Gupta, A.R., Narwal, R.P., Antil, R.S. and Dev, S. 1992. Sustaining soil fertility with organic C, N, P and K using FYM and fertilizer N in semi acid zone. *And Soil Res. Rabil.* 6 : 243-251

Gupta, R.P. and Dakshinamoorthy, C. 1980. Procedures for physical analysis of soil and collection of agrometeorological data, IARI, New Delhi

Gupta, V.V.S.R. and Germida, J.J. 1988. Distribution of microbial biomass and its activity in different soil aggregate single classes as affected by cultivation. *Soil Biol. Biochem.* 20 : 777-786

Haider, J., Marumoto, T. and Azad, A.K. 1991. Estimation of microbial biomass C and N in Bangladesh soils. Soil Science Plant Nutrition 37 : 591-599

Halm, B.J., Stewart, J.W.B. and Halstead, R.L. 1971. The phosphorus cycle in a native grassland ecosystem. In "Isotopes and Radiation in Soil plant relationship including forestry". Proc. Symp. pp. 571-586. IEAE, Vienna

Halstead, R.L and Mc. Kercher, F.J. 1975. Effect of long term additions of organic matter on crop yields and soil properties. *Can. J. Soil. Sci.* 48 : 341-348

Halstead, R.L. and Sowden, F.J. 1968. Effect of long term additions of organic matter on crop yields and soil properties. *Canadian Journal of Soil Science* 48 : 341-348

Harrison, A.F. 1983. Relationship between intensity of phosphatase activity and physico chemical properties in woodland soils. *Soil Biology and Biochemistry* 15 : 93-99

Hart, M.R. and Brookes, P.C. 1996. Soil microbial biomass and mineralization of soil ortganic matter after 19 years of cumulative field applications of pesticides. *Soil Biology and Biochemistry* 28 : 12, 1641-1649

Haussling, M. and Marschnner, H. 1989. Organic and inorganic soil phosphatase and acid phosphatase activity in the rhizosphere of 80 year old Norway Spruse trees. *Biol. and Fertil. Soils* 8 : 128-133

Hayano, K. 1977. Extraction and properties of phosphodiesterase from a forest soil. *Soil Bology and Biochemistry* 9 : 221-223

Hayano, K. 1993. Protease activity in paddy field soil : Orgin and some properties. *Soil Science and Plant Nutrition* 39 : 539-546

Hayano, K. and Tubaki, K. 1985. Origin and properties of B-glucosidase activity of a tomato field soil. *Soil Biology and Biochemistry* 18 : 215-219

Hayano, K. and Tubaki, K. 1995. Extraction of B-Glucosidase activity from peat soil. *Soil Biol. Biochem.* 9 : 349-351

Hayano, K. and Tubaki. 1995. Origin and properties of B-Glucosidase activity of tomato field soil. *Soil Biology and Biochemistry* 17 : 553-557

Haynes, R.J. 1986. The decomposition proess : mineralization, immobilization, humus formation and dehydration. In : Haynes, R.J. (Ed.) Mineral in the plant soil system. Academic Press, Orlands, pp 52-126

Haynes, R.J. and Swift, R.S. 1988. Effects of lime and P addition changes in enzyme activities, microbial biomass and levels of available N, S and P in an acid soil. *Biology of soils* 6 : 153-158

Haynes, R.J. and Swift, R.S. 1990. Effect of soil amendmentts and sawdust mulching on growth, yield and leaf nutrient content of high bush blue berry plants. *Scientia Hort.* 29 : 229-238

He, N., Lin, Z., and Beri, V. 1992. Effect of organic and chemical fertilizers on the grain yields and soil properties. Proc. of Int. Symp. on Nut. Mgt. Sus. Prod. : Vol. 2 130-132

Hersman, L.E. and Temple, K.L. 1979. Comparison of ATP, phosphatase, pictinolyase and respiration as indireators of soil microbial activity in reclaimed coal mine spoils. *Soil science* 127 : 70-73

Hesse, P.R. 1971. *A text book of soil chemical analysis*. John Murray, London

Hope, C.F.A. and Burns, R.G. 1989. Activity, origins and location of cellulases in silt loam soil. *Biology and Fertility of soils* 164-170

Howard, P.J.A. 1972. Problems in the estimation of biological activity in soil Oikos 23, 235-240

Ischii, T. and Hayano, K. 1974. A method for estimation of phosphodiesterase activity in soil. *Japanese Journal of Soil Sicence and Plant Nutrition* 45 : 405-508

Ishaque, M. and Kluepfel, D. 1990. Cellulase complex of a mesophilic streptomyces strain : *Canadian Journal of microbiology* 26 : 183-189

Jackson, M.L. 1973. Soil chemical analysis. Prentice Hall of India Private Ltd., New Delhi

Janzen, H.H., Campbell, C.A., Brandt, S.A., Lafond, G.P. and Towky-Smith, L. 1992. Light fractions organic matter in soils from term rotations. Soil Science Society, *Am. J.* 56 : 1799-1806

Jenkinson, D.S. 1988. Determination of microbial biomass carbon and nitrogen in soil. In : Advances in nitrogen cycling agricultural ecosystems. J.R. Wikson (ed.) CAB international, Wallingford pp 368-386

Jenkinson, D.S. and Coleman, K. 1994. Calculating the annual input of organic matter to soil from measurements of total organic carbon and radiocarbon. Europeal *Journal of Soil Science* 45, 167-174

Jenkinson, D.S. and Johnson, A.E. 1977. Soil organic matter in the Hoosfield continuous barley experiment : In : Report for 1976, Part 2. Rothamsted Experimental station, Harpenden, U.K., pp 81-101

Jenkinson, D.S. and Ladd, J.N. 1981. Microbial biomass in soil : measurement and turover. In : Paul, E.A., Ladd, J.N. (Eds.). Soil biochemistry, Vol. 5. Dekker, NY. pp 415-471

Jenkinson, D.S. and Powlson, D.S. 1976. The effect of biocidal treatments on metabolism in soil-V. A. method for measuring soil biomass- *Soil Biology and Biochemistry* 8, 209-213

Jha, K.L. 1992. Activities of enzymes in North Eastern hilly regions. *Soil Biology and Biochemistry* 34 : 115 - 117

Jones, A.L., Johnson, D.B. and Suett, D.L. 1991. Effects of soil treatments with atdicash, carbofuran and chlorfevinphos on the size and composition of microbial biomass. In : British crop protection council monograph No. 47. (Ed. A. Walker). pp 15-82. Lavenham press, Lavenham

Jordan, D., Kremer, R.J., Bergfield, W.A., Kim, K.Y. and Cacnio, V.N. 1995. Evaluation of microbial methods as potential indicators of soil quality in historical agricultural soils. *Biology and Fertility of Soils* 19 : 297-303

Jordan, D., Kremner, R.J. and Pankhurst, C.E, Doube, B.M. and Gupta, V.V.S.R. and Grace, P.R. 1994. Potential use of soil microbial activity as an indicator of soil quality. Soil biota : management in sustainable farming system. 18 : 245-249

Jorgansen, L.A. 1992. Role of Lumbricus terrestrius L. burrows on the quality of infiltrating water. *Soil Biology and Biochemistry* 24, 1555-1561

