THE EFFECTS OF SOIL AMENDMENTS ON SELECTED PROPERTIES OF TEA SOILS AND TEA PLANTS (*Camellia sinensis* L.) IN AUSTRALIA AND SRI LANKA

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AFFECTIONATELY DEDICATED

TO

MY PARENTS, TEACHERS AND MY FAMILY

ABSTRACT

Organic matter transformations and nutrient cycling in soils depend on the activity of soil organisms. Deterioration of soil fertility lowers the biological activity and results in lower productivity. In the absence of adequate organic matter, the processes of conversion of nutrients to plant-available forms and their retention are very low. To enhance the activity of soil organisms especially that of beneficial microbial populations, the addition of high quality organic amendments is very important. Even though there are plenty of organic materials available in tea lands, there is inadequate information on their suitability and influence on the biological properties of soils.

The present research has attempted to determine the extent to which the microbial activity and productivity of tea soils in Australia and Sri Lanka can be manipulated by use of readily available soil amendments. Trials were conducted using grass and leguminous mulching materials with different C/N ratios, in combination with two pH amendments (dolomite and 'Minplus^{TM}' – a finely ground volcanic rock dust - both applied at rates of 2500 kg ha^{-1} only for the pot trials and 1000 kg ha^{-1} for the other trials) and an inoculum of biologically active rain forest soil. The nursery stage of tea (*Camellia sinensis*) propagation was studied in a shadehouse at James Cook University, Townsville, Australia, and young tea (unpruned, 1 year after planting) and mature tea (pruned bushes, 5 years since planting) were studied in a field in Sri Lanka.

In the nursery trial, mulching materials consisting of finely chopped *Brachiaria decumbens* (a grass), *Calliandra calothyrsus* (a legume), and tea prunings were applied at a rate of 35 tonnes fresh weight ha^{-1} $year^{-1}$ to pots with or without tea seedlings.

The grass mulch with Minplus improved soil organic carbon, CEC, soil pH, microbial biomass carbon, plant available phosphorus, and total nitrogen contents of the soil and enhanced the growth of plants when compared to the effects of *Calliandra* legume and tea mulch. All the combinations of mulches with dolomite reduced plant growth even though they enhanced some soil properties. Application of grass and legume mulches increased the beneficial population of gram positive bacteria, fungi, and mycorrhiza. Grass mulch also improved the growth of tea as measured by shoot weight and total biomass. The addition of a rainforest inoculum to the soils of the nursery tea plants increased the soil microbial biomass carbon and growth of tea plants even in the absence of any mulch.

The field trials in Sri Lanka demonstrated the extent of the changes induced by mulches and soil pH modifiers in soil microbial properties, including the abundances of functional groups of microbes (bacteria, fungi, mycorrhizae), soil microbial biomass carbon, and microbial respiration. In addition organic carbon, soil pH, nitrogen and mulch decomposition rate were measured. The mulching materials tested were: refuse tea (25 tonnes ha^{-1} $year^{-1}$), Mana grass (*Cymbopogon confertiflora*), and branches of Dadap (*Erythrina lithosperma*), a leguminous tree (35 tonnes fresh weight ha^{-1} $year^{-1}$). In addition to these treatments, lemon grass (*Cymbopogan nardus*) 20,000 plants ha^{-1} as live mulch in young tea and a *Trichoderma* fungal culture in the mature tea were used. For young tea and mature tea, Mana and Dadap were applied four times and Refuse tea three times per study period and the lemon grass was planted at the start of the

trial on a 15 x 15 cm spacing in the young tea; *Trichoderma* was applied once to the mature tea trial at a rate of 500 g of spore culture / plant.

The results indicated that Dadap and Refuse tea raised the yield of tea significantly by 16% and 19% respectively in young tea, and by 14% in mature tea for both mulches. The mulches enhanced soil pH, microbial biomass carbon, soil respiration and also suppressed the most detrimental gram negative bacterial populations one year after the application of treatments in young tea and increased soil nitrogen by refuse tea in mature tea trial. The quality of tea increased in tea grown under the control and lemon grass mulch treatment in young tea and in *Trichoderma* fungus-treated plots in mature tea.

Minplus rock dust and the rainforest soil inoculum enhanced the growth of the nursery plants. The most suitable mulching materials to accelerate the biological activity were found to be those with C/N ratios below 20, and low in lignin and unoxidisable polyphenol. Therefore, the suitable materials for use as mulches on tea lands are *Brachiaria grass*, refuse tea, and Dadap legume. They also suppressed the development of pathogenic bacterial populations particularly gram negative bacteria. These materials also improved the biological properties of soil and thereby enhanced the growth and yield of tea.

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Glossary of terms

AP	Available phosphorus
CEC	Cation Exchange Capacity determined as the sum of the basic cations (Ca^{2+}, Mg^{2+}, K^{+}, Na^{+}) and the acidic cations (H^{+}, Al^{3+})
Dadap	The small leguminous tree *Erythrina lithosperma*
Dolomite	Calcium and magnesium carbonate: $CaMg(CO_3)_2$
FAME	Fatty Acid Methyl Ester
Guatemala grass	*Tripsacum laxum*
Immature tea	Young tea (1-2 yrs old)
JCU	James Cook University
Lemon grass	*Cymbopogon nardus*
Made tea	Black tea after processing
Mana grass	*Cymbopogon confertiflorus*
Mature tea	More than 20 years old tea
MBC	Microbial biomass carbon
MBN	Microbial biomass nitrogen
Minplus	Finely ground volcanic rock dust produced by Pacific Mineral Developments Pty Ltd, Innisfail.
N	Nitrogen
NH_4^+	Ammonium cation
Nursery tea	1-9 months old tea
OC	Organic carbon
P	Phosphorus
Plucking Table	Top part of the canopy where flush is harvested

PO_4^{3-}	Phosphate anion
Refuse tea	Waste product of the tea manufacturing process and is the partly ground, brown stalk and fibrous particles remaining after separating the commercial components of the manufactured tea
S	Sulphur
SO_4^{2-}	Sulphate anion
TRI	Tea Research Institute of Sri Lanka
Young tea	1-2 years old tea

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CHAPTER 1

INTRODUCTION

1.1 AIMS OF THE RESEARCH

The responses of tea plant growth to soil chemical and microbiological properties and amendments by organic and inorganic materials in the tea lands of north Queensland and Sri Lanka have been studied.

The research has been carried out in shadehouse pot trials at James Cook University Townsville, Queensland, to determine the impacts of a range of soil amendments on selected chemical and microbial properties of an Australian tea-growing soil, and thereby on the growth of tea seedlings. Some of the successful treatments from the Australian pot trial were applied to young tea plants (less than 2 years old) and to mature, pruned tea plants (more than 5 years old) on a plantation of the Tea Research Institute at Talawakelle, in upland Sri Lanka.

The specific aims of the research project were:

- To quantify the effects of adding different organic mulches (refuse tea, a grass, a legume, and live grass) and soil acidity modifiers (dolomite and 'Minplus' volcanic rock dust) on soil chemical and biological properties, and on the growth of juvenile and mature tea plants;

- To study the influence of selected soil microbiological properties (microbial biomass carbon and nitrogen; microbial functional group structure) on the growth of tea plants;
- To identify cultural practices to improve the productivity of tea soils and provide information as a basis for formulating preliminary guidelines for the effective management of upland soils in Sri Lanka.

1.2 BACKGROUND

In this chapter the literature on the biological and chemical properties of the soils of the humid tropics is reviewed with special reference to tea-growing soils, and the beneficial and pathogenic activities of microflora are discussed. The effect on soil fertility of continuous cultivation in acid soils and the feasibility of manipulating soil fertility by using soil amendments are also addressed.

1.2.1 Tea soils

Tea is grown in soils of the humid tropics (eg. Sri Lanka and India) and the subtropics (eg. Japan, and Georgia in Russia) in soils that are generally highly weathered and strongly acidic. Soils of tea lands in different countries differ widely in parent material and morphological characteristics but the most important requirement is for soil pH to be low, generally 4.5-5.5. Soils should also be deep, permeable, and well drained (Somaratne 1986). The soils of the agro-ecological regions suitable for tea in Sri Lanka fall into three major soil groups; red yellow podsolic (Ultisol), reddish brown latosolic soils (Ultisol), and immature brown loams (Entisol), of which the largest extent is red-yellow podsolic soils (Watson 1986).

Tea cultivation is practiced in different countries and on different types of soils. In the highly weathered tea soils, high organic matter decomposition rates, coupled with low ion retention capacity determine low soil fertility (Gillman and Sumpter 1986). The development of land for agricultural purposes has often resulted in significant soil degradation, including the gradual deterioration of soil structure, a decline in soil organic matter, loss of nutrients especially nitrogen and phosphorus through erosion and leaching, a reduction of soil organism biomass, and in some soils, acidification associated with increased inputs of nitrogenous fertilisers such as ammonium sulphate (Sandanam *et al*. 1978; Lee and Pankhurst 1992). All the tea soils are acidic and acidity is usually associated with high exchangeable Al^{3+} and low exchangeable bases, especially (Ca^{2+}, Mg^{2+}). Physical, chemical and biological conditions should be suitable for the initial establishment of tea plants in soils as well as for achieving high yields subsequently. The physical and chemical properties suitable for tea are well-established by Ranganathan (1977), Natesan *et al*. (1985), Anandacumaraswamy and Amarasekera (1986), Anandacumaraswamy *et al.* (1989), Barua (1989) and Othieno (1992), whereas only limited information is available on the biological conditions of soils suitable for tea (Bezbaruah 1994). Management practices that contribute to land degradation such as excessive cultivation of soil, continuous cropping, removal of crop residues, and excessive uses of pesticides and fertilisers all contribute to reduced sustainability of agriculture in different parts of the world. (Dalal *et al*. 1995; Lee and Pankhurst 1992). Among some beneficial microbial species, *Bacillus mycoides* and *Bacillus subtilis* have been identified in the rhizosphere of Indian tea *Camellia sinensis* and *C. assamica* (Pandey and Palni 1997). Rhizobacteria promoting plant growth increase the crop yield by production of plant growth hormones, and suppression of minor plant pathogens (Kloepper *et al*. 1980).

1.2.2 Soil acidity

The pH of microbial cytoplasm is close to neutral and the majority of soil microbes grow best at pH values close to 7 (Gray and Williams 1971; Alexander 1977). Acidophilic organisms can grow at pH 6.0 or less. Soil bacteria and actinomycetes are generally less tolerant of acid conditions than are fungi. The critical pH level for most bacteria and actinomycetes is around 5 (Gray and Williams 1971), and only a few bacteria can grow in strongly acidic soils (pH 3.0) (Alexander 1977). The most acid-tolerant S-oxidising bacteria (*Thiobacillus* spp.) can grow at pH 1, while the most alkali-tolerant *Streptomyces* can grow at pH 10 (Killham 1994).

Microbial decomposition of organic matter tends to decrease soil pH through the production of organic acids. Ammonium-based fertiliser and sulphide-rich areas also increase the acidity of soils, thus adversely affecting nutrient availability to plants.

Application of lime ($CaCO_3$) or dolomite [Ca Mg $(CO_3)_2$] reduces the acidity of tea soils. Liming increases the bacterial population substantially in the short-term and to a lesser extent in the long-term, because bacterial species are recognized to be acid-intolerant (Alexander 1977). Shah *et al.* (1990) showed that liming increased the bacterial population by 20-fold in the short term (32 days), but the increase did not persist in a brown podsolic soil near Aberystwyth, Wales; liming had no significant effect on the fungal population, either in the short or long term in the same soil. Liming caused a rapid and substantial increase in both soil respiration and net nitrogen mineralisation in solonetzic soils in northeastern Alberta, Canada (Carter 1986). In general, positive

effects on the accumulation of mineral N have been reported following the liming of acid mineral soils (Ishaque and Cornfield 1972).

1.2.2.1 *Soil amendments used in tea soils of Sri Lanka*

Soil acidity is one of the most important soil properties influencing tea growth. Generally the pH of the soils in tea-growing countries varies from 3.3 to 6.0 (Natesan 1999), and a soil pH of 4.5 - 5.5 is considered to be the optimum for the utilization of nutrients especially nitrogen (Natesen 1999), and for the growth of tea (Sandanam *et al*. 1978). While most tea varieties yield best in the soil pH range of 4.5-5.0 (Saikh 2001), certain tolerant varieties can flourish at a higher pH of 6.0-6.5 (Natesen 1999). Continued addition of nitrogen fertilizers to tea fields eventually causes a reduction in soil pH, with an associated decline in fertility, and liming is therefore practiced to maintain the soil pH at the optimum level. Presently, dolomite found in certain parts of central province of Sri Lanka is the only liming material applied to tea fields to improve the soil pH of the highland tea estates of Sri Lanka (Wickramasinghe *et al*. 1981).

The Sri Lankan tea industry uses very large quantities of dolomite, around 2000 tonnes annually, and although locally sourced, its availability may become limited with time. Limestone is also locally available but it is not used by the tea industry due to its very high calcium content. Dolomite gives magnesium, calcium, and soil pH corrections whereas, lime gives calcium and pH correction only. Magnesium is important as a tea nutrient (Wickramasighe, and Krisnapillai 1986). Given the possibility of a decline in the availability of dolomite, it is important to look for future supplies of Sri Lankan dolomite, or for an alternative liming material.

Farmers may use large amounts of lime and fertilisers to encourage high crop production from the acidic soils of the humid tropics. In such farming systems, the input of soil organic matter is important, yet the processes of reduction of soil acidity, the optimisation of soil organic matter contents, and the retention and efficient use of soil nutrients are poorly understood. Conventional agriculture practiced over many years has resulted in reduced organic matter levels and degraded soil structure, resulting in the decline of crop productivity in other parts of the world (Dalal and Mayer 1987).

Organic matter turnover and soil structure are regulated by micro- and macro-organisms and, in order to achieve sustainable land use, it is essential to stimulate biological activity of soils (Alexander 1977; Gupta and Roper 1994; Killham 1994). Crop residue retention (Doran 1980; Roper 1983; Powlson *et al*.1987; Dalal *et al.*1995; Amir and Pineau 1998) and inorganic amendments such as lime (Ishaque and Cornfield 1976; Sandanam *et al.*1978; Carter 1986; Shah *et al.*1990) have been shown to maintain or improve soil organic matter levels and soil structure and crop productivity. But the associated changes in biological processes under different soil amendments are poorly understood (Sivapalan *et al*. 1985; Gupta and Roper 1994).

1.2.3 Microbial decomposition and the utilisation and release of soil nutrients

Nutrients such as carbon, nitrogen, sulphur, and phosphorus are important in the life cycles of plants, animals, and micro-organisms, and are cycled in the soil between organic matter residues and the plant available nutrient pool. The death and microbiologically-mediated decay of organisms result in release of inorganic ions which can be readily taken up by plants (Paul and Clark 1989; Killham 1994). Thus, decomposing organic matter acts as a slow release fertiliser.

Some soil microbes obtain a significant proportion of their nutrient requirements directly from the weathering of soil minerals, sometimes accelerating weathering by the production of organic acids (Killham 1994). Decaying organic matter, consisting of plant, animal, and microbial residues represents a source of microbial nutrition by mineralisation (which involves the degradation of proteins, amino sugars, and nucleic acids with their nitrogen components changed to cationic or anionic mineral forms), or converting nutrients locked up in organic matter to a "mineralised" plant available form (Killham 1994). Nitrification i.e. conversion of NH_4^+ to NO_2^- and then to NO_3^- (available form to plant) and immobilisation of nitrogen by i.e. conversion to ammonium (NH_4^+) or to ammonia (NH_3) forms is driven by many genera of micro-organisms in the soil (Paul and Clark 1989). However in Sri Lankan tea soils, the amount of nitrogen immobilisation taking place is negligible (Wickramasinghe *et al*., 1985b). Micro-organisms can also directly acquire nutrients from other organisms. For example, phagotropic soil protozoa such as *Sarcodina* may directly ingest many thousands of bacteria in a life cycle, and some fungi can entrap nematodes with their hyphae and slowly digest them (Alexander 1977; Paul and Clark 1989; Killham 1994). Fungi, actinomycetes, and most bacteria are heterotrophs, requiring organic matter as a source of energy and carbon, and their distribution is determined by the availability of an oxidisable, organic substrate (Alexandra 1977; Lee and Pankhurst 1992). Certain bacteria can fix molecular nitrogen and others are able to utilise methane (Lynch 1983).

Soil organisms such as bacteria, actinomycetes, fungi, algae, protozoa, nematodes, annelids, and micro- and macro-arthropods play a major role in the optimisation of soil organic matter in the soil, and in the retention and efficient use of soil nutrients in

sustainable production. Many soil micro-organisms are heavily involved in the primary processes of direct nutrient cycling (Alexander 1977; Paul and Clark 1989; Lee and Pankhurst 1992; Dalal 1998).

In a fertile soil in the temperate zone, biomass contributed by microflora (bacteria, fungi, actinomycetes, and algae) may exceed 20 tonnes ha^{-1}, but their contribution in the tropics has not been estimated (Lee and Pankhurst 1992).

Micro- and macro-organisms in soils regulate organic matter turnover and affect soil structure thereby influencing the productivity of soils. It is therefore essential to stimulate biological activity if soil productivity is to be maintained (Alexander 1977; Gupta and Roper 1994; Killham 1994). Crop residue retention (Doran 1980; Roper 1983; Powlson *et al*. 1987; Dalal *et al*. 1995; Amir and Pineau 1998) and the use inorganic soil amendments such as lime (Ishaque and Cornfield 1976; Sandanam *et al*. 1978: Carter 1986; Shah *et al*. 1990) have been shown to maintain or improve soil organic matter levels, soil structure, and crop productivity. But the associated changes in biological processes under different soil amendments are poorly understood (Sivapalan *et al*.1985; Gupta and Roper 1994).

Soil organisms play a major role in mineralisation of organic matter, the production of humus, and the recycling of nutrients for sustainable production. Plant roots, fungi, actinomycetes, and bacteria, provide most of the biomass and biological activity in soils (Pankhurst 1994). Though the soil biota contains approximately 1-8 % of the total organic carbon of the soil (Doran and Smith 1987; Dalal 1998), the importance of the

soil animals lies not in their absolute biomass but their functional activities, such as carbon, nitrogen, phosphorus cycling (Pankhurst and Lynch 1994).

Bacteria, fungi, actinomycetes, and algae constitute functional groups of microflora. Bacteria are very important because they are the principal agents for the global cycling of many inorganic compounds such as inorganic nitrogen, sulphur, and phosphorus (Lynch 1983). The soil organic matter must be mineralised by micro-organisms to an inorganic form for plant uptake. The biological mineralisation of organic residues producing NH_4^+, NO_3^-, PO_4^{-3}, and SO_4^{-2} ions is evident from the linking of inorganic nutrient cycles that are driven by microbial utilisation of carbon for energy (Smith *et al.*1993). It has been estimated that in the Pacific Northwest of the USA, a winter wheat crop will produce 16 tonnes ha^{-1} of dry matter and it will contribute 302, 36, and 32 kg ha^{-1} of N, P, and S, respectively (Smith and Paul 1990). The readily available average N, P, and S of soil organic matter is 180, 17, and 9 kg ha^{-1} respectively which is merely 60, 47 and 28 % of the N, P, and S requirements for the crop (Smith and Paul 1990). Microbial biomass N represents 1-5% of the total soil nitrogen and studies conducted in a Canadian soil showed that in arable systems, biomass nitrogen is 40-385 kg ha^{-1}, in forest systems 130-216 kg ha^{-1}, and in grasslands 40-496 kg ha^{-1} (the average biomass N is 195, 170, and 225 kg ha^{-1} for arable, forest and grassland systems, respectively (Smith and Paul 1990).

Bacteria, numbering 10^6-10^9 gram $^{-1}$ of soil, contribute less than half of the total biomass in aerated soil (Alexander 1977). However in conditions of low oxygen availability, bacteria may account for most of the microbial biomass. Soil fungi are numerically less {(10^4 - 10^9) $gram^{-1}$ of the soil} than bacteria (Lynch 1983), but are as important as

bacteria because their filamentous growth habit enables them to exploit a greater soil volume. Like bacteria, soil fungi become active in favourable environmental conditions and they are the main agents for decomposition of cellulose and lignin in organic matter residues. Actinomycetes play a major role in the decomposition of organic compounds in soil; cyanobacteria have the ability also to fix atmospheric nitrogen making a valuable input and contributing a significant organic carbon input to the soil (Lynch, 1983). Numbers of protozoa (10^4 - 10^5 gram $^{-1}$ soil) and algae (10^1 -10^6 gram -^{1} soil) have both been estimated in dry soils (Lynch, 1983). Abiotic parameters such as sunlight, carbon dioxide level, and oxygen level, regulate the growth of algae and protozoa in soils. Protozoa are important predators in soils and regulate the size of the bacterial population (Alexander 1977). Algae contribute organic carbon in the form of extra cellular polymers to the soil help to enhance soil structure through the production of extra cellular polymers (Alexander 1977). The cycle of nitrogen transformation is shown in Fig. 1.1. NH_4 is automatically converted to NH_3 form.

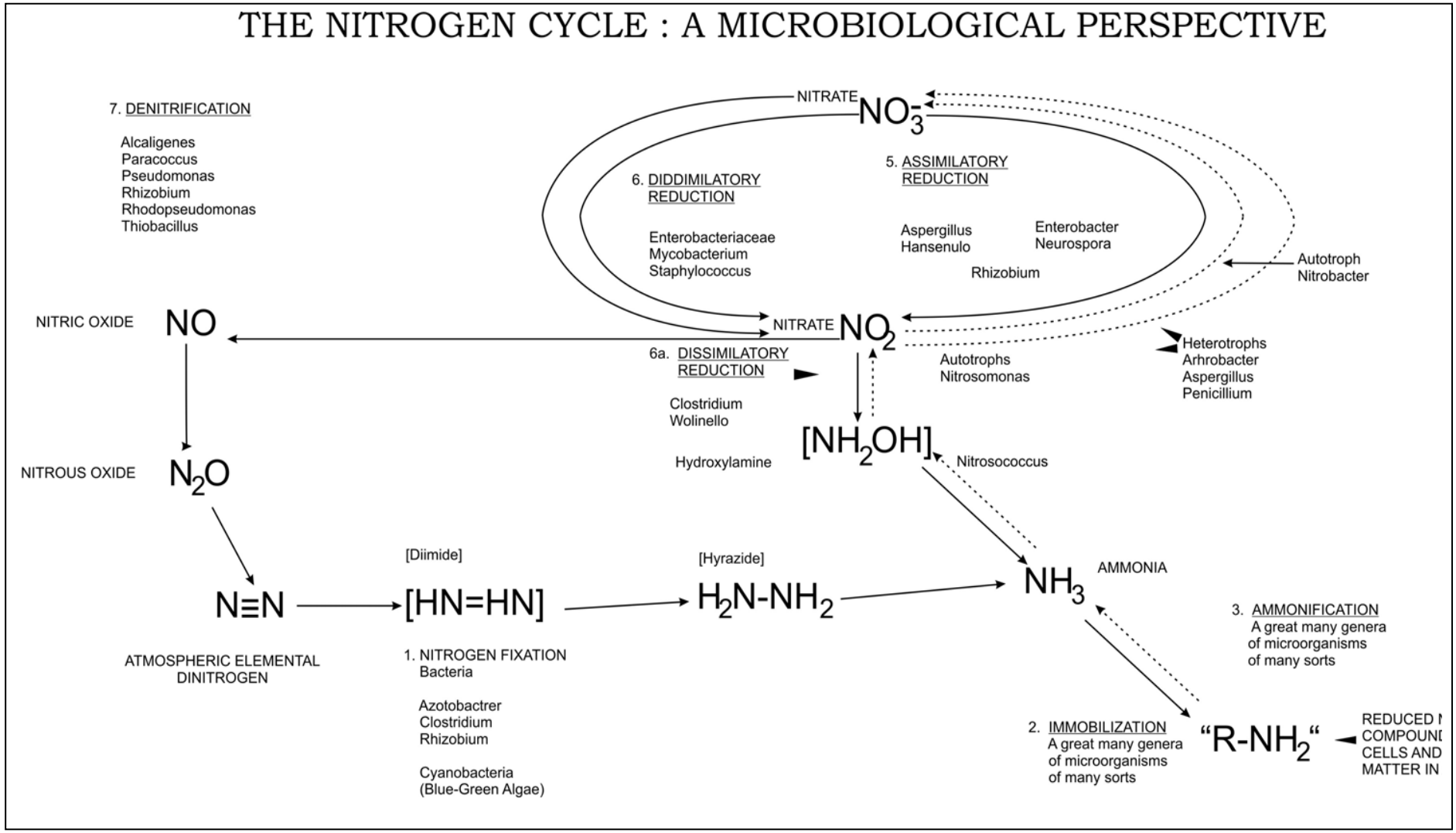


Fig. 1.1 Microbial transformation of nitrogen (Source: Paul and Clark 1989)

Microbial transformation includes mineralisation, immobilisation and denitrification (Fig.1.1). Mineralisation (ammonification and nitrification) refers to the degradation of organic nitrogen (proteins, amino sugars, and nucleic acid) to NH_4^+. It involves autotroph *Nitrobacter* and heterotrophs *Arthobacter*, *Penicillium*, and *Aspergillus*. Immobilisation refers to the incorporation of NH_4^+ into amino acid and depends largely on the availability of substrate carbon; it involves many genera of microorganisms. Microbial reduction of NO_3^- to NO_2^- and then to gaseous N_2O and N_2, which are commonly lost to the atmosphere, is referred to as denitrification. This process involves certain microorganisms in the absence of oxygen (Paul and Clark 1989).

Energy and nutrients from organic residues flow through the microflora to the predatory micro-, meso-, and macrofauna during decomposition in the detritus food web cycle. The functioning of the food web is driven by inputs of organic matter. Microorganisms utilise the energy, carbon, and nutrients available in the organic matter to produce new microbial biomass. Therefore, the mineralisation of nutrients by micro-organisms, and the turn over and release of nutrients contained within the microbial biomass by soil fauna, are the key factors sustaining soil fertility (Pankhurst and Lynch 1994).

1.2.4 Soil environmental factors affecting microbes

Environmental conditions as well as the non-biological factors affecting the density and composition of microbes are discussed in the following sections. Non-biological factors can alter the nature of the microbial population and its biochemical composition. The primary environmental factors influencing soil microbial populations include moisture, aeration, temperature, acidity, organic matter, and inorganic nutrient supply (Alexander

1977). Other factors include cultivation, season, and depth, and their influence may be enhanced by interactions with primary factors (Alexander 1977).

1.2.4.1 *Moisture and aeration*

Many microbial cells die with desiccation and only those with resistant propagules can survive long periods of drought. Actinomycetes in Kenyan soils (Meiklejohn 1957) are known to survive as spores, sclerotia, rhizomorphs, and resting hypae. McLaren and Skujins (1968) showed that when soil was stored dry for ten years, a hundredfold decrease in microbial population occurred; the survival of the remaining micro-organisms was due to the presence of resistant structures, or possibly to the presence of residual osmotic and hygroscopic water in the soil (Gray and Williams 1971). The optimal level for the activities of aerobic microbes lies in the range of 50-70% of the soil's water holding capacity (Alexander 1977).

Microbes respond to changes in the concentration of gases in the soil atmosphere. Some bacteria such as *Clostridium botulinum* are strictly anaerobic, and are unable to grow in the presence of oxygen, while others are strictly aerobic; *Psedomonas fluorescens* and the actinomycetes and cannot tolerate waterlogged conditions. Fungi are strictly aerobes, but the quantitative relationship between growth and oxygen supply varies in different species (Gray and Williams 1971). Some fungi such as *Fusarium* spp. tolerate low oxygen and high carbon dioxide concentrations than either bacteria or actinomycetes, although other actinomycetes and bacteria grow well only in very low oxygen tensions (Gray and Williams 1971).

Anaerobic microenvironments occur at various times in many soils, allowing the growth of anaerobic bacteria, including those which fix nitrogen (eg. *Clostridium pasteurianum*). Anaerobic conditions often lead to the accumulation of sulphides and ferrous iron, with a decrease of nitrate and available phosphates. The concentration of oxygen affects the redox potential of soil, but it is not known whether this in itself is a determining factor in microbial distributions (Alexander 1977).

The concentration of CO_2 in the soil atmosphere influences microbial populations in three ways: by affecting the pH of micro-habitats; by providing a source of carbon for autotrophic microbes, and by the inhibiting activity of the heterotrophic microflora (Alexander 1977). In general, soil microbes are tolerant of high carbon dioxide and low oxygen concentrations, and even extreme fluctuations in the soil atmosphere do not inhibit microbial development.

1.2.4.2 *Temperature*

Temperature has both direct and indirect effects on micro-biological activity in soils. The indirect effects are those caused mainly through temperature induced-changes to other aspects of soil physiochemical environment such as gas and water diffusion rates, weathering rates, and water activity (Killham 1994).

Temperature governs all biological processes and the optimal temperature range for most soil micro-organisms (mesophiles) is between 25^{o} C and 35^{o} C, and they have a capacity to grow at temperatures up to about 45^{o} C. There are also cold-tolerant micro-organisms known as psychrophiles, which can grow at low temperatures below 20^{o} C and 'snow-mould' fungi can decompose leaves buried under snow (Killham 1994). There is no

evidence for the presence of true psychrophilic bacteria in soil; the bacteria found to be active even in winter are called tolerant mesophiles rather than psychrophiles (Alexander 1977).

There is an approximate doubling of activity in the mesophilic microbial communities of most soils for each 10^{o} C rise in temperature up to 35^{o} C. This is followed by a dramatic fall in activity if temperature increases any further, caused mainly as a result of denaturation of proteins and membranes of micro-organisms (Killham 1994). Thermophiles, however, can tolerate temperatures from 45^{o} C to 65^{o} C, with some species able to tolerate temperatures higher than 90^{o} C (Gray and Williams 1971; Alexander 1977, Lindsay and Creaser 1977).

Sarathchandra *et al.* (1989) observed seasonal changes of microbial biomass, with higher biomass values in autumn and late spring compared to those of winter and early spring. Soil light (total solar radiation at the earth's surface - reflected) represents about 5 % of net solar radiation and also affects the photoautotrophic (plants, soil algae, and photoautotrophic soil bacteria) component of the soil biota. Light provides the energy source for the photoautotrophic soil bacteria that convert sunlight to energy for use by plants (Killham 1994).

1.2.4.3 *Soil properties*

Crop residues have significant effects on soil organic matter contents, microbial biomass levels, decomposition rates, and nutrient dynamics (Smith *et al*. 1993). Crop residues are used and managed to protect soils from wind and water erosion, to maintain productivity and soil organic matter content, and to improve soil physical properties such

as bulk density, water retention, and soil aggregation – all as a result of microbial production of mucilage (Parr and Papendick 1978).

The drastic decrease of soil organic matter in many cropping soils of the world over the last 80 years has focused attention on the importance of crop residues being returned to land (Smith *et al.* 1993). One of the factors improving the soil organic carbon pool is by adding crop residues to maintain soil fertility. Soil carbon sequestration through integrated nutrient management for 20 years in some soils of India showed a 1.0 -1.8 % increment of carbon by adding 30 (DW) Mg/ha/yr of farmyard manure (Lal 2004). In tea crops, yield and use efficiency of inputs are adversely affected by low levels of the soil organic carbon pool (Anandacumaraswamy *et al*., 2001). Ideal conditions for increasing microbial activity occur when surface residues are dispersed and come into contact with soil particles. Although the burning of stubble and removal of residues can cause the loss of soil organic matter, cultivation is the major cause of loss soil organic matter in agricultural land (Smith *et al.* 1993). Residues and their decomposition alter the composition of soil organic matter and affect the release of N, P, and S through microbial activity. If the carbon content of the residues is high compared to their N, P, and S contents, the residues will cause significant immobilisation of nutrients that would otherwise be available for plant uptake (Paul and Clark1989, Smith *et al.* 1993). This could be overcome by the addition of fertiliser or other residues with N and P contents higher than 1.5% and 0.3% respectively (Power and Legg 1978).

Organic matter is a dominant food reservoir for soil microbes and the microbial biomass is influenced by land use patterns. McLaughlin *et al*. (1988) and Robertson *et al*. (1997) showed that the microbial biomass is 20-80% higher in grasslands and forest soils than

in arable soils mainly because of the larger organic carbon inputs. The average microbial biomass carbon has been estimated at 560, 680, and 870 mg C kg^{-1} soil in cultivated land, forest, and grassland respectively in a temperate Canadian soil (Smith and Paul 1990). Although soil microbial biomass levels were lower in the forest land than in the grassland, the potential metabolic activity of the microorganisms tended to be greater in the forest (Ross *et al*. 1996). In southern Queensland, Dalal and Mayer (1987) estimated that microbial biomass carbon varied between 361- 508 mg kg^{-1} in forest soils which is lower than in temperate soils. The microbial biomass carbon content of a soil reflects the long-term amount of carbon input into the soil (Dalal and Mayer 1987). Agricultural practices such as tillage (McGill *et al.* 1986; Doran and Smith 1987), manuring, or residue incorporation (Powlson *et al.* 1987) influence microbial biomass content which, in a monoculture, was significantly lower than that of land under crop rotations. Similarly, microbial biomass carbon contents are lower in mineral-fertilised soils compared to organically manured plots (Insam and Parkinson 1989).

1.2.5. The effects of management practices on tea soils

1.2.5.1 *Fertiliser*

Tea soils are usually acidic due to the prolonged use of nitrogenous fertilisers such as urea and ammonium sulphate to obtain high crop production (Ishaque and Cornfield 1974; Sanadanam *et al*. 1978; Walker and Wickramasinghe 1979; Golden *et al*. 1981; Wickramasinghe *et al*. 1981; Nioh *et al.* 1995). Information on urea hydrolysis, nitrification, denitrification, and immobilisation of nitrogen fertilisers is available for some Sri Lankan tea soils (Sandanam *et al*. 1978; Wickramasinghe and Talibudeen 1981; Wickramasinghe *et al*. 1984; Wimaladasa and Wickramasinghe 1986).

In the tea plantations of Sri Lanka, continuous use of ammonium sulphate fertilisers at rates of 200 - 300 kg N ha^{-1} over the past 20 - 30 years has resulted in increasing acidity in the soil. Currently, urea is being increasingly used as an alternative source of nitrogen because urea acidifies the soil less than ammonium sulphate or nitrate-based fertilisers and releases potassium, calcium, and magnesium into the soil solution from cation exchange sites (Wickramasinghe *et al*. 1985a). Sri Lankan tea soils treated with urea and ammonium sulphate behaved similarly with negligible immobilisation of nitrate-nitrogen (Wickramasinghe *et al*. 1985a).

Limited information is available, however, on microbial activity in Sri Lankan tea soils (Walker and Wickramasinghe 1979; Sivapalan 1982). In Sri Lankan and Bangladesh tea soils, some nitrifying (ammonium-oxidising) bacteria have been isolated (Walker and Wickramasinghe 1979). All the Bangladesh nitrifiers were *Nitrosospira* species and Sri Lanka isolates were identified as *Nitrosolobus* species, *Nitrosospira* spp, and *Nitrosovibrio* (Walker and Wickramasinghe 1979).

Suppression of bacteria may occur as a result of increased acidity due to the use of ammonium-based fertilisers in tea lands (Ishaque and Cornfield 1974; Wickramesinghe *et al*. 1985a; Nioh *et al*. 1995). In contrast, Hayatsu and Kosuge (1993) showed that ammonium supplied to tea field soils as ammonium-based fertilisers was rapidly transformed into nitrate nitrogen by acid-tolerant autotrophic nitrifiers of the kind found in acid soils in Japan.

Fungi are more able to compete than bacteria and actinomycetes in an acid environment when supplied with adequate nitrogen (Alexander 1977). Actinomycetes become

prominent and more efficient when nutrients become limiting and there is less competition with other micro-organisms (Alexander 1977; Nioh *et al*. 1995).

Wicramasinghe *et al*. (1985b) showed that at pH 4, acidic St Coombs Soil, a Sri Lankan tea soil, had lower microbial activity than the neutral Highfield soil at pH 6.8 in the United Kingdom. They also showed that urea-treated St Coombs soil provides more nitrate nitrogen, than does the soil when treated with ammonium sulphate. This is probably because of the temporary rise in soil pH associated with urea hydrolysis (Overrein 1967) favoured the activity of nitrifying bacteria in this strongly acid soil. In contrast, Hayatsu and Kosuge (1993) showed that nitrogen supplied in the ammonium form to acidic tea soils in Japan was rapidly transformed into the nitrate form by acid-tolerant autotrophic nitrifiers in the soil.

The rate of nitrification in the red-yellow podsolic tea soils of Sri Lanka, as is common in other soils, is governed by factors such as aeration, temperature, soil pH, and soil moisture content. In Sri Lanka, nitrification rates may result in the production of 20 ppm nitrate-nitrogen, even at a pH of 3.7 (Sandanam *et al.* 1978). Increasing the soil pH increased nitrification appreciably in a good soil in Sri Lankan tea lands but only to a limited extent in a poor soil (Sandanam *et al.* 1978), and there was substantial nitrification (resulting in 39% recovery of applied nitrogen as nitrate-nitrogen) in a good soil even in the absence of any other soil amendments.

1.2.5.2 *Mulching*

Mulching is the application of organic or inorganic materials to the soil surface to provide a ground cover, reduce weed growth, conserve soil moisture, and moderate extremes of soil temperature.

'Thatching' is the common word for mulching in tea lands. The effect of mulching or thatching on plant growth and soil properties has been well documented, with beneficial effects of suppression of weed growth (Manipura 1972; Sandanam and Rajasingham 1982; Somaratne 1986); conservation of soil moisture by reducing evaporation and runoff (Sandanam and Anandacumaraswamy 1982); protection from erosion (Hasello and Sikurajapathy 1965; Sandanam and Rajasingham 1982; Basnayake 1985); increased infiltration rates (Ungei, 1975; Sandanam *et al.* 1978; Acosta-Martinez *et al.* 1999); reduction in soil temperature fluctuation (Grice 1990); and enhanced nitrification of organic matter (Sandanam *et al*.1978; Krishnapillai 1984).

Mulching is a preferred practice when there is inadequate soil cover and poor erosion control. A layer of mulch is considered to be a good physical barrier, increasing the boundary layer resistance at the soil surface (Anandacumaraswamy 1988), hence reducing loss of soil moisture through evaporation. Mulches recommended for tea include Guatemala grass (*Tripsicum laxum*) or Mana grass (*Cymbopogan confertiflorus* (Basnayake 1985). Manipura *et al.* (1969) have measured soil erosional losses of 40 tonnes ha^{-1} on cleared, weeded plots, and 0.07 ha^{-1} for mulched plots. Studies conducted by Manipura (1972) and Krishnarajah (1985) showed the importance of ground cover in reducing soil loss by mulching. 'Thatching', especially in young tea mainly protects water in tea plantations. The yield responses of tea during the wet

season are determined by the degree of stress during the previous dry period (Anandacumaraswamy 1988), and mulches are essential for reducing such stress and for effective water use efficiency (Shaxol and Hall 1968; Othieno *et al*. 1980).

In Sri Lanka, tea is grown as a plantation crop in the central and western parts of the country where annual rainfall varies from 1500 mm to 5000 mm. The tea-growing lands have undulating to mountainous and steeply dissected terrains (Panabokke 1970). The more intense rain (>25 mm hr^{-1}) in such sloping lands makes the soil vulnerable to erosion (Hasello and Sikurajapathy 1965; Sandanam and Rajasingham 1982; Basnayake 1985). Incorporation of organic material such as compost, mulches, and recycled tea prunings is practised in tea fields to help reduce erosion, conserve soil moisture, suppress weed growth, and sustain the soil fertility by improving other physical, chemical, and biological properties of soil. Earlier studies have been conducted to investigate the effect of mulching materials such as refuse tea, mana grass, and compost in the tea lands of Sri Lanka (Anon. 1988, 1991). In some studies, locally available material such as sawdust, coir dust, and paddy straw have also been used in fields of young tea (Anandacumaraswamy 1988), mainly to determine their effects on soil moisture conditions rather than on soil fertility.

Large amounts of polyphenol-rich residues are returned to the soil by leaf-fall and prunings in tea lands (Sivapalan *et al*. 1983). Observations made over long periods have indicated that these polyphenol-rich residues do not inhibit nitrification in Sri Lankan tea lands (Sandanam *et al*. 1978; Sivapalan *et al*. 1985). On the other hand, however Vallis and Jones (1973) showed that, although both legume species *Desmodium intortum* and *Phaseolus atropurpureus* have similar nitrogen contents, nitrogen mineralisation of

Desmodium intortum was less because of its higher polyphenol content. Olson and Reiners (1983) and Baldwin *et al*. (1983), investigating possible factors inhibiting nitrification in a sub-alpine balsam fir forest, suggested that tannins and phenolics specifically inhibited nitrification. The inhibition was thought to be a result of the higher polyphenol content in the decomposing plant residue leading to the formation of nitrogen-rich humic matter (in the alkali-extractable fraction); nitrogen is thus locked up in the humus fraction and less readily mineralised (Sivapalan, 1982). The studies of Olson and Reiners (1983) and Baldwin *et al*. (1983), indicated that tannins and other protein-binding phenolics which are relatively insoluble polyphenols, are potent inhibitors of nitrification, while water soluble polyphenolic compounds are less reactive. This provides an explanation for the absence of nitrification inhibition in soil by water soluble polyphenol-rich tea residues (Sivapalan *et al.*1985).

Sandanam *et al.* (1978), Sivapalan (1982) and Sivapalan *et al.* 1983 showed that mulches of Guatemala grass (*Tripsicum laxum*) and Mana grass (*Cymbopogon confertiflorus*), which are currently used for rehabilitation of tea lands in Sri Lanka, have high C/N ratios (30 and 40, respectively) and therefore low nitrification rates when compared to mulches of the legume Dadap (*Erythrina lithosperma*) which has a high soluble nitrogen content. Addition of nitrogen-poor residues to the soil resulted in nitrogen immobilisation as a consequence of assimilation of nitrogen by heterotrophic organisms (Sivapalan 1982).

The presence of autotrophic nitrifiers and the evidence of their activity in the soils suggest that nitrification, and the subsequent denitrification or leaching of nitrate, may be partly responsible for losses of fertiliser nitrogen even in the acid tea soils of Sri

Lanka (Walker and Wickramasinghe 1979). In a Bangladesh soil, Ishaque and Cornfield (1974) showed that the nitrification rate of applied organic nitrogen by heterotrophic microbes was considerably slower than that of ammonium-nitrogen by autotrophis microbes. Eylar and Schmidt (1959) found that strains of *Penicillium, Cephalosporium,* and several strains of *Aspergillus flavus* and *Pseudomonas,* all derived from soils, were able to produce nitrate from nitrogen fertilisers applied to the soil.

1.2.5.3 *Rock dust soil amendments*

Practically no research has been conducted on the benefits of the use of alternative liming materials on tea production. In the present study, both glasshouse and field trials were conducted to examine the effects of different mulch materials and pH amendments on soil chemical and biological properties, and on tea production.

"Minplus", a finely crushed basaltic rock (all particles finer than 0.25 mm), is a soil conditioner, which has been used in Australia as a lime substitute on sugarcane and banana farms (Campe 1993; Gillman *et al*. 2001). Similar materials have also been used in USA, Europe, South East Asia and Middle East (Coventry *et al*. 2001). Minplus consists predominantly of calcium, magnesium, and potassium silicates and provides a range of macro- and micro-nutrient elements. Minplus is able to reduce soil acidity by exchanging its calcium and magnesium cations for hydrogen ions in acidic soils (McSkimming 1998). It can also raise the negative charge of the soil thereby increasing its cation exchange capacity (Gillman *et al*. 2001, 2002). Hence Minplus improves the ability of soil to hold nutrient ions supplied from the breakdown of organic matter (humus) or mineral particles in the soil, or those derived from fertilisers applied to the soil (Coventry *et al*. 2001). Use of Minplus as a soil amendment has also resulted in a significant improvement in crop production (Edwards 1993; Coventry *et al*. 2001). It

has the added advantage of supplying low levels of sulphur, iron, cobalt, copper, zinc and other trace elements to the soil. The application of Minplus to highly weathered soils can eliminate the problem of phosphate fixation where phosphorus-bearing anions are chemically bound to soil particle surfaces (Coventry *et al* 2001). Minplus is not locally produced in Sri Lanka and the product used in the present study was supplied by Pacific Mineral Developments Pty. Ltd., PO Box 594, Innisfail, Queensland, Australia.

1.2.6 Pesticides

Pesticides are chemicals which are classified according to their target population (eg. herbicides, fungicides, insecticides). These chemicals are ultimately broken down and detoxified by the soil microbial biomass (Killham 1994).

Perucci and Scarponi (1994) found that the herbicide Imazethapyr (an amidazolinone derivative), used to control a wide spectrum of broad leaf weeds and grasses, when applied at the recommended field rate (50 g a.i. ha^{-1}) had no effect on the soil's microbial biomass. At a rate 100-fold higher, however, Imazethapyr had an adverse effect on the microbial biomass. Similar results were obtained by Voos and Groffman (1947) for the microbial biomass breakdown of the herbicides 2,4-D and Dicamba. In a Sri Lankan tea soil, herbicides such as Paraquat, Diuron, and Glyphosate were tested both at the recommended and 10-fold higher levels and were found to have no adverse effect on microbially-mediated urea hydrolysis (Wimaladasa and Wickramasinghe 1986).

Generally, pesticides which are the most mobile in the soil and plant environment tend to have most non-target effects. Systemic fungicides, which are readily translocated by the

plant, may affect non-target soil fungi in the rhizosphere as well as the targeted fungal disease. Some systemic plant growth regulators are also strongly fungicidal and may, also, adversely affect the mycorrhizal symbiosis as well as the free-living soil fungi in the rhizosphere (Killham 1994). Application of Captan (a non-systemic, non-selective fungicide) is known to have detrimental effects on soil rhizobial populations (Killham 1994).

Soil fumigants are often used in nurseries in order to prevent pathogenic attack on seedlings. In general, these fumigants temporarily suppress most of the soil microbial and animal communities, but the soil microbes tend to recover quickly (Killham 1994).

General toxins such as soil fumigants, nematocides, and fungicides more often cause non-target effects, whereas most insecticides and herbicides (under normal application rates) tend to have much fewer non-target effects (Killham 1994). Pesticides however, readily kill some nematodes, fungi, and nitrifying bacteria, whereas spore-forming bacteria and some actinomycetes are more robust (Killham 1994).

1.2.7 Increasing microbial activity and promoting soil quality

The fertility of soil may be defined as the ability of soil to provide all essential plant nutrients in available forms and adequate amounts for plant growth (Tamhane *et al.* 1970). The soil should be kept in a fertile condition if high yields are to be produced. Thus good management practices which promote optimal soil fertility and availability of water also lead to high soil productivity (Tamhane *et al.* 1970).

Conversion of nutrients to plant available forms is mediated by the micro-organisms in the soil (Alexander 1977; Gray and Williams 1971), and the subsequent death and decay of these organisms, results in the release of inorganic and organic ions in a plant-available form (Paul and Clark 1989; Killham 1994).

Soil quality is an indication of the capacity of a soil to function within natural ecosystem boundaries, to sustain biological productivity, maintain environmental quality, and promote plant and animal health (Doran and Perkin 1994; Karlen *et al.*1997). Soil quality is sensitive to the different types of land use management such as conversion of virgin land to cultivation (Dalal and Mayer 1987; Doran *et a.l* 1994 a), crop rotation (Mc Gill *et al.* 1986; Doran *et al.* 1994 b), land restoration (Staben *et al.* 1997), heavy metal contamination (Brookes and McGrath 1984), and pesticide residues (Alexander, 1977).

Soil physical indicators of soil quality include how well a soil accepts, retains, and transmits water to crops, soil structure (especially pore sizes and distribution), aggregate stability, saturated hydraulic conductivity, and inter-particle bonding. Soil organic matter content is important to soil quality and its contribution to soil structure and the reservoir of nutrients. Other chemical properties important in defining soil quality might include ionic exchange capacity, pH, carbon content, and water adsorption capacity. Biological indicators of soil quality include the soil microbial biomass and / or respiration rates, mycorrhizal associations, nematode communities, enzymes, and fatty-acid profiles (Karlen *et al.*1997).

The soil microbial biomass has been suggested as an important biological indicator of soil health (Pankhurst 1997; Sparling 1992, 1997) and soil quality (Jordan *et al.* 1994;

Karlen *et al.* 1997) for a given soil and cultural practice. Microbial biomass does not provide threshold values, however, against which soil quality can be evaluated (Dalal 1998).

Applications of microbial biomass as a management tool have been limited because analytically soil microbial biomass needs to have measurements of its three functions simultaneously: source size, sink size, and turnover rate for carbon, nitrogen, phosphorus, and sulfur (Dalal 1998). Some of the laboratory measurements of microbial biomass are expensive, time consuming, and several methods (e.g. chloroform fumigation incubation, chloroform fumigation extraction, surface-induced respiration, adenosine triphosphate ATP analysis) may be required to estimate the size or activity of the microbial populations (Paul and Clark 1989). Thus, there are some limitations in the evaluation of the functions of soil microbial biomass and its use as an indicator of soil quality (Dalal 1998). According to Dalal (1998), the capacity of a soil to protect the microbial biomass, and how it relates to the optimum organic matter content, are unresolved questions.

On the other hand, however, a soil can not be considered as a ‘quality soil’ while it contains soil-borne pathogens (Cook 1994). By eliminating an inoculum of soil-borne plant pathogens, Cook (1994) found roots became healthier and took up more nutrients as the crop flourished. Root diseases are indirectly responsible for the decline of organic matter by reducing crop yield and the return of crop residues to the soil. Subsequent tillage of infested crop residues similarly accelerates the decline of organic matter (Cook 1994).

Soil quality can be expressed two distinct ways (Karlen *et al*. 1997): as an inherent characteristic of a soil; or as the overall condition or health of soil which assumes that if a soil is functioning at full potential for a specific land use (best management practices) it has excellent quality; whereas, if a soil is functioning well below its potential, it can be concluded to have poor quality (Karlen *et al*. 1997).

In the present study, the focus was to improve soil quality by using cultural techniques that are appropriate for tea lands.

1.2.8 Conclusions

The soil treatments applied in the present project involve organic and inorganic soil amendments. Organic amendments (organic mulches) directly involve soil organic matter dynamics by priming the microbial activity. Inorganic amendments such as dolomite and Minplus rock dust manipulate the soil acidity to offer optimal conditions for microbial activity, which also helps to sustain soil fertility.

Crop residues can have a significant effect on soil organic matter content, microbial biomass levels, decomposition rates and nutrient availability. Similarly, crop residues protect soils from wind and water erosion, especially in sloping lands.

The critical factor in ecosystem stability is the interaction of soil organic matter with micro-organisms, soil particles, nutrients, abiotic factors, and plants. The loss of soil organic matter content affects both soils and plant growth (Smith *et al*. 1993). Therefore, more efficient nutrient cycling processes could be created by manipulation of the soil organic matter.

Disturbances like deforestation and agricultural production cause biological, chemical, and physical instability in the soil because of massive carbon losses and altered nutrient cycling. Therefore, research needs to focus on the quantification of aspects of the soil system relative to its health and performance. Changes in soil organism diversity and functioning will help to maintain the stability of disturbed and regenerating ecosystems.

1.3 THE TEA PLANT

Throughout the world, tea is the cheapest, most readily available beverage next to water (Amarakoon, 2004). The tea plant belongs to the genus *Camellia* in the family Theaceae (Willson, 1999). Tea is the most valuable plantation crop in the world and is the highest net foreign exchange earner for Sri Lanka. The first commercial planting in Sri Lanka was made in 1867 by James Taylor (Nathaniel 1986). In 2004, tea production had exceeded 308 million kg of which more than 290 million kg was exported by Sri Lanka. The foreign earning generated was 53,133 million Rupees (about $ AUD 1,003 million) (Anon. 2004 a).

Tea is produced by cultivars of *Camellia sinensis* which is a big shrub, growing to 6 – 9 m with numerous vegetative stems arising from the base. Leaves are erect, small, 1.5-1.4 cm long and 1.0-2.5 cm wide, leathery, with a dark green colour. It is a free growing plant and can reach a height of 9 m if allowed to grow unrestricted. Tea has been domesticated into a bush 1 m tall in commercial production systems. The economic portion of the bush is the terminal bud of the young shoots and the adjacent two-three succulent immature leaves.

Tea is propagated either from seeds or by vegetative means. In seed tea, a mixture of characters may be seen whereas vegetatively propagated tea has characters identical to the mother plant. Plants produced vegetatively from a single parent plant are referred to as cultivars, and may show large variability in yield under different climate or soil conditions. Of 200 approved cultivars, only about 40 have been used widely in Sri Lanka, and only 10-15 are now popularly used (Anandappa 1986). Over 95% of the replanted clones represent the TRI 2020 series with the remainder represented by estate clones. Tea is grown in Sri Lanka under different agro-climatic zones, with different mean temperatures, rainfall distributions, and pest and disease problems; hence specific clones are suited for planting in particular areas; genotype and environment interactions occur within the clones (Anandappa 1986).

Tea plants are raised in a nursery either from single node cuttings or from seeds in polythene sleeves filled with soil. After 9 - 10 months following their establishment in the nursery, the seedlings (20-30 cm high) are planted in the field at the onset of the monsoonal rain; the plantings are in rows 120 cm apart along the contour and at 60 cm spacings along the rows. Soon after planting, the exposed soil is mulched with grass loppings from the inter-rows to conserve soil and moisture. After about 8-12 weeks the plant is cut at a height of 30 cm to encourage lateral branching. After another 3 months the plant is cut again at 45 cm height in order to develop more lateral branches. The tea is known as ‘young tea’ for the first two years after planting. During this period, the soil is exposed; therefore, mulch is applied at 3-4 month intervals.

After two years, commercial tea harvesting commences and continues for another 3 years. This growth phase is called the immature phase and, again, the soil remains

incompletely covered. After 5 years the tea is pruned at a height of 45 cm, and tea plant litter is left on the soil surface. Three months after pruning, new shoots grow to a height of 60-70 cm. The shoots are then cut at a height of 55-60 cm leaving 4-5 leaves on the branches. After that, tea leaves are regularly harvested by hand plucking of the terminal bud and adjacent leaves at 5-9 day intervals, depending on the growth rate. This phase is called 'mature phase' which lasts for another 4-5 years depending on growth rate. After that, the tea is pruned again and the cycle continues. In general, individual tea plants have remained in production in Sri Lanka for as long as 100 -150 years (Anandacumaraswamy *et al*. 2003).

The yield of clonal tea in the higher elevations of Sri Lanka is around 2500 kg ha^{-1} $year^{-1}$of made tea (in a fresh crop, 9 kg of harvested tea leaves turns into 2 kg of made tea), while in the lower elevations, yields of 5000-6000 kg ha^{-1} $year^{-1}$ of made tea are common (Kulasegaram 1986).

1.4 SCOPE OF THIS THESIS

This thesis describes the effects of a range of soil mulches with different C/N ratios and lignin contents, and two soil acidity or pH modifiers (dolomite and Minplus rock dust) on the biological, physical, and chemical properties of tea soils in Australia and Sri Lanka. It also attempts to apply these results to the development of new management strategies, which may promote greater productivity and enhanced sustainability of tea soils in Sri Lanka.

The research project was carried out in 2 stages:

- Nursery trial (under greenhouse conditions) at James Cook University, Townsville, Australia, and
- Field trials implementing the results of the pot trial on young tea, and established, pruned tea at the St Coombs Estate under the auspices of the Tea Research Institute, Talawakelle, Sri Lanka.

Seedlings of *Camellia assamica* were used for a nursery trial at James Cook University. *C. sinensis* was used in the Sri Lanka field trials. TRI 4071 clone was used for a young tea trial, and the TRI 2025 clone was used for a pruned tea trial. Cultural practices such as land preparation, forking, fertilizing, tipping, pruning, weeding, and harvesting were carried out using the recommendations of Tea Research Institute, Talawakelle, Sri Lanka.

In the following chapter, the nursery trial is discussed. The nature and results of the young tea experiments are presented in chapter 3, and parallel studies on the established, mature tea are dealt with in chapter 4. Some general comments are made in chapter 5 where guidelines for the effective management of tea soils are provided. In the final chapter, conclusions from the study are reiterated and some directions for future research are offered. Most of the primary data, on which the results depend, are presented in a set of appendices on a CD-ROM at the back of the thesis.

CHAPTER 2

NURSERY TRIAL AT JAMES COOK UNIVERSITY, AUSTRALIA

2.1 BACKGROUND

Each year about 14,000 tonnes of tea is brewed in Australia (Burdon 2000). Until the 1980s, overseas producers met the entire tea demand of Australia, but by the late 1990s, 1840 tonnes at a value of $AUD 13.3 million, had been exported. Australia's total annual tea imports of 17,556 tonnes were worth $ 84.6 m (Anon 1998).

Despite a number of failed early attempts to establish an Australian tea industry, commercial tea cultivation in North Queensland is only about 20 years old (Burdon 2000). While tea plantations in Australia are relatively young and small, they are free from pests and diseases, but they require regular cultural practices such as pruning, fertilizing, and weeding. Fresh tea leaves are harvested every 20-35 days by mechanical harvester with green leaf yields of up to 15 tonnes ha^{-1} per harvest (Burdon 2000).

Australia has two tea-growing regions, one in north eastern New South Wales, and a bigger area on the coastal lowlands near Innisfail and on the adjacent Atherton Tableland in North Queensland. Together they produce about 1600 tonnes annually. Both have regular and well distributed annual rainfall (2000 - 3000 mm $annum^{-1}$ in North Queensland) and high humidity ideal for tea cultivation. The soils are acidic Oxisols with relatively low fertility.

The primary objective of the experiments reported in this chapter was to improve soil fertility by determining the conditions that maximize soil microbial biomass in an Australian tea soil from Innisfail. Such information will provide a basis for improving the ratio of 'beneficial' to pest micro-organisms, the soil nutrient supply, and tea growth. In the present study, organic and inorganic soil amendments were compared in a factorial trial to observe their effects on soil chemical and biological properties and on tea growth.

2.2 AIMS OF THE STUDY

The experiments discussed in this chapter aimed to examine the effects of mulching materials with varying C/N ratios and lignin contents, in combination with soil pH amendments and rainforest inoculum, on the biological and chemical properties of soil with or without tea plants. A forest soil inoculum was used to introduce beneficial micro-organisms into the tea soils in the expectation that it would increase the soil microbial activity. The nursery trial with or without plants was used to investigate the influence of tea plants on soil microbial activity.

Hypothesis to be tested was that soil and cultural treatments change the chemical and biological properties of the soil and thereby enhance the growth of young tea plants.

2.3 MATERIALS AND METHODS

A pot trial was conducted on soils in a shadehouse, at James Cook University, Townsville (Latitude 19° 20' S; Longitude 146° 46; 27 m above sea level), with or without tea seedlings, for a period of 28 weeks commencing on 19 March 1999.

2.3.1 Soil

The soil used in the trial was collected from the Nerada Tea Plantation, Innisfail, North Queensland (latitude 17° 33' S; longitude 145° 53' E; altitude 100 m above sea level). The soil has formed on metamorphic rocks, belongs to the Galmara Series (Murtha 1986), and is classified as a Tropeptic Haplorthox (Soil Survey Staff 1975). It has a granular, reddish brown topsoil (10 – 17 cm thick) of loam to clay loam texture and pH of 4.7 - 4.9 that overlies a red to reddish yellow, well-structured, clay-rich subsoil with pH of 4.7 - 5.0. It occurs in the humid tropical lowlands of North Queensland where the mean annual rainfall is approximately 3500 mm, with prominent summer dominance, and a mild, relatively dry winter (Murtha 1986).

2.3.2 Plant Material

Tea seedlings (six months old) were collected from the Nucifora Tea Plantation, Innisfail, where Assam seed tea (*Camellia assamica*) was planted on a commercial basis in 1986. An ammonium-based fertilizer mixture (N 320, P 30, K 140 kg ha^{-1} yr^{-1}) had been used and the herbicide Simazine had ever been applied (S. Nucifora, 1999, personal communication). The seedlings were transported to Townsville, 250 km to the south, in an insulated box and were planted directly into soils prepared for the experimental treatments that are described in the following section.

2.3.3 Experimental treatments

The treatments consisted of a combination of four mulching materials, three pH amendments, with or without rainforest inoculum, and with or without tea plants (Table 2.1). There were 48 treatment combinations in a factorial design and each treatment was replicated five times requiring a total of 240 pots of 18 cm diameter for the trial.

The experiment included a control treatment that received no mulch, no soil pH modifier, and no rainforest soil inoculum.

Table 2.1 Experimental treatments applied to 5 replicates of pots of Nerada Tea Plantation topsoil (0-15 cm depth) in two randomized factorial blocks. The experimental treatments were duplicated in pots with and without tea seedlings, bringing the total number of treatments to 48, each replicated 5 times; the total number of pots used in the trial was 240.

No	Treatment code	Mulch	Soil pH modifier	**Forest soil inoculum** used (+), not used (-)
1	M0 pH0 In -	Zero mulch	Not treated	In -
2	M0 pH0 In +	Zero mulch	Not treated	In +
3	M1 pH0 In-	Grass mulch	Not treated	In -
4	M1 pH0 In +	Grass mulch	Not treated	In +
5	M2 pH0 In -	Legume mulch	Not treated	In -
6	M2 pH0 In +	Legume mulch	Not treated	In +
7	M3 pH0 In-	Tea mulch	Not treated	In -
8	M3 pH0 In +	Tea mulch	Not treated	In +
9	M0 pH1 In -	Zero mulch	Dolomite	In -
10	M0 pH1 In +	Zero mulch	Dolomite	In +
11	M1 pH1 In -	Grass mulch	Dolomite	In -
12	M1 pH1 In +	Grass mulch	Dolomite	In +
13	M2 pH1 In -	Legume mulch	Dolomite	In -
14	M2 pH1 In +	Legume mulch	Dolomite	In +
15	M3 pH1 In -	Tea mulch	Dolomite	In -
16	M3 pH1 In +	Tea mulch	Dolomite	In +
17	M0 pH2 In -	Zero mulch	Minplus	In -
18	M0 pH2 In +	Zero mulch	Minplus	In +
19	M1 pH2 In -	Grass mulch	Minplus	In -
20	M1 pH2 In +	Grass mulch	Minplus	In +
21	M2 pH2 In -	Legume mulch	Minplus	In -
22	M2 pH2 In +	Legume mulch	Minplus	In +
23	M3 pH2 In -	Tea mulch	Minplus	In -
24	M3 pH2 In +	Tea mulch	Minplus	In +

The mulching materials used were leaves of:

- the grass *Brachiaria decumbens*,
- the shrub legume *Calliandra calothyrsus*,
- tea *Camellia assamica*.

Brachiaria decumbens was included because it is a grass with widespread occurrence in the humid tropics. The legume *Calliandra calothyrsus* was included because its leaves contain relatively high amounts of nitrogen. The tea mulch consisted of a mixture of tea leaves and twigs less than 3 mm diameter that had been collected from underneath an abandoned part of the Nerada Tea Plantation, near Innisfail. The tea litter was included because it is naturally added to soil in a tea plantation by leaf fall. The mulch materials were dried at 40 ^{o}C, crushed using a grinder (2.0 mm mesh), and applied to the pots at a rate of 28 g pot^{-1} which was equivalent to the recommended rate of 35 tonnes ha^{-1} fresh weight of mulch material per hectare; the depth of the mulch material was around 1 cm in each pot. The chemical compositions of the mulch materials are presented in Table 2.2.

Table 2.2 Chemical composition of oven dried (85 ^{o}C) mulch materials that were applied to the Nursery Trial at James Cook University.

Mulch material	**N %**	**C %**	**C/N ratio**	**P %**	**K %**	**Lignin %**	**Polyphenol %**
Grass: *Brachiaria decumbens*	1.9	35.3	18.6	0.17	2.13	7.5 (Monteiro *et al.* 2002)	2.2 (Monteiro *et al.* 2002)
Legume: *Calliandra calothyrsus*	3.9	36.1	9.2	0.19	1.87	17.0 (De Costa *et al.* 2001)	18.2 (De Costa *et al.* 2001)
Tea litter: *Camellia assamica*	3.5	40.5	11.6	0.20	1.50	11.1 (Sivapalan 1982)	19.0 (Sivapalan 1982)

The amount of mulch, soil pH amendments, and rainforest soil inoculum used are shown in Table 2.3.

The three soil pH modifiers used were dolomite applied at an equivalent of 2500 kg ha^{-1}, Minplus at 2500 kg ha^{-1}, and an untreated control. Dolomite was used in this experiment because it is commonly used as a pH modifier in tea soils of Sri Lanka and has an acid neutralizing capacity of 119. The other pH modifier used was Minplus, containing high levels of calcium, magnesium, and potassium silicates, with a slightly lower acid neutralizing capacity (Coventry *et al*. 2001).

The forest soil inoculum, containing a naturally high population of beneficial micro-organisms, was applied at a rate of 200 g forest soil to 1500 g of tea soil, and a control with no inoculum was also included in the treatment (Table 2.3).

Table 2.3. Treatment codes. The pots used had a diameter at 18 cm.

Code	Treatment	Rate of application
M0	No mulch	0
M1	Grass (*Brachiaria decumbens*)	35,000 kg ha^{-1} = 28g pot^{-1}
M2	Legume (*Calliandra calothyrsus*)	35,000 kg ha^{-1} = 28g pot^{-1}
M3	Tea (*Camellia assamica*)	35,000 kg ha^{-1} = 28g pot^{-1}
PH0	No pH modifier	0
PH1	Dolomite	2500 kg ha^{-1} = 2 g pot^{-1}
PH2	Minplus	2500 kg ha^{-1} = 2 g pot^{-1}
In-	No treatment	0
In+	Rainforest soil inoculum	200 g pot^{-1}

2.3.4 The pot experiment

Bulk samples of the surface soil (0-15 cm depth) from the Nerada Tea plantation were collected and passed through a 2 mm mesh sieve. The soil was treated with the nematocide Phenamiphos (5% granular formulation; trade name "Nemacur") at a rate of 1 g pot^{-1} to eliminate any influence of nematodes. Appropriate masses of nematocide and inorganic soil amendments (dolomite, Minplus, and forest soil inoculum) were mixed in bulk with the tea soil in a mechanical cement mixer before being weighed into the pots.

Two sets of 120 plastic pots (18 cm diameter ; 19 cm deep) were each filled with 1500 g of soil and the first set of pots (Block 1) was planted with nursery tea plants (established seedlings six months old), and the second set of pots (Block 2) kept without tea plants.

Because of a shortage of soil, pots without nursery plants were filled with only 1 kg of the appropriate soil mixture.

The three mulching materials were dried at 40 ^{o}C for 24 hours, ground to pass through a 2 mm mesh then applied at the rate of 28 g to each pot (equivalent to 35 tonnes ha^{-1}). The pots were maintained at 50-80% of field capacity by weighing the pots and adding appropriately masses of water regularly during the 28 week experiment.

A basal application of fertilizer mixture (T65) for the nursery tea was added to all pots one month after planting of tea seedlings according to TRI recommendations (Wickremasinghe and Krishnapillai 1986). One seventh of the total N-fertilizer (ammonium sulphate) of a basal fertilizer mixture was added every month throughout the experimental period of 28 weeks. The T65 mixture was dissolved (35 g in 5 L of water) and applied to the foliage of 120 plants at monthly intervals, and washed off with clean water after each application.

The composition of the T65 fertilizer is given below:

15 parts ammonium sulphate --------- 20.6% N

20 parts ammonium phosphate --------- 20.0% N and 35% $P_2 O_5$

15 parts potassium sulphate---------- 48.8% K_2O

15 parts magnesium sulphate ---------- 16.0% MgO

The mixture contains approximately 10.4% N, 10.6 % P_2O_5, 1.1% K_2O and 3.7% MgO.

Maximum and minimum temperatures and humidities were recorded daily at 8.30 am and 3.30 pm (local time) throughout the duration of the shadehouse experiment.

2.3.5 Sample collection and analysis

Soil samples were taken from pots on weeks 0, 10, 20, and 28 from the commencement of the experiment. At time zero, the analysis of key chemical and biochemical characteristics of bulk samples of the tea soils used to fill the pots and the forest soil that supplied the inoculum were carried out. They included organic carbon, pH, total nitrogen, total phosphorus, plant available phosphorus, cation exchange capacity, and microbial biomass nitrogen. In order to obtain some information on the functional group composition of the microbial biomass, FAME (Fatty Acid Methyl Ester) analyses were carried out by Dr Clive Pankhurst, CSIRO Division of Land and Water, Adelaide.

Plant growth measurements such as plant diameter 3 cm above the soil surface, and the height of plant, number of leaves, length of the leaves, leaf area were recorded fortnightly throughout the experiment. A destructive sampling was carried out at the end of the 28th week after the application of treatments, and root and shoot masses, and total biomass (dry weights of plant at 85 ^{o}C) were determined. Leaf areas of the tea plants were measured by the leaf area grid method (Pethiyagoda and Rajendran 1965). Soil chemical characteristics, together with FAME analyses and soil microbial biomass nitrogen, were also measured at the end of the experiment.

All of the chemical analyses of soil and plant materials were made within the School of Marine and Tropical Biology, James Cook University. Soil pH was determined using a soil-to-water ratio of 1: 5 (Rayment and Higginson 1992). Total nitrogen content of the soil and mulch material, and total phosphorus content of the soil were determined using the salicylate hypochlorite method and single solution method respectively (Anderson and Ingram 1989). Plant-available soil phosphorus was determined by a weak acid

extraction method (Rayment and Higginson 1992). Soil and plant organic carbon contents were analysed by a modified Walkley Black method (Rayment and Higginson 1992). Cation exchange capacity was determined by the compulsive exchange method, with no pre-treatment for soluble salts, as described by Gillman and Sumpter (1986**).**

Total leaf nitrogen contents of the organic mulches (*Brachiaria decumbens*, *Calliandra calothyrsus,* and tea mulch) were determined by the salicylate hypochlorite method, and total phosphorus determined by the single solution method (Anderson and Ingram 1989).

2.3.5.1 Microbial biomass determinations

Thin wall, stainless steel tubing 14 cm long by 1.9 cm diameter was used to collect two core samples (each of 39.7 ml volume and containing approximately 80 g of soil) from the soil of each pot at each sampling time (weeks 0, 10, 20, and 28) for microbial biomass analysis. Soil microbial biomass nitrogen was measured using the chloroform fumigation – extraction method (Amato and Ladd 1988) whereby the amount of ninhydrin reactive nitrogen contained in extracts of fumigated soil is determined and compared with that of non-fumigated, similarly analysed samples (Appendix 1).

Microbial biomass carbon was determined by a combination of chloroform fumigation – extraction method and a modified Walkley Black method (Sparling *et al*. 1990); see Appendix 2.

Soils were passed through a 2 mm sieve and moisture content was adjusted to 40% of field capacity just prior to being fumigated under an atmosphere of chloroform for 10

days. This was followed by an extraction of the fumigated soil with 0.5 M K_2SO_4, and reaction of the extract with ninhydrin. Because of the highly acidic soils used in this study, a conversion factor of 18.7 was calculated using 20 random samples of the bulk tea soils used instead of factor 21 (Sparling *et al*. 1990) to convert the ninhydrin-reactive N (microbial biomass nitrogen; MBN) to microbial biomass carbon (MBC). In the present study, microbial biomass nitrogen was analysed using the initial, 1st, 2nd, and 3rd soil samples (0, 10, 20, and 28 weeks after application of treatments) and the final values of microbial biomass nitrogen converted to microbial biomass carbon using the following equation (Joergensen, 1996).

Microbial biomass carbon = 18.7 x microbial biomass nitrogen

This technique for estimating microbial biomass is simpler than direct microscopy and also permits measurement of biomass carbon and nitrogen incorporated into the soil organisms (Paul and Clark 1989).

2.3.5.2 *FAME (Fatty Acid Methyl Ester) analysis*

A molecular technique, Fatty Acid Methyl Ester (FAME) analysis, has been used to investigate the functional group composition of the soil microbial communities. This technique uses marker fatty acids from live cells, dead cells, humic materials and plant and root exudates to provide information on the functional group composition of the soil biomass (Ibekwe and Kennedy 1999).

The marker fatty acids in lipids of various micro-organisms are different and specific for them. Specific fatty acids especially those in phospholipids and lipopolysaccharides of cell walls have been found to be biomarkers for identification of micro-organisms (Zelles *et al*. 1992). This has been made possible by FAME analysis which identifies

signature fatty acids that can be readily volatilized following methylation, and then analysed by gas chromatography (Moss *et al*. 1980; Moss 1981; Vestal and White 1989).

Generally, the odd numbered, branched-chain fatty acids are produced by Gram-positive bacteria, while the even numbered straight-chain and cyclopropyl fatty acids are from Gram-negative bacteria. Gram-positive bacteria are represented by 15:0, a15:0, i15:0, i16:0, a17:0, i17:0 peaks in the FAME analysis, and Gram-negative bacteria by short chain hydroxy acids (10.03 OH and 12:0 OH) and cyclopropane acids (cy 17:0, cy 19:0) (Zogg *et al*. 1997; Ibekwe and Kenedy 1999; Pankhurst *et al*. 2001 a). The fungi fatty acid profile is 18:2 w6c (Zogg *et al*. 1997; Pankhurst *et al*. 2001 a) and mycorrhizal fungi fatty acid profile is 16:1 w5c (Olsson 1999). Most actinomycetes contain iso- and anteiso- fatty acids (Zelles *et al*. 1992). The fatty acid profile of eukaryotes is 12:0 (White 1983). Prokaryotic cells generally lack polyunsaturated fatty acids in their membranes (Zelles *et al*. 1992).

2.3.5.3 Statistical analysis

Principal component analysis (PCA) was used to analyse the multivariate data generated by the nursery trial since there are many independent variables operating simultaneously. The analysis is an ordination technique, used to display the relative positions of points in multivariate space in fewer dimensions, while retaining as much information as possible. Principal component analysis is an exploratory technique and allows investigation of patterns of variation and relationships among the points of an ordination plot. The most important test of the analysis is its interpretability. The original variables (such as soil parameters) can be correlated to the new axes. Vectors

(arrows) represent the orientation of the original variables within the new ordination plot; their lengths and directions reflect where these variables are important in the reduced space plot (McArdle 1999).

An extraction method was used in the data analysis to demonstrate the interaction effects of all variables. A Kaiser-Mayer-Olkin (KMO) measure of sampling and Bartlett's test was carried out to evaluate the overall accuracy. KMO is an index for comparing the magnitudes of the observed correct coefficient to the magnitude; KMO values of the partial correlation coefficients greater than 0.5 imply that the metrics are acceptable for the factor analysis. The minimum requirement for subjecting the data set to a principal component analysis was fulfilled in this trial since it had a Measure of Sampling Adequacy of more than 0.5 (Appendix 3).

Where probability levels were less than 5%, the null hypothesis that the dependent variables are not correlated was rejected, and the alternate hypothesis that the correlation matrices are identity matrices was accepted. Since the variables have been measured using different units in the principal component analysis performed on correlation matrices, the eigen values provide the variance of the scores of the observations on each of the new axes. The first component lying along the axis of the data will have the largest amount of associated variance and is known as the first principal component. The next largest is known as the second principal component, and so on.

2.4 RESULTS

2.4.1 Changes in soil parameters – JCU Nursery Trial

Initially, the principal component analysis was performed on the pooled data with or without plants. The results indicated that there was a distinct separation in the spread of data in soil parameters with or without plants (Appendix 4). Therefore, the rest of analysis was carried out separately both with or without tea plants. Subsequently, a factor analysis was performed to test the levels of the significance of the differences observed.

2.4.1.1 Principal component analysis

Soil organic carbon, soil pH, microbial biomass carbon, plant available phosphorus, soil total nitrogen, and cation exchange capacity were measured as soil parameters and the data were analysed by principal component analysis. In the present analysis, different mulch treatments were considered with each pH modifier. While preparing the data set, outliers and missing values were identified and were removed before analysis, where appropriate. In order to achieve the normal distributions required for standard statistical techniques, some of the data were transformed using square root, fourth root, and logarithm transformations; the transformed data have been analysed statistically and back-transformed data are discussed below, where appropriate.

Three components that described 75.1 % of variance of data set were extracted for the plant available phosphorus, soil total nitrogen, soil pH, microbial biomass carbon, soil organic carbon, and cation exchange capacity information. After performing the principal component analysis, a scatter plot in which the first and second principal

components (representing the largest fraction of the overall variability) were plotted on the vertical and horizontal axes respectively. Component 1 (37.9 %) included organic carbon, cation exchange capacity, and available phosphorus of the tea soils; component 2 (23.2 %) included soil pH, and microbial biomass carbon; component 3 included (14.0 %) total phosphorus of soil (Appendix 5).

To determine which of the soil parameters contributes most significantly to the variability of the soil, two criteria were used. One is Kaisers' criteria where variables with eigen values greater than 1 are considered as main contributory factors when number of variables are less than or equal to 20 (McArdle 1999). In this analysis, two eigen values were greater than 1 (Appendix 3), and the third eigen value, 0.9, were all considered to express the total variability of the data-set. This cut-off point is called Kaiser's criterion.

Another way of identifying the cut off point is by using a scree graph technique but it usually results in fewer acceptable components (McArdle 1999). The scree graph identifies the position where large eigen values end and small ones begin. This position is called the 'scree plot elbow'. In the present analysis (Appendix (3), a scree graph produced two components from the data, and up to the point of the scree plot elbow, 61.1 % of the variance was explained. It has been suggested that two components will give better reliance than more (McArdle 1999). The third component on the scatter graph of Appendix 3 showed a distinct pattern of variables (75.1 %), and therefore only the nature of the two components was considered further.

After performing the principal components analysis, a scatter plot was prepared with the first and the second principal components on the vertical and horizontal axis respectively for all the mulch treatments with pH modifiers (Fig. 2.1). In this scatter plot, each soil parameter is represented by a vector which is a set of co-ordinates defining a point in space relative to the set of axes. The length and direction of each vector reflects where the variables are important in the reduced space plot and gives a relative interpretation of the responses, with longer vectors representing larger responses than shorter vectors.

The pooled data with all pH modifiers for each mulching materials is presented in Figure 2.1a. However, for ease of interpretation, the principal component analysis graphs for each soil pH modifier with mulching treatments have been plotted separately (Figs 2.1b – 2.1d).

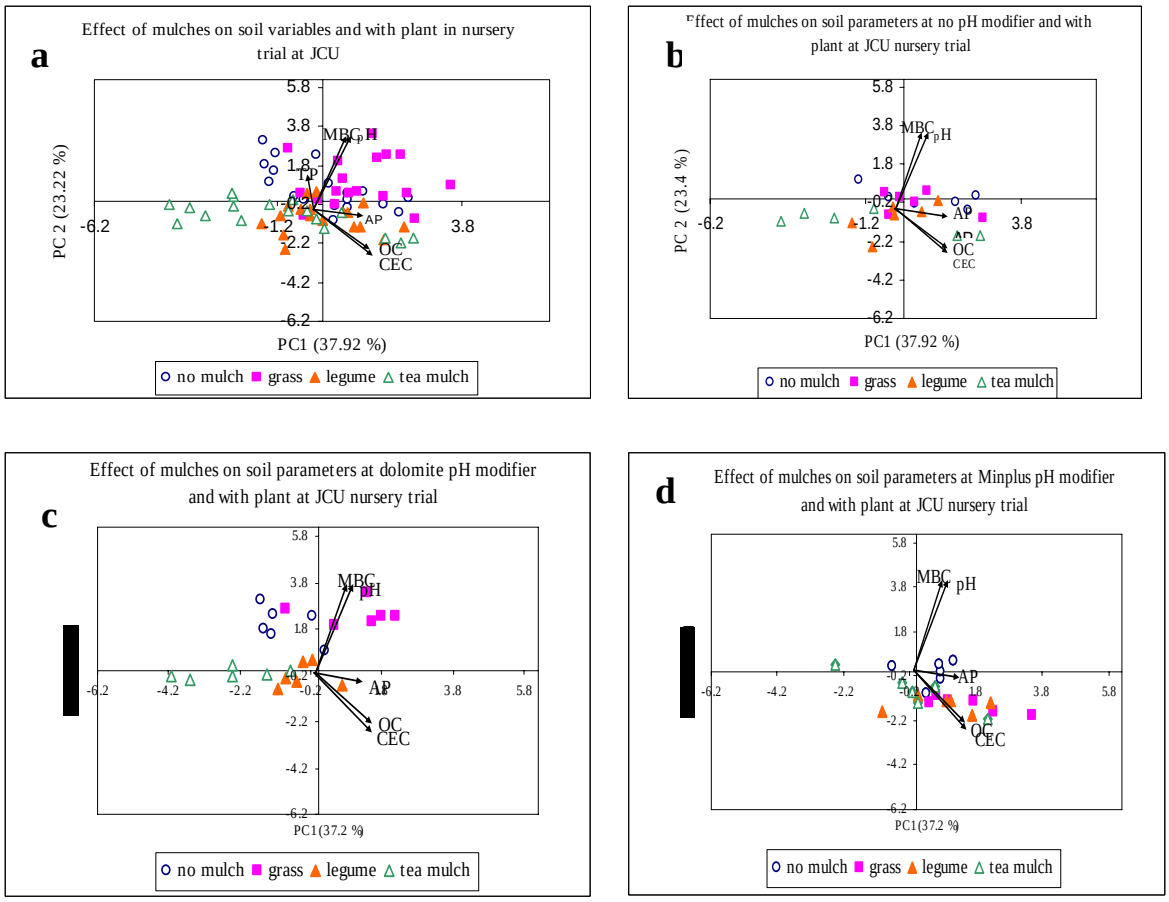


Figure 2.1 Effects of mulches on soil parameters in the James Cook University Nursery Trial with plants:

(a) The analysed data include all soil pH modifier treatments;
(b) The analysed data excludes all soil pH modifier treatments;
(c) The analysed data embraces dolomite as the soil pH modifier treatment,
(d) The analysed data embraces Minplus as the soil pH modifier treatment.

The grass mulch had a strong influence on soil microbial biomass carbon and pH and weak influence on plant available phosphorus compared to legume and tea mulches (Fig. 2.1 a), whereas the tea and legume mulches had greater influence on organic carbon and cation exchange capacity, than did the control (Fig. 2.1a).

In the absence of pH modifiers, the grass and tea mulches had a stronger influence on soil organic carbon and cation exchange capacity, and the legume and tea mulches reduced the microbial biomass carbon and pH (Fig. 2.1b). The tea mulch improved total phosphorus content of the soil (Appendix 5).

In the presence of dolomite, the grass mulch increased both the soil microbial biomass and pH than legume and tea mulches (Fig. 2.1c), and the tea mulch improved the total phosphorus content of the soil (Appendix 5).

In the presence of Minplus, the soils under the grass, legume, and tea mulches all produced strong positive responses to soil organic carbon, cation exchange capacity and plant available phosphorus parameters (Fig. 2.1d; Appendix 5).

Principal component analyses were also performed on the results of the experiment that excluded tea plants (Appendices 6 and 7) and they showed similar, but lower overall responses with shorter vectors for microbial biomass carbon and pH than for the soils that supported plant growth.

2.4.1.2 Factor Analysis: *JCU Nursery Trial*

Multivariate analysis (MANOVA; McArdle, 1999) showed that there were significant differences in soil pH, microbial biomass carbon, soil organic carbon, and cation exchange capacity of the soils produced by the mulch, pH, mulch x pH, and mulch x pH x inoculum treatments (LSD at $p < 0.05$ using F value and Wilk's Lambda test). The main treatment effects on soil parameters are presented in Table 2.4 for the trial with tea plants.

Table 2.4 Summary results of analyses of variance, Nursery Trials with or without plants at James Cook University, Australia, showing the effects of soil amendments on soil chemical, biological and plant growth parameters, 28 weeks after mulch emplacement. Where the soils without plants produced a result different from those with plants, the "without plants" data are shown in parentheses.

Treatments		Soil parameters							Plant parameters				
		OC	**PH**	**TN**	**TP**	**AP**	**CEC**	**MB-C**	**DM**	**Ht**	**SW**	**RW**	**TB**
Mulch	No mulch	-	-	-	-	-	-	-	-	-	-	-	-
		(-)	(-)	(-)	(-)	(-)	(-)	(-)					
	Grass	*	*	Ns	Ns	Ns	*	*	#	*	*	Ns	*
		(*)	(*)	Ns	Ns	(*)	(*)	(*)					
	Legume	Ns	#	Ns	Ns	#	Ns	#	Ns	Ns	Ns	Ns	Ns
		(*)	(#)	(#)	(Ns)	(Ns)	(*)	(*)					
	Tea	#	#	Ns	Ns	#	#	#	Ns	Ns	Ns	#	Ns
		(*)	(#)	(#)	(Ns)	(Ns)	(*)	(*)					
pH	No pH treatments	-	-	-	-	-	-	-	-	-	-	-	-
		(-)	(-)	(-)	(-)	(-)	(-)	(-)					
	Dolomite	Ns	*	Ns	*	Ns	Ns	*	Ns	#	#	#	#
		(*)	(*)	(Ns)	(Ns)	(#)	(*)	(*)					
	Minplus	*	*	Ns	*	Ns	*	Ns	Ns	Ns	Ns	Ns	Ns
		(*)	(Ns)	(Ns)	(Ns)	(#)	(*)	(Ns)					
Inoculum	Without Inoculum	-	-	-	-	-	-	-	-	-	-	-	-
		(-)	(-)	(-)	(-)	(-)	(-)	(-)					
	With Inoculum	Ns	Ns	Ns	Ns	#	Ns	*	Ns	Ns	Ns	Ns	Ns
		(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)					
Interaction	Mulch x pH	*	*	Ns	Ns	*	*	*	Ns	Ns	Ns	Ns	Ns
		(*)	(*)	(*)	(Ns)	(Ns)	(*)	(*)					
	Mulch x Inoculum	Ns	*	Ns	Ns	Ns	Ns	Ns	Ns	*	*	*	*
		(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)					
	pH x Inoculum	*	Ns	Ns	Ns	Ns	*	Ns	Ns	Ns	Ns	Ns	Ns
		(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)					
	Mulch x pH x Inoculum	Ns	*	Ns	Ns	Ns	Ns	*	Ns	Ns	Ns	Ns	Ns
		(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)	(Ns)					

OC: Soil organic carbon, pH: Soil pH, TN: Total nitrogen, TP: Total phosphorus, AP: Plant available phosphorus, CEC: Cation exchange capacity, MB-C: Microbial Carbon, DM: diameter, Ht: Height, LA: leaf area, SW: Shoot mass, RW: Root mass, TB: Total biomass of plant.

A significant increase ($p < 0.05$) over the relevant control is shown as: *; A significantly decrease ($p < 0.05$) below the control is shown as #, no significant difference from control ($p < 0.05$) is shown as Ns, control treatments are shown as - .

The parallel responses of the soil and plant growth parameters are also shown, in parentheses, from the trial without tea plants.

Table 2.4 shows the soil parameters that responded most strongly to the mulch, soil pH, and rainforest inoculum treatments under tea plants were: organic carbon, pH, CEC, and microbial biomass carbon (Table 2.4). Under grass mulch, soil parameters such as organic carbon, cation exchange capacity, soil pH, microbial biomass carbon were significantly improved (Table 2.4). Among the pH modifiers, Minplus significantly increased several soil parameters, including organic carbon, soil pH, total phosphorus and cation exchange capacity, while dolomite improved the microbial biomass carbon and pH of the soil. Presence of the rain forest inoculum improved soil microbial biomass carbon significantly.

Under legume and tea mulches, there were no significant improvements of soil parameters with tea plants (Table 2.4). But in the absence of tea plants, organic carbon, cation exchange capacity, and microbial biomass carbon were increased (Appendix 7). All these responses are also evident in the principal component analysis plot (Figs 2.1a – 2.1d).

2.4.1.3 ***Factor analysis: JCU Nursery Trial- with tea plants***

Use of standard statistical techniques requires data that are normally distributed. In order to obtain a normal distribution of the results of the Nursery Trial with tea plants at James Cook University, the data were transformed to square root values. Therefore, in the tables of data presented in this section, the treatment means are presented as square root transformed data, and as back- transformed values.

At the end of the 28th week, visual observations have revealed that some of the undecomposed legume (*Calliandra)* and tea mulches were still present on the surface of the soils in the pots, but the grass mulch had completely decomposed. This gives an indication of the decomposition rate of the mulches. The grass mulch increased the soil organic carbon, cation exchange capacity, pH and microbial biomass carbon, while tea mulch reduced all of them (Table 2.5). The legume mulch did not change the organic carbon and cation exchange capacity but reduced the soil pH, microbial biomass carbon, and plant available phosphorus (Table 2.5).

Table 2.5 Effect of mulches on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter within a column are not significantly different ($p > 0.05$).

Treatments	**Soil parameters**				
	Organic carbon (mg/g)	**CEC** (meq/100g soil)	**Soil pH**	**Soil microbial biomass carbon** (µg/g)	**Plant available phosphorus** (mg/g)
No mulch	6.11 b (37.3)	1.750 b (3.06)	2.11 b (4.43)	17.37 b (301.7)	0.601 a (0.361)
Grass	6.35 a* (40.3)	1.761 a* (3.10)	2.13 a * (4.54)	18.25 a* (333.1)	0.609 a (0.371)
Legume	6.14 b (37.7)	1.752 b (3.07)	1.98 c# (3.92)	16.47 c# (271.3)	0.557 b# (0.310)
Tea mulch	5.89 c# (34.7)	1.742 c# (3.03)	1.99 c# (3.97)	16.17 c# (261.5)	0.513 c# (0.263)
LSD at p=0.05	0.03	0.002	0.006.	0.052	0.019
CV%	5.2	1.1	2.7	20.3	11.9

Of the soil pH amendments, Minplus significantly increased the organic carbon, cation exchange capacity, and soil pH. Dolomite applications resulted in a significant increase in soil pH and soil microbial carbon (Table 2.6).

Table 2.6 Effect of mulch on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*'. Means followed by the same lower-case letter within a column are not significantly different ($p > 0.05$).

Treatments	**Soil parameters** (square root transformation and back-transformed data)				
	Organic carbon (mg/g)	**CEC** (meq/100g soil)	**Soil pH**	**Soil microbial biomass carbon** (µg/g)	**Soil total phosphorus** (mg/g)
No soil pH modifier	6.062 b (36.75)	1.749 b (3.06)	2.009 c (4.04)	16.63 b (276.56)	1.863 b (3.47)
Dolomite	6.004 b (36.05)	1.746 b (3.06)	2.123 a* (4.51)	18.13 a* (328.69)	1.983 a* (3.93)
Minplus	6.298 a* (39.66)	1.759 a* (3.09)	2.026 b* (4.11)	16.44 b (270.27)	2.015 a* (4.06)
LSD at p=0.05	0.16	0.006	0.015	0.445	0.08
CV%	5.2	1.1	2.7	20.3	8.7

The microbial biomass carbon of the treated soil increased significantly under the rain forest inoculum, and the plant available phosphorus content of the soil increased significantly without rain forest inoculum in the presence of tea plant (Table 2.7). In the absence of tea plant there were no significant difference showed on soil parameters with the presence of rain forest inoculum (Appendix 8).

Table 2.7 Effect of inoculum on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$.

Inoculum Treatments	**Soil parameters** (square root transformation and back-transformed data)	
	Soil microbial Biomass carbon (µg/g)	**Plant available phosphorus** (mg/g)
(Without) In-	16.79 b (281.9)	0.593 a (0.352)
(With) In+	17.34 a* (300.7)	0.547 b # (0.299)
LSD at p=0.05	0.36	0.02
CV%	20.3	11.9

When the interaction between mulch and pH modifiers is considered, only pH and microbial biomass carbon had a significant effect (Table 2.8). Grass mulch improved the soil pH irrespective of pH modifiers, while no mulch and the grass mulch increased the soil pH in the presence of dolomite only (Table 2.8).

Table 2.8 Effect of mulch x pH combination on soil properties at the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$

Treatments		**Soil parameters** (square root transformation and back-transformed data)				
Mulch	**pH**	**Soil organic carbon** (mg/g)	**CEC** (meq/100g soil)	**Soil pH**	**Soil microbial biomass carbon** (µg/g)	**Available Phosphorus** (mg/g)
No mulch	Nil	6.20 ab (38.47)	1.753 ab (3.07)	2.04 c (4.16)	17.67 bc (312.23)	0.618 ab (0.382)
	Dolomite	5.97 bc (35.64)	1.745 ab (3.04)	2.21 a* (4.89)	18.34 b (336.36)	0.546 b (0.298)
	Minplus	6.16 ab (37.90)	1.753 ab (3.07)	2.06 c (4.26)	16.10 de # (259.21)	0.640 a (0.409)
Grass	Nil	6.20 ab (38.40)	1.754 ab (3.08)	2.09 b * (4.36)	17.36 bc (301.4)	0.593ab (0.352)
	Dolomite	6.33 a (40.13)	1.761 a (3.10)	2.21 a* (4.87)	19.81 a* (392.4)	0.649 a (0.421)
	Minplus	6.52 a (42.51)	1.769 a (3.13)	2.09 b * (4.38)	17.59 bc (309.4)	0.584ab (0.341)
Legume	Nil	5.98 bc (35.78)	1.746 ab (3.05)	1.95 e # (3.77)	16.17 de # (261.5)	0.586ab (0.343)
	Dolomite	6.10 abc (37.16)	1.750 ab (3.06)	2.03 d # (4.11)	17.52 bc (306.9)	0.536 bc (0.287)
	Minplus	6.33 a (40.09)	1.761 a (3.10)	1.96 e # (3.86)	15.73 de # (247.4)	0.548 b (0.300)
Tea mulch	Nil	5.87 c # (34.41)	1.741abcd (3.03)	1.96 e # (3.84)	15.32 e # (234.7)	0.586 ab (0.343)
	Dolomite	5.62 c # (31.54)	1.731 cd # (2.99)	2.04 c (4.16)	16.85 c (283.9)	0.536 bc (0.287)
	Minplus	6.18 ab (38.20)	1.754 ab # (3.08)	1.98 e # (3.92)	16.34 d # (266.9)	0.548 b (0.300)
LSD at p=0.05		0.31	0.012	0.03	0.89	0.05
CV%		5.2	1.1	2.7	20.3	11.9

Table 2.9 Effect of pH x inoculum amendments on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*'. Means followed by the same lower-case letter within column are not significantly different $p > 0.05$.

Treatments	Soil Organic Carbon (square root transformation and back-transformed data)		Cation Exchange Capacity (square root transformation and back-transformed data)	
	Without Inoculum	**With Inoculum**	**Without Inoculum**	**With Inoculum**
No pH modifier	6.227 ab (38.78)	5.897 c (34.77)	1.756 ab (3.08)	1.741bc (3.03)
Dolomite	6.035 ab (36.42)	5.974 bc (35.69)	1.748 b (3.06)	1.745bc (3.05)
Minplus	6.179 ab (38.18)	6.416 a * (41.17)	1.754 b (3.08)	1.764 a* (3.11)
LSD at p=0.05	0.22		0.01	
CV%	5.2		1.1	

The interaction between pH modifiers and forest inoculum shows that Minplus, in the presence of the rainforest soil inoculum, increased the soil organic carbon and cation exchange capacity of the treated soils, whereas dolomite had no effect (Table 2.9).

When the interaction between mulch x inoculum is considered, grass mulch increased the soil pH in the presence of forest inoculum, while grass mulch and no mulch improved soil pH compared to legume and tea mulches irrespective the forest inoculum (Table 2.10).

Table 2.10 Effect of mulch x inoculum amendments on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$.

Treatments	**Soil pH** (square root, transformation and back-transformed data)	
	Without Inoculum	**With Inoculum**
No mulch	2.086 b (4.35)	2.126 a * (4.52)
Grass	2.121 a ***** (4.50)	2.140 a * (4.58)
Legume	1.988 d # (3.95)	1.971 d # (3.88)
Tea mulch	2.014 c # (4.06)	1.975 d # (3.90)
LSD at p = 0.05	0.02	
CV%	2.7	

Tea and legume mulch reduced soil pH with all pH modifiers except dolomite with rainforest inoculum under the tea mulch, and dolomite without rainforest inoculum under the legume mulch. Microbial biomass carbon increased in the dolomite and grass mulch treatments, both with or without the rainforest soil inoculum (Table 2.11).

Table 2.11 Effect of mulch x pH x inoculum on soil properties in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter are not significantly different $p > 0.05$.

Treatments		**Soil parameters**(square root transformation and back-transformed data- average values)			
		Soil pH		**Soil microbial biomass carbon** (μg/g)	
Mulch	**pH**	**Without Inoculum**	**With Inoculum**	**Without Inoculum**	**With Inoculum**
No mulch	Nil	2.04 d (4.2)	2.03 d (4.1)	16.9 bc (285.6)	18.4 ab (338.6)
	Dolomite	2.16 b * (4.7)	2.26 a* (5.1)	18.2 b (331.2)	18.4 ab (338.6)
	Minplus	2.05 cd (4.2)	2.08 c * (4.3)	15.2 dc (231.0)	16.9 bc (285.6)
Grass	Nil	2.09 c * (4.4)	2.09 c * (4.4)	16.9 bc (285.6)	17.7 b (313.3)
	Dolomite	2.18 b * (4.8)	2.23 a* (5.0)	20.0 a* (400.0)	19.6 a (384.2)
	Minplus	2.09 c * (4.4)	2.10 c* (4.4)	16.9 c (285.6)	18.2 b (331.2)
Legume	Nil	1.94 f # (3.8)	1.89 g # (3.6)	16.1 c (259.2)	16.2 c # (262.4)
	Dolomite	2.01ed (4.0)	2.05 d * (4.2)	16.8 c (282.2)	18.3 b (334.9)
	Minplus	1.96 ef # (3.8)	1.97 ef # (3.9)	15.9 c (252.8)	15.6 dc # (243.4)
Tea mulch	Nil	1.94 f # (3.8)	1.98 e # (3.9)	15.4 dc (237.2)	15.2 dc # (231.0)
	Dolomite	2.10 c * (4.4)	1.98 e # (3.9)	16.1 c (259.2)	17.5 b (306.3)
	Minplus	2.00 ed (4.0)	1.96 ef # (3.8)	16.9 bc (285.6)	15.7 c # (246.5)
LSD (p=0.05)		0.04		1.25	
CV%		2.7		20.3	

When the effects of dolomite with the mulch x pH modifiers x forest inoculum interaction are considered, dolomite had increased the pH, even in the absence of the grass mulch and irrespective of rain forest inoculum. The grass mulch also improved the soil pH and soil microbial biomass carbon in the presence of dolomite and with or without the rainforest soil inoculum (Table 2.11).

2.4.1.4 *Microbial dynamics: JCU Nursery Trial*

The microbial activity of the soil is a dynamic property and changes with time. Therefore, microbial dynamics were studied by measuring the microbial biomass nitrogen with time up to 28 weeks after the application of treatments. The results are presented in Figs 2.2 a-c.

There was an initial decline in the microbial biomass nitrogen of the soil up to week 10 followed by a slightly increase up to week 20 in all the treatments (Fig. 2.2 a). At the end of the trial, the microbial biomass nitrogen of the soil under the grass mulch was higher than the initial value and that of all of other treatments. The initial (Week 0) value of the microbial biomass nitrogen content of the soil under the tea mulch was significantly lower than that of all of the other treatments (Fig. 2.2 a).

Under all pH amendments, there was an initial decline in soil microbial nitrogen up to 10 weeks after application of the mulches (Fig. 2.2 b). However, the decline was significantly lower in the Minplus and the control. The soil microbial biomass nitrogen under dolomite recovered to the initial level at the end of 28 weeks, but the soil microbial nitrogen contents under the Minplus and the control were significantly lower than initial value (Fig. 2.2 b).

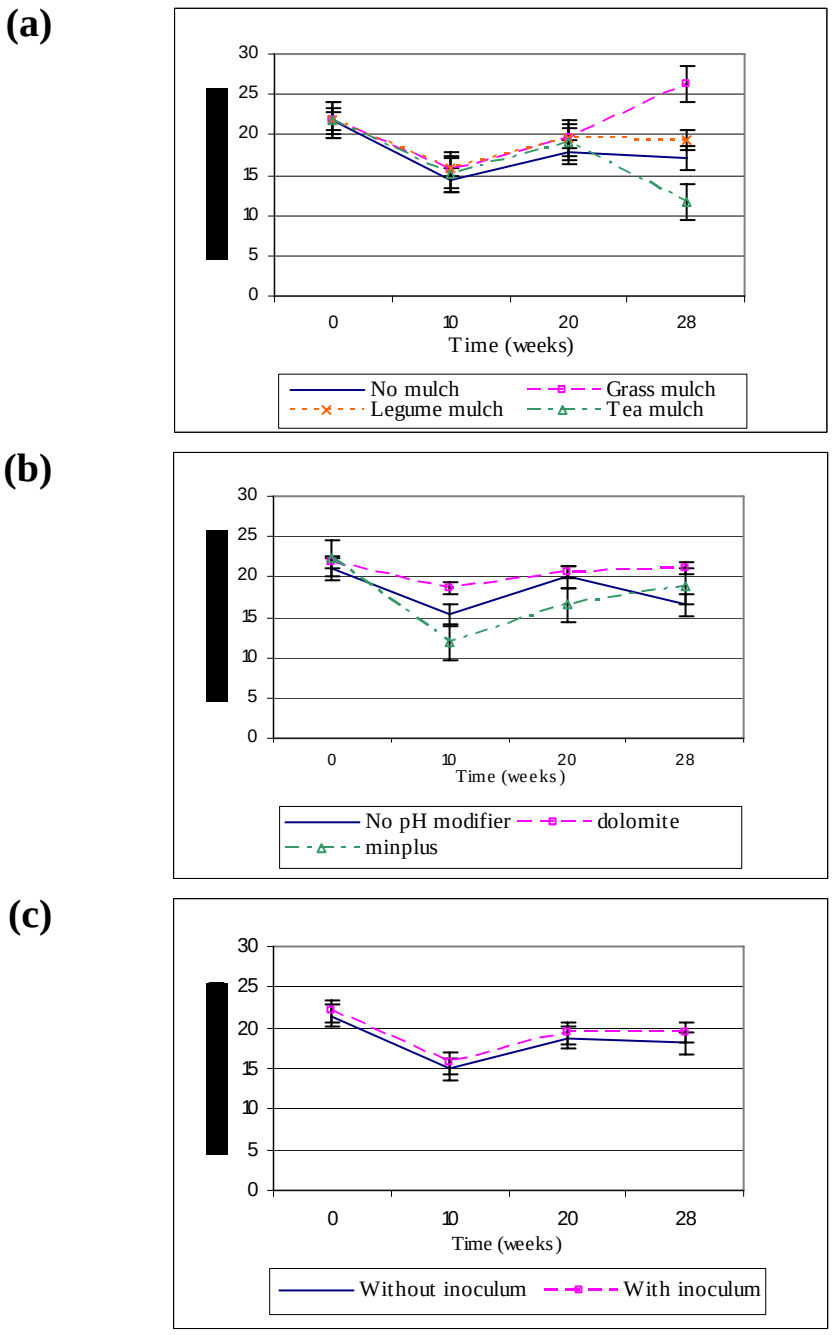


Figure 2.2 Changes in soil microbial biomass nitrogen over the 28 week period of the James Cook University Nursery Trial (with plants):

(a) Effect of mulches on microbial biomass nitrogen,
(b) Effect of soil pH modifiers on microbial biomass nitrogen
(c) Effect of rainforest soil inoculum on microbial biomass nitrogen

The influence of forest inoculum on soil microbial biomass nitrogen also followed a similar pattern to that of mulching and pH amendment treatments by showing an initial decline up to 10 weeks (Fig. 2.2c). At the end of 28 weeks, however, the differences disappeared and microbial biomass nitrogen recovered to the initial value. In the absence of tea plants, the microbial biomass nitrogen was lower than the initial value at the end of the 28th week (Appendix 9).

2.4.2 Changes in soil microbial functional groups

An initial FAME (fatty acid methyl ester) analysis was carried out by Dr. C. Pankhurst, CSIRO Land and Water, Adelaide, on samples of the soils used in the JCU Nursery Trial after mixing the soils with pH amendments, but not with mulching materials. A second FAME analysis was carried out on the soils from the pot trial at the end of week 28, after growing tea in the soil treated with pH amendments, mulching materials, and forest inoculum (Fig. 2.3).

A principal component analysis showed that the grass and legume mulch had the greater effect on the FAME profiles of the treated soil than tea mulch and no mulch. It was found that 14 attributes of the microbial populations represented 90% of the total variability, and those microbial groups were significantly different from other groups of micro-organisms (Appendix 10).

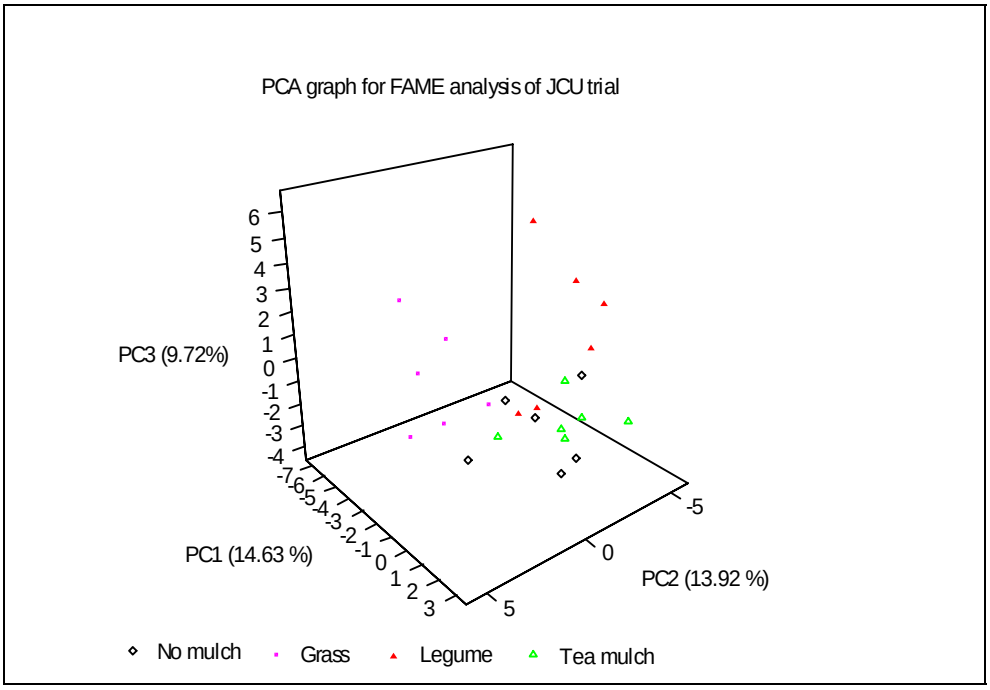


Figure 2.3 Principal component analysis of FAME profiles of microbial communities in soils collected after 28 weeks from the nursery trail at James Cook University in Australia.

A principal components analysis of the fatty acids from the soils of the JCU Nursery Trial showed that the microbial populations of the soils amended with dolomite and rainforest soil inoculum were different from those of the rest of the treatments, and from each other (Fig. 2.4); they had lower counts of selected total bacterial components (Table 2.12).

Gram negative bacteria were present in the initial soil analysis (Table 2.12), but at the end of the trial after an incubation period of 28 weeks after application of treatments, none of the gram negative bacteria were observed in the treated soils (Table 2.13). This suggests that the pathogenic micro-organisms had been suppressed by the beneficial micro-organisms whose populations have developed within a favourable environment provided by the amended soils.

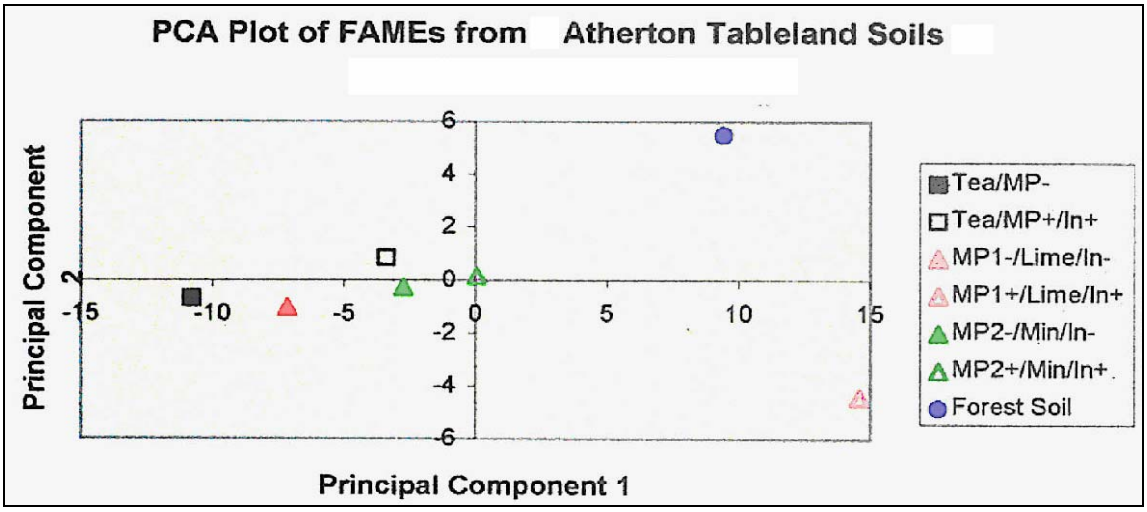


Figure 2.4 Principal component analysis of bacterial communities in the soils used at the start of the Nursery Trial at James Cook University, Australia. Data from Dr. C. Pankhurst, CSIRO Land and Water, Adelaide.

Legend:
Tea/MP- = *tea soil + no pH modifier + without inoculum*
Tea/MP+/In+ = *tea soil + no pH modifier + with inoculum*
MP1/lime/In- = *tea soil + dolomite + without inoculum*
MP1/lime/In+ = *tea soil + dolomite + with inoculum*
MP2/Min/In- = *tea soil + Minplus + without inoculum*
MP2/Min/In+ = *tea soil + Minplus + with inoculum*

Table 2.12 Abundances of bacteria, fungi, ratio of fungi/bacteria, and mycorrhizal components of the soil at the start of the Nursery Trial at James Cook University.

Treatments	Total bacteria (%)	Total fungi (%)	Gram positive bacteria (%)	Gram negative bacteria (%)	Ratio: Gram positive / Gram negative bacteria	Mycorrhizae (%)
Soil, without inoculum	52.08	5.05	48.12	3.96	12.15	5.31
Soil, with inoculum	41.66	8.03	38.68	2.98	12.98	4.31
Soil, Dolomite and without inoculum	48.62	5.65	44.85	3.77	11.90	4.63
Soil, Dolomite and inoculum	18.85	2.7	17.87	0.98	18.23	2.23
Soil, Minplus and without inoculum	45.08	4.66	41.74	3.34	12.50	4.72
Soil, Minplus and inoculum	39.17	5.51	36.55	2.62	13.95	3.94

Fungal population numbers were higher in the soil under the grass and *Calliandra* legume treatments compared to those under the tea mulch (Table 2.13) suggesting that

Table 2.13 Abundances of bacteria, fungi, ratio of fungi/bacteria, and mycorrhizal components of the soil at the end of the 28 week Nursery Trial at James Cook University.

Treatments	**Total Bacteria** (all Gram positive) (%)	**Total fungi (%)**	**Ratio Fungi/Bacteria**	**Mycorrhizae (%)**
		Mulch		
No mulch	28.39	1.79	0.06	1.75
Grass	29.67	4.36	0.15	1.92
Calliandra	28.27	3.76	0.13	1.58
Tea mulch	27.66	1.65	0.06	1.61
		Soil pH modifiers		
No pH modifiers	28.76	2.87	0.10	1.59
Dolomite	28.50	2.83	0.10	1.93
Minplus	28.23	2.96	0.10	1.64
		Forest inoculum		
Without inoculum	28.60	2.67	0.09	1.63
With inoculum	28.40	3.10	0.11	1.81

there has been an increase in fungal populations in the treatments. It was present in much higher amounts in the soil that had received the grass and *Calliandra* mulches compared to that under the tea mulch. There was no difference in the ratio of fungi to bacteria population in the other treatments (Table 2.13), but the mycorrhizal numbers had increased in the dolomite and Minplus treated plots after 28 weeks.

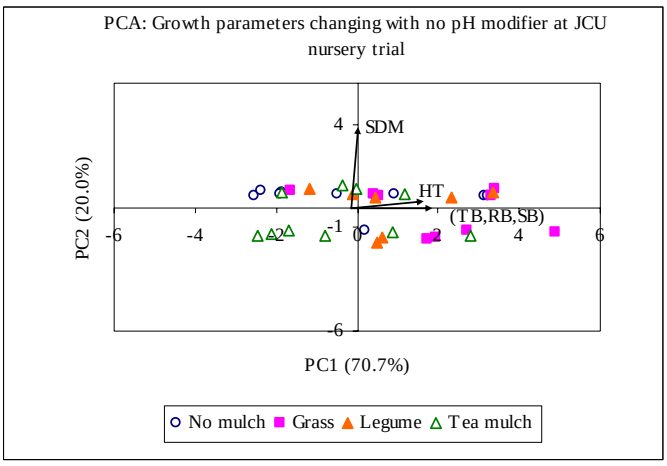
2.4.3 Plant growth parameters

2.4.3.1 Principal component analysis of plant growth parameters - JCU Nursery Trial

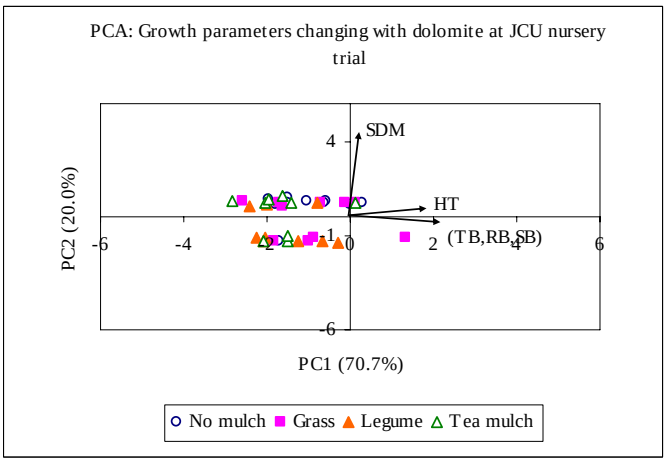
Plant stem diameter, height, number of shoots, number of leaves per shoot, leaf area, dry shoot mass, dry root mass, and total dry biomass were measured as growth parameters and were analysed by principal component analysis. Leaf area was analysed separately since that data includes zeros due to the death of plants in the dolomite treatments. This may be because the rate of dolomite (2500 kg ha^{-1}) applied resulted in excess calcium accumulating in the plant.

Two components that described 90.9% of variance of data set were extracted from the stem diameter, height, shoot, root and total biomass. After performing a principal component analysis, in which the first and second principal components (representing the largest fraction of the overall variability) were plotted on vertical and horizontal axes respectively (Figs 2.5a – c). Component 1, explaining 70.7 % of the variance, included plant height, shoot and root mass, and total biomass. Component 2, explaining an additional 20.2 % of the variance, included stem diameter (Figs 2.5 a – c).

(a)



(b)



(c)

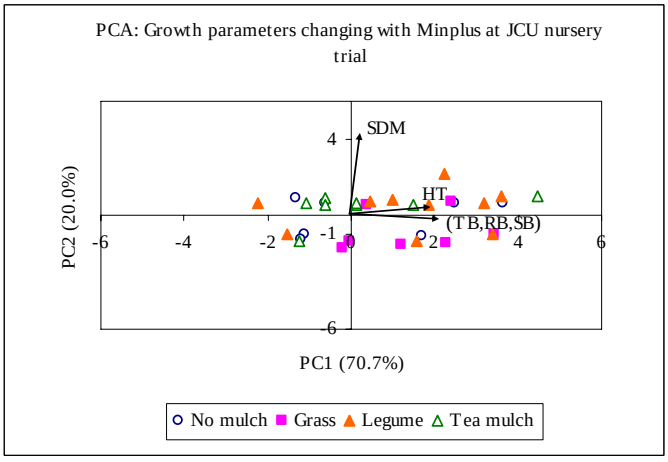


Figure 2.5 Effects of mulches on soil parameters in the JCU Nursery Trial with plants:

(a) Data exclude soil pH modifiers.

(b) Data include Minplus as the soil pH modifier;

(c) Data include dolomite as the soil pH modifier

The grass and legume mulches had a stronger influence than no mulch and tea mulch treatments on plant height, stem diameter, and shoot, root, and total biomasses (Fig. 2.5 a). In the dolomite-treated pots, all of the treatments had very little influence on plant height, and on shoot, root, and total biomasses compared with plants grown in the soils to which no pH modifier had been applied (Fig. 2.5b). In the Minplus treatments, the plants grown under grass and legume mulches showed responses similar to those of the plants grown with no soil pH modifier (Fig 2.5a and c), and plant height and shoot, root, and total biomasses showed the greatest responses (Fig. 2.5c).

2.4.3.2 *Factor analysis of growth parameters: JCU Nursery Trial*

The grass mulch improved the dry shoot biomass and total biomass which were trends also shown by the principal component analysis; none of the other mulches had a significant effect on the plant growth parameters (Table 2.14).

Plant shoot biomass and total dry biomass showed significant responses to grass mulch compared with the growth responses of the plants under the control tea and legume mulch treatments (Table 2.14). Plant height was not affected by the mulching treatments (Table 2.14) and this was also demonstrated in the principal component analyses (Figs 2.5a,b,c). The grass mulch produced smaller stem diameters in the tea plants; the legume mulch had no effect on any of the measured growth parameters; and the tea mulch reduced dry root biomass (Table 2.14).

Table 2.14 Effect of mulch on growth parameters in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the fourth root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by same the lower-case letters within a column are not significantly different $p > 0.05$.

Treatments	**Mean growth parameters** (fourth root transformation and back-transformed data)				
	Height of plant (cm)	**Stem diameter at 3 cm above the soil surface** (cm)	**Dry shoot biomass** (g)	**Dry root biomass** (g)	**Total dry biomass** (g)
No mulch	1.98 (15.4)	0.65 a (0.2)	0.87 b (0.6)	0.80 a (0.4)	0.99 b (1.0)
Grass	1.99 (15.7)	0.62 b # (0.1)	1.04 a* (1.2)	0.88 a (0.6)	1.16 a* (1.8)
Legume	1.96 (14.8)	0.63 ab (0.2)	0.95 a b (0.8)	0.81 a (0.4)	1.07 ab (1.3)
Tea mulch	1.94 (14.2)	0.64 ab (0.2)	0.84 b (0.5)	0.73 b # (0.3)	0.94 b (0.8)
Lsd (p=0.05)	Ns	0.03	0.14	0.10	0.14
CV%	9.8	10.3	15.9	7.4	6.2

Plant height, and dry shoot, dry root, and total dry biomasses were all smaller in the plants grown in the dolomite-treated soils compared with those grown under the control and Minplus treatments (Table 2.15).

Table 2.15 Effect of pH on plant growth parameters in the JCU Nursery Trial, with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the fourth root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by same the lower-case letter within a column are not significantly different $p>0.05$.

Treatments	**Mean growth parameters** (fourth root transformation and back-transformed data)				
	Height of plant (cm)	**Stem diameter at 3 cm above the soil** (cm)	**Dry shoot mass** (g)	**Dry root mass** (g)	**Total dry biomass** (g)
No pH modifier	1.97 ab (15.1)	0.64 (0.2)	1.01 a (1.0)	0.85 a (0.5)	1.13 a (1.6)
Dolomite	1.88 b (12.2)	0.64 (0.2)	0.70 b # (0.2)	0.67 b # (0.2)	0.82 b # (0.5)
Minplus	2.04 a 17.3	0.64 (0.2)	1.07 a (1.3)	0.90 a (0.7)	1.19 a (2.0)
LSD (p=0.05)	0.09	Ns	0.12	0.09	0.12
CV%	9.8	10.3	15.9	7.4	6.2

The presence of forest inoculum increased the plant height and the shoot biomass of tea plant (Table 2.16). Interactions of the mulch and pH amendments were significant at $p < 0.01$ (Table 2.17).

Table 2.16 Effect of an inoculum of rainforest soil on growth parameters of plants used in the JCU Nursery Trial, 28 weeks after mulch emplacement. Each cell represents the mean values of the fourth root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*'. Means followed by the same lower-case letter within a column are not significantly different at $p > 0.05$.

Treatments	**Plant growth parameters** (fourth root transformation and back-transformed data)				
	Height of plant (cm)	**Stem diameter at 3 cm above the soil** (cm)	**Shoot dry weight** (g)	**Root dry weight** (g)	**Total dry biomass** (g)
In- (without inoculum)	1.93 b (13.9)	0.63 a (0.2)	0.87 b (0.6)	0.78 a (0.4)	1.00 a (1.0)
In+ (with inoculum)	2.00 a * (16.0)	0.64 a (0.2)	0.97 a * (0.9)	0.83 a (0.5)	1.09 a (1.4)
LSD (p=0.05)	0.07	Ns	0.09	0.07	0.10
CV%	9.8	10.3	15.9	7.4	6.2

Grass and legume mulches in the presence of Minplus increased the dry shoot and root biomasses (Table 2.17). The grass and legume mulches in the absence of pH modifiers also increased the dry shoot biomass. All the mulch treatments with dolomite significantly reduced total dry biomass (Table 2.17).

Grass and legume mulches increased dry shoot biomass and total dry biomass irrespective of the presence or absence of the rainforest soil inoculum. Dry root mass was higher in the plants grown under the grass mulch with no rainforest soil inoculum (Table 2.18).

Table 2.17 Effect of mulch and soil pH amendments on plant growth parameters in the JCU Nursery Trial, 28 weeks after mulch emplacement. Each cell represents the mean values of the fourth root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.10$) differences greater than those obtained by the control treatment are shown by '*' and significant decreases by '#'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.10$

Treatments		**Growth parameters**				
		Plant height (cm)	**Stem diameter** (cm)	**Dry shoot mass** (g)	**Dry root mass** (g)	**Total dry biomass** (g)
No mulch	Nil	1.90 abc (13.03)	0.66 a (0.19)	0.91bcd (0.66)	0.81 c (0.43)	1.03 ab (1.13)
	Dolomite	1.95ab (14.46)	0.66 a (0.19)	0.71e # (0.25)	0.69 cd (0.23)	0.84 c # (0.50)
	Minplus	2.10 a (19.45)	0.65 a (0.18)	1.00 bc (1.00)	0.91 a * (0.69)	1.14 a (1.69)
Grass	Nil	2.10 a (19.45)	0.63 abc (0.16)	1.19 a * (2.01)	1.00 a * (1.00)	1.33 a (3.13)
	Dolomite	1.90ab (13.03)	0.64 abc (0.17)	0.80 de (0.41)	0.69 cd (0.23)	0.89 c # (0.63)
	Minplus	2.00 a (16.00)	0.61 c # (0.14)	1.15 a * (1.75)	0.94 a * (0.78)	1.26 a (2.52)
Legume	Nil	2.00 a (16.00)	0.64 abc (0.17)	1.10 a * (1.46)	0.84 abc (0.50)	1.19 a (2.01)
	Dolomite	1.80 c (10.50)	0.61 c # (0.14)	0.65 e # (0.18)	0.68 cde (0.21)	0.80 c # (0.41)
	Minplus	2.10 a (19.45)	0.65 a (0.18)	1.10 a * (1.46)	0.91 a * (0.69)	1.22 a (2.22)
Tea mulch	Nil	1.90 abc (13.03)	0.62 abc (0.15)	0.85 bcd (0.52)	0.74 dc (0.30)	0.96 abc (0.85)
	Dolomite	1.90 abc (13.03)	0.64 abc (0.17)	0.66 e # (0.19)	0.60 e # (0.13)	0.76 c # (0.33)
	Minplus	2.00 a (16.00)	0.66 a (0.19)	1.02 ab (1.08)	0.84 abc (0.50)	1.13 ab (1.63)
LSD (p=0.10)		0.15	0.04	0.19	0.10	0.20
CV%		9.8	10.3	15.9	7.4	6.2

Table 2.18 Effect of mulch x inoculum amendments on plant growth parameters in the JCU Nursery Trial with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root x square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences greater than those obtained by the control treatment are shown by '*'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$.

Treatments	**Dry shoot mass** (fourth root transformation and back-transformed data)		**Dry root mass** (fourth root transformation and back-transformed data)		**Total dry biomass** (fourth root transformation and back-transformed data)		
Mulch	Without Inoculum	With Inoculum	Without Inoculum	With Inoculum	Without Inoculum	With Inoculum	Mean
No mulch	0.73 b (0.28)	1.01 a * (1.05)	0.71 b (0.26)	0.89 a * (0.63)	0.87 b (0.56)	1.14 a* (1.72)	(0.75)
Grass	1.10 a * (1.44)	0.99 a * (1.00)	0.93 a * (0.75)	0.83 ab (0.47)	1.22 a * (2.20)	1.10 a* (1.49)	(1.22)
Legume	0.95 a * (0.83)	0.95 a * (0.82)	0.80 ab (0.41)	0.82 ab (0.45)	1.07 a * (1.30)	1.07 a* (1.32)	(0.85)
Tea mulch	0.76 b (0.34)	0.92 ab (0.73)	0.67 b (0.20)	0.79 ab (0.38)	0.86 b (0.55)	1.03 ab (1.14)	(0.56)
Mean	(0.72)	(0.90)	(0.39)	(0.48)	(1.15)	(1.41)	
LSD at p=0.05	0.19		0.14		0.19		
CV%	15.9		7.4		6.2		

Plant height was not affected by soil pH amendments in the absence of the rainforest soil inoculum, but was significantly higher in no pH modifier and Minplus treatments than in the plants grown in the soils treated with dolomite in the presence of rainforest soil inoculum (Table 2.19).

Table 2.19 Effect of pH x inoculum amendments on plant growth parameters in the JCU Nursery Trial with plants, 28 weeks after mulch emplacement. Each cell represents the mean values of the square root x square root transformed data; back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences lower than those obtained by the control treatment are shown by '#'. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$.

Treatments	**Plant height** (Fourth root transformation and back-transformed data)	
pH modifiers	Without Inoculum	With Inoculum
No pH modifier	1.90 b (13.03)	2.04 a * (17.32)
Dolomite	1.90 b (13.03)	1.87 b # (12.23)
Minplus	1.98 ab (15.37)	2.11 a * (19.82)
LSD (p=0.05)	0.13	
CV%	9.8	

2.5 DISCUSSION

2.5.1 Soil Parameters

2.5.1.1 *Soil organic carbon*

Organic carbon levels depend on the amount and decomposition rate of the added mulch materials (Joergensen *et al*. 1994). In turn, organic matter decomposition rates depend on the biochemical composition of the substrate (Trinsoutrot *et al.* 2000), the physical availability of those components to decomposer micro-organisms (Swift *et al*. 1979), the priming effect of added nitrogen fertilizers (Recous *et al*. 1995) and carbon materials (Bending *et al*. 2002), particle size of the organic debris (Christensen 1987; Ambus and Jensen 1997), the area of contact between the mulching materials and soil surface (Ambus and Jensen 1997; Rovira and Vallejo 2002), soil pH (Alexander 1977; Grey and Williams 1981; Paul and Clark 1989; Shah *et al*. 1990; Neale *et al*. 1997), and the soil water potential and soil temperature (Lal 1974; Anderson and Nilsson 2001). These factors interact to determine the size of the microbial population (Paul and Clark 1989, Dalal 1998).

In the present study, 10-11 g pot^{-1} (equivalent to 35 tonnes fresh weight ha^{-1}) of carbon was added to each pot, and all of the factors mentioned above were kept constant, except for the quality of the organic materials which depends on their C : N ratios (Trinsoutrot *et al*. 2000); on their nitrogen concentrations or (lignin + polyphenol) : N ratios (Hayes 1986); on their polyphenol : N ratios (Oglesby and Fownes 1992; Constantinides and Fownes 1994; Handayanto *et al*. 1994; Seneviratne *et al*. 1998); or on their lignin concentrations or lignin : N ratios (Oglesby and Fownes 1992). The C/N ratio of the incorporated plant material, are often the best predictors of the rate of

organic matter decomposition and N release for a wide range of residue N concentration (Mullar *et al*. 1988; Seneviratne 2000). Plant nutrients and critical level of carbon which limit the enzyme activities of microbial decomposers were the important factors in determining nutrient release (Seneviratne 2000).

In the present study, the C : N ratios of the grass *Brachiaria*, legume *Calliandra,* and tea mulches at the start of the 28 week trial, were 17, 10, and 11 respectively (Table 2.2). All the mulching materials had C : N ratios lower than the critical value of 25 (Mullar *et al.* 1988). The physical qualities of the plant residues and their area of contact with the soil were similar because they were all ground to similar sizes and uniformly placed on the surface of the soil. Therefore, the foregoing parameters were not likely to contribute to the observed variations in soil properties.

The soil water potential was also constant across the experiment since the experimental soils were kept at or close to 50 - 80% of field capacity, and temperature fluctuations in the shadehouse would have affected all of the treatments equally. All the treatments were fertilized by nitrogenous fertilizer (T65 mixture) at monthly intervals and the added nitrogen would have had a 'priming effect' on the microbial activity and mineralization (Jenkinson 1988).

Among the mulching materials, the grass mulch increased the organic carbon content of the treated soils more than did the legume mulch (Table 2.5). The lignin content of *Calliandra* mulch (22%) was higher than that of the other mulches used (Table 2.2) which may have affected its decomposition rate (Fox *et al*. 1990; Handayanto *et al*. 1997). Therefore the decomposition rate of the grass mulch was higher than that of the

Calliandra legume mulch. Sivapalan (1982) reported that the tea mulch has a polyphenol content of (19%) which is higher than that of *Brachiaria decumbens* grass mulch (3.4%) (Table 2.2). Soluble polyphenols slow the mineralization of residual nitrogen by forming complexes with proteins, which are less accessible to micro-organism decomposition (Mafongoya *et al*. 1998). *Calliandra* also has a polyphenol content (18.2%) closely similar to that of tea (Sivapalan 1982). Further, it also has higher lignin content (17%) than tea mulch (11%) and *Brachiaria decumbens* grass mulch (9%) (Table 2.2). These may be the factors contributing the to the high organic carbon contents in the soils under the grass mulch (Tables 2.4 and 2.5).

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The soil pH and microbial biomass carbon contents were higher under the grass mulch than under the other mulches (Tables 2.8, 2.5). Among the pH amendments only Minplus at 2.5 tonnes ha^{-1}, produced significantly higher organic carbon contents (Table 2.6), possibly as a result of higher soil pH produced by the treatment, and possibly as a consequence of the range of other nutrients available in the Minplus.

In the absence of tea plants, the organic carbon content of the soils was increased by all of the mulching materials (Table 2.4). This increase in organic carbon may be due to relatively lower microbial activity compared to that of the other treatments (Bezbaruah 1999).

2.5.1.2 *Soil pH*

Of the mulch treatments, only the grass mulch increased the soil pH significantly (Tables 2.4 and 2.6) in the presence of tea plants. The change in soil pH may have been caused by a number of factors among which are the addition of cations and

neutralization of H^+ ions by the soil pH amendments and mulching materials (Yan *et al*. 1996; Marschner and Noble 2000); the addition of acidifying nitrogen fertilisers and nitrification of the added fertilisers (Wickramasinghe *et al*. 1985 b); the liberation of carbon dioxide by root and microbial respiration (Paul and Clark 1989; Blum and Shafer 1988); or by the formation of humic and fulvic acids (Hayes 1986). The resultant change in soil pH is most likely the cumulative effect of all of the above processes.

The increase in soil pH may also be due in part to higher ash alkalinity of the grass mulch compared to the other mulches, as a consequence of cations being converted to oxidisable forms during the microbial decomposition of the organic litter (Tables 2.4, 2.5). Under the legume and tea mulches there was an accumulation of H^+ ions in the soil as a result of nitrification and the removal of nitrate ions, which produced the lower soil pH conditions. The retention of ammonium ions generated from mulch decomposition and fertiliser inputs was also lower compared to that of the grass mulch and the control (Table 2.4, 2.5). Similarly, Whitehead *et al*. (1981) found that tea mulch reduced the soil pH in most instances, probably as a result of phenolic substances being added to the soil.

Dolomite and Minplus treatments both increased the soil pH (Table 2.6), and grass mulches that were used in conjunction with these soil pH modifiers also increased soil pH (Table 2.8). On the other hand, legume and tea mulches, in conjunction with pH modifiers, reduced the soil pH significantly (Table 2.8) showing that the effect of mulches was predominant.

When the mulch x inoculum interactions were considered, the grass mulch without the rainforest inoculum increased the soil pH, while tea and legume mulches, both with and without the soil inoculum, reduced the soil pH (Table 2.10). This may be a consequence of the lower ash alkalinity of the legume and tea mulches, and the higher polyphenol content of the tea mulch.

2.5.1.3 *Cation Exchange Capacity*

The clay mineralogy of the Galmara Series soil from Innisfail is dominated by approximately equal proportions of kaolinite and vermiculite, with traces of gibbsite and goethite (Murtha 1983). These clays generally have lower cation exchange capacity than that of the reactive, smectic clays. However, with the increases in organic carbon content and pH of the soils as a response in the soil pH amendments, the cation exchange capacity of the experimental soil may have been increased. Ananthacumaraswamy and Baker (1991) reported that the addition of lime increased the effective cation exchange capacity of a similar soil in Sri Lanka. Calcium and magnesium, derived from dolomite, may also increase the basic cation content of the soil, and with humic and fulvic acids from decomposing mulching materials, may also contribute to the cation exchange capacity of the soil (Sivapalan *et al.* 1983). The other pH modifier used was Minplus, which provides abundant surface area in the colloids in the soil, thereby raising the negative charge and increasing the cation exchange capacity of soil (Coventry *et al*. 2001).

The cation exchange capacity of the soil also depends on the soil texture and mineralogy, organic carbon content, and pH (Gillman and Sumpter 1986). In the present study, soil texture and mineralogy were kept constant across the treatments.

Therefore, the variables that may have contributed most to changes in cation exchange capacity are organic matter content and pH, a conclusion supported by the results of this study, in both the presence or absence of tea plants (Tables 2.4, 2.5, and 2.6).

2.5.1.4 *Total Nitrogen*

In most of the treatments considered, no significant changes were induced in the soil nitrogen contents, either with or without tea plants present. In the absence of tea plants, however, total nitrogen contents were significantly lower in the soils under the legume and tea mulches compared to those of the control and grass mulch treatments (Table 2.4). The total nitrogen content of the soil is influenced by the addition of fertilisers, mineralisation of organic compounds derived from the soil and the mulching materials, and denitrification and leaching processes. Although *Calliandra* legume and tea mulches had relatively high leaf nitrogen contents (3.5-4.0 %) and low C/N ratios, their decomposition rates were influenced by other biochemical compounds, specifically polyphenol contents (Palm and Sanchez 1991). The low C/N ratio of legume and tea mulch with pH modifiers increased the mineralisation (Constantinides and Fownes 1994, Seneviratne 2000) and nitrate ions may have been leached though the system since the pots had been maintained 40% field capacity. This may be the reason for the lower nitrogen contents of the soil under the legume and tea mulches compared to the grass mulch treated plots (Table 2.4).

2.5.1.5 *Total phosphorus*

All the mulches had about 0.1-0.2 % phosphorus (Table 2.2), and Minplus contains about 0.5% phosphorus (Coventry *et al.* 2001). None of the soils under the mulch treatments showed any significant differences in total nitrogen contents (Table 2.4)

However, there was a significant increase in total phosphorus contents only in the soils under the dolomite and Minplus treatments and in the presence of tea plants (Table 2.4, 2.6). The total phosphorus in the soil is derived from fertilizer inputs and the soil pH amendments, and from mineralization of phosphorus in the mulching materials. The phosphorus from fertiliser additions was similar in all the cases. The higher soil pH produced by the dolomite treatment may have resulted in stronger mineralization of phosphorus from the mulches, thus contributing to higher total phosphorus contents of the treated soils.

2.5.1.6 *Plant available phosphorus*

The grass mulch increased the plant available phosphorus in the soils (weak sulphuric acid extraction method) in the absence of tea plants (Tables 2.4), while the legume and tea mulches reduced the plant available phosphorus in the soil (Table 2.4). It has been shown that soil pH, mineralization of mulches, and uptake by plants are the main processes regulating phosphorus availability (McLaughlin *et al*. 1988). The dolomite and Minplus treated soils had lower plant available phosphorus contents compared to the soils in the control treatment in both the presence and absence of tea plants (Table 2.4 and 2.5). This is most likely a consequence of the fixation of phosphorus by an excess of calcium ions in the soil pH modifier treatments, but the processes are controlled by organic matter content, soil pH levels, and microbial activity. Despite a constant addition of phosphorus from the T 65 fertiliser mixture (Section 2.3.4), there was a reduction in plant available phosphorus in the soil treated with forest inoculum in the presence of tea plant (Tables 2.4, and 2.7) possibly due to fixation by the clay minerals, gibbsite, and goethite (Noble *et al*. 1996).

2.5.1.7 *Soil microbial biomass*

In the presence of tea plants, only grass mulch significantly increased the microbial biomass carbon content of the soil while the tea and legume mulches reduced the microbial biomass carbon (Tables 2.4 and 2.5). This may have been partly a consequence of the nutrient availabilities in the substrates. In the case of the legume mulch, faster decomposition may have reduced the substrate availability. The presence of unoxidised polyphenols in the tea mulch may be another reason for lower soil microbial biomass numbers in the underlying soil. In the soils under the legume and tea mulches, organic carbon and soil pH were also lower than in the untreated controls, and may provide be another reason for the lower microbial biomass under these mulches.

The microbial biomass carbon was significantly increased by all of the mulch treatments in the absence of tea plants. Among the soil pH amendments, only dolomite increased the microbial biomass carbon in both the presence and absence of tea plant (Tables 2.4, 2.5).

In the absence of tea plants, the soil organic carbon content was significantly increased by the grass and tea mulches (Table 2.4) which, with their decomposition products, provided a substrate containing carbon and nitrogen for the soil microbial populations. It has been shown that the addition of carbon and nitrogen to the soil have a 'priming' effect on the growth of microbial populations and on the mineralization of organic matter (Jenkinson 1988). Therefore, the substrate availability such as carbon, nitrogen, and phosphorus contents of the soil, and the soil pH, are the dominant factors affecting the soil microbial populations, and therefore influence the microbial biomass carbon

contents of the soil (Lynch, 1983, Diaz-Ravina *et al*. 1988; Dalal 1998; Shah *et al*. 1990; Anderson and Nilsson 2001).

Dolomite also increased microbial biomass carbon probably due to an increase in pH (Table 2.4, 2.6). Most microbes prefer soils that are not strongly acidic (Alexander 1977). The soil microbial biomass carbon also increased in the soils under the rainforest soil inoculum treatment in the presence of tea plants. This may have been result of incorporation of microbial biomass by the inoculum (Table 2.7). Further, grass mulch and dolomite combination increased the soil microbial biomass, while the legume and tea mulches with Minplus combinations reduced the microbial biomass carbon in the presence of tea plants (Tables 2.8) possibly as a consequence of the lower soil pH (Table 2.11).

Microbial biomass nitrogen was significantly increased in the soils under the grass mulch (Fig. 2.2a). There were no significant difference in microbial biomass nitrogen in the presence of soil pH modifiers (Fig. 2.2b) and inoculum with tea plants (Fig. 2.2c).

2.5.1.8 *Soil bacteria and fungi*

Table 2.12 shows the initial composition of soil microbial populations including Gram-positive and -negative bacteria, fungi, and mycorrhizae. Twenty eight weeks after application of treatments, there were no Gram-negative bacteria, but only Gram-positive-bacteria which are beneficial to the soil (Table 2.13). This confirms the suppression of most of the pathogenic (Gram-negative) bacteria by the addition of mulch as was found by Cook (1994). In the present trial, the soils under the grass and legume mulches showed higher abundances of Gram-positive bacteria than in those

under the tea mulch and no mulch treatments. This is due to the unoxidised polyphenolic compounds in the tea mulch suppressing the bacterial growth (Sivapalan 1982, Constantinides and Fownes 1994).

Application of mulching materials may have suppressed the growth of pathogenic organisms (Table 2.12) and such was also found by Kloepper *et al*. (1980). There is also evidence to show that balanced fertilizer applications also can suppress pathogenic organisms (Anonymous 1999). In the FAME analysis, the fatty acid of 12:0 indicates the presence of eukaryotic cells (White 1983). This peak was absent in the analyses of the initial pot soil of the JCU Nursery Trial, which suggests that it has been derived from the plant residues in the mulches added to the pots. Fatty acid biomarker 16.1 w5c was interpreted as evidence for arbuscular mycorrhizal fungi in plant roots and in soil (Olsson 1999).

Increasing fungal populations in mulch treatments (Table 2.13) compare to those under the tea mulch, is also suggested by the presence of the main discriminating peak 18:2 w6c, which is commonly used as a fungal biomarker (Pankhurst *et al*. 2001b). Fungal fatty acid biomarker 18:2 w6c was low at the end of the experiment compared to its initial value (Table 2.12 and 2.13), possibly as a consequence of damage to filamentous fungi caused by sieving and disturbing the soil (Petersen and Klug 1994).

Favourable niches for the propagation of microbial populations are indicated by the increases in total fungi and in the fungi/bacteria ratio in the soils under the grass and *Calliandra* mulches (Table 2.13), and by the increased mycorrhizae under the grass mulch and pH amendments (Table 2.8). Similarly, the high soil microbial carbon

contents and high fungal and mycorrhizal population abundances also indicate the favourable nature of the treatments that included the rainforest inoculum (Table 2.7).

2.5.2 Plant growth parameters

Of all the mulch treatments, only the grass mulch increased the dry shoot and total biomasses of the indicator plants, and the tea mulch reduced the dry root masses (Table 2.14). Growth of tea plants is highly dependent on soil moisture, nutrient availability, and soil pH. Since all the soils were kept at 50-80% of field capacity there was no limitation in soil moisture. The optimum soil pH for tea is 4.5 -5.5. Of the mulch treatments, only the grass mulch increased the initial soil pH from 4.43 to 4.54 (Table 2.5), which is closer to the desired value for tea (Natesan 1999). The grass treatment also increased the organic carbon, cation exchange capacity, soil pH and soil microbial biomass carbon of the soil (Table 2.5). Soil microbial biomass nitrogen significantly increased in the soils under the grass mulch while those under the tea and legume mulches showed a rapid decline of microbial biomass nitrogen (Fig. 2.2a). The soils under the pH modifiers and rainforest inoculum treatments did not show any significant differences in microbial biomass nitrogen contents (Figs 2.2b, 2.2c).

Improvements in plant growth parameters induced by Minplus applications may have been influenced by associated increases in organic carbon contents, cation exchange capacity, soil pH, and total phosphorus in the treated soils (Table 2.6, 2.16). Unlike dolomite, Minplus has 0.7% of P_2O_5 as one of its constituents, a factor which may have contributed to the higher total phosphorus in the soils under the Minplus treatments compared to that of the soils under the control treatments (Table 2.6).

There was a significant reduction of plant growth parameters, such as height, shoot and root masses, and total biomass, under the dolomite treatments (Table 2.15). It was found that, in the dolomite-treated soils, the calcium levels were about 800 ppm – a level which is thought to be excessive for optimal growth of tea given that the optimal level of calcium of 100 - 300 ppm is adequate to sustain growth of tea plants (Sivasubramaniam 1981).

The rainforest soil inoculum may have been responsible for increasing the soil microbial biomass, which in turn has increased the nutrient recycling and availability (Table 2.7).

2.6 CONCLUSIONS

1. The results from the present study suggest that the **optimal treatments** for the early growth of transplanted tea seedling are grass mulch (FW 3500 kg ha^{-1}) and Minplus rock dust (2500 kg ha^{-1}), or a legume mulch (FW 3500 kg ha^{-1}) and Minplus rock dust, and the addition of a rainforest soil inoculum in combination with either of the Minplus treatments.

2. Among the mulching materials, grass had the largest effect on the soil properties of interest and increased the soil organic carbon, pH, cation exchange capacity, and microbial biomass carbon both in the presence and absence of tea plants.

3. Application of grass and legume mulches increased the beneficial populations of gram positive bacteria, fungi, mycorrhizae, and also increased the fungi/bacteria ratio.

4. Grass mulch improved the growth of tea as measured by dry shoot mass and total dry biomass.

5. Among the pH amendments, Minplus increased the soil organic carbon, pH, and cation exchange capacity and total phosphorus in the soils in the presence of tea plants, while dolomite increased the soil pH, soil microbial biomass carbon and total phosphorus in the presence of tea plants.

6. Dolomite and the rainforest soil inoculum together raised the soil pH in the no mulch, grass, and legume mulch treatments. The combination of Minplus with the rainforest inoculum increased soil pH only under the grass mulch, while the other mulches reduced both the pH and soil microbial biomass carbon contents of the underlying soils.

7. In the presence of the rainforest soil inoculum, Minplus increased the soil organic carbon and cation exchange capacity.

8. There were no differences in soil organic carbon contents and cation exchange capacity of the soils among the treatments involving grass and legume mulches and both of the pH amendments in the presence of tea plants.

9. The addition of an inoculum of rainforest soils increased the soil microbial biomass carbon.

10. Only grass mulch increased soil pH and soil microbial biomass carbon with dolomite, and increased soil pH with Minplus.

Treatments to be avoided because they do not produce active tea seedling growth are: grass mulch and dolomite, tea mulch in any combination with dolomite, Minplus rock dust, tea mulch, and rainforest soil inoculum.

1. Legume mulch reduced soil pH, soil microbial biomass carbon, and plant available phosphorus, while tea mulch reduced the values of all of the tested soil parameters.

2. Tea mulch reduced organic carbon and cation exchange capacity of the soils under dolomite, and reduced cation exchange capacity under the Minplus treatments.

3. In the presence of the rainforest soil inoculum, the legume and tea mulches reduced soil pH.

4. Though the presence of the rainforest soil inoculum led to higher dry shoot masses, it was not reflected in the total biomass produced by the plants.

5. There was no significant interaction of mulching materials x rainforest soil inoculum on the growth of tea plants.

6. The improvement in organic carbon content of the soil under the grass mulch and dolomite combination was not reflected in the growth of tea plants in terms of height, shoot mass, root mass and total biomass. In fact, the growth parameters were reduced by the grass mulch - dolomite combination when compared to the control.

CHAPTER 3

THE YOUNG TEA TRIAL, ST. COOMBS ESTATE, SRI LANKA

3.1 INTRODUCTION

"Young tea" is regarded as a growth stage of the tea plant soon after seedlings are planted in the field. For the initial two years after planting it is referred to as "young tea"; from when harvesting commences in the 3rd year after planting until pruning begins in the 5th year, it is known as "immature tea". After raising the plant for 9 months in the nursery, the tea is planted in the field along the contour at a spacing of 1.2 m between the rows and 0.6 m between the plants along the row. The planting hole has dimensions of 30 cm diameter and 45 cm deep. Generally, tea planting is carried out during the monsoon season. After planting, the inter-row space is mulched with grasses to give an adequate soil cover for erosion control. The amount of grass required to mulch is approximately 30-35 tonnes fresh weight ha^{-1}. Before planting the tea seedlings, the soil is usually conditioned with mana grass (*Cymbopogon confertiflorus)* or Guatemala grass (*Tripsacum laxum*) for 18-24 months to reduce toxic effects of previous old tea plants; to improve the porosity and structure of the compacted soil of the tea land; and to reduce the pest and disease populations in the soil, especially root disease pathogens and nematodes. Before planting tea seedlings, grasses are planted for soil conditioning, and both medium height and tall shade trees are planted in the inter-row spaces. By the time the tea is planted, the shade trees provide the young tea plants with some protection from excessive insolation. The commonly used medium shade trees are dadap *Erythrina lithosperma*, or *Calliandra calothyrsus* (Somaratne 1986). They are planted in every third inter-row at spacings of 3.6 m between rows of trees,

and at 3.0 m intervals along the rows. Similarly *Grevillea robusta* is planted as a high shade tree at spacings of 6 m x 6 m.

Mulching is a cultural practice undertaken to conserve the soil and soil moisture in young tea, where the soil is exposed without cover immediately after planting and in mature tea where there is bare soil (Grice, 1990). Mulching also improves the organic carbon content, bulk density, and structure of the soil (Gupta *et al*. 1977). Soil chemical properties such as pH, cation exchange capacity, the availability of nutrients, and biological properties such as microbial biomass and enzyme activities are also enhanced by mulching (Gianfreda and Bollag 1996).

Tea soils are usually acidic due to the strong weathering environment of the humid tropics and to the presence of aluminium-rich soil minerals such as gibbsite. Soil acidity is further aggravated by the extended use of nitrogenous fertilisers such as urea and ammonium sulfate to obtain high crop production in most tea estates (Ishaque and Cornfield 1974; Sandanam *et al*. 1978; Walker and Wickramasinghe 1979; Golden *et al.* 1981; Wickramasinghe *et al.* 1981; Nioh *et al*. 1995).

In young tea, the soils are exposed as a consequence of the inadequate development of a canopy cover of the tea plants. Since urea-based fertilisers suffer high volatilization losses due to higher temperature in the exposed area, ammonia-based fertilisers are applied instead to minimize the volatilization losses of nitrogen. Continued use of ammonium sulfate fertilisers at rates of 200-300 kg nitrogen ha^{-1} over the past 20-30 years has contributed to increasing acidity in the tea soils of the highlands of Sri Lanka. Sandanam *et al*. (1978), Sivapalan (1982), and Sivapalan *et al*. (1983) showed that

mulches of Guatemala grass and mana grass which are currently used for the rehabilitation of tea estates, have high C/N ratios (greater than 25), and therefore low nitrification rates compared with mulches of the legume dadap, which has a high soluble nitrogen content and a C/N ratio of 8.

The addition of nitrogen-poor residues to the soil results in nitrogen immobilization as a consequence of assimilation of nitrogen by heterotrophic organisms (Sivapalan 1982; Wickemasinghe *et al*. 1985). Manipura *et al*. (1969) and Wijeratne *et al.* (1994) have shown that, in young tea, the incorporation of Guatemala grass mulch may produce significant increases in the nitrogen and exchangeable potassium contents of the top layer of the soil compared with those of the untreated controls.

The maintenance of an optimal soil pH (4.5-5.5) is an important soil management strategy in tea cultivation (Natesan, 1999). Generally dolomite or lime is applied to tea soil as an amendment when the pH is less than 4.5.

Very little is currently known about the effects on the fertility of tea land soils of specific mulches and soil pH amendments (other than dolomite) in terms of their quantity, quality, and rates of application. Therefore, studies were carried out to determine the effects of a combination of mulch materials and soil pH amendments on both young tea plants that are discussed in the present chapter, and on older, mature tea that are discussed in Chapter 4.

3.2 AIMS OF THE STUDY

- To study the effects of mulching materials with a range of C/N ratios on the chemical and biological properties of the soil of a young tea field;
- To study the effects of soil pH amendments (dolomite and Minplus rock dust) on the chemical and biological properties of the soil in a young tea field;
- To study the effects of combinations of mulches and soil pH amendments on the chemical and biological properties of the soil in a young tea field;
- To study the effects of mulches and soil pH amendments on the growth and yield of young tea.

3.3 MATERIALS AND METHODS

3.3.1 Study site

The experimental plots were located in Field No. 10 of the St. Coombs Estate of the Tea Research Institute of Sri Lanka, at Talawakelle, Sri Lanka (longitude 80^0 41' E; latitude 6^0 55' N; altitude 1382 above mean sea level). The topography is steeply dissected with an average slope of 22 degrees.

The soil is classified as a fine mixed Tropudult (Panabokke 1970) of the Mattakelle series (Dassanayake and Hettiarachchi 1999). This site belongs to up-country wet zone ;WU2 (Watson, 1986). The soils are well drained with a weak, subangular blocky surface soil structure grading to a moderate subangular blocky structure in the subsoil. The surface soil colour is dark yellowish brown moist (10YR 4/6 moist, 10YR 4/4 dry) grading to strong brown (7.5YR, 4/6 – 7.5YR 5/6) in the moist subsoil. The soil has many properties in common with those of the Galamara Series soil, Innisfail, Australia

(Murtha 1983) that was used in the Nursery Trial at James Cook University and described in the previous chapter.

3.3.2 Climate

The study site is situated in an up-country wet zone with an annual average rainfall of 2250 mm. This zone is characterised by dry weather with cool nights and warm days from mid January to late March. There are usually two monsoonal periods with high humidity between April and June and between October and December (Fig 3.1 a, b).

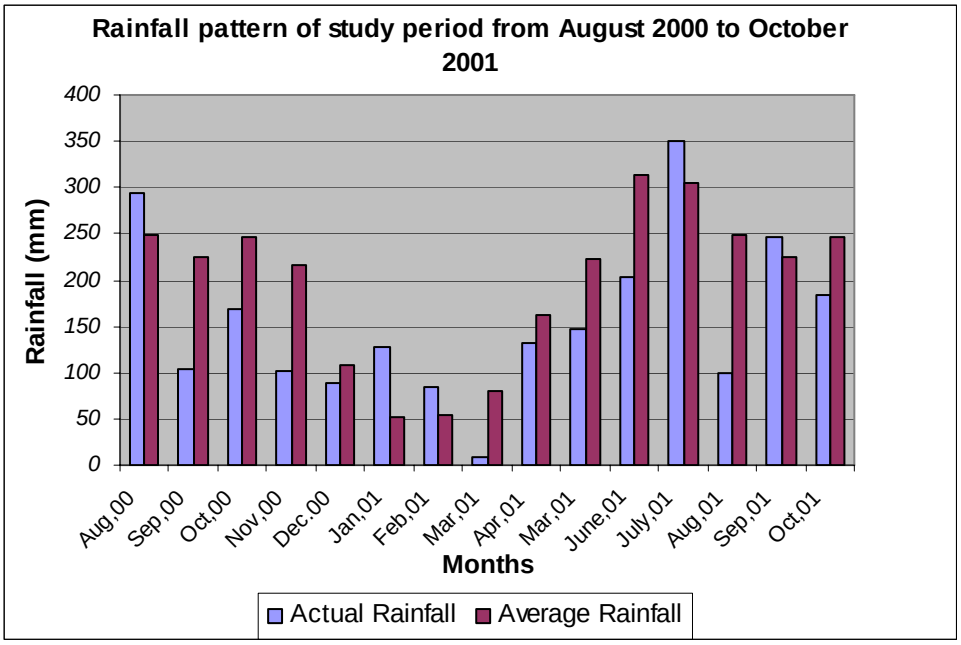


Figure 3.1 a The rainfall pattern at the experimental site at Talawakelle, over the period of the trial (August 2000 to October 2001). Source: Annual Report of Tea Research Institute of Sri Lanka (2000 and 2001)

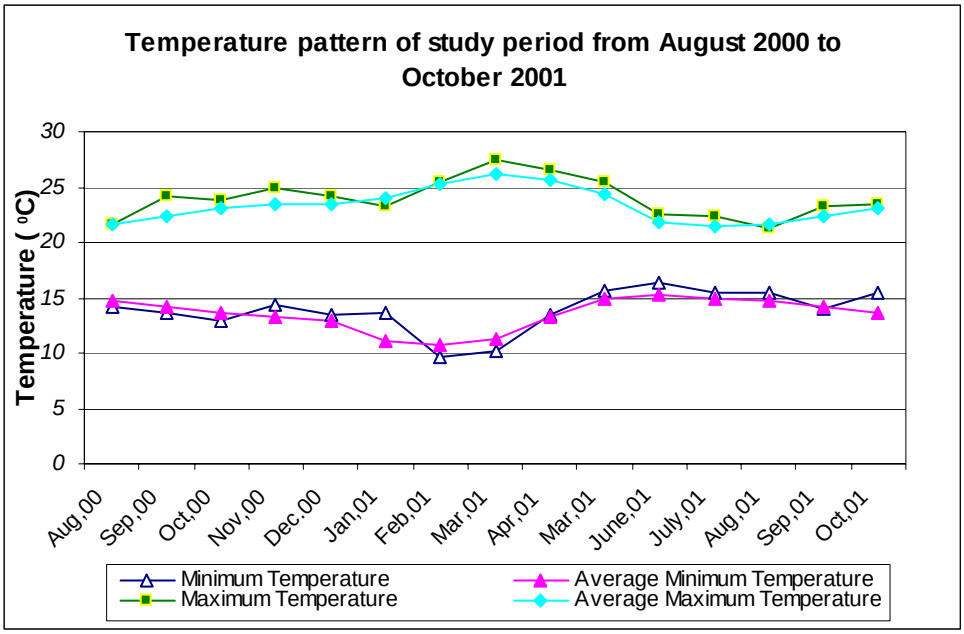


Figure 3.1 b The temperature pattern at the experimental site at Talawakelle, over the period of the trial (August 2000 to October 2001). Source: Annual Report of Tea Research Institute of Sri Lanka (2000 and 2001)

3.3.3 Plant material

The experiments were carried out using the tea clone TRI 4071. The plants were two years old (young tea) when the trial was initiated on August 2000. The trial ended in October 2001.

3.3.4 Experimental Design

A randomized complete block design with two factors was used in the experimental area at St. Coombs Estate of the Tea Research Institute of Sri Lanka. Field plots were established (4.5 m x 3.5 m) for 15 soil treatments with 5 replicates in 5 blocks. The 75 plots were marked out in June 2000 over an area of approximately 60 m by 40 m of Field No. 10. Each plot was surrounded by a guard row which separated the treated area in order to prevent treatment effects in any adjacent plots to influence the experiment.

The assessment area of each plot was 15.5 m^2 and the individual plots contained 15-16 young tea plants.

3.3.5 Treatments

The treatments were combinations of mulching materials and soil pH amendments. The five mulching materials used were selected on the basis of their availability in the Sri Lankan tea gardens, and to give a wide range of C/N ratios, as follows:

- mana grass (*Cymbopogon confertiflorus*) at 35 tonnes (fresh weight) ha^{-1}. The mana grass was cut at 30 cm above the ground before it flowered, and was transported from the places where it had been established for rehabilitation purposes.
- dadap legume (*Erythrina lithosperma*) at 35 tonnes (fresh weight) ha^{-1}. The dadap legume mulch consisted largely of leaves and some branches up to 2.5 cm diameter that had been lopped from shade trees available in the tea estates, and were transported to the experimental plots and spread over the surface of the plot.
- refuse tea (*Camellia sinensis*) at 25 tonnes (fresh weight) ha^{-1}. "Refuse tea" is a waste product of the tea manufacturing process and is the partly ground, brown stalk and fibrous particles remaining after separating the commercial components of the manufactured tea.
- lemon grass (*Cymbopogon nardus*) used as live mulch at the rate of 20,000 plants ha^{-1} with a spacing of 15x15 cm in the inter-rows according to the recommendations of Tea Research Institute of Sri Lanka. Lemon grass was used as live mulch because its profuse root system promotes the growth of soil microbial populations. Also, lemon grass has commercial value for extensive use as an indigenous medicine and as an oil extract.

- untreated control plots maintained without adding any mulch but subjected to the normal leaf fall from the canopy of the tea plants.

The pH amendments used were dolomite and Minplus (crushed basaltic rock) both as once-only application of 1000 kg ha^{-1}. The soil pH modifiers were applied according to the recommendations of the Tea Research Institute of Sri Lanka and the rate of dolomite application was based on the pH value of the soil:

- if the soil pH was less than 3.8, 2500 kg dolomite ha^{-1} was used
- if the soil pH was 3.9 - 4.2, 2000 kg dolomite ha^{-1} was used
- if the soil pH was 4.2 - 4.5, 1500 dolomite kg ha^{-1} was used
- if the soil pH was greater than 4.5, 1000 kg dolomite ha^{-1} was used

The dolomite application is recommended once per cycle (once every 5 years) in tea to correct the soil pH condition, but not as a split dosage. Minplus was applied at the same application rate (1000 kg ha^{-1}) that was used for the dolomite, but it is now recognised that this was a much lighter rate than that recommended for Minplus use in a North Queensland agricultural context (Coventry *et al.* 2001).

All the other cultural operations such as fertilizing, pest control, weeding, etc., were carried out in accordance with the recommendations of the Tea Research Institute of Sri Lanka. Treatments were imposed in a randomized factorial design in August 2000 and the plants were harvested 14 months later in October 2001.

Fertiliser was applied at three-month intervals using 1200 kg ha^{-1} of the T 200 fertiliser mixture and it split into four doses of 300 kg ha^{-1} (Sulfate of Ammonia 100 parts,

Saphos Phosphate (Superphosphate) 50 parts, Muriate of Potash 25 parts, Magnesium Sulphate 25 parts) according to the recommendation of Tea Research Institute of Sri Lanka. The T 200 fertiliser mixture contains 20.6 % N, 32.5 % P_2O_5, 60 % K_2O, and 24 % MgO.

Details of the mulch and fertiliser applications are set out in Tables 3.1 and 3.2, respectively. Fertiliser was applied immediately following the collection of soil samples for analysis throughout the experimental period. The complete list of treatments of mulch and soil pH amendments used in the field trial is set out in Table 3.2. The chemical properties of the mulches are shown in Table 3.3

Table 3.1 Time of application of mulch and fertiliser, and of soil sampling of the young tea trial in Sri Lanka

Treatments	**Times of Application**				
Refuse tea Mulch	August 2000	February 2001	October 2001		
Mana grass Mulch	August 2000	December 2000	May 2001	September 2001	
Dadap legume Mulch	August 2000	December 2000	May 2001	September 2001	
lemon grass (live) mulch	Planted in August 2000	-	-		
Dolomite and Minplus soil pH modifiers	August 2000	-	-		
Fertiliser (T200 @ 300 kg/ha)	June 2000	October 2000	January 2001	April 2001	July 2001
Soil Sampling	June 2000	October 2000	April 2001	October 2001	
Growth assessment of tea plant	-	-	-	October 2001	

Table 3.2 The treatment combinations used on the young tea trial in Sri Lanka

	Treatment Code *	Mulch treatment	Soil pH modifier treatment
1	MoPo	No mulch	No pH modifier
2	MoPd	No mulch	Dolomite
3	MoPm	No mulch	Minplus
4	RtPo	Refuse tea	No pH modifier
5	RtPd	Refuse tea	Dolomite
6	RtPm	Refuse tea	Minplus
7	MaPo	Mana grass	No pH modifier
8	MaPd	Mana grass	Dolomite
9	MaPm	Mana grass	Minplus
10	DaPo	Dadap legume	No pH modifier
11	DaPd	Dadap legume	Dolomite
12	DaPm	Dadap legume	Minplus
13	LePo	Live lemon grass	No pH modifier
14	LePd	Live lemon grass	Dolomite
15	LePm	Live lemon grass	Minplus

* Organic mulches: Mo- No mulch, Rt- Refuse tea (25,000 kg ha^{-1}), Ma- Mana 35,000 f.wt. kg ha^{-1}, Da- Dadap (35,000 f.wt. kg ha^{-1}, Le- lemon grass (20,000 plants ha^{-1})

* Soil pH modifier: Po – No mulch, Pd – Dolomite (1000 kg ha^{-1}), Pm – Minplus (1000 kg ha^{-1})

Table 3.3 Chemical compositions of oven dried (85 ^{0}C) mulch materials applied to the young tea trial, Sri Lanka

Mulch material	Total N %	Carbon %	C/N ratio	Total P %	Total K %	Polyphenol %	Lignin %
Tea litter (control)	3.50	40.5	11.6	0.20	1.50	11.1	19.0
Refuse tea	3.15	34.44	10.93	0.28	1.85	22.53	9.52
Mana	1.40	40.32	28.80	0.14	1.40	1.03	29.68
Dadap	4.55	38.22	8.40	0.39	2.15	2.60 (Sivapalan 1982)	13.6 (Sivapalan 1982)

3.3.6 Soil and plant analysis

3.3.6.1 Soil sample preparation

Soil samples of 0-15 cm depth (approximately 1 kg) were collected from four randomly selected locations within each plot, bulked, and a sub-sample for analysis was taken from the bulk sample. Part of the sub-sample was air dried and passed through a 2 mm sieve prior to chemical and physical analysis. The remaining soil was sieved through 2 mm mesh and stored at 4 ^{0}C for microbial biomass carbon analysis.

3.3.6.2 *Plant sample preparation*

Samples of mulching materials (dadap legume, mana grass, lemon grass, refuse tea) of approximately 100 g were collected from the field , cut into 2 cm lengths, and dried for 48 hours at 85 ^{0}C then ground to pass a 0.4 mm mesh according to the procedures for chemical analysis that are set out in the following section. All samples were kept in desiccators for further chemical analysis.

3.3.6.3 *Chemical and microbial analysis*

The methods used for soil and plant analysis have been described above (Sections 2.3.4.1 and 2.3.4.2). Soil microbial biomass carbon was measured by the chloroform fumigation extraction method (section 2.3.4.3.1) and soil organic carbon content was determined by the method described by Sparling *et al*. (1990), which is based on a modified Walkley Black method (Rayment and Higginson 1992b). Additionally, six months after application of treatments, soil respiration rate was also measured using the method of carbon dioxide evolution from undisturbed soils (Black 1968) (Appendix 11). Decomposition rates of mulch materials were measured using a litter-bag technique (Wardle *et al*. 1999) (Appendix 12). Soil texture was assessed by the method described

by Gee and Bauder (1979) (Appendix 13). Chlorophyll content of leaves was measured by using a chlorophyll meter (Spectrum model SPAD-502) (Appendix 14).

A functional analysis of the structure of the soil microbial community was carried out by Dr C. Pankhurst, using the FAME (Fatty Acid Methyl Ester) analysis in the laboratories of CSIRO Land and Water, Adelaide, Australia.

3.3.7 Decomposition rates of mulching materials

The decomposition rate of mulching materials under the dolomite treatments, the normal practice recommended by Tea Research Institute of Sri Lanka, was determined by a litter-bag technique as set out in the following section.

3.3.7.1 Litter- bag technique

Litter decomposition rates were determined following the litter-bag methods of Wardle *et al*. (1999), by employing 30 x 45 cm bags made from nylon mesh with mesh openings of 0.5 x 0.5 mm. Leaves of dadap and mana grass were kept and withered in the sun to a constant air-dry weight. Refuse tea was used at 5% moisture content. A sample (200 g) of each of the mulches was put into the bags, which were then placed directly on the soil surface after scraping away any mulch that was already there. Four bags were used for each mulch, and the bags touched only the surface soil**.** Therefore, the litter in the bags take into account the influences of the meso- and macrofauna of the soil. The oven dry weights of remaining mulches in the litter bags (85° C, in 48 hrs) were determined at four different times during the period of experiment (Wardle *et al.* 1999).

The decomposition rates of each of the mulches were determined according to the method set out in Appendix 12, and were used to assess the effect of mulch composition on the half-life of the mulching materials.

3.3.8 Tea quality analysis

Tea was harvested and manufactured by an 'orthodox' manufacturing procedure (Dahanayake and Ziyad 2002) (Appendix 15). One year after application of the treatments and the tea quality was tested by preparing the made tea using the mini manufacture method described by Samaraweera (1986) and sending the samples to three professional tea tasters. Tea quality was assessed in terms of appearance, colour, strength, flavour, and infusion (Keegel 1983).

3.3.9 Statistical analysis

Principal components and factor analyses were used draw out the effects and possible interactions of all of the measured soil and plant attributes. A Kaiser-Mayer-Olkin (KMO) measure of sampling and Bartlett's tests (McArdle 1999) were used to ensure the overall accuracy of the analyses. The minimum requirements of the data for a principal components analysis were fulfilled in this trial since it had a Measure of Sampling Adequacy greater than 0.5. A significance level of $p < 0.05$ was used in all of the statistical comparisons made.

3.4 RESULTS

The effects of treatments on some soil chemical and biological parameters and growth parameters of the young tea, Sri Lanka, are summarised in Table 3.4. Only a limited number of treatments affected soil properties (pH, microbial biomass carbon, and respiration rates) and fewer treatments affected plant properties (chlorophyll content of leaves, and the yield responses of the young tea plants).

Table 3.4 Summary of the response of treatments on soil chemical and biological parameters and plant growth parameters

Treatment	Soil Parameters									Plant Parameters		
	OC	**pH**	**TN**	**TP**	**AP**	**CEC**	**BD**	**MB-C**	**Res**	**Chl**	**Cir**	**Yld**
Mulch												
No mulch	-	-	-	-	-	-	-	-	-	-	-	-
Refuse tea	Ns	*	Ns	Ns	Ns	Ns	Ns	*	*	Ns	Ns	*
Mana grass	Ns	*	Ns	Ns	Ns	Ns	Ns	*	Ns	Ns	Ns	Ns
Dadap Legume	Ns	Ns	Ns	Ns	Ns	Ns	Ns	*	Ns	Ns	Ns	*
lemon Grass	Ns	Ns	Ns	Ns	Ns	Ns	Ns	*	ND	*	Ns	Ns
Soil pH modifier												
No pH modifier	-	-	-	-	-	-	-	-	-	-	-	-
Dolomite	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
Minplus	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
Interactions												
pH x mulch	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns

* = significantly increased compared to the control, $p < 0.05$
ND= Not determined
- = Control plots
OC: Soil organic carbon, pH: Soil pH, TN: Total nitrogen, TP: Total phosphorus,
AP: Plant available phosphorus, CEC: Cation exchange capacity, BD: bulk density, MB-C: Microbial biomass carbon, Res: Soil respiration, Chl: Chlorophyll content, Cir: Stem circumference, Yld: yield of made tea

3.4.1 Mulch applications

According to the mulching material applied and chemical analysis of plant material and refuse tea, the amounts of organic carbon, total nitrogen, and total phosphorus derived from the mulch materials were calculated and are presented in Table 3.5. Lemon grass was used as live mulch and as clippings that were cut and placed on the soil surface at the 9th month.

Table 3.5 Actual dry masses of mulch applied to the experimental plots (dry weight; tonnes ha^{-1}) and cumulative amounts of the nutrients applied (kg/ha)

Time of mulch application	Refuse tea				Dadap legume				Mana grass			
	DW	C	N	P	DW	C	N	P	DW	C	N	P
August 2000 (Month 1)	10	340	30	3	6	138	16.8	1.2	10.6	455	10.6	1.06
December 2000 (Month 4)	0	-	-	-	6	276	33.6	2.4	10.6	911.6	21.2	2.12
February 2001 (Month 6)	10	680	60	6	0	-	-	-	0	-	-	-
May 2001 (Month 9)	0	-	-	-	6	414	50.4	3.6	10.6	1367.4	31.8	3.18
October 2001 (Month 14)	0	680	60	6	0	414	50.4	3.6	0	1367.4	31.8	3.18

DW= Dry weight (t/ha), C= Organic Carbon, N= Nitrogen, P= Phosphorus
0 = No application of mulch material
- = No added nutrients

Results of the litter decomposition experiment are shown in Fig. 3.2, and the calculated decay constants are shown in Table 3.6.

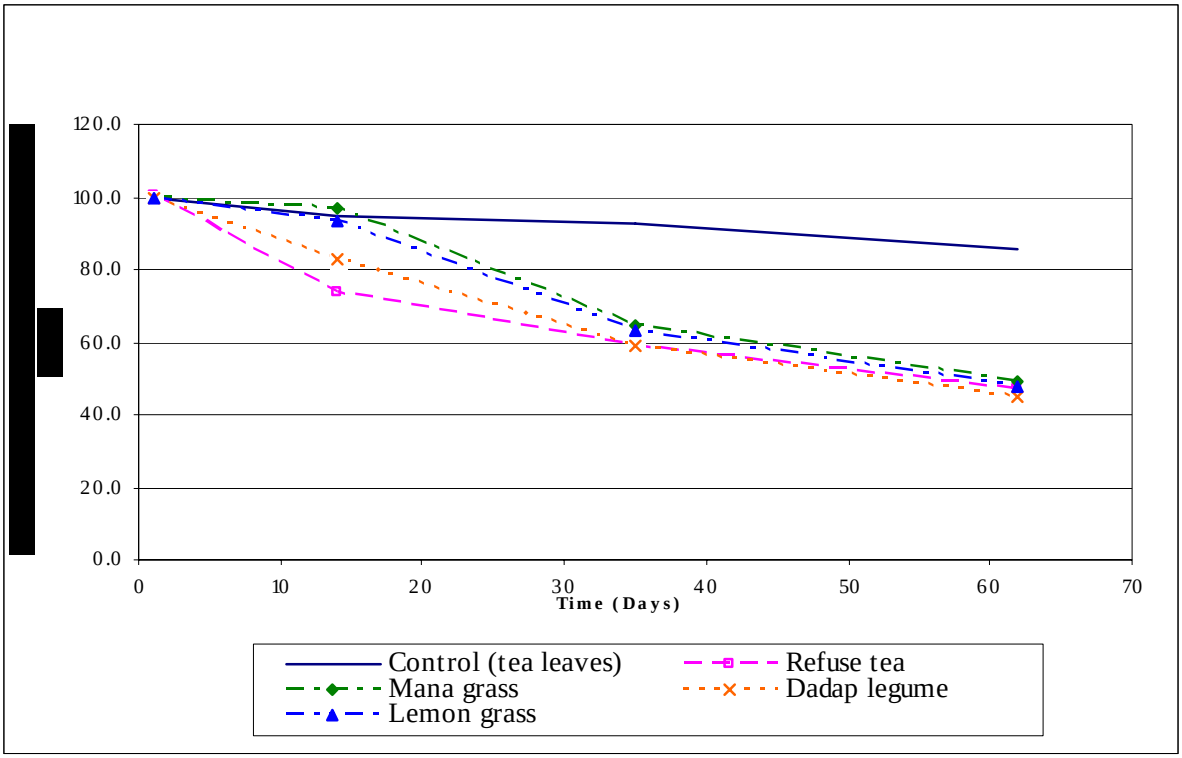


Figure 3.2 Residual oven dried mass of litter remaining in the litter bags after specific time intervals on the bare soil surface of experimental plots in young tea (% of original weight). Treatment control refers fallen tea leaves.

Table 3.6 Decay constants and related parameters for mulch materials on the soil surface under the young tea, Sri Lanka.

Material	**C/N ratio**	**Coefficient of determination** (R^2)	**Decay constant, (k)** [day^{-1}]	**Half life** [days]
Control (fallen tea leaves and twigs)	10	0.96	0.0023	210.4
Refuse tea	11	0.95	0.0119	58.4
Mana grass	29	0.96	0.0125	55.4
Dadap legume	8	0.99	0.0133	52.1
Lemon grass	14	0.98	0.0130	53.3

Except for the control (fallen tea leaves), all the mulching materials had half lives in the range of 52-58 days (Table 3.6). Visual observations of mulch disappearance as decomposed, powder-like material in the field agreed well with the calculated decay constants.

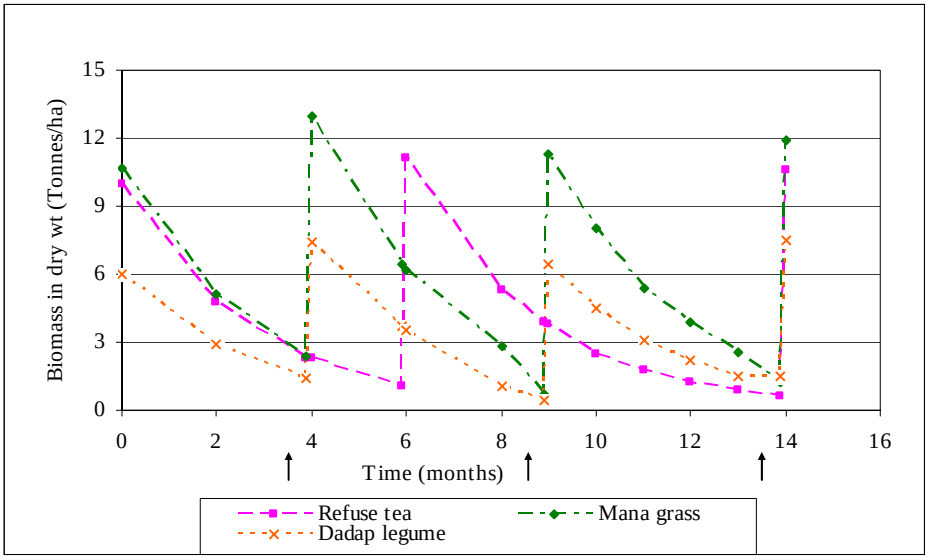


Figure 3.3 Dry biomass (tonnes ha^{-1}) of mulch remaining on the soil surface with time. Arrows indicate the months that soil samples for analysis were taken.

The application of mulching materials was determined by the amount of soil cover provided to the young tea, and was based on the Tea Research Institute's recommendations regarding mulch covers to the growers. Because the decomposition rates of the mulching materials differed, the refuse tea was replaced once in 6 months compared to dadap and mana which were replaced at 4 month intervals (Fig. 3.3). Therefore, refuse tea was added only twice and mana and dadap were added three times during the experimental period. The total amount of carbon added to the soil surface in the refuse tea,dadap, and mana grass treatments were 6.8, 6.9 and 12.9 tonnes ha^{-1} , respectively during the experimental period (Table 3.5). Lemon grass was tested as

living mulch and it was lopped once and mulched; the amount of carbon added was only 0.4 tonnes ha^{-1}.

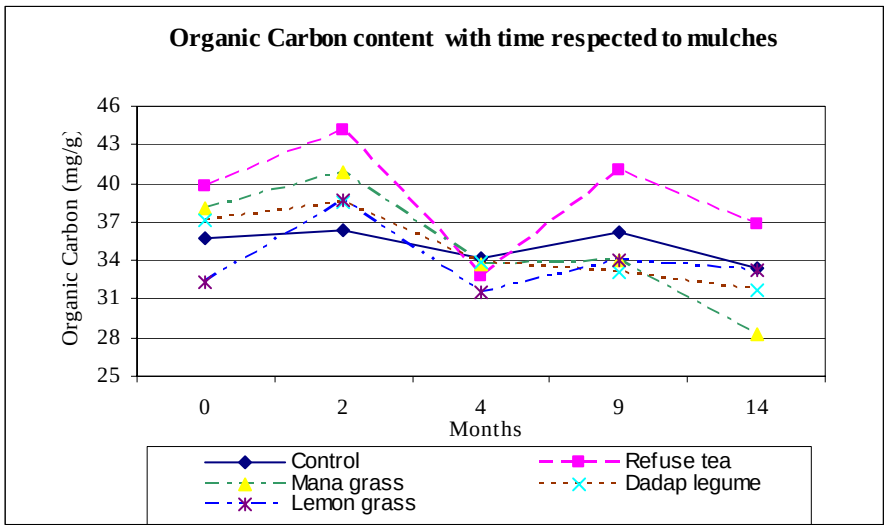


Figure 3.4 (a) Changes of organic carbon with remaining biomass of mulch material in the soil.

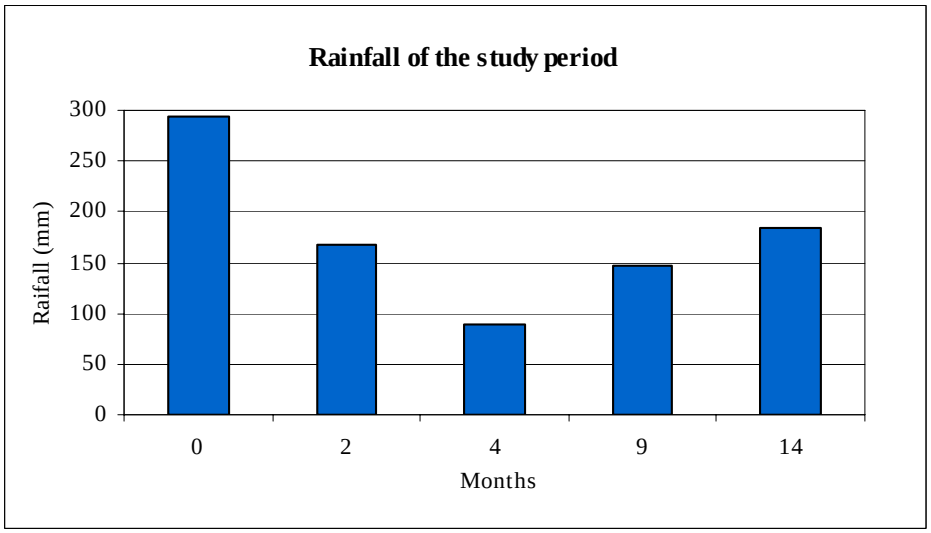


Figure 3.4 b. Rainfall at the study site between August 2000 and October 2001. Source: Unpublished data from the Sri Lanka tea Research Centre, Talawakelle.

The organic carbon content of the soil below the various mulch treatments fluctuated primarily with rainfall (Fig. 3.4a and b). The highest soil organic carbon level was maintained under the refuse tea plot, and the lowest was in the soil under the lemon grass plots (Fig. 3.4a)

Fig. 3.5 indicates a slight positive relationship between the soil organic carbon levels of the various mulch plots and rainfall during the study period. There was a higher mineralisation rate in the soils under the refuse tea, followed by that under the mana and dadap plots compared to the soils of the control and lemon grass treatments (Fig. 3.5). Soil moisture is an attribute closely associated with the mineralisation process, but since no soil moisture measurements were routinely carried out on the mulch plots, actual rainfall has been used as a proxy for soil moisture.

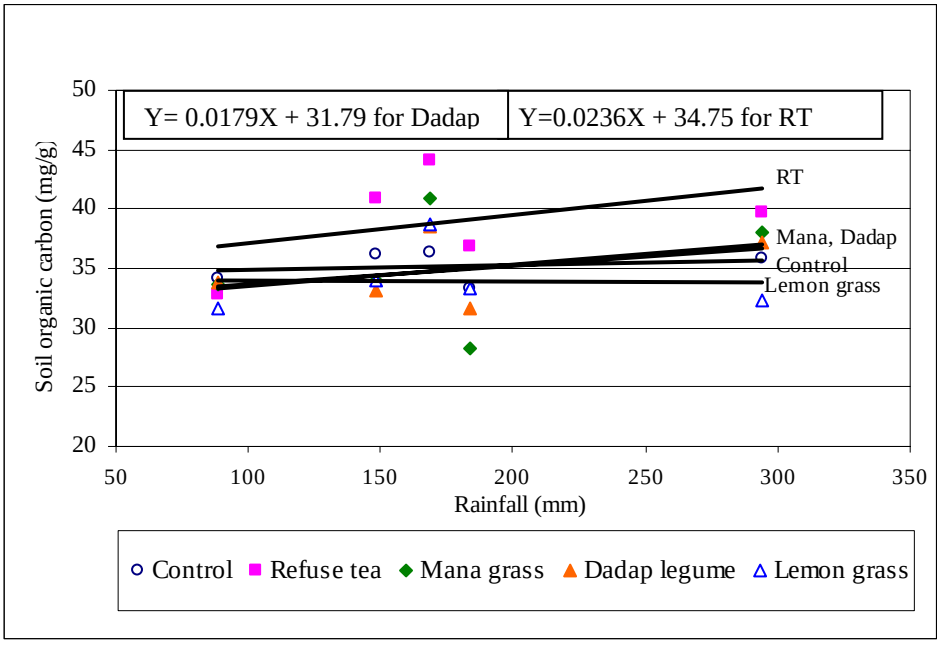


Figure 3.5 Changes in rainfall and organic carbon content of the soils under mulch-treated plots in young tea, Sri Lanka.

3.4.3 Effects of mulches and pH amendments on soil properties

The mulch x pH treatment combinations produced no significant differences in soil properties (Appendix 16). Therefore, all of the soil pH amendment data were pooled and subjected to a principal components analysis (Fig 3.6 and Appendix 16).

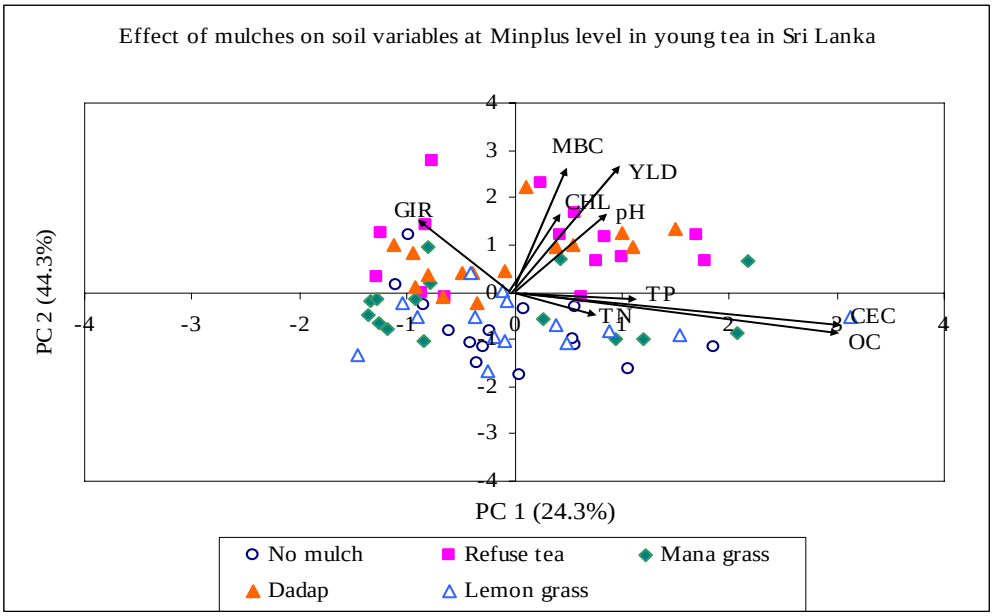


Figure 3.6 Principal components analysis of the effects of various mulches on soil properties and on growth parameters with pooled pH modifiers in Young Tea, Sri Lanka.

Soil pH and microbial biomass carbon contents showed stronger responses to mulching treatments than did organic carbon, cation exchange capacity, total nitrogen, and total carbon. Similarly, plant growth parameters such as yield and chlorophyll showed stronger responses under the refuse tea and legume mulches than under the other mulches (Fig. 3.6). The control, and mana and lemon grass mulch treatments did not influence the soil microbial biomass carbon content, soil pH, and tea yield (Fig. 3.6). The soil organic carbon content and cation exchange capacity showed moderate responses to all of the mulches and the untreated control. Stem diameter showed

moderate responses to no mulch (control) and to the lemon and mana grass mulches (Fig. 3.6).

The results of the factor analysis of the effect of mulching treatments on soil properties and growth parameters are summarized in Table 3.7. These results also show that pH amendments applied to young tea had not altered the soil pH significantly. In fact, none of the pH amendment treatments of the present study were seen to have had any influence on the soil properties or tea growth parameters.

Refuse tea mulch significantly increased the soil pH, soil microbial biomass, soil respiration, and yield of tea compared to the untreated control soil (Table 3.7). The dadap legume mulch raised the soil microbial biomass and the tea yield. This was also evident from the principal components analysis (Fig. 3.6). Soil pH increased under the mana grass and refuse tea mulches, and the soil microbial biomass carbon was higher under all of the mulch materials, and particularly under the refuse tea and dadap legume mulches (Table 3.7).

3.4.3.1 *Relationships between organic carbon and microbial carbon contents*

Figure 3.7 shows a plot of pooled data for 10 estimates of organic carbon and microbial biomass carbon averaging from 30 measurements taken except no mulch plots under the young tea trial, 9 months after the application of treatments.

Table 3.7 Effects of mulches on soil and plant properties in Young Tea, Sri Lanka. Each cell represents the mean values of log transformed data for soil pH and soil microbial biomass; square root transformed data for soil respiration; and raw, untransformed data for chlorophyll content and made tea yield. Back-transformed data are shown in parentheses. Significant ($p < 0.05$) differences from the control treatment are shown by * significantly increased, and # significantly decreased. Means followed by the same lower-case letter within a column are not significantly different $p > 0.05$. ND: not determined

	Soil parameters			**Plant parameters**		
Treatments	**Soil pH** (log)	**Soil microbial biomass –C** (log)	**Soil respiration** kg ha^{-1} day^{-1} (square root)	**Chlorophyll**	**Yield** (made tea kg ha^{-1})	**Yield increases** with respect to "No mulch" (%)
No mulch	0.646 b (4.42)	2.079 d (119.9)	3.2 b (10.24)	60.13 a	1750 b	--
Refuse tea mulch	0.677 a* (4.75)	2.421 a* (263.6)	4.1 a* (16.81)	61.74 a	2089 a*	19.2
Mana grass mulch	0.667 a* (4.64)	2.282 b* (191.4)	3.1 b (9.61)	58.55 b #	1785 b	2.0
Dadap legume mulch	0.662 ab (4.59)	2.460 a* (288.4)	3.7 a b (13.69)	60.40 a	2030 a*	16.0
Lemon grass mulch	0.642 b (4.38)	2.173 c* (148.9)	ND	59.28 ba	1761 b	0.6
LSD ($p < 0.05$)	0.03	0.09	0.60	2.35	180.0	--
CV%	6.4	5.9	1.7	4.0	9.7	

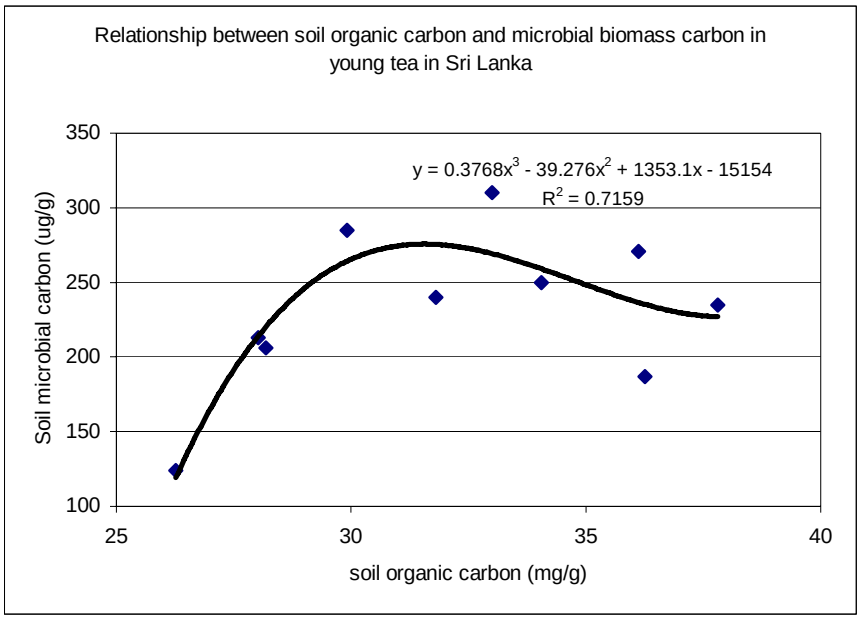


Fig. 3.7 Relationship between soil organic carbon and the microbial biomass carbon content of the soil, 9 months after the application of treatments to the Young Tea, Sri Lanka.

The soil microbial biomass carbon content increased with soil organic carbon and reached a peak value of 275 μg g^{-1} at an organic carbon content of 33 mg g^{-1}, and decreased a little thereafter (Fig 3.7**.**

3.4.4 Effects of mulches on soil microbial population structures

Both bacterial and fungal populations were higher in the soil under the dadap legume mulch and lower under the refuse tea mulch (Table 3.8 and Appendix 17). The ratio of Gram-positive: Gram-negative bacteria was highest in the soils under the dadap legume mulch, followed by those under the mana and lemon grass and then under the refuse tea, and finally the control. This implies that application of dadap and grass mulches have reduced most of the pathogenic bacteria in the soil because Gram-negative bacteria are comparatively low. Beare (1997) reported that surface placement of mulching materials may enhance the fungal populations, while incorporating the mulching materials into the soil may improve the soil bacterial populations (Beare 1997). In the present study,

the mulching materials were placed on the soil surface. Only the dadap legume mulch enhanced the fungal populations, probably due to its rapid decomposition rate, and the relatively rapid loss of the treatment effects from the underlying soils.

Table 3.8 Nature of bacterial and fungal populations in Sri Lankan Young Tea soils, 14 months after application of mulch treatments. Data obtained from a FAME analysis carried out by Dr C. Pankhurst, CSIRO Land and Water, Adelaide.

Treatments	Bacteria Gram-positive	Bacteria Gram-negative	Total Bacteria	Ratio of Gram-positive / Gram-negative bacteria	Fungi	Fungi / bacteria ratio
No mulch	23.74	1.55	25.29	15.32	2.81	0.11
Refuse tea	22.13	1.44	23.57	15.37	1.84	0.08
Mana	23.00	1.25	24.25	18.40	2.91	0.12
Dadap	24.86	1.10	25.96	22.60	3.05	0.12

3.4.5 Effect of soil amendments on the growth of young tea

3.4.5.1 *Chlorophyll content of tea plant leaves*

The chlorophyll contents of the leaves of the young tea plants grown in all of the mulch treatments were similar, except that of the plants grown under the mana grass mulch which had significantly lower chlorophyll contents than the other plants (Table 3.7). All the treatments were fertilized with 437 kg ha^{-1} $application^{-1}$ of the T 750 mixture (containing 240 kg N and 28 kg of MgO) which appears to have been instrumental in maintaining leaf chlorophyll across the treatments, except for the mana and lemon grass treatments which also had the lowest nitrogen and phosphorus contents (Table 3.8). This suggests that the young tea growing in the less fertile soils were less vigorous and consequently produced less chlorophyll.

3.4.6 Effects of soil amendments on the quality of made tea

The assessment by tea tasters on leaf infusion, colour, strength, quality, and flavours of made tea produced from the young tea grown under different mulches and soil pH modifiers indicated that only the leaf infusion was affected by any of the treatments. The leaf infusion was assessed on the tea leaves after brewing the tea and gives an indication of its colour and its evenness. A score was given to each batch of made tea, made from leaves harvested from plants grown under each experimental treatment (Table 3.9).

Table 3.9 Effects of mulch materials and pH amendments on infusions of tea made from the Young Tea, Sri Lanka. The numbers indicate the scores for colour for samples of the made tea: 1=very dull, 2=dull, 3=fair colour, 4=fairly bright, 5=quite bright, 6=bright, and 7=very bright. The number of samples producing the mean values are shown in parentheses. Mean values followed by the same letter are not significantly different ($p > 0.05$).

Treatment	pH amendments			
	Control	**Dolomite**	**Minplus**	**Mean**
No mulch	3.62	3.10	3.04	3.25 (A)
Refuse tea	3.29	3.00	3.32	3.20 (B)
Mana	3.09	3.00	3.00	3.03 (B)
Dadap	2.99	3.42	3.27	3.22 (B)
Lemon grass	3.32	3.97	3.56	3.61 (A)
Mean	3.26 (A)	3.29 (A)	3.23 (A)	
LSD ($p < 0.05$)				0.37
CV%	10.2			

The results indicated that tea made from plants grown under the lemon grass and the control treatments had significantly higher leaf infusions compared to that made from tea grown under all the other mulch treatments. There were no significant differences in tea quality among the soil pH modifier treatments (Table 3.9).

3.5 DISCUSSION

The different rates of decomposition of mulches, indicated by their decay constants was highest for the mulch made from dadap legume, followed by lemon grass, mana grass and refuse tea; the decay constant for the control, consisting of fallen senescent tea leaves and twigs, was the lowest (Table 3.6). The decay constants depend on the quality of the mulching materials, especially their C/N ratio (Seligman and van Keulen 1981), and polyphenol and lignin contents (Tian *et al*. 1997).

Visual observations of the relative order of decomposition of mulch in the field agreed well with the numerical values of the decay constants. The decomposition of mulches is largely controlled by microbial processes, which are influenced by the quantity and quality of substrate availability (Swift *et al*. 1979), soil pH, soil moisture and temperature (Anderson and Nilsson 2001). Further, the quality of plant residues is determined by relative contents of carbohydrates, cellulose, lignin, polyphenol fractions and C/N ratios (Tian *et al*. 1997).

In the young tea field, mulching materials were exposed to solar radiation since there was an incomplete tea canopy. With the exception of the control, the decay constants for all of the mulching materials were in the range of 0.012 - 0.013 d^{-1}. The reported decay constant for carbohydrates is 0.2 d^{-1}, for cellulose 0.05 d^{-1}, and for lignin 0.0095 d^{-1} (Seligman and van Keulen 1981). All the mulching materials used in the present study were composed of carbohydrates, cellulose, hemi-cellulose, lignin, and polyphenols (Seneviratne *et al*. 1998; De Costa *et al* 2001).

Seligman and van Keulen (1981) also reported that the C/N ratio of a mulch will determine the decay constant only when the C/N ratio is greater than 25. The decay constants reported for the mulching materials in this study (Table 3.6) were between those of lignin and polyphenol. Therefore, the differences in the decay constant may be due to variations in the proportion of carbohydrates, lignin, cellulose, and polyphenols in the mulching materials. The dadap legume, with high nitrogen, low lignin, and low polyphenol contents, decomposed rapidly, as was also found by Handayanto *et al.* (1997) in *Erythrina* plant litter. Even though mana grass mulch had a C/N ratio greater than 25 (Table 3.4), there were regular fertiliser additions to the tea at 90 kg N ha^{-1} per application every 3-4 months (Table 3.4). Adding a nitrogenous fertiliser may have reduced the C/N ratio of mana grass mulch to below 25, thus promoting a faster decomposition. This is a case of a non-limiting condition of nitrogen which is known to enhance microbial activity (Trinsoutrot *et al*. 2000).

It is also known that the larger the exposed surface area of the mulch, the faster the mineralisation of nitrogen (Marchner and Noble 2000). In the refuse tea mulch, the exposed area was large but it was not direct contact with soil surface. Further, it had a high polyphenol content, which reduces the decomposition rate (Sivapalan 1982). The control treatment, consisting of fallen senescent tea leaves and twigs had the longest half life probably due to nitrogen limitations. Usually before leaf senescence in the tea plant, nitrogen is reallocated to aerial parts (Sivapalan 1982), and the tea leaves contain 22% polyphenol and 9.5% crude fibre that is rich in lignin and carbohydrates (Sivapalan 1982). The stem contains mostly celluloses and lignin, and Fox *et al*. (1990) suggested that in litter with a high polyphenol and lignin content, the decomposition rate would be

slower. Hence, the low decay constant for the refuse tea treatment is a direct consequence of its biochemical composition.

In the young tea, the mulching materials were well exposed and decomposition rates were therefore fast. The addition of mulching materials had not changed the organic carbon content of the soil significantly at the end of the 14 months field trial (Table 3.4). The amounts of carbon added from refuse tea, dadap, and mana grass mulches were 6.8, 6.9, and 12.9 tonnes ha^{-1} respectively during the study period (Table 3.6).

The contribution from lemon grass mulch to organic carbon enrichment was by the addition of clipped leaves (0.4 carbon tonnes ha^{-1} per application) and rhizo-deposition by root exudates. The amount of carbon contributed from this source was negligible compared to that from the other mulches since lemon grass was used as live mulch and its cover under the young tea was only 20-30%.

The addition of mulching materials did result in an increase in the soil microbial biomasses under all of the mulching treatments compared to the control (Table 3.7). The fatty acid methyl ester analysis at the end of the Young Tea trial indicated that, in the tea rhizosphere, the bacterial population was higher than the fungal population. Pandy (1971) also observed a similar trend of higher bacterial populations compared to fungal populations under different mulch conditions. Pandy (1971) showed that addition of mulching materials to the soil surface stimulates fungal populations compared to their initial levels.

In the present study, the total fungal population was higher under more conventional mulches of dadap and mana compared to those under refuse tea and the control. A similar decline was observed in bacterial populations under the refuse tea mulch. This may possibly be due to anti-microbial properties of the refuse tea mulch (Sivapalan 1982). The ratio of Gram-positive to Gram-negative bacterial populations was higher in the soils under dadap and mana mulches compared to mulch of refuse tea and the untreated control (Table 3.8). This implies that the numbers of Gram-negative bacteria in the soil under dadap and mana mulches are less than in mulches under refuse tea and the control. Even though the soil microbial biomass and soil respiration were higher in soils under the refuse tea mulch (Table 3.7), this was not reflected in the fatty acid methyl ester analysis (Table 3.8).

At the end of fourth months after application of mulch, the weather was dry with the mean soil temperature at 0-15 cm depths rising to 22.2^{o} C. Therefore, a decline in the soil organic carbon content may be mainly due to the rapid decomposition of organic matter on the soil surface where moist, warm soil conditions prevailed (Figs 3.1 a, b).

3.5.1 Soil microbial biomass

In general, microbial biomass carbon contents of the soil reflect the long-term amount of carbon input into soil (McGill *et al*. 1986). The addition of mulching materials enhances the soil microbial activity due to a 'priming' effect. The young tea was fertilized with nitrogen fertiliser at two month intervals, which also contributed to the 'priming' effect. Therefore, the decomposition rates of mulching materials were not limited by the availability of soil nitrogen. Further, the present study was carried out under field conditions where optimal soil moisture and temperature conditions did not

exist all the time, even though mulching materials were added at regular intervals. The variation in soil microbial biomass may be due to a combined effect of quality and quantity of organic substrate available (Swift *et al*. 1979; Recous 2000), soil pH (Alexander 1977; Grey and Williams 1981; Paul and Clark 1989; Shah *et al*. 1990; Neale *et al*. 1997), soil moisture (Insam *et al*. 1989), and soil temperature (Lal 1974; Anderson and Nilsson 2001).

The high soil microbial biomass of the soils under all of the mulching materials (Table 3.7) is most likely due to the addition of substrates to sustain microbial populations. The refuse tea and dadap legume mulch produced higher soil microbial biomasses and higher soil respiration rates than did the other mulches (Table 3.7). Although the refuse tea mulch had a lower carbon content (34.4%) compared to the mana (40.3%) and dadap (38.2%) mulches, its nitrogen content was higher than that of the mana grass mulch and therefore its C/N ratio was close to that of the dadap legume (Table 3.6). Therefore, the availability of microbial substrate in the mulch of refuse tea is similar to that of dadap. As a result, the soils under both treatments had higher soil microbial biomass carbon contents compared to those under the rest of the treatments (Table 3.7). The slight increase in soil microbial biomass carbon under the mana grass mulch compared to that of the control is probably due to extra carbon added from the mulching materials (Table 3.7).

As the soil organic carbon level increased, the microbial biomass carbon also increased (Fig 3.6) due to the availability of nitrogen and phosphorus of the substrate. However, further increase in organic carbon tended to decrease the microbial biomass carbon (Fig 3.6). This may be due to the limitations posed by either nitrogen or phosphorus

availability which tend to increase competition among the microbial populations and thus decrease the microbial populations.

Figure 3.7 reflects the microbial biomass carbon content of the soil nine months after application of the mulch and pH modifier treatments, and just prior to a fertiliser application. It can therefore be assumed that there is no fertiliser effect in this relationship. Here, the mineralization of organic matter depends on microbial transformations which are influenced by the soil factors that affect microbial activity such as nitrogen availability, temperature, moisture, and soil pH (Paul and Clark 1989). Powlson and Jenkinson (1981) and Holt (1997) showed that there may be rapid responses of microbial biomass carbon to changes in the management of the soil in trials conducted in cropping and grazing systems respectively. Figure 3.7 provides a sensitive measure of changes in the organic matter status of the tea soils.

In general, the microbial biomass carbon content of the soil reflects the long-term amounts of carbon input into soil (McGill *et al*. 1986). This is due to higher availability of energy released from the decomposition of organic matter. But the size of the soil microbial biomass depends on the environment, soil, cultural practices, and soil additives (Dalal 1998).

To maintain the carbon level, the soil microbes need access to nitrogen, but there is a competition with plant roots for nitrogen (Neale *et al*. 1997). This usually leads to a slight reduction with time in microbial biomass carbon content as was found in the soils under the young tea (Fig. 3.7). Here the conversion of organic matter to available forms depends on microbial transformations and is influenced by the factors that affect

microbial activity, primarily soil temperature, moisture, pH, and available nitrogen. If the C/N ratio of applied mulch is greater than 30, and the total soil nitrogen content is about 1.5% or less, lower nitrogen reserves cause net immobilization (Stevenson and Cole 1999). This means the substrate nitrogen is utilized by microbes for their activity. On the other hand, however, residues with C/N ratios of less than about 20, and with nitrogen contents more than 2.5%, often result in an increase in mineral nitrogen levels through net mineralisation by microbial activity (Stevenson and Cole 1999).

3.5.2 Soil respiration

Soil respiration depends on the microbial biomass present, the organic carbon content of the soil, and its C/N ratio, pH, moisture, and temperature (Paul and Clark, 1989). The soil respiration rate represents the microbial activity in the soil rhizosphere and the respiration of tea plant roots. Since the tea plants were small, their contribution to soil respiration by roots should have been very low compared to the treatment effects. Further, there was no moisture limitation at the time of measurement. Therefore, higher soil respiration in the soils under the refuse tea mulch may be a result of favourable soil pH and higher microbial biomass carbon (Table 3.7).

3.5.3 Soil pH

The dolomite and Minplus rock dust treatments produced no significant differences in soil or plant properties in the present study (Table 3.5). This may be a result of insufficient amounts of soil pH modifiers being added to the experimental soils. In studies reported by Coventry *et al.* (2001), Minplus application rates of 2.5 to 5 times those of the present study on young tea plants (1 tonne ha^{-1}) were used. The application

rates of soil pH modifiers have been increased over the experimental plots on the St. Coombs Estate, but no results are currently available from these recently modified trials.

The increase in soil pH under the refuse tea and mana grass mulch may be mainly due to the ash alkalinity of the added materials (Noble *et al*. 1996). Refuse tea has 1.85 % K and 0.20 % Mg which have contributed to ash alkalinity (Wijeratne 1999). Similarly, the mana grass mulch also contains a relatively high content of basic cations, (2.4 % K and 2.1 % Mg). Other reasons may be the protonation of organic anions and microbial decarboxylation of soluble organic anions (Marschner and Noble, 2000). In the case of dadap mulch, due to its faster decomposition rates, there may be leaching effects which gave rise to a similar soil pH as that of the control (Table 3.7). Lemon grass treatments had the lowest soil pH which was not significantly different from the control (Table 3.7), most likely because it was a live mulch excreting root exudates containing H^+ ions.

3.5.4 Chlorophyll

The chlorophyll content of the young tea plant leaves is an important component of dry matter production because it is involved in photosynthetic processes (Kulasegaram 1986). The chlorophyll content of the leaves of the young tea plants reflects nitrogen and magnesium status of the soil which is the main source of the nitrogen and magnesium constituents of the chlorophyll molecule (Manivel and Hussain 1982). The chlorophyll content of tea was higher in the plants grown under the control, refuse tea, and dadap mulches compared to those under the mana and lemon grass mulch. The lower leaf chlorophyll contents produced by the mana grass may be due to its higher C/N ratio and, therefore, immobilization of nitrogen by microbial populations, and an associated decrease in the nitrogen availability to the tea plant. Since the lemon grass

was live mulch, its roots would have taken up some of the available nitrogen and magnesium from the soil, and thus may be the reason for low chlorophyll contents of the tea plants grown under the lemon grass treatment (Table 3.7).

3.5.5 Yield of tea

The dadap legume mulch had the highest nitrogen, phosphorus and potassium contents which are essential for growth and yield of tea (Table 3.4). The the refuse tea mulch and the dadap mulch produced the highest leaf chlorophyll contents, and the highest yields of useable tea leaves (19 and 16%, respectively more than the control; Table 3.7). The yield increase may be a consequence of high soil microbial biomass carbon contents in the underlying soil which also enhance nutrient recycling and availability.

3.5.6 Tea quality

Leaf infusion is one of the parameters that determine the quality of the made tea. It is related to the appearance of the tea leaf after brewing the tea, and is an important factor in determining tea prices. A bright, even infusion is considered to be a specific assessment for high quality tea (Keegal 1983). Plants grown under stressed conditions generate leaf infusions that are higher than those produced by plants from a no stress condition. The leaf flush picked from the control and lemon grass treatments had higher leaf infusions than did the tea from the rest of the mulching treatments, probably as a result of assimilated stress conditions. Generally, when the yield is higher, the quality is lower, and vice-versa. The quality of the tea produced by the present study reflected this trend (Table 3.7 and 3.9). This may be a consequence of either the stressed condition of the flush leaves, and may be related to the availability of the substrates, polyphenols, and enzyme polyphenol oxidase in the flush, or to processing procedures

in manufacturing made tea (Dahanayake and Ziyad 2002). Since the processing procedures were similar for all the treatments, the differences between treatments are more likely to be a consequence of the condition of the leaves and suggests that the absence of mulch and the living lemon grass mulch may have contributed to the development of stressed conditions in the young tea plants reflected the higher quality (Table 3.9). The stress may have been due to inadequate availability of soil moisture nutrients or higher vapour deficit.

3.6 CONCLUSIONS

1. All of the mulching materials tested on the young tea plants of Sri Lanka had higher decay constants compared to that of the control which was produced by the natural accumulation of tea leaves and twigs under the plants.

2 All of the mulching materials tested increased the soil microbial biomass over that of the control. The order of increase was refuse tea mulch (greatest), dadap, mana grass, and lemon grass (least); soil respiration was increased only by the refuse tea mulch.

3. There was no response evident in either soil or plant growth parameters to the application of the soil pH amendments (dolomite and Minplus rock dust). The amount of Minplus applied in the Young Tea trial in Sri Lanka was extraordinarily low and at only 10 – 20% of the recommended application rate needed to induce significant changes in plant growth in highly weathered soils in North Queensland.

4. A fatty acid methyl ester (FAME) analysis indicated that the bacterial populations were larger than the fungal populations at the end of 14 months in the soils under the young tea. The dadap legume mulch increased both bacterial and fungal populations while the refuse tea mulch reduced them compared to the control. All of the mulches, except for the dadap legume mulch, also produced a low ratio of Gram-positive to Gram-negative bacteria indicating their efficacy at reducing the populations of bacterial plant pathogens in the soil.

5. There was no change in cation exchange capacity, total nitrogen, total phosphorus, and plant-available phosphorus in the underlying soils by added mulching materials, soil pH amendments, or their combinations. This may be largely a reflection on the low application rates of Minplus rock dust and dolomite that were used in the present study.

6. Among the growth parameters studied, only the chlorophyll content of leaves and the yields of the young tea plants responded to the mulching treatments; mana grass mulch significantly reduced the leaf chlorophyll content of plants growing in the underlying soil.

7. Both refuse tea (25 tonnes ha^{-1}) and dadap legume (35 tonnes ha^{-1}) mulches improved the yield of young tea by 19% and 16% respectively.

8. The quality of made tea in terms of leaf infusion characteristics was higher in tea grown under the control and lemon grass mulch treatments which suggests that the stress condition was created by the control and lemon grass mulch treatments.

9. Since refuse tea from tea factories and dadap legume from shade trees are both readily available at tea estates in Sri Lanka, they could be used as mulching materials to enhance soil microbial properties, and to increase the yields from young tea plants.

CHAPTER 4

MATURE TEA TRIAL AT ST. COOMBS ESTATE, TALAWAKELLE, SRI LANKA

4.1 INTRODUCTION

"Mature tea" is considered to be commercially productive tea plants, at least 5 years old, and having canopies that cover the ground almost completely. In the upland areas of Sri Lanka, higher than about 1200 m a.s.l., pruning is carried out every 4-5 years in order to control the height of the plucking table of the tea plants. The pruning is done at a height of 45-55 cm and removes all of the foliage and branches above this height, leaving one branch with 200 - 300 leaves to allow the plant to recover.

Tea leaves and prunings themselves serve as a mulch material after pruning. However, the workers take most of the thick woody branches out of the tea field for use as firewood. As a result, the soil below the plants is not usually completely covered by prunings. The exposed soils of the fields are vulnerable to weed growth and soil erosion until the mature canopy regenerates. The time taken for regeneration of the canopy varies, but is usually 3-4 months. Hence, a pruned mature tea field is similar to a young tea fields in terms of inadequate soil cover. Some 2-3 weeks after pruning, the frame of the bush that remains is manually cleaned with clean water and brushes to remove mosses and ferns. The manual method ensures that there is no contamination to the microbial population of the soil.

Since tea is fertilised with high doses of nitrogen (as high as 360 kg nitrogen ha^{-1} $annum^{-1}$ in 4 split applications), soil acidity tends to be high and requires regular

amendment with dolomite. Soil acidity and any magnesium deficiency are corrected by "liming" with dolomite which is usually applied by broadcasting in tea lands at a rate of 2500 kg ha^{-1}. The decomposition of polyphenol-rich tea residues leads to the formation of nitrogen-rich humic matter in the soil (Sivapalan 1982), a reduction in soil urease activity (Sivapalan *et al*. 1983), and contributes to increasing soil acidity.

Decomposition of plant litter and humus are fundamental ecosystem processes which maintain a continuous supply of essential nutrients to plants (Satchell 1974; Swift *et al*. 1979). Krishnapillai (1984) showed that, from well decomposed tea mulch, and from the soil collected beneath the layer of mulch, there was a release of about 180-250 kg nitrogen (as ammonium and nitrate ions) over a period of six weeks. Similarly, the exchangeable potassium released during this period was 200-250 kg, and exchangeable magnesium 90-100 and phosphate was 60-90 kg respectively.

In mature tea, plant available nutrients are produced by mineralisation of an adequate supply of mulch, other than from applications of fertiliser. Tolhurst (1960) observed that there was rapid recovery of mature tea plants in spite of the cessation of manuring for about 4-6 months before the pruning stage (at 4-5 year intervals). This shows that the tea plants have been solely utilizing the nutrients released from the mineralisation of mulch for their growth. However, no detailed study has been carried out on the effects of mulches, either individually or in combination with soil pH amendments, on the chemical, biological, and physical properties of the tea soils, nor on the growth and yield of tea. Therefore, the present study was undertaken with the following aims.

4.2 AIMS OF THE STUDY

- To examine the effects of mulching materials and soil pH amendments with different compositions and qualities on the chemical and biological properties of the soils under mature tea plants.
- To examine the effects of applied mulch and soil pH amendments individually and together on the growth, yield, and quality of the products of a mature tea plantation.

4.3 MATERIALS AND METHODS

4.3.1 Study site

The experimental plots were located in Field No. 3, St. Coombs Estate of the Tea Research Institute of Sri Lanka, Talawakelle. The average slope was 26.5° at the site which was 1 km from the young tea trial described in Chapter 3, above. The soil was classified as a fine mixed Tropudult (Panabokke 1996).

4.3.2 Soil and plant materials

Soil samples were collected from a depth of 0-15 cm from below the canopy of the tea plants, and immediately before the application of mulch treatments (Table 4.2). Chemical, physical and biological analyses were performed on all of the soil samples using the methods outlined in section 3.3.5.

The mature tea experiments were carried out with clone TRI 2025 tea plants that were 20 years old and in the first year after pruning. Measurements were made of mean branch circumference taken from the average circumferences of three branches per bush

randomly selected from the periphery of the canopy and 6 cm from the healed pruning cut.

4.3.3 Experimental treatments

Treatments were combinations of mulching materials and pH amendments as used on the young tea (section 3.3.2), with the omission of the lemon grass treatment. In addition, a *Trichoderma herzianum* fungal inoculum mixture was applied as a soil amendment to help accelerate plant litter breakdown and its mineralization (Anonymous 1985). The *T. herzianum* inoculum was prepared by mixing 20 kg of dry cow dung with 10 kg of refuse tea, and 15 kg of soil to which had been added 500 g of pure *T. herzianum* culture; 100 g of this medium contained approximately 11 x 10^5 spores. The mixture was applied to the soil around the mature tea plants at a rate of 300 g $plant^{-1}$.

As in the young tea trial, all of the mulches and soil pH amendments were applied in August 2000. Following the initial application, mulches were applied at 4-5 month intervals (depending on their decomposition rate) except for the refuse tea which was applied at 6 month intervals during the 14 months of the experimental period (August 2000-October 2001). The rates of mulching materials were applied in accordance with the recommendations of the Tea Research Institute. The soil pH amendments were dolomite and Minplus (crushed basaltic rock) applied at the low rate of 1000 kg ha^{-1}. Fertiliser applications in the mature tea were carried out using the same scheduled as for young tea (Table 3.4). The treatment combinations are set out in Table 4.1 and the times of treatment applications, made immediately after sampling the soils, are shown in Table 4.2

Table 4.1 The treatment combinations of mature tea trial in Sri Lanka

No.	Treatment code	Mulch or inoculum used	Soil pH modifiers used
1	MoPo	No mulch	No pH modifier
2	MoPd	No mulch	Dolomite
3	MoPm	No mulch	Minplus
4	RtPo	Refuse tea	No pH modifier
5	RtPd	Refuse tea	Dolomite
6	RtPm	Refuse tea	Minplus
7	MaPo	Mana grass	No pH modifier
8	MaPd	Mana grass	Dolomite
9	MaPm	Mana grass	Minplus
10	DaPo	Dadap legume	No pH modifier
11	DaPd	Dadap legume	Dolomite
12	DaPm	Dadap legume	Minplus
13	TrPo	*Trichoderma* fungus	No pH modifier
14	TrPd	*Trichoderma* fungus	Dolomite
15	TrPm	*Trichoderma* fungus	Minplus

Table 4.2. Time of application of mulch material and soil conditioners to mature tea in Sri Lanka

Number of each application	Refuse tea	Mana grass	Dadap legume	*Trichoderma* inoculum	Dolomite/ Minplus
1	August 2000	August 2000	August 2000	Incorporated in August 2000	August 2000
2	February 2001	December 2000	December 2000	-	-
3	October 2001	May 2001	May 2001	-	-
4		September 2001	September 2001	-	-

4.3.4 Crop yield

Each plot was plucked separately at weekly intervals and the fresh weight of the flush was recorded with the yield expressed as made tea per hectare, as follows:

Yield (made tea kg ha^{-1}) = Total flush fresh weight (g)/ (Number of plants in the plucked plot x 12,500 / 1000 x 0.22).

A factor of 0.22 is used to convert the fresh weight of plucked tea leaves to made tea and 12,500 is the recommended number of plants planted per hectare.

4.3.5 Mulch decomposition rates

The litter bag measurement technique used was described in Section 3.4.2.1 (above).

4.3.6 Soil and Plant analysis

The soil and plant analysis methods used are described in section 3.3.5, and the statistical analyses of the results were carried out as for the young tea (Section 3.3.6).

4.4 RESULTS

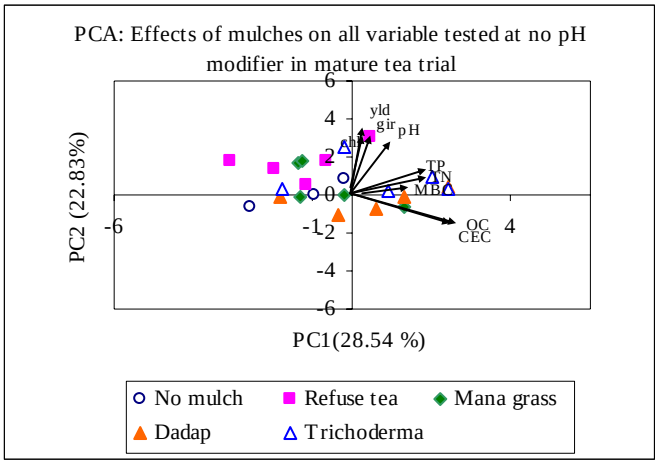
4.4.1 Principal components analysis – mature tea, Sri Lanka

After performing the principal components analysis, a scatter plot was prepared with the first and the second principal components on the vertical and horizontal axis respectively for all the mulch treatments with pH modifiers (Fig. 4.1a – 4.1c).

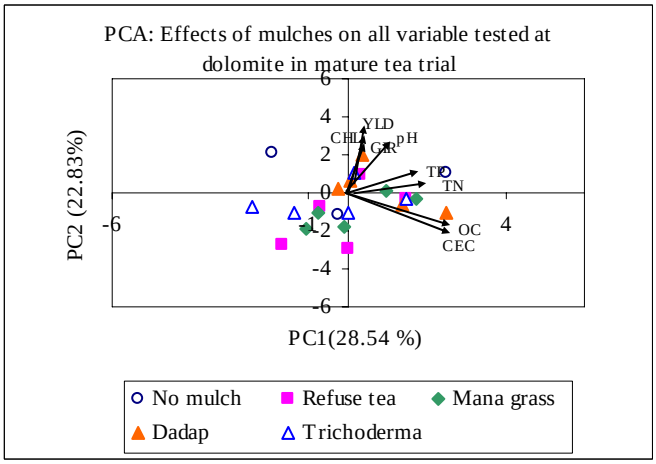
The soil organic carbon, soil pH, CEC, total nitrogen and total phosphorus were the main soil properties that varied with the application of mulches (Fig.4.1 a) in the absence of soil pH modifiers. The plants grown under the dadap legume, refuse tea, and mana grass mulches produced medium growth responses to the changes in soil pH, total phosphorus and total nitrogen (Fig. 4.1 a). The plants under the dadap legume mulch produced greater responses in organic carbon and CEC than did those under the other treatments. The plant factors chlorophyll, branch circumference, and yield were increased by the dadap and refuse tea mulches (Fig. 4.1 a).

In the presence of dolomite, the plants grown under the dadap legume mulch produced higher responses in soil pH, total nitrogen and total phosphorus than did the control (Fig. 4.1 b). Soil organic carbon and CEC responded in a moderate way to the dadap legume and mana grass mulches; similar responses were also observed in plant growth factors such as leaf chlorophyll, branch girth, and tea yield.

(a)



(b)



(c)

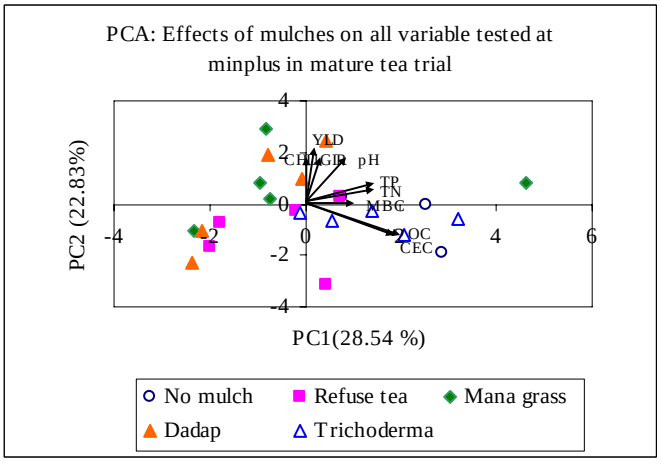


Figure 4.1 Effect of mulches on soil properties in mature tea, Sri Lanka:
(a) The data exclude all soil pH modifier treatments
(b) The data include dolomite as the soil pH modifier treatment,
(c) The data include Minplus as the soil pH modifier treatment.

In the Minplus-treated plots, the plants grown under the dadap legume and mana grass mulches produced stronger responses to soil pH, total nitrogen, and total phosphorus (Fig. 4.1 c). Similar responses were seen in the plant growth factors such as leaf chlorophyll, branch circumference, and tea yield. The plants grown under the *Trichoderma* treatments showed strong responses to organic carbon and CEC (Fig. 4.1 c). Relatively low responses were shown by the plants grown under the refuse tea mulch to chlorophyll, branch circumference, soil pH, and tea yield.

4.4.2 Factor analysis

The results of a multivariate analysis of all the soil parameters are shown in Table 4.3. Significant increases in total soil nitrogen and plant yield were achieved under the refuse tea mulch, and in tea yield by the dadap legume mulch; a significant decrease in soil pH and in tea yield were produced by the Minplus treatment (Table 4.3). No other statistically significant results were observed (Table 4.3).

Table 4.3. Summary of an analysis of variance: mature tea trial at St. Coombs Estate, Talawakelle, Sri Lanka.

Treatments		Soil parameters									Growth parameters		
		OC	**pH**	**TN**	**TP**	**AP**	**CEC**	**BD**	**MB-C**	**Res**	**Chl**	**Cir**	**Yld**
Mulch	No mulch	-	-	-	-	-	-	-	-	-	-	-	-
	Refuse tea	Ns	Ns	*	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	*
	Mana grass	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
	Dadap	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	*
	Legume	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
	Trichoderma fungus												
pH	No pH	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
	Dolomite	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
	Minplus	Ns	#	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	#
Inter-actions	pH x mulch	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns

* Significantly increased ($p < 0.05$), # Significantly decreased ($p < 0.05$)

OC: Soil organic carbon, pH: Soil pH, TN: Total soil nitrogen, TP: Total soil phosphorus, AP: Plant-available soil phosphorus, CEC: Soil cation exchange capacity, BD: Soil bulk density, MB-C: Soil microbial carbon, Res: Soil respiration, Chl: Leaf chlorophyll content, Cir: branch circumference, Yld: yield of made tea, - = Control treatments.

4.4.3 Decomposition rates of mulches

The decomposition rates of the different mulching materials, as determined by the litter bag technique, are presented in Table 4.4.

Since the decomposition of mulches is controlled by soil microbial processes, the decay constant gives an indication of the efficiency of microbial activity under various treatments. The legume mulch, with low C/N ratio and low lignin content, decomposed faster than the other mulch materials (Table 4.4). Though the mana grass mulch had a

Table 4.4 Decomposition of mulching materials under mature tea at St Coombs, Sri Lanka from September 2001 to December 2001.

Mulch used	**C/N ratio at the start of the trial**	**Coefficient of determination** (R^2)	**Decay constant** (d^{-1})	**Half life** (days)
Control (untreated)	10	0.91	0.0019	328
Refuse tea	11	0.83	0.0033	210
Mana grass	29	0.77	0.0095	73
Dadap legume	8	0.94	0.0138	50

high C/N ratio, it was fertilised with nitrogen at 3 month intervals, and it decomposed faster than refuse tea. The refuse tea mulch had the highest polyphenol content (Sivapalan 1982) and lowest surface area contact with the soil, and both factors inhibit decomposition. The lowest decomposition rate was shown in the control treatment which received the natural fall of leaf and twigs of litter during the experimental period (Table 4.4).

4.4.4 Effects of mulches on soils and plant growth

Statistically significant impacts of the mulch treatments were made from the dadap legume and refuse tea mulches on the yield of made tea (Table 4.5). The refuse tea mulch produced significantly higher soil nitrogen contents and yields of made tea compared to the responses under all of the other treatments (Table 4.5). Significant yield improvements from the mature tea plants were attained over the study period under the refuse tea and dadap treatments compared to the control.

Table 4.5 The main effect of mulch on soil properties and yields of mature tea, Sri Lanka. Each cell represents the mean values of soil total nitrogen and yield. Means followed by the same letter within a column are not significantly different (LSD; $p > 0.05$).

Mulch treatment	**Soil parameters**			
	Soil total nitrogen ($mg\ g^{-1}$)	**Yield of made tea** ($kg\ ha^{-1}$)	**Percentage increase of yield over the control**	**Percentage increase of nitrogen over the control**
No mulch	0.428 B	3604 B	--	--
Refuse tea	0.523 A	4110 A	14	22
Mana grass	0.443 B	3640 B	0.9	3.5
Dadap	0.456 B	4105 A	14	6.5
Trichoderma	0.445 B	3558 B	-1.3	3.9
LSD ($p = 0.05$)	0.05	397		
CV%	2.7	13.7		

The uncharacteristically light application of Minplus reduced the soil pH compared to those produced by the untreated control and dolomite treatments (Table 4.6). Neither the Minplus nor dolomite soil pH conditioners produced yields of made tea that were significantly different from that of the untreated control (Table 4.6).

Table 4.6 The main effect of pH amendments on soil properties and yield of mature tea, Sri Lanka. Each cell represents the mean values of soil pH and yield of tea. Mean followed by the same letter within a column are not significantly different (LSD; $p > 0.05$).

Treatments	Soil pH	Yield of made tea (kg ha^{-1})
No pH amendments	4.90 A	3827 AB
Dolomite	4.99 A	3964 A
Minplus	4.73 B	3620 B
LSD ($p < 0.05$)	0.12	321
CV%	4.0	13.7

The weak positive relationship between the organic carbon contents of the soils under each mulch treatment with the corresponding yields of made tea is shown in Fig. 4.2, which indicates that a change of 1 mg/g in the organic carbon content of the soil could change the yield of made tea by about 26 kg ha^{-1}. Although this is a weak relationship, it does indicate the importance of maintaining the organic carbon of the soil through mulching if commercial crop yields are to be sustained.

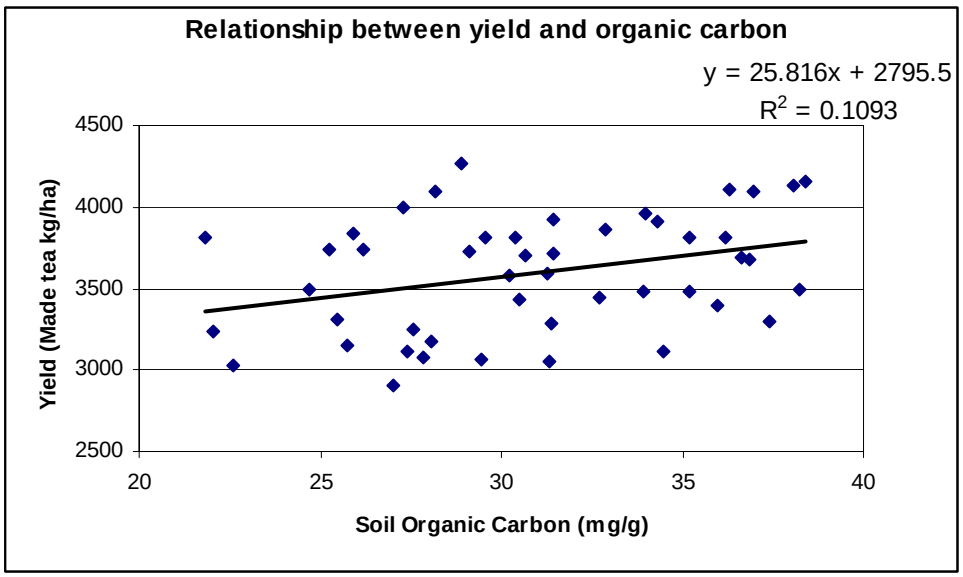


Fig. 4.2 Relationship between organic carbon contents of soils and the yields of made tea.

The initial total nitrogen content of the soil under mature tea lies in the range 0.34-0.35 mg g^{-1}(Fig 4.3). After four months of mulch treatments there were increases in the total nitrogen in the soil under the refuse tea and mana grass mulches with no pH modifiers (Fig. 4.3). These changes were sustained across all of the treatments over the 15 months of the mulching trial (Fig. 4.3). Other studies such as that of Ranganathan (1977) in southern India also reported the return of large amounts of nitrogen to the soil by way of tea prunings.

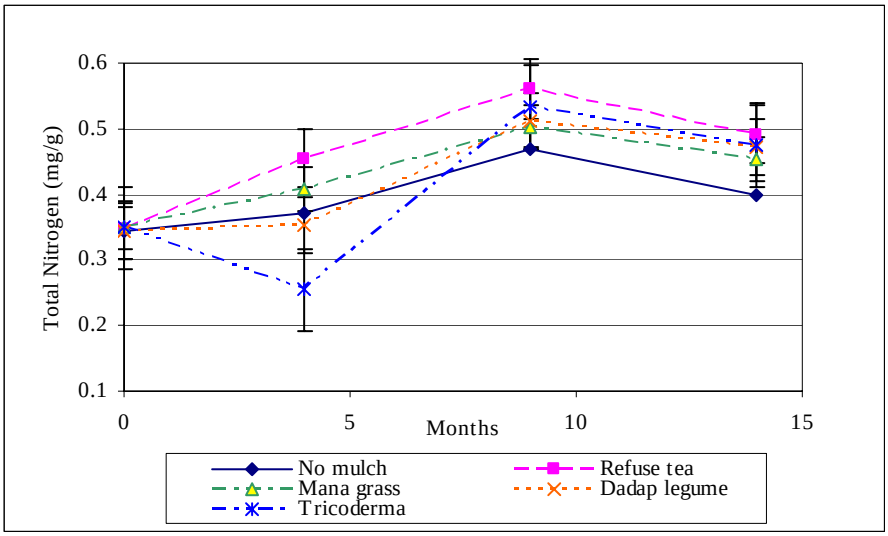


Fig. 4.3 Changes with time in total nitrogen content of the soil to which no soil pH modifier was added under mature tea, Sri Lanka.

4.4.5 Quality of tea produced

Tea quality was measured by the strength, colour, and taste of the infused leaf (Sections 3.3.7 and 3.4.6). Only the infused leaf data showed significant differences between the mulch and soil pH treatments.

Of the quality parameters assessed by the tea tasters, only the infused leaf scores showed significant differences between treatments (Table 4.7); tea produced by the no

mulch and *Trichoderma* fungus treatments had significantly higher scores than the rest of the treatments. As in the young tea trial, this again may be due to the condition of the leaves with stress, the main factor contributing to the higher infused leaf value.

Table 4.7 Scores for quality (infused leaf) of tea in mulch and pH modifier treatments (ranges of values 1-7: Section 3.4.6). Mean values followed by same letter are not significantly different ($p > 0.05$).

Treatments	pH amendments			
Mulch	Control	Dolomite	Minplus	Mean
No mulch	2.57	3.06	2.97	2.87 A
Refuse tea	3.27	2.95	3.12	2.12 D
Mana grass	3.00	2.55	2.86	2.80 B
Dadap legume	2.46	2.78	2.85	2.69 C
Trichoderma fungus	2.85	2.96	2.90	2.90 A
Mean	2.83 B	2.86 B	2.94 A	
LSD (0.05)				0.04
CV%				9.2

4.5 DISCUSSION

4.5.1 Tea Yield

Tea yield is the net result of the interaction of soil properties, plant physiology, and climatic conditions. The harvested economic portion of the tea is its young, vegetative leaf growth, which needs high levels of nitrogen, and to a lesser extent, stores of plant-available potassium and phosphorus in the soil. It takes about 3-4 months for the plant to refoliate after pruning but still it may not produce sufficient canopy to cover the entire ground area for about 12-18 months. Therefore, immediately following pruning, the tea plant shows an investment-type strategy whereby it diverts energy into building its branches and leaves (Kulasegaram 1986). This requires access to substantial levels of plant nutrients, especially nitrogen. Ideally, the nitrogen should be released slowly to match the needs of the tea plant. Chemical fertilisers added in excess of requirement could be easily lost due to leaching or erosion. The tea soil where the study was undertaken has an inherently low CEC. Therefore, slow release of nutrients is essential for the commercial viability of tea plantations and continued sustainable production.

The best option in terms of fertiliser application is to use both organic mulches and inorganic fertilisers. Readily available sources of organic mulches include refuse tea, dadap legume, and mana grass all of which can be obtained from the tea estate itself. Refuse tea is a by product obtained after processing tea in the factory, the dadap legume is a medium shade tree and it could be lopped periodically before the onset of wet weather, and mana grass could be obtained from a ‘thatch bank’. By applying such materials to the soil surface, the soil microbial activity is enhanced by a ‘priming’ effect.

4.5.2 Total Soil Nitrogen

The total soil nitrogen content represents both organic and inorganic nitrogen sources in the soil. Higher total nitrogen in the soil under the refuse tea mulch may be due to the considerably higher amounts of nitrogen (315 kg N ha^{-1}) added to the soil by refuse tea compared to 148 kg N ha^{-1} from the mana grass, and 273 kg N ha^{-1} from the dadap legume mulches (Table 4.5). Yields of made tea were higher from the plants grown under the refuse tea and dadap legume mulches mainly due to the extra nitrogen added to the soil by these treatments (Table 4.5). A part of the new growth of leaves and stems of tea plants is removed continuously by harvesting. Mature leaf flushes contains 4-5 % N, 2.0-2.5% of K, and 0.15-0.20% P on a dry weight basis (Wickremasinghe 1985). This results in 40-50 kg of nitrogen, 20-25 kg of potassium, and 1.5-2.0 kg of phosphorus being removed from the tea field for every 1000 kg of crop harvested. Since the harvest index of tea is about 10-15%, and nitrogen use efficiency is about 30-35%, the balance the nitrogen is locked up in the plant frame and/or is leached below the root zone.

The addition of nitrogen from the dadap legume is approximately 273 kg ha^{-1} (calculated from values in Tables 3.3 and 3.5). Because of its low C/N ratio, it is easily mineralisable. This explains the higher contents of nitrogen observed in the soils under the dadap legume treatment compared with that under the mana grass treatment. Up to the 4^{th} month after pruning, the tea plants were recovering and, after the 4^{th} month, the plants were generally adequately refoliated; the harvesting of tea commences between 4^{th} and 5^{th} months after pruning.

For refoliation and continued tea crop production, adequate quantities of soil nitrogen have to be supplied by fertiliser and the mineralization of mulches. In the control treatments, the nitrogen was supplied by fertiliser and mature tea leaves in the soil litter layer, which are high in polyphenols (Sivapalan, 1982). Therefore, mineralisation may be slow and the amount of nitrogen required in the soil for adequate plant growth may not be available. That may be the reason for lower total nitrogen in the soil under the control. In the other treatments, there was extra nitrogen coming from the mineralisation of mulching materials, therefore total soil nitrogen contents are higher than that of the control.

In dolomite-treated plots, the dadap legume mulch gave rise to lower soil nitrogen contents than did the refuse tea mulch. This effect is most likely a result of the low C/N ratio of the dadap mulch, coupled with its rapid decomposition rate (Table 4.4). The faster release of nitrogen may then have led to some losses from the soil through leaching. The reduction in total nitrogen content of soils under the *Trichoderma* fungus was higher than that occurred in the other mulches within the initial period of 6-7 months (Fig 4.3). This suggested that immobilisation of nitrogen was occurring in the soils under the plots treated with *Trichoderma* (Fig. 4.3).

The nitrogen supply from the mulch materials was unable to match the demand from the tea plant, especially given the continued harvesting and the possible loss of nitrogen through leaching and volatilization (Kulasegaram 1982). Mulch effects depend on their C/N ratios and correlate well (Tables 4.4 and 4.5) with nitrogen mineralisation (Ford *et al*. 1989). Unlike the young tea trial, a urea-based fertiliser (U 709) was applied according to the recommendations of the Tea Research Institute, Sri Lanka. It consists

of 28.4 % N, 4.72 % P_2O_5 and 14.2 % K_2O. In each fertiliser application, 90 kg of N was applied per hectare from U 709 along with 45 kg ha^{-1} of K_2O from muriate of potash.

In soils treated with a urea-based fertiliser, there is usually a higher nitrate-nitrogen content than that in soils treated with ammonium sulphate, because of the temporary rise of pH associated with urea hydrolysis (Wickramasinghe *et al*. 1985). This favours the activity of nitrifying bacteria (autotrophic bacteria) in acid soils (Overrein 1967).

None of the soil pH amendments influenced the soil nitrogen contents significantly. This is possibly the result of very low initial application rates of dolomite and Minplus, and of the regular applications of fertiliser U 709.

4.6 CONCLUSIONS

1. All of the mulching materials tested had a higher decay constant than the control, and the refuse tea treatment was found to have had a lower decay constant than that of all of the other mulches tested (Table 4.4.)

2. The application of mulching materials to mature tea plantations did not change the soil organic carbon content or pH of the soil. But, the refuse tea mulch increased the soil nitrogen content over that of the control and other mulches. There was a significant reduction of pH in Minplus-treated plots. No yield improvement was demonstrated in either the dolomite- or the Minplus- treated plots (Table 4.6).

3. No changes were induced in soil organic carbon, cation exchange capacity, microbial biomass carbon, total phosphorus, and plant available phosphorus contents by adding mulches or soil pH amendments, individually or in combinations, to the soils of the mature tea.

4. Total soil nitrogen contents were significantly increased by the refuse tea treatment in mature tea.

5. Among the tea plant growth parameters studied, only the yield of made tea responded to the mulching treatments and pH amendments of the mature tea. The refuse tea and dadap legume mulches produced significant improvements of yield by 14 - 19% (Table 4.6).

6. The quality of made tea, in terms of leaf infusion scores, were higher in the plants grown under the control and *Trichoderma* fungus treatments which reflect the higher quality of tea.

7. Since mulches of refuse tea from tea factories and of dadap legumes from shade trees are readily available in tea estates, they could be used as cheap and effective mulching materials to enhance the made tea yields and total soil nitrogen contents in mature tea.

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

5.1 GENERAL CONCLUSIONS

The growth and productivity of a perennial rain-fed crop like tea depends mainly on soil fertility. Three separate research projects were carried out as parts of the present study of methods to improve the productivity of tea plants at different growth stages on similar soils:

- a nursery pot trial in a shade house at James Cook University, Townsville, Australia,
- field trials on young tea at the Tea Research Institute, Talawakelle, Sri Lanka,
- field trials on mature tea at the Tea Research Institute, Talawakelle, Sri Lanka.

Improvements in soil fertility were found to depend on initial soil fertility, soil pH, the quality and quantity of mulching materials used, and on the size and decomposing activity of the soil microbial populations. Soil microbial populations were enhanced significantly under refuse tea, and dadap legume mulches where they attained 263.6 µg C g^{-1} and 288.4 µg C g^{-1}, respectively in young tea (control = 120 µg C g^{-1}; Table 3.7).

The mulching materials used in the studies had different decomposition rates depending on their chemical composition and physical state. In the JCU Nursery Trials, there was a clear indication that legume and grass mulch were superior to tea mulch in increasing the organic carbon, pH, CEC, plant available phosphorus and microbial biomass carbon of the soil.

Similarly, the young and mature tea trials conducted in Sri Lanka under field conditions revealed that soil fertility in terms of organic carbon and soil pH were not altered significantly by the addition of mulching materials. However, in the soils of all three studies, the soil microbial biomass carbon was increased by the mulches due to the priming effect of added organic carbon and nitrogen from the mulching materials.

The soil microbial populations drive the nutrient recycling processes. The extent of the improvement in soil properties depends on the quality and quantity of mulching materials used. In the JCU Nursery Trial, both grass and legume mulches increased the microbial biomass compared to the effects of the tea mulch. This was also reflected in the growth of the tea seedlings under the mulches (Sections 2.4.1.2 and 2.4.2.2). However, in the Sri Lankan field-based studies, the dadap legume and refuse tea mulches raised the soil microbial biomass carbon contents it was reflected in the enhanced yields of tea (Section 3.4.3).

The different responses of tea plant growth in the Sri Lankan field studies might be readily explained by the differences in chemical compositions of the mulches used. The tea mulch was rich in unoxidised polyphenol, while the refuse tea mulch was rich in oxidised polyphenols (22%) with a relatively high nitrogen content (Sivapalan 1982); the dadap legume mulch had the highest nitrogen and relatively low polyphenol and lignin contents (Table 3.3).

The JCU Nursery Trial showed that the microbial biomass carbon and nitrogen contents of the underlying soils increased with the applications of grass mulch (Tables 2.4 and 2.5),

rainforest inoculum (Table 2.7), and dolomite (Table 2.8). The microbial biomass is composed of both beneficial and pathogenic populations and applications of mulching materials were shown to have increased the ratio of beneficial Gram-positive to detrimental Gram-negative bacteria, expanded the fungal and mycorrhizal numbers, and reduced the bacteria/fungi ratio. Similar microbial responses occurred under the dadap legume mulch in the soil under young tea in Sri Lanka.

In the Nursery Trial, it was observed that the addition of a rainforest inoculum to the soil in the experimental pots increased the microbial biomass, the organic carbon content and the cation exchange capacity of the soil (Table 2.14), but these effects were not reflected in the tea seedlings (Table 2.21).

There was a variable response to the soil pH modifiers used in the different components of the present study. In the JCU Nursery Trials, the Minplus rock dust produced superior tea seedling growth to those under the equivalent application rate of dolomite, while in the Sri Lankan field studies; dolomite produced superior growth in young and mature tea plants. These different responses may be due to level of calcium in the soil, or more likely, to the very low application rates of Minplus used in Sri Lanka which were less than one quarter of the rates used to demonstrate clear growth responses in other field crops in North Queensland (Coventry *et al*. 2001). Grass or legume mulches used in conjunction with Minplus applications produced positive responses in the growth of tea seedlings in the Nursery Trial at JCU.

There was no response to the soil pH modifiers in tea growth parameters or tea yield in either the young or mature tea trials in Sri Lanka where the dadap legume and refuse tea mulches produced the greatest responses in the growth of tea plants. Enhanced growth was reflected in high chlorophyll content of the leaves, and in high nitrogen contents of the soil under the mature tea, and in high tea yields from both the young and mature tea plants.

In terms of product quality, the refuse tea and dadap legume mulches produced tea with poorer leaf infusions than did plants grown under the other mulches. The control and lemon grass treatments in the young tea trial, and the *Trichoderma*-treated plots in the mature tea trial produced enhanced leaf infusions suggesting that the plants, and the quality of their made tea, had benefited from stress conditions at some stage during growth.

5.2 RECOMMENDATIONS FOR TEA GROWERS

The present study has shown that the use of mulches, soil pH modifiers, and rainforest soil inoculums all have a place in improving the growth and productivity of tea plants.

In the nursery and young tea stages of propagation of tea seedlings, mulching materials such as grass (e.g. *Brachiaria decumbens*, or a shrub legume (e.g. *Calliandra calothyrsus*, *Erythrina lithosperma*) may be applied at rates of up to 35 t ha^{-1} fresh weight to enhance growth rates and provide benefits to the tea grower.

In young and mature tea fields, readily available mulches such as refuse tea at a rate of 25 t ha^{-1} and dadap legume at a rate of 35 t ha^{-1} could be used to enhance soil properties, especially soil microbial populations, and to improve tea plant growth.

The use of soil pH amendments such as Minplus rock dust applied at a rate of at least 2.5 t ha^{-1}, could be used more beneficially than dolomite in nursery plots.

An inoculum of undisturbed rainforest soil could be added to the highly modified soils of tea plantations in order to enhance the soil microbial populations and to promote the decomposition of plant litter on the soil surface and nutrient cycling. Application rates of 13% were found to be beneficial in the present study.

Except for the Minplus rock dust, all of the foregoing treatments are cheap and readily available to the commercial tea growers of the highlands of Sri Lanka. The effect of higher rates of Minplus (5-10 t ha^{-1}) are being investigated at the Tea Research Institute, Talawakelle, and this soil conditioner may have great potential in improving the productivity of the tea lands of Sri Lanka and elsewhere in the world.

5.3 FUTURE STUDIES

- Higher rates of dolomite and Minplus on tea soil should be investigated
- Root study of the field trials should be carried out with different treatment combinations and should analyse soil biological properties, particularly for carbon dioxide liberation.
- Studies of decomposition rates of mulch materials with different C/N ratios should be carried out in conjunction with pH amendments at glasshouse and field levels.
- Mixing different mulch materials to achieve synchrony between nutrient release and its demand by tea could be investigated under glasshouse conditions.

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Appendix 1

Determination of Microbial Biomass Nitrogen: - Chloroform-Fumigation Extraction Method (Amato and Ladd 1988).

i) Fumigation

Twenty gram soil samples (passed through 2 mm mesh) are weighed into a plastic chipette and placed in a vacuum desiccator along with a beaker containing 50 mL of chloroform and some glass beads. The desiccator is then evacuated with a vacuum pump until the chloroform boils, sealed, and placed in a dark cupboard, and left for 10 days.

ii) *Extraction*

After transferring the fumigated soil to a 100 mL centrifuge tube, 63 mL of 0.5 M K_2SO_4 is added and the tube is then placed in an end-over-end shaker and mixed for 1 hour. Following shaking, the tubes are then centrifuged for 3 minutes at 2000 g. Ten mL of supernatant liquid from the centrifuge tube is then passed through a Millipore pre-filter (GF/F filter) and stored in vials at –15 °C pending analysis. At the same time, 20 g of an unfumigated sample of each soil is extracted in the same way.

iii) *Analysis*

One mL samples of the fumigated and unfumigated extracts are pipetted into 10 mL test tubes prior to adding 1 mL of freshly prepared ninhydrin reagent (see below). The tubes are then mixed on a vortex mixer before being placed for 15 minutes in a boiling water bath, and then cooled in a cold water bath before diluting with 5 mL of 50% ethanol in distilled water. Each tube is then sealed with para film and shaken vigorously for 5 seconds. At the same time, a series of standards and a blank solution is also prepared. A Beckman Infrared Spectrometer is then used to measure the optical density at 570 nm (readings were taken soon after colour development, before fading occurs).

iv) *Reagents*

a) Acetate buffer 4N

Add 2720 g of sodium acetate (CH_3cooNa.3 H2O (AR) or 1720 g of sodium acetate (anhydrous) to 2 L of distilled water and stir while heating until dissolved. Cool at room temperature, add 500 mL of glacial acetic acid and make up to 5 L with distilled water. The solution should have a pH of 5.5 +/- 0.03 (for the adjustment of pH, 0.1 M sodium acetate or 0.1% glacial acetic acid could be used).

b) Diluent

50% ethanol (AR) in distilled H_2O.

c) Ninhydrin reagent

For 100 mL of reagent (volume determined by number of assays required) dissolve 2 g ninhydrin($C_9H_6O_4$ -2,2, dihydroxy-1,3-indanedione-) and 0.2 g hydrindantin ($C_{18}H_{10}O_6$ -2,2'-dihydroxy-[2,2'-bi-IH-in-dene]-1,1',3,3'-(2H,2'H)-tetrone) in 50 mLs methoxyethanol ($C_2H_5OCH_3$). Add 50 mL of acetate buffer and mix.

Prepare ninhydrin reagent fresh on the day of assay and store in a dark glass reagent bottle.

d) Standards

L-leucine ($C_6H_{13}NO_2$-2-amino-4-methylvaleric acid-) prepare stock solution (2.5 x 10-2 M) by dissolving 0.3279g L-leucine in 100 mL 0.5M K_2SO_4

For the working solution, dilute the stock solution by 100 x with 0.5 M K_2SO_4 (concentration of 2.5 x 10^{-4} M). 2 mL of this solution = 7 μg N

Store all standards in a refrigerator.

(Both amino acid radicals and NH^{4+} ions are released during fumigation, but since the amino acid radical and the NH^{4+} ion both have the same optical density, L-leucine is used as the standard. NH_4Cl could also be used in the same concentrations.)

v) *Extractant*

Extract soils with 0.5M K_2SO_4 (87.13 g of K_2SO_4 L^{-1})

vi) Calculation of results

Absorbance (fumigated) - absorbance (unfumigated)/Dry Weight of soil used x 63/10 x 1000 = Microbial biomass Nitrogen (μg/g).

Appendix 2

Determination of Microbial biomass Carbon; Chloroform Fumigation–Extraction method (Sparling *et al.* 1990)

Analysis

A 60 mLs of soil extract was collected using 0.5 M K_2SO_4 as described in chloroform fumigation – extraction method (Appendix 1). Dispense 15 mL of extract into a 75 mL digestion tube, add about 5 glass boiling beads then add 10 mL of 0.167 M $K_2Cr_2O_7$. Add 20 mL of H_2SO_4 while swirling tube to prevent violent reaction from localized boiling. Heat the tubes at 150^0 C for one hour in a block digester. Remove and allow to cool, then make up to 75 mL with distilled water. Stopper and invert several times to ensure proper mixing. Leave the tubes to cool before reading absorbance on a spectrophotometer at 600 nm.

Prepare standards for each set of analyses by dispensing 0, 0.2, 0.4, 0.6, 0.8 and 1.0 mL of standard sucrose solution into a 75 mL digestion tube. Add 15 mL of 0.5 M K_2SO_4 solution (to maintain constant volume). The volume of Standards sucrose solution, ie. 0, 0.2, 0.4, 0.6, 0.8 and 1.0 mls referred 0,1, 2, 3, 4, and 5 µg of carbon/g respectively. Standard graph can be drawn, spectrometer reading Vs concentration (µg of carbon/g) and can be find the gradient for calculation.

Reagents

1 Concentrated sulphuric acid (AR Grade)

2 0.167 M potassium dichromate ($K_2Cr_2O_7$). Dissolve 49.1 g of K2Cr2O7 in distilled water and make up to 1 L.

3 Sucrose stock standard. Dissolve 11.8745 g A.R grade sucrose ($C_{12}H_{22}O_{11}$) – previously dried in a desiccator) in 1 L of distilled water. 1mL contains 5 mg C. Note. It may be necessary to further dilute the standard solution x 10 (ie. Take 10 mL and dilute to 100mL) for the working standard. This will enable 2, 4, 6, 8 and 10 mL to be used instead of the smaller volumes)

3 Extraction: Extract soils with 0.5 M K_2SO_4 (87.13 g of K_2SO_4/L).

Calculation of results

Absorbance (Fumigation) - absorbance (unfumigation)/15 (dry weight of soil) x 100/15 x 1000 = microbial biomass carbon ($\mu g\ g^{-1}$)

Factor Analysis

Appendix 3

KMO and Bartlett's Test

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.536
Bartlett's Test of Sphericity	Approx. Chi-Square	273.085
	df	21
	Sig.	.000

Communalities

	Initial	Extraction
MBC1	1.000	.691
OC1	1.000	.967
CEC1	1.000	.954
PHV1	1.000	.785
TN1	1.000	.495
TP1	1.000	.764
AP1	1.000	.604

Extraction Method: Principal Component Analysis.

Total Variance Explained

	Initial Eigenvalues		
Component	Total	% of Variance	Cumulative %
1	2.654	37.918	37.918
2	1.625	23.218	61.136
3	.981	14.008	75.144
4	.826	11.798	86.942
5	.543	7.752	94.694
6	.355	5.067	99.761
7	1.670E-02	.239	100.000

Extraction Method: Principal Component Analysis.

Total Variance Explained

Component	Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	2.654	37.918	37.918	2.284	32.631	32.631
2	1.625	23.218	61.136	1.869	26.696	59.327
3	.981	14.008	75.144	1.107	15.817	75.144
4						
5						
6						
7						

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component		
	1	2	3
MBC1	.429	.710	4.213E-02
OC1	.889	-.328	.263
CEC1	.815	-.446	.302
PHV1	.538	.698	-9.239E-02
TN1	.431	.258	-.492
TP1	-9.066E-02	.494	.716
AP1	.730	-.126	-.235

Extraction Method: Principal Component Analysis.

a. 3 components extracted.

Rotated Component Matrix[a]

	Component		
	1	2	3
MBC1	6.385E-02	.806	.194
OC1	.975	.128	-2.778E-02
CEC1	.976	-1.710E-02	-1.706E-02
PHV1	.117	.877	4.514E-02
TN1	9.103E-02	.533	-.451
TP1	-6.128E-02	.227	.842
AP1	.592	.314	-.393

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

a. Rotation converged in 5 iterations.

Component Transformation Matrix

Component	1	2	3
1	.840	.511	-.184
2	-.437	.837	.329
3	.322	-.196	.926

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

General Linear Model

Between-Subjects Factors

		N
MULCH	1.00	16
	2.00	16
	3.00	15
	4.00	18
PH	1.00	19
	2.00	23
	3.00	23
INOC	1.00	33
	2.00	32

Multivariate Tests[c]

Effect		Value	F	Hypothesis df	Error df	Sig.
Intercept	Pillai's Trace	1.000	4388796.1[a]	7.000	35.000	.000
	Wilks' Lambda	.000	4388796.1[a]	7.000	35.000	.000
	Hotelling's Trace	877759.22	4388796.1[a]	7.000	35.000	.000
	Roy's Largest Root	877759.22	4388796.1[a]	7.000	35.000	.000
MULCH	Pillai's Trace	1.608	6.104	21.000	111.000	.000
	Wilks' Lambda	.021	13.801	21.000	101.051	.000
	Hotelling's Trace	20.782	33.318	21.000	101.000	.000
	Roy's Largest Root	19.612	103.662[b]	7.000	37.000	.000
PH	Pillai's Trace	1.295	9.443	14.000	72.000	.000
	Wilks' Lambda	.071	13.715[a]	14.000	70.000	.000
	Hotelling's Trace	7.879	19.135	14.000	68.000	.000
	Roy's Largest Root	7.163	36.838[b]	7.000	36.000	.000
INOC	Pillai's Trace	.323	2.386[a]	7.000	35.000	.042
	Wilks' Lambda	.677	2.386[a]	7.000	35.000	.042
	Hotelling's Trace	.477	2.386[a]	7.000	35.000	.042
	Roy's Largest Root	.477	2.386[a]	7.000	35.000	.042
MULCH * PH	Pillai's Trace	1.663	2.191	42.000	240.000	.000
	Wilks' Lambda	.117	2.312	42.000	167.617	.000
	Hotelling's Trace	2.867	2.275	42.000	200.000	.000
	Roy's Largest Root	1.075	6.140[b]	7.000	40.000	.000
MULCH * INOC	Pillai's Trace	.535	1.148	21.000	111.000	.312
	Wilks' Lambda	.516	1.246	21.000	101.051	.231
	Hotelling's Trace	.840	1.347	21.000	101.000	.164
	Roy's Largest Root	.713	3.770[b]	7.000	37.000	.004
PH * INOC	Pillai's Trace	.291	.875	14.000	72.000	.588
	Wilks' Lambda	.724	.875[a]	14.000	70.000	.589
	Hotelling's Trace	.360	.873	14.000	68.000	.590
	Roy's Largest Root	.287	1.475[b]	7.000	36.000	.207
MULCH * PH * INOC	Pillai's Trace	1.154	1.361	42.000	240.000	.080
	Wilks' Lambda	.202	1.623	42.000	167.617	.017
	Hotelling's Trace	2.464	1.956	42.000	200.000	.001
	Roy's Largest Root	1.863	10.644[b]	7.000	40.000	.000

a. Exact statistic

b. The statistic is an upper bound on F that yields a lower bound on the significance level.

c. Design: Intercept+MULCH+PH+INOC+MULCH * PH+MULCH * INOC+PH * INOC+MULCH * PH * INOC

Levene's Test of Equality of Error Variances[a]

	F	df1	df2	Sig.
MBC1	3.322	23	41	.000
OC1	3.624	23	41	.000
CEC1	3.489	23	41	.000
PHV1	3.358	23	41	.000
TN1	1.752	23	41	.058
TP1	3.238	23	41	.001
AP1	1.728	23	41	.062

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.

a. Design: Intercept+MULCH+PH+INOC+MULCH * PH+MULCH * INOC+PH * INOC+MULCH * PH * INOC

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	MBC1	111.567[a]	23	4.851	3.268	.000
	OC1	7.591[b]	23	.330	3.704	.000
	CEC1	9.550E-03[c]	23	4.152E-04	2.743	.002
	PHV1	.520[d]	23	2.263E-02	37.738	.000
	TN1	9.517E-02[e]	23	4.138E-03	1.576	.100
	TP1	.742[f]	23	3.225E-02	1.844	.043
	AP1	.201[g]	23	8.750E-03	4.642	.000
Intercept	MBC1	17674.453	1	17674.453	11906.360	.000
	OC1	2178.661	1	2178.661	24452.387	.000
	CEC1	185.964	1	185.964	1228725.0	.000
	PHV1	255.150	1	255.150	425575.36	.000
	TN1	14.267	1	14.267	5433.492	.000
	TP1	230.150	1	230.150	13161.455	.000
	AP1	19.719	1	19.719	10461.682	.000
MULCH	MBC1	36.347	3	12.116	8.162	.000
	OC1	4.280	3	1.427	16.012	.000
	CEC1	3.202E-03	3	1.067E-03	7.052	.001
	PHV1	.282	3	9.391E-02	156.643	.000
	TN1	1.766E-02	3	5.886E-03	2.242	.098
	TP1	1.937E-02	3	6.456E-03	.369	.776
	AP1	9.821E-02	3	3.274E-02	17.368	.000
PH	MBC1	54.447	2	27.223	18.339	.000
	OC1	1.092	2	.546	6.129	.005
	CEC1	2.149E-03	2	1.075E-03	7.100	.002
	PHV1	.157	2	7.863E-02	131.146	.000
	TN1	4.694E-03	2	2.347E-03	.894	.417
	TP1	.250	2	.125	7.151	.002
	AP1	1.026E-02	2	5.128E-03	2.721	.078
INOC	MBC1	3.868	1	3.868	2.605	.114
	OC1	1.094E-02	1	1.094E-02	.123	.728
	CEC1	3.627E-05	1	3.627E-05	.240	.627
	PHV1	1.166E-04	1	1.166E-04	.194	.662
	TN1	5.894E-04	1	5.894E-04	.224	.638
	TP1	3.677E-02	1	3.677E-02	2.103	.155
	AP1	2.826E-02	1	2.826E-02	14.994	.000
MULCH * PH	MBC1	13.991	6	2.332	1.571	.180
	OC1	.548	6	9.134E-02	1.025	.423
	CEC1	1.150E-03	6	1.917E-04	1.267	.294
	PHV1	1.948E-02	6	3.247E-03	5.416	.000
	TN1	4.658E-02	6	7.764E-03	2.957	.017
	TP1	.185	6	3.085E-02	1.764	.131
	AP1	4.325E-02	6	7.208E-03	3.824	.004
MULCH * INOC	MBC1	1.468	3	.489	.330	.804
	OC1	.114	3	3.796E-02	.426	.735
	CEC1	2.388E-04	3	7.959E-05	.526	.667
	PHV1	1.400E-02	3	4.666E-03	7.783	.000
	TN1	3.047E-03	3	1.016E-03	.387	.763
	TP1	4.720E-02	3	1.573E-02	.900	.450
	AP1	9.429E-03	3	3.143E-03	1.667	.189

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.
PH * INOC	MBC1	1.433	2	.716	.483	.621
	OC1	.714	2	.357	4.008	.026
	CEC1	1.348E-03	2	6.738E-04	4.452	.018
	PHV1	1.065E-03	2	5.325E-04	.888	.419
	TN1	2.537E-03	2	1.268E-03	.483	.620
	TP1	4.477E-02	2	2.239E-02	1.280	.289
	AP1	3.394E-03	2	1.697E-03	.900	.414
MULCH * PH * INOC	MBC1	7.761	6	1.293	.871	.524
	OC1	.361	6	6.012E-02	.675	.671
	CEC1	6.751E-04	6	1.125E-04	.743	.618
	PHV1	3.275E-02	6	5.459E-03	9.105	.000
	TN1	4.762E-03	6	7.937E-04	.302	.932
	TP1	.128	6	2.138E-02	1.223	.315
	AP1	6.536E-03	6	1.089E-03	.578	.746
Error	MBC1	60.863	41	1.484		
	OC1	3.653	41	8.910E-02		
	CEC1	6.205E-03	41	1.513E-04		
	PHV1	2.458E-02	41	5.995E-04		
	TN1	.108	41	2.626E-03		
	TP1	.717	41	1.749E-02		
	AP1	7.728E-02	41	1.885E-03		
Total	MBC1	19174.420	65			
	OC1	2352.740	65			
	CEC1	199.491	65			
	PHV1	274.620	65			
	TN1	15.460	65			
	TP1	249.440	65			
	AP1	21.093	65			
Corrected Total	MBC1	172.429	64			
	OC1	11.244	64			
	CEC1	1.575E-02	64			
	PHV1	.545	64			
	TN1	.203	64			
	TP1	1.459	64			
	AP1	.279	64			

a. R Squared = .647 (Adjusted R Squared = .449)

b. R Squared = .675 (Adjusted R Squared = .493)

c. R Squared = .606 (Adjusted R Squared = .385)

d. R Squared = .955 (Adjusted R Squared = .930)

e. R Squared = .469 (Adjusted R Squared = .171)

f. R Squared = .508 (Adjusted R Squared = .233)

g. R Squared = .723 (Adjusted R Squared = .567)

1. MULCH

Dependent Variable	MULCH	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
MBC1	1.00	17.576	.310	16.949	18.202
	2.00	17.963	.310	17.337	18.590
	3.00	16.695	.342	16.004	17.385
	4.00	16.061	.287	15.481	16.641
OC1	1.00	6.057	.076	5.904	6.211
	2.00	6.347	.076	6.194	6.501
	3.00	5.932	.084	5.763	6.101
	4.00	5.640	.070	5.498	5.782
CEC1	1.00	1.751	.003	1.744	1.757
	2.00	1.762	.003	1.755	1.768
	3.00	1.751	.003	1.744	1.758
	4.00	1.742	.003	1.736	1.748
PHV1	1.00	2.108	.006	2.095	2.121
	2.00	2.130	.006	2.118	2.143
	3.00	1.973	.007	1.959	1.987
	4.00	1.994	.006	1.982	2.006
TN1	1.00	.509	.013	.482	.535
	2.00	.494	.013	.467	.520
	3.00	.464	.014	.435	.493
	4.00	.474	.012	.449	.498
TP1	1.00	1.950	.034	1.882	2.018
	2.00	1.925	.034	1.857	1.993
	3.00	1.945	.037	1.870	2.020
	4.00	1.973	.031	1.910	2.036
AP1	1.00	.603	.011	.581	.625
	2.00	.607	.011	.585	.630
	3.00	.558	.012	.533	.583
	4.00	.513	.010	.492	.534

2. PH

Dependent Variable	PH	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
MBC1	1.00	16.482	.298	15.880	17.085
	2.00	18.366	.256	17.848	18.883
	3.00	16.373	.256	15.855	16.890
OC1	1.00	5.919	.073	5.772	6.067
	2.00	5.888	.063	5.761	6.015
	3.00	6.176	.063	6.049	6.303
CEC1	1.00	1.748	.003	1.742	1.754
	2.00	1.746	.003	1.741	1.752
	3.00	1.759	.003	1.754	1.765
PHV1	1.00	2.006	.006	1.994	2.018
	2.00	2.121	.005	2.111	2.132
	3.00	2.027	.005	2.017	2.038
TN1	1.00	.476	.013	.450	.501
	2.00	.497	.011	.475	.519
	3.00	.483	.011	.461	.505
TP1	1.00	1.853	.032	1.788	1.919
	2.00	1.988	.028	1.931	2.044
	3.00	2.004	.028	1.948	2.060
AP1	1.00	.584	.011	.563	.606
	2.00	.553	.009	.534	.571
	3.00	.574	.009	.555	.592

3. INOC

Dependent Variable	INOC	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
MBC1	1.00	16.821	.223	16.370	17.272
	2.00	17.326	.219	16.883	17.769
OC1	1.00	6.008	.055	5.897	6.118
	2.00	5.981	.054	5.872	6.089
CEC1	1.00	1.752	.002	1.748	1.757
	2.00	1.751	.002	1.746	1.755
PHV1	1.00	2.050	.004	2.041	2.059
	2.00	2.053	.004	2.044	2.062
TN1	1.00	.488	.009	.469	.507
	2.00	.482	.009	.463	.501
TP1	1.00	1.924	.024	1.875	1.973
	2.00	1.973	.024	1.925	2.021
AP1	1.00	.592	.008	.576	.608
	2.00	.549	.008	.533	.564

4. MULCH * PH

Dependent Variable	MULCH	PH	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
MBC1	1.00	1.00	17.840	.556	16.717	18.963
		2.00	18.803	.497	17.799	19.808
		3.00	16.084	.556	14.961	17.207
	2.00	1.00	17.077	.556	15.954	18.200
		2.00	19.820	.556	18.697	20.943
		3.00	16.992	.497	15.987	17.996

4. MULCH * PH

Dependent Variable	MULCH	PH	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
MBC1	3.00	1.00	15.692	.746	14.186	17.199
		2.00	17.942	.497	16.937	18.946
		3.00	16.450	.497	15.445	17.454
	4.00	1.00	15.320	.497	14.316	16.325
		2.00	16.897	.497	15.893	17.902
		3.00	15.965	.497	14.960	16.970
OC1	1.00	1.00	6.142	.136	5.867	6.417
		2.00	5.929	.122	5.683	6.175
		3.00	6.101	.136	5.826	6.376
	2.00	1.00	6.222	.136	5.947	6.497
		2.00	6.301	.136	6.026	6.576
		3.00	6.519	.122	6.273	6.766
	3.00	1.00	5.710	.183	5.341	6.079
		2.00	5.928	.122	5.682	6.174
		3.00	6.159	.122	5.913	6.405
	4.00	1.00	5.604	.122	5.358	5.850
		2.00	5.392	.122	5.146	5.638
		3.00	5.925	.122	5.679	6.171
CEC1	1.00	1.00	1.755	.006	1.744	1.766
		2.00	1.745	.005	1.735	1.755
		3.00	1.752	.006	1.741	1.764
	2.00	1.00	1.756	.006	1.745	1.767
		2.00	1.760	.006	1.748	1.771
		3.00	1.769	.005	1.759	1.780
	3.00	1.00	1.741	.008	1.726	1.756
		2.00	1.750	.005	1.740	1.761
		3.00	1.761	.005	1.751	1.771
	4.00	1.00	1.741	.005	1.731	1.752
		2.00	1.730	.005	1.720	1.740
		3.00	1.754	.005	1.744	1.765
PHV1	1.00	1.00	2.042	.011	2.020	2.065
		2.00	2.213	.010	2.193	2.234
		3.00	2.068	.011	2.045	2.091
	2.00	1.00	2.094	.011	2.071	2.116
		2.00	2.203	.011	2.180	2.225
		3.00	2.094	.010	2.074	2.115
	3.00	1.00	1.926	.015	1.896	1.957
		2.00	2.028	.010	2.008	2.049
		3.00	1.965	.010	1.945	1.985
	4.00	1.00	1.960	.010	1.940	1.980
		2.00	2.041	.010	2.021	2.061
		3.00	1.981	.010	1.961	2.002
TN1	1.00	1.00	.508	.023	.461	.556
		2.00	.534	.021	.492	.576
		3.00	.484	.023	.436	.531
	2.00	1.00	.420	.023	.373	.467
		2.00	.539	.023	.491	.586
		3.00	.523	.021	.481	.565
	3.00	1.00	.492	.031	.429	.555
		2.00	.447	.021	.405	.489
		3.00	.453	.021	.411	.496

4. MULCH * PH

Dependent Variable	MULCH	PH	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
TN1	4.00	1.00	.482	.021	.439	.524
		2.00	.468	.021	.425	.510
		3.00	.472	.021	.429	.514
TP1	1.00	1.00	1.770	.060	1.648	1.892
		2.00	2.033	.054	1.924	2.142
		3.00	2.045	.060	1.923	2.167
	2.00	1.00	1.761	.060	1.639	1.883
		2.00	2.007	.060	1.886	2.129
		3.00	2.007	.054	1.898	2.116
	3.00	1.00	1.920	.081	1.756	2.083
		2.00	1.963	.054	1.854	2.072
		3.00	1.953	.054	1.844	2.062
	4.00	1.00	1.962	.054	1.853	2.071
		2.00	1.947	.054	1.838	2.056
		3.00	2.010	.054	1.901	2.119
AP1	1.00	1.00	.625	.020	.585	.665
		2.00	.546	.018	.510	.582
		3.00	.637	.020	.597	.677
	2.00	1.00	.589	.020	.549	.629
		2.00	.649	.020	.609	.689
		3.00	.584	.018	.549	.620
	3.00	1.00	.590	.027	.536	.643
		2.00	.536	.018	.500	.572
		3.00	.548	.018	.512	.584
	4.00	1.00	.534	.018	.498	.569
		2.00	.481	.018	.445	.517
		3.00	.525	.018	.489	.561

5. MULCH * INOC

Dependent Variable	MULCH	INOC	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
MBC1	1.00	1.00	17.211	.439	16.325	18.096
		2.00	17.941	.439	17.055	18.827
	2.00	1.00	17.613	.406	16.793	18.433
		2.00	18.313	.469	17.366	19.260
	3.00	1.00	16.403	.524	15.344	17.462
		2.00	16.986	.439	16.100	17.872
	4.00	1.00	16.057	.406	15.237	16.877
		2.00	16.064	.406	15.244	16.885
OC1	1.00	1.00	6.075	.107	5.858	6.292
		2.00	6.040	.107	5.823	6.257
	2.00	1.00	6.371	.099	6.170	6.572
		2.00	6.323	.115	6.091	6.556
	3.00	1.00	5.877	.128	5.618	6.136
		2.00	5.988	.107	5.771	6.205
	4.00	1.00	5.708	.099	5.507	5.909
		2.00	5.572	.099	5.371	5.773
CEC1	1.00	1.00	1.752	.004	1.743	1.761
		2.00	1.750	.004	1.741	1.759

5. MULCH * INOC

Dependent Variable	MULCH	INOC	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
CEC1	2.00	1.00	1.763	.004	1.754	1.771
		2.00	1.761	.005	1.751	1.770
	3.00	1.00	1.749	.005	1.738	1.759
		2.00	1.753	.004	1.744	1.762
	4.00	1.00	1.745	.004	1.737	1.754
		2.00	1.739	.004	1.730	1.747
PHV1	1.00	1.00	2.089	.009	2.071	2.106
		2.00	2.127	.009	2.109	2.145
	2.00	1.00	2.121	.008	2.105	2.138
		2.00	2.139	.009	2.120	2.158
	3.00	1.00	1.977	.011	1.955	1.998
		2.00	1.970	.009	1.952	1.988
	4.00	1.00	2.014	.008	1.997	2.030
		2.00	1.975	.008	1.958	1.991
TN1	1.00	1.00	.523	.018	.486	.561
		2.00	.494	.018	.457	.531
	2.00	1.00	.497	.017	.462	.531
		2.00	.491	.020	.451	.530
	3.00	1.00	.461	.022	.417	.506
		2.00	.467	.018	.430	.505
	4.00	1.00	.471	.017	.437	.506
		2.00	.476	.017	.441	.510
TP1	1.00	1.00	1.884	.048	1.788	1.981
		2.00	2.015	.048	1.919	2.111
	2.00	1.00	1.914	.044	1.825	2.003
		2.00	1.936	.051	1.834	2.039
	3.00	1.00	1.916	.057	1.801	2.030
		2.00	1.975	.048	1.879	2.071
	4.00	1.00	1.981	.044	1.892	2.070
		2.00	1.965	.044	1.876	2.054
AP1	1.00	1.00	.627	.016	.595	.658
		2.00	.579	.016	.548	.611
	2.00	1.00	.610	.014	.581	.640
		2.00	.604	.017	.570	.638
	3.00	1.00	.581	.019	.543	.618
		2.00	.535	.016	.504	.567
	4.00	1.00	.550	.014	.521	.579
		2.00	.476	.014	.447	.506

6. PH * INOC

Dependent Variable	PH	INOC	Mean	Std. Error	95% Confidence Interval	
					Lower Bound	Upper Bound
MBC1	1.00	1.00	16.282	.431	15.412	17.152
		2.00	16.683	.412	15.850	17.516
	2.00	1.00	17.914	.352	17.203	18.624
		2.00	18.818	.373	18.064	19.571
	3.00	1.00	16.267	.373	15.514	17.021
		2.00	16.478	.352	15.768	17.188
OC1	1.00	1.00	6.061	.106	5.848	6.274
		2.00	5.778	.101	5.574	5.982
	2.00	1.00	5.914	.086	5.740	6.088
		2.00	5.862	.091	5.677	6.046
	3.00	1.00	6.049	.091	5.864	6.233
		2.00	6.303	.086	6.129	6.477
CEC1	1.00	1.00	1.755	.004	1.746	1.763
		2.00	1.742	.004	1.734	1.751
	2.00	1.00	1.748	.004	1.741	1.755
		2.00	1.745	.004	1.737	1.752
	3.00	1.00	1.754	.004	1.746	1.761
		2.00	1.765	.004	1.758	1.772
PHV1	1.00	1.00	2.009	.009	1.991	2.026
		2.00	2.003	.008	1.986	2.019
	2.00	1.00	2.114	.007	2.100	2.129
		2.00	2.128	.007	2.113	2.143
	3.00	1.00	2.027	.007	2.012	2.042
		2.00	2.027	.007	2.013	2.042
TN1	1.00	1.00	.479	.018	.442	.515
		2.00	.472	.017	.437	.508
	2.00	1.00	.507	.015	.478	.537
		2.00	.486	.016	.454	.518
	3.00	1.00	.479	.016	.447	.510
		2.00	.487	.015	.457	.517
TP1	1.00	1.00	1.806	.047	1.712	1.901
		2.00	1.900	.045	1.810	1.991
	2.00	1.00	1.948	.038	1.871	2.025
		2.00	2.027	.040	1.946	2.109
	3.00	1.00	2.017	.040	1.935	2.098
		2.00	1.991	.038	1.914	2.068
AP1	1.00	1.00	.617	.015	.586	.648
		2.00	.552	.015	.522	.582
	2.00	1.00	.571	.013	.546	.597
		2.00	.535	.013	.508	.561
	3.00	1.00	.588	.013	.561	.615
		2.00	.560	.013	.534	.585

7. MULCH * PH * INOC

Dependent Variable	MULCH	PH	INOC	Mean	Std. Error	95% Confidence Interval	
						Lower Bound	Upper Bound
MBC1	1.00	1.00	1.00	17.676	.703	16.255	19.096
			2.00	18.004	.862	16.264	19.744
		2.00	1.00	18.764	.703	17.343	20.184
			2.00	18.843	.703	17.423	20.264

7. MULCH * PH * INOC

Dependent Variable	MULCH	PH	INOC	Mean	Std. Error	95% Confidence Interval	
						Lower Bound	Upper Bound
MBC1	1.00	3.00	1.00	15.192	.862	13.453	16.932
			2.00	16.976	.703	15.555	18.396
	2.00	1.00	1.00	16.546	.703	15.126	17.967
			2.00	17.608	.862	15.868	19.348
		2.00	1.00	19.360	.703	17.940	20.781
			2.00	20.280	.862	18.540	22.020
		3.00	1.00	16.932	.703	15.512	18.353
			2.00	17.051	.703	15.631	18.472
	3.00	1.00	1.00	15.461	1.218	13.001	17.922
			2.00	15.924	.862	14.184	17.664
		2.00	1.00	17.405	.703	15.984	18.825
			2.00	18.478	.703	17.058	19.899
		3.00	1.00	16.344	.703	14.923	17.764
			2.00	16.555	.703	15.135	17.976
	4.00	1.00	1.00	15.445	.703	14.024	16.866
			2.00	15.195	.703	13.775	16.616
		2.00	1.00	16.126	.703	14.705	17.546
			2.00	17.669	.703	16.248	19.089
		3.00	1.00	16.601	.703	15.180	18.022
			2.00	15.329	.703	13.908	16.750
OC1	1.00	1.00	1.00	6.323	.172	5.975	6.672
			2.00	5.961	.211	5.534	6.387
		2.00	1.00	5.975	.172	5.627	6.323
			2.00	5.884	.172	5.536	6.232
		3.00	1.00	5.926	.211	5.500	6.352
			2.00	6.276	.172	5.928	6.624
	2.00	1.00	1.00	6.310	.172	5.962	6.658
			2.00	6.134	.211	5.707	6.560
		2.00	1.00	6.281	.172	5.933	6.629
			2.00	6.321	.211	5.894	6.747
		3.00	1.00	6.523	.172	6.175	6.871
			2.00	6.516	.172	6.168	6.864
	3.00	1.00	1.00	5.711	.298	5.108	6.313
			2.00	5.710	.211	5.283	6.136
		2.00	1.00	5.888	.172	5.540	6.236
			2.00	5.969	.172	5.621	6.317
		3.00	1.00	6.033	.172	5.685	6.381
			2.00	6.285	.172	5.937	6.633
	4.00	1.00	1.00	5.900	.172	5.552	6.248
			2.00	5.308	.172	4.960	5.656
		2.00	1.00	5.511	.172	5.163	5.859
			2.00	5.273	.172	4.925	5.621
		3.00	1.00	5.714	.172	5.366	6.062
			2.00	6.136	.172	5.788	6.484
CEC1	1.00	1.00	1.00	1.764	.007	1.749	1.778
			2.00	1.746	.009	1.729	1.764
		2.00	1.00	1.747	.007	1.732	1.761
			2.00	1.743	.007	1.729	1.757
		3.00	1.00	1.745	.009	1.727	1.762
			2.00	1.760	.007	1.746	1.775
	2.00	1.00	1.00	1.760	.007	1.746	1.774
			2.00	1.752	.009	1.734	1.770

7. MULCH * PH * INOC

Dependent Variable	MULCH	PH	INOC	Mean	Std. Error	95% Confidence Interval	
						Lower Bound	Upper Bound
CEC1	2.00	2.00	1.00	1.759	.007	1.744	1.773
			2.00	1.760	.009	1.743	1.778
		3.00	1.00	1.769	.007	1.755	1.784
			2.00	1.769	.007	1.755	1.784
	3.00	1.00	1.00	1.741	.012	1.716	1.766
			2.00	1.741	.009	1.724	1.759
		2.00	1.00	1.749	.007	1.734	1.763
			2.00	1.752	.007	1.738	1.766
		3.00	1.00	1.756	.007	1.741	1.770
			2.00	1.766	.007	1.752	1.780
	4.00	1.00	1.00	1.753	.007	1.739	1.768
			2.00	1.730	.007	1.715	1.744
		2.00	1.00	1.737	.007	1.723	1.752
			2.00	1.723	.007	1.709	1.738
		3.00	1.00	1.746	.007	1.731	1.760
			2.00	1.763	.007	1.749	1.777
PHV1	1.00	1.00	1.00	2.045	.014	2.017	2.074
			2.00	2.039	.017	2.005	2.074
		2.00	1.00	2.166	.014	2.138	2.195
			2.00	2.261	.014	2.232	2.289
		3.00	1.00	2.054	.017	2.019	2.089
			2.00	2.082	.014	2.053	2.110
	2.00	1.00	1.00	2.091	.014	2.063	2.120
			2.00	2.096	.017	2.061	2.131
		2.00	1.00	2.182	.014	2.154	2.211
			2.00	2.223	.017	2.188	2.258
		3.00	1.00	2.090	.014	2.061	2.118
			2.00	2.099	.014	2.071	2.128
	3.00	1.00	1.00	1.962	.024	1.913	2.012
			2.00	1.891	.017	1.856	1.926
		2.00	1.00	2.011	.014	1.982	2.039
			2.00	2.046	.014	2.018	2.075
		3.00	1.00	1.957	.014	1.928	1.985
			2.00	1.973	.014	1.944	2.002
	4.00	1.00	1.00	1.936	.014	1.908	1.965
			2.00	1.984	.014	1.956	2.013
		2.00	1.00	2.098	.014	2.069	2.126
			2.00	1.984	.014	1.955	2.013
		3.00	1.00	2.007	.014	1.978	2.036
			2.00	1.956	.014	1.927	1.984
TN1	1.00	1.00	1.00	.519	.030	.460	.579
			2.00	.497	.036	.424	.571
		2.00	1.00	.556	.030	.496	.616
			2.00	.512	.030	.452	.571
		3.00	1.00	.494	.036	.421	.568
			2.00	.473	.030	.413	.533
	2.00	1.00	1.00	.410	.030	.350	.470
			2.00	.430	.036	.357	.503
		2.00	1.00	.558	.030	.498	.618
			2.00	.519	.036	.446	.593
		3.00	1.00	.523	.030	.463	.583
			2.00	.523	.030	.463	.582

7. MULCH * PH * INOC

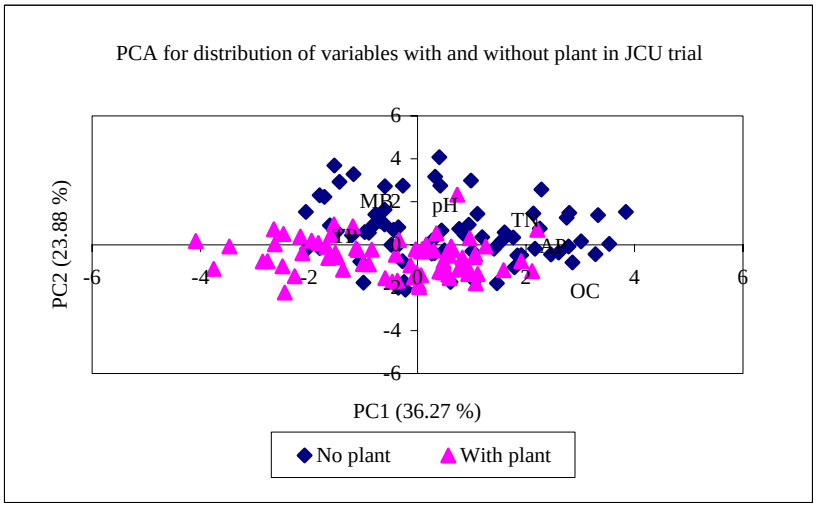
Dependent Variable	MULCH	PH	INOC	Mean	Std. Error	95% Confidence Interval	
						Lower Bound	Upper Bound
TN1	3.00	1.00	1.00	.510	.051	.406	.613
			2.00	.474	.036	.401	.547
		2.00	1.00	.434	.030	.374	.494
			2.00	.460	.030	.400	.520
		3.00	1.00	.439	.030	.380	.499
			2.00	.467	.030	.408	.527
	4.00	1.00	1.00	.475	.030	.415	.535
			2.00	.488	.030	.429	.548
		2.00	1.00	.482	.030	.422	.542
			2.00	.453	.030	.394	.513
		3.00	1.00	.457	.030	.398	.517
			2.00	.486	.030	.426	.546
TP1	1.00	1.00	1.00	1.706	.076	1.551	1.860
			2.00	1.835	.094	1.646	2.024
		2.00	1.00	1.980	.076	1.826	2.134
			2.00	2.086	.076	1.932	2.241
		3.00	1.00	1.967	.094	1.779	2.156
			2.00	2.123	.076	1.969	2.277
	2.00	1.00	1.00	1.796	.076	1.642	1.950
			2.00	1.727	.094	1.538	1.916
		2.00	1.00	1.885	.076	1.731	2.039
			2.00	2.130	.094	1.941	2.319
		3.00	1.00	2.062	.076	1.907	2.216
			2.00	1.952	.076	1.798	2.106
	3.00	1.00	1.00	1.819	.132	1.552	2.086
			2.00	2.020	.094	1.831	2.209
		2.00	1.00	1.971	.076	1.817	2.125
			2.00	1.955	.076	1.801	2.109
		3.00	1.00	1.956	.076	1.802	2.110
			2.00	1.950	.076	1.796	2.104
	4.00	1.00	1.00	1.905	.076	1.751	2.059
			2.00	2.020	.076	1.865	2.174
		2.00	1.00	1.956	.076	1.801	2.110
			2.00	1.938	.076	1.784	2.092
		3.00	1.00	2.082	.076	1.927	2.236
			2.00	1.939	.076	1.785	2.093
AP1	1.00	1.00	1.00	.668	.025	.618	.719
			2.00	.582	.031	.520	.644
		2.00	1.00	.559	.025	.509	.610
			2.00	.533	.025	.483	.584
		3.00	1.00	.652	.031	.590	.714
			2.00	.622	.025	.571	.672
	2.00	1.00	1.00	.600	.025	.549	.651
			2.00	.578	.031	.516	.640
		2.00	1.00	.636	.025	.586	.687
			2.00	.661	.031	.599	.723
		3.00	1.00	.595	.025	.544	.646
			2.00	.574	.025	.523	.625
	3.00	1.00	1.00	.627	.043	.539	.715
			2.00	.552	.031	.490	.614
		2.00	1.00	.569	.025	.518	.620
			2.00	.503	.025	.452	.554

7. MULCH * PH * INOC

Dependent Variable	MULCH	PH	INOC	Mean	Std. Error	95% Confidence Interval	
						Lower Bound	Upper Bound
AP1	3.00	3.00	1.00	.546	.025	.495	.597
			2.00	.551	.025	.500	.601
	4.00	1.00	1.00	.571	.025	.521	.622
			2.00	.496	.025	.445	.546
		2.00	1.00	.520	.025	.470	.571
			2.00	.441	.025	.391	.492
		3.00	1.00	.558	.025	.507	.608
			2.00	.492	.025	.441	.543

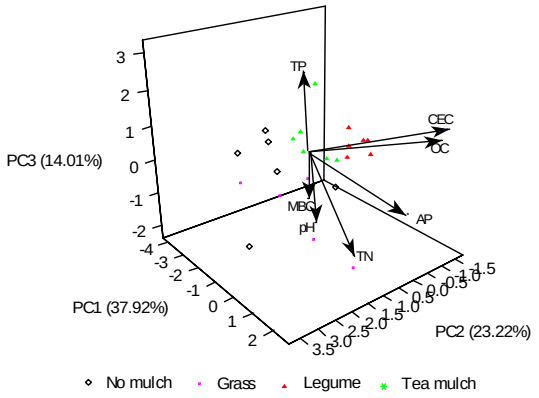
Appendix 4

Principal Component Analysis (PCA) graph for with and without tea plants of nursery trial at JCU, Australia

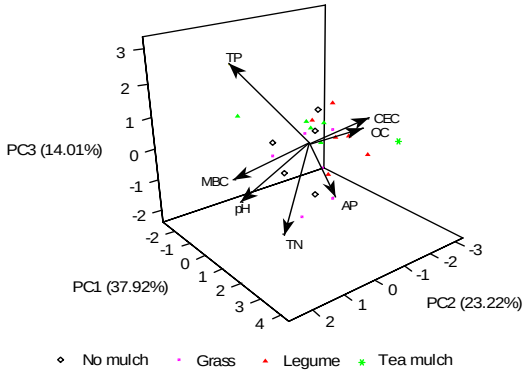


Appendix 5

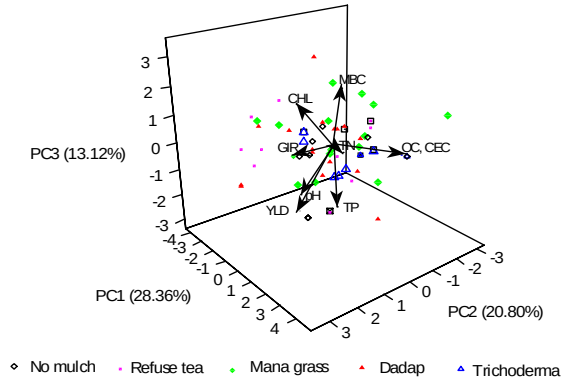
Effect of mulches on soil parameters at dolomite in nursery trial at JCU



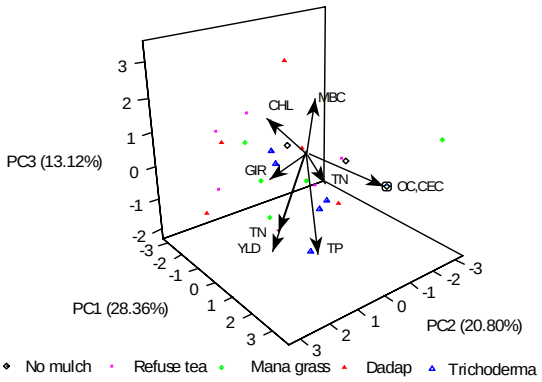
Effect of mulches on soil properties at minplus in nursery trail at JCU



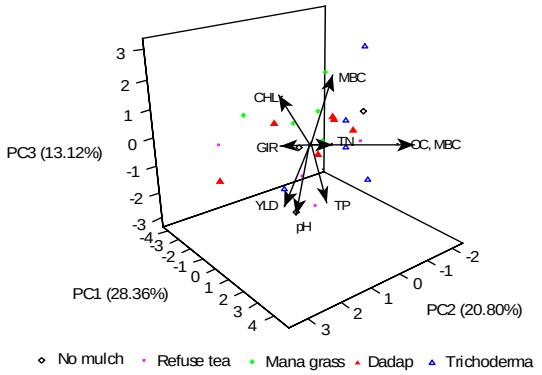
Effect of mulches on all variables in mature tea



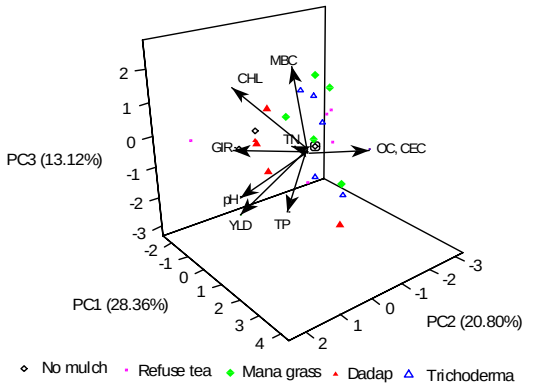
Effect of mulches on soil parameters at no pH modifier in mature tea



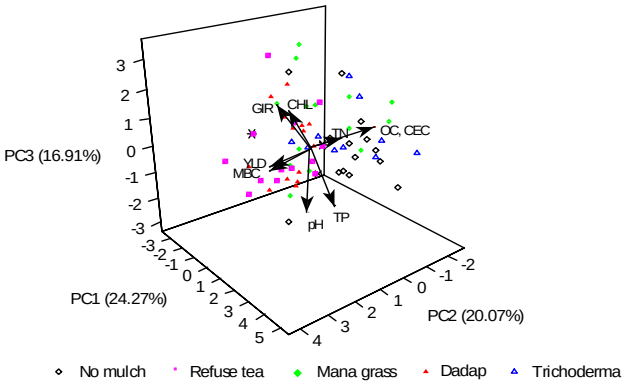
Effect of mulches on soil parameters at dolomite modifier in mature tea



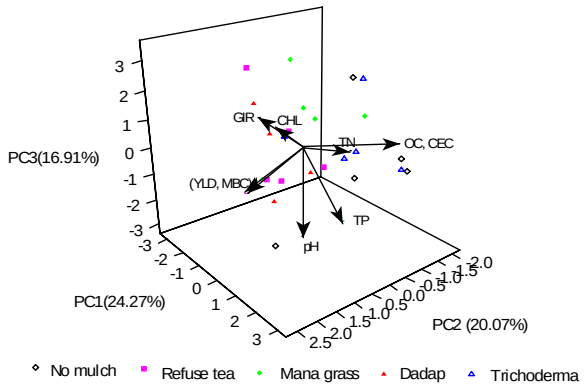
Effect of mulches on soil parameters at minplus modifier in mature tea



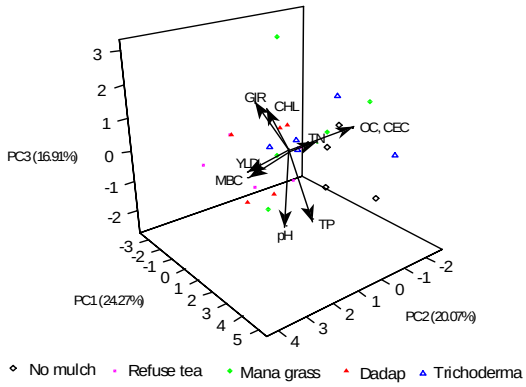
Effect of mulches on all variables in young tea



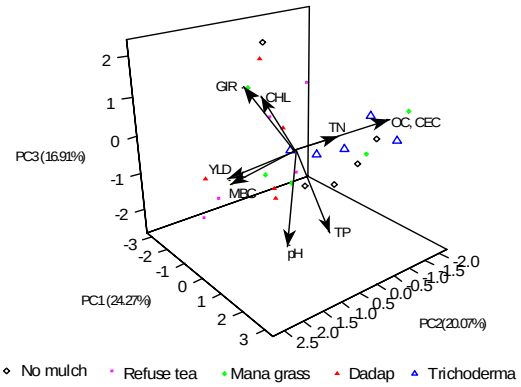
Effect of mulches on soil parameters at no pH modifier in young tea



Effect of mulches on all variable at dolomite in young tea

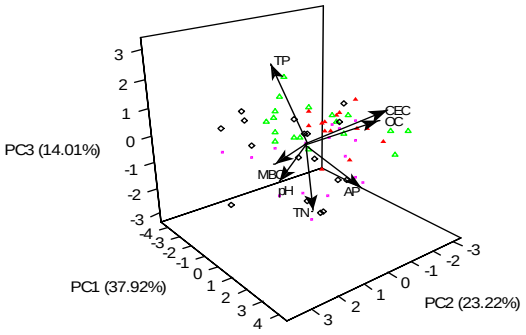


Effect of mulches on all variable tested at minplus in young tea



No mulch Refuse tea Mana grass Dadap Trichoderma

Effect of mulches on soil properties in nursery trial at JCU



No mulch Grass Legume Tea mulch

Appendix 6

Factor Analysis

KMO and Bartlett's Test

Kaiser-Meyer-Olkin Measure of Sampling Adequacy.		.502
Bartlett's Test of Sphericity	Approx. Chi-Square	431.901
	df	21
	Sig.	.000

Communalities

	Initial	Extraction
FORTHOC	1.000	.913
FORTHCEC	1.000	.911
FORTHPHV	1.000	.574
FORTHTN	1.000	.536
FORTHTP	1.000	5.001E-02
FORTHAP	1.000	.493
FORTHMBC	1.000	.414

Extraction Method: Principal Component Analysis.

Total Variance Explained

	Initial Eigenvalues		
Component	Total	% of Variance	Cumulative %
1	2.495	35.647	35.647
2	1.395	19.931	55.579
3	1.181	16.877	72.455
4	.864	12.336	84.791
5	.704	10.051	94.843
6	.359	5.130	99.973
7	1.914E-03	2.734E-02	100.000

Extraction Method: Principal Component Analysis.

Total Variance Explained

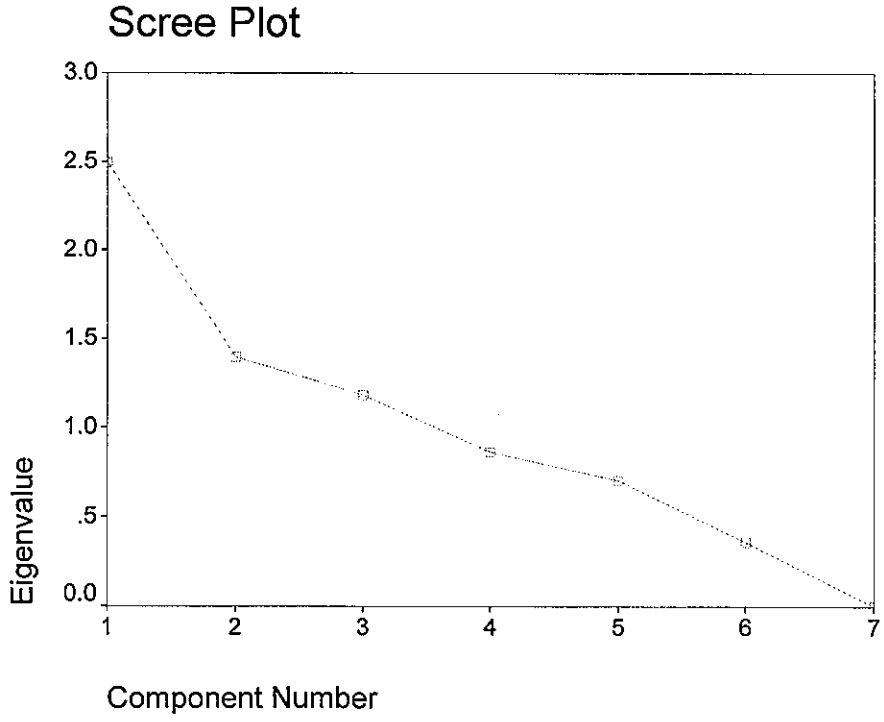
Component	Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %
1	2.495	35.647	35.647
2	1.395	19.931	55.579
3			
4			
5			
6			
7			

Extraction Method: Principal Component Analysis.

Total Variance Explained

Component	Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %
1	2.335	33.364	33.364
2	1.555	22.215	55.579
3			
4			
5			
6			
7			

Extraction Method: Principal Component Analysis.



Component Matrix[a]

	Component	
	1	2
FORTHOC	.899	-.322
FORTHCEC	.903	-.310
FORTHPHV	.550	.521
FORTHTN	.346	.645
FORTHTP	-4.220E-02	-.220
FORTHAP	.284	.643
FORTHMBC	.606	-.217

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

Rotated Component Matrix[a]

	Component	
	1	2
FORTHOC	.954	4.515E-02
FORTHCEC	.953	5.784E-02
FORTHPHV	.310	.691
FORTHTN	7.355E-02	.728
FORTHTP	4.470E-02	-.219
FORTHAP	1.746E-02	.702
FORTHMBC	.643	2.979E-02

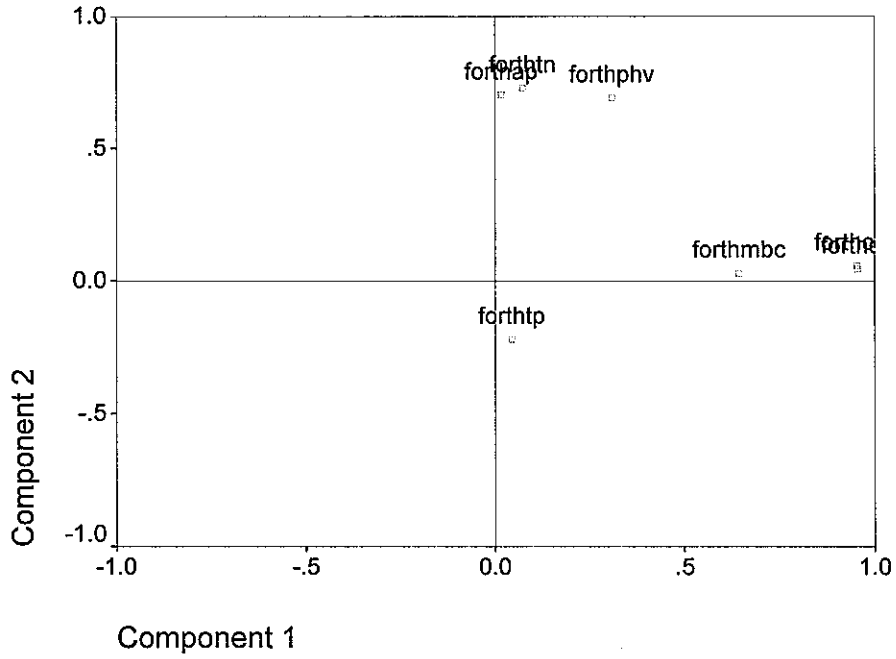
Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.
a. Rotation converged in 3 iterations.

Component Transformation Matrix

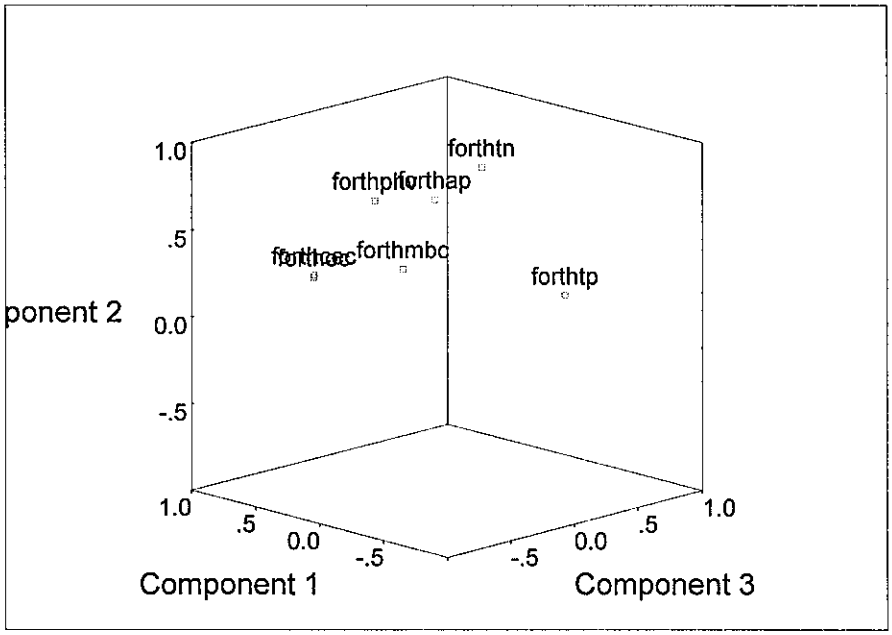
Component	1	2
1	.924	.381
2	-.381	.924

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

Component Plot in Rotated Space



Component Plot in Rotated Space



General Linear Model

Between-Subjects Factors

		N
MULCH	1.00	18
	2.00	18
	3.00	18
	4.00	18
PH	1.00	24
	2.00	24
	3.00	24
INOCU	1.00	36
	2.00	36

Multivariate Tests[c]

Effect		Value	F	Hypothesis df
Intercept	Pillai's Trace	1.000	1.44E+08[a]	7.000
	Wilks' Lambda	.000	1.44E+08[a]	7.000
	Hotelling's Trace	24045632	1.44E+08[a]	7.000
	Roy's Largest Root	24045632	1.44E+08[a]	7.000
MULCH	Pillai's Trace	1.884	10.606	21.000
	Wilks' Lambda	.022	16.109	21.000
	Hotelling's Trace	10.575	20.478	21.000
	Roy's Largest Root	6.664	41.887[b]	7.000
PH	Pillai's Trace	1.235	9.912	14.000
	Wilks' Lambda	.074	16.023[a]	14.000
	Hotelling's Trace	8.310	24.336	14.000
	Roy's Largest Root	7.774	47.757[b]	7.000
INOCU	Pillai's Trace	.245	1.946[a]	7.000
	Wilks' Lambda	.755	1.946[a]	7.000
	Hotelling's Trace	.324	1.946[a]	7.000
	Roy's Largest Root	.324	1.946[a]	7.000
MULCH * PH	Pillai's Trace	1.985	3.319	42.000
	Wilks' Lambda	.038	4.810	42.000
	Hotelling's Trace	7.028	6.749	42.000
	Roy's Largest Root	5.088	34.159[b]	7.000
MULCH * INOCU	Pillai's Trace	.575	1.490	21.000
	Wilks' Lambda	.498	1.583	21.000
	Hotelling's Trace	.865	1.675	21.000
	Roy's Largest Root	.674	4.233[b]	7.000
PH * INOCU	Pillai's Trace	.645	2.922	14.000
	Wilks' Lambda	.444	3.009[a]	14.000
	Hotelling's Trace	1.056	3.092	14.000
	Roy's Largest Root	.810	4.976[b]	7.000
MULCH * PH * INOCU	Pillai's Trace	1.388	2.020	42.000
	Wilks' Lambda	.153	2.352	42.000
	Hotelling's Trace	2.741	2.633	42.000
	Roy's Largest Root	1.646	11.053[b]	7.000

Multivariate Tests[c]

Effect		Error df	Sig.
Intercept	Pillai's Trace	42.000	.000
	Wilks' Lambda	42.000	.000
	Hotelling's Trace	42.000	.000
	Roy's Largest Root	42.000	.000
MULCH	Pillai's Trace	132.000	.000
	Wilks' Lambda	121.151	.000
	Hotelling's Trace	122.000	.000
	Roy's Largest Root	44.000	.000
PH	Pillai's Trace	86.000	.000
	Wilks' Lambda	84.000	.000
	Hotelling's Trace	82.000	.000
	Roy's Largest Root	43.000	.000
INOCU	Pillai's Trace	42.000	.086
	Wilks' Lambda	42.000	.086
	Hotelling's Trace	42.000	.086
	Roy's Largest Root	42.000	.086
MULCH * PH	Pillai's Trace	282.000	.000
	Wilks' Lambda	200.450	.000
	Hotelling's Trace	242.000	.000
	Roy's Largest Root	47.000	.000
MULCH * INOCU	Pillai's Trace	132.000	.091
	Wilks' Lambda	121.151	.064
	Hotelling's Trace	122.000	.044
	Roy's Largest Root	44.000	.001
PH * INOCU	Pillai's Trace	86.000	.001
	Wilks' Lambda	84.000	.001
	Hotelling's Trace	82.000	.001
	Roy's Largest Root	43.000	.000
MULCH * PH * INOCU	Pillai's Trace	282.000	.000
	Wilks' Lambda	200.450	.000
	Hotelling's Trace	242.000	.000
	Roy's Largest Root	47.000	.000

a. Exact statistic

b. The statistic is an upper bound on F that yields a lower bound on the significance level.

c. Design: Intercept+MULCH+PH+INOCU+MULCH * PH+MULCH * INOCU+PH * INOCU+MULCH * PH * INOCU

Levene's Test of Equality of Error Variances[a]

	F	df1	df2	Sig.
FORTHOC	2.164	23	48	.012
FORTHCEC	2.096	23	48	.015
FORTHPHV	2.579	23	48	.003
FORTHTN	4.297	23	48	.000
FORTHTP	9.448	23	48	.000
FORTHAP	2.679	23	48	.002
FORTHMBC	4.063	23	48	.000

Tests the null hypothesis that the error variance of the dependent variable is equal across groups.

a. Design: Intercept+MULCH+PH+INOCU+MULCH * PH+MULCH * INOCU+PH * INOCU+MULCH * PH * INOCU

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	df
Corrected Model	FORTHOC	.292[a]	23
	FORTHCEC	2.463E-03[b]	23
	FORTHPHV	4.993E-02[c]	23
	FORTHTN	8.056E-02[d]	23
	FORTHTP	.117[e]	23
	FORTHAP	.137[f]	23
	FORTHMBC	1.103[g]	23
Intercept	FORTHOC	501.128	1
	FORTHCEC	128.933	1
	FORTHPHV	145.160	1
	FORTHTN	37.361	1
	FORTHTP	134.712	1
	FORTHAP	44.503	1
	FORTHMBC	1167.038	1
MULCH	FORTHOC	.140	3
	FORTHCEC	1.243E-03	3
	FORTHPHV	1.580E-02	3
	FORTHTN	2.537E-02	3
	FORTHTP	4.455E-02	3
	FORTHAP	5.936E-02	3
	FORTHMBC	.374	3
PH	FORTHOC	2.019E-02	2
	FORTHCEC	1.513E-04	2
	FORTHPHV	2.906E-02	2
	FORTHTN	5.020E-03	2
	FORTHTP	7.992E-03	2
	FORTHAP	5.795E-03	2
	FORTHMBC	.224	2
INOCU	FORTHOC	2.605E-07	1
	FORTHCEC	6.664E-07	1
	FORTHPHV	3.592E-04	1
	FORTHTN	1.304E-03	1
	FORTHTP	9.809E-04	1
	FORTHAP	2.732E-05	1
	FORTHMBC	4.452E-04	1
MULCH * PH	FORTHOC	8.299E-02	6
	FORTHCEC	6.521E-04	6
	FORTHPHV	2.917E-03	6
	FORTHTN	3.403E-02	6
	FORTHTP	3.631E-02	6
	FORTHAP	8.811E-03	6
	FORTHMBC	.386	6
MULCH * INOCU	FORTHOC	1.368E-02	3
	FORTHCEC	1.044E-04	3
	FORTHPHV	2.781E-04	3
	FORTHTN	7.133E-04	3
	FORTHTP	4.125E-03	3
	FORTHAP	1.011E-02	3
	FORTHMBC	1.771E-02	3
PH * INOCU	FORTHOC	1.676E-02	2
	FORTHCEC	1.397E-04	2
	FORTHPHV	2.820E-04	2
	FORTHTN	3.746E-03	2
	FORTHTP	9.125E-03	2
	FORTHAP	1.690E-02	2
	FORTHMBC	7.339E-02	2

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	df
MULCH * PH * INOCU	FORTHOC	1.858E-02	6
	FORTHCEC	1.719E-04	6
	FORTHPHV	1.237E-03	6
	FORTHTN	1.037E-02	6
	FORTHTP	1.434E-02	6
	FORTHAP	3.567E-02	6
	FORTHMBC	2.751E-02	6
Error	FORTHOC	7.209E-02	48
	FORTHCEC	6.531E-04	48
	FORTHPHV	4.757E-03	48
	FORTHTN	8.985E-02	48
	FORTHTP	.119	48
	FORTHAP	3.937E-02	48
	FORTHMBC	.285	48
Total	FORTHOC	501.492	72
	FORTHCEC	128.936	72
	FORTHPHV	145.215	72
	FORTHTN	37.531	72
	FORTHTP	134.948	72
	FORTHAP	44.679	72
	FORTHMBC	1168.426	72
Corrected Total	FORTHOC	.364	71
	FORTHCEC	3.116E-03	71
	FORTHPHV	5.469E-02	71
	FORTHTN	.170	71
	FORTHTP	.236	71
	FORTHAP	.176	71
	FORTHMBC	1.387	71

Tests of Between-Subjects Effects

Source	Dependent Variable	Mean Square	F	Sig.
Corrected Model	FORTHOC	1.271E-02	8.463	.000
	FORTHCEC	1.071E-04	7.871	.000
	FORTHPHV	2.171E-03	21.905	.000
	FORTHTN	3.502E-03	1.871	.034
	FORTHTP	5.105E-03	2.060	.017
	FORTHAP	5.942E-03	7.244	.000
	FORTHMBC	4.795E-02	8.084	.000
Intercept	FORTHOC	501.128	333660.63	.000
	FORTHCEC	128.933	9476660.7	.000
	FORTHPHV	145.160	1464600.1	.000
	FORTHTN	37.361	19959.631	.000
	FORTHTP	134.712	54350.658	.000
	FORTHAP	44.503	54255.113	.000
	FORTHMBC	1167.038	196770.80	.000
MULCH	FORTHOC	4.672E-02	31.106	.000
	FORTHCEC	4.144E-04	30.455	.000
	FORTHPHV	5.266E-03	53.129	.000
	FORTHTN	8.457E-03	4.518	.007
	FORTHTP	1.485E-02	5.991	.001
	FORTHAP	1.979E-02	24.121	.000
	FORTHMBC	.125	21.024	.000
PH	FORTHOC	1.009E-02	6.720	.003
	FORTHCEC	7.565E-05	5.561	.007
	FORTHPHV	1.453E-02	146.621	.000
	FORTHTN	2.510E-03	1.341	.271
	FORTHTP	3.996E-03	1.612	.210
	FORTHAP	2.897E-03	3.532	.037
	FORTHMBC	.112	18.887	.000
INOCU	FORTHOC	2.605E-07	.000	.990
	FORTHCEC	6.664E-07	.049	.826
	FORTHPHV	3.592E-04	3.624	.063
	FORTHTN	1.304E-03	.697	.408
	FORTHTP	9.809E-04	.396	.532
	FORTHAP	2.732E-05	.033	.856
	FORTHMBC	4.452E-04	.075	.785
MULCH * PH	FORTHOC	1.383E-02	9.210	.000
	FORTHCEC	1.087E-04	7.988	.000
	FORTHPHV	4.862E-04	4.906	.001
	FORTHTN	5.672E-03	3.030	.014
	FORTHTP	6.052E-03	2.442	.039
	FORTHAP	1.469E-03	1.790	.121
	FORTHMBC	6.427E-02	10.836	.000
MULCH * INOCU	FORTHOC	4.560E-03	3.036	.038
	FORTHCEC	3.478E-05	2.557	.066
	FORTHPHV	9.269E-05	.935	.431
	FORTHTN	2.378E-04	.127	.944
	FORTHTP	1.375E-03	.555	.647
	FORTHAP	3.371E-03	4.109	.011
	FORTHMBC	5.904E-03	.995	.403
PH * INOCU	FORTHOC	8.381E-03	5.580	.007
	FORTHCEC	6.984E-05	5.133	.010
	FORTHPHV	1.410E-04	1.423	.251
	FORTHTN	1.873E-03	1.001	.375
	FORTHTP	4.562E-03	1.841	.170
	FORTHAP	8.449E-03	10.301	.000
	FORTHMBC	3.670E-02	6.187	.004

Tests of Between-Subjects Effects

Source	Dependent Variable	Mean Square	F	Sig.
MULCH * PH * INOCU	FORTHOC	3.097E-03	2.062	.075
	FORTHCEC	2.866E-05	2.106	.070
	FORTHPHV	2.061E-04	2.079	.073
	FORTHTN	1.728E-03	.923	.487
	FORTHTP	2.389E-03	.964	.460
	FORTHAP	5.945E-03	7.247	.000
	FORTHMBC	4.584E-03	.773	.595
Error	FORTHOC	1.502E-03		
	FORTHCEC	1.361E-05		
	FORTHPHV	9.911E-05		
	FORTHTN	1.872E-03		
	FORTHTP	2.479E-03		
	FORTHAP	8.203E-04		
	FORTHMBC	5.931E-03		
Total	FORTHOC			
	FORTHCEC			
	FORTHPHV			
	FORTHTN			
	FORTHTP			
	FORTHAP			
	FORTHMBC			
Corrected Total	FORTHOC			
	FORTHCEC			
	FORTHPHV			
	FORTHTN			
	FORTHTP			
	FORTHAP			
	FORTHMBC			

a. R Squared = .802 (Adjusted R Squared = .707)

b. R Squared = .790 (Adjusted R Squared = .690)

c. R Squared = .913 (Adjusted R Squared = .871)

d. R Squared = .473 (Adjusted R Squared = .220)

e. R Squared = .497 (Adjusted R Squared = .256)

f. R Squared = .776 (Adjusted R Squared = .669)

g. R Squared = .795 (Adjusted R Squared = .696)

Estimated Marginal Means

1. Grand Mean

Dependent Variable	Mean	Std. Error	95% Confidence Interval	
			Lower Bound	Upper Bound
FORTHOC	2.638	.005	2.629	2.647
FORTHCEC	1.338	.000	1.337	1.339
FORTHPHV	1.420	.001	1.418	1.422
FORTHTN	.720	.005	.710	.731
FORTHTP	1.368	.006	1.356	1.380
FORTHAP	.786	.003	.779	.793
FORTHMBC	4.026	.009	4.008	4.044

2. MULCH

Dependent Variable	MULCH	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHOC	1.00	2.579	.009	2.561	2.597
	2.00	2.698	.009	2.679	2.716
	3.00	2.619	.009	2.600	2.637
	4.00	2.657	.009	2.639	2.676
FORTHCEC	1.00	1.333	.001	1.331	1.335
	2.00	1.344	.001	1.342	1.346
	3.00	1.336	.001	1.334	1.338
	4.00	1.340	.001	1.338	1.342
FORTHPHV	1.00	1.419	.002	1.414	1.423
	2.00	1.444	.002	1.440	1.449
	3.00	1.406	.002	1.401	1.410
	4.00	1.411	.002	1.406	1.416
FORTHTN	1.00	.732	.010	.711	.752
	2.00	.745	.010	.725	.766
	3.00	.702	.010	.681	.722
	4.00	.702	.010	.682	.723
FORTHTP	1.00	1.348	.012	1.325	1.372
	2.00	1.354	.012	1.331	1.378
	3.00	1.358	.012	1.335	1.382
	4.00	1.410	.012	1.387	1.434
FORTHAP	1.00	.785	.007	.771	.798
	2.00	.833	.007	.820	.847
	3.00	.768	.007	.755	.782
	4.00	.759	.007	.745	.772
FORTHMBC	1.00	3.913	.018	3.877	3.950
	2.00	4.091	.018	4.054	4.127
	3.00	4.013	.018	3.976	4.049
	4.00	4.087	.018	4.051	4.124

3. PH

Dependent Variable	PH	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHOC	1.00	2.615	.008	2.599	2.631
	2.00	2.645	.008	2.629	2.661
	3.00	2.655	.008	2.639	2.670
FORTHCEC	1.00	1.336	.001	1.335	1.338
	2.00	1.339	.001	1.337	1.340
	3.00	1.340	.001	1.338	1.341
FORTHPHV	1.00	1.406	.002	1.401	1.410
	2.00	1.448	.002	1.444	1.452
	3.00	1.406	.002	1.402	1.410
FORTHTN	1.00	.718	.009	.700	.736
	2.00	.732	.009	.714	.749
	3.00	.712	.009	.694	.729
FORTHTP	1.00	1.377	.010	1.357	1.397
	2.00	1.353	.010	1.333	1.374
	3.00	1.373	.010	1.353	1.394
FORTHAP	1.00	.797	.006	.785	.808
	2.00	.775	.006	.763	.786
	3.00	.787	.006	.776	.799
FORTHMBC	1.00	3.996	.016	3.965	4.028
	2.00	4.104	.016	4.073	4.136
	3.00	3.978	.016	3.946	4.009

4. INOCU

Dependent Variable	INOCU	Mean	Std. Error	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHOC	1.00	2.638	.006	2.625	2.651
	2.00	2.638	.006	2.625	2.651
FORTHCEC	1.00	1.338	.001	1.337	1.340
	2.00	1.338	.001	1.337	1.339
FORTHPHV	1.00	1.418	.002	1.414	1.421
	2.00	1.422	.002	1.419	1.425
FORTHTN	1.00	.725	.007	.710	.739
	2.00	.716	.007	.702	.731
FORTHTP	1.00	1.372	.008	1.355	1.388
	2.00	1.364	.008	1.347	1.381
FORTHAP	1.00	.787	.005	.777	.796
	2.00	.786	.005	.776	.795
FORTHMBC	1.00	4.029	.013	4.003	4.054
	2.00	4.024	.013	3.998	4.049

5. MULCH * PH

Dependent Variable	MULCH	PH	Mean	Std. Error
FORTHOC	1.00	1.00	2.478	.016
		2.00	2.604	.016
		3.00	2.655	.016
	2.00	1.00	2.697	.016
		2.00	2.698	.016
		3.00	2.698	.016
	3.00	1.00	2.613	.016
		2.00	2.630	.016
		3.00	2.613	.016
	4.00	1.00	2.672	.016
		2.00	2.648	.016
		3.00	2.652	.016
FORTHCEC	1.00	1.00	1.324	.002
		2.00	1.335	.002
		3.00	1.340	.002
	2.00	1.00	1.344	.002
		2.00	1.344	.002
		3.00	1.344	.002
	3.00	1.00	1.336	.002
		2.00	1.337	.002
		3.00	1.336	.002
	4.00	1.00	1.341	.002
		2.00	1.339	.002
		3.00	1.339	.002
FORTHPHV	1.00	1.00	1.402	.004
		2.00	1.457	.004
		3.00	1.397	.004
	2.00	1.00	1.427	.004
		2.00	1.478	.004
		3.00	1.428	.004
	3.00	1.00	1.395	.004
		2.00	1.431	.004
		3.00	1.391	.004
	4.00	1.00	1.398	.004
		2.00	1.427	.004
		3.00	1.408	.004
FORTHTN	1.00	1.00	.692	.018
		2.00	.764	.018
		3.00	.740	.018
	2.00	1.00	.763	.018
		2.00	.774	.018
		3.00	.698	.018
	3.00	1.00	.701	.018
		2.00	.702	.018
		3.00	.702	.018
	4.00	1.00	.715	.018
		2.00	.686	.018
		3.00	.706	.018
FORTHTP	1.00	1.00	1.371	.020
		2.00	1.331	.020
		3.00	1.342	.020
	2.00	1.00	1.351	.020
		2.00	1.353	.020
		3.00	1.358	.020
	3.00	1.00	1.328	.020
		2.00	1.345	.020
		3.00	1.403	.020

5. MULCH * PH

Dependent Variable	MULCH	PH	Mean	Std. Error
FORTHTP	4.00	1.00	1.458	.020
		2.00	1.383	.020
		3.00	1.390	.020
FORTHAP	1.00	1.00	.798	.012
		2.00	.763	.012
		3.00	.793	.012
	2.00	1.00	.846	.012
		2.00	.833	.012
		3.00	.820	.012
	3.00	1.00	.792	.012
		2.00	.753	.012
		3.00	.759	.012
	4.00	1.00	.750	.012
		2.00	.750	.012
		3.00	.776	.012
FORTHMBC	1.00	1.00	3.791	.031
		2.00	3.961	.031
		3.00	3.988	.031
	2.00	1.00	4.022	.031
		2.00	4.162	.031
		3.00	4.088	.031
	3.00	1.00	3.989	.031
		2.00	4.120	.031
		3.00	3.929	.031
	4.00	1.00	4.183	.031
		2.00	4.173	.031
		3.00	3.906	.031

5. MULCH * PH

Dependent Variable	MULCH	PH	95% Confidence Interval	
			Lower Bound	Upper Bound
FORTHOC	1.00	1.00	2.446	2.510
		2.00	2.572	2.636
		3.00	2.623	2.687
	2.00	1.00	2.666	2.729
		2.00	2.666	2.730
		3.00	2.666	2.730
	3.00	1.00	2.581	2.645
		2.00	2.598	2.661
		3.00	2.581	2.645
	4.00	1.00	2.641	2.704
		2.00	2.616	2.679
		3.00	2.620	2.684
FORTHCEC	1.00	1.00	1.321	1.327
		2.00	1.332	1.338
		3.00	1.337	1.343
	2.00	1.00	1.341	1.347
		2.00	1.341	1.347
		3.00	1.341	1.347
	3.00	1.00	1.333	1.339
		2.00	1.334	1.340
		3.00	1.333	1.339
	4.00	1.00	1.338	1.344
		2.00	1.336	1.342
		3.00	1.336	1.342
FORTHPHV	1.00	1.00	1.394	1.411
		2.00	1.448	1.465
		3.00	1.389	1.405
	2.00	1.00	1.419	1.435
		2.00	1.470	1.486
		3.00	1.420	1.436
	3.00	1.00	1.386	1.403
		2.00	1.423	1.440
		3.00	1.382	1.399
	4.00	1.00	1.390	1.406
		2.00	1.419	1.435
		3.00	1.400	1.416
FORTHTN	1.00	1.00	.656	.727
		2.00	.728	.799
		3.00	.705	.776
	2.00	1.00	.728	.799
		2.00	.739	.810
		3.00	.663	.734
	3.00	1.00	.666	.737
		2.00	.667	.738
		3.00	.667	.738
	4.00	1.00	.680	.751
		2.00	.651	.722
		3.00	.670	.741
FORTHTP	1.00	1.00	1.330	1.412
		2.00	1.290	1.372
		3.00	1.302	1.383
	2.00	1.00	1.310	1.392
		2.00	1.312	1.394
		3.00	1.318	1.399
	3.00	1.00	1.287	1.368
		2.00	1.304	1.386
		3.00	1.362	1.444

5. MULCH * PH

Dependent Variable	MULCH	PH	95% Confidence Interval	
			Lower Bound	Upper Bound
FORTHTP	4.00	1.00	1.417	1.499
		2.00	1.342	1.424
		3.00	1.349	1.431
FORTHAP	1.00	1.00	.775	.822
		2.00	.739	.786
		3.00	.770	.817
	2.00	1.00	.823	.870
		2.00	.810	.857
		3.00	.797	.844
	3.00	1.00	.769	.816
		2.00	.730	.777
		3.00	.736	.783
	4.00	1.00	.726	.773
		2.00	.726	.773
		3.00	.753	.800
FORTHMBC	1.00	1.00	3.727	3.854
		2.00	3.898	4.024
		3.00	3.925	4.052
	2.00	1.00	3.959	4.085
		2.00	4.099	4.226
		3.00	4.024	4.151
	3.00	1.00	3.925	4.052
		2.00	4.057	4.184
		3.00	3.866	3.992
	4.00	1.00	4.120	4.247
		2.00	4.109	4.236
		3.00	3.842	3.969

6. MULCH * INOCU

Dependent Variable	MULCH	INOCU	Mean	Std. Error
FORTHOC	1.00	1.00	2.556	.013
		2.00	2.602	.013
	2.00	1.00	2.709	.013
		2.00	2.687	.013
	3.00	1.00	2.630	.013
		2.00	2.608	.013
	4.00	1.00	2.658	.013
		2.00	2.657	.013
FORTHCEC	1.00	1.00	1.331	.001
		2.00	1.335	.001
	2.00	1.00	1.345	.001
		2.00	1.343	.001
	3.00	1.00	1.337	.001
		2.00	1.335	.001
	4.00	1.00	1.340	.001
		2.00	1.340	.001
FORTHPHV	1.00	1.00	1.415	.003
		2.00	1.422	.003
	2.00	1.00	1.445	.003
		2.00	1.443	.003
	3.00	1.00	1.402	.003
		2.00	1.409	.003
	4.00	1.00	1.408	.003
		2.00	1.414	.003
FORTHTN	1.00	1.00	.736	.014
		2.00	.727	.014
	2.00	1.00	.751	.014
		2.00	.739	.014
	3.00	1.00	.701	.014
		2.00	.703	.014
	4.00	1.00	.710	.014
		2.00	.695	.014
FORTHTP	1.00	1.00	1.347	.017
		2.00	1.350	.017
	2.00	1.00	1.359	.017
		2.00	1.349	.017
	3.00	1.00	1.354	.017
		2.00	1.362	.017
	4.00	1.00	1.426	.017
		2.00	1.395	.017
FORTHAP	1.00	1.00	.767	.010
		2.00	.802	.010
	2.00	1.00	.841	.010
		2.00	.825	.010
	3.00	1.00	.782	.010
		2.00	.755	.010
	4.00	1.00	.757	.010
		2.00	.760	.010
FORTHMBC	1.00	1.00	3.924	.026
		2.00	3.903	.026
	2.00	1.00	4.067	.026
		2.00	4.115	.026
	3.00	1.00	4.019	.026
		2.00	4.006	.026
	4.00	1.00	4.104	.026
		2.00	4.071	.026

6. MULCH * INOCU

Dependent Variable	MULCH	INOCU	95% Confidence Interval	
			Lower Bound	Upper Bound
FORTHOC	1.00	1.00	2.530	2.582
		2.00	2.576	2.628
	2.00	1.00	2.683	2.735
		2.00	2.661	2.713
	3.00	1.00	2.604	2.656
		2.00	2.582	2.634
	4.00	1.00	2.632	2.684
		2.00	2.631	2.683
FORTHCEC	1.00	1.00	1.328	1.333
		2.00	1.332	1.337
	2.00	1.00	1.343	1.347
		2.00	1.340	1.345
	3.00	1.00	1.335	1.340
		2.00	1.333	1.338
	4.00	1.00	1.337	1.342
		2.00	1.337	1.342
FORTHPHV	1.00	1.00	1.408	1.422
		2.00	1.416	1.429
	2.00	1.00	1.439	1.452
		2.00	1.436	1.450
	3.00	1.00	1.395	1.409
		2.00	1.402	1.416
	4.00	1.00	1.401	1.415
		2.00	1.407	1.421
FORTHTN	1.00	1.00	.707	.765
		2.00	.698	.756
	2.00	1.00	.722	.780
		2.00	.710	.768
	3.00	1.00	.672	.730
		2.00	.674	.732
	4.00	1.00	.681	.739
		2.00	.666	.724
FORTHTP	1.00	1.00	1.313	1.380
		2.00	1.317	1.383
	2.00	1.00	1.326	1.392
		2.00	1.316	1.383
	3.00	1.00	1.321	1.388
		2.00	1.329	1.396
	4.00	1.00	1.393	1.459
		2.00	1.362	1.428
FORTHAP	1.00	1.00	.748	.786
		2.00	.783	.822
	2.00	1.00	.822	.861
		2.00	.806	.844
	3.00	1.00	.763	.801
		2.00	.736	.774
	4.00	1.00	.738	.777
		2.00	.741	.779
FORTHMBC	1.00	1.00	3.873	3.976
		2.00	3.851	3.954
	2.00	1.00	4.015	4.118
		2.00	4.063	4.166
	3.00	1.00	3.968	4.071
		2.00	3.954	4.058
	4.00	1.00	4.052	4.155
		2.00	4.019	4.122

7. PH * INOCU

Dependent Variable	PH	INOCU	Mean	Std. Error
FORTHOC	1.00	1.00	2.595	.011
		2.00	2.636	.011
	2.00	1.00	2.648	.011
		2.00	2.641	.011
	3.00	1.00	2.671	.011
		2.00	2.638	.011
FORTHCEC	1.00	1.00	1.334	.001
		2.00	1.338	.001
	2.00	1.00	1.339	.001
		2.00	1.338	.001
	3.00	1.00	1.341	.001
		2.00	1.338	.001
FORTHPHV	1.00	1.00	1.405	.003
		2.00	1.406	.003
	2.00	1.00	1.443	.003
		2.00	1.453	.003
	3.00	1.00	1.405	.003
		2.00	1.407	.003
FORTHTN	1.00	1.00	.731	.012
		2.00	.704	.012
	2.00	1.00	.735	.012
		2.00	.728	.012
	3.00	1.00	.707	.012
		2.00	.716	.012
FORTHTP	1.00	1.00	1.397	.014
		2.00	1.357	.014
	2.00	1.00	1.348	.014
		2.00	1.358	.014
	3.00	1.00	1.370	.014
		2.00	1.377	.014
FORTHAP	1.00	1.00	.817	.008
		2.00	.777	.008
	2.00	1.00	.774	.008
		2.00	.775	.008
	3.00	1.00	.770	.008
		2.00	.805	.008
FORTHMBC	1.00	1.00	4.021	.022
		2.00	3.971	.022
	2.00	1.00	4.129	.022
		2.00	4.079	.022
	3.00	1.00	3.935	.022
		2.00	4.020	.022

7. PH * INOCU

Dependent Variable	PH	INOCU	95% Confidence Interval	
			Lower Bound	Upper Bound
FORTHOC	1.00	1.00	2.572	2.617
		2.00	2.613	2.658
	2.00	1.00	2.626	2.671
		2.00	2.619	2.664
	3.00	1.00	2.649	2.694
		2.00	2.616	2.661
FORTHCEC	1.00	1.00	1.332	1.337
		2.00	1.336	1.340
	2.00	1.00	1.337	1.341
		2.00	1.336	1.340
	3.00	1.00	1.339	1.343
		2.00	1.336	1.340
FORTHPHV	1.00	1.00	1.399	1.410
		2.00	1.401	1.412
	2.00	1.00	1.438	1.449
		2.00	1.448	1.459
	3.00	1.00	1.399	1.411
		2.00	1.401	1.412
FORTHTN	1.00	1.00	.706	.756
		2.00	.679	.729
	2.00	1.00	.710	.760
		2.00	.703	.753
	3.00	1.00	.682	.733
		2.00	.691	.741
FORTHTP	1.00	1.00	1.368	1.425
		2.00	1.328	1.386
	2.00	1.00	1.319	1.377
		2.00	1.329	1.387
	3.00	1.00	1.341	1.399
		2.00	1.348	1.406
FORTHAP	1.00	1.00	.800	.833
		2.00	.760	.793
	2.00	1.00	.757	.791
		2.00	.759	.792
	3.00	1.00	.753	.786
		2.00	.788	.821
FORTHMBC	1.00	1.00	3.977	4.066
		2.00	3.926	4.016
	2.00	1.00	4.084	4.174
		2.00	4.035	4.124
	3.00	1.00	3.890	3.980
		2.00	3.976	4.065

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	Mean	Std. Error
FORTHOC	1.00	1.00	1.00	2.418	.022
			2.00	2.537	.022
		2.00	1.00	2.586	.022
			2.00	2.623	.022
		3.00	1.00	2.665	.022
			2.00	2.646	.022
	2.00	1.00	1.00	2.664	.022
			2.00	2.731	.022
		2.00	1.00	2.737	.022
			2.00	2.659	.022
		3.00	1.00	2.725	.022
			2.00	2.671	.022
	3.00	1.00	1.00	2.618	.022
			2.00	2.608	.022
		2.00	1.00	2.634	.022
			2.00	2.625	.022
		3.00	1.00	2.636	.022
			2.00	2.590	.022
	4.00	1.00	1.00	2.679	.022
			2.00	2.666	.022
		2.00	1.00	2.637	.022
			2.00	2.659	.022
		3.00	1.00	2.658	.022
			2.00	2.646	.022
FORTHCEC	1.00	1.00	1.00	1.319	.002
			2.00	1.329	.002
		2.00	1.00	1.333	.002
			2.00	1.337	.002
		3.00	1.00	1.341	.002
			2.00	1.339	.002
	2.00	1.00	1.00	1.340	.002
			2.00	1.347	.002
		2.00	1.00	1.348	.002
			2.00	1.340	.002
		3.00	1.00	1.347	.002
			2.00	1.341	.002
	3.00	1.00	1.00	1.336	.002
			2.00	1.335	.002
		2.00	1.00	1.338	.002
			2.00	1.337	.002
		3.00	1.00	1.338	.002
			2.00	1.333	.002
	4.00	1.00	1.00	1.342	.002
			2.00	1.341	.002
		2.00	1.00	1.338	.002
			2.00	1.340	.002
		3.00	1.00	1.340	.002
			2.00	1.339	.002
FORTHPHV	1.00	1.00	1.00	1.395	.006
			2.00	1.410	.006
		2.00	1.00	1.456	.006
			2.00	1.457	.006
		3.00	1.00	1.394	.006
			2.00	1.400	.006
	2.00	1.00	1.00	1.433	.006
			2.00	1.421	.006
		2.00	1.00	1.468	.006
			2.00	1.488	.006

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	Mean	Std. Error
FORTHPHV	2.00	3.00	1.00	1.435	.006
			2.00	1.420	.006
	3.00	1.00	1.00	1.394	.006
			2.00	1.395	.006
		2.00	1.00	1.428	.006
			2.00	1.435	.006
		3.00	1.00	1.385	.006
			2.00	1.396	.006
	4.00	1.00	1.00	1.397	.006
			2.00	1.399	.006
		2.00	1.00	1.421	.006
			2.00	1.433	.006
		3.00	1.00	1.406	.006
			2.00	1.410	.006
FORTHTN	1.00	1.00	1.00	.723	.025
			2.00	.660	.025
		2.00	1.00	.759	.025
			2.00	.768	.025
		3.00	1.00	.727	.025
			2.00	.754	.025
	2.00	1.00	1.00	.756	.025
			2.00	.770	.025
		2.00	1.00	.791	.025
			2.00	.757	.025
		3.00	1.00	.706	.025
			2.00	.690	.025
	3.00	1.00	1.00	.701	.025
			2.00	.701	.025
		2.00	1.00	.710	.025
			2.00	.695	.025
		3.00	1.00	.692	.025
			2.00	.712	.025
	4.00	1.00	1.00	.745	.025
			2.00	.685	.025
		2.00	1.00	.680	.025
			2.00	.693	.025
		3.00	1.00	.704	.025
			2.00	.707	.025
FORTHTP	1.00	1.00	1.00	1.397	.029
			2.00	1.345	.029
		2.00	1.00	1.330	.029
			2.00	1.332	.029
		3.00	1.00	1.313	.029
			2.00	1.372	.029
	2.00	1.00	1.00	1.360	.029
			2.00	1.342	.029
		2.00	1.00	1.360	.029
			2.00	1.346	.029
		3.00	1.00	1.357	.029
			2.00	1.360	.029
	3.00	1.00	1.00	1.322	.029
			2.00	1.333	.029
		2.00	1.00	1.324	.029
			2.00	1.366	.029
		3.00	1.00	1.417	.029
			2.00	1.388	.029
	4.00	1.00	1.00	1.507	.029
			2.00	1.409	.029

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	Mean	Std. Error
FORTHTP	4.00	2.00	1.00	1.379	.029
			2.00	1.387	.029
		3.00	1.00	1.392	.029
			2.00	1.389	.029
FORTHAP	1.00	1.00	1.00	.834	.017
			2.00	.762	.017
		2.00	1.00	.742	.017
			2.00	.783	.017
		3.00	1.00	.724	.017
			2.00	.862	.017
	2.00	1.00	1.00	.856	.017
			2.00	.836	.017
		2.00	1.00	.825	.017
			2.00	.841	.017
		3.00	1.00	.842	.017
			2.00	.798	.017
	3.00	1.00	1.00	.810	.017
			2.00	.775	.017
		2.00	1.00	.797	.017
			2.00	.709	.017
		3.00	1.00	.738	.017
			2.00	.781	.017
	4.00	1.00	1.00	.765	.017
			2.00	.734	.017
		2.00	1.00	.732	.017
			2.00	.768	.017
		3.00	1.00	.775	.017
			2.00	.778	.017
FORTHMBC	1.00	1.00	1.00	3.848	.044
			2.00	3.734	.044
		2.00	1.00	3.996	.044
			2.00	3.927	.044
		3.00	1.00	3.929	.044
			2.00	4.048	.044
	2.00	1.00	1.00	4.018	.044
			2.00	4.026	.044
		2.00	1.00	4.179	.044
			2.00	4.146	.044
		3.00	1.00	4.003	.044
			2.00	4.172	.044
	3.00	1.00	1.00	3.992	.044
			2.00	3.986	.044
		2.00	1.00	4.133	.044
			2.00	4.108	.044
		3.00	1.00	3.934	.044
			2.00	3.925	.044
	4.00	1.00	1.00	4.228	.044
			2.00	4.138	.044
		2.00	1.00	4.209	.044
			2.00	4.137	.044
		3.00	1.00	3.874	.044
			2.00	3.937	.044

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHOC	1.00	1.00	1.00	2.373	2.463
			2.00	2.492	2.582
		2.00	1.00	2.541	2.631
			2.00	2.578	2.668
		3.00	1.00	2.620	2.710
			2.00	2.601	2.691
	2.00	1.00	1.00	2.619	2.709
			2.00	2.686	2.776
		2.00	1.00	2.692	2.782
			2.00	2.614	2.704
		3.00	1.00	2.680	2.770
			2.00	2.626	2.716
	3.00	1.00	1.00	2.573	2.663
			2.00	2.563	2.653
		2.00	1.00	2.589	2.679
			2.00	2.580	2.670
		3.00	1.00	2.591	2.681
			2.00	2.545	2.635
	4.00	1.00	1.00	2.634	2.724
			2.00	2.621	2.711
		2.00	1.00	2.592	2.682
			2.00	2.614	2.704
		3.00	1.00	2.613	2.703
			2.00	2.601	2.691
FORTHCEC	1.00	1.00	1.00	1.315	1.323
			2.00	1.325	1.333
		2.00	1.00	1.329	1.337
			2.00	1.332	1.341
		3.00	1.00	1.336	1.345
			2.00	1.334	1.343
	2.00	1.00	1.00	1.336	1.345
			2.00	1.343	1.352
		2.00	1.00	1.344	1.352
			2.00	1.336	1.344
		3.00	1.00	1.342	1.351
			2.00	1.337	1.345
	3.00	1.00	1.00	1.332	1.341
			2.00	1.331	1.339
		2.00	1.00	1.333	1.342
			2.00	1.332	1.341
		3.00	1.00	1.334	1.342
			2.00	1.329	1.338
	4.00	1.00	1.00	1.338	1.346
			2.00	1.336	1.345
		2.00	1.00	1.334	1.342
			2.00	1.336	1.344
		3.00	1.00	1.336	1.344
			2.00	1.334	1.343
FORTHPHV	1.00	1.00	1.00	1.383	1.406
			2.00	1.398	1.422
		2.00	1.00	1.445	1.468
			2.00	1.445	1.468
		3.00	1.00	1.382	1.406
			2.00	1.389	1.412
	2.00	1.00	1.00	1.421	1.445
			2.00	1.409	1.432
		2.00	1.00	1.456	1.479
			2.00	1.477	1.500

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHPHV	2.00	3.00	1.00	1.424	1.447
			2.00	1.409	1.432
	3.00	1.00	1.00	1.382	1.406
			2.00	1.384	1.407
		2.00	1.00	1.416	1.439
			2.00	1.424	1.447
		3.00	1.00	1.373	1.396
			2.00	1.385	1.408
	4.00	1.00	1.00	1.386	1.409
			2.00	1.388	1.411
		2.00	1.00	1.410	1.433
			2.00	1.421	1.445
		3.00	1.00	1.394	1.417
			2.00	1.398	1.422
FORTHTN	1.00	1.00	1.00	.673	.773
			2.00	.610	.711
		2.00	1.00	.709	.810
			2.00	.718	.818
		3.00	1.00	.676	.777
			2.00	.704	.804
	2.00	1.00	1.00	.706	.806
			2.00	.720	.820
		2.00	1.00	.741	.841
			2.00	.707	.807
		3.00	1.00	.656	.757
			2.00	.640	.740
	3.00	1.00	1.00	.651	.751
			2.00	.651	.752
		2.00	1.00	.660	.760
			2.00	.644	.745
		3.00	1.00	.642	.742
			2.00	.662	.763
	4.00	1.00	1.00	.695	.796
			2.00	.635	.736
		2.00	1.00	.630	.730
			2.00	.642	.743
		3.00	1.00	.654	.755
			2.00	.657	.757
FORTHTP	1.00	1.00	1.00	1.339	1.455
			2.00	1.287	1.403
		2.00	1.00	1.272	1.388
			2.00	1.275	1.390
		3.00	1.00	1.255	1.370
			2.00	1.315	1.430
	2.00	1.00	1.00	1.303	1.418
			2.00	1.284	1.400
		2.00	1.00	1.302	1.418
			2.00	1.288	1.404
		3.00	1.00	1.299	1.415
			2.00	1.302	1.418
	3.00	1.00	1.00	1.264	1.380
			2.00	1.275	1.391
		2.00	1.00	1.266	1.382
			2.00	1.308	1.424
		3.00	1.00	1.359	1.475
			2.00	1.331	1.446
	4.00	1.00	1.00	1.449	1.565
			2.00	1.352	1.467

8. MULCH * PH * INOCU

Dependent Variable	MULCH	PH	INOCU	95% Confidence Interval	
				Lower Bound	Upper Bound
FORTHTP	4.00	2.00	1.00	1.321	1.437
			2.00	1.329	1.445
		3.00	1.00	1.334	1.450
			2.00	1.331	1.447
FORTHAP	1.00	1.00	1.00	.801	.868
			2.00	.728	.795
		2.00	1.00	.709	.775
			2.00	.750	.816
		3.00	1.00	.691	.757
			2.00	.829	.896
	2.00	1.00	1.00	.823	.890
			2.00	.803	.869
		2.00	1.00	.792	.859
			2.00	.808	.874
		3.00	1.00	.809	.876
			2.00	.765	.832
	3.00	1.00	1.00	.777	.843
			2.00	.741	.808
		2.00	1.00	.764	.830
			2.00	.676	.742
		3.00	1.00	.705	.771
			2.00	.747	.814
	4.00	1.00	1.00	.732	.799
			2.00	.701	.767
		2.00	1.00	.698	.765
			2.00	.735	.801
		3.00	1.00	.742	.808
			2.00	.744	.811
FORTHMBC	1.00	1.00	1.00	3.758	3.937
			2.00	3.644	3.823
		2.00	1.00	3.906	4.085
			2.00	3.838	4.016
		3.00	1.00	3.840	4.019
			2.00	3.958	4.137
	2.00	1.00	1.00	3.929	4.107
			2.00	3.936	4.115
		2.00	1.00	4.090	4.268
			2.00	4.057	4.235
		3.00	1.00	3.914	4.092
			2.00	4.083	4.262
	3.00	1.00	1.00	3.902	4.081
			2.00	3.896	4.075
		2.00	1.00	4.044	4.222
			2.00	4.018	4.197
		3.00	1.00	3.844	4.023
			2.00	3.835	4.014
	4.00	1.00	1.00	4.139	4.318
			2.00	4.049	4.228
		2.00	1.00	4.119	4.298
			2.00	4.047	4.226
		3.00	1.00	3.785	3.964
			2.00	3.848	4.026

Appendix 7

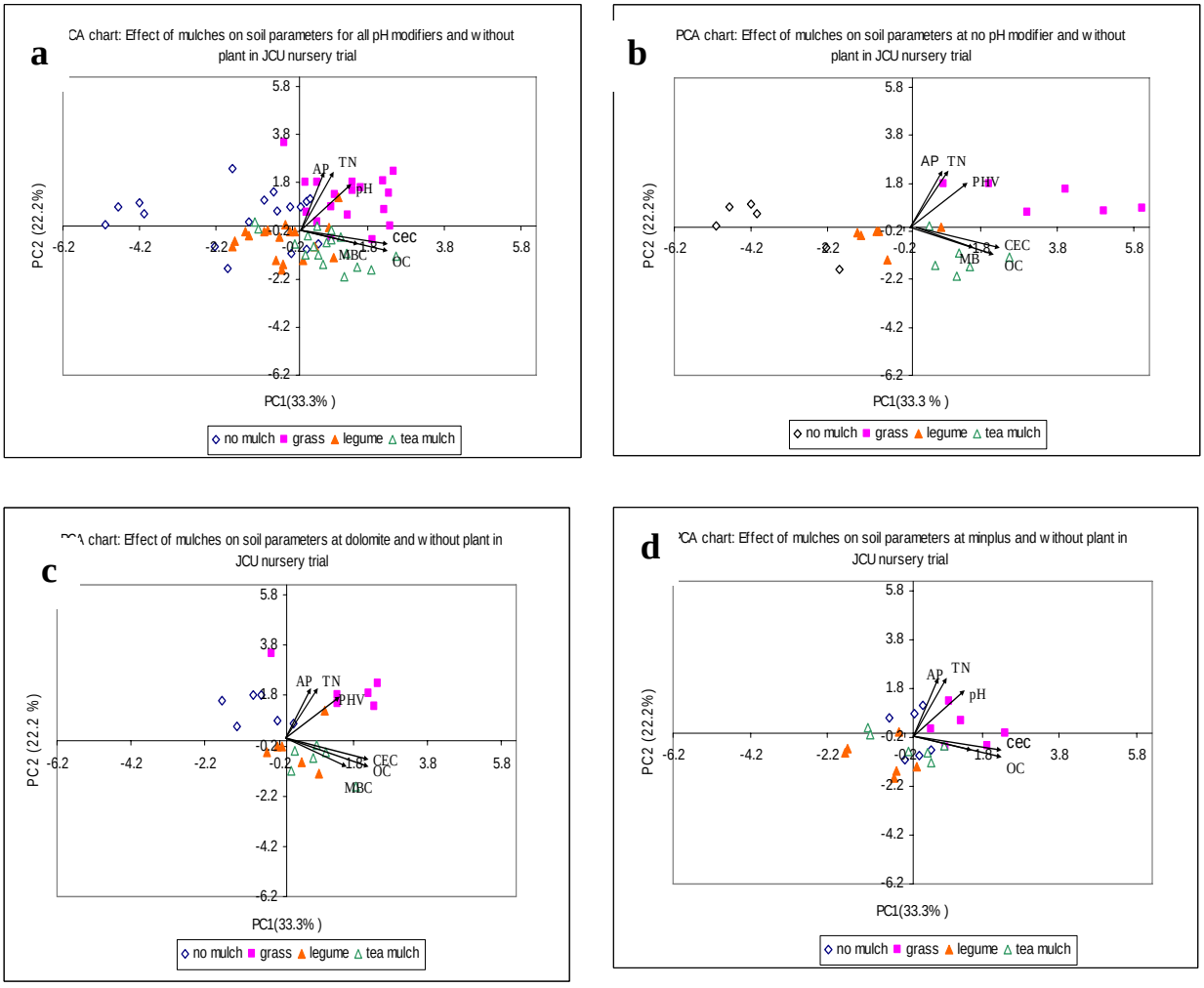


Figure Effects of mulches on soil parameters in the James Cook University Nursery Trial without plants:

(a) The analysed data include all soil pH modifier treatments;
(b) The analysed data excludes all soil pH modifier treatments;
(c) The analysed data embraces dolomite as the soil pH modifier treatment,
(d) The analysed data embraces Minplus as the soil pH modifier treatment.

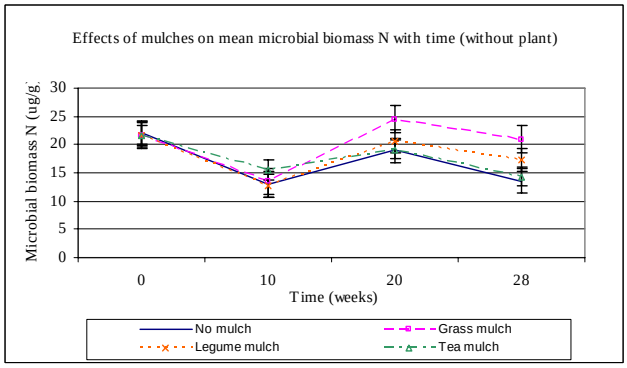
Appendix 8

Effect of inoculum on soil properties in nursery trial at JCU – without plants at JCU. Each cell represents the mean values of the fourth root transformed data; back-transformed data are shown in parentheses. Significant ($p = 0.05$) differences from the control treatment are shown by '*'. Means followed by same lower-case letter within a column are not significantly different ($p < 0.05$)

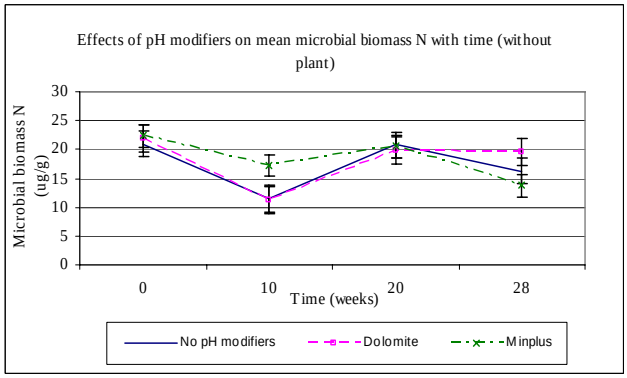
Treatments	Soil parameters (sqrt x sqrt transformation data)				
	Organic carbon (mg/g)	CEC (meq/100g soil)	Soil pH	Soil microbial Biomass (µg/g)	Plant available phosphorus (mg/g)
Without inoculum	2.63 ns	1.33 ns	1.418 ns	4.029 ns	0.787 ns
With inoculum	2.63 ns	1.33 ns	1.422 ns	4.024 ns	0.786 ns

Appendix 9

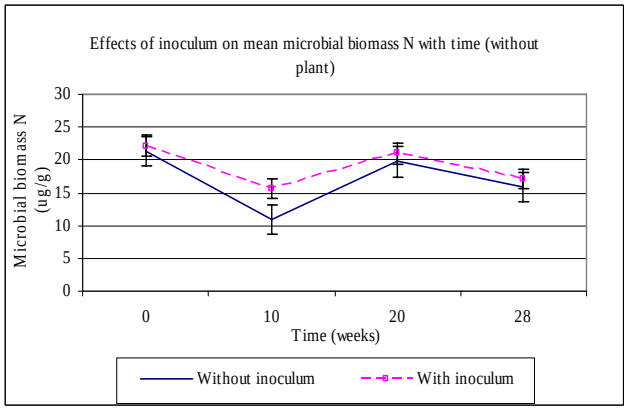
Changes in soil microbial biomass nitrogen over the period of the James Cook University Nursery Trial (without plants):



(a) Effect of mulches on microbial biomass nitrogen,



(b) Effect of soil pH modifiers on microbial biomass nitrogen,



(c) Effect of rainforest soil inoculum on microbial biomass nitrogen.

Component Matrix[a]

	Component				
	1	2	3	4	5
A	-7.652E-03	-.724	-.390	-.211	-2.327E-02
B	4.792E-02	-.340	.639	.251	.112
C	.534	4.662E-02	-.128	.551	-.110
D	-.331	6.878E-02	.202	.361	-.392
E	-.156	-.252	-.242	-.661	.248
F	-3.664E-02	-.621	-.541	-.353	-9.672E-02
G	.758	.133	-.163	.447	.101
H	.148	-.349	-.110	4.762E-02	-.163
I	.550	.209	-.159	.367	9.438E-02
J	.701	.238	-1.438E-02	-4.534E-02	.265
K	-.380	.333	.298	-.221	.506
L	5.019E-03	9.764E-02	-.559	-.269	.565
M	7.452E-02	.205	8.859E-02	.386	.736
N	-.113	.801	.163	-.375	-.272
O	-2.450E-02	.733	.106	-.298	-.298
P	6.937E-02	.111	9.526E-02	-.510	-6.620E-02
Q	-1.658E-02	.140	-.440	.268	-.207
R	.137	-.367	-.515	-1.857E-02	-.425
S	.281	.444	8.399E-02	7.170E-02	-.221
T	-1.748E-02	3.082E-03	.172	-.345	.245
U	.153	-.447	5.358E-03	-.185	-.519
V	-.428	.572	6.757E-02	-.418	-.271
W	-.157	.658	-.249	.409	-4.296E-02
X	.400	-.655	-.132	.129	.211
Y	.416	.377	.226	-.109	-2.666E-02
Z	.336	-.468	.596	-.235	.126
A1	.367	.409	-.122	-.371	.232
B1	-.241	-5.052E-03	.330	.196	.587
C1	.767	-5.170E-02	-6.578E-02	-8.240E-02	4.784E-02
D1	-9.388E-02	.186	2.810E-02	.332	-5.165E-02
E1	.632	.355	-.180	.149	6.788E-02
F1	.178	.202	-.163	6.788E-02	-8.268E-02
G1	.263	.101	.342	.593	.133
H1	-.877	.207	.132	8.780E-02	-4.830E-02
I1	-.203	-4.499E-02	-.522	4.896E-02	.232
J1	8.267E-03	.314	.569	-7.164E-02	-3.763E-02
K1	.220	-.519	.654	-.133	5.279E-02
L1	-.133	.409	-.190	.515	-6.781E-02
M1	-.165	4.039E-02	8.158E-02	.160	-.356
N1	.733	4.518E-02	.296	-.265	-.448
O1	.810	.188	.254	-9.677E-02	-.247
P1	.171	-.418	.616	-5.251E-02	.176
Q1	.423	-.183	-.286	-5.054E-03	.251
R1	-.458	5.513E-02	-2.398E-02	.114	.234
S1	-.607	-.340	.178	.502	-.115
T1	.299	.545	-.111	-.326	8.259E-02
U1	5.115E-03	.297	-.380	2.313E-02	.161
V1	-3.661E-02	-.424	-1.572E-02	.388	-.336

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component				
	6	7	8	9	10
A	.245	-.198	-.124	.109	6.822E-02
B	-.285	-3.162E-02	.261	9.645E-02	-.185
C	-.162	-.301	-.111	.426	.122
D	-1.509E-02	.209	3.166E-02	-1.369E-02	-.539
E	.182	.214	-.105	.289	-9.432E-02
F	.244	7.178E-02	6.041E-02	.171	2.340E-02
G	.285	-8.975E-02	5.740E-02	-4.039E-04	-3.343E-02
H	-.464	-.507	-.321	3.088E-02	-.123
I	.459	-.113	3.359E-02	-.297	-7.043E-02
J	.305	.130	9.989E-02	-2.109E-02	2.307E-02
K	.190	-.154	.124	-.203	-7.687E-02
L	-.193	-2.050E-02	.246	-.113	-.217
M	.127	.124	-5.204E-02	-7.812E-02	.144
N	1.510E-02	-.124	-.121	.143	7.581E-02
O	.146	5.380E-02	-7.966E-02	8.830E-02	.183
P	.182	-.296	.187	-.387	-2.704E-02
Q	8.461E-02	.297	-9.969E-02	.451	-.312
R	1.665E-02	.284	.166	-.334	.206
S	-.591	.331	.192	-2.972E-02	.103
T	-.143	-.411	.651	3.844E-02	8.027E-02
U	.164	.350	-.314	-.158	.157
V	-8.843E-02	-.352	-3.674E-03	1.314E-02	.162
W	.112	6.881E-02	.212	-.159	-9.697E-03
X	-.164	-.309	-3.496E-02	-.216	-.106
Y	-.287	7.160E-02	-.227	7.508E-02	-.112
Z	-2.231E-02	.416	-8.778E-02	9.505E-02	-9.872E-03
A1	-.129	.169	.116	.520	-2.812E-02
B1	.382	7.678E-02	-.133	.372	5.307E-02
C1	.340	-.280	-.157	.119	8.335E-02
D1	.391	5.331E-02	-.115	2.535E-02	.263
E1	-.253	-.146	4.875E-02	.315	-.117
F1	-.440	.344	9.314E-03	-8.446E-02	.395
G1	-.238	2.115E-02	-1.922E-02	-7.459E-02	.367
H1	8.294E-02	2.392E-02	-.198	9.395E-02	-1.198E-02
I1	-.397	.235	.296	6.741E-02	.289
J1	-1.536E-02	-.174	-.447	-.139	-.222
K1	7.373E-02	.273	.116	.110	3.113E-02
L1	.369	.184	4.388E-02	-.228	-.170
M1	.481	2.217E-02	.338	.301	.253
N1	1.609E-02	-.122	5.867E-02	-6.796E-02	6.652E-02
O1	.155	-.138	.175	-1.082E-02	1.159E-02
P1	-6.228E-02	.392	9.068E-04	-.148	7.396E-03
Q1	-.108	.106	-.452	-.143	9.897E-02
R1	-.116	-3.320E-02	-.193	-.222	.275
S1	-.136	-.208	4.843E-02	.209	.118
T1	-2.786E-02	.452	9.160E-02	-.132	-.317
U1	-.251	-9.660E-03	-.405	-.105	-.220
V1	-4.682E-02	-6.347E-03	.412	-2.076E-02	-.383

Extraction Method: Principal Component Analysis.

Component Matrix[a]

	Component			
	11	12	13	14
A	.258	-.111	-.159	-1.126E-02
B	-.129	-6.973E-02	-7.533E-02	.249
C	-.167	-5.149E-02	2.377E-02	-1.777E-02
D	-1.254E-02	-.194	.138	-5.501E-02
E	.200	-3.571E-03	-.165	-8.195E-02
F	-1.056E-02	-.175	.108	-5.522E-02
G	-3.680E-02	7.759E-02	9.280E-02	-9.399E-03
H	-9.222E-03	6.462E-02	-.191	9.889E-02
I	.152	.228	4.382E-02	-.178
J	-.394	-.219	.120	.149
K	7.302E-02	.195	-.355	-.115
L	-1.986E-02	2.818E-02	-8.236E-02	.131
M	-5.832E-02	.174	-5.462E-02	-.117
N	-.104	.128	6.500E-02	-3.399E-03
O	5.070E-02	.319	.146	-3.380E-04
P	-.424	-.308	.169	.136
Q	7.448E-02	.137	.138	-.122
R	-.135	-4.956E-03	-.183	-.110
S	-.101	-6.256E-02	-6.222E-02	-.192
T	.138	7.710E-03	.171	-.205
U	1.709E-02	.211	3.395E-02	5.598E-02
V	.144	6.185E-02	4.294E-02	-1.014E-02
W	.246	.149	-.229	2.377E-02
X	.201	-7.460E-02	-1.864E-02	.151
Y	-.212	.135	-.244	.437
Z	-3.951E-03	6.819E-02	-4.109E-03	5.782E-02
A1	-.126	-.114	.143	7.584E-02
B1	-.120	-9.834E-02	.151	-.139
C1	8.317E-02	-8.298E-02	-2.950E-03	5.215E-02
D1	-.371	-.154	-.410	4.070E-02
E1	.346	9.311E-02	-.156	4.016E-02
F1	.200	-.473	1.499E-02	-.150
G1	.302	-.248	1.629E-02	-.106
H1	3.180E-02	-.105	-.113	-9.091E-03
I1	-6.257E-02	.254	2.903E-03	.345
J1	.326	-.225	5.414E-02	.103
K1	.158	2.518E-02	5.741E-02	9.038E-03
L1	.166	-.199	7.511E-02	.340
M1	.114	3.769E-02	-.117	.316
N1	1.183E-02	.148	-6.336E-02	-3.752E-02
O1	-2.515E-02	6.190E-02	7.123E-02	-.211
P1	-7.102E-02	.276	-.128	-.103
Q1	.105	.204	.235	7.176E-02
R1	3.624E-02	.194	.576	.214
S1	-9.597E-02	7.087E-02	6.140E-02	-.102
T1	.313	-.153	2.141E-02	2.664E-02
U1	-.460	3.119E-02	-4.159E-02	-.329
V1	-.126	.259	.176	-3.747E-02

Extraction Method: Principal Component Analysis.

a. 14 components extracted.

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Appendix 11

Determination of soil respiration (Black, C.A , 1968)

One of the simplest methods for estimating the rate of carbon dioxide (CO2) evolution from undisturbed soil was used (Black 1968). Alkali of a defined concentration (NaOH) was placed in an open jar above the soil surface and the area to be measured was covered with a metal cylinder that was closed at the upper end. As CO_2 evolves from the soil surface it becomes trapped in the cylinder and absorbed by the alkali. After a given period of time (24 hrs), the alkali was removed and the un-reacted portion was determined by titration using 1 N HCl. By means of subtraction, the amount of CO_2 that combined with alkali was determined.

Special apparatus

1. Metal cylinder with one sealed (airtight) end: the open end should be at least 25 cm in diameter and the cylinder should be at least 30 cm high.
2. Screw-capped glass jars: The openings should be at least 6.5cm in diameter, and the jar should be at least 7cm high.
3. Tripods made from heavy gauged wire or plastic: these should be constructed to hold the base of the jars about 2 cm above the surface soil

Reagents

1. Sodium hydroxide (NaOH) solution,1.0N
2. Barium chloride ($BaCl_2$) , 3.0N
3. Hydrochloric acid (HCl), 1.0N
4. Phenopthline indicator: Dissolve 1g of Phenopthline in 100mL of 95% ethanol.

Procedure

Before keeping the cylinder, mulches should be removed. After selecting an appropriate sight, prepare a $CO_{2\ trap}$ by pippeting 20 mL of 1.0 NaOH solution into a glass jar and place this, plus a tripod on the soil surface. Immediately place he metal cover over the trap and press the edge about 2 cm into the surface soil. The cylinder should cover from direct sun light by either covering with an appropriately sized sheet of wood or weighted piece of Aluminum foil. After keeping this trap for 24 hors, remove the jar and cover with lids and take to the laboratory analysis. The controls for this experiment, keep one jar on polytheen sheet at same field covered by metal cylinder.

In the laboratory,

1. Add excess amount of 3N of $BaCl_2$ to the NaOH solution to precipitate the carbonate as insoluble $BaCO_3$.
2. Add few drops of Phenopthaline as indicator.
3. Titrate the untreated NaOH with 1N HCl directly in the jar. Note the volume of acid needed to titrate the alkali.

Calculation

The following formula can be used to calculate the amount of CO_2 evolved from the soil during the exposure to alkali

Milligrams of C or CO_2 = (B-V)NE

B = Volume (mL) of acid needed to titrate the NaOH in the jars from control cylinders to the end point,

V = Volume (mL) of acid needed to titrate the NaOH in the jars exposed to the soil atmosphere to end point

N = Normality of the acid

E = Equivalent weight. To express the data in, in Carbon: E=6, To express it as CO_2 E=22

The units of this formula can express as milligrams of CO_2 per square meter per hour (mg/m^2 hour).

Appendix 12

Determination of mulch decomposition rate

The litterbag technique of (Wardle *et al*. 1999) was used to determine the mulch decomposition rate. Five samples of the mulches namely, dry tea leaf mulch (control), Refuse tea, Mana grass, Dadap, Lemon grass were placed in the 30 x 45 cm^2 nylon mesh bags (mesh size of 1mm) keeping the thickness to 2.5 cm then placed randomly on the soil surface in three replicates of each mulch treatment without a pH modifier. Existing litter was removed from a small area prior to placing the litter bags against the bare soil thereby ensuring maximum influence of microorganisms with leaf litter. Litter bag measurements were not done with other pH modifiers due to a practical difficulty of obtaining replicates. The above measurements, however, provide an indication of mulch decomposition rate. Litterbags were sampled at after 0 (the fresh sample), 14, 35, and 62 days. After bringing litterbags to the laboratory, litter losses were determined from the oven dry weight (85^0 C, after 48 hrs). A graph was drawn of days (t) again the mass of litter remaining in the bag: ln (Yt), and a decay constant was calculated by using the equation of

$Y(t) = Y(0)\, e^{-kt}$ ----------(1)

Y(0)= original mulching material at time 0,

Y(t)= mulching material present at a time t (days),

k is the decay constant,

Taking the logarithm of both sides: ln Yt=ln Y0-kt

This is in the form of Y=mX+C

From this data, the graph of t (days) was plotted against ln Yt to determine gradient k.

Calculation of half life (Yt) = Y0/2, by substituting in equation (1)

Ln (Y0/2)=Ln Y0-kt

Kt=ln 2,

t=ln 2/k

Where t is time and k is decay constant

Appendix 13

Soil texture analysis (Gee, G.W. and Bauder, J.W. 1986)

Hydrometer method

Particle size analysis can be done conveniently with a hydrometer, which allows for nondestructive sampling of suspensions undergoing settling. This method provides for multiple measurements on the same suspension so that detailed particle-size distributions can be obtained.

Apparatus and Reagents

1. Standard hydrometer, ASTM no 152 H, with Bouyoucos scale in g/L
2. Electric stirrer (malted-milk-mixer type, with 10000-rpm motor)
3. Plunger or rubber stoppers for 1000-mL sedimentation cylinders
4. Sedimentation cylinders with 1-L mark $36^{+}_{-}2$ cm from the bottom of the inside
5. Metal dispensing cups and 600 mL beakers
6. Amyl alcohol
7. Sodium-hexametaphosphate (HMP) solution (50 g/L)
8. Electric oven and weighing jars.

Procedure

1. Add 100 mL of the (HMP) solution to a cylinder and make the volume to 1L with room temperature distilled water.
2. Mix thoroughly with plunger and record temperature. Lower the hydrometer into the solution and determine Rt, the hydrometer reading of the blank solution
3. Weigh 40 g of air dry soil into a 600 mL beaker add 250 mL of distilled water and 100 mL of HMP solution, and allow soaking over-night.
4. Dry overnight at 105^{0}C, Cool, and weigh.
5. Transfer the HMP-treated sample to a dispersing cup and mix for 5 min with the electric mixer, or transfer the suspension to shaker bottles and shake overnight on a horizontal shaker.
6. Transfer the suspension to a sedimentation cylinder and add distilled water to bring up the volume to 1 L.
7. Allow time for the suspension to equilibrate thermally and record temperature.
8. Insert plunger into a cylinder and mix the contents thoroughly by holding bottom of cylinder and dislodging sediment from the bottom using strong upward strokes of plunger. This should be used end-over-end shaking for 1 min.
9. Add a drop of Amyl alcohol if the surface of the suspension is covered with foam.
10. As soon as mixing is completed, lower the hydrometer into the suspension and take readings after 30 s (R_{30}) and again at the end of 1 min.
11. Remove the hydrometer, rinse, and wipe it dry. Reinsert the hydrometer carefully about 10 s before each reading and take readings at 1.5 hrs ($R_{1.5}$) and 24 hrs (R_{24}). Record the Blank reading as R_L

Calibration
Particles sediment per 30 seconds
$P_{30\ sec} = [(R_{30}-R_l) /30] \times 100$

Particles sediment per 1.5 hrs
$P_{1.5\ hrs} = [(R_{1.5}-R_l) /30] \times 100$

Particles sediment per 24 hrs
$P_{24\ hrs} = [(R_{24}-R_l) /30] \times 100$

% Sand = 100-P30

% Clay = $[P_{24\ hrs} + (0.876 \times P_{1.5\ hrs})] / 2$

% Silt = $P_{30\ sec}$ - % Clay

Appendix 14

Determination of chlorophyll content

The chlorophyll content of leaves of the young tea was measured by a chlorophyll meter (Spectrum model SPAD-502). The SPAD-502 determines the relative amount of chlorophyll present by measuring the absorbance of leaf at red and near infrared regions. Using these two values it calculates a numerical 'SPAD value' that is proportional to the amount of chlorophyll present. Chlorophyll content was measured in 5 leaves per plot.

Appendix 15

Orthodox procedure of Tea Manufacture

(Dahanayaka, D.L.D.H. and Ziyad Mohamed, M.T. (2002). Tea leaf to the cup. The tea Research Institute of Sri Lanka.)

Tender leaf (bud and two leaves) is defined as good leaf for processing of black tea.

1. Withering

The moisture content of green leaves varies from 70-83% depending on the climatic conditions and it's influenced by cultivar, cultural practices etc. The moisture content should be brought down to about 55% by withering green leaves in troughs using an air air flow. It is very important to achieve a uniform wither without damaging the leaf. A series of biochemical reactions take place during withering.

2. Rolling

The rolling is carried out to initiate fermentation i.e. oxidation of polyphenols by polyphenol oxidase enzyme in the presence of oxygen. The polyphenols and polyphenol oxidase enzyme are spatially separated in the leaf. During rolling, the cell walls are broken so that the substrate and the enzyme are brought into contact to initiate fermentation. The machines used for this operation are orthodox rollers and rotorvanes. Once the leaf is rolled, it will break into particles of different sizes. Roll breaking is carried out to separate fine particles.

Then passing the rolled leaf through a roll breaker consisting of wire meshes, the small particles (dhools) are spread either on fermenting beds and or on racks for further fermentation.

3. Fermentation

The objective of fermentation is to allow necessary chemical changes to take place in tea to meet the consumer requirements. During fermentation, these chemical reactions will result in the development of flavour, colour, and taste of the tea liquor.

4. Drying

The objective of drying is to stop fermentation at the right time and bring down the moisture content to 3%. Odourless hot air is needed for drying tea. The source of energy for air heaters is either by liquid fuel or firewood. Conventional ECP (Endless Chain Pressure drier or FED (Fluidized Bed Drier) or a combination drier is used for drying tea.

5. Grading

Grading of tea is done by sifters consisting of different size mesh to separate particles according to their sizes. The quality of tea thus produced also varies with the grade. The 3T Electrostatic Stalk Extractor is used to separate stalks and fiber from tea.

The main tea grades produced in up country, Sri Lanka, can categorized as Broken Orange Pekoe (BOP), Broken Orange Pekoe Fannings (BOPF) and Dust 1.

Appendix 16

Factor analysis of young tea data showing no significant difference in mulch x pH interactions

General Linear Model

Between-Subjects Factors

		N
MULCH	1.00	12
	2.00	12
	3.00	12
	4.00	12
PH	1.00	16
	2.00	16
	3.00	16

Multivariate Tests

Effect		Value	F	Hypothesis df	Error df	Sig.
Intercept	Pillai's Trace	1.000	9846.441	12.000	25.000	.000
	Wilks' Lambda	.000	9846.441	12.000	25.000	.000
	Hotelling's Trace	4726.292	9846.441	12.000	25.000	.000
	Roy's Largest Root	4726.292	9846.441	12.000	25.000	.000
MULCH	Pillai's Trace	1.438	2.071	36.000	81.000	.004
	Wilks' Lambda	.040	4.089	36.000	74.593	.000
	Hotelling's Trace	13.221	8.691	36.000	71.000	.000
	Roy's Largest Root	12.453	28.019	12.000	27.000	.000
PH	Pillai's Trace	.668	1.087	24.000	52.000	.389
	Wilks' Lambda	.441	1.054	24.000	50.000	.425
	Hotelling's Trace	1.020	1.020	24.000	48.000	.463
	Roy's Largest Root	.619	1.341	12.000	26.000	.255
MULCH * PH	Pillai's Trace	1.606	.914	72.000	180.000	.665
	Wilks' Lambda	.125	.916	72.000	141.819	.656
	Hotelling's Trace	2.829	.917	72.000	140.000	.655
	Roy's Largest Root	1.351	3.378	12.000	30.000	.003

a Exact statistic
b The statistic is an upper bound on F that yields a lower bound on the significance level.
c Design: Intercept+MULCH+PH+MULCH * PH

Tests of Between-Subjects Effects

Source	Dependent Variable	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	OC	394.261	11	35.842	.550	.855
	GI	4.981	11	.453	.507	.886
	CEC	5.488	11	.499	.532	.868
	YLD	1711418.667	11	155583.515	3.453	.002
	CHLO	156.217	11	14.202	1.874	.077
	TN	1.055E-02	11	9.591E-04	.379	.956
	LOGPH	2.308E-02	11	2.098E-03	2.113	.045
	LOGTP	5.960E-02	11	5.418E-03	1.088	.398
	LOGMBC	1.306	11	.119	12.130	.000
	SQRTRES	13.277	11	1.207	1.868	.078
	SQRTTEMP	.188	11	1.711E-02	.867	.578
	MOIS	193.294	11	17.572	.225	.994
Intercept	OC	48618.416	1	48618.416	745.767	.000
	GI	3939.650	1	3939.650	4412.705	.000
	CEC	699.366	1	699.366	746.422	.000
	YLD	177669856.333	1	177669856.333	3943.173	.000
	CHLO	175172.085	1	175172.085	23115.915	.000
	TN	11.976	1	11.976	4729.966	.000
	LOGPH	20.832	1	20.832	20976.184	.000
	LOGTP	.644	1	.644	129.367	.000
	LOGMBC	255.335	1	255.335	26081.650	.000
	SQRTRES	601.207	1	601.207	930.655	.000
	SQRTTEMP	968.702	1	968.702	49116.071	.000
	MOIS	39802.753	1	39802.753	510.721	.000
MULCH	OC	2.281	3	.760	.012	.998
	GI	.721	3	.240	.269	.847
	CEC	3.654E-02	3	1.218E-02	.013	.998
	YLD	1187517.167	3	395839.056	8.785	.000
	CHLO	61.509	3	20.503	2.706	.060
	TN	1.617E-03	3	5.389E-04	.213	.887
	LOGPH	7.480E-03	3	2.493E-03	2.511	.074
	LOGTP	1.629E-02	3	5.430E-03	1.090	.366
	LOGMBC	1.248	3	.416	42.502	.000
	SQRTRES	8.496	3	2.832	4.384	.010
	SQRTTEMP	8.224E-02	3	2.741E-02	1.390	.262
	MOIS	109.829	3	36.610	.470	.705
PH	OC	86.811	2	43.406	.666	.520
	GI	1.783	2	.891	.998	.378
	CEC	1.195	2	.598	.638	.534
	YLD	88160.167	2	44080.083	.978	.386
	CHLO	13.983	2	6.991	.923	.407
	TN	3.762E-03	2	1.881E-03	.743	.483
	LOGPH	7.647E-03	2	3.823E-03	3.850	.031
	LOGTP	1.011E-02	2	5.056E-03	1.015	.373
	LOGMBC	1.868E-02	2	9.341E-03	.954	.395
	SQRTRES	3.550	2	1.775	2.747	.078
	SQRTTEMP	1.685E-02	2	8.425E-03	.427	.656
	MOIS	14.285	2	7.142	.092	.913

Tests of Between-Subjects Effects Contd……..

MULCH * PH	OC	305.169	6	50.862	.780	.591
	GI	2.478	6	.413	.463	.831
	CEC	4.256	6	.709	.757	.608
	YLD	435741.333	6	72623.556	1.612	.172
	CHLO	80.725	6	13.454	1.775	.132
	TN	5.171E-03	6	8.618E-04	.340	.911
	LOGPH	7.957E-03	6	1.326E-03	1.335	.267
	LOGTP	3.320E-02	6	5.533E-03	1.111	.375
	LOGMBC	3.935E-02	6	6.558E-03	.670	.674
	SQRTRES	1.231	6	.205	.318	.924
	SQRTTEMP	8.910E-02	6	1.485E-02	.753	.611
	MOIS	69.180	6	11.530	.148	.988
Error	OC	2346.930	36	65.193		
	GI	32.141	36	.893		
	CEC	33.731	36	.937		
	YLD	1622073.000	36	45057.583		
	CHLO	272.807	36	7.578		
	TN	9.115E-02	36	2.532E-03		
	LOGPH	3.575E-02	36	9.931E-04		
	LOGTP	.179	36	4.981E-03		
	LOGMBC	.352	36	9.790E-03		
	SQRTRES	23.256	36	.646		
	SQRTTEMP	.710	36	1.972E-02		
	MOIS	2805.640	36	77.934		
Total	OC	51359.607	48			
	GI	3976.772	48			
	CEC	738.584	48			
	YLD	181003348.000	48			
	CHLO	175601.110	48			
	TN	12.078	48			
	LOGPH	20.891	48			
	LOGTP	.883	48			
	LOGMBC	256.993	48			
	SQRTRES	637.740	48			
	SQRTTEMP	969.600	48			
	MOIS	42801.686	48			
Corrected Total	OC	2741.191	47			
	GI	37.122	47			
	CEC	39.218	47			
	YLD	3333491.667	47			
	CHLO	429.025	47			
	TN	.102	47			
	LOGPH	5.884E-02	47			
	LOGTP	.239	47			
	LOGMBC	1.659	47			
	SQRTRES	36.533	47			
	SQRTTEMP	.898	47			
	MOIS	2998.933	47			

Appendix 17

Relative amounts of selected fatty acids extracted from soils collected in dolomite plots after 56 weeks application of treatments in young tea trial in Sri Lanka. A FAME (Fatty acid methyl ester) analysis was carried out by Dr. C. Pankhurst, CSIRO Land and Water, Adelade.

Treatments	Gram positive bacteria									Gram negative bacteria		Fungi 18: 2 w6c	Mycchо -rrhiza 16: 1 w5c	?? 19:1 w8t/S
	15: 0 ISO	15: 0 ANTE ISO	15: 0	16: 0 ISO	16: 0	17: 0 ISO	17: 0 ANTE ISO	21: 0 ISO (unknown)	Total Gram + ve Bacteria.	10: 0 3OH	12: 0 3OH			
No mulch No pH modifier	2.94	1.21	0.60	1.39	8.71	0.78	0.79	7.32	23.74	0.70	0.85	2.81	1.78	21.25
Refuse tea Dolomite	2.88	1.16	0.62	1.28	7.48	0.72	00	7.99	22.13	0.50	0.94	1.84	1.82	26.70
Mana grass Dolomite	2.59	1.02	0.66	1.16	7.62	0.75	0.41	8.79	23.00	0.47	0.78	2.91	1.18	21.99
Dadap legume Dolomite	3.34	1.26	0.54	1.56	9.35	0.87	0.48	7.46	24.86	0.44	0.66	3.05	1.93	19.13

* = Fatty acids are designated by the number of carbon atoms, followed by a colon, the number of double bonds and then by the position of the first double bond from the methyl (w) end of the molecules. *Cis* and *trans* isomers are indicated by C or t. Branched-chain fatty acids are indicated by the prefixes i and a for iso and anteiso-branching, respectively. The prefix designates cyclopropane fatty acid.