OPTIMIZATION OF SOIL ENVIRONMENT AND CROP RESPONSE FOR MAGNESIUM NUTRITION IN ULTISOL

by

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THESIS

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DECLARATION

I, hereby declare that the thesis entitled 'Optimization of soil environment and crop response for magnesium nutrition in Ultisol' is a bonafide record of research done by me during the course of study and the thesis has not previously formed the basis for the award of any degree, diploma, fellowship or other similar title, of any other University or Society.

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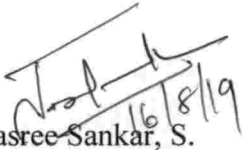
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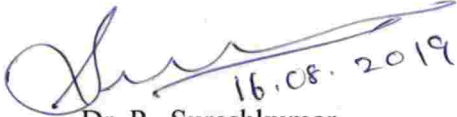
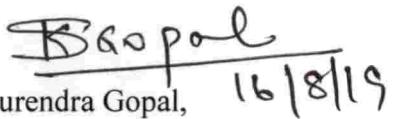Agronomic Research Station,

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CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

Introduction

1. INTRODUCTION

Magnesium is an essential macronutrient with respect to plant nutrition. It is required by plants in quantities lesser than nitrogen, potassium and calcium. Magnesium along with calcium and sulphur are referred to as secondary nutrients because they are less yield limiting than the macronutrients (N, P, and K), yet are required by crops in relatively large amounts.

Magnesium is the eighth most abundant element constituting two per cent by weight of earth's crust. But about 98 per cent of the element is incorporated into the crystal lattice structure of layer silicate minerals and are not easily available for plant absorption. Magnesium is present in soil as free ions (Mg^{2+}), as ions adsorbed on to mineral or organic surfaces, as dissolved compounds, as part of lattice structures or contained within the soil biota. The most important soil properties governing magnesium availability are soil pH, texture, cation exchange capacity, organic matter, and soil moisture.

Plants require magnesium for many physiological and biochemical processes *viz.*, photophosphorylation, photosynthetic carbon dioxide fixation, phloem loading and partitioning of photo assimilates, protein synthesis *etc*. Apart from the physiological functions, magnesium modifies the rhizosphere environment preferably for the growth of beneficial micro flora. It is also a structural component of chlorophyll. Application of magnesium has profound impact on alleviating soil acidity along with the nutrient supply.

The soils of Kerala have been developed from parent rocks of acid igneous nature under tropical humid climate. Formed under the influence of high rainfall and undulating topography, soils are largely acidic due to leaching of bases. Soils of Kerala can be classified under Ultisols, Alfisols, Inceptisols, Entisols and Histosols, of which Ultisols occupy more than fifty per cent of total geographical area of the State. Ultisols

1

are characterized by low pH, low cation exchange capacity and low base saturation due to dominance of kaolinite clays and oxides and hydrous oxides of iron and aluminium. The deficiency of magnesium is a common nutritional disorder in these soils.

Magnesium present as a divalent cation in the soil solution, are bound less avidly to soil particles due to greater hydrated radius than other cations and is prone to leaching. This is considered to be an important factor influencing Mg phytoavailability especially in coarse-textured soils (Wilkinson *et al.*, 1990). Magnesium deficiency is also aggravated by high concentrations of competing cations, particularly H^+, Al^{3+} and Mn^{2+} in soil solution (Hailes *et al.*, 1997). Recent reports project 65 per cent of soils to be deficient in available magnesium in tropical soils of Kerala (Bhindhu and Sureshkumar, 2016). Crops are found to respond to applied magnesium fertilizers with an increase in quantity and quality of produce.

Various magnesium fertilizers are used for correcting magnesium deficiency of which kieserite or magnesium sulphate is widely used. But the application of soluble fertilizers under high rainfall situations prevailing in Kerala aggravates the loss. Hence a less soluble source is more congenial to improve nutrient use efficiency. Magnesium carbonate or dolomite can be promising sources in acid lateritic soil due to sparingly soluble nature and sufficient magnesium content (Dechen *et al.*, 2015).

The determination of critical level of nutrient in soil and plant helps to manage nutrient deficiency and to avoid crop loss. According to the *adhoc* recommendations for management of secondary and micronutrients in Kerala, soils with exchangeable magnesium level less than 120 mg kg^{-1} are classified as deficient, which is seldom attained under acidic soil environment. Hence, the management of magnesium deficiency in acid soils require the improvement in soil pH. Application of dolomite or magnesium carbonate can raise the soil pH apart from increasing the availability of magnesium in soil. The improvement in soil pH can have a positive influence on plant

growth and population of soil micro flora. Hence, the present study was undertaken with the following objectives:

- Evaluate the modifications of the acidic soil environment through added magnesium source
- Validate the critical level of magnesium in soil and plant thereby optimizing magnesium nutrition for cowpea

Review of Literature

2. REVIEW OF LITERATURE

The essentiality of magnesium as a plant nutrient was established as early as 1839 by Sprengel. But as the relative abundance of magnesium in plant is far less in comparison to nitrogen, potassium and calcium, the importance of the nutrient in crop production was over looked during the past decades. But recently magnesium deficiency has been identified as a limiting factor under intensive cropping systems with little response to NPK fertilizers.

In highly weathered tropical acid soils with high leaching potential depletion of magnesium from soil is a growing concern. The negative balance of magnesium in acidic soils is jeopardizing crop yield. Research has been augmented to reveal the significance of magnesium for enhancing crop yield and ways to optimize crop nutrition. A review of research that supports the present investigation is detailed here.

2.1. Soil- a store house of magnesium

Magnesium is the eighth most abundant element in the earth's crust comprising about two per cent by weight and one among the dominant cations on the exchange complex of soil. About 90–98 per cent of the soil Mg is incorporated in the crystal lattice structure of minerals, thus not directly available for plant uptake.

Appreciable quantities of magnesium exists in soil minerals like olivine, amphibole, pyroxene, biotite, chlorite, serpentine, montmorillonite, vermiculite, brucite, schoenite, and epsom salt. Additionally, carbonates such as magnesite ($MgCO_3$) and dolomite ($MgCO_3 + CaCO_3$) also provide reasonable quantities of Mg ranging from 10 to 30 g kg^{-1} (Dechen *et al.*, 2015). Bioavailable forms of Mg originates from weathering of magnesium containing minerals. Owing to the variation of magnesium in parent material and their degree of weathering, the total content of Mg in soil varies considerably between 0.05 per cent and 0.5 per cent (Grimme, 1991; Maguire and Cowan, 2002).

2.1.1. Forms of magnesium in soil

Magnesium is a highly reactive metal and all magnesium in the biosphere is either present as free cation (Mg^{2+}) in aqueous solutions or as some salt or mineral form. In soil, magnesium is present mainly in inorganic compounds, although considerable amounts also appear associated with the organic material in humus (Jodral-Sedago, *et al.*, 2006). Primary and secondary minerals comprises the major source of magnesium in soil. The content and availability depends on the parent material and their degree of weathering. According to Pasricha and Sarkar (2009) magnesium is present in soil as water soluble, exchangeable and non- exchangeable forms; the water soluble and exchangeable magnesium are always in equilibrium with non-exchangeable form. They also observed that magnesium can occur in soil as its oxides with the content of MgO ranging between 0.24 to 1.2 per cent in acid soils.

Roy *et al.* (2006) reported that the non-exchangeable Mg accounts to more than 90 per cent of total Mg; exchangeable Mg about 5 per cent of total Mg and water-soluble Mg about 1–10 per cent of exchangeable Mg. While non-exchangeable Mg is contained in the primary minerals and in secondary clay minerals, Mg on the exchange complex and in the soil solution is most important for plant nutrition.

Only a small per cent of the available nutrients move freely in the soil solution, while most of them are loosely bound on mineral and organic surfaces in exchangeable form. In soils of neutral soil reaction, about 75 per cent of the adsorbed cations are Ca and Mg. According to Barber (1984) the concentration of magnesium in the soil solution varies from 0.5 to 2 mmol L^{-1} and 2-8 mmol L^{-1} in leached and unleached soils respectively. The concentration of magnesium ranging between 125 and 8.5 mM in soil solution is sufficient for mass flow to supply magnesium to plant roots for growth requirements (Barber, 1995).

A fractionation scheme was proposed by Mokwunye and Melsted (1972) for separating total soil Mg into exchangeable, acid-soluble, organic complexed, and

mineral fractions in tropical and temperate soils. They recorded the order of abundance of different fractions to be: mineral Mg> acid-soluble Mg> exchangeable Mg> organic complexed Mg. According to Hailes *et al.* (1997) the contribution of various fractions to total Mg was in the order mineral > acid-soluble $\approx$ exchangeable $\gg$ organic-complexed. The acid soluble magnesium fraction is considered by Metson and Brooks (1975) to be a measure of potentially available magnesium that can replenish the exchangeable fraction as it gets depleted. Stahlberg (1960) determined the amount of slowly exchangeable Mg released from several Swedish top soils (boiling them in 1 N HCl) and concluded that vermiculite and chlorite are the main sources of acid-soluble Mg. The chlorites have silicate layers with brucite-like (essentially Mg $(OH)_2$) interlayers, but vermiculites contain appreciable magnesium in the interlayer positions.

The ferromagnesian minerals (olivines, pyroxenes, and amphiboles) are the main mineral forms in basic igneous rocks which would have largely disappeared from older, more developed soil because of their ease of weathering. The finely divided clay minerals of the layer-lattice type, such as montmorillonite and illite, contain lattice magnesium substituting for aluminium in octahedral positions. Dioctahedral micas (muscovite-type) have magnesium substituting for part of the aluminium in octahedral positions and trioctahedral (biotite-type) micas, magnesium and ferrous iron are the dominant octahedral cations (Metson, 1974).

2.1.2. Factors affecting availability of magnesium in soil

Availability of magnesium mainly depends on its form and content in soil. According to Barber, (1984) water soluble and exchangeable forms of magnesium maintain equilibrium in soil and constitute the labile pool of soil magnesium. The important factors that determine magnesium availability are discussed here after.

2.1.2.1. Soil pH

Soil pH has a direct effect on the availability of magnesium in soils. In acidic soils magnesium availability is reduced by competition from H, Al and Mn whereas in alkaline soil carbonate formation and excess concentrations of Na, K, and Ca reduces the availability. Mg deficiency can be widespread in acid soils as a consequence of low supply and leaching losses (Roy *et al.*, 2006). The availability of Ca and Mg reduces with decrease in pH (<pH 6) as a consequence of their inability to build up and maintain sufficient pH and electrochemical gradient across the plasma membrane of root cells (Schubert *et al.* 1990). Fageria (1998) also reported a decrease in uptake of Ca and Mg with decrease in pH. The solubility of magnesium minerals are high below pH 7.5 and helps to maintain a reference level of magnesium (10^{-3} M) in soil solution. In alkaline soil, dolomite is the solid phase which controls soil solution Mg whereas in acid soils, the content of Mg in soil solution is buffered by exchangeable phase (Lindsay, 1979).

2.1.2.2. Soil type

Soil texture is a key variable that affects plant-available Mg. Because Mg is located in clay minerals and associated with cation exchange sites on clay surfaces, clayey soils generally contain adequate Mg for plant requirements, whereas sandy soils are frequently deficient in magnesium (Mayland and Wilkinson, 1989). Chung-Ho and Johnson (1985) studied the relationship between exchangeable and total Mg in Alfisols of Pennsylvania and found close correlation between exchangeable Mg and Mg in sand and silt but not clay, which revealed that sand and silt fractions are important source of exchangeable Mg in soil.

Magnesium availability to plants are influenced by parent material, intensity of weathering as well as capacity of soil to retain and supply magnesium to plants as and when it is needed. Structural and exchangeable magnesium is highly dependent on clay mineralogy of soil. The order of abundance of Mg in some of the Mg containing

minerals are muscovite > biotite > hornblende > augite > olivine. Chlorite, vermiculite, and montmorillonite clays have undergone intermediate weathering and still contain some Mg as part of their structure. Illite contain Mg in their structure but potassium is released more easily than Mg. Soils rich in kaolinitic clays are highly weathered and have low Ca and Mg availability (Baker, 1972).

Mg deficiencies are less likely in Alfisols than in Ultisols, and less likely in Mollisols than Alfisols. An important aspect of soil type is rate and amount of Mg released from nonexchangeable sources (Wilkinson et al., 1990).

2.1.2.3. Cation Exchange Capacity of soil

Cation exchange capacity is the important property that determines retention of basic cations in soil. Tropical soils are intensely weathered and dominated by variable charged clays having low cation exchange capacity which varies with the soil pH (Uehara and Gillman, 1981). According to Akselsson et al. (2007) cation exchange complex in soil is the source and sink reservoir of basic cations. Exchangeable fraction of Mg is a good indicator of available pool in soil.

Low CEC implies lower retention of basic cations and results in deficiencies of Ca, Mg and K. But in contrast to other cations like K, Ca, and NH_4^+, Mg have a smaller ionic radius and higher hydrated radius as a result, magnesium is loosely bound to soil charges (CEC) which leads to higher concentration in soil solution and contributes to higher leaching losses (Shaul, 2002). Apart from this, increase in calcium saturation of cation exchange sites also reduces magnesium availability.

A severe magnesium deficiency in most of the crop plants were observed when soil exchange capacity is saturated with less than 6 per cent by magnesium (Wilkinson et al., 1990). According to Roy et al. (2006) the ratio of the nutrients is considered well-balanced when about 65 per cent of CEC is saturated by Ca, 10 per cent by Mg, 5 per cent by K and the remaining 20 per cent by others (H, Na, etc.).

2.1.2.4. Interaction with other ions in soil solution

Mg^{2+} uptake by plants is reduced with levels of K^+, Ca^{2+}, Al^{3+}, H^+, Mn^{2+} and NH_4^+. Soluble or exchangeable Al in soil interferes with uptake of Ca and Mg more than that of K, thereby increasing K/ (Ca + Mg) values. Ohno and Grunes (1985) studied the K-Mg interactions and reported that increasing K supply depressed Mg concentrations in plant tops, but not in roots. They concluded that K inhibits translocation of Mg from the root to plant top.

Wilkinson *et al.* (1990) reported that the rate of Mg uptake doubled when K concentration at the root surface in soil solution decreased below 20 μmol L^{-1}. They also observed that presence of aluminium on exchange sites reduces Mg uptake more than K and Ca. Hecht- Buchholz *et al.* (1987) found that increasing Mn concentrations in the nutrient solution from 3-920 mM reduced the concentration of K by 5 per cent, Ca by 35 per cent and magnesium by 72 per cent in plants. Mayland and Wilkinson (1989) also observed that magnesium uptake at root surface was inhibited due to Mn toxicity at low pH.

2.1.2.5. Organic matter

Soil organic matter contain more carboxylic group contributing to CEC at pH < 8 and phenolic hydroxyl group contributing to CEC at pH above 7. In most of the mature tropical soils, pH dependent variable charge of organic matter contributes to CEC. Increasing soil organic matter concentrations increases the cation exchangeable capacity and improves the Mg supply available for plant uptake (Mathan and Rao, 1982). Stofella and Kahn (2001) reported that Mg concentration in compost ranges from 1-5 g kg^{-1} with a mean value of 3.5 g kg^{-1}. Addition of organic manure at the rate of 300 kg ha^{-1} was found to raise exchangeable soil magnesium by 5.5, 2.4, and 0.6 meq/100 g in the 0-4, 4-8, and 8-15 cm layers respectively, which persisted for at least 9 months (Metson, 1974).

2.1.2.6. Fertilizer applications

Crop fertilization with acid forming fertilizers increases soil acidity and thus loss of Mg (Ellis, 1979). Magnesium deficiency can be induced by excess potassium fertilizer application which is a common phenomenon in banana and coffee plantations where fertilizers high in K are being used.

In sandy soils, application of high rates of potassium or ammonium fertilizer enhances the risk of Mg deficiency as high concentrations of these cations in the soil solution interfere with Mg uptake. But the antagonistic effect of potassium on magnesium do not occur when the soil contains more exchangeable Mg than exchangeable K (Senbayram et al., 2015).

Mengel and Kirkby (2001) also reiterated that the application of N, P, K fertilizers without sufficient supply of magnesium fertilizers accompanied with profound leaching in lateritic and sandy soil makes the deficiency of magnesium a major concern in crop nutrition.

Ammoniacal N is antagonistic to the uptake of Mg by plants, whereas NO_3 fertilization often enhances Mg uptake (Mayland and Grunes, 1979). Lasa et al. (2000) found that sufficiently high Mg supply along with NH_4 fertilization reduced the toxicity of NH_4 in sunflower grown in sandy soil.

2.2. Magnesium status in soils of Kerala

Soils of Kerala are classified under the soil orders- Ultisols, Alfisols, Entisols, Inceptisols and Vertisols (Sureshkumar et al., 2018) of which Ultisols occupy more than 50 per cent of the total geographical area of the state (Krishnan et al. 1996).

The Ultisols of Kerala have low pH, low cation exchange capacity, low effective cation exchange capacity and base saturation, with dominant presence of 1:1 clays and gibbsite along with hydroxy-interlayered vermiculites (HIV), Mica-HIV, and

mica in the silt and clay fractions indicating the presence of significant amounts (>10%) of weatherable minerals (Chandran et al., 2005). The nutrient supply of a soil is mainly by the weathering of soil minerals and further release to available pool. The Al^{3+} released during the weathering of these minerals occupies the interlayer position of clays making it very difficult to weather.

Based on the studies conducted on the magnesium status of acidic soil in Kerala, Prema (1992) reported that the soils are deficient in total magnesium reserves with a mean value of 963.7 mg kg^{-1}. In general, about 24 per cent of the total Mg reserves in the soil were considered available and 11 per cent of the total Mg was considered exchangeable.

Nair et al. (2013) reported magnesium deficiency to the tune of 74 per cent in the soils of Kerala with only exemption in the soils of Central and Eastern Palakkad and Attapady hills. According to Bhindhu and Sureshkumar (2016) available magnesium status in soil samples collected from different agro-ecological units of Kerala varied widely from as low as 9.56 to 1931.25 mg kg^{-1} with 68 per cent of the soils recorded as deficient. The inability of lateritic soils to retain Mg on the exchange surface reveals the importance of managing magnesium nutrition in these soils.

2.3. Effect of magnesium on nutrient availability in soil

2.3.1. Macronutrients

There exists a positive interaction between nitrogen and magnesium in soil. If nitrogen is available in soil it releases NO_3^- and electrical neutrality is maintained internally by its reduction in synthesizing organic acids or by release of root anions such as OH^- or HCO_3^-, these are bound with cations such as Ca^{2+}, Mg^{2+} and Na^+ (Barber, 1995).

Adams (1980) reported the occurrence of positive correlation and interactions between phosphorus and magnesium in soil. Magnesium helps in greater solubilization

of phosphorus in soil and also act as a carrier of phosphorus which then contributes to improving availability P in soil (Jacob, 1958).

Interaction of Mg and potassium are very important in maintaining soil health, quality and production in leafy vegetables. Generally Mg has an effect on K translocation in soil. The low magnesium status in soil may decreases the available K (Hannaway *et al.*, 1982).

Barber (1995) reported the existence of negative interaction between calcium and magnesium in soil. Kene *et al.* (1990) observed that the reduced uptake of calcium by plants in high magnesium containing soil and the plants grown under such conditions showed calcium deficiency. Fageria (1974) reported that high levels of Ca^{2+} in the nutrient solution affected the absorption of Mg^{2+} and K^+ by rice plants, and showed that as the concentration of Ca^{2+} rises, absorption rates of both Mg^{2+} and K^+ increase.

Halvin *et al.* (2004) found that magnesium sulphate application increases suphur content in different soils.

2.3.2. Micronutrients

Magnesium shows synergistic effect with manganese in soil. In wheat, Mg increased the tolerance of plants to high concentrations of manganese in shoot tissue and also increased the ability of the plant to discriminate against manganese ions in translocation of nutrients from roots to shoots (Goss, *et al.* 1992). Malcova *et al.* (2002) found the toxicity produced by excess Mn to be alleviated with the application of magnesium.

Disch *et al.* (1994) reported that iron content was increased with the application of magnesium fertilizers. According to Krauskop (1972) magnesium and zinc exhibits synergistic or antagonistic effect.

2.4. Magnesium and rhizosphere microflora

Magnesium is also an essential element for growth of beneficial microbes in soil. Jones and Huber (2007) reported an increase in the reproduction of soil bacteria with the application of magnesium carbonate. Calcium and magnesium are essential elements for efficient nitrogen fixation by rhizobia and magnesium deficiency results in reduced nitrogen fixation (Dechen *et al.*, 2015).

Vincent (1962) observed that either magnesium or calcium deficiency can decrease the total amount of growth in *Rhizobium trifolii* and he also concluded that an equal increment of total growth of Rhizobium requires about eight times greater molar concentration of Mg than Ca. Sufficient Mg supply considerably increased nodule number, size, weight, mass, nodulation index and nitrogenase activity in nodules of soybean plant validating that Mg supply plays crucial role in nodule formation and development (Khaitov, 2018).

Edwards and Kelly (1992) studied the effect of added magnesium on per cent root colonization of mycorrhiza in pine roots and observed a greater colonization of morphotype-A mycorrhiza in seedlings grown with 35 mg kg^{-1} magnesium than that of seedlings grown in soil having 15 mg kg^{-1} Mg.

2.5. Effect of magnesium on nutrient uptake by plants

2.5.1. Macronutrients

Magnesium shows synergistic effect with nitrogen in plants. Nitrogen metabolism, subsequently protein metabolism was disturbed in magnesium deficient plants. Adequate Mg nutrition of crop plants is necessary for better nitrogen- use efficiency and grain N accumulation. Low availability of Mg during plant development influence the rate of NO_3^- absorption and leaf growth, as well as the supply of assimilates to the roots (Cakmak and Kirkby, 2008).

Kumar *et al.* (1981) reported that the uptake of phosphorus was not affected at low magnesium application rates and a decrease in phosphorus uptake was there at high dose of magnesium application.

Potassium and magnesium exhibit antagonistic effect and antagonistic effect of K on Mg uptake was more significant when compared to Mg on K uptake (Ding *et al.* 2006). The work conducted by Nguyen *et al.* (2017) in Pummelo showed that high exchangeable Na and low K/ Mg ratio inhibited the uptake of K from soil even though the soil was abundant in K. They concluded that the content of Mg and K in plant leaves proved the negative correlation between them.

Excess magnesium over calcium decreases the productivity of crops, health of roots was reduced with excess application of magnesium and thus leads to a reduction in turgidity (Bolton, 1973).

The significant increase in magnesium content in mustard seed was observed with the application of Mg (Gupta and Singh, 1990). Singh and Singh (1990) also reported the similar results in linseed.

The content of sulphur in leaves increased with the application of magnesium as magnesium sulphate. Sulphur content was increased from 0.34% to 0.40% S after the application $MgSO_4$ (Lopez, 2010).

2.5.2. Micronutrients

Manganese and magnesium concentrations in leaves varied in opposite directions, suggesting an inverse relationship in the absorption of these elements (Pakrasi *et al.*, 2001). Khan *et al.* (1997) observed that there exists a higher positive correlation between magnesium and Cu content in plants. Nayana *et al.* (1985) reported the beneficial effect of Mg in increasing leaf zinc concentration of Coorg mandarin.

2.6. Role of magnesium nutrition in plant

Magnesium is second most important cation in plants. Major proportion of total magnesium exist as mobile forms (Marschner and Rengel, 2012) and the share of Mg bound to chlorophyll depends on the Mg status and ranges from about 6 to 25 %, showing highest values in Mg-deficient plants. Apart from that, 5–10 % of total Mg is firmly bound to cell wall pectins and sparingly soluble salts in the vacuole. The remaining fraction fulfils further roles in plant physiology, some of which are also associated with photosynthesis. In contrast to the function of Mg in light interception these additional aspects are related to the charge and high trans-membrane mobility of Mg^{2+}.

Magnesium plays unreplaceable role in crop nutrition as it forms the central atom in porphyrin ring structure of chloroplast, a key component in photosynthetic carbon dioxide fixation (Cakmak and Kirkby, 2008). A significant increase in total chlorophyll, chlorophyll a, and chlorophyll b concentrations in mature leaves of soybean and maize plants were noted with the addition of magnesium fertilizer to low-magnesium containing soil as magnesium carbonate (Chen *et al.*, 2017).

Magnesium helps in phloem loading and transport of photoassimilates from source to sink organs such as roots, shoot tips and seeds (Cakmak *et al.*, 1994). The high phloem mobility of magnesium ions ensures the faster transport of photo assimilates to all sink organs such as root, shoot tips and leaves for their growth and reproduction.

Magnesium act as co-factor for >300 enzymes which includes peroxidase (involved in plant defence mechanism), Rubisco (ribulose-1,5-bisphosphate carboxylase/ oxygenase), kinases, ATPase, RNA polymerases etc (Cowan, 2002). It also takes the role as allosteric modulator of many enzymes.

15

Magnesium plays an important role in protein synthesis through the binding of ribosomal subunits. The absence of magnesium during protein synthesis results in irreversible unfolding of ribosomal subunits and loss of protein polymerizing activity leading to increased concentrations of the precursor amino acids (Fischer *et al.*, 1998). Magnesium helps to stabilize the conformational structures of nucleic acids and it also plays a major role in functioning of nucleic acid synthesizing polymerases and degrading nucleases (Sreedhara and Cowan, 2002). In addition, Mg along with K serves as cation in the regulation of the cation-anion balance and also osmotically active ion in turgor regulation of cells (Marschner and Rengel, 2012).

Magnesium plays noteworthy function in cell energy balance through its interaction with pyrophosphate structure of nucleotide tri and di- phosphates. The energy rich compounds Mg- ATP and Mg- ADP represent the main complexed Mg pools in the cytosol and they maintain balance with the free Mg^{2+} pool under the control of adenylate kinase (Igamberdiev and Kleczowski, 2003).

Maintaining sufficiently high supply of magnesium for crops through Mg fertilization is important in crop production to minimize heat and radiation related losses. The expression of Mg deficiency-induced leaf chlorosis in common bean plants was markedly prevented by providing partial shading or promoted by the partial exposure to high light (Cakmak, 2013).

Another impressive function of adequate Mg supply is its mitigating effect on aluminum (Al) toxicity in plants which is a common growth-limiting factor in acidic soils. The probable mechanisms in plants which impart resistance on exposure to high Al content are exudation of organic acid anions, better carbon partitioning from shoots to roots, maintenance of H^+-ATPase activity on plasma membranes and better cytoplasmic pH regulation (Cakmak, 2013). Silva *et al.* (2001) observed that tap root tips of two soybean genotypes grown in 4.6 μM Al activity solution produced high citrate and malate concentrations after additions of either Ca up to 3mM or Mg up to

50 µM. The findings proved the superior effectiveness of Mg over Ca in alleviating Al rhizotoxicity.

Magnesium content in the plants also alters the pest and disease incidence depending on the pathogen/pest, plant species and environmental conditions. High accumulation of sugars in source leaves due to impaired phloem transportation under Mg deficiency may promote pathogen invasion and infection (Cakmak, 2013).

Magnesium along with calcium makes middle lamella more resistant to degradation by pectolytic ezymes produced by various bacterial and fungal pathogens (Bateman, 1965). A high foliar Mg concentration decreased the leaf scald symptoms in addition to partially preserving the photosynthetic performance of rice leaves attacked with *Monographella albescens* (Tatagiba *et al.*, 2016).

2.7. Effect of magnesium on growth and yield parameters

Magnesium plays important role in transportation of phosphate in plants and thus contributes to higher yields (Russel, 1975). Kiss (1977) reported that use of magnesium sulphate for seed dressing and for foliar spray increased plant height, pods per plant, seeds per pod thousand seed weight and fresh seed weight in pea. Soil application of magnesium increased the growth of the plants and better root nodulation in ground nut (Kulkarni *et al.*, 1986). Mani and Halder (1996) documented improved shoot and root dry matter yield in green gram (*Vigna radiata* L) as a result of dolomite application.

Magnesium application as magnesium sulphate produced higher number of branches per plant, leaf dry weight, photosynthetic pigment content, hundred seed weight, harvest index, seed, straw and biological yield in soyabean (Saad *et al.*, 2000). Riga and Anza (2003) observed a reduction in relative growth rate, total dry weight, and total leaf area under magnesium deficiency. Foliar application of magnesium in mung bean improved growth and yield components significantly (Kassab, 2005).

Application of magnesium at 4 per cent dose resulted in better growth and yield in tomato (Ilyas *et al.* 2014).

In banana, Mg fertilization showed positive effect on vegetative growth parameters, nitrogen, magnesium, chlorophyll a and b content in leaves, and also improved the yield and fruit characteristics (Mostafa *et al.*, 2007). Vegetative growth, green pod yield and pod quality of snap bean were significantly enhanced by the increased levels of magnesium fertilizer application (Huda *et al.*, 2010).

Foliar application of magnesium fertilizers significantly improved plant height, leaf dry weight and curd yield in cauliflower. It also enhanced the chemical composition of leaves and curd of cauliflower (Ahmad *et al.*, 2011).

Effect of magnesium was more pronounced in respect of plant height, number of branches per plant, width of flower, number of flowers per plant, fresh weight of flower, and oil content. The growth and yield parameters of Matricaria increased with increase in application rate of magnesium (Upadhyay and Patra, 2011).

Ahmed *et al.* (2012) reported that foliar application of Mg (137 ppm), Cu (97ppm), and growth regulators (20ppm 2, 4-D, 30ppm GA3 or 10ppm BA) improved growth characters and yield of Washington Navel orange trees.

Qubaie (2013) observed a significant improvement in plant height and leaf area upon spraying with 0.5% magnesium sulphate at vegetative shoot stage. Venkataramana (2014) reported improved yield in Black pepper as a result of magnesium sulphate application at the rate of 200g per wine.

2.8. Effect of magnesium on quality parameters

In potato tubers, fertilization with magnesium sulphate resulted in a reduction of enzymatic discoloration and concentration of phenolics whereas the content of crude lipid and phospholipids were increased (Klein *et al.*, 1982). Villarias *et al.* (2000)

observed an increase in sugar content of beet root from 13.6 to 16.9 per cent upon application of Mg (0-40 kg Mg per ha).

Application of 5 per cent Mg increased the seed yield, protein and oil concentration in soybean (Vrataric *et al.*, 2006). Dris *et al.* (1999) reported that Mg fertilization improves taste, flavor and storage characteristics like titrable acidity (TA), total soluble solid (TSS) concentration, fruit firmness and starch degradation in apple.

Foliar application of Mg enhanced shoot length and lignification and also increased number of roots which contributes to production of high quality grapes (Moretti, 2002). The degradation of anthocyanins were reduced upon magnesium application (Sahked- Sachray *et al.*, 2002).

Azizi *et al.* (2011) evaluated different mode of application (seed treatment, soil, foliar application and their combinations) of $MgSO_4$ and concluded that all the methods produced improved yield and quality of lentil (*Lens culinaris* L). But the highest percentage of crude protein content in seeds was from foliar application.

2.9. Magnesium deficiency

Magnesium deficiency is a common nutritional disorder in acidic soils with low organic matter content and dominant kaolinitic minerals. The two major reasons behind magnesium deficiency are absolute deficiency of the element in soil or due to competition of other cations.

Low magnesium contents in the source rocks, mobilization and subsequent leaching loss of Mg from soil and long term unbalanced fertilization practice without considering magnesium depletion from soil (van der Pol and Traore, 1993) are the major causes of absolute deficiency. Cation competition is a consequence of nutrient imbalances in soils. The uptake of Mg is strongly influenced by the availability of other cations like NH_4, Ca and K (Jacob, 1958).

2.9.1. Physiological effects of magnesium deficiency in plants

Inadequate supply of magnesium disturbs the equilibrium existing in partitioning of assimilates between roots and shoots that results in accumulation of photo-assimilates in the source organs and reduced growth of sink organs (Cakmak and Kirkby, 2008). Disturbance in carbon partitioning is regarded as a hidden deficiency symptom (Gransee and Fuhrs, 2013) and it occurs long before the appearance of visible symptoms.

The impaired phloem loading leads to accumulation of carbohydrates in magnesium deficient leaves and results in a decrease of CO_2 fixation by Ribulose-1, 5-bisphosphate carboxylase/oxygenase (Rubisco), at the early stages of the deficiency. This happens through two mechanisms and they are feedback inhibition of sucrose synthesis and accumulation of starch in the chloroplast that affects CO_2 conductance of the chloroplast membrane which leads to lesser CO_2 partial pressure at the catalytic site of Rubisco. As a result, there arises an imbalance between light capture and its utilization which triggers the production of reactive oxygen species in magnesium deficient plants (Cakmak and Yazici, 2010).

Depending on the equilibrium between enhanced ROS production and their scavenging, the increased ROS would serve as a signaling molecule and cause oxidative damages to the chlorophyll molecules (Asada and Takahashi, 1987). Inter-veinal chlorosis of older leaves due to excessive ROS production is one of the early visual symptoms of magnesium deficiency (Mengutay *et al.* 2013). Along with reduction in plant growth, photosynthetic rates were also reduced to half due to magnesium deficiency.

2.10. Response of cowpea to magnesium nutrition

Cowpea (*Vigna unguiculata*) is an annual herbaceous legume belonging to fabaceae family. Roots of leguminous plants have higher cation exchange capacity than

graminaceous plants. They requires higher proportion of basic cations in their nutrition when compared to grasses. Magnesium is an important mineral that helps in symbiotic nitrogen fixation which is an additional function carried out by leguminous plants.

Inhibition of plant growth and development due to copper toxicity is found to be alleviated by magnesium treatments in cowpea which is due to non specific reduction in the negativity of electrical potential at the outer surface of the plasma membrane and thereby decrease in the activity of Cu^{2+} at the outer surface of the plasma membrane (Kopittke et $al.$, 2011).

Chen et $al.$ (2017) suggested the presence of Mg^{2+} mediated Al-tolerance mechanisms in legumes as the addition of micromolar concentrations of Mg in the rooting media while maintaining constant Al^{3+} activity could enhance root growth.

2.11. Critical nutrient level

Critical level of a nutrient in soil refers to the level below which crops readily respond to applied nutrient. According to White and Brown (2010) the critical concentration for sufficiency is defined as the concentration in the diagnostic tissue that allows a crop to achieve 90 per cent of its maximum yield and the critical concentration for toxicity is defined as the concentration in a diagnostic tissue above which yield is decreased by more than 10 per cent.

The determination of critical level of nutrient in soil and plant helps to manage the nutrient deficiency and avoid crop loss. A graphical approach to determine the critical limit of nutrient in soil and plant was proposed by Cate and Nelson (1965). The critical soil Mg concentration for maximum crop yield is higher in acid than in neutral soils (Ferrari and Sluisman, 1955). Adams and Henderson, (1962) reported that the critical magnesium saturation levels are in between 5-10 per cent of CEC of the soil. The critical soil magnesium levels, for better performance of plants, will range from

25 to 100 mg Mg kg^{-1} soil, which depend on the soil and soil testing procedure used (Nelson and Jones, 1972).

Critical limit of Mg for higher plants vary with the plant part and stages of development. Generally, the concentration of magnesium in plant leaves below 0.2% are considered as deficient and above 0.4% Mg as sufficient. Critical magnesium concentration is lower in the case of monocotyledonous plants compared to dicotyledonous plants. Hailes *et al.* (1997) derived the critical soil test values for 90% relative yield of maize in strongly acidic soils to be 0·21 cmol(+) kg^{-1} of exchangeable Mg or 7% Mg saturation of CEC.

Kasinath *et al.* (2014) found the critical limit of magnesium in soil and plant to be 74 mg kg^{-1} and 0.39% respectively for tomato crop in Alfisols of Southern Karnataka. Venkatesh *et al.* (2018) also worked on 30 soils from Imphal west district having a mean pH and CEC value of 5.43 and 16.28 cmol (p+) kg^{-1} respectively and determined critical limit of available Mg as 164 mg kg^{-1} for soil and 0.12% for 45 days old green gram plants. According to KAU (2018) the critical level of available magnesium in soil for yard long bean is 125 mg kg^{-1}. Yield improvement up to 22% could be realized by the application of magnesium through magnesium sulphate at the rate of 60 -100 kg ha^{-1} in lateritic soils of Kerala.

2.12. Magnesium fertilization

Amount of magnesium released from weathering of primary and secondary minerals may not be sufficient for plant nutrition. Even though soil solution concentration is high, low soil pH, drought and high content of competing cations in the soil solution limits magnesium availability which necessitates the external application to meet crop demand. According to Metson (1974) magnesium deficiency could be averted by the liberal use of farmyard manure (with around 0.34 per cent Mg

on a dry-weight basis), or by using magnesium fertilisers such as magnesian limestone, kieserite, epsom salts, or calcined magnesite.

Common magnesium fertilizers are distinguished into soluble and semi-soluble sources. Minerals like dolomite and magnesite comes under semi- soluble sources while kieserite (includes magnesium sulphate monohydrate and magnesium sulphate hepta hydrate) and magnesium nitrate makes soluble source of magnesium. Kieserite application increased the exchangeable Mg much larger compared to magnesium oxide. Application of magnesium fertilizers such as magnesium carbonate, calcined magnesium carbonate and magnesium oxide increases the available magnesium status in soils (Heming and Hollis, 1995).

Water solubility of magnesium fertilizers are determined by the chemical composition (oxide, sulfate, carbonate, nitrate, chloride, phosphate or silicate) of the fertilizers and its availability to plants depends on solubility and particle size of mineral fertilizers (Mayland and Wilkinson, 1989). Magnesium sulphate is completely soluble and forms a best source of Mg in deficient soils. Synthetic magnesium sulphate produced by reacting magnesium oxide and sulphuric acid (Hardter, et al. 2004) are also commercially available now. Senbayram, et al. (2015) reviewed the solubility of different magnesium minerals at 20°C and found that kieserite have highest solubility followed by struvite, magnesite and dolomite.

Unlike other basic cations magnesium is less strongly bound to soil charges and have high mobility in soils causing high risk of leaching losses. Application of soluble fertilizers like kieserite and magnesium sulphate poses problems related to leaching especially when applied to sandy soils having high hydraulic conductivity and lateritic soils having low cation exchange capacity. There arises the importance of slow release magnesium fertilizers like dolomite, magnesite and calcined magnesite which helps to mitigate risk of leaching losses and deliver sufficient quantities as per requirement

(Hardter *et al*. 2004). The efficiency of slow release fertilizers are slightly high in acidic soils when applied as ground form (Heming and Hollis, 1995).

Materials and Methods

3. MATERIALS AND METHODS

The present study entitled "Optimization of soil environment and crop response for magnesium nutrition in Ultisol" was carried out at Radiotracer Laboratory, College of Horticulture, Kerala Agricultural University during 2017-2019. Two experiments comprising of an incubation study and a pot culture study with cowpea as test crop were conducted to accomplish the objectives. The materials used and the methods adopted are summarised below.

3.1. Collection and characterization of soil

Top soil from 0-15 cm depth representing Ultisols was collected from Water Management Research Unit, Vellanikkara to conduct the incubation study and pot culture experiment. A fresh sample of the soil was taken for the microbiological analysis and the remaining soil was air dried, ground with wooden mortar and pestle and sieved through 2 mm sieve.

3.1.1. Analysis of physico-chemical properties

A representative sample was taken and characterized for various physico-chemical properties following standard procedures given in Table 3.1.

Table 3.1. Procedures followed for physico-chemical characterization of soil

Parameter	Value	Method used for extraction and estimation	Reference
1. Physical properties			
Sand (%)	46.90	International pipette method	Robinson (1922)
Silt (%)	11.60		
Clay (%)	40.30		
Texture	Sandy clay		

Parameter	Value	Method used for extraction and estimation	Reference
2. Chemical properties			
pH	4.70	The H^+ ion activity in 1: 2.5 soil water homogeneous suspension was measured potentiometrically using pH meter	Jackson (1958)
Electrical Conductivity (EC) (dS m^{-1})	0.07	The electrical conductivity (EC) of the supernatant solution after pH measurement was measured using conductivity meter	Jackson (1958)
Organic carbon (OC) (%)	1.32	Wet oxidation method	Walkley and Black (1934)
Available nitrogen (Av. N) (kg ha^{-1})	476.67	Alkaline permanganometry	Subbiah and Asija (1956)
Available phosphorus (Av. P) (kg ha^{-1})	98.04	Bray No.1 extraction and colorimetric estimation of intensity of blue color complex developed by reduced molybdate ascorbic acid at 660 nm	Bray and Kurtz (1945) Watanabe and Olsen (1965)
Available potassium (Av. K) (kg ha^{-1})	240.18	Extraction with neutral normal ammonium acetate and estimation using flame photometer	Jackson (1958)
Available calcium (Av. Ca) (mg kg^{-1})	429.30	Extraction with neutral normal ammonium acetate and estimation using atomic absorption spectrometer	
Available magnesium (Av. Mg) (mg kg^{-1})	64.53		
Available sulphur (Av. S) (mg kg^{-1})	5.00	Extracted with 0.15% $CaCl_2$ and turbidimetric estimation using spectrophotometer at 440 nm	Tabatabai (1982) Massoumi and Cornfield (1963)

Parameter	Value	Method used for extraction and estimation	Reference
Available iron (Av. Fe) (mg kg^{-1})	12.41	Extraction with 0.1M HCl and estimation using atomic absorption spectrophotometer	Sims and Johnson (1991)
Available manganese (Av. Mn) (mg kg^{-1})	16.26		
Available zinc (Av. Zn) (mg kg^{-1})	3.81		
Available copper (Av. Cu) (mg kg^{-1})	8.08		
Available boron (Av. B) (mg kg^{-1})	0.24	Hot water extractable boron determined colorimetrically by using azomethine-H reagent at 420 nm	Berger and Troug (1939) Gupta (1972)
Effective cation exchange capacity (cmol(+)kg^{-1})	5.63	Displacement of exchangeable cations with 0.1M Barium chloride and summation of the cations in the extract estimated by atomic absorption spectrophotometer	Hendershot and Duquette (1986)

3.1.2. Microbial population in soil

3.1.2.1. Population of Rhizobium

Serial dilution and plating method using selective media, Yeast extract mannitol agar media with congo-red dye. Serial dilution was done up to 10^{-5} and pour plate method was used for plating.

3.1.2.2. Population of free living nitrogen fixing bacteria

Serial dilution and plating method in Jenson media (selective media for free living nitrogen fixing microorganisms). Serial dilution was done up to 10^{-6} and pour plate method was used for plating.

3.1.2.3. Spore count of Arbuscular Mycorrhizal Fungi

Spore count of AMF was determined in the rhizosphere soil of plants using wet sieving and decanting method (Gerdmann and Nicolson, 1963). Hundred gram of soil sample was taken 1litre beaker and made up to 1000ml with water. Stirred well, heavier particles were allowed to settle for a few seconds. Then the suspension was passed through a series of different size sieves (250µm, 106 µm, 75 µm, 45 µm, 37 µm) arranged in descending order of their mesh size. Again water was added to 1000ml, stirred well and allowed for few seconds and this procedure was repeated for 5-6 times, till the suspension appeared clear. Seivates were collected from each sieve separately in beakers. Supernatant from each beaker was separately filtered through Whatman No.1 filter paper and the content of the filter papers were examined for spores under stereo zoom microscope (LABOMED).

3.2. Experiment 1: Incubation Experiment

An incubation experiment was conducted with lateritic soil to study the release pattern of magnesium from magnesium carbonate. Different levels of magnesium carbonate was added to 1 kg of soil with the addition of magnesium sufficient to theoretically raise the available magnesium status to 120 mg kg^{-1} as the optimum dose and one level above and one level below the optimum dose was added with and without the addition of recommended dose of calcium carbonate and organic manure. The details of the treatments applied are given below.

3.2.1. Treatments details:

Levels of organic manure: 2

O_0: 0 t ha^{-1}
O_1: 20 t ha^{-1}

Levels of calcium carbonate: 2

L_0: 0 kg ha^{-1}

L_1: 250 kg ha^{-1}

Levels of magnesium carbonate: 3

M_1: 50 per cent of optimum dose

M_2: Optimum dose

M_3: 150 per cent of optimum dose

Design: CRD

No. of treatments: 2x2x3=12

No. of replications: 3

The available magnesium content in the soil was 64 mg kg^{-1} soil. Thus the amount of magnesium carbonate (AR grade; 28.82 % Mg) required to raise available magnesium status to 120 mg kg^{-1} of soil was 0.1943g. The characteristics of organic manure is given in Table 3.2. The quantity of organic manure (vermicompost- ground and sieved through 0.5 mm sieve), calcium carbonate (AR grade) and magnesium carbonate supplied to 1kg soil as per the treatment combinations are given in Table 3.3.

Table 3.2. Characteristics of organic manure

Parameters	Content	Procedure
pH	7.10	FCO (1985)
EC (dS m^{-1})	0.81	
Nitrogen (%)	1.79	As described in table 3.5 for plant analysis
Phosphorus (%)	0.30	
Potassium (%)	0.61	
Calcium (%)	1.97	

43

Parameters	Content	Procedure
Magnesium (%)	0.28	As described in table 3.5 for plant analysis
Sulphur (%)	0.25	
Iron (mg kg^{-1})	1000.00	
Manganese (mg kg^{-1})	290.60	
Zinc (mg kg^{-1})	80.50	
Copper (mg kg^{-1})	24.00	
Boron (mg kg^{-1})	64.40	

Table 3.3. Treatment combinations of incubation experiment

Treatments	Treatment combination	Organic manure (O) (g kg^{-1})	Calcium carbonate (L) (g kg^{-1})	Magnesium carbonate (M) (g kg^{-1})
T_1	$O_0L_0M_1$	0	0	0.0972
T_2	$O_0L_0M_2$	0	0	0.1943
T_3	$O_0L_0M_3$	0	0	0.2915
T_4	$O_0L_1M_1$	0	0.1116	0.0972
T_5	$O_0L_1M_2$	0	0.1116	0.1943
T_6	$O_0L_1M_3$	0	0.1116	0.2915
T_7	$O_1L_0M_1$	8.93	0	0.0972
T_8	$O_1L_0M_2$	8.93	0	0.1943
T_9	$O_1L_0M_3$	8.93	0	0.2915
T_{10}	$O_1L_1M_1$	8.93	0.1116	0.0972
T_{11}	$O_1L_1M_2$	8.93	0.1116	0.1943
T_{12}	$O_1L_1M_3$	8.93	0.1116	0.2915

Treatments were applied to 1kg soil and mixed thoroughly to ascertain uniform dispersion of the inputs. The incubation study was carried out for four months (16 weeks). Soil in the plastic jars were maintained at field capacity and mixed thoroughly over the incubation period.

Representative samples were taken at weekly intervals and analyzed for pH, EC and available magnesium. The fractions of magnesium was determined initially and after the completion of incubation study. The procedure followed for the sequential fractionation of magnesium is detailed below.

3.2.2. Fractionation of magnesium in soil

The fractionation of soil magnesium into exchangeable, organic-complexed, acid soluble and mineral forms was done as per the procedure outlined by Mokwunye and Melsted (1972). Fractionation scheme followed for the extraction of magnesium fractions is as detailed below. The soil samples were thoroughly ground and passed through 0.417 mm sieve.

3.2.2.1. Exchangeable fraction

Exchangeable fraction was extracted by shaking 1g of soil sample with 20 mL neutral 1N ammonium acetate for 45 min followed by centrifuging for 10 minutes at 2000 rpm and decanting the supernatant. Additional 20 mL aliquots of ammonium acetate were used with 10 minute shaking periods followed by centrifuging until a total of 100 mL of the supernatant solution was collected.

3.2.2.2. Organic-complexed fraction

The residue after extracting exchangeable fraction was oxidized with 10% H_2O_2 solution as per the procedure of Jackson (1958). Ten mL of H_2O_2 solution was added until foaming ceased and the excess of H_2O_2 in the mixture was removed by heating in a water bath. The oxidized mixture was centrifuged, decanted into 100 mL volumetric

flask. The residue was washed consecutively using IN neutral ammonium acetate until 100 mL of the supernatant solution was obtained.

3.2.2.3. Acid soluble fraction

The residue after extracting organic-complexed fraction was treated with 30 mL 1N HNO_3 following the procedure of Rouse and Bertramson (1949). The mixture was gently boiled for 15 minutes on an electric hot plate and filtered through Whatmann No. 42 filter paper. The residue was then washed with aliquots of 0.2 N HNO_3 until a total of 100mL of extract was obtained.

3.2.2.4. Mineral fraction

The residue from the previous extraction including the filter paper was transferred to a 250 mL beaker. The fraction associated with primary minerals were extracted by digesting the mixture with 25 mL of tri-acid mixture (23 parts of concentrated HNO_3, 23 parts of 85% H_3PO_4, and 54 parts of 70% $HClO_4$) on an electric hot plate under a hood until completely decomposed. The digested mixture was cooled to which 5 mL of 5N HCl was added and the mixture filtered and washed with distilled water until 100 mL of filtrate was collected.

3.2.2.5. Water soluble fraction

Water soluble fraction was estimated by the modified procedure given by Baruah *et al.* (2011) where five grams (2mm sieved) of soil sample with 25 mL deionized water was centrifuged at 4000 rpm for 30 minutes, decanted and residue was rinsed with 25 mL of deionized water followed by shaking, centrifugation and filtration.

3.2.2.6. Total content

Total content of magnesium was determined on a separate sample by digesting one gram of soil sample with 70% $HClO_4$ following the procedure of Jackson (1958).

On cooling, 5 mL of 5N HCl was added to the digested mixture followed by filtration and washing with distilled water until 100 mL of filtrate was collected.

The different fractions of Mg were analyzed using ICP-OES (PerkinElmer - model Optima 8000).

Fig. 3.1. Fractionation scheme of magnesium

48
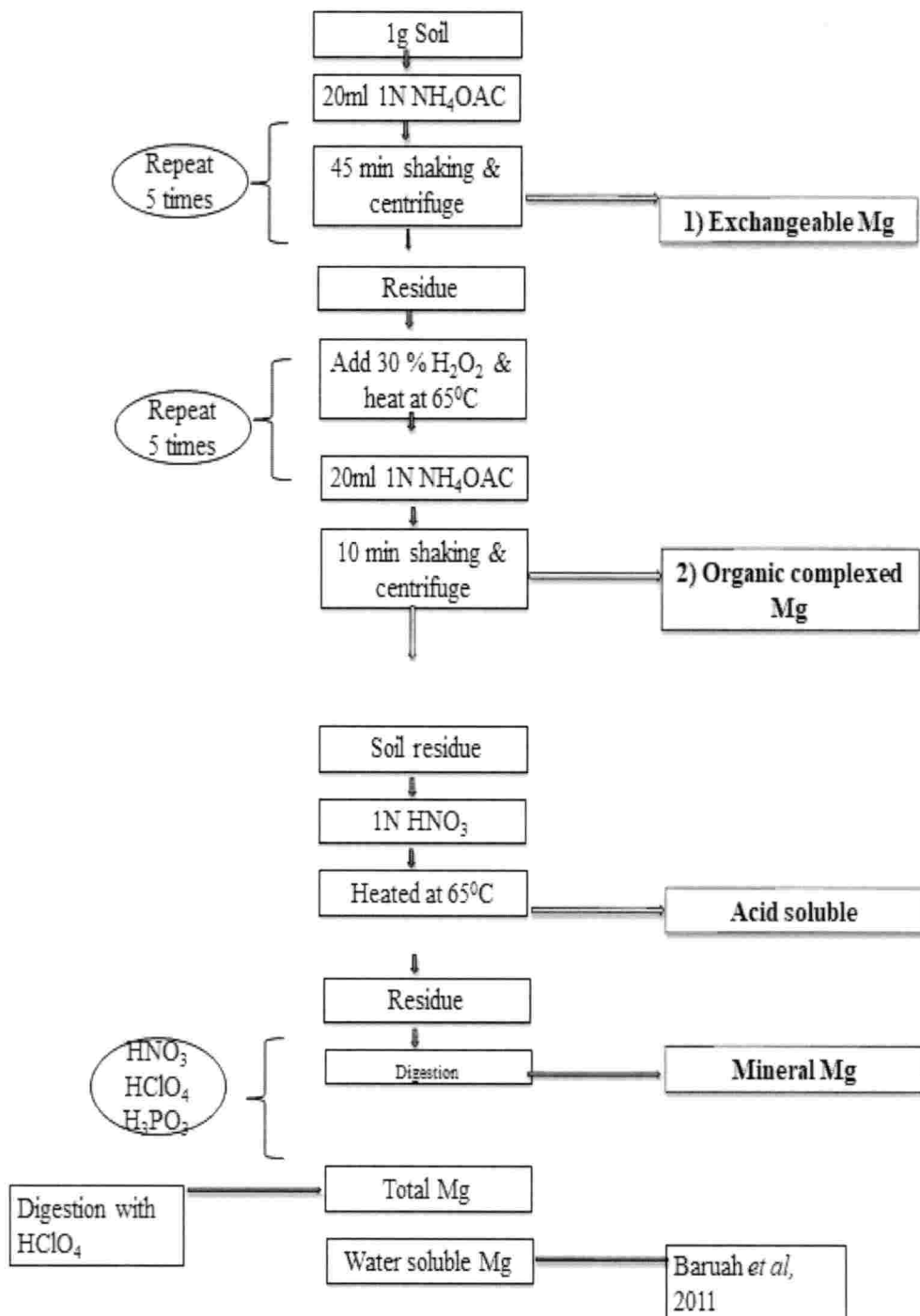
3.3. Experiment 2: Pot culture experiment

A pot culture experiment to investigate the response of cowpea to magnesium nutrition and to validate critical level of magnesium in soil and plant using lateritic soil was conducted at Radiotracer Laboratory, Kerala Agricultural University. The details of the experiment conducted are as follows:

Crop   : Cowpea
Variety   : Bhagyalakshmi
Design   : CRD
Treatments   : 12
Replications   : 4

3.3.1. The treatment details are as follows:

T_1: Absolute control

T_2: Organic manure @ 20 t ha^{-1}

T_3: POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}

T_4: POP recommendation with dolomite @ 400 kg ha^{-1}

T_5: T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}

T_6: T_3+ $MgCO_3$ @ 10 % of T_5

T_7: T_3+ $MgCO_3$ @ 20 % of T_5

T_8: T_3+ $MgCO_3$ @ 40 % of T_5

T_9: T_3+ $MgCO_3$ @ 60 % of T_5

T_{10}: T_3+ $MgCO_3$ @ 80 % of T_5

T_{11}: T_3+ $MgCO_3$ @ 125 % of T_5

T_{12}: T_3+ $MgCO_3$ @ 150 % of T_5

Number of pots per treatment: 5

Quantity of soil per pot: 5 kg

Number of plants per pot: 1

3.3.2. Filling of pots

Earthen pots of 5 kg capacity were filled with 2 mm sieved soil and then treatments were applied accordingly.

3.3.3. Application of lime and organic manure

Calcium carbonate (AR grade) was applied @ 250 kg ha^{-1} to the treatments T_3 to T_{12} excepting T_4 where dolomite was applied as the liming material. The quantity of dolomite applied was corrected for its neutralizing value (106.32 %). Organic manure in the form of vermicompost was applied @ 20 t ha^{-1} after one week of lime application. Different doses of magnesium carbonate was applied two weeks after organic manure application.

3.3.4. Application of magnesium sources

Based on the initial characterization of soil, $MgCO_3$ required to raise available magnesium to 120 mg kg^{-1} was determined and the quantities were modified based on the different treatments (Table 3.4).

3.3.5. Variety

Bhagyalakshmi, a dwarf bush type vegetable cowpea variety was used in this study. It takes 41 days to flowering and produces white colored flowers and bold pods.

3.3.6. Seed treatment

Presoaked cowpea seeds were treated with Rhizobium culture at the rate of 100 g kg^{-1} before sowing. The treated seeds were allowed to air dry for some time before sowing.

Table 3.4. Treatments applied in pot culture experiment

Treatments	Description of treatments	Quantity of inputs (g) added per pot		
		Organic manure	Calcite/ Dolomite	Magnesium carbonate
T_1	Absolute control	-	-	-
T_2	Organic manure @ 20 t ha^{-1}	44.64	-	-
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	44.64	0.5580	-
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	44.64	0.9153	-
T_5	T_3+MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	44.64	0.5580	0.9715
T_6	T_3+ MgCO$_3$ @10% of T_5	44.64	0.5580	0.0971
T_7	T_3+ MgCO$_3$ @ 20% of T_5	44.64	0.5580	0.1942
T_8	T_3+ MgCO$_3$ @ 40% of T_5	44.64	0.5580	0.3884
T_9	T_3+ MgCO$_3$ @ 60% of T_5	44.64	0.5580	0.5828
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	44.64	0.5580	0.7768
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	44.64	0.5580	1.2141
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	44.64	0.5580	1.4570

3.3.7. Crop culture

Rhizobium treated seeds were sown at the rate of three seeds per pot and sufficient irrigation was given thereafter. One week after emergence one healthy seedling was maintained in each pot. Fertilizers were applied as per the Package of Practices Recommendations - Crops, KAU (2016) modified based on soil test results (Table 3.5). The recommended dose of fertilizers as per the Package of Practices of KAU includes the application of 20t ha^{-1} of organic manure, 250 kg ha^{-1} calcium

carbonate or 400 kg ha^{-1} dolomite and 20: 30: 10 kg ha^{-1} of N, P_2O_5 and K_2O. Complete dose of phosphorus, potassium and half split of nitrogen was given after thinning of plant population. Second dose of nitrogen was applied after 15 days. The nutrients were supplied through water soluble sources. Irrigation with de-ionized water, weed control and plant protection measures were adopted uniformly in each pot.

Table 3.5. Quantity of fertilizers added to each pot as per soil test results

Initial status of available nutrients	Value	Rating	Modified POP recommendation of 20:30:10	Quantity of fertilizers added per pot
Organic carbon (%)	1.32	Medium	78 % N- 15.6 kg ha^{-1}	0.076 g urea
Phosphorus (kg ha^{-1})	98.04	High	25 % P_2O_5 -7.5 kg ha^{-1}	0.032 g KH_2PO_4
Potassium (kg ha^{-1})	240.18	Medium	60% K_2O - 6 kg ha^{-1}	0.0043 g K_2SO_4
Calcium (mg kg^{-1})	429.30	Sufficient	As per treatments	
Magnesium (mg kg^{-1})	64.53	Deficient	As per treatments	
Sulphur (mg kg^{-1})	5.0	Sufficient	-	
Iron (mg kg^{-1})	12.41	Sufficient	-	
Manganese (mg kg^{-1})	16.26	Sufficient	-	
Zinc (mg kg^{-1})	3.81	Sufficient	-	
Copper (mg kg^{-1})	8.08	Sufficient	-	
Boron (mg kg^{-1})	0.24	Deficient	10 kg borax ha^{-1}	0.0111g Solubor

3.3.8. Biometric observations

1. Days to germinations
2. Plant height (cm)
3. Number of branches per plant
4. Number of pods per plant
5. Length of pods (cm)

6. Number of seeds per pod
7. Yield per plant (g)
8. Root nodules per plant at flowering and after harvest

3.3.8. Soil analysis

Soil samples drawn from each treatment during flowering and harvest were air dried under shade and analyzed for pH, EC, Organic carbon, available N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu and B as per the procedure given in Table 3.1.

3.3.8.1. Microbial population in soil

Population of Rhizobium, free-living nitrogen fixing bacteria and total spore count of arbuscular mycorrhizal fungi in soil was taken during flowering and harvest as per the procedure in section 3.1.2.

3.3.8.1.1. Per cent root colonization of AMF

The roots used to measure AMF colonization were cut into approximately 1 cm length and were cleared in 10% (w/ v) KOH at 90 °C in a water bath for 60 minutes or 121^0C for 10 minutes to remove host cytoplasm and nuclei. Remove the KOH and rinse with water to remove KOH completely. Followed by acidifying with 1 per cent HCl for 10 minutes to neutralize the extreme KOH. Then root bits were stained with 0.05% (w/v) Trypan blue in lactophenol and heated gently for 10 minutes. The excess stain was removed by lactophenol and 30 root segments of each sample were examined microscope for per centage of root length colonized by Arbuscular Mycorrhizal Fungi (Philips and Haymen, 1970).

Per cent root colonization = No. of root segments colonized x100

Total no. of root segments examined

39

3.3.9. Plant analysis

The plant samples representing various treatments were collected during flowering and harvest. Samples were drawn at flowering by destructive sampling of two replications. Leaves, stem and pods were separated and washed twice with tap water followed by rinsing with 0.2 per cent detergent solution and later with 0.1N HCl and finally with distilled water. Samples were dried to constant weight at 65^0 C in hot air oven. Finally the samples were ground thoroughly in mixer grinder and stored in moisture free condition.

3.3.9.1. Chlorophyll content in cowpea leaves

Chlorophyll a and chlorophyll b in index leaves of cowpea was determined during flowering and harvest of the crop as per the procedure given by Hiscox and Israelstem (1979). DMSO (Di methyl sulphoxide- 10 mL) was added to 100 mg of leaf sample in a test tube and kept overnight in dark. The sample was later filtered and final volume made up to 25mL. The intensity of green colour was measured at two wavelengths *viz.*, 645nm and 663nm (model: Systronix 128). The content of chlorophyll was determined from the following formulae and expressed as mg g^{-1} of fresh weight of plant tissue.

Chlorohyll a   = [(12.7x A663) – (2.69x A645)] x (volume/1000 x weight)

Chlorophyll b   = [(22.9x A645) – (4.68x A663)] x (volume/1000 x weight)

Where, A663 and A645 indicate absorbance at the corresponding wavelength

3.3.8.2. Analysis of plant nutrient content

Plant nutrient content was determined following the procedures given in Table 3.6. Ground plant samples were digested with di- acid mixture (9: 4 nitric acid and perchloric acid) in a hot plate under fume hood. Digested samples were filtered and made up to 100mL and used for the analysis of different elements.

Table 3.6. Methods used in analysis of plant nutrient content

Sl No.	Element	Method
1	Nitrogen	Nitrogen content in ground plant sample was analyzed in CHNS Analyzer (Elemeter Vario EL Cube)
2	Phosphorus	Di-acid digestion of leaf, stem and pod samples followed by filtration (Piper, 1966). Intensity of yellow coloured vanadomolybdate complex was determined colorimetrically at 420 nm (Spectrophotometer model: Systronics 169).
3	Potassium	Di-acid digestion of leaf, stem and pod sample followed by filtration. The content of potassium was determined by flame photometer.
4	Calcium and Magnesium	Di-acid digestion of leaf, stem and pod sample followed by filtration. The content in the filtrate was determined by Atomic Absorption Spectrophotometer (Model: Perkin Elmer-PinAAcle 500).
5	Sulphur	Sulphur content in leaf, stem and pod was analyzed in CHNS Analyzer (Elemeter Vario EL Cube).
6	Micronutrients (Fe, Mn, Zn, Cu)	Di-acid digestion of leaf, stem and pod sample followed by filtration. The content in the filtrate was determined by Atomic Absorption Spectrophotometer (Model: Perkin Elmer-PinAAcle 500).
7	Boron	Determined by dry ashing of plant sample (Gaines and Mitchell, 1979) and then analysed colorimetrically by Azomethine-H (Spectrophotometer model: Systronics 169) (Bingham, 1982).

3.3.10. Critical level in plant and soil

Cate and Nelson (1965) demonstrated that scatter points on a graphical plot of relative yield (Y- axis) and soil or plant nutrient content (X- axis) gives critical limit of the nutrient in soil or plant. In order to find the critical limit the scatter points are divided into two populations graphically by using Olmstead and Tukey's nonparametric test of association (1947). Two perpendicular lines were drawn to produce four quadrants having same relative size so that maximum number of points fall in positive quadrants and fewest in negative quadrants. The intercept of vertical line on X-axis gives the critical level of a nutrient in soil or plant. The relative yield is calculated as

Relative per cent yield = $\dfrac{\text{Yield at each level of a nutrient}}{\text{Maximum yield achieved}} \times 100$

As the nutrient composition of a plant changes with age, the critical levels are defined for a specific stage of maturity. Flower initiation is taken as the diagnostic stage of growth for plant leaf analysis of cowpea (Nelson, 1989: Fageria, 2009).

3.4. Statistical Analysis

The data obtained from the incubation experiment was analyzed as factorial CRD with the treatments imposed and incubation time as main factors. Analysis of variance in CRD was done using OPSTAT software package (Sheoran *et al.*, 1998) in pot culture experiment. Duncan's multiple range test was employed to test the significance of difference between means of treatments.

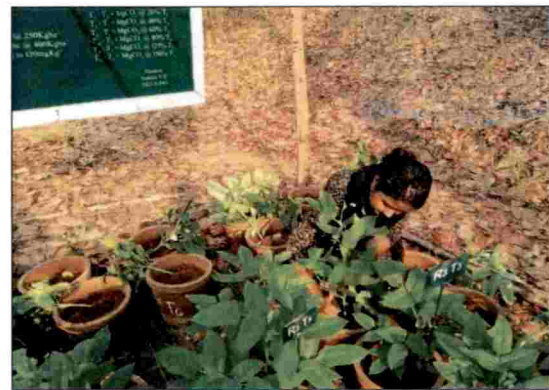Plate 1. Experimental view of incubation study

Plate 2. Over view of pot culture experiment

Results

4. RESULT

Results obtained from the present study entitled "Optimization of soil environment and crop response for magnesium nutrition in Ultisol" are documented in this chapter.

4.1. Experiment 1. Incubation experiment

An incubation experiment was conducted with lateritic soil (Ultisol) collected from Water Management Research Unit, Vellanikkara to study the release pattern of magnesium from magnesium carbonate. Different levels of magnesium carbonate was added to 1kg soil considering the addition of magnesium sufficient to theoretically raise the available magnesium status to 120 mg kg^{-1} as the optimum dose and one level above and one level below the optimum dose, with and without the addition of recommended dose of calcium carbonate (250 kg ha^{-1}) and organic manure (20 t ha^{-1}). The dose of $MgCO_3$ required to raise available magnesium to 120 mg kg^{-1} was determined based on the initial characterization of soil.

Soil was maintained at field capacity and soil pH, EC and available magnesium was determined in the samples drawn at weekly intervals. The data generated were analyzed for variance as factorial CRD with the treatments imposed (T) and the time interval (W) as main factors.

4.1.1. Effect of treatments on pH of soil

The data on soil pH at weekly intervals of incubation with different levels of calcium carbonate, organic matter and magnesium carbonate for sixteen weeks are presented in Table 4.1. The initial status of soil pH was 4.70. The treatments imposed could produce significant changes in soil pH over the period of time. The treatment T_{12} with the addition of calcium carbonate (250 kg ha^{-1}), organic manure (20 t ha^{-1}) and magnesium carbonate at 150% of the optimum dose required to theoretically raise the available magnesium status to 120 mg kg^{-1} recorded significantly higher soil pH (5.14)

and the lowest pH was recorded by T_1 (treated with lowest dose of magnesium carbonate along with no application of calcium carbonate and organic manure). The effect of incubation period on soil pH revealed significantly higher soil pH (5.06) at one week after incubation.

The interaction effect of treatments and period of incubation showed that significantly higher soil pH was recorded in treatment T_{12}, one week after incubation (5.32).

4.1.2. Effect of treatments on electrical conductivity of soil

The data on soil EC at weekly intervals of incubation with different levels of calcium carbonate, organic matter and magnesium carbonate for sixteen weeks are presented in Table 4.2. The initial status of soil EC was 0.07 dS m^{-1}. The treatments imposed produced significant changes in soil EC over the period of time. The treatment T_{12} ($O_1L_1M_3$) with the addition of calcium carbonate, organic manure and magnesium carbonate at 150% of the optimum dose required to theoretically raise the available magnesium status to 120 mg kg^{-1} recorded significantly higher (0.18 dS m^{-1}) soil EC. Effect of incubation period on soil EC revealed significantly higher EC (0.20 dS m^{-1}) at the end of the incubation period.

The interaction effect of treatments and period of incubation showed significantly higher EC to be recorded in T_9 (0.25 dS m^{-1}) at sixteen weeks after incubation which was on par with EC recorded at thirteen weeks after incubation in treatment T_{12} (0.23 dS m^{-1}).

4.1.3. Effect of treatments on available magnesium content in soil

Available magnesium showed significant difference among treatments as per Table 4.3. The highest available Mg was recorded in T_{12} (142.07 mg kg^{-1}) with the addition of calcium carbonate, organic manure and magnesium carbonate at 150% of optimum dose required to theoretically raise the available Mg status to 120 mg kg^{-1} and

was on par with T_9 (141.64 mg kg^{-1}) with the addition of organic manure and magnesium carbonate at 150% of the optimum dose required to theoretically raise the available magnesium status to 120 mg kg^{-1} without the addition of calcium carbonate. The lowest value of available Mg was recorded by T_1 with a mean of 88.95 mg kg^{-1}.

On analyzing the effect of incubation period on available magnesium, highest content was recorded at eight weeks after incubation (133.89 mg kg^{-1}) which was on par with available magnesium content at seven and nine weeks after incubation. The interaction effect of treatments and period of incubation showed significantly higher available magnesium content in treatment T_{12} (170.26 mg kg^{-1}) at nine weeks after incubation.

4.1.4. Effect of treatments on fractions of magnesium in soil

The different fractions of magnesium were estimated in all the treatments after completion of the incubation experiment. The total magnesium in soil was partitioned into water soluble, exchangeable, organic-complexed, acid soluble and mineral fractions as per the procedure outlined in section 3.2.2. The fractions of magnesium in the initial soil sample was found in the order mineral Mg> exchangeable Mg> acid soluble Mg> organic complexed Mg> water soluble Mg. The treatments imposed were found to have significant influence on all the fractions of magnesium in soil. The effect of treatments on different fractions of magnesium are presented in Table 4.4.

Water soluble fraction was smallest of all the fractions of magnesium. The initial content of water soluble magnesium was 3.52 mg kg^{-1}. After the incubation experiment the water soluble fraction in soil ranged from 9.19 to 18.82 mg kg^{-1}. Significantly higher content of this fraction was recorded in treatment T_{12} with the treatment combination of $O_1L_1M_3$ and was on par with T_8 ($O_1L_0M_2$).

Table 4.1. Effect of treatments on soil pH during incubation

Treatments (T)		Weeks after incubation (W)																Mean
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	
T_1	$O_0L_0M_1$	4.82	4.81	4.81	4.75	4.71	4.73	4.80	4.78	4.82	4.65	4.79	4.81	4.72	4.72	4.74	4.73	4.76
T_2	$O_0L_0M_2$	4.94	4.85	4.86	4.83	4.75	4.82	4.88	4.86	4.84	4.79	4.81	4.84	4.77	4.74	4.79	4.77	4.82
T_3	$O_0L_0M_3$	5.02	4.87	4.95	4.82	4.82	4.96	4.91	4.92	4.88	4.89	4.92	4.87	4.87	4.85	4.82	4.87	4.89
T_4	$O_0L_1M_1$	4.83	4.77	4.86	4.81	4.77	4.96	4.82	4.81	4.85	4.75	4.81	4.82	4.77	4.73	4.84	4.75	4.81
T_5	$O_0L_1M_2$	5.04	4.81	4.93	4.87	4.86	4.83	4.88	5.02	4.93	4.76	4.84	4.90	4.84	4.82	4.86	4.77	4.87
T_6	$O_0L_1M_3$	5.12	4.95	5.06	4.90	5.04	4.95	4.90	5.01	4.97	4.93	4.94	5.01	4.94	4.91	4.93	4.86	4.96
T_7	$O_1L_0M_1$	4.88	4.85	4.93	4.94	4.85	4.85	4.82	4.93	4.93	4.83	4.85	4.88	4.82	4.84	4.74	4.83	4.86
T_8	$O_1L_0M_2$	5.11	4.92	5.01	4.94	5.01	4.94	4.92	5.02	5.06	4.80	4.96	4.94	4.88	4.90	4.91	4.93	4.95
T_9	$O_1L_0M_3$	5.16	5.07	5.09	4.98	5.06	4.98	5.02	5.06	5.07	5.03	5.02	5.03	5.05	5.03	4.96	5.08	5.04
T_{10}	$O_1L_1M_1$	5.24	4.95	5.09	4.95	5.00	5.02	5.00	5.04	5.08	4.99	4.96	5.04	4.95	4.91	4.86	4.94	5.00
T_{11}	$O_1L_1M_2$	5.24	5.04	5.13	5.01	5.13	5.09	5.05	5.09	5.11	5.03	5.05	5.06	4.97	5.01	4.96	5.03	5.06
T_{12}	$O_1L_1M_3$	5.32	5.12	5.24	5.15	5.14	5.13	5.10	5.14	5.15	5.11	5.12	5.09	5.12	5.08	5.16	5.10	5.14
	Mean	5.06	4.91	4.99	4.91	4.93	4.94	4.92	4.97	4.97	4.88	4.92	4.94	4.89	4.88	4.88	4.89	

CD-T (0.05) -0.01 CD-W(0.05) -0.011 CD – T x W (0.05) -0.039

Table 4.2. Effect of treatments on electrical conductivity (dS m^{-1}) of soil during incubation

Treatments (T)		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	Mean
T$_1$	O$_0$L$_0$M$_1$	0.11	0.09	0.10	0.10	0.12	0.14	0.12	0.12	0.14	0.14	0.14	0.18	0.16	0.17	0.16	0.16	**0.13**
T$_2$	O$_0$L$_0$M$_2$	0.10	0.09	0.10	0.11	0.13	0.14	0.14	0.14	0.14	0.15	0.16	0.13	0.18	0.16	0.14	0.20	**0.14**
T$_3$	O$_0$L$_0$M$_3$	0.10	0.10	0.10	0.12	0.13	0.13	0.11	0.13	0.14	0.14	0.14	0.14	0.17	0.16	0.18	0.17	**0.13**
T$_4$	O$_0$L$_1$M$_1$	0.10	0.09	0.10	0.11	0.12	0.11	0.14	0.13	0.14	0.15	0.15	0.14	0.15	0.15	0.16	0.17	**0.13**
T$_5$	O$_0$L$_1$M$_2$	0.11	0.09	0.10	0.11	0.12	0.13	0.13	0.13	0.12	0.14	0.16	0.13	0.18	0.16	0.17	0.17	**0.13**
T$_6$	O$_0$L$_1$M$_3$	0.11	0.10	0.11	0.12	0.12	0.13	0.12	0.13	0.14	0.15	0.15	0.15	0.17	0.15	0.19	0.19	**0.14**
T$_7$	O$_1$L$_0$M$_1$	0.12	0.12	0.13	0.13	0.14	0.15	0.16	0.15	0.17	0.17	0.18	0.16	0.19	0.15	0.18	0.21	**0.16**
T$_8$	O$_1$L$_0$M$_2$	0.13	0.14	0.12	0.14	0.14	0.16	0.15	0.17	0.17	0.19	0.20	0.18	0.21	0.19	0.17	0.22	**0.17**
T$_9$	O$_1$L$_0$M$_3$	0.13	0.13	0.13	0.14	0.16	0.16	0.16	0.17	0.19	0.19	0.19	0.17	0.19	0.17	0.21	0.25	**0.17**
T$_{10}$	O$_1$L$_1$M$_1$	0.13	0.13	0.13	0.15	0.16	0.16	0.17	0.17	0.19	0.19	0.17	0.18	0.21	0.21	0.19	0.21	**0.17**
T$_{11}$	O$_1$L$_1$M$_2$	0.12	0.12	0.13	0.14	0.15	0.15	0.16	0.16	0.16	0.19	0.17	0.17	0.19	0.20	0.21	0.22	**0.16**
T$_{12}$	O$_1$L$_1$M$_3$	0.13	0.12	0.14	0.15	0.16	0.17	0.18	0.17	0.19	0.19	0.19	0.19	0.23	0.21	0.20	0.22	**0.18**
	Mean	0.11	0.11	0.11	0.13	0.13	0.14	0.14	0.15	0.16	0.17	0.16	0.16	0.18	0.17	0.18	0.20	

CD (0.05)- T - 0.006   CD (0.05)-W-0.007   CD (0.05) T x W- 0.024

47

64

Table 4.3. Effect of treatments on available magnesium (mg kg^{-1}) in soil during incubation

Treatments (T)	Weeks after incubation (W)															Mean
	1	2	3	4	5	6	7	8	9	10	11	12	13	15	16	
T$_1$	81.90	96.68	81.05	83.56	81.61	81.35	107.60	112.26	96.36	78.86	82.6	82.25	99.66	90.15	83.01	88.95
T$_2$	97.01	116.15	113.75	100.00	102.35	103.9	126.10	136.80	121.86	99.31	108.51	94.95	120.05	116.41	108.46	110.58
T$_3$	114.80	135.96	151.41	145.56	142.01	113.81	133.20	150.80	136.71	109.21	116.05	114.45	129.12	131.31	137.98	130.21
T$_4$	76.11	103.25	88.86	85.71	87.00	82.46	105.15	111.16	102.30	83.71	89.10	88.10	101.75	88.31	95.21	92.22
T$_5$	92.55	113.40	98.96	97.56	97.20	112.75	125.35	127.66	126.16	98.06	106.06	103.90	121.55	110.91	108.30	108.90
T$_6$	111.48	142.61	133.65	125.9	116.16	121.51	144.05	147.85	152.51	114.91	128.15	120.88	144.38	131.05	141.80	131.57
T$_7$	91.61	112.25	102.15	94.00	86.51	97.20	115.00	104.95	105.60	89.16	100.70	92.81	104.75	94.40	82.85	98.27
T$_8$	111.63	141.55	124.35	113.85	110.35	109.91	131.11	138.40	135.56	112.71	129.30	121.50	128.88	127.60	117.40	123.30
T$_9$	130.75	158.13	148.10	132.51	140.26	135.01	148.96	158.88	166.91	123.16	132.15	133.20	143.11	141.05	136.70	141.64
T$_{10}$	95.90	108.76	104.11	92.80	94.05	97.11	117.71	116.90	107.55	97.56	93.40	94.61	107.88	106.45	101.40	102.11
T$_{11}$	111.01	137.76	136.76	110.45	104.65	118.40	143.06	138.10	137.85	103.43	116.76	115.46	129.61	124.86	124.66	123.13
T$_{12}$	117.41	155.26	153.80	138.11	132.60	131.51	157.31	163.00	170.26	123.26	138.10	136.6	140.63	132.26	142.95	142.07
Mean	102.68	126.81	119.74	110.00	107.89	108.74	129.55	133.89	129.97	102.78	111.74	108.22	122.61	116.23	115.06	

CD(0.05)- T- 1.075   CD(0.05)- W- 1.241   CD(0.05)- TxW- 4.299

Table 4.4. Effect of treatments on fractions of magnesium

Treat ments	Treatment combinations			WS Mg	Ex Mg	Or-c Mg	Ac-s Mg	Min Mg	Tot Mg
	Organic manure	Calcium carbonate	Magnesium Carbonate						
	(g kg^{-1})			(mg kg^{-1})					
T$_1$	0	0	0.0972	9.19^g	94.00^f	9.46^e	47.53^{cd}	986.77^d	1154.36^f
T$_2$	0	0	0.1943	14.43^{ef}	111.23^d	9.63^e	47.50^d	984.81^d	1175.01^{de}
T$_3$	0	0	0.2915	17.44^{bc}	124.03^c	10.03^e	47.33^{de}	992.87^{cd}	1186.68^d
T$_4$	0	0.1116	0.0972	13.88^f	102.83^e	10.10^{de}	47.63^{cd}	1001.03^{bc}	1182.87^{de}
T$_5$	0	0.1116	0.1943	14.02^{ef}	111.03^d	10.53^{cde}	44.30^{fg}	984.60^d	1171.88^e
T$_6$	0	0.1116	0.2915	14.14^{ef}	136.53^b	11.26^{bc}	45.50^{ef}	985.25^d	1186.68^d
T$_7$	8.93	0	0.0972	15.21^{de}	109.93^d	11.23^{bcd}	43.50^g	999.40^{bc}	1199.10^c
T$_8$	8.93	0	0.1943	17.58^{ab}	124.40^c	10.30^{cde}	44.93^{fg}	1009.27^b	1213.88^b
T$_9$	8.93	0	0.2915	16.50^{bc}	138.73^{ab}	11.30^{bc}	44.93^{fg}	1022.23^a	1241.03^a
T$_{10}$	8.93	0.1116	0.0972	17.37^{bc}	109.90^d	9.73^e	50.40^b	991.87^{cd}	1200.09^c
T$_{11}$	8.93	0.1116	0.1943	16.28^{cd}	118.06^c	12.30^{ab}	53.93^a	1005.91^b	1213.88^b
T$_{12}$	8.93	0.1116	0.2915	18.82^a	143.53^a	12.53^a	49.40^{bc}	1009.42^b	1241.10^a
Treatment means with common superscript do not differ significantly				3.52	77.30	9.00	38.33		
Initial soil fractions								981.930	1142.26

The exchangeable fraction of Mg (Ex-Mg) in soil increased from the initial value of 77.30 mg kg^{-1} and ranged from 94.00 mg kg^{-1} (T$_1$) to 143.53 mg kg^{-1} (T$_{12}$). The highest content of exchangeable magnesium was observed in T$_{12}$ (O$_1$L$_1$M$_3$) and was on par with T$_9$ (O$_1$L$_0$M$_3$). The organic-complexed (Or-c-Mg) fraction varied from 9.46 to 12.53 mg kg^{-1} compared to the initial status of 9.00 mg kg^{-1} and T$_{12}$ and T$_{11}$ were on par in recording higher content of this fraction. Significantly higher content of acid soluble (Ac-s-Mg) fraction was recorded in T$_{11}$ (O$_1$L$_1$M$_2$) and mineral fraction in T$_9$ (O$_1$L$_0$M$_3$). Higher content of acid soluble and mineral fraction (Min- Mg) was recorded in all treatments in comparison to the initial status.

The total magnesium content (Tot- Mg) in all the treatments were higher than the initial value of 1142.26 mg kg^{-1}. The highest content was observed in T$_{12}$ (1241.10 mg kg^{-1}).

4.2. Experiment 2. Pot culture experiment

The results obtained from pot culture experiment done with the objective to optimize magnesium nutrition for cowpea and to determine the critical level of Mg in soil and plant are presented here. Soil and plant samples were analyzed during flowering and after harvest and the results are summarized below.

4.2.1. Effect of treatments on soil properties during flowering and harvest of cowpea crop

Soil samples collected during flowering through destructive sampling of two replications and after harvest were analyzed for various parameters and results are presented below.

4.2.1.1. Electro-chemical properties and organic carbon (OC) content in soil
4.2.1.1.1. Flowering stage

The effect of treatments on electrochemical properties and organic carbon status in soil at flowering stage is presented in Table 4.5. The initial soil pH recorded was 4.70, while at flowering stage of the crop, the soil pH ranged from 4.75 to 5.20. Treatments imposed were found to have significant influence on soil

pH. Significantly higher soil pH of 5.20 was recorded in the treatment T_{12} (T_3+ $MgCO_3$ @ 150% of T_5), while the lowest pH was recorded in the absolute control treatment which was on par with T_2 where only organic manure was added.

Table 4.5. Effect of treatments on pH, EC and organic carbon of soil during flowering

	Treatments	pH	EC (dS m^{-1})	OC (%)
T_1	Absolute control	4.75^g	0.047^c	1.27^g
T_2	Organic manure @ 20 t ha^{-1}	4.76^g	0.046^c	2.51^a
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	4.88^f	0.059^b	2.54^a
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	4.88^{ef}	0.063^{ab}	2.54^a
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	4.92^{de}	0.064^b	2.23^b
T_6	T_3+ MgCO$_3$ @10% of T_5	4.94^d	0.059^b	2.02^c
T_7	T_3+ MgCO$_3$ @ 20% of T_5	4.96^d	0.062^{ab}	1.43^f
T_8	T_3+ MgCO$_3$ @ 40% of T_5	4.94^d	0.059^b	1.51^f
T_9	T_3+ MgCO$_3$ @ 60% of T_5	5.01^c	0.060^b	1.48^f
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	5.04^c	0.062^{ab}	1.87^d
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	5.10^b	0.068^a	1.77^{de}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	5.20^a	0.064^{ab}	1.74^e
Treatment means with common superscript do not differ significantly				

The initial soil EC was recorded as 0.07 dS m^{-1}. Highest EC was observed in T_{11} (0.068 dS m^{-1}) and was on par with T_{12} (0.064 dS m^{-1}), T_{10} (0.062 dS m^{-1}), T_4 (0.063 dS m^{-1}) and T_7 (0.062 dS m^{-1}). Lowest EC was recorded by absolute control (T_1) and was on par with T_2 (Table 4.5).

Significant difference between treatments were recorded in the organic carbon content in soil during flowering (Table 4.5). Organic carbon was significantly higher in treatment T_3 (2.54%) and T_4 (2.54%) which was on par with T_2 (2.51%) and the lowest value was observed in absolute control (T_1- 1.27%). The

organic carbon content in soil increased from the initial value of 1.32 % in all treatments except absolute control.

4.2.1.1.2. After harvest

The pH, EC and organic carbon status in soil after harvest of the crop are depicted in Table 4.6. Soil pH ranged from 4.72 to 5.20 after harvest of the crop. Treatment T_{12} (T_3+ MgCO$_3$ @ 150% of T_5) was found to be superior in increasing the soil pH. The lowest pH was recorded in the absolute control. A slight increase in the soil pH was observed at harvest in comparison to the flowering stage in all treatments except absolute control. The electrical conductivity in soil was not found to be significantly influenced by the treatments at this stage.

Table 4.6. Effect of treatments on pH, EC and organic carbon of soil after harvest

Treatments		pH	EC (dS m^{-1})	OC (%)
T_1	Absolute control	4.72^g	0.04	1.08^g
T_2	Organic manure @ 20 t ha^{-1}	4.83^f	0.05	1.67^{de}
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	4.92^e	0.05	2.12^a
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	4.91^e	0.05	1.90^{bc}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	4.95^{de}	0.06	1.72^{cd}
T_6	T_3+ MgCO$_3$ @10% of T_5	4.96^{de}	0.06	1.70^{cde}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	4.97^{de}	0.05	1.67^{def}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	4.98^{cd}	0.05	1.52^{def}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	5.02^c	0.05	1.50^{ef}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	5.14^b	0.05	1.47^f
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	5.14^b	0.05	1.95^{ab}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	5.20^a	0.05	1.99^{ab}
Treatment means with common superscript do not differ significantly				

The organic carbon content in soil ranged between $1.08 - 2.12$ % at harvest of cowpea and the lowest content was recorded in absolute control. Significantly higher value of organic carbon was noted in T_3 which was on par with T_{11} and T_{12}. A decline in the organic carbon status was recorded in all the treatments in comparison to the flowering stage.

4.2.1.2. Status of primary nutrients in soil

The effect of treatments on the status of available nitrogen, phosphorus and potassium are depicted in Table 4.7 and 4.8.

4.2.1.2.1. Flowering stage

The initial status of available nitrogen, phosphorus and potassium in soil was 476.67 kg ha^{-1}, 98.04 kg ha^{-1} and 240.18 kg ha^{-1}. The initial status of available nitrogen and potassium in soil was medium while phosphorus content was high. The treatments imposed produced significant variations in available nitrogen, phosphorus and potassium status in soil. Significantly higher content of available nitrogen was recorded in the treatment T_3 (534.55 kg ha^{-1}) while significantly higher content of available phosphorus (131.76 kg ha^{-1}) and potassium (198.74 kg ha^{-1}) was recorded in treatment T_{12} ($T_3 + MgCO_3$ @ 150% of T_5). The availability of primary nutrients was the lowest in the absolute control treatment. The available status of phosphorus in soil during flowering was found to increase from the initial status in all the treatments except T_1 whereas available potassium was found to decline.

4.2.1.2.1. After harvest

Data on available N content in soil after crop harvest shows significant variations between treatments (Table 4.8). The available nitrogen content in soil varied from 286.12 kg ha^{-1} in the absolute control treatment (T_1) to 502.94 kg ha^{-1} in T_9 ($T_3 + MgCO_3$ @ 60% of T_5). Treatments $T_2, T_3, T_5, T_6, T_8, T_{11}$ and T_{12} were on par with T_9 in available nitrogen content. The available phosphorus status in soils was high and ranged from 72.23 kg ha^{-1} in treatment T_1 to 137.24 kg ha^{-1} in T_3

(POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}). The available potassium status in soil was found to increase from the flowering stage in all the treatments and treatment T_{12} recorded significantly higher content of available potassium in soil (290.64 kg ha^{-1}).

Table 4.7. Effect of treatments on available primary nutrient status in soil during flowering

Treatments		Nitrogen	Phosphorus	Potassium
		(kg ha^{-1})		
T_1	Absolute control	347.72^g	82.89^g	139.38^f
T_2	Organic manure @ 20 t ha^{-1}	467.49^d	118.21^d	166.43^c
T_3	POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}	534.55^a	122.55^{bc}	183.29^b
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	503.52^{bc}	121.50^c	148.06^e
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	429.98^e	123.83^b	179.42^b
T_6	T_3+ $MgCO_3$ @10% of T_5	502.46^{bc}	116.89^d	149.01^e
T_7	T_3+ $MgCO_3$ @ 20% of T_5	512.37^b	107.14^f	150.02^e
T_8	T_3+ $MgCO_3$ @ 40% of T_5	371.02^f	113.31^e	156.96^d
T_9	T_3+ $MgCO_3$ @ 60% of T_5	506.38^b	108.35^f	157.86^d
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	509.52^b	107.69^f	165.20^c
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	512.09^b	106.71^f	166.65^c
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	491.40^c	131.76^a	198.74^a
Treatment means with common superscript do not differ significantly				

4.2.1.3. Status of secondary nutrients in soil

The effect of treatments on availability of calcium, magnesium and sulphur in soil during flowering and after harvest are presented in Tables 4.9 and 4.10 respectively.

4.2.1.3.1. Flowering stage

The initial status of available calcium, magnesium and sulphur in soil was 429.30 mg kg^{-1}, 64.53 mg kg^{-1} and 5.00 mg kg^{-1} respectively. The treatments

imposed was found to significantly influence the status of secondary nutrients in soil at flowering stage. Significantly higher content of available calcium was recorded in T_{10} (T_3+ $MgCO_3$ @ 80% of T_5) and was on par with T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) with values 538.25 mg kg^{-1} and 520.00 mg kg^{-1} respectively.

Table 4.8. Effect of treatments on available primary nutrient status in soil after harvest

Treatments		Nitrogen	Phosphorus	Potassium
		(kg ha^{-1})		
T_1	Absolute control	286.12^c	72.23^h	191.99^i
T_2	Organic manure @ 20 t ha^{-1}	458.03^a	123.28^c	264.76^{cd}
T_3	POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}	501.72^a	137.24^a	268.24^{bc}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	332.41^{bc}	125.11^c	259.56^{cde}
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	495.48^a	125.20^c	250.82^{ef}
T_6	T_3+ $MgCO_3$ @10% of T_5	489.21^a	98.28^e	234.24^{gh}
T_7	T_3+ $MgCO_3$ @ 20% of T_5	345.18^b	98.79^e	244.49^f
T_8	T_3+ $MgCO_3$ @ 40% of T_5	482.94^a	88.11^g	229.60^h
T_9	T_3+ $MgCO_3$ @ 60% of T_5	502.94^a	94.48^f	242.42^{fg}
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	348.45^b	98.29^e	256.92^{de}
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	470.39^a	109.73^d	276.92^b
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	472.39^a	129.88^b	290.64^a
Treatment means with common superscript do not differ significantly				

Available Mg content was found to have increased in all treatments except absolute control (T_1) during this stage of crop. The content of available magnesium ranged between 55.57 mg kg^{-1} and 123.47 mg kg^{-1}. The lowest value was recorded in absolute control and the highest in T_{12} (T_3+ $MgCO_3$ @ 150% of T_5) which was significantly above all other treatments.

Treatments imposed was found to cause significant variations in available sulphur content in soil. Available sulphur ranged between $1.91 - 2.29$ mg kg^{-1} which comes under deficient category.

Table 4.9. Effect of treatments on available secondary nutrient status in soil during flowering

Treatments		Calcium	Magnesium	Sulphur
		mg kg^{-1}		
T_1	Absolute control	390.67^d	55.57^i	1.91^c
T_2	Organic manure @ 20 t ha^{-1}	487.42^c	68.90^h	2.59^a
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	511.75^b	73.77^g	2.50^{ab}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	502.40^{bc}	76.90^{fg}	2.40^{ab}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	520.00^{ab}	105.25^b	2.20^{bc}
T_6	T_3+ MgCO$_3$ @10% of T_5	502.00^{bc}	67.20^h	2.33^{ab}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	510.75^b	75.90^{fg}	2.21^{bc}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	504.75^{bc}	77.85^f	2.30^{ab}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	502.07^{bc}	81.77^e	2.24^{bc}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	538.25^a	93.05^d	2.48^{ab}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	499.92^{bc}	101.10^c	2.42^{ab}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	507.25^{bc}	123.47^a	2.22^{bc}
Treatment means with common superscript do not differ significantly				

4.2.1.3.2. After harvest

Status of available calcium, magnesium and sulphur in soil after crop harvest are shown in Table 4.10. All these nutrients in soil showed an increase when compared to the flowering stage. Treatment T_4 (627.25 mg kg^{-1}) recorded highest available Ca and was on par with T_7 (624.00 mg kg^{-1}), T_9 (616.50 mg kg^{-1}) and T_{11} (614.00 mg kg^{-1}).

Available Mg varied from 63.10 mg kg^{-1} to 130.95 mg kg^{-1}. The highest value was recorded in T_{12} (T_3+ MgCO$_3$ @ 150% of T_5) which was on par with T_{11} that recorded 126.82 mg kg^{-1}.

All the treatments except T_1 and T_3 were on par in recording significantly higher content of available sulphur in soil at this stage. Invariably the lowest content of available primary and secondary nutrients was recorded in the absolute control treatment.

Table 4.10. Effect of treatments on available secondary nutrient status in soil after harvest

Treatments		Calcium	Magnesium	Sulphur
		mg kg^{-1}		
T_1	Absolute control	467.35^g	63.10^f	2.75^c
T_2	Organic manure @ 20 t ha^{-1}	583.87^{de}	79.22^e	3.49^{ab}
T_3	POP recommendation with CaCO3 @ 250 kg ha^{-1}	575.02^e	80.90^e	3.36^b
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	627.25^a	93.10^d	3.48^{ab}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	597.50^{cd}	110.40^b	3.43^{ab}
T_6	T_3+ MgCO$_3$ @10% of T_5	580.50^e	70.52^f	3.4^{ab}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	624.00^a	79.45^e	3.49^{ab}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	598.25^{cd}	82.55^e	3.41^{ab}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	616.50^{ab}	89.42^d	3.46^{ab}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	605.30^{bc}	100.80^c	3.45^{ab}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	614.00^{abc}	126.82^a	3.55^{ab}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	557.25^f	130.95^a	3.63^a
Treatment means with common superscript do not differ significantly				

4.2.1.4. Status of micronutrients in soil

4.2.1.4.1. Flowering stage

The effect of treatments on the available status of micronutrients in soil at flowering stage of the crop is presented in Table 4.11. The data shows significant variations in micronutrient status in soil at this stage. Upon analyzing the results of available Fe in soil during flowering, highest content was observed in T_2 (Organic manure @ 20 t ha^{-1}) with a value of 26.94 mg kg^{-1}, followed by T_3 (POP

recommendation with $CaCO_3$ @ 250 kg ha^{-1}) and T_6 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}). All other treatments were on par with respect to available Fe content. The initial status of available iron was 12.41 mg kg^{-1}.

Available Mn content ranged from 33.09 mg kg^{-1} to 50.27 mg kg^{-1}. An increase in the status of available manganese was observed from the initial value of 16.26 mg kg^{-1}. Treatment T_3 (50.27 mg kg^{-1}) recorded the highest value and T_{11} (33.09 mg kg^{-1}) the lowest. Treatments T_5 (44.81 mg kg^{-1}), T_7 (42.39 mg kg^{-1}) and T_{10} (43.18 mg kg^{-1}) were on par with respect to available Mn.

A decline in the available zinc status was recorded from the initial value of 3.81 mg kg^{-1}. The highest value of available Zn was recorded in T_3 (2.78 mg kg^{-1}) and was on par with T_{12} (2.61 mg kg^{-1}). Absolute control showed lowest content of Zn during flowering of cowpea (2.05 mg kg^{-1}). All treatments were sufficient with respect to availability of Zn during flowering of cowpea.

The initial value of available copper in soil was 8.08 mg kg^{-1}. Available Cu content in soil during flowering varied between 6.00 mg kg^{-1} (T_1) to 13.33 mg kg^{-1} (T_6). Significantly lower content of available copper was recorded in absolute control (T_1). Available boron in soil was initially deficient at 0.24 mg kg^{-1}, while the data shows an increase in boron availability during flowering except in absolute control (T_1). Lowest content was recorded in absolute control (0.22 mg kg^{-1}) and highest in T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) which was on par with T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}).

4.2.1.4.2. After harvest

The data in Table 4.12 shows the micronutrient status in soil after crop harvest. Available iron ranged from 11.19 to 19.40 mg kg^{-1}. The highest content of available Fe was recorded by T_6 (T_3+ $MgCO_3$ @10% of T_5) and lowest by T_{10} (T_3+ $MgCO_3$ @ 80% of T_5). Available Mn content in soil ranged between 37.80 to 23.33 mg kg^{-1} after harvest of cowpea. A significantly higher Mn content was recorded in

T_2 (Organic manure @ 20 t ha⁻¹) and lowest content in T_{12} (T_3+ MgCO₃ @ 150% of T_5) which was on par with T_{10} (T_3+ MgCO₃ @ 80% of T_5).

Table 4.11. Effect of treatments on available micronutrient status in soil during flowering

Treatments		Fe	Mn	Zn	Cu	B
		mg kg⁻¹				
T_1	Absolute control	13.79^c	40.41^{cd}	2.05^f	6.00^h	0.22^d
T_2	Organic manure @ 20 t ha⁻¹	26.94^a	38.68^{de}	2.35^{cde}	11.01^g	0.68^{bc}
T_3	POP recommendation with CaCO₃ @ 250 kg ha⁻¹	16.12^b	50.27^a	2.78^a	11.38^f	0.70^{bc}
T_4	POP recommendation with dolomite @ 400 kg ha⁻¹	12.85^c	36.41^e	2.24^{def}	11.80^e	0.88^a
T_5	T_3+ MgCO₃ to raise available Mg to 120 mg kg⁻¹	13.76^c	44.81^b	2.53^{bc}	11.78^e	0.97^a
T_6	T_3+ MgCO₃ @10% of T_5	13.68^c	39.21^{de}	2.30^{de}	13.33^a	0.73^b
T_7	T_3+ MgCO₃ @ 20% of T_5	16.14^b	42.39^{bc}	2.22^{def}	12.42^{bc}	0.57^c
T_8	T_3+ MgCO₃ @ 40% of T_5	13.23^c	41.12^{cd}	2.16^{ef}	11.91^{de}	0.60^{bc}
T_9	T_3+ MgCO₃ @ 60% of T_5	12.81^c	40.39^{cd}	2.39^{cd}	12.23^{bc}	0.67^{bc}
T_{10}	T_3+ MgCO₃ @ 80% of T_5	13.26^c	43.18^{bc}	2.43^{bcd}	12.49^b	0.69^{bc}
T_{11}	T_3+ MgCO₃ @ 125% of T_5	12.06^c	33.09^f	2.28^{de}	12.16^{cd}	0.57^c
T_{12}	T_3+ MgCO₃ @ 150% of T_5	13.94^c	41.59^{cd}	2.61^{ab}	12.49^b	0.72^b
Treatment means with common superscript do not differ significantly						

The available Zn content in soil was found to range from 2.22 mg kg⁻¹ to 3.32 mg kg⁻¹. Treatment T_8 (T_3+ MgCO₃ @ 40% of T_5) recorded the highest value and was on par with T_3 (POP recommendation with CaCO₃ @ 250 kg ha⁻¹) and T_5 (T_3+ MgCO₃ to raise available Mg to 120 mg kg⁻¹). Available copper content in

soil ranged from 5.05 mg kg^{-1} to 15.32 mg kg^{-1}. Significantly lower content of available copper was recorded in absolute control treatment.

Treatments showed significant difference in available B in soil and the content ranged between 1.23- 0.51 mg kg^{-1}. Treatment T_9 (T_3+ MgCO$_3$ @ 60% of T_5) recorded highest and T_1 (absolute control) recorded the lowest content of available B.

Table 4.12. Effect of treatments on available micronutrient status in soil after harvest

Treatments		Fe	Mn	Zn	Cu	B
		mg kg^{-1}				
T_1	Absolute control	13.92^{cde}	33.70^{bc}	2.22^c	5.05^f	0.39^g
T_2	Organic manure @ 20 t ha^{-1}	15.34^{bc}	37.80^a	2.60^c	13.04^{cd}	1.01^c
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	16.67^b	30.56^{ef}	3.10^{ab}	13.40^c	1.00^c
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	14.79^{bc}	32.03^{de}	2.62^c	14.66^b	0.68^{de}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	14.52^{bcd}	30.24^f	3.30^a	12.68^{de}	0.71^d
T_6	T_3+ MgCO$_3$ @10% of T_5	19.40^a	32.18^{cd}	2.55^c	15.23^a	1.12^b
T_7	T_3+ MgCO$_3$ @ 20% of T_5	13.70^{cdef}	34.47^b	2.52^c	15.32^a	1.19^{ab}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	13.30^{cdefg}	32.85^{cd}	3.32^a	12.53^{de}	1.08^{bc}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	11.73^{fg}	28.33^g	2.67^{bc}	13.46^c	1.23^a
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	11.19^g	23.89^h	2.62^c	12.48^e	0.66^{de}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	12.53^{defg}	29.95^f	2.66^{bc}	12.57^{de}	0.59^{ef}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	11.89^{efg}	25.33^h	2.60^c	12.66^{de}	0.51^f
Treatment means with common superscript do not differ significantly						

4.2.2. Effect of treatments on microbial population in soil

The population of free living nitrogen fixing bacteria, Rhizobium/ bradyrhizobium and spore count and per cent root colonization of arbuscular mycorrhizal fungi in soil was recorded during flowering and after harvest of the crop and the results of the same are presented below.

Table 4.13. Effect of treatments on microbial population during flowering

	Treatments	Rhizobium (cfu/ g of soil)	Total N-fixing bacteria (cfu/ g of soil)	Arbuscular Mycorrhizal Fungi	
				Spores /g soil	Root colonization (%)
T_1	Absolute control	2.00×10^{5f}	40.00×10^{5g}	23.45	75^c
T_2	Organic manure @ 20t ha^{-1}	8.00×10^{5e}	40.50×10^{5g}	22.93	80^{bc}
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	6.50×10^{5e}	47.00×10^{5fg}	23.30	80^{bc}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	4.50×10^{5ef}	47.50×10^{5fg}	24.30	85^{ab}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	18.00×10^{5cd}	59.50×10^{5ef}	22.98	85^{ab}
T_6	T_3+ MgCO$_3$ @10% of T_5	28.50×10^{5b}	115.00×10^{5b}	24.15	80^{bc}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	41.50×10^{5a}	149.00×10^{5a}	24.65	90^a
T_8	T_3+ MgCO$_3$ @ 40% of T_5	20.50×10^{5c}	84.50×10^{5d}	22.77	90^a
T_9	T_3+ MgCO$_3$ @ 60% of T_5	16.50×10^{5d}	88.50×10^{5d}	23.18	90^a
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	20.50×10^{5c}	50.50×10^{5fg}	23.99	90^a
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	25.50×10^{5b}	68.50×10^{5e}	23.3	85^{ab}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	28.00×10^{5b}	102.00×10^{5c}	25.33	90^a
Treatment means with common superscript do not differ significantly					
Initial population		Absent	8.00×10^5	21	N. A.

61

Table 4.14 .Effect of treatments on microbial population after harvest

Treatments		Rhizobium (cfu /g soil)	Total N fixers (cfu / g of soil)	Arbuscular Mycorrhizal Fungi	
				Spores /g soil	Root colonizati on (%)
T_1	Absolute control	1×10^{5e}	16.00×10^{5g}	22.23	75^c
T_2	Organic manure @20 t ha^{-1}	1.5×10^{5de}	21.00×10^{5fg}	23.28	80^{bc}
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	2.00×10^{5de}	27.00×10^{5ef}	21.28	80^{bc}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	1.50×10^{5de}	29.50×10^{5ef}	23.01	80^{bc}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	4.00×10^{5bcd}	74.50×10^{5b}	23.62	85^{ab}
T_6	T_3+ MgCO$_3$ @10% of T_5	3.00×10^{5cde}	29.50×10^{5ef}	22.60	80^{bc}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	12.00×10^{5a}	140.50×10^{5a}	21.72	85^{ab}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	6.00×10^{5b}	35.00×10^{5e}	23.5	90^a
T_9	T_3+ MgCO$_3$ @ 60% of T_5	3.00×10^{5cde}	69.00×10^{5bc}	24.30	90^a
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	4.00×10^{5bcd}	36.00×10^{5e}	22.02	90^a
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	5.00×10^{5bc}	46.50×10^{5d}	23.62	90^a
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	2.50×10^{5cde}	59.50×10^{5c}	22.84	90^a
Treatment means with common superscript do not differ significantly					
Initial population		Absent		21	N.A.

4.2.2.1. During flowering

Microbial population in soil at flowering stage are depicted in Table 4.13. The initial population of Rhizobium was absent in soil. However during flowering stage of the crop, rhizobial population could be recorded and significant variations were observed between treatments. Similarly, the population of free living nitrogen fixers also increased from the initial population of 8.00×10^5 cfu/g of soil. Significantly higher number of Rhizobium population and free living nitrogen fixers were recorded in T_7 (149.00×10^5). However, the spore count of arbuscular mycorrhizal fungi (AMF) showed no significant variations among the treatments, though an increase was observed from the initial value (1067). All the treatments

except T_1, T_2, T_3, and T_6 were on par in recording significantly higher per cent of root colonization with AMF.

4.2.2.2. After harvest

The microbial population in rhizosphere soil at harvest are depicted in Table 4.14. The population of Rhizobium and free living nitrogen fixers were found to be significantly higher in T_7 (T_3+ $MgCO_3$ @20% of T_5 -12.00x10^5) when compared to other treatments. The spore count of arbuscular mycorrhizal fungi (AMF) showed no significant variations between treatments. All the treatments except T_1 (75%), T_2 (80%), T_3 (80%) T_4 (80%) and T_6 (80%) were on par in recording significantly higher per cent of root colonization with AMF.

4.2.3. Effect of treatments on plant nutrient content during flowering and harvest

The content of nutrients in different plant parts (stem and leaf during flowering and stem, leaf and pod after harvest) of cowpea was analyzed during flowering and harvest stage of cowpea and the results are presented hereafter.

4.2.3.1. Nitrogen

Data on nitrogen content in different plant parts are given in Table 4.15.

4.2.3.1.1. During flowering

The nitrogen content in the stem of cowpea plant during flowering was significantly influenced by the treatments imposed. The nitrogen content ranged from 1.83 % to 3.18 % in the stem of cowpea. The highest content (3.18%) was observed in T_2 (Organic manure @ 20 t ha^{-1}). No significant variations were recorded in the leaf nitrogen content of cowpea during flowering.

Table 4.15. Effect of treatments on nitrogen content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	2.25^{bc}	2.90	2.21^{a}	2.87^{a}	2.99
T_2	Organic manure @ 20 t ha^{-1}	3.18^{a}	3.52	1.60^{bc}	2.62^{abc}	3.12
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	2.02^{cd}	3.43	1.08^{d}	2.36^{cd}	3.16
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	1.96^{cd}	3.48	1.76^{abc}	2.64^{abc}	3.09
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	2.08^{cd}	3.41	1.53^{cd}	2.75^{ab}	3.30
T_6	T_3+ MgCO$_3$ @10% of T_5	1.89^{cd}	3.34	1.81^{abc}	2.60^{abc}	3.17
T_7	T_3+ MgCO$_3$ @ 20% of T_5	1.83^{d}	3.36	1.53^{c}	2.26^{d}	3.30
T_8	T_3+ MgCO$_3$ @ 40% of T_5	2.22^{bc}	3.52	1.67^{bc}	2.88^{a}	3.16
T_9	T_3+ MgCO$_3$ @ 60% of T_5	2.20^{bc}	3.50	1.84^{abc}	2.69^{ab}	3.17
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	2.15^{bcd}	3.09	1.45^{cd}	2.24^{bcd}	2.92
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	2.06^{cd}	3.48	2.00^{ab}	2.49^{d}	3.04
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	2.49^{b}	3.62	1.58^{bc}	2.76^{ab}	3.30
Treatment means with common superscript do not differ significantly						

4.2.3.1.2. After harvest

The nitrogen content in the stem and leaf of the crop varied significantly between treatments while no difference in the nitrogen content in pods was evident. Highest nitrogen content in stem at harvest of cowpea was obtained in absolute control (2.21%), which was on par with T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}), T_6 (T_3+ MgCO$_3$ @10% of T_5), T_9 (T_3+ MgCO$_3$ @ 60% of T_5) and T_{11} (T_3+ MgCO$_3$ @ 125% of T_5).

Treatment T_8 recorded the highest value of leaf nitrogen content (2.88%) but was on par with T_1 (Absolute control), T_2 (Organic manure @ 20 t ha^{-1}), T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}), T_5 (T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}), T_6 (T_3+ MgCO$_3$ @10% of T_5) and T_9 (T_3+ MgCO$_3$ @ 60% of T_5) and T_{12} (T_3+ MgCO$_3$ @ 150% of T_5). There was no significant difference between the treatments in content of nitrogen in pods.

4.2.3.2. Phosphorus

The phosphorus content in different plant parts are given in Table 4.16.

Table 4.16. Effect of treatments on phosphorus content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	0.20	0.19^c	0.16^{bc}	0.23	0.29^{bc}
T_2	Organic manure @ 20 t ha^{-1}	0.22	0.19^c	0.12^{ef}	0.22	0.32^{ab}
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	0.18	0.32^a	0.13^{def}	0.21	0.27^{cd}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	0.24	0.26^b	0.12^{ef}	0.21	0.36^a
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	0.21	0.31^a	0.11^f	0.24	0.26^d
T_6	T_3+ MgCO$_3$ @10% of T_5	0.22	0.25^b	0.16^{bcd}	0.23	0.32^{ab}
T_7	T_3+ MgCO$_3$ @ 20% of T_5	0.20	0.27^b	0.14^{cdef}	0.28	0.28^{cd}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	0.18	0.26^b	0.15^{bcde}	0.23	0.30^{bc}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	0.18	0.24^b	0.28^a	0.22	0.30^{bc}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	0.23	0.26^b	0.18^b	0.22	0.32^{ab}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	0.19	0.24^b	0.15^{bcde}	0.24	0.28^{cd}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	0.21	0.26^b	0.13^{def}	0.21	0.26^d
Treatment means with common superscript do not differ significantly						

4.2.3.2.1. During flowering

Analysis of the data on plant nutrient content during flowering showed no significant variation in phosphorus content in stem. The phosphorus content of cowpea leaves ranged from 0.19% to 0.32%. Highest content of 0.32 % was recorded by T_3 (POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}) and was on par with T_5 (T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}) respectively. Significantly lower content (0.19%) was observed in T_1 (Absolute control) and T_2 (Organic manure @ 20 t ha^{-1}).

4.2.3.2.2. After harvest

Treatments differed significantly in phosphorus content of cowpea stem and highest among treatments was recorded by T_9 (T_3+ $MgCO_3$ @ 60% of T_5) with a value of 0.28%.

The phosphorus content in cowpea leaves ranged between 0.21 % and 0.28 %, but did not differ significantly between treatments.

Phosphorus content in cowpea pods exhibited significant difference among treatments. T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}) recorded highest phosphorus content and was on par with T_2 (Organic manure @ 20 t ha^{-1}), T_6 (T_3+ $MgCO_3$ @10% of T_5), and T_{10} (T_3+ $MgCO_3$ @ 80% of T_5). The lowest content among treatments was recorded by T_{12} (T_3+ $MgCO_3$ @ 150% of T_5) and was on par with T_{11} (T_3+ $MgCO_3$ @ 125% of T_5), T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and T_3 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}).

4.2.3.3. Potassium

The plant content of potassium analyzed during flowering and harvest is depicted in Table 4.17.

4.2.3.3.1. During flowering

The content of potassium in stem of plants ranged between 1.60 to 2.73 % and highest was in T_2 (Organic manure @ 20 t ha^{-1}) and was on par with treatment T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}).

Potassium content of cowpea leaves differed significantly among treatments during flowering stage, with significantly higher value (2.58%) in T_2 (Organic manure @ 20 t ha^{-1}).

Table 4.17. Effect of treatments on potassium content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	2.09^{cd}	2.24^c	2.08^{bc}	1.49^{abc}	1.46^{cde}
T_2	Organic manure @ 20 t ha^{-1}	2.73^a	2.s58^a	1.82^{cde}	1.33^{de}	1.45^{de}
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	2.19^c	2.46^b	1.97^{bcd}	1.38^{cd}	1.44^{de}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	2.24^{bc}	2.09^d	1.74^{de}	1.47^{abc}	1.60^c
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	2.58^{ab}	2.26^c	1.67^e	1.52^{ab}	1.38^e
T_6	T_3+ MgCO$_3$ @10% of T_5	2.02^{cd}	1.81^f	2.37^a	1.56^a	1.78^b
T_7	T_3+ MgCO$_3$ @ 20% of T_5	1.83^{de}	1.80^f	1.73^{de}	1.24^{ef}	1.91^{ab}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	1.92^{cde}	2.09^{de}	1.85^{bcde}	1.43^{bcd}	1.94^a
T_9	T_3+ MgCO$_3$ @ 60% of T_5	1.91^{cde}	2.26^c	1.75^{de}	1.20^f	1.87^{ab}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	1.60^e	1.98^e	2.11^{ab}	1.44^{bcd}	1.55^{cd}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	1.98^{cd}	1.77^f	1.78^{de}	1.45^{abc}	1.46^{cde}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	2.22^c	2.05^{de}	1.81^{de}	1.51^{ab}	1.51^{cde}
Treatment means with common superscript do not differ significantly						

4.2.3.3.2. After harvest

Treatments differed significantly in potassium content of cowpea stem at harvest stage. Treatment T_6 (T_3+ MgCO$_3$ @10% of T_5) recorded highest potassium content (2.37%) in stem and was on par with T_{10} (T_3+ MgCO$_3$ @ 80% of T_5). Treatment T_5 recorded lowest content of potassium (1.67 %) and was on par with T_7 (T_3+ MgCO$_3$ @ 20% of T_5), T_8 (T_3+ MgCO$_3$ @ 40% of T_5), T_9 (T_3+ MgCO$_3$ @ 60% of T_5), T_{11} (T_3+ MgCO$_3$ @ 125% of T_5), T_{12} (T_3+ MgCO$_3$ @ 150% of T_5), and T_2 (Organic manure @ 20 t ha^{-1}).

Potassium content in leaf varied from 1.2% to 1.56%. Highest content was recorded in T_6 (T_3+ MgCO$_3$ @10% of T_5) and was on par with T_5 (T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}), T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}), T_{11} (T_3+ MgCO$_3$ @ 125% of T_5), T_{12} (T_3+ MgCO$_3$ @ 150% of T_5) and T_1

(absolute control). The lowest content was obtained in T_9 (T_3+ $MgCO_3$ @ 60% of T_5) and was on par with T_7 (T_3+ $MgCO_3$ @ 20% of T_5).

In the case of potassium content in pods a significantly higher content was recorded in T_8 (T_3+ $MgCO_3$ @ 40% of T_5) and was on par with T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and T_9 (T_3+ $MgCO_3$ @ 60% of T_5). The content of potassium varied from 1.38% to 1.94%.

4.2.3.4. Calcium

Data pertaining to the calcium content in plant is shown in Table 4.18.

Table 4.18. Effect of treatments on calcium content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	1.06^b	2.32^{def}	0.83^{cd}	3.53^c	0.16
T_2	Organic manure @ 20 t ha^{-1}	0.98^{cd}	2.69^{ab}	1.14^{ab}	3.68^{abc}	0.15
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	1.03^{bc}	2.21^{ef}	1.05^{abc}	3.23^d	0.15
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	0.97^{cd}	2.43^{cd}	0.87^{bcd}	3.23^d	0.15
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	0.94^{de}	2.38^{cde}	0.78^{cd}	3.27^d	0.16
T_6	T_3+ $MgCO_3$ @10% of T_5	1.21^a	2.73^a	1.22^a	3.77^{ab}	0.15
T_7	T_3+ $MgCO_3$ @ 20% of T_5	1.10^b	2.53^{bc}	1.27^a	3.87^a	0.15
T_8	T_3+ $MgCO_3$ @ 40% of T_5	0.87^{efg}	2.31^{def}	0.91^{bcd}	3.53^c	0.16
T_9	T_3+ $MgCO_3$ @ 60% of T_5	0.92^{def}	2.35^{cdef}	0.77^{cd}	3.59^{bc}	0.15
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	0.92^{def}	2.48^{cd}	0.73^d	3.66^{bc}	0.16
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	0.84^g	2.33^{def}	0.78^{cd}	3.49^c	0.16
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	0.85^{fg}	2.16^f	0.74^d	3.12^d	0.16
Treatment means with common superscript do not differ significantly						

4.2.3.4.1. During flowering

Significant variations between treatments were observed in the calcium content of stem and leaf of cowpea during flowering. The highest Ca content in

stem was recorded in T_6 (1.21%). Compared to stem, leaves recorded higher content of Ca and values ranged between 2.73 – 2.16 %.

Treatment T_6 (T_3+ $MgCO_3$ @10% of T_5) recorded significantly higher content of calcium in leaves and was on par with T_2.

4.2.3.4.2. After harvest

The calcium content in stem and leaf varied significantly between treatments. The calcium content in stem ranged from 0.73% to 1.27% and between 3.12 % and 3.87% in leaves. The content of calcium in leaves showed a significantly higher value than stem. The calcium content in stem and leaves at harvest were significantly superior in treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and was on par with T_6 and T_2. Calcium content in pods was not found to vary with respect to treatment imposed.

4.2.3.5. Magnesium

4.2.3.5.1. During flowering

Analysis of the results (Table 4.19) obtained for magnesium content in stem of cowpea showed significantly higher value in T_{12} (0.55% -T_3 +$MgCO_3$ @ 150% of T_5) which was on par with T_5 (0.53% -T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}).

Treatments differed significantly with regard to magnesium content in leaves. The highest value of magnesium in leaves was recorded in T_{11} (T_3+ $MgCO_3$ @ 125% of T_5) which was on par with T_{10} (T_3+ $MgCO_3$ @ 80% of T_5) and T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) while the lowest value was observed in absolute control (T_1).

4.2.3.5.2. After harvest

The effect of treatments on magnesium content in stem, leaf and pods of cowpea at harvest is evident from the data (Table 4.19). Significantly higher

content (0.73%) of magnesium was recorded in T_{11} (T_3+ MgCO$_3$ @ 125% of T_5) when compared to other treatments. The lowest content (0.48%) was recorded by T_2 (Organic manure @ 20 t ha^{-1}).

Analysis of data on magnesium content of cowpea leaves showed that significantly higher content (0.50%) was obtained in T_{11} (T_3+ MgCO$_3$ @ 125% of T_5) and was on par with T_{12} (T_3+ MgCO$_3$ @ 150% of T_5), T_5 (T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}), T_{10} (T_3+ MgCO$_3$ @ 80% of T_5), T_8 (T_3+ MgCO$_3$ @ 40% of T_5) and T_7 (T_3+ MgCO$_3$ @ 20% of T_5). However the treatments imposed did not show any significant effect on magnesium content in pods.

Table 4.19. Effect of treatments on magnesium content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	0.43^d	0.35^f	0.54^{ef}	0.38^e	0.25
T_2	Organic manure @ 20 t ha^{-1}	0.42^d	0.44^{bc}	0.48^g	0.42^{cde}	0.25
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	0.45^{cd}	0.44^{bc}	0.53^f	0.39^{de}	0.25
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	0.48^{bc}	0.41^{de}	0.54^{ef}	0.43^{bcd}	0.26
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	0.52^{ab}	0.46^{ab}	0.56^{de}	0.47^a	0.26
T_6	T_3+ MgCO$_3$ @10% of T_5	0.45^{cd}	0.36^f	0.61^{bc}	0.42^{bcde}	0.26
T_7	T_3+ MgCO$_3$ @ 20% of T_5	0.42^d	0.39^e	0.57^d	0.46^{abc}	0.26
T_8	T_3+ MgCO$_3$ @ 40% of T_5	0.47^{bc}	0.43^{cd}	0.60^c	0.47^a	0.27
T_9	T_3+ MgCO$_3$ @ 60% of T_5	0.46^{cd}	0.43^{cd}	0.54^{ef}	0.41^{de}	0.26
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	0.48^{bc}	0.46^{ab}	0.57^d	0.46^{ab}	0.27
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	0.49^{bc}	0.47^a	0.73^a	0.50^a	0.27
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	0.55^a	0.45^{bc}	0.63^b	0.48^a	0.27
Treatment means with common superscript do not differ significantly						

4.2.3.6. Sulphur

The effect of treatments on sulphur content in plant parts are shown in Table 4.20.

4.2.3.6.1. During flowering

No significant difference between treatments were observed in the sulphur content of plant stem. However significantly higher content of sulphur in leaves was recorded in treatment T_8 (0.27%) which was on par with T_{11} and T_2. The sulphur content in leaves ranged from 0.21 % to 0.27 %.

4.2.3.6.2. After harvest

The analysis of plant sulphur content in stem, leaf and pods at harvest of the crop showed no significant variations between treatments.

Table 4.20. Effect of treatments on sulphur content (%) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	0.36	0.24^{bc}	0.40	0.28	0.16
T_2	Organic manure @ 20 t ha^{-1}	0.34	0.26^{ab}	0.39	0.29	0.17
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	0.33	0.24^{bc}	0.37	0.26	0.22
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	0.37	0.24^{bc}	0.40	0.28	0.20
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	0.36	0.24^{bc}	0.39	0.27	0.18
T_6	T_3+ MgCO$_3$ @10% of T_5	0.38	0.21^{d}	0.44	0.29	0.18
T_7	T_3+ MgCO$_3$ @ 20% of T_5	0.30	0.23^{cd}	0.31	0.35	0.18
T_8	T_3+ MgCO$_3$ @ 40% of T_5	0.40	0.27^{a}	0.40	0.27	0.17
T_9	T_3+ MgCO$_3$ @ 60% of T_5	0.40	0.24^{bc}	0.40	0.32	0.16
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	0.38	0.22^{cd}	0.37	0.24	0.16
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	0.42	0.25^{ab}	0.37	0.39	0.16
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	0.40	0.24^{bc}	0.34	0.27	0.17
Treatment means with common superscript do not differ significantly						

4.2.3.7. Iron

Data on iron content in plant at flowering and harvest of crop is given in Table 4.21.

4.2.3.7.1. During flowering

The data showed non-significant difference among treatments with respect to iron contents in stem. Whereas the iron content in leaves varied significantly between treatments. The highest content was recorded in T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) with a mean value of 3466.12 mg kg^{-1} and was on par (3060.25 mg kg^{-1}) with T_3 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}). The lowest content of iron in leaves was recorded in T_{12} (1122 mg kg^{-1}) which was on par with T_{11}.

4.2.3.7.2. After harvest

Iron content in stem, leaf and pod varied significantly with treatments. T_{10} (T_3+ $MgCO_3$ @ 125% of T_5) recorded highest content of iron (2484.25 mg kg^{-1}) and was on par with treatments T_9 (T_3+ $MgCO_3$ @ 60% of T_5), T_8 (T_3+ $MgCO_3$ @ 40% of T_5), T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and T_6 (T_3+ $MgCO_3$ @10% of T_5).

The effects of treatments on iron content of cowpea leaves during harvest showed that treatment T_2 (Organic manure @ 20 t ha^{-1}) recorded highest content (1713.37 mg kg^{-1}) of iron in leaves which was on par with T_3 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}). Iron content in pods of cowpea was ranged between 244.37 mg kg^{-1} (T_7) and 1163.62 mg kg^{-1} (T_6). Treatment T_6 (T_3+ $MgCO_3$ @10% of T_5) was on par with T_4 (POP recommendation with dolomite @ 400 kg ha^{-1}), and T_2 (Organic manure @ 20 t ha^{-1}).

Table 4.21. Effect of treatments on iron content (mg kg⁻¹) in plant

Treatments	Flowering		Harvest		
	Stem	Leaf	Stem	Leaf	Pod
T_1 Absolute control	2219.38	2619.50^{bc}	1294.00^{b}	1436.75^{b}	569.75^{de}
T_2 Organic manure @ 20 t ha⁻¹	2075.25	2387.00^{cd}	1152.75^{b}	1713.37^{a}	1122.00^{ab}
T_3 POP recommendation with CaCO$_3$ @ 250 kg ha⁻¹	1934.25	3060.25^{ab}	1269.50^{b}	1588.25^{ab}	614.75^{de}
T_4 POP recommendation with dolomite @ 400 kg ha⁻¹	2164.00	2408.00^{cd}	1162.00^{b}	820.25^{cd}	1101.87^{ab}
T_5 T_3+ MgCO$_3$ to raise available Mg to 120 mg kg⁻¹	2628.00	3466.12^{a}	1114.50^{b}	889.00^{cd}	683.50^{cde}
T_6 T_3+ MgCO$_3$ @10% of T_5	2493.00	2171.25^{cd}	2021.00^{a}	752.75^{cde}	1163.62^{a}
T_7 T_3+ MgCO$_3$ @ 20% of T_5	1918.75	2219.75^{cd}	2036.50^{a}	544.37^{d}	244.37^{f}
T_8 T_3+ MgCO$_3$ @ 40% of T_5	1807.75	2243.00^{cd}	1968.25^{a}	624.50^{de}	439.62^{ef}
T_9 T_3+ MgCO$_3$ @ 60% of T_5	1759.25	2423.00^{cd}	2125.12^{a}	668.25^{de}	581.25^{de}
T_{10} T_3+ MgCO$_3$ @ 80% of T_5	2221.50	2109.25^{d}	2484.25^{a}	847.50^{cd}	902.25^{bc}
T_{11} T_3+ MgCO$_3$ @ 125% of T_5	2253.75	1261.50^{e}	1180.50^{b}	969.50^{c}	745.12^{cd}
T_{12} T_3+ MgCO$_3$ @ 150% of T_5	2263.00	1122.00^{e}	1190.25^{b}	629.75^{de}	636.25^{de}

Treatment means with common superscript do not differ significantly

Table 4.22. Effect of treatments on manganese content (mg kg^{-1}) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	183.37^a	596.25^a	216.51^b	845.25^a	88.50^a
T_2	Organic manure @ 20 t ha^{-1}	150.12^b	415.25^{ef}	134.37^{gh}	796.75^b	76.00^b
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	112.75^g	511.50^b	161.50^d	671.75^{fg}	70.75^{cd}
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	130.00^e	473.00^c	158.00^{de}	647.37^{gh}	70.00^{cde}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	139.75^{cd}	430.50^{ef}	192.00^c	692.12^{ef}	66.00^f
T_6	T_3+ MgCO$_3$ @10% of T$_5$	147.87^{bc}	385.75^{gh}	236.12^a	706.25^{de}	70.25^{cde}
T_7	T_3+ MgCO$_3$ @ 20% of T$_5$	130.75^{de}	380.50^{gh}	150.00^{ef}	740.50^c	71.625^c
T_8	T_3+ MgCO$_3$ @ 40% of T$_5$	135.25^{de}	424.25^{ef}	139.87^{fg}	729.75^{cd}	66.75^{ef}
T_9	T_3+ MgCO$_3$ @ 60% of T$_5$	117.50^{fg}	402.50^{fg}	132.62^{gh}	626.25^h	68.00^{cdef}
T_{10}	T_3+ MgCO$_3$ @ 80% of T$_5$	126.75^{ef}	436.75^{de}	128.25^h	714.37^{cde}	79.00^b
T_{11}	T_3+ MgCO$_3$ @ 125% of T$_5$	113.25^g	376.00^h	140.25^{fg}	710.25^{cde}	68.00^{cdef}
T_{12}	T_3+ MgCO$_3$ @ 150% of T$_5$	126.25^{ef}	454.25^{cd}	142.50^{fg}	614.50^h	67.5^{def}

Treatment means with common superscript do not differ significantly

4.2.3.8. Manganese

Data on manganese content in plant at flowering and harvest of crop is given in Table 4.22.

4.2.3.8.1. During flowering

Significantly higher content of manganese in stem (183.37 mg kg^{-1}) and leaf (596.25 mg kg^{-1}) was recorded in treatment T_1 (absolute control). Significantly lower content (112.75 mg kg^{-1}) was recorded in T_3 and was on par with T_{11} and T_9.

Manganese content in leaves during flowering of cowpea was higher compared to stem. The content ranged from 376.00 to 596.25 mg kg^{-1} with significantly higher value in absolute control (T_1). Among the treatments the treatment T_{11} (T_3+ $MgCO_3$ @ 125% of T_5) recorded the lowest content of manganese and was on par with T_6 and T_7.

4.2.3.8.2. After harvest

On analyzing the result of manganese content in cowpea stem at crop harvest, T_6 recorded higher value of 236.12 mg kg^{-1}. Lowest content of manganese (128.29 mg kg^{-1}) was obtained in T_{10} (T_3+ $MgCO_3$ @ 80% of T_5) which was on par with T_9 (T_3+ $MgCO_3$ @ 60% of T_5) and T_2 (Organic manure @ 20 t ha^{-1}).

Content of manganese in cowpea leaves ranged between 614.50 mg kg^{-1} and 845.25 mg kg^{-1}. Absolute control recorded highest content of manganese followed by T_2 (Organic manure @ 20 t ha^{-1}). T_{12} (T_3+ $MgCO_3$ @ 150% of T_5) recorded lowest manganese content in cowpea leaves (614.50 mg kg^{-1}) and was on par with T_9 and T_4.

All the treatments differed significantly with regard to Mn content in pods. Significantly higher content was recorded in pods obtained from absolute control followed by T_2 (Organic manure @ 20 t ha^{-1}).

4.2.3.9. Zinc

4.2.3.9.1. During flowering

The zinc content in stem and leaf of cowpea during flowering did not show significant variations between treatments.

4.2.3.9.2. After harvest

Similar to the flowering stage no significant variations in the zinc content of stem, leaf and pods of cowpea could be recorded between treatments (Table 4.23).

Table 4.23. Effect of treatments on zinc content (mg kg^{-1}) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	104.16	39.01	54.75	31.57	51.66
T_2	Organic manure @ 20 t ha^{-1}	104.60	38.90	54.74	31.62	51.64
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	104.82	39.01	54.50	31.35	51.52
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	104.72	39.00	54.24	31.62	51.73
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	104.39	39.00	54.43	31.64	51.70
T_6	T_3+ MgCO$_3$ @10% of T_5	104.75	39.05	54.70	31.64	51.57
T_7	T_3+ MgCO$_3$ @ 20% of T_5	104.39	39.01	54.66	31.50	51.56
T_8	T_3+ MgCO$_3$ @ 40% of T_5	104.35	39.00	54.12	31.83	51.38
T_9	T_3+ MgCO$_3$ @ 60% of T_5	104.33	39.08	54.38	31.67	51.55
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	104.54	38.96	54.53	31.77	51.50
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	104.44	39.04	54.82	31.45	51.61
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	104.66	39.00	54.92	31.42	51.50
Treatments do not differ significantly						

4.2.3.10. Copper

4.2.3.10.1. During flowering

The copper content in stem and leaf of cowpea during flowering did not show significant variations between treatments (Table 4.24).

4.2.3.10.2. After harvest

Similar to the flowering stage no significant variations in the copper content of stem, leaf and pods of cowpea could be recorded between treatments (Table 4.24).

Table 4.24. Effect of treatments on copper content (mg kg^{-1}) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	11.63	13.34	21.64	23.72	19.25
T_2	Organic manure @ 20 t ha^{-1}	11.81	13.80	21.30	23.25	19.17
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	11.37	13.48	21.50	23.4	19.27
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	11.20	13.42	21.66	23.34	19.33
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	11.63	13.50	21.42	23.46	19.32
T_6	T_3+ MgCO$_3$ @10% of T_5	11.75	13.26	21.61	23.26	19.35
T_7	T_3+ MgCO$_3$ @ 20% of T_5	11.63	13.66	21.65	23.21	19.47
T_8	T_3+ MgCO$_3$ @ 40% of T_5	11.50	13.32	21.21	23.19	19.38
T_9	T_3+ MgCO$_3$ @ 60% of T_5	11.72	13.36	21.51	23.25	19.29
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	11.49	13.73	21.47	23.19	19.34
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	11.55	13.25	21.48	23.15	19.33
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	11.51	13.5	21.47	23.23	19.26
Treatments do not differ significantly						

4.2.3.11. Boron

The boron content in plant during flowering and after harvest is shown in Table 4.25.

4.2.3.11.1. During flowering

All treatments except T_1, T_8 and T_{12} were on par in recording significantly higher content of boron in stem and all the treatments except T_1 recorded significantly higher content of boron in leaf at flowering. The boron content in stem and leaf of cowpea during flowering did not show significant variations between treatments.

Table 4.25. Effect of treatments on boron content (mg kg^{-1}) in plant

Treatments		Flowering		Harvest		
		Stem	Leaf	Stem	Leaf	Pod
T_1	Absolute control	14.98^c	28.23^b	53.25^c	66.00^b	71.79^b
T_2	Organic manure @ 20 t ha^{-1}	25.5^{ab}	48.05^a	72.64^b	95.89^a	84.00^a
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	25.80^{ab}	46.27^a	73.25^b	97.25^a	83.99^a
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	28.00^a	47.48^a	71.86^b	95.37^a	84.06^a
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	25.25^{ab}	47.02^a	71.03^b	95.62^a	83.83^a
T_6	T_3+ MgCO$_3$ @10% of T_5	25.1^{ab}	45.91^a	72.09^b	76.56^b	83.04^a
T_7	T_3+ MgCO$_3$ @ 20% of T_5	24.81^{ab}	48.03^a	72.42^b	94.77^a	84.28^a
T_8	T_3+ MgCO$_3$ @ 40% of T_5	21.95^b	48.27^a	73.72^b	94.41^a	83.71^a
T_9	T_3+ MgCO$_3$ @ 60% of T_5	26.5^{ab}	52.56^a	72.5^b	95.24^a	82.88^a
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	27.17^{ab}	50.25^a	72.67^b	95.69^a	83.62^a
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	24.02^{ab}	46.33^a	72.55^b	94.80^a	83.95^a
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	22.55^b	46.18^a	77.02^a	95.07^a	83.40^a
Treatment means with common superscript do not differ significantly						

4.2.3.11.2. After harvest

The boron content in stem was significantly higher in treatment T_{12} while all the treatments except T_1 were on par with respect to boron content in leaf and pod.

4.2.4. Effect of treatments on chlorophyll content in leaves

Content of chlorophyll a and b were analyzed in recently matured leaves of cowpea during flowering and harvest and are presented in Table 4.26.

4.2.4.1. During flowering

Chlorophyll a showed significant variation in response of treatments in which highest content was recorded in T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and was on par with T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}). There was no significant variation among treatments in chlorophyll b during flowering.

Table 4.26. Effect of treatments on chlorophyll content (mg g^{-1}) of cowpea during flowering and harvest

	Treatments	Flowering		Harvest	
		Chl a	Chl b	Chl a	Chl b
T_1	Absolute control	1.33^f	0.842	0.842	0.122
T_2	Organic manure @ 20 t ha^{-1}	1.40^{def}	0.965	0.95	0.159
T_3	POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}	1.42^{def}	0.74	0.74	0.084
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	1.68^{bcd}	0.751	0.751	0.122
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	1.92^{ab}	0.721	0.721	0.097
T_6	T_3+ $MgCO_3$ @10% of T_5	1.67^{bcde}	0.633	0.633	0.19
T_7	T_3+ $MgCO_3$ @ 20% of T_5	2.12^a	0.95	0.95	0.558
T_8	T_3+ $MgCO_3$ @ 40% of T_5	1.80^{bc}	0.699	0.699	0.168
T_9	T_3+ $MgCO_3$ @ 60% of T_5	1.41^{def}	0.687	0.687	0.143
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	1.57^{cdef}	0.724	0.724	0.165
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	1.56^{cdef}	0.626	0.626	0.345
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	1.37^{ef}	0.674	0.674	0.073
Treatment means with common superscript do not differ significantly					

4.2.4.2. After harvest

The treatments did not show any variation in the chlorophyll content in leaves after harvest of the crop.

4.2.5. Effect of treatments on nutrient uptake by crop

The effects of treatments on nutrient uptake by the test crop cowpea is presented in Table 4.27 and 4.28.

4.2.5.1. Macronutrients

The perusal of data on nutrient uptake by crop reveals, treatment T_6 (T_3+ $MgCO_3$@10% of T_5) to be superior over other treatments to significantly increase the uptake of nitrogen, phosphorus and potassium. The comparison of treatments on uptake of calcium showed T_7 to be superior over other treatments. Treatment T_7 recorded significantly higher uptake of magnesium and was on par with T_8 and T_{11}. The crop uptake of sulphur was significantly higher in T_7 and was on par with T_5, T_6 T_8 and T_{11}.

4.2.5.2. Micronutrients

The uptake of iron was significantly higher in treatment T_6 while T_7 recorded significantly higher uptake of manganese and was on par with T_8. The data on the uptake of zinc also show significantly higher uptake in T_7 on par with T_6. Similarly significantly higher uptake of copper was recorded in T_7 on par with T_6 and T_8. Treatment T_6 and T_7 were on par in recording higher uptake of boron.

Table 4.27. Effect of treatments on crop uptake of macronutrients (mg per plant dry weight)

Treatments		N	P	K	Ca	Mg	S
T_1	Absolute control	187.98[i]	15.36[h]	117.54[i]	114.13[i]	26.67[i]	18.03[h]
T_2	Organic manure @ 20 t ha^{-1}	252.70[g]	22.66[g]	156.59[i]	176.74[g]	38.39[i]	28.16[g]
T_3	POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}	234.57[h]	22.14[g]	171.29[h]	166.71[h]	42.57[h]	29.61[fg]
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	349.01[e]	32.76[cd]	221.68[e]	191.64[e]	55.59[ef]	38.65[bc]
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	378.45[cd]	30.98[ef]	223.14[de]	203.67[d]	61.25[cd]	39.59[ab]
T_6	T_3+ $MgCO_3$ @10% of T_5	416.72[a]	40.90[a]	292.11[a]	213.72[c]	58.88[de]	41.40[ab]
T_7	T_3+ $MgCO_3$ @ 20% of T_5	397.81[b]	38.92[b]	267.09[b]	261.65[a]	66.62[a]	42.78[a]
T_8	T_3+ $MgCO_3$ @ 40% of T_5	386.55[c]	33.65[c]	256.42[c]	255.86[b]	63.40[abc]	39.58[ab]
T_9	T_3+ $MgCO_3$ @ 60% of T_5	302.72[f]	31.88[de]	189.45[g]	182.19[f]	48.34[g]	33.40[de]
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	298.45[f]	32.31[cde]	219.69[e]	183.56[f]	52.45[f]	31.81[ef]
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	350.99[e]	31.39[def]	212.58[f]	216.88[c]	64.94[ab]	41.61[ab]
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	370.06[d]	29.82[f]	227.77[d]	201.53[d]	62.07[bcd]	35.84[cd]

Treatment means with common superscript do not differ significantly

Table 4.28. Effect of treatments on crop uptake of micronutrients (mg per plant dry weight)

	Treatments	Fe	Mn	Zn	Cu	B
T_1	Absolute control	8.99^f	2.61^h	0.30^g	0.13^g	0.46^d
T_2	Organic manure @ 20 t ha^{-1}	15.26^{bc}	3.74^{de}	0.43^f	0.20^f	0.90^c
T_3	POP recommendation with CaCO$_3$ @ 250 kg ha^{-1}	14.03^{bcd}	3.23^{fg}	0.46^f	0.22^{ef}	0.94^c
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	14.05^{bcd}	3.81^{de}	0.62^{cd}	0.27^{cd}	1.09^{bc}
T_5	T_3+ MgCO$_3$ to raise available Mg to 120 mg kg^{-1}	12.38^{cde}	4.44^{bc}	0.66^{bc}	0.30^{bcd}	1.21^{bc}
T_6	T_3+ MgCO$_3$ @10% of T_5	19.61^a	4.32^c	0.71^{ab}	0.32^{ab}	1.56^a
T_7	T_3+ MgCO$_3$ @ 20% of T_5	13.44^{bcde}	5.18^a	0.73^a	0.36^a	1.30^{ab}
T_8	T_3+ MgCO$_3$ @ 40% of T_5	13.12^{cde}	4.81^{ab}	0.66^{bc}	0.31^{abc}	1.18^{bc}
T_9	T_3+ MgCO$_3$ @ 60% of T_5	12.84^{cde}	3.17^g	0.54^e	0.26^{de}	0.97^{bc}
T_{10}	T_3+ MgCO$_3$ @ 80% of T_5	16.35^b	3.63^{ef}	0.58^{de}	0.26^{de}	1.06^{bc}
T_{11}	T_3+ MgCO$_3$ @ 125% of T_5	12.13^{de}	4.31^c	0.64^{cd}	0.27^d	1.19^{bc}
T_{12}	T_3+ MgCO$_3$ @ 150% of T_5	10.89^{ef}	4.11^{cd}	0.65^c	0.29^{bcd}	1.12^{bc}
	Treatment means with common superscript do not differ significantly					

4.2.6. Effect of treatments on biometric parameters

The effect of treatments on the biometric parameters of crop are shown in Table 4.29.

4.2.6.1 Days to germination of cowpea seeds

The results of pot culture experiment showed that there was no significant difference among treatments with respect to no of days to germination of seeds.

4.2.6.2. Plant height

Significantly higher plant height was obtained in T_7 (T_3+ MgCO$_3$ @ 20% of T_5) with a mean value of 61.65cm and followed by T_9 which was on par with T_4. The lowest plant height was recorded in absolute control.

4.2.6.3. Number of branches per plant

Treatments T_4, T_7, T_9, T_{10}, T_{11}, and T_{12} were on par in recording significantly higher number of branches at harvest stage

4.2.6.4. Number of root nodules

Analysis of the data showed a significant influence of treatments on root nodule formation. The number of nodules was higher during flowering than after harvest. Among various treatments significantly higher number of root nodules was recorded in treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) during flowering stage. Similarly after harvest of crop, significantly higher number of nodules was recorded in T_7 which was on par with T_4, T_5, T_8 and T_9. Absolute control (T_1) and T_2 were on par in recording significantly lower number root nodules at harvest stage.

4.2.6.5. Number of pods per plant

The treatments differed significantly with respect to number of pods per plant. Significantly higher number of pods per plant was obtained in treatment T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) and was on par with T_8, T_6, T_7, T_4, and T_{12}.

4.2.6.6. Length of pods

Length of pods in response of various treatments are furnished in the table 4.30. Significantly longer pods were observed in T_{11} and was on par with T_9 and T_6.

4.2.6.7. Seeds per pod

The data on number of seeds in each pod showed that treatments varied significantly and highest number of seeds per pod were obtained in T_{11} (T_3+ $MgCO_3$ @ 125% of T_5) and was on par with T_9, T_6 and T_5. Number of seeds per pod obtained from absolute control was the lowest and was on par with T_2 (Organic manure @ 20 t ha^{-1}).

Table 4.29. Effect of treatments on biometric parameters of cowpea

Treatments	Days to germination	Plant height (cm)	Branches per plant	Pods per plant	Length of pods (cm)	Seeds per pod	Yield (g plant^{-1})	No of root nodules At flowering	No of root nodules After harvest
T$_1$ -Absolute control	5.5	37.90^f	2.0^c	8.25^f	9.60^g	8.00^e	33.04^e	20.5^g	5.0^f
T$_2$ -Organic manure @ 20 t ha^{-1}	4.5	45.70^e	3.0^b	10.5^{ef}	11.90^f	8.50^e	60.84^d	27.0^{efg}	8.5^{ef}
T$_3$ -POP recommendation with CaCO3 @250 kg ha^{-1}	5.0	44.00^e	2.0^c	9.00^{de}	12.80^{ef}	10.50^d	63.28^d	29.5^{def}	12.5^{cde}
T$_4$ -POP recommendation with dolomite@ 400kg ha^{-1}	4.5	56.95^b	4.0^a	18.75^{abc}	15.95^{bc}	12.50^c	73.33^{bc}	41.0^b	18.5^{ab}
T$_5$ -T$_3$+ MgCO$_3$ to raise available Mg to 120mg kg^{-1}	5.0	49.80^d	3.0^b	21.75^a	16.35^{abc}	14.50^{ab}	76.12^{abc}	35.0^{bcd}	17.0^{ab}
T$_6$ -T$_3$+ MgCO$_3$ @10% of T$_5$	4.5	52.80^c	3.0^b	19.5^{abc}	17.70^a	14.50^{ab}	70.39^c	32.0^{cde}	14.5^{bcd}
T$_7$ -T$_3$+ MgCO$_3$ @ 20% of T$_5$	4.5	61.65^a	4.0^a	19.50^{abc}	15.80^{bc}	13.50^{bc}	79.33^a	50.0^a	20.5^a
T$_8$ -T$_3$+ MgCO$_3$ @ 40% of T$_5$	5.0	49.65^d	3.0^b	20.25^{ab}	15.05^{cd}	13.00^{bc}	76.57^{ab}	36.5^{bc}	17.0^{ab}
T$_9$ -T$_3$+ MgCO$_3$ @ 60% of T$_5$	4.0	58.95^b	4.0^a	17.25^{bc}	17.00^{ab}	14.50^{ab}	75.28^{abc}	26.0^{efg}	17.5^{ab}
T$_{10}$ -T$_3$+ MgCO$_3$ @ 80% of T$_5$	4.0	48.40^d	4.0^a	18.00^{bc}	15.50^c	13.50^{bc}	73.30^{bc}	27.5^{ef}	15.5^{bc}
T$_{11}$ -T$_3$+ MgCO$_3$ @125% of T$_5$	4.0	48.25^d	3.5^{ab}	16.50^{cd}	17.70^a	15.50^a	74.64^{abc}	27.5^{ef}	11.0^{de}
T$_{12}$ - T$_3$+ MgCO$_3$@150% of T$_5$	4.5	49.55^d	3.5^{ab}	18.75^{abc}	13.95^{de}	12.50^c	74.19^{abc}	25.0^{fg}	12.5^{cde}

Treatment means with common superscript do not differ significantly

4.2.6.8. Yield per plant

The treatments differed significantly with respect to the yield per plant. Treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) recorded significantly higher yield but was on par with T_5, T_8, T_9, T_{11} and T_{12} and the absolute control treatment recorded the lowest yield.

4.2.6. Critical limit of magnesium in soil and cowpea plant

The computed data for delineation of critical level of magnesium in soil and plant are presented in table 4.30. Critical limit of Mg was worked out by the method proposed by Cate and Nelson (1965)

Table 4.30. Parameters for determination of critical level

	Treatments	Relative Yield (%)	Av. Mg in soil at Flowering (mg kg^{-1})	Mg in leaf at flowering (%)
T_1	Absolute control	41.65	55.58	0.35
T_2	Organic manure @ 20 t ha^{-1}	76.68	68.90	0.44
T_3	POP recommendation with $CaCO_3$ @ 250 kg ha^{-1}	79.76	73.78	0.44
T_4	POP recommendation with dolomite @ 400 kg ha^{-1}	92.43	76.90	0.41
T_5	T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}	95.90	105.25	0.46
T_6	T_3+ $MgCO_3$ @10% of T_5	88.73	67.20	0.36
T_7	T_3+ $MgCO_3$ @ 20% of T_5	100	75.90	0.39
T_8	T_3+ $MgCO_3$ @ 40% of T_5	96.52	77.85	0.43
T_9	T_3+ $MgCO_3$ @ 60% of T_5	94.89	81.78	0.43
T_{10}	T_3+ $MgCO_3$ @ 80% of T_5	92.39	93.05	0.46
T_{11}	T_3+ $MgCO_3$ @ 125% of T_5	94.08	101.10	0.47
T_{12}	T_3+ $MgCO_3$ @ 150% of T_5	93.51	123.48	0.45

Fig. 4.1. Critical level of magnesium in soil and leaves of cowpea at flowering

Fig 4.1a Critical level of Mg in soil

Fig 4.1b Critical level of Mg in cowpea leaves at flowering
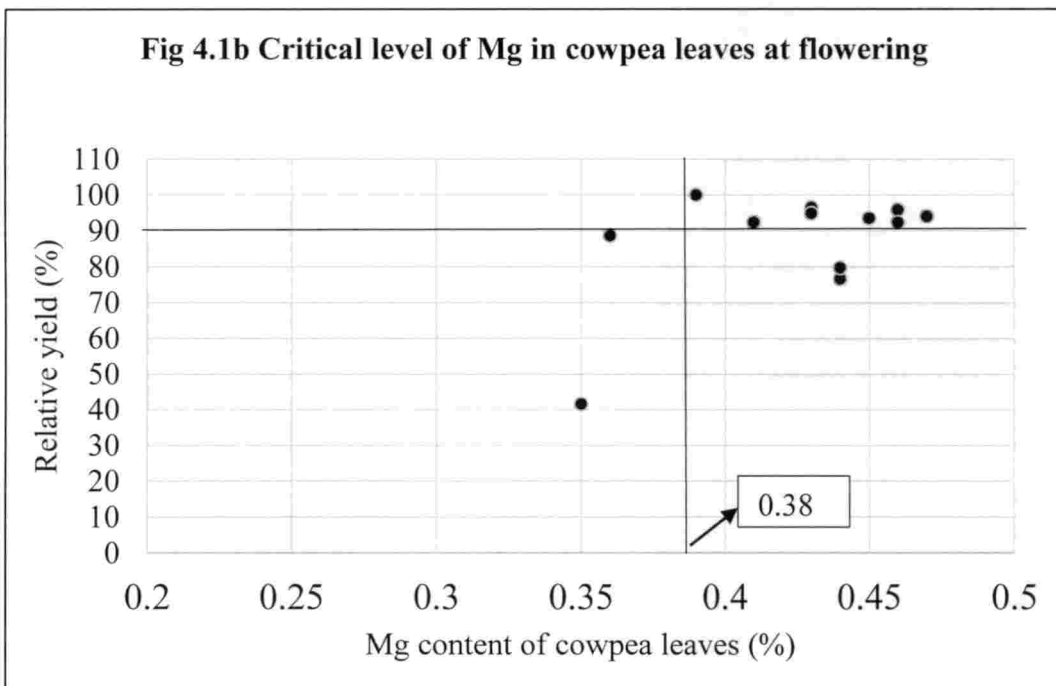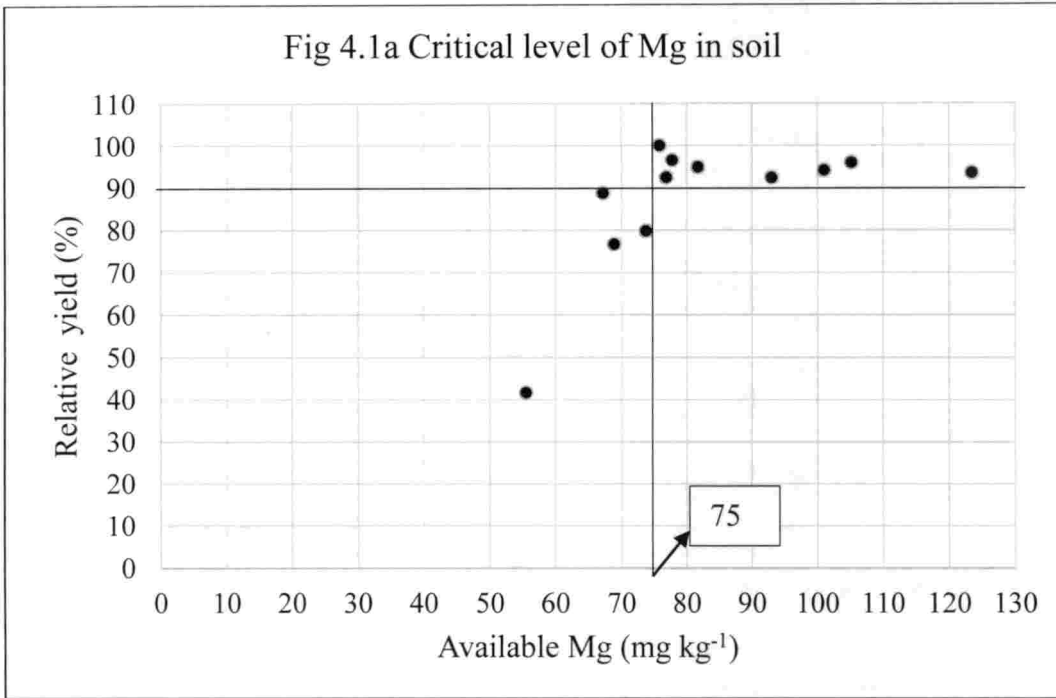
Scattered plot of soil magnesium and relative per cent yield revealed the critical level of magnesium in soil at flowering to be 75 mg kg^{-1} (Fig. 4.1a). Similarly the plots of relative per cent yield and magnesium content in leaves (Fig 4.1b) identified critical level of magnesium in cowpea leaves at flowering as 0.38 %.

Discussion

5. DISCUSSION

Ultisols occupying more than fifty per cent of the total geographical area of Kerala are characterized by low pH, low effective cation exchange capacity and base saturation. Under acidic conditions, crop yield is often limited due to nutritional disorders. Earlier studies have documented magnesium to be a critical nutrient posing constraint to soil fertility in Kerala. Under these circumstances the present study entitled "Optimization of soil environment and crop response for magnesium nutrition in Ultisol" was undertaken at Radiotracer Laboratory", College of Horticulture, Vellanikkara.

The study comprised of two experiments- 1. Incubation study to analyse the release of magnesium from magnesium carbonate 2. Pot culture experiment with cowpea as a test crop to study the response of crop to magnesium nutrition. The results of the experiments presented in chapter 4 are discussed here with supporting literature wherever possible.

5.1. Experiment 1. Incubation experiment

5.1.1. Effect of treatments on pH of soil

Data in table 4.1 shows the effect of different levels of magnesium carbonate with or without calcium carbonate and organic matter on soil pH at weekly intervals of incubation for sixteen weeks. Significantly higher soil pH was recorded in treatment T_{12} ($O_1L_1M_3$) with the addition of calcium carbonate (250 kg ha^{-1}), organic manure (20 t ha^{-1}) and magnesium carbonate at 150% of the optimum dose required to theoretically raise the available magnesium status to 120 mg kg^{-1}. The significant rise in soil pH is due to the addition of both calcium carbonate with neutralizing value of 100% and magnesium carbonate with 118.61%. Significant difference in soil pH between T_6 ($O_0L_1M_3$) and T_{12} ($O_1L_1M_3$) indicates the release of basic cations from vermicompost. Rise in pH due to application of vermicompost was attributed by Bekele

88

et al. (2018) to its high content of basic cations and slightly alkaline pH, which could reduce soil acidity through replacing the acidic cations from the exchange sites.

Over the period of incubation significantly higher soil pH (5.06) was observed at one week after incubation indicating the efficacy of the liming materials added. The calcium and magnesium released from $CaCO_3$ and $MgCO_3$ replace H^+ ions on the exchange sites which is neutralized by the anion CO_3^{2-}. Watling *et al.* (2010) showed that liming materials of size less than 0.5mm can raise soil pH within one week of application. Fig. 5.1 shows the effect of treatments on soil pH during the incubation period. The soil pH at the end of the experiment is higher than the initial soil pH in all the treatments indicating the persistence of magnesium carbonate throughout the incubation period.

5.1.2. Effect of treatments on EC of soil

Table 4.2 shows the variation in the electrical conductivity of soil as influenced by different treatments over the period of incubation. The treatments imposed produced significant changes in soil EC over the period of time. The treatment T_{12} with the addition of calcium carbonate (250 kg ha^{-1}), organic manure (20 t ha^{-1}) and magnesium carbonate at 150% of the optimum dose required to theoretically raise the available magnesium status to 120 mg kg^{-1} recorded significantly higher (0.18 dS m^{-1}) soil EC, which can be attributed to the addition of higher quantity of bases to the soil. The effect of incubation period on soil EC revealed significantly higher EC (0.20 dS m^{-1}) at the end of the incubation period. This substantiates the release of basic cations throughout the incubation period.

5.1.3. Effect of treatments on available magnesium content in soil

Available magnesium showed significant difference among treatments. Significantly higher status of available magnesium was recorded in treatment T_{12} ($O_1L_1M_3$) which was on par with T_9 ($O_1L_0M_3$) which is obviously due to the addition

of higher level of magnesium carbonate in these treatments. However T_6 ($O_0L_1M_3$) with same level of magnesium carbonate recorded significantly lower content of available magnesium indicating the release of magnesium from added vermicompost.

Over the period of incubation significantly higher content of available magnesium was recorded at eighth weeks after incubation (133.89 mg kg^{-1}) and was on par with the status of the nutrient at seven and nine weeks after incubation. This indicates the period of maximum release from magnesium carbonate added. A further reduction noted might be due to release of other cations from the exchange sites to maintain the equilibrium between the soil solid phase and solution phase.

Fig. 5.1. Effect of treatments on soil pH over the incubation period

Fig. 5.1a. Effect of different levels of $MgCO_3$ on soil pH at 0kg ha^{-1} $CaCO_3$ and 0t ha^{-1} organic manure
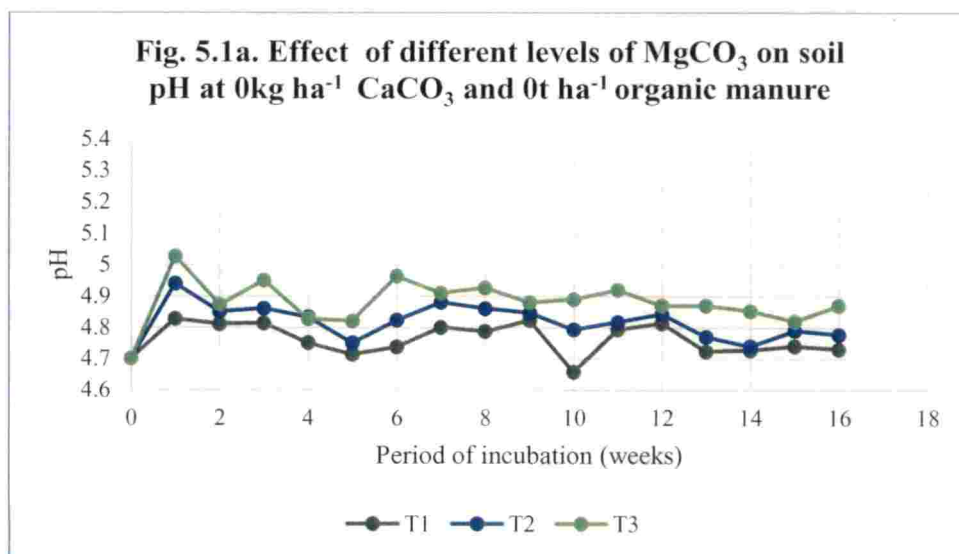
Fig. 5.1b. Effect of different levels of $MgCO_3$ on soil pH at 250kg ha^{-1} $CaCO_3$ and 0t ha^{-1} organic manure
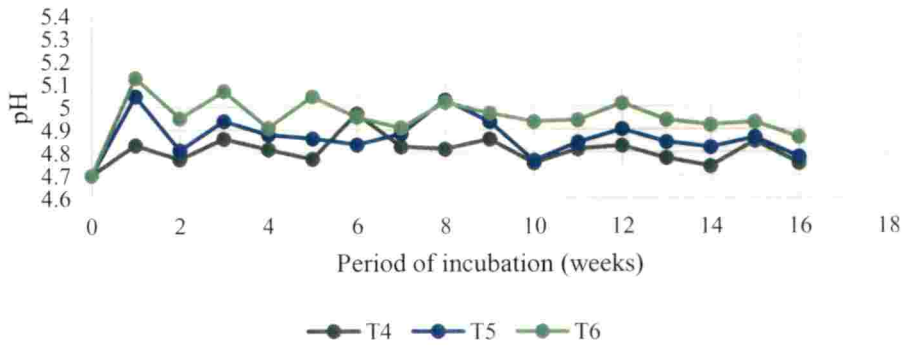
Fig. 5.1c. Effect of different levels of $MgCO_3$ on soil pH at 0kg ha^{-1} $CaCO_3$ and 20t ha^{-1} organic manure
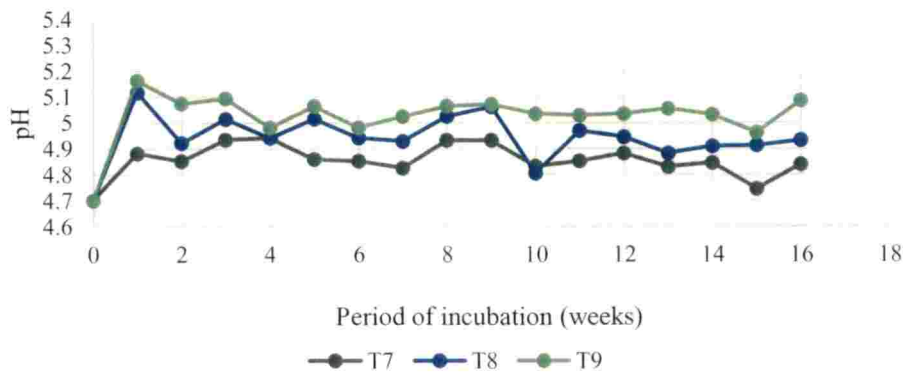
Fig. 5.1d. Effect of different levels of $MgCO_3$ on soil pH at 250kg ha^{-1} $CaCO_3$ and 20t ha^{-1} organic manure
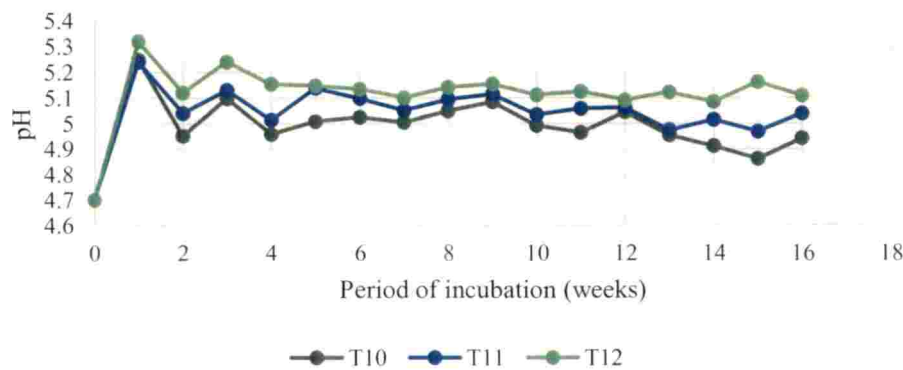
Fig. 5.2. Effect of treatments on soil EC over the period of incubation

Fig. 5.2a. Effect of different levels of $MgCO_3$ on soil EC at 0kg ha^{-1} CaCO$_3$ and 0t ha^{-1} organic manure

Fig. 5.2b. Effect of different levels of $MgCO_3$ on soil EC at 250kg ha^{-1} CaCO$_3$ and 0t ha^{-1} organic manure

Fig. 5.2c. Effect of different levels of $MgCO_3$ on soil EC at 0kg ha^{-1} CaCO$_3$ and 20t ha^{-1} organic manure

Fig. 5.2d. Effect of different levels of MgCO$_3$ on soil EC at 250kg ha^{-1} CaCO$_3$ and 20t ha^{-1} organic manure

Period of incubation (weeks)

— T10 — T11 — T12

Fig. 5.3. Effect of treatments on available Mg over the period of incubation

Fig.5.3a. Effect of different levels of MgCO$_3$ on soil available Mg at 0kg ha$^{-1$ CaCO$_3$ and 0t ha^{-1} organic manure

Period of incubation (weeks)

— T1 — T2 — T3

93
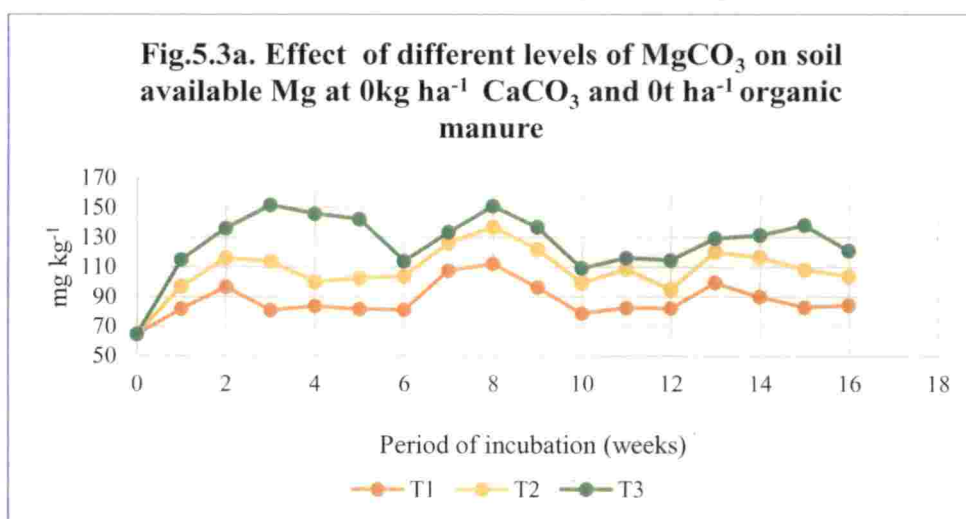
Fig. 5.3b. Effect of different levels of $MgCO_3$ on soil available Mg at 250kg ha^{-1} $CaCO_3$ and 0t ha^{-1} organic manure
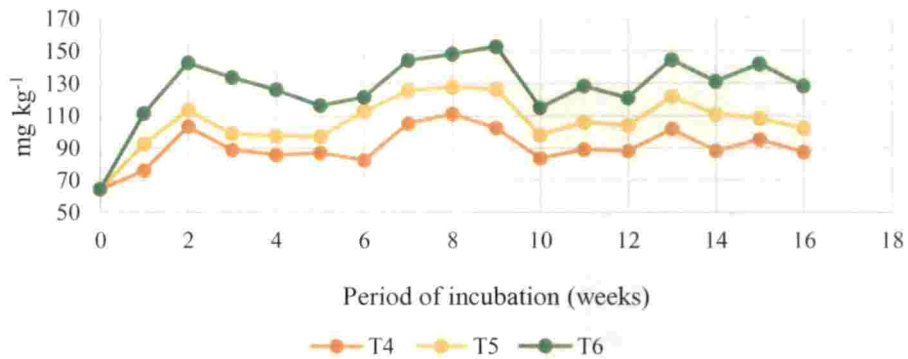
Fig. 5.3c. Effect of different levels of $MgCO_3$ on soil available Mg at 0kg ha^{-1} $CaCO_3$ and 20t ha^{-1} organic manure

Fig. 5.3d. Effect of different levels of $MgCO_3$ on soil available Mg at $0kg\ ha^{-1}$ $CaCO_3$ and $20t\ ha^{-1}$ organic manure
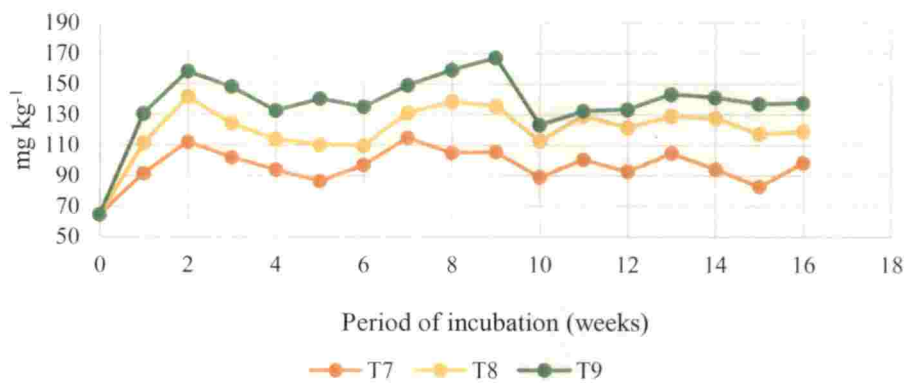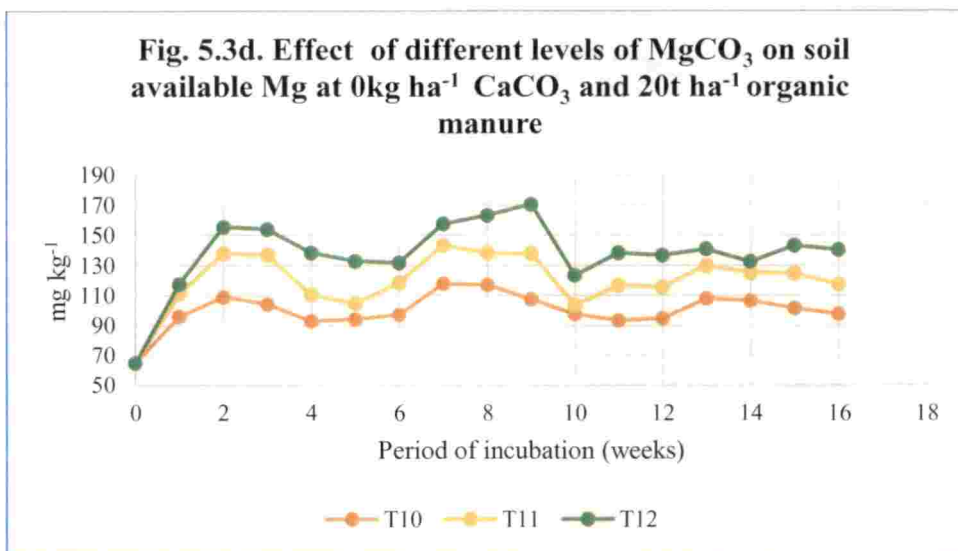
5.1.4. Effect of treatments on fractions of magnesium in soil

The effect of treatments on different fractions of magnesium are presented in Table 4.4.

The addition of different levels of magnesium carbonate produced significant differences in the water soluble fraction. Significantly higher content of this fraction was recorded in treatment T_{12} with the treatment combination of $O_1L_1M_3$ and was on par with T_8 ($O_1L_0M_2$). The highest content of exchangeable magnesium was also observed in T_{12} ($O_1L_1M_3$) and was on par with T_9 ($O_1L_0M_3$). This might be because of higher release of magnesium from this treatment with higher level of added magnesium carbonate. The water soluble fraction is always in a state of dynamic equilibrium with exchangeable fraction.

The organic-complexed fraction was significantly higher in T_{12} and T_{11} which might have been contributed through higher microbial biomass in these treatments due to a significant increase in soil pH. The higher content of acid soluble and mineral fraction in the treatments in comparison to the initial status substantiates the presence of magnesium carbonate as a solid phase. White and Munro (1981) reported a release

of 43 % of added magnesium from dolomite after 200 days in a pot culture experiment with soil of pH 5.5.

5.2. Experiment 2. Pot culture experiment

5.2.1. Effect of treatments on soil properties during flowering and harvest of cowpea crop

Soil samples were collected during flowering and after harvest and were analyzed for various parameters and results obtained are discussed below.

5.2.1.1. Electro-chemical properties and organic carbon (OC) content in soil

Soil pH measured during flowering and after harvest was found to differ significantly between treatments. At both the stages treatment T_{12} recorded significantly higher soil pH. The highest dose (150 % of $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) of magnesium carbonate was applied in this treatment (Fig 5.4). Magnesium carbonate with a neutralizing value of 118.61% had resulted in the increase in soil pH. An increase in the soil pH was observed after crop harvest in comparison to the flowering stage in all treatments except absolute control. This might be due to slow solubility of magnesium carbonate. The solubility of dolomite/ magnesite was found to be 87 % less than kieserite three weeks after application (Senbayram *et al.,* 2015).

The organic carbon content in soil increased from the initial value of 1.32 % in all treatments except absolute control which can be attributed to the addition of vermicompost @ 20 t ha^{-1}. Significantly higher content of organic carbon was recorded in T_3 (2.54%) and T_4 (2.54%) which was on par with T_2 (2.51%) (Fig. 5.5). This can be attributed to the lower population of free living nitrogen fixers in these treatments as these organisms derive energy from organic molecules in soil (Wagner, 2011) (Table 4.13). Similar results were reported by Swanepoel *et al.* (2011). Though there was a reduction in the organic carbon status after crop harvest, significantly higher content was recorded in treatment T_3.

Fig. 5.4 Effect of treatments on soil pH

Fig. 5.5 Effect of treatments on soil organic carbon
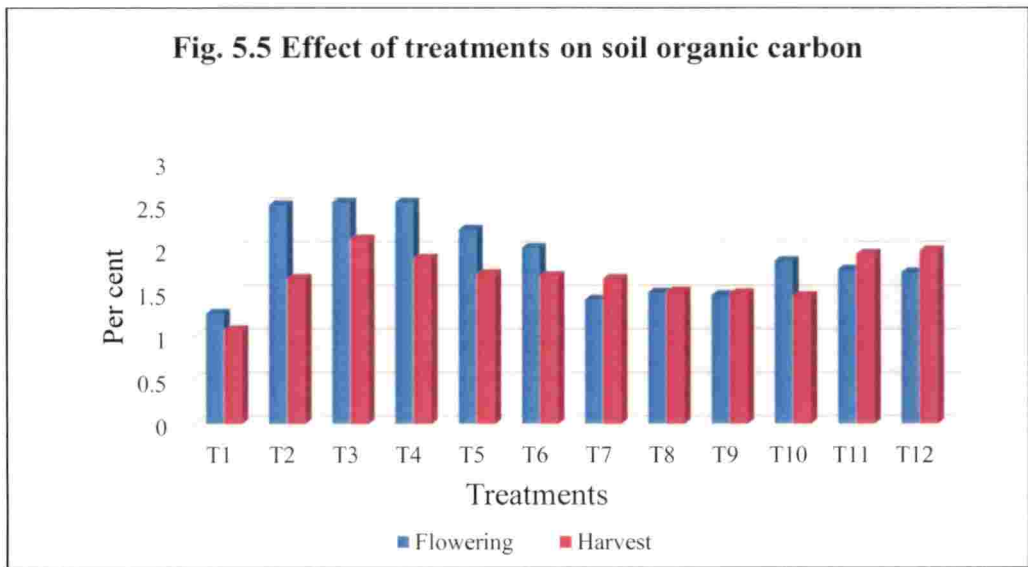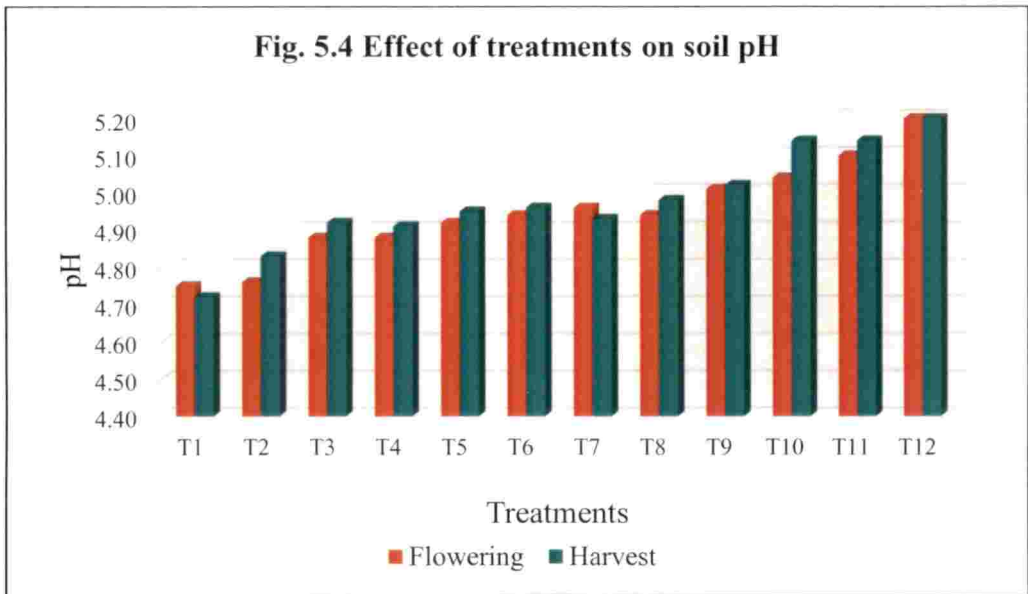
5.2.1.2. Status of primary nutrients in soil

Available nitrogen in soil was significantly higher in treatment T_3 during flowering and after harvest of crop which can be attributed to the higher organic carbon status of soil in this treatment. Organic carbon content of the soil is taken as the index of nitrogen supplying power as the C: N ratio is usually stabilized at 10:1 to 12:1 under

tropical humid climate (Sureshkumar *et al.*, 2018; John, 2014). Fig 5.6 shows the effect of treatments on primary nutrient status of soil.

Fig. 5.6. Effect of treatments on available N, P and K in soil

Fig. 5.6a. Effect of treatments on available N, P and K at flowering

Fig. 5.6b. Effect of treatments on available N, P and K at harvest
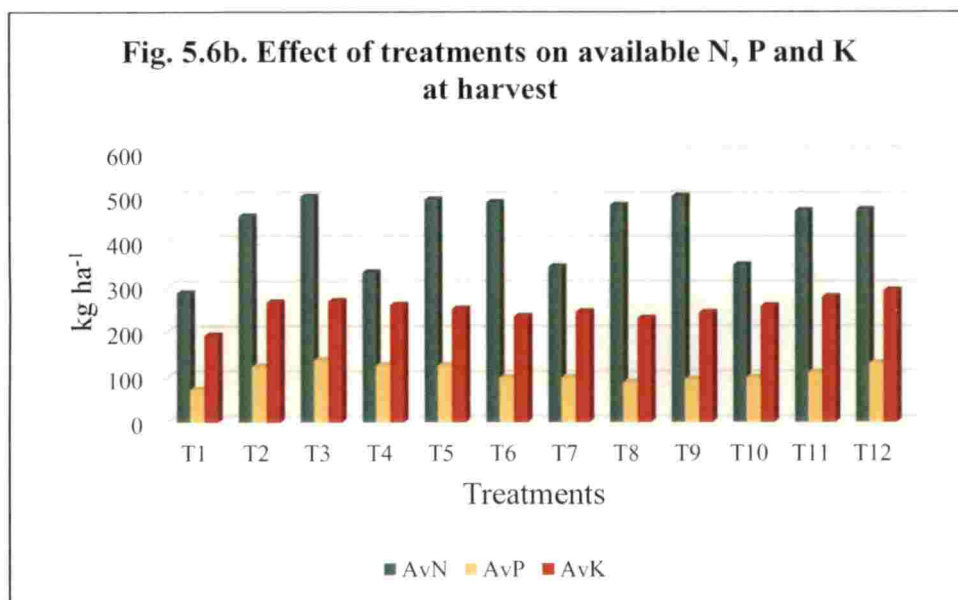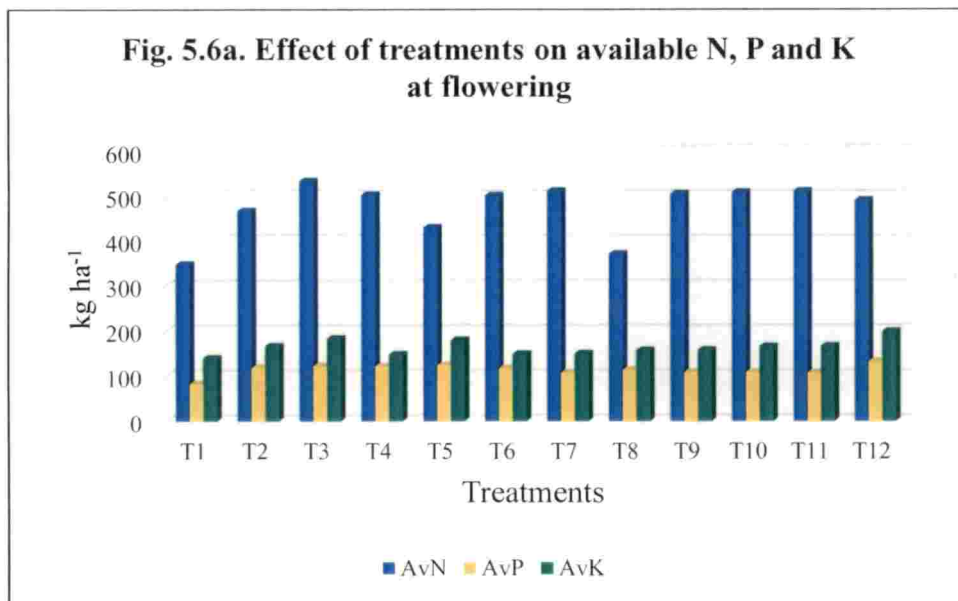
Significantly higher content of available phosphorus was recorded in treatment T_{12} which might be due to the increase in soil pH. Fageria *et al.* (2008) reported an increase in available phosphorus as pH increased to above 5.0, due to release of P ions from Al and Fe oxides. Adams (1980) reported the occurrence of positive correlation and interactions between phosphorus and magnesium in soil and that Mg helps in greater solubilisation of phosphorus in soil. The available phosphorus content in soil was significantly higher in T_3 after harvest of crop, which can be attributed to the lower uptake of phosphorus by crop in this treatment (Table 4.27). Available potassium was significantly higher in T_{12} where the highest level of magnesium carbonate was applied (T_3+ 150% of $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) at both flowering and after harvest. This might be due to release of potassium from the exchange sites to maintain the equilibrium between the soil solid phase and solution phase. According to Schofield's ratio law the ratio of cations held by the soil and the ratio in equilibrium solution is constant (Sanyal *et al.*, 2009). Hannaway *et al.* (1982) studied the effect of Mg on K translocation in soil and reported that low magnesium status in soil decreases the available K.

5.2.1.3. Status of secondary nutrients in soil

Initial status of available calcium in soil was sufficient at 429.30 mg kg^{-1}. The treatments imposed resulted in significant variations between treatments. Available calcium level increased from the initial level in all treatments except absolute control (T_1) due to release of calcium from calcium carbonate and/or organic manure (Fig. 5.7). Significantly higher content of available calcium was recorded in T_{10} (T_3+ $MgCO_3$ @ 80% of T_5) and was on par with T_5, followed by all other treatments except T_1 and T_2, where calcium carbonate was not added. Further increase in available calcium in soil was observed after the crop harvest, which might indicate the release of calcium from calcium carbonate and/or organic manure. Though there are conflicting information concerning reaction time of limestone in acid soils, Jones and Mallarino (2018)

reported significant influence of reagent grade calcium carbonate in soil after 200 days of incubation though significant increase in pH was realized within 10 days.

Fig 5.7. Effect of treatments on available calcium

Fig. 5.7. Effect of treatments on available Ca during flowering and harvest

Fig. 5.8. Effect of treatments on available magnesium

Fig. 5.8. Effect of treatments on available Mg during flowering and harvest
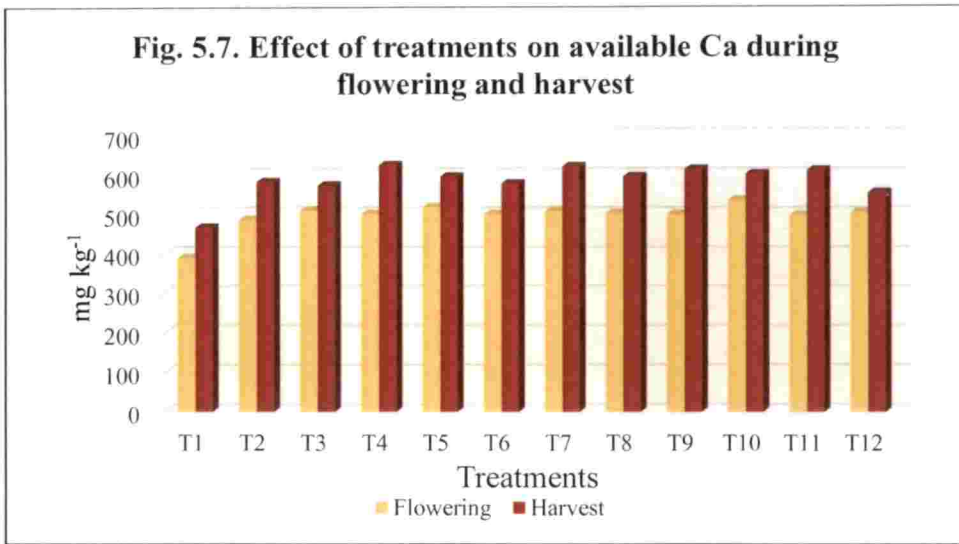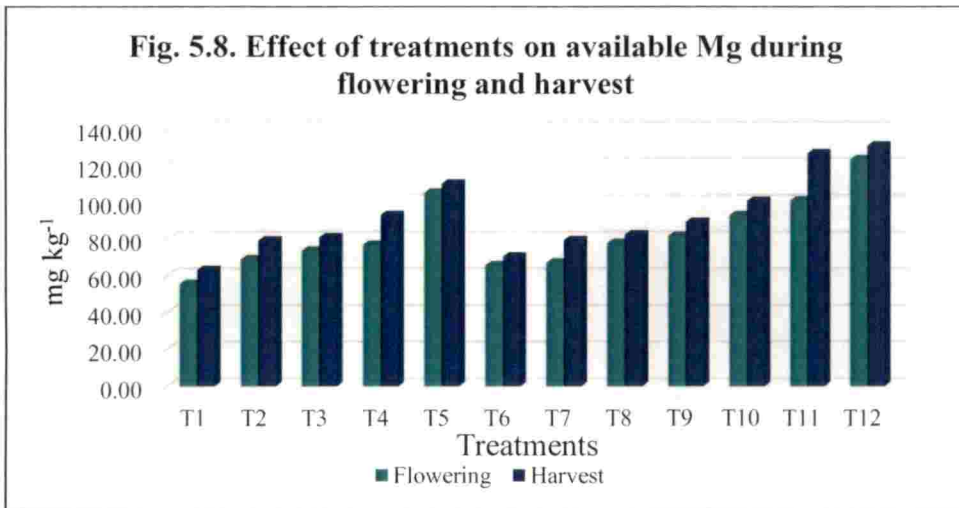
The variations in available Mg content in soil at both the stages of analysis corresponded to the gradation in magnesium through added sources with the highest content in T_{12} (T_3 + $MgCO_3$ @ 150% of T_5) (Fig. 5.8). An increase in available magnesium status at crop harvest when compared to flowering stage indicates the

release of magnesium from magnesium carbonate. Further, increase in available pool of nutrients on maintaining soil at field capacity was reported by Salmon (1963). Soil was initially found to be sufficient with respect to available sulphur, while a decline was recorded at flowering stage which might be due to crop uptake (Table 4.9). Significantly lower status was recorded in absolute control. This might be because of crop uptake. A further increase in available status at crop harvest might be due to release from organic pools in soil.

5.2.1.4. Status of micronutrients in soil

The available status of micronutrients (Fe, Mn, Zn and Cu) was found to be sufficient in soil initially as well as during different crop stages. Though the treatments imposed were found to produce significant variations in their status in soil, no definite pattern could be identified (Table 4.11 and 4.12). Available boron was deficient initially whereas it was found to be sufficient in all the treatments except absolute control during both the stages. This might be due to the release from organic manure added (20 t ha^{-1}) which contained 64.40 mg kg^{-1} of boron. Further the application of solubor as per *adhoc* recommendation of POP would have contributed to available pool. The increase in the available pool from flowering stage to harvest points to the release from organic pools.

5.2.2. Effect of treatments on microbial population in soil

Microbial population during flowering are presented in Table 4.13. The Rhizobium population was absent in initial soil. However during flowering stage of the crop rhizobial population could be recorded and significant variations were observed among treatments (Fig.5.9a). This might be due to seed inoculation with Rhizobium culture. Similarly the population of free living nitrogen fixers also showed an increase from the initial population of 8.00×10^5 cfu/ g to 149×10^5 cfu/ g of soil (Fig. 5.9b). Significantly higher population of both Rhizobium and free living nitrogen fixers were

recorded in treatment T_7 (T_3+ $MgCO_3$ @20% of T_5) both during flowering and harvest (Plate 3 and 4). Dechen *et al.* (2015) reported that calcium and magnesium are essential elements for efficient nitrogen fixation by rhizobia and Mg deficiency results in reduced nitrogen fixation. However, a linear response of nitrogen fixers to magnesium availability could not be recorded in this study. According to Peng *et al.* (2018), nodule growth under nitrogen limited conditions was enhanced by external Mg supply due to higher partitioning of photosynthates to roots as nitrogen fixation require large amount of energy. The medium to high status of available N in the experimental soil can be attributed to the lack of linear response to graded dose of magnesium.

Fig 5.9. Effect of treatments on microbial population in soil

Fig. 5.9a. Effect of treatments on Rhizobium in soil

Fig. 5.9b. Effect of treatments on freeliving N fixing bacteria in soil
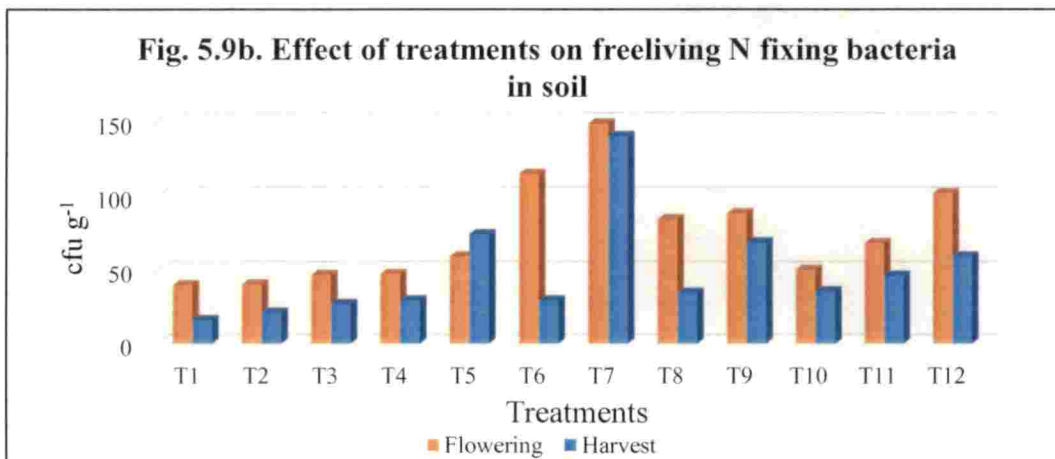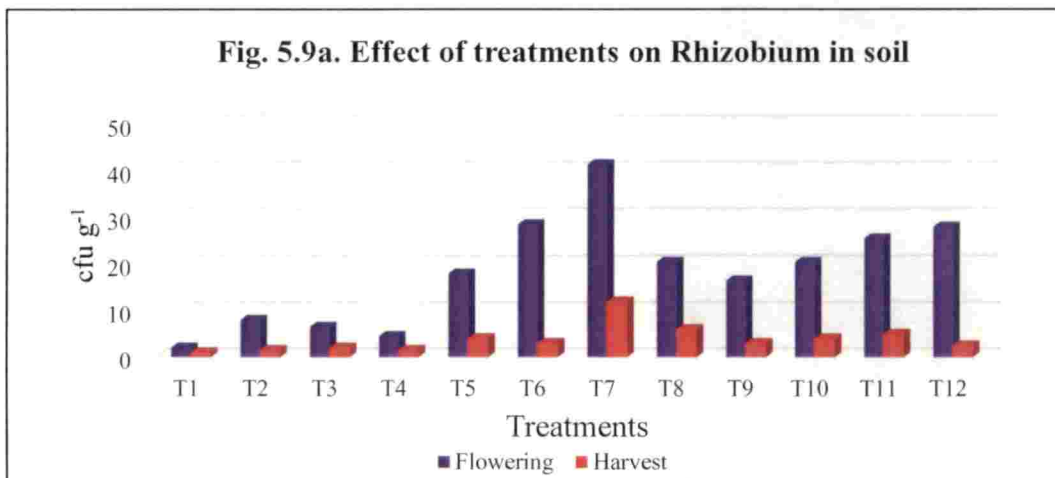
T_7 (T_3+ MgCO$_3$ @20% of T$_5$) T_6 -T_3+ MgCO$_3$ @10% of T$_5$
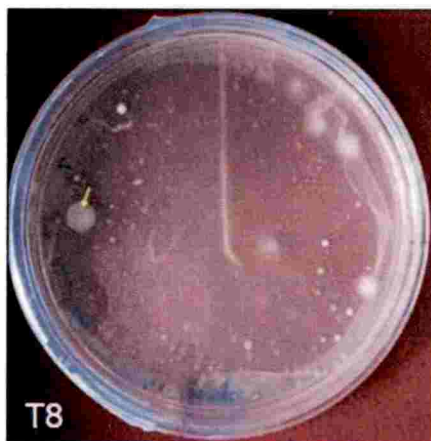
Flowering

T_7 (T_3+ MgCO$_3$ @20% of T$_5$) T_8- T_3+ MgCO$_3$ @ 40% of T$_5$

Harvest

Plate 3. Effect of treatments on Rhizobium population in soil

T7 (T3+ MgCO3 @20% of T5)   T6 -T3+ MgCO3 @10% of T5

Flowering

T7 (T3+ MgCO3 @20% of T5)   T5 - MgCO3 to raise available Mg to

120 mg kg^{-1}

Harvest

Plate 4. Effect of treatments on population of free living N fixing bacteria in soil
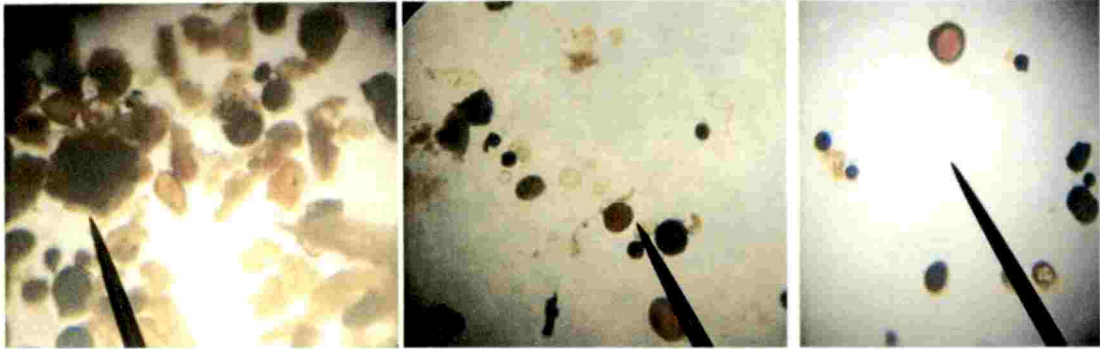
Initial   Flowering   Harvest

T_7 (T_3+ $MgCO_3$ @20% of T_5)

Plate 5 Spore count of Arbuscular Mycorrizal Fungi in soil
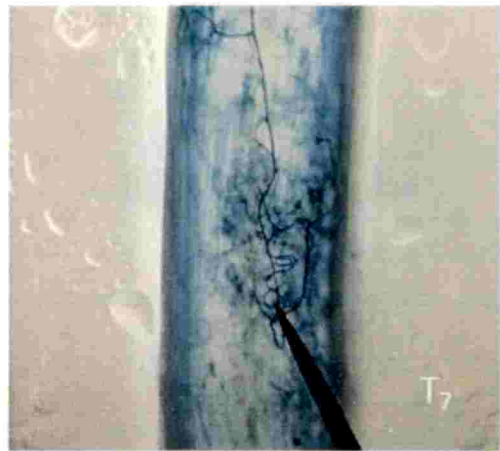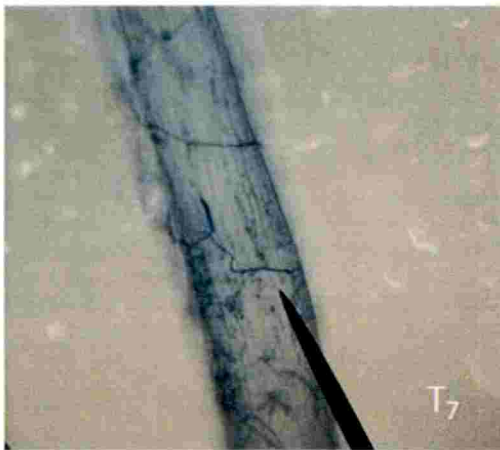
Per cent root colonization

During flowering   During harvest

Plate 5. Effect of treatments on per cent root colonization of Arbuscular Mycorrhizal Fungi

However, the spore count of arbuscular mycorrhizal fungi (AMF) showed no significant variations between treatments, though an increase was observed from the initial value (1067) (Plate 5). However, per cent root colonization with AMF was significantly higher in treatments with higher availability of magnesium. Similar reports were given by Gryndler *et al.* (1992) where they observed pronounced positive effect of magnesium on per cent root colonization of maize and substitution of magnesium by potassium or calcium significantly reduced infection.

5.2.3. Effect of treatments on plant nutrient content during flowering and harvest

5.2.3.1. Primary nutrients

The nitrogen content in the stem of cowpea plant during flowering and stem and leaf during harvest were significantly influenced by the treatments imposed. Whereas the nitrogen content in leaves during flowering and pods did not differ significantly (Fig. 5.10). The highest content (3.18%) of nitrogen in stem was observed in T_2 (Organic manure @ 20 t ha^{-1}). Correspondingly a higher content of available nitrogen was recorded in soil at this stage. While at crop harvest higher content recorded in T_1 (absolute control) indicates a dilution effect of nutrient in other treatments with higher biomass than T_1. According to Fageria, *et al.* (2006) the response to nitrogen in legumes is evidenced through increase in pod number and seed weight. The decrease in nitrogen content in stem and leaf at harvest (Table 4.15) when compared to flowering with higher nitrogen content in pods is due to translocation of N from shoot to grain (Wilhelm *et al.,* 2002). The medium to high status of organic carbon and available nitrogen in soil indicates that nitrogen was not a constraint for crop growth.

Phosphorus content in plant during flowering and harvest (Table 4.16) showed no significant variation in stem during flowering and leaf at harvest. The phosphorus content in cowpea leaves during flowering ranged from 0.32% to 0.19%. Significantly

103

lower content (0.19%) was observed in T_1 (Absolute control) and T_2 (Organic manure @ 20 t ha^{-1}). This might be in response to the soluble source of phosphorus added as per POP recommendation in treatments T_3 to T_{12}. The decrease in P content in stem and leaf at harvest is the dilution effect due to increase in biomass and due to translocation of larger amount to grains (Fageria, 2009). The variations observed in the phosphorus content in pods does not follow a definite pattern.

Fig 5.10. Effect of treatments on nitrogen content in cowpea

Fig 5.10a Effect of treatments on content of N in stem and leaf of cowpea at flowering

5.10b. Effect of treatments on content of N in stem, leaf and pod of cowpea at harvest

Fig. 5.11 Effect of treatments on content of K in cowpea

Fig. 5.11a. Effect of treatments on content of K in cowpea at flowering
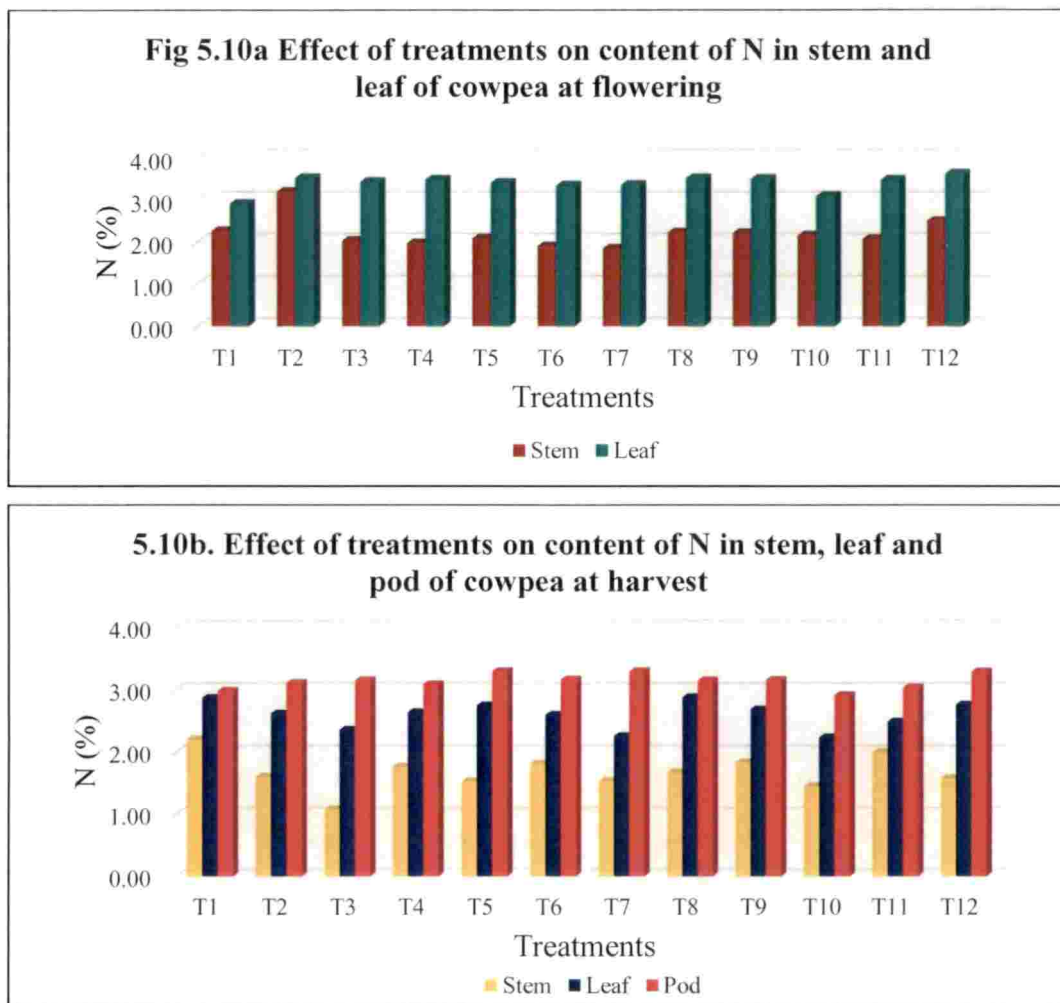
Fig. 5.11b. Effect of treatments on content of K in cowpea at harvest
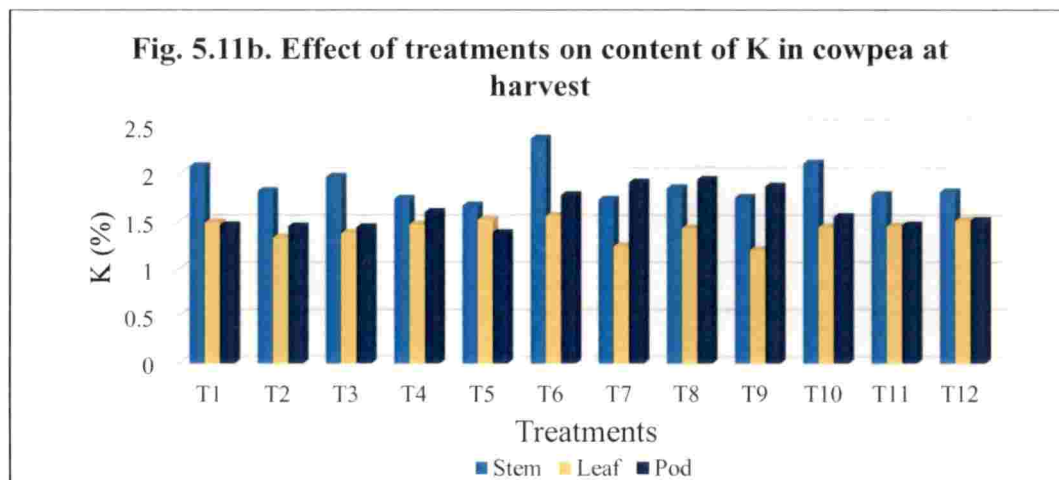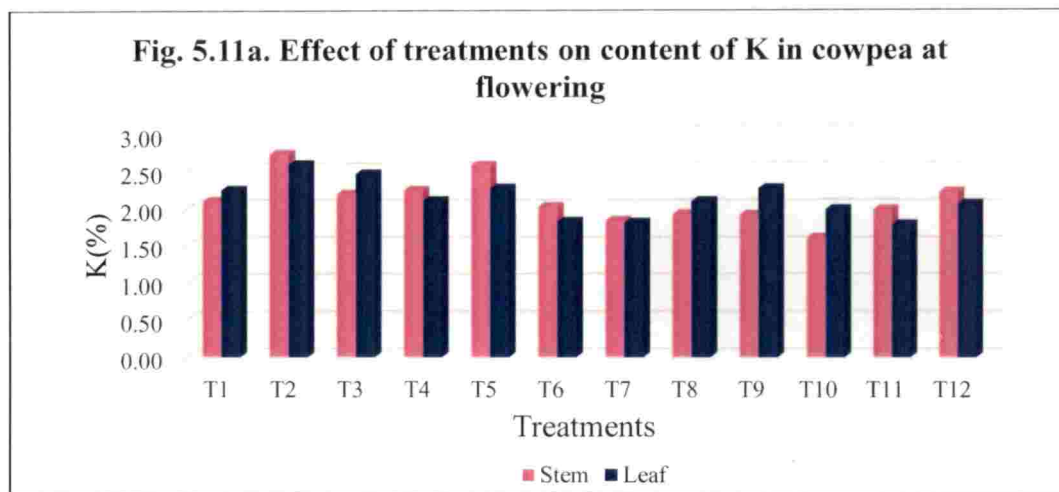
The highest content of potassium in stem and leaf of plants during flowering were in T_2 (Organic manure @ 20 t ha^{-1}) which indicates a dilution effect due to higher biomass in treatments where fertilizers were applied as per recommendation (T_1 to T_{12}). Potassium content in stem and leaf decreased towards harvest of crop (Fig. 5.11). Fageria and Santos (2008) reported that about 50 per cent of accumulated potassium was translocated from shoot to grain in soybean. Potassium content in pods was significantly higher in T_8 (T_3+ MgCO$_3$ @ 40% of T_5) and was on par with treatments T_7 (T_3+ MgCO$_3$ @ 20% of T_5) and T_9 (T_3+ MgCO$_3$ @ 60% of T_5). According to

Narwal *et al.* (1985) Mg up to a concentration of 20 ppm had a synergistic effect on concentration of K in all plant parts but had antagonistic effect at higher concentration.

5.2.3.2. Secondary nutrients

Significant variations between treatments were observed in the calcium content of stem and leaf of cowpea during flowering. Significantly higher content of calcium in stem and leaf during flowering was recorded in T_6 while after harvest higher content of calcium in stem and leaf at harvest was recorded in T_7 (T_3+ $MgCO_3$ @ 20% of T_5) (Fig. 5.12). Treatment T_6 (T_3+ $MgCO_3$ @10% of T_5) recorded significantly higher content and was on par with T_2. Compared to stem, leaves recorded higher content of Ca at both the stages. Calcium content in pods did not differ significantly. According to Karley and White (2009), calcium tends to be present at low concentrations in phloem fed tissues and is retained more in leaves. Fageria (1974) reported suppression in the uptake of calcium at higher concentration of magnesium in groundnut.

Magnesium content in stem and leaf of cowpea at flowering showed magnesium content to increase with the addition of graded dose of magnesium. Similar response to added doses were also observed in the nutrient content analyzed after harvest (Fig 5.13). However the magnesium content in pods did not differ significantly with treatments. Karley and White (2009) reported that magnesium absorbed in excess is stored in the leaves of plants. Canizella *et al.* (2017) reported high positive and significant correlation between rates of magnesium applied and magnesium content in leaves.

The sulphur content in stem at flowering and stem, leaf and pod after harvest did not differ significantly over different treatments. The variations in sulphur content in leaf at flowering is very narrow and cannot be attributed to changes in magnesium

doses. This might be because the available sulphur content at the start of the experiment was in the sufficient range.

Fig. 12. Effect of treatments on calcium content of cowpea

Fig. 5.12a. Effect of treatments on content of Ca in cowpea at flowering

Fig. 5.12b. Effect of treatments on content of Ca in cowpea at harvest

Fig. 5.13. Effect of treatments on content of Mg in cowpea

Fig. 5.13a. Effect of treatments on content of Mg in cowpea at flowering
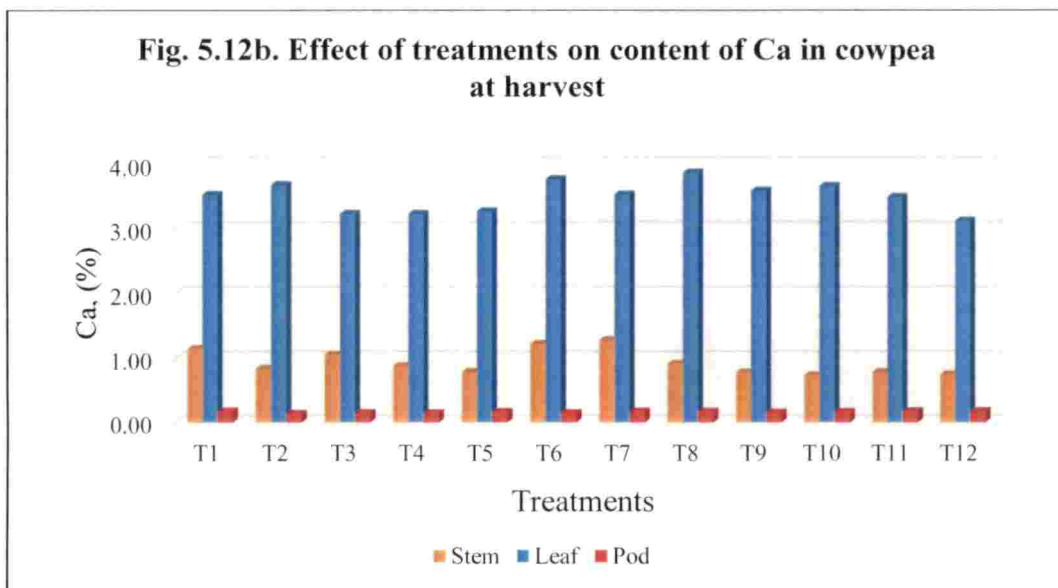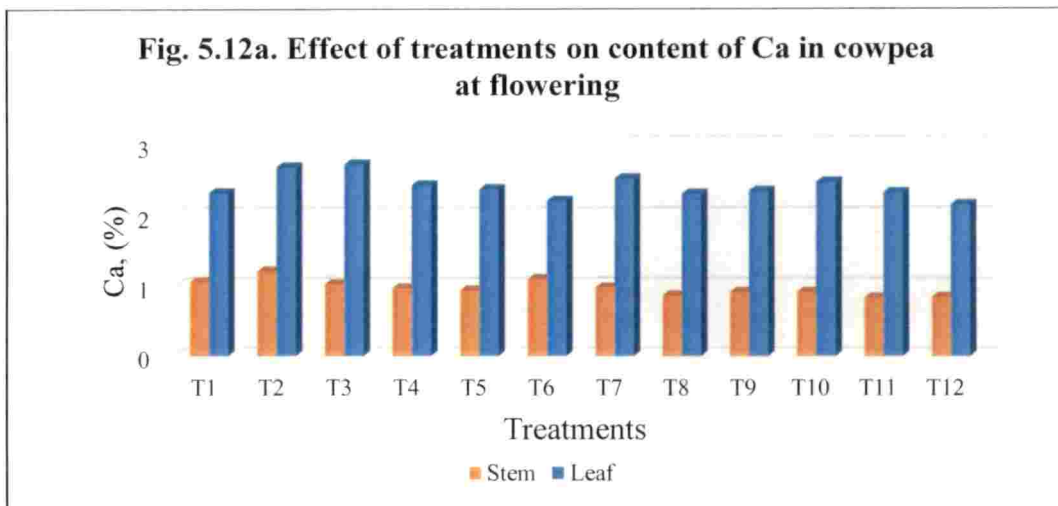
Fig. 5.13b. Effect of treatments on content of Mg in cowpea at harvest
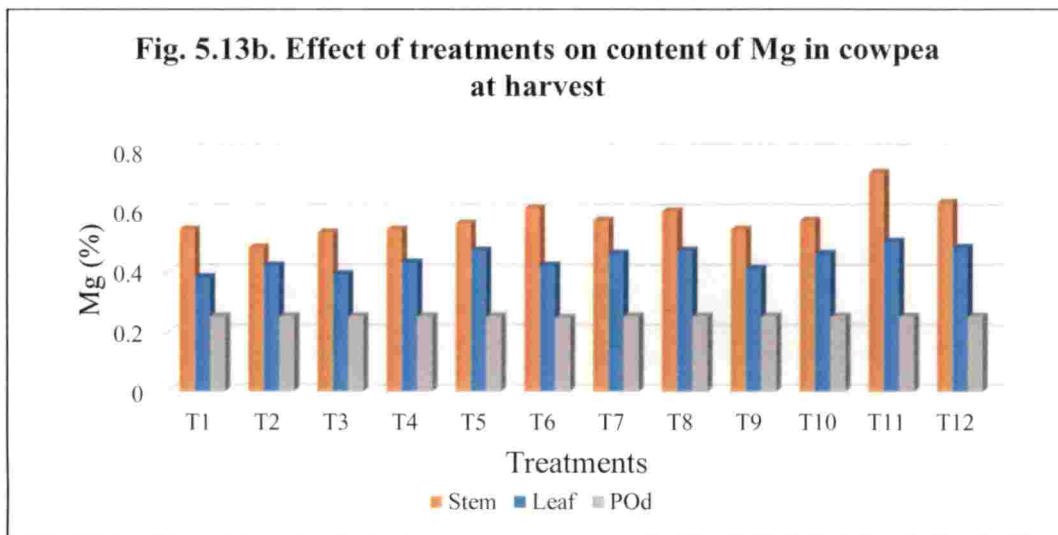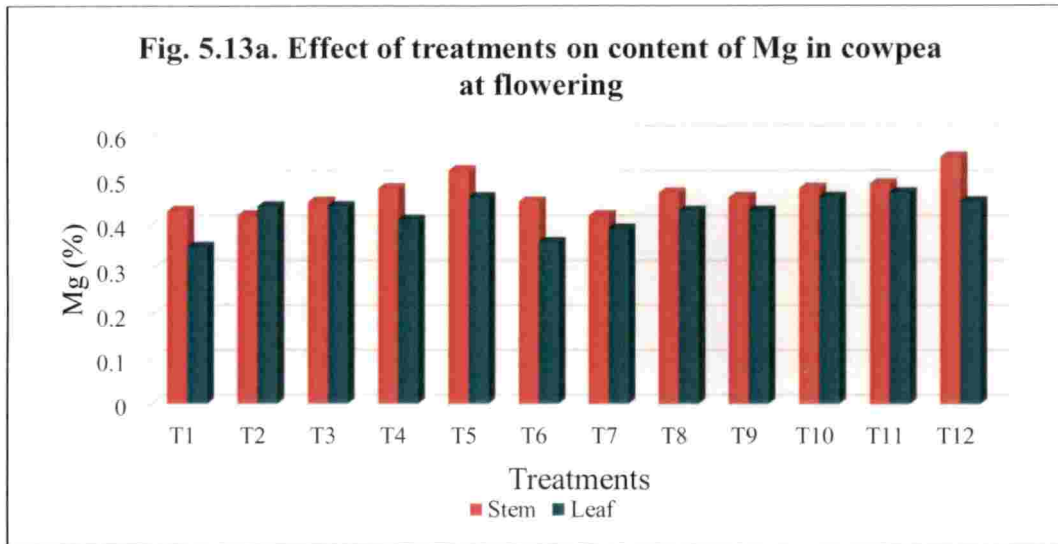
5.2.4.3. Micronutrients

The iron content in the stem of crop at flowering did not vary significantly (Table 4.21). But the treatments imposed produced significant variations in the content of iron in leaf at flowering, stem leaf and pod after crop harvest. The perusal of data shows decrease in plant content of iron with increasing concentration of added

magnesium. Similar was the result in the concentration of manganese in tissue at different stages (Table 4.22). The decrease in concentration of iron and manganese at higher doses of magnesium might be due to cationic competition. Maas *et al.* (1969) reported inhibitory effect of calcium and magnesium on manganese absorption.

The content of zinc and copper in different plant parts did not differ significantly with the treatments imposed. This might be due lower requirement of these nutrients by crop. The availability of both the nutrients were found to be sufficient in soil throughout the crop duration. The boron concentration in tissue at both the stages varied between treatments and significantly lower concentration was recorded in absolute control (T_1). This might be because the available boron status in soil was found to be sufficient in treatments T_2 to T_{12} at both stages. This can be attributed to application of soluble boron source and/or organic manure (20 t ha^{-1}) in all other treatments.

5.2.4. Effect of treatments on chlorophyll content

Chlorophyll a showed significant variation in response of treatments in which highest content was recorded in T_7 (T_3+ $MgCO_3$ @ 20% of T_5) and was on par with T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}). There was no significant variation among treatments in chlorophyll b content in leaf during flowering and chlorophyll a and b in leaf at harvest. No deficiency symptom was noticed in any of the treatments.

5.2.6. Effect of treatments on nutrient uptake

The perusal of data on nutrient uptake by crop reveals, treatment T_6 (T_3+ $MgCO_3$@10% of T_5) to be superior over other treatments to significantly increase the uptake of nitrogen, phosphorus and potassium (Fig. 14.). The comparison of treatments on uptake of calcium showed T_7 to be superior over other treatments. Treatment T_7 recorded significantly higher uptake of magnesium and was on par with T_8 and T_{11}.

The crop uptake of sulphur was significantly higher in T_7 and was on par with T_5, T_6 T_8 and T_{11}.

The uptake of iron was significantly higher in treatment T_6 while T_7 recorded significantly higher uptake of manganese and was on par with T_8. The data on the uptake of zinc also show significantly higher uptake in T_7 on par with T_6. Similarly significantly higher uptake of copper was recorded in T_7 on par with T_6 and T_8. Treatment T_6 and T_7 were on par in recording higher uptake of boron. This can be attributed to higher biomass produced in these treatments. Canizella *et al.* (2017) reported a depressive effect of application of higher dose of magnesium on shoot dry weight of common bean varieties in tropical soils.

Fig. 5.14. Effect of treatments on uptake of macronutrients

Fig. 5.14a. Effect of treatments on uptake of N, P and K

Fig. 5.14b. Effect of treatments on uptake of Ca and Mg
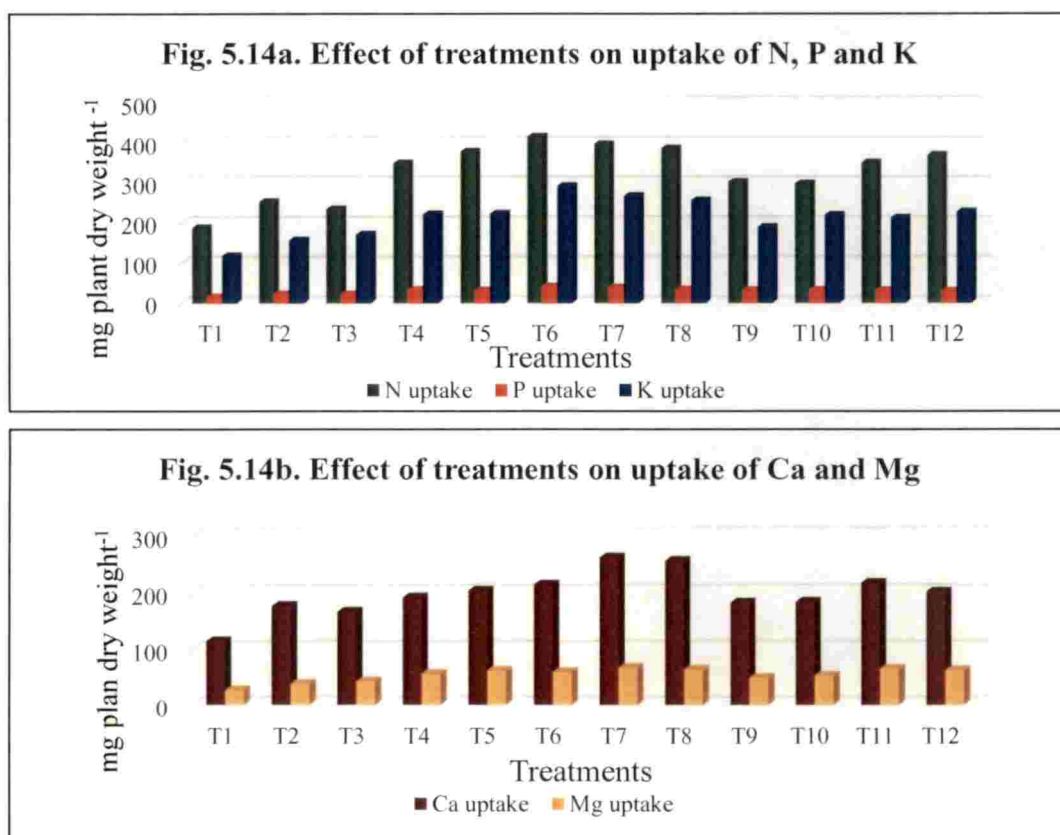
5.2.6. Effect of treatments on biometric parameters

The data on biometric parameters are shown in table 4.29. The results of pot culture experiment showed that there was no significant difference among treatments with respect to no of days to germination of seeds. Significantly higher plant height was obtained in T_7 (T_3+ $MgCO_3$ @ 20% of T_5) with a mean value of 61.65cm and followed by T_9 which was on par with T_4. The lowest plant height was recorded in absolute control (Fig 5.15a). Treatments T_4, T_7, T_9, T_{10}, T_{11}, and T_{12} were on par in recording significantly higher number of branches at harvest stage. Analysis of the data showed a significant influence of treatments on root nodule formation. The number of nodules was higher during flowering than after harvest. Among various treatments significantly higher number of root nodules was recorded in treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) during flowering stage. Similarly after harvest of crop, significantly higher number of nodules was recorded in T_7 which was on par with T_4, T_5, T_8 and T_9. Absolute control (T_1) and T_2 were on par in recording significantly lower number root nodules at this stage. The treatments differed significantly with respect to number of pods per plant (Fig. 5.15b.). Significantly higher number of pods per plant was obtained in treatment T_5 (T_3+ $MgCO_3$ to raise available Mg to 120 mg kg^{-1}) and was on par with T_8, T_6, T_7, T_4, and T_{12}. A significantly long pods was observed in T_{11} and was on par with T_9 and T_6. The data on number of seeds in each pod showed that treatments varied significantly and highest number of seeds were obtained in T_{11} (T_3+ $MgCO_3$ @ 125% of T_5) and was on par with T_9, T_6 and T_5. Number of seeds in the pods obtained from absolute control was the lowest and was on par with T_2 (Organic manure @ 20 t ha^{-1}). The treatments differed significantly with respect to the yield per plant (Fig. 5.15c). Treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) recorded significantly higher yield but was on par with T_5, T_8, T_9, T_{11} and T_{12} and the absolute control treatment recorded the lowest yield.

Fig. 15. Effect of treatments on biometric parameters of cowpea

Fig. 5.15a. Effect of treatments on plant height of cowpea

Fig. 5.15b. Effect of treatments on number of nodules per plant

Effect of treatments on yield of cowpea
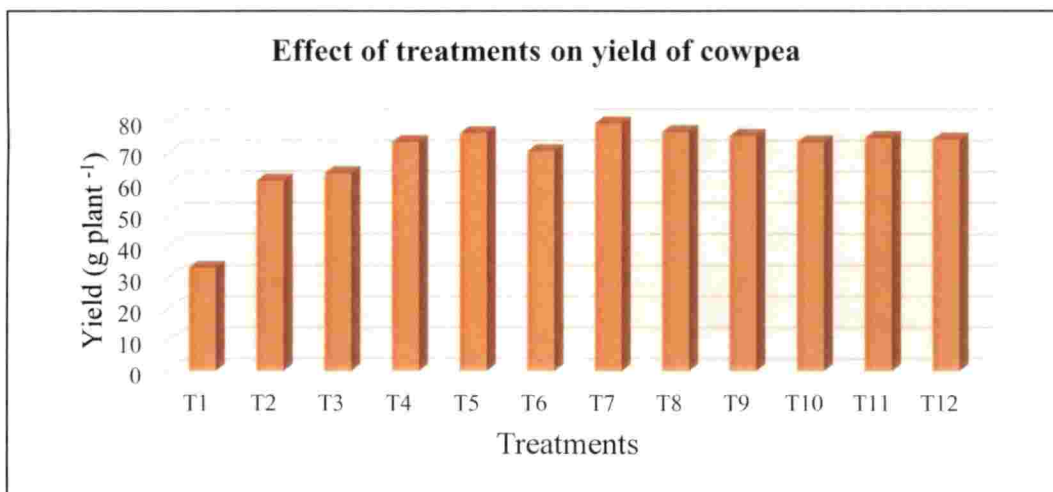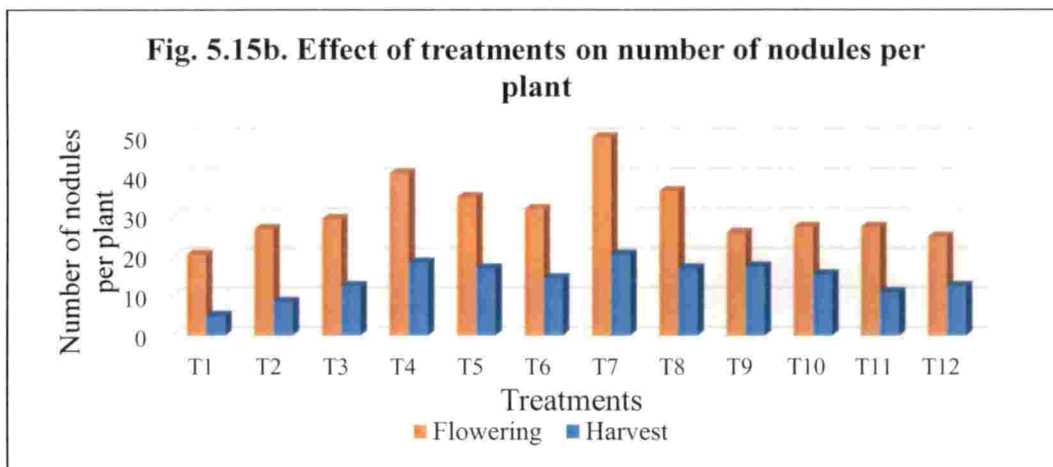