Jose, A., Andrew, M.G., Jodi, B.L. and Josef, H.G. 1997. Spatial Distribution of phosphatase activity within a reparian forest. *Soil Science* 231 : 808-825

Joseph, P.A. and R. Prasad. 1993. The effect of dicyandiamide and neem cake on the nitrification on Urea derived ammonium under field conditions. *Biology and Fertility of soils* 15 : 149-152

Juma, N.G., Tabatabai, M.A. 1977. Effect of trace elements on phosphatase activity in soils

Kalidurai, M. 1988. Studies on nitrogen fixation and nitrogen contribution by stem nodulating begume. *Sesbania rostrata* in rice soil ecosystem. M.Sc. (Ag) Thesis, TNAU, Coimbatore

Kandeler, E. 1990. Characterization of free and absorbed phosphatases in soils. *Biol. Fertil. Soils* 9 : 199-202

Kandeler, E. and Gerber, H. 1988. Short term assay of soil urease activity using colorimetric determination of ammonium. *Biol. Fertil. Soils* 6 : 68-72

Kanzawa, S. and Miyashita, K. cellulase activity in forest soils. *Soil Science and plant nutrition* 33 (3) : 399-406

Katayama, A. and Kuwatsuka, S. 1991. Effect of pesticides on cellulase degradation in soil under upland and flooded condition. *Soil Sci. Plant Nutr.* 37 : 1-6

Kennedy, A.C. and Papendick, R.I. 1995. Microbial characteristics of soil quality. *Journal of Soil Water Conservation* 50 : 243-248

Khaziyev, E. and Gulko, A.Y. 1990. Some properties of humus peroxidase complex. *Soviet Soil Science* 22 : 30-36

Khaziyev, F. and Khabirov, I.K. 1983. Physiographic factors and enzymatic activity of soils. *Soviet Soil Science* 15 : 6, 23-31

Kiss, S., Boaru, M. and Stoiata, M. 1969. Enzyme activity in vineyard soils. Review Roumaine de Biologie, Serie de Botanique 14, 127-132

Kiss, St. Stefonia and Dragan Buralda, M. 1975. Soil Enzymology in Romania. Part 2 Contrib. Bot. Univ. Babes Bolyai duj Napoca Gradina Bot., 197-207

Kiss. S., Dragan-Bularda, M. and Radulescu. D. 1975. Biological significance of enzymes accumulated in soils. *Advances in Agronomy* 27 : 25-87

Konanova, A.S. 1971. Physico-chemical properties and enzymatic activity of light grey forest soils upon cultivation. *Soviet Soil Sci.* 13 (2) : 39-44

Kozlov, K.A. and Kislitsina, V.C. 1967. Effect of ecological conditions on enzymatic activity of soils and micro organisms. *Stiinta Solution* 5, 217-226

Krishnamoorthy, R.V., Vajranabhaiah, S.N. 1986. Biological activity of earthworm casts : An assessment of plant growth promoter levels in the casts. *Proc. Indian Acad. Science. Anim. Sci.* 95 : 341-352

Kudzin, Y.K., Yaroshevich, I.V. and Gubenko. 1980. Mobilization of phosphorus reserves in chernozem during prolonged application of nitrogen and phosphorus fertilizers. Dokl. Vses. Akad. Sel 'sko Khoz. Nauk. 12 : 6-8

Kumar, S. and Kapoor, K.K. 1995. Soil microbial biomass, dehydrogenase and Urease activities and decomposition of Sesbania green manure in alkaline soils. Indian Journal of Microbiology 35 : 39-42

Ladd, J.N. 1985. Soil enzymes. In : Soil organic matter and Biological activity. Ed. D. Vanghan and E. Malcom. Matinus Nighoff Dr. W. Junk Publishers, Netherlands, pp. 175-221

Ladd, J.N. and Butler, J.H.A. 1972. Short term assays of soil proteolytic enzyme activities using proteins and dipeptide derivatives as substrates. *Soil Biology and Biochemistry* 4 : 19-30

Ladd, J.N. and Paul, E.A. 1973. Changes in enzymic activity and distribution of acid soluable, amino acid nitrogen in soil during N immobilization and mineralization. *Soil Biol. Biochem.* 5: 825-840

Ladd, J.N., Forster, R.C. and Ksjemstad, J.O. 1993. Soil structure : C and N metabolism. In : Workshop on methods of research soil structure. Ed. L. Brussard and M.J. Kooinstra, Geoderma 56 : 401-434

Lahdesmaki, P. and Piispanen, R. 1992. Soil enzymology : Role of protective colloid systems in the preservation of exo enzyme activities in soil. *Soil Biology and Biochemistry* 24 : 1173-1177

Lai, C.M. and Tabatabai, M.A. 1994. Kinetic parameter of immobilized urease. Soil *Biology and Biochemistry* 24 : 225-228

Landis, W.G. 1991. Distribution and nature of aquatic organo phosphorus acid anhydrases : enzymes for organo phosphate detoxification. *Reviews in aquatic Sciences.* 5 : 267-285

Lann, D.M. and Doran, J.W. 1984. Effect of water filled pore spare on CO2 and NO2 production in tilled and no tilled soils. *Soil Sci. Soc. Am. J.* 48 : 1267-1270

Lebedeva, L.A. and Gommonova, N.F. 1972. Effect of the long term use of mineral fertilizers and lime on the properties of sod-podzolic soil and the yield of plants. Khini. Sel., Khoz 9 : 2-8

Lee. 1993. The chemical composition of earthworm calts. *Soil Science* 58 : 367-375

Lehlinger, A.L. 1981. *Sci. Am.* 205 : 63

Lenhard, G. 1966. The dehydrogenase activity for the study of soils and river deposits. *Soil Science* 101, 400-402

Leonowiez, A. and Bollag, J.M. 1997. Laccases in soil and the feasibility of their extractions. *Soil Biology and Biochemistry* 1 : 237-242

Lethbridge, G., Bull, A.T. and Burns, R.G. 1981. Effect of pesticides on 1, 3-B-Glucanase and urease activities in soil in the presence and absence of fertilizers, lime and organic materials. *Pesticide Science* 12 : 147-155

Linkins, A.E. and Fetcher, N. 1993. Effect of surface applied prudhoe Bay crude oil on vegetaation and soil processes in tussock tendra. Proceedings of National *Academy of Sciences* 6 : 723-728

Ma Lachlan, K.D. 1994. Acid phosphatase activity of intact roots and phosphorus nutrition in plants. Australian Journal of Agricultural Research. 31 : 429-440

Mackay, A.D., Kladviko, E.J. 1985. Earthworms and the rate of breakdown of soyabean and maize residues in soil. *Soil Biology and Biochemistry* 17 : 851-857

Macura, J. and Vagernova, K. 1969. Colorimetric determination of the activity of the proteolyte enzymes in soil. Rostt. Vyroba 15, 173-180

Malcom, R.E. 1983. Assessment of phosphatase activity in soils. Soil Biology and Biochemistry

Mandal and Pani. 1965. Effect of continuous application of organic manures and ammonium sulphate in mulberry field on some soil properties. *J. Indian Soc. Soil Sci.* 13 : 37

Margesin, R. and Schinner, F. 1994. Phospho monoesterase, phosphodiesterase, phosphotriesterase and inorganic pyrophosphate activies in forest soils in an alphine area. Effect of pH on enzyme activity and extractability. *Biol. Fertil. Soils* 18 : 320-326

Maris kevich, D.G. 1992. Enzyme activity of soils in conditions of a pasture load. *Soviet Journal of Ecology* 23 : 5, 294-298

Markus, L. 1955. Determination of carbohydrates from plant materials with anthrone reagent II. Assay of cellulase activity in soil and farm yard manure. *Agrokem.* Talajt 4, 207-216

Martens, D.A. and Bremner, J.M. 1994. Effects of pre emergence and post emergence herbicides on urea hydrolysis and nitrification of urea nitrogen in soil. B*iology and fertility of soils* 17 : 309-313

Martens, D.A., Johnson, J.B. and Frankenberger, W.T. 1992. Production and persistence of soil enzymes with repeated additions of organic residues. *Soil Science* 153 : 53-61

Martin, A. and Marinissen, J.C.Y. 1993. Biological and Physico-Chemical processes in excrements of soil animals. *Geoderma* 56 : 331-347

Martin, J.K. and Haider, K. 1971. Microbial activity in relation to humus formation. *Soil Science* 111 : 54-63

Martin, K.K. 1977. Factors influencing the lossof organic arbon from wheat roots. *Soil Biology and Biochemistry* 9 : 1-7

Marumoto, T. 1984. Mineralization of C and N from microbial biomass in paddy soil. Plant and soil. 76 : 165-173

Mathan, K.K., Bankaran, K., Kanaka bushini, N. and Krishnamoorthy, K.K. 1978. Available N status of black soils under the influence of continuous cropping. *J. Indian Soc. Soil Sci.* 26 : 166-168

Maurya, R.R. and Ghosh, A.B. 1972. Effect of long term manuring and rotational cropping on fertility status of alluvial calearious soil. *J. Indian Soc. Soil Sci.* 20 : 31-43

Mayoudan, J., Balistic, L. and Sarkar, J.M. 1975. Proprietes des activities protelytiquies extraites des sols frais. *Soil Biology and Biochemistry* 7 : 281-286

Mc claugherty, C.A. and Linkins, A.E. 1988. Extractability of cellulase in forest litter and soil. *Biology and Fertility of soils* 6 : 322-325

Mc Gill, W.B., Cannon, K.R., Robertson, J.A., and Cook, F.D. 1986. Dynamics of soil microbial biomass and water soluble organic in Brection L after 50 years of cropping to two rotations. *Canadian journal of Soil Science* 66 : 1-19

Mc Lean, A.A. and Doyle, J.J. 1963. The evaluation of the residual effect of fertilizers in long term fertility plots 1. Potassium. *Canadian Journal of Soil Science* 43 : 229-234

Mc Lean, E.O., Eckert, D.J., Reddy, G.Y. and Triewicler, J.f. 1978. An imrpoved SMP soil lime requirement method incorporating double buffer and quick test fetures. *Soil Science Scoeity of America Journal* 42 : 311-316

Mc. Laren. 1985. Soil as system of humus and clay immobilized enzymes. *Chemical Science* 8 : 97-99

Medina, R. and Muller, K. 1990. Molecular modelling studies on Urease active site and the enzyme catalyzed Urease hydrolysis. Journal of computer aided Molecular Design, 4 : 355-367

Miller and Dick. 1995. Dynamics of soil C and microbial biomass on whole soil and aggregates in two cropping systems different C inputs. *Applied Soil Biology*

Mishra, C., Deshpande, V. and Rao, M. 1993. Immobilization of *Penicillium funiculosm* on a soluble polymer. *Enzyme Microbiology Technology* 5 : 342-345

Mitterer, M., Bayer, H. and Schinner, F. 1981. The influence of fungicides on microbial activity of soil. Zentral Pflanzeenern Bodenkundes 144 : 463-471

Mobley, H.L.T. and Hausinger, R.P. 1989. Microbial ureases : Significance, regulation and molecular characterization. *Microbiology Reviews* 53 : 85-108

Monreal, C.M., Dinel, H., Schritzer, M., Ganible, D.S. and Bieder beck, V.O. 1998. Importance of C Sequestration on functional indicators of soil quality as influenced by management in sustainable agriculture. *Soil Biology and Biochemistry* 26 : 1033-1040

Mortland, M.M. 1970. Clay organic complexes and interactions. *Advances in Agronomy* 22 : 75-177

Mulvaney, R.A. and Bremner, J.M. 1979. A modified diacetyll monoxime method for colonimetric determination of urea in soil extracts. *Commun. Soil. Sci. Plant Analy.* 10 : 1163-1170

Muthuvel, P., Kandaswamy, P. and Krishnamoorthy, K.K. 1977. Availability of NPK under long term fertilization. *Madras Agric. J.* 64 (6) : 358-362

Nakamura, T., Mochida, K., Ozoe, Y., Ykawa, S., Sakai, M. and Mitsugi, S. 1990. Enzymological properties of three soil hydrolases and effects of pesticides on their activites. *Journal of pesticide science* 15 (4) : 593-598

Nanni pieri, P., Cecanti, B and Grego, S. 1990. Ecological significance of the biological activity on soil. In : Soil Biochemistry Vol. 6. Eds. J.M. Bolag and G. Stotzky. pp 293-355. Marcel Dekker, New York

Nannipieri, P. 1994. Enzyme activity. In : The Encyclopedia of Science and Technology. Ed. C.W. Finkl. Van Nosterand Reihold Company, New York

Nannipieri, P. 1997. The potential use of soil enzymes as indicator of productivity, sustainability and pollution. *Soil Biota.* 28 : 238-244

Nannipieri, P. and Bollag, J.M. 1991. Use of enzymes to detoxify pesticide contaminated soils and water. *Journal of environmental quality.* 20 : 510-517

Nannipieri, P. and Bollag, J.M. 1997. Use of enzymes to detoxify pesticide contaminated soils and waters. *Journal of environmental quality* 20 : 510-517

Nannipieri, P., Ceccanti, B. and Bianchi, D. 1988. Characterization of humus-phosphotase complexes extracted from soil. *Soil Biology and Biochemistry* 20 : 683 -691

Nannipieri, P., Ceccanti, B., Cervelli, S. and Sesqui, P. 1978. Stability and kinetic properties of humus-urease complexes. *Soil Biol. Biochem.* 10 : 143-147

Nannipieri, P., Cervelli, S. and Pedrazzini, F. 1976. Concerning the extraction of enzymatically active organic matter from soil. *Experiantia* 31, 513-515

Nannipieri, P., Grego, S. and Ceccanli, B. 1990. Ecological significance of biological activity in soil. In : Soil Biochemistry (Eds.) Bollag, J.M. and Stotzky, G. Vol. 6. Marcel Dekker, New York, pp 293-355

Nannipieri, P., Johnson, R.L., Paul, E.A.B. 1978. Criteria for microbial growth and activity in soil. *Soil Biol. Biochem.* 10 : 223-228

Nannipieri, P., L. Muccini and C. Ciardi. 1983. Microbial biomass and enzyme actiities : Production and persistence *Soil Biol. Biochem.* 10 : 223-229

Nannipieri. 1984. Microbial biomass and activity measurements in soils : Ecological significance. In : Current perspectives in microbial ecology. Ed. Klug, M.J. and Reddy, A. American Society of Microbiology, Washington DC, pp 512-521

Narain Rai. 1992. Effects of long term 2,4-D application on microbial populations and biochemical processes in a cultivated soil. *Biology and Fertility of soils.* 13 : 187-191

Nohrstedt, H.O. 1995. Biological activity in soil from forest stands in central Sweden. 11 : 259-266

Nowak, A. and Michaluwicz. 1988. Effect of Lerbicide Patoran 50 WP on amlylolytic, proteolytic and lipolytic activity of soil actinomycetes. Z. Pflkrankh Pflschutz, Sonderheft. XI : 295-300

O'Toole, P. 1991. Assay of enzyme activity in an ammonium fixing soil. Commun. Soil Sci. Plant Anal. 22 (3 & 4) : 213-224

Ohlinger, R. 1986. Effect of simulated acid rain on soil and young Norway Spruce plants in a pot experiment. *Canadian journal of Soil Science.* 66 : 303-313

Olson, B.M. and Lindwall, C.W. 1991. Soil microbial activity under chemical follow conditions : Effects of 2,4-D and glyphosate. *Soil Biology and Biochemistry.* 23 : 1071-1075

Palaniswami, C. and Ramulu, V.S.S. 1995. Effect of continuous flow of effluent in a channel on the available nutrient status and on certain soil enzymes activities of the adjoining fields. *Journal of the India Society of Soil Science.* 42 : 3, 468-470

Pancholy, S.K. and Rice, E.L. 1972. Effect of storage conditions on activities of urease, invertase, amylase and dehydrogenase in soil. Soil Society of America. Proc. 36, 536-537

Pancholy, S.K. and Rice, E.L. 1973. Soil enzymes in relation to old field succession : analyse, cellulase, invertase, dehydrogenase and urease. *Soil Sciece Soc. Am. Proceedings* 37 : 47-50

Panse, V.G. and Sukhatme, P.V. 1967. *Statistical Methods ofor Agricultural Workers.* ICAR Pub. House, New Delhi

Pantosh Derimova, T.D. 1983. Enzymatic activity of soils in certain Hungarian forest biogeo cenoses. *Biology Bulletin* 10(3) : 203-207

Parent, L.F. and A.F. Mackenzie. 1985. Rate of pyrophosphate hydrolysis in organic soils. *Canadian Journal of Soil Science* 65 : 497-506

Park, J. and Seaton, R.A.F. 1996. Integrative research and sustainable agriculture. *Agriculture Systems.* 50 : 81-100

Parkin, T.B. 1993. Spatial variability of microbial processess in soil. *A review journal of Environmental Quality* 22 : 409-417

Parks, F.P. 1974. The sorption of *Ezcherichia coli* alkaline phosphatase by selected clay minerals. Ph.D. Thesis, University of Idaho

Patel, S.P., Ghosh, A.P. and Snes, S. 1963. *Journal of Indian Society of Soil Science* 11 : 225

Paul, E.A. and Mc. Laren, A.D. 1975. Biochemistry of the soil subsystem. In : E.A. Paul and A.D. Mc Laren (Editors) Methods of Soil analysis Part 2. 2nd edn., Agron. Manogr. 9 ASA and SSSA, Madison, W.I. pp 403-430

Pedersen, J.P. and Hendriksen, N.B. 1993. Effect of passage through the intestinal tract of detrivore earthworms (Lumbricus sp.) on the number of selected gram negative and total bacteria. *Biology and ertility of soils* 16 : 227-232

Pedrazzini, F.R. and Mc. kee, K.L. 1984. Effect of flooding on activities of soil dehydrogenase and alcohol dehydrogenase in rice. *Soil Sci. Plant Nutr.* 30 : 359-365

Pennock, D.J, Anderson, D.W. and Jong, E. 1994. Landscape scale changes in indicators of soil quality due to cultivation in Saskatchewan Geoderma. 64 : 1-19

Perez Mateos, M. and Gonzales Cercedo, S. 1985. Effect of fractionation on location enzyme activities in soil structural units. *Biology and Fertility of Soils* 1 : 153-159

Perucci, P. 1992. Enzyme activity and microbial biomass in a field soil amended with municipal refuse. *Biology and Fertility of soils* 14 : 14-60

Perucci, P. and Guisquiani, P.L. 1990. Influence of municipal waste compost addition on chemical properties and soil phosphatase activity. *Zentral bi Microbiol.* 145 : 615-620

Perucci, P. and Searponni, L. 1983. Effect of crop residue addition on arylsulphatase activity in soils. *Plant and Soil* 73 : 323-326

Peshakov, G., Barov, V., Rankov, V., Ampova, G., Toskov, N., Tzirkov, T. 1986. An attempt at optimizing fertilization with a view to maximum capacity of soil enzymes. *Soil Biology and Conservation of the biosphere* 1 : 37-44

Pickett, S.T.A. and M.L. Cadenasso. 1995. Landscape Ecology : Spatial heterogenity in ecological systems. *Science* 269 : 331-334

Pier Lodovico, G. 1994. Long term effects of heavy metals from composted municipal waste on some enzyme activities in a cultivated soil. *Biology and Fertility of soils* 17 : 257-262

Polland, E. and Riha, S.J. 1990. Surface soil acidification under red pine Norway sprice. *Soil Science Society of America Journal* 54 : 1124-1130

Pototskaya, L.A. and Tyul menkov, E.L. 1980. The enzyme actiity of soil and potato producitivity in relation to herbicide application. Fiziologo biokhimic cheskie osnovy povsheniya produktiv rastenic. 21 : 144-153

Powlson, D.S., Brookes, P.C., Christenen, B.T. 1987. Measurement of soil microbial biomass provided an early indication of changes in total soil organic matter due to straw incorporation. *Soil Biology and Biochemistry* 19 : 159-164

Prasad, C.R., Shamiuddin, Y., Singh, B.N., Jha, K.K. and Mandal, S.C. 1971. *Proc. Int. Sym. Soil Ferti. Evaln.* New Delhi 1 : 866

Quastel, J.H. 1946. Soil metabolism. The Royal Inst. of Chem. of Great Britain and Ireland, London

Quilt, D.., Gross bard, E and Wright, S.J.L. 1979. Effects of herbicide Barban and its commercial formulation carbyne on soil micro-organisms. *J. Appl. Bacteriol.* 46 : 431-442

Quiquampoix, H., Stanton, S., Baron, S and Ratcliffe, R.G. 1993. Interpretation of the pH dependence of protein adsorption on clay mineral surface. *Physico chem. Eng. Asp.* 75 : 85-93

Raheja, P.C. 1966. Phosphatyase in crop production (In) Soil productivity and crop growth. Asia Publishing House, 152-204

Rai, R.K., Sinha, M.N. and Mahatim Singh. 1982. Studies on direct and residual effect of phosphorus on grwoth and yield of maize and wheat in sesquence. *Indian Journal of Agronomy.* 27 : 354-362

Rajendran, P. 1992. Electrochemical properties of selected oxisols and ultisols of Kerala with special reference to charge characteristics and surface minerology. Ph.D Thesis, KAU

Ramirez-Martinez, J.R. and Mc. Laren, A.D. 1966. Determination of phosphetase activity by a flourimeter technique. Enzyumologia 31 : 23-28

Rao, D.L.N. and Ghai, S.K. 1989. Urease and dehydrogenase activity of alkali and reclaimed soils. Australian *J. Soil Research* 23 : 661-665

Rao, L.N. and Burns, R.G. 1990. The effect of surface growth of blue green algae and bryophytes on some microbiological, triochemical and physical soil properties. *Biology and Fertility of soils* 9 : 239-244

Ray Chaudhury, S.P. 1977. Intergrated nutrient supply for increased soil productivity. *J. Indian Soc. Soil Sci.* 25 : 444-456

Reddy, B.B., Reddy, S.N., Reddy, V.M., Reddy, M.R., Kumar, A. and Swamy, K.B. 1987. Effect of plant population on the performace of maize hybrids at different fertility levels in a semi arid environment. *Indian J. Agric. Sci.* 57 : 705-709

Reddy, G.B., Faza, A. and Bennett, R. 1987. Activity of enzymes rhirosphere and non-rhirosphere soil amended with sludge. *Soil Biology and Biochemistry* 19 : 203-205

Reicosky, D.C., Kemper, W.D., Langdale, G.W., Douglas, C.L. and Rasmussen, P.E. 1995. Soil organic matter changes resulting from tillage and biomass production. *Journal of soil and water conservation* 50 : 254-261

Rhoades, J.D. 1982. Soluble slots. In : Method of soil analyses Ed. Page, A. Land Miller, R.A. Agronomy I, Americal Society of Agronomy Madison, Wisconsen. pp 167-179

Robson, A.D. and Abbot, L.K. 1989. The effect of soil activity on microbial activity in soil. In : Robson, A.D. (Ed.) Soil acidity and plant growth. Academic Press, London, pp-139-165

Rogar, L., Bernard, G. and Regis, R. 1998. Microbial biomass C and alkaline phosphatase activity in two compost amended soils. *Canadian Journal of Soil Science* 54 : 581-586

Roper, M.M., Turpin, J.E. and Thompson, J.P. 1994. Nitrogenase activity by free living bacteria in soil in a long term tillage and stubble management experiment in a vertisol. *Soil Biology and Biochemistry* 26 : 1087-1091

Ross, D.J. 1970. Effects of storage of dehydrogenase activities of soils. *Soil Biology and Biochemistry* 2 : 55-61

Ross, D.J. and Cairns, A. 1992. Effects of earthworms and ryegrass on respiratory and enzyme activies of soil. *Soil biology and biochemistry* 14 : 583-587

Ross, D.J., Speir, T.W., Cowling, J.C. and Whale, K.N. 1984. Influence on field applications of oxyml and Fenamiphos on biochemical activities of soil under pasture. *N.Z. J. Sci.* 27 : 247

Ross, D.J., T.W. Speir, H.A. Kettles and A.D. Mackay. 1995. Soil microbial biomass, C and N mineralization and enzyme activities in a hill pasture. Influence of season and slow release P and S fertilizer. *Soil Biology and Biochemistry* 27 : 1431-1443

Rossi, D.E., Mulla, D.J., Journel, A.G. and Franz, E.H. 1992. Geostatistical tools for modeling and interpreting ecological spatial dependence. *Ecology Monograph* 62 : 277-314

Rowell, M.J., Ladd, J.N. and Paul, E.A. 1973. Enzymatically active complexes of proteases and humic acid analogues. *Soil Biol. Biochem.* 5 : 699-703

Russel, E.W. 1973. Soil Conditions and Plant Growth Tenth Edition. ETBS and Longman, U.K

Ryu, D.D.Y., Kini, C. and Mandels, M. 1994. Competitive adsorption of Cellulase components and its significance in a synergative mechanism. *Biotechnology and Bioenergy* 26 : 488-496

Sahu, B.N. and Nayak, B.C. 1971. Proc. Int. Symp. *Soil Fert. Evaln.* 1 : 878

Sakamoto, K. and Oba, Y. 1991. Relationship between the amount of organic material applied and soil biomass content. *Soil Science Plant Nutrition* 37 : 387-397

Sandor, J. 1987. Intial investigations of soils in agricultural terraces in the colca valley., Peru In : W.M. Denevan, K. Mathewson and G. Knapp (Eds.) Pre-Hispanic Agricultural Fields in the Andean Region. British Archaeological Reports International Socies. 359 (1), Oxford, U.K. 163-192

Sandor, J.A. and Eash, N.S. 1991. Significance of ancient agricultural soil for long term agreonomic studies and sustainable agriculture research. *Agronomy Journal* 83 : 29-37

Sanyasi Raju, M.S. 1952. The role of organic manures and inorganic fertilizers in soil fertility. *Madras Agric. J.* 39 : 130-147

Sarkar, J.M., Leonowitz, A. and Bollag, J.M. 1989. Immobilization of enzymes on clays and soils. *Soil Bio. Bio. Chem.* 21 : 223-230

Satchell, J.E. and Martin, K. 1994. Phosphatase activity in earthworm faces. *Soil Biology and Biochemistry.* 16 : 191-194

Satchell, J.E., Martin, K. and Krishnamoorthy, R.V. 1986. Stimulation of Microbial phosphatase production by earthworm activity. 16 : 2, 195

Sato, F. and Omura, H. 1989. Soil enzyme activity in Andisol paddy fields. Relationship between soil enzyme activity and microbial count. *Japanese Journal of Soil Science and Plant Nutrition* 60 : 37-41

Sato, K. 1981. Relations between soil microflora and CO_2 evolution upon decomposition of cellulase. *Plant and Soils* 61 : 251-254

Schaffer, A. 1993. Pesticide effects on enzyme activities in the soil eco systems. In : Soil Biochemistry, Vol. 9. Ed . Bollay, J.M. and Stotzky, G. pp 273-340, Marcel Dekker, New York

Schinner, F. 1993. Litter decomposition CO_2 release and enzyme activities in a snowbed and on a wide swept ridge in an alphine environment. *Oceologia* 59 : 288-291

Schinner, F. 1993. Litter decomposition; CO_2 release and enzyme activities in a snow bed and on a wind swept ridge in an alphine environment. *Decologia* 59 : 288-291

Schinner, F., Ohlinger, R., Kandeler, E and Margesin, R. 1993. Bodenbiologische Arbeitsmethoden. 2, Auflage. Springer Verlag, Berlin

Schuster, E. and Schroder, D. 1990. Side effects of sequentially and simultaneously applied pesticides on non target soil micro organisms. *Soil Biology and Biochemistry* 22 : 375-383

Selvi, S. and Ramaswami, P.P. 1995. Residual effect of integrated nutrient management in rice-rice pulse cropping sequences. *Madras Agric. J.* 82 (1) : 24-26

Sequi, P., Cerciganni, G., Nobili, M. and Paglai, M. 1995. A positive trend among two soil enzyme activities and a range of soil porosity under zero and conventional tillage. *Soil Biol. Biochem.* 17 : 2, 255-256

Serban, A and Nissembaum, A. 1996. Humic acid association with peroxidase and catalase. *Soil Biology and Biochemistry* 1 : 41-44

Serra-Wittling, C., Honot, S. and Barriuso, E. 1995. Soil enzymatic response to addition of municipal solid-waste compost. Biology and Fertility of soils 20 : 226-236

Sevinnen, J., Van Veen, J.A. and Merckx, R. 1995. Carbon fluxes in the rhirosphere of winter wheat and spring barley with conventional Vs integrated farming. *Soil Biology and Biochemistry* 27 : 811-820

Sharpley, A.A. and Syess, J.K. 1987. Seasonal variation in casting activity and in the amounts and release to solution of phosphorus forms in earthworms casts. *Soil Biol. Biochemistry* 9: 227-231

Sharpley, A.N. 1985. Phosphorus cycling in unfertilized and fertilized agricultural soils. *Soil Science Society of America Journal* 49 : 4, 905-911

Sharpley, A.N. and Syers, J.K. 1976. Potential role of earthworm cast for the phosphorus enrichment of run off waters. *Soil Biology and Biochemistry* 8 : 341-346

Shinde, D.A. and A.B. Ghosh. 1971. Proc. Int. Symp. Soil Ferti. Evaln. New Delhi, 1 : 905

Sikora, L.J., Kawfman, D.D. and Hrong, L.C. 1990. Enzyme activities in soil showing enhanced degradation of OP insecticies. *Biology and Fertility of soils* 9 : 14-18

Silva, C.G. and Perera, A.M.A. 1971. A study of urease activity in the rubber soils of ceylon. *Quast, J. Rubber Res. Inst : Ceylon* 47, 30-36

Singaram, P. and Kamalakumari, K. 1995. Long term effect of FYM and fertilizers on enzyme dynamics of soi. *Journal of Indian Society of Soil Science*. 43 : 3, 378-381

Singh, H. and Singh, K.P. 1993. Effect of residue placement and chemical fertilizer on soil microbial biomass under tropical dryland cultivation. *Biology and Fertility of soils* 16 : 275-281

Sinsabaugh, R.L., Antibus, R.K., Linkins, A.E., Mc Claugnerly, C.A., Rayburn, L and Weiland, T. 1992. Wood decomposition over a first order water shed : mass loss as a function of lignocellulase activity. *Soil Biology and Biochemistry* 24 : 743-749

Skogland, T., Lomeland, S. and Goksoyr, J. 1998. Respiratory activity during freezing and thowing of soil : Experiments with soil bacteria. *Soil Biology and Biochemistry* 20 : 851-856

Skujins, J. 1978. History of a biontic soil enzyme research. In : Soil Enzymes ed. R.G. Burns. pp 1-49. Academic Press, New York

Skujins, J.J. 1976. Extracellular enzymes in soil CRC critical Reviews in Microbiology. pp 383-421

Skujins, J.J. and Mc Laren, A.D. 1969. Assay of urease activity using ^{14}C-urea in stored, geologically preserved and in irradiated soils. *Soil Biology and Biochemistry* 1 : 88-89

Skujins, J.J., Braal, L and Mc Laren, A.D. 1962. Characterization of phosphatase in a terrestrial soil sterilized with an electron beam. *Enzymologia* 25 : 125-133

Skujins, J.S. 1976. Extra cellular enzymes in soil. CRC Critical Review of Microbiology. 4 : 383-427

Slott, D.E. Martin, J.P., Fochdt., D.D., Haider, K. 1983. Microbial biomass act as a source of plant nutrient in dry biopical forests of savanna. Nature (London) 399 : 499-500

Smith, A.E., Aubin, A.J. and Biederbeck, V.O. 1989. Effects of long term application of 2,4-D and MPCA on soil residues and their break down. *Journal of Environmental quality* 18 : 299-302

Song, Z., Chen, X.Z. and Huang, T.Z. 1994. Soil enzyme activities and chemical properties in the soil infected by Tylenchulus semipenetrans. *Journal of Fujean College of Forestry* 14 : 81-85

Songyin, G., Guigin, S., Zhaopeng, M., Zaohua, Y. and Jiukang, M. 1994. Enzyme actiities in main soils in China. *Acta-Pedologica-Sinica* 41 : 4, 368-381

Sparrow, S.D. and Cochran, V.L. 1998. Carbon and nitrogen mineralization of subaritice agricultural and forest soils. *Biology and Fertility of Soils* 6 : 33-38

Speir, T.W. 1977. Studies on cliniosequence of soils in tussock grasslands. 11 Urease, phosphatase and sulfatase activities of topsoils and their relationship with other properties including plant available S. *Newzealand Journal of Science* 20 : 159-166

Speir, T.W. and J.C. Cowling. 1991. Phosphatase activities of pasture plants and soils. *Biol. Fertil. Soils* 12 : 189-194

Speir, T.W. and Ross, D.J. 1976. Studies on a dimosequence of soils in tussock grass land. Influence of age of chinochloa regida leaves on enzyme activities. *Newzealand Journal of Science* 19 : 159-166

Speir, T.W. and Ross, D.J. 1978. Soil phosphatase and sulphatase. p. 197-250. In. R.G. Burns (Eds.). *Soil Enzymes*, Acad. Press, New York

Spot helfer-Magana, J., Thalmam, A., Schweikle, V. 1993. Methode Zur Bestimmung der Dehydrogenase aktivitat von Boden Unter Einsatz Von Idonitro tetra zolium chlorid (INT)

Snedecor, G.W. and W.G. Cochran. 1973. *Statistical methods*. Ames. low. Iowa state Univ. Press

Sreelatha, A.K. 1993. Potassium dynamics in Neyyattinkara soil series under coconut cultivation. M.Sc. (Ag.) Thesis. KAU

Stehouwer, R.C., Dick, W.A. and Traina, S.J. 1994. Sorption and retention of herbicides in vertically oriented earthworm and artificial burrows. *Journal of Environemntal quality* 23 : 286-292

Stevenson, F.J. 1982. Humus Chemistry. Witey, New York, pp 55-92

Stot, D.E., Martin, J.P., Focht, D.D. and Haider, K. 1983. Edaphic aspects and the disposal of unused pesticides. Pesticde water and pesticides containers. *Journal of Environmental quality* 1 : 54-62

Subbiah, B.V. and G.S. Asija. 1956. A rapid procedure for the estimation of available nitrogen in soils. *Current Science*, 37 : 328-332

Subbiah, K., Sundarajan, S. and Muthuswami, S. 1983. Effect of varying levels of organic and inorganic fertilizers on the yield and nutrient uptake in India. *South Indian Horticulture* 31 (b) : 287-290

Szegi, J. 1988. Cellulase decomposition and soil fertility. Akademiai Kiado, Budapest.

Tabatabai, A. and Fu, M. 1992. Extraction of enzymes from soils. In : Soil Biochemistry Eds. G. Stotzky and J.M. Bollay. Vol 7, Marcel Pekker, New York. pp 197-227

Tabatabai, M.A. 1982. Soil enzymes. In : Methods of Analysis Ed : A.L. Pade *et al.*, Agronomy. 9 : 903-947

Tabatabai, M.A. 1992. The nature of the catalyst causing the hydrolysis of urea in soil. *Soil Science* 256 : 119-134

Tabatabai, M.A. and Bremner, J.M. 1972. Assay of Urease activity in soils. *Soil Biology and Biochemistry* 4 : 479-487

Tarafdar, J.C. and Junk, A. 1988. Influence of humic substances on growth and physiological processes. *Biol. Fertil. soils* 3 : 199-204

Tate, K. 1987. Effects of toluene on Eischerichia coli. *Journal of Bacteriology* 90 : 1420-1425

Tate, R.L. 1984. soil organic Matter, Biological and Ecological Effects. John Wiley, New York

Tateno, M. 1988. Limitations of available substrates for the expression of cellulase and protease activities in soil. *Soil Biology and Biochemistry* 20 : 117-18

Tateno, M. 1998. Limitations of available substarates for the expression of cellulase and protease activities in soil. *Soil Biology and Biochemistry* 40 : 117-118

Tha, D.K., Sharma, G.D. and Mishra, R.R. 1992. Soil microbial population numbers and enzyme activities in relation to attribute and forest degradation. *Soil Biol. Biochem* 24 : 761-767

Thalman, A. 1968. Zur Methodik der Beswtimung der Dehydrogenase aktivitat im Boden mittels Triphenyl Tetra Zoluim chlorid. Landwirtsch Forsch. 21 : 249-258

Thente, B. 1970. Effects of toluene and high energy radiation on urease activity in soil Lant br. Hegsk. Anntr. 36, 401-418

Thister, J.D., Shippen, R.S. and Calpan, J. 1980. A new method for measuring cellulose and starch degradation in soils. *Bull. Environ. Contam. Toxicol.* 24 : 570-574

Tisdale, S.L., Nelson, W.L. and Beaton, J.D. 1990. Soil fertility and fertilizer. IVth Edn. Maxwell Macmillon International Editors

Tisdale, S.L., Nelson, W.L., Beaton, J.D. and Haolin, J.L. 1995. Soil fertility and fertilizers. Fifth Edn., Prentice Hall of India Pvt. Ltd., New Delhi

Tiwari, S.C., Tiwari, B.K. and Mishra, R.R. 1989. Microbial populations, enzyme activities and Nitrogen Phospheros-potassium enrichment in earthworkm casts and in the surrounding soil of pine apple plantations. *Biology and fertility of soils* 81 : 177-182

Tomesue, E. 1970. Contributii fa metoda de determinare a celulazei din Sol. Microbiol. Lucr. Cont. Nat. Microbiol. Gen. Appl. Bucharest 509-513

Torello, W.A. and Wehner, D.J. 1993. Urease activity in a kentucky bluegrass test *Agronomy Journal* 75 (4) : 654-656

Torrani. 1968. Alkaline phosphatase of *Escherichia coli.* Methods of enzymology 128 : 212-213

Trasar cepeda, M.A. and Gil stores, F. 1988. Kinetics of acid phosphatase activities in various soils of Galicia. *Soil Biology and Biochemistry* 20 : 275-280

Trasar Cepeda, M.A. and Gilstores, F. 1987. Phosphatase activity in acid high organic matter soils in Galicia (NW Spain) *Soil Biology and Biochemistry* 19 : 281-284

Trasar Cepeda, M.C., Carballas, T., Gill-stores, F., De Blas, E. 1991. Liming and phosphatase activity and mineralization of P in an acidic soil. *Soil Biology and Biochemistry* 23 : 209-215

Trevors, J.T. 1984. Effect of substrate concentration, inorganic nitrogen, O2 concentration, temperature and pH on dehydrogenase activity in soil. *Plant and Soil* 77 : 285-293

Trevors, J.T., Mayfield, C.I., Innins, W.E. 1982. Measurement of electron transport system actively in soil. *Microbial Ecology.* 15 : 18-16

Tu, C.M. 1987. Effect of some pesticides on enzyme activieis in an organic soil. Bulletin of Environmental contanimation and Toxicology 27 : 109-144

Tu, C.M. 1991. Effects of pesticides on enzyme activities in an organic soil. Bulletin of Environmental Contamenation and Texicology. 27 : 109-114

Tyler, G. 1991. Heavy metals in soil biology and biochemistry. In : Soil Biochemistry, Vol. 5. Ed. Paul, P.A. and Ladd, J.N. pp 371-414. Marcel Dekker, New York

Van Cleve, K., Oliver, L., Schlentnor, R. and C.T. Dugmess. 1997. Productivity and nutrient cycling in Tara forest ecosystems. *Canadian Journal of Forest Research* 13 : 747-766

Van Huystee, R.B. and Cairns, W.I. 1982. Humus chemistry : Genesis, Composition, reactions. *Soil Science* 24 : 15-16

Vardavakis, E. 1989. Seasonal varion in heterotropic soil bacteria and some soil enzyme activities in a typical calcixeroll soil in Greece. *Rev. Ecol. Biol. Soil.* 26 : 233-247

Vaughan, D. and Ord, B.G. 1991. Influence of natural and synthetic humic substances on the activity of urease. *Journal of Soil Science* 42, 17-23

Vaughan, D., Maliom, R.E., Ord, B.G. 1985. Influence of humic substances on biochemical processes in plants. In : Vaughan, D., Malcom, R.E. (Eds.) Soil organic matter and biological activity. Junk Publishers, Dordrecht Boston Laneaster, pp 77-108

Verma, L., Martin, J.P. and Haider, K. 1975. Decomposition of carbon-14-labelled proteins, peptides and amino acids : free and complexed with humic polyness. *Soil Sci. Soc. Am. Proc.* 39, 279-284

Verstrete, W. and Voets, J.P. 1977. Soil microbial and biochemical characters in relation to soil management. *Soil Biology and Biochemistry* 9 : 253-258

Vlasyuk, P.A. and Lisoval, A.P. 1964. Effect of fertilizers on phosphatase activity of soil. Vestn Skh Naukhi, Vses Akad Skh Nauk 9 : 52-55

Voets, J.P. and Dedeken, M. 1964. Studies on biological phenomena of proteolysis in soil. Ann. Ist. Pasteur. Paris 107, Suppl. 3, 320-329

Von Mersi, W. and Schinner, F. 1991. An improved and accurate method for determining the dehydrogenase activity of soils with iodonitro tetrazolium chloride. *Biol. Fertil. Soils* 11 : 216-220

Votes, J.P. and Dedeker, M. 1965. Influence of high frequency and gamma radiation on the microflora and enzymes of soil. Meded. Landbouhogesch, opzoeking Sta Statt Gent. 30, 2037-2049

Walkley, A. and Black. 1934. An examination of the degtijareff method for determining soil organic matter and a proposed modification of the chromic acid titration method. *Soil Science* 37 : 93-101

Wang, Y.Z., Pan, T.G., Ke, Y.Q. and Zheng, S.Q. 1992. Effects of applying the used bagasse substrate on sugarcane mycorrhizal rhozasphere microbes and soil fertility. *Journal of Fujean Agricultural College* 21 : 4, 424-429

Watanabe, K. and Hayano, K. 1994. Source of Soil protease based on splitting sites of a poly peptide. *Soil Science and Plant Nutrition* 40 : 4, 697-701

Watanabe, K., Asakawa, S. and Hayano, K. 1994. Evaluation of extra cellular protease activities of soil bacteria. *Soil Biology and Biochemistry* 26 : 479-482

Wheatley, R., Ritz, K. and Griffiths, B. 1990. Microbial biomass and mineral N transformations in soils planted with barley, regegrass, pea or turnip. *Plant and soils* 127 : 157-167

Wilke, B.M. 1988. Effects of sodium selenite on microbial activity of mill, moder and mor soils. *Biology and fertility of soils* 6 : 148-152

Wolters, V. 1991. Biological processes in two beech forest soils treated with simulated and rain : A laboratory experiment with *Isotoma ligrina. Soil Biology and Biochemistry* 9 : 78-82

Wong, K.K.Y., Tan, L.U.L. and Saddler, J.N. 1998. Multiplicity of B-1-4-xylanase in micro organisms functions and applications. *Microbiology Review* 52 : 305-317

Wood, T.M. Mc Crae, S.I. and Mac Farlane, C.C. 1980. The isolation, purification and properties of the celluxo hydrolase component of *penicillium funiculosum* cellulase. *Biochem. J.* 189 : 51-65

Wu, J., Joergensen, R.G., Pommerning, B., Chaussool, R. and Brookes, P.C. 1990. Measurement of soil microbial biomass C by fumigation-extraction an automated procedure. *Soil Biology and Biochemistry* 22 : 1167-1169

Yadav, D.S., Kumar, Alok, Singh, R.M. and Acha, Ram. 1991. Yield, economics and nutrient balance in cropping systems based on rice (oryza sativa) *Indian J. Agric. Sci.* 61 (12) : 872-876

Yifong, T. and Ching Wen, C. 1993. The effect of dairy compost on crop production and soil characteristics. Bulletin of Taichung District Agricultural Improvement station, No. 40. p : 9-16

Zantua, M.I. and Bremner, J.M. 1975. Comparision of methods of assaying urease activity in soils. *Soil Biology and Biochemistry* 7 : 291-325

Zantua, M.I. and Bremner, J.M. 1975. Preservation of soil samples for assay of Urease activity. *Soil Biology and Biochemistry* 7 : 297-299

Zantua, M.I. and Bremner, J.M. 1977. Factors affecting urease activity in soils. *Soil Biol. Biochem.* 9 : 135-140

Zelles, L., Bai, Q.Y., Beck, T. and Beese, F. 1992. Signature fatty acids in phospholepids and biopolysacharides as indicators of microbial becomes and community structure in agricultural soils. *Soil Biology and Biochemistry* 24 : 317-323

Zelles, L., Stepper, K. and Zsolnay, A. 1990. The effect of lime on microbial activity in sprice picea abies L. forests. *Biology and Fertility of soils* 9 : 78-82

Zhang, R.S., Rahman, G.F., Vance, C and L.C. Munn. 1995. Geostatistical analyses of trace elements in soils and plants. *Soil Science* 159 : 383-390

Zukova, P.S., Kharitonova, A.P. and Pototskaya, L.A. 1993. Effects of herbicides on fermentative activity of soils under onion. Khimya-v-soil. Skom-khozyaistve. 21 : 5, 58-60

DISTRIBUTION, CHARACTERIZATION AND DYNAMICS OF SOIL ENZYMES IN SELECTED SOILS OF KERALA

by

B. APARNA

ABSTRACT OF A THESIS

Submitted in partial fulfilment of the requirement
for the degree
DOCTOR OF PHILOSOPHY
Faculty of Agriculture
Kerala Agricultural University

Department of Soil Science & Agricultural Chemistry
College of Agriculture
Vellayani, Thiruvananthapuram

2000

ABSTRACT

Biologically and biochemically mediated processes in soils are fundamental to the functioning of terrestial ecosystem. Understanding the underlying biological processes in tandem with identification of indicators of ecosystem stress is inevitable to provide strategies and approaches to promote long term sustainability of the fragile ecosystem. All members of the food chain are dependent on the soil as a source of nutrients, support and sustenance besides their reliance on soil as a sink for terrestial wastes. Now there is growing evidence to believe that soil biological parameters can hold potential as early and sensitive indices of soil degradation or restoration. In this context, soil enzymes play an active role in the microbial ecology and transformations brought about by catalysing enumerable reactions in soils. The activity of soil enzymes thus hold potential as soil quality indicators though they are sensitive to temporal changes due to environmental and management factors. Thus the long term effects of soil management techniques such as crop rotation, liming, fertilization and pesticide applications on the biological regimes need detailed investigations. Hence, with the overriding objective of projecting the effects of long term fertilization, application of soil amendments and agrochemicals on enzyme activities in relation to soil fertility, the present investigation was carried out.

Analysis of soil samples collected from the experimental plots of five permanent manurial trials distributed in the coastal, midland and mid-upland regions of Kerala reveal drastic differences in the activities of five major soil enzymes, viz., urease, phosphatase, protease, dehydrogenase and cellulase. It is obvious from the study of permanent manurial plots of the Rice Research Station, Kayamkulam that the activity with respect to urease, protease and cellulase is low compared to the corresponding samples collected from the long term experimental plots of Pattambi, Balaramapuram and Chethackal. The activity of phosphatase and dehydrogenase reported in this case was moderate to high compared to Pattambi and Balaramapuram. In the case of protease and cellulase the highest activity was represented by samples collected from the Central Experimental Station of the Rubber Research Institute of India, Chethackal. In general, the highest activity with respect to all enzymes studied have been reported in the intercropping experiment at the Central Experimental Station, Chethackal, representing the hilly terrain and the lowest values were seen for the permanent manurial experiment at Balaramapuram.

In the permanent manurial experiments, activities of the enzymes were found to be higher in plots receiving treatments involving combinations of organic manures and chemical fertilizers. It is evident from the study that the total quantity of organic amendments added to the soil *per se* could not sustain a higher microbial activity coupled with a higher enzyme activity, but required supplemental addition of mineral nutrients through chemical fertilizers also. Further, the role of supplemental addition of chemical fertilizers in the enhancement of enzyme activities was very clear from the stastically significant results obtained for the permanent manurial experiment at Pattambi.

Significant differences in the enzyme activities have been observed in the case of the permanent manurial experiment at Balaramapuram consequent to the main and interaction effects of chemical fertilizers at higher levels of addition especially for urease, phosphatase and dehydrogenase. Hence it is clear from the study that the application of mineral fertilizers over a basal dose of organics could enhance the microbial activity of the soil to significantly higher levels yielding higher enzyme activities even in a highly weathered low activity clay soil like the red loam soil of Balaramapuram.

Enzyme activities observed were the highest under the rubber based cropping system at Chethackal followed by the rice based experiments at Pattambi and Kayamkulam while the lowest activity was noticed at Balaramapuram. Of the physico-chemical characteristics studied through path co-efficient analysis and regression analysis, OC, CEC, R_2O_3, available N, available P, available K and respiratory activity were found to influence the enzyme activities to the maximum extent.

Pot culture experiments to study the effect of amendments and agrochemicals on enzyme activities revealed significant effects due to treatments. In all these cases, the maximum inhibitory effect on enzyme activities in general, were observed consequent to the application of antibiotics followed by fungicides, insecticides and herbicides. Immediately after the application of treatments there was a decline in the enzyme activity during the active tillering, the activity gradually resumed during the panicle initiation and then decreased asymptotically towards the harvest stage. Thus it is evident from the present study that considerable suppression of the enzyme activity is

observed consequent to the application of agrochemicals though it is rendered relatively harmless over a period of time within the life span of the crop. However, the long term effect of this application need to be investigated in detail.

Among the various amendments used in the present study, treatment involving the application of organic amendment as vermicompost in combination with lime and fertilizers was found to be significantly superior. All the five enzymes thus registered the highest value in this treatment irrespective of the stage of the crop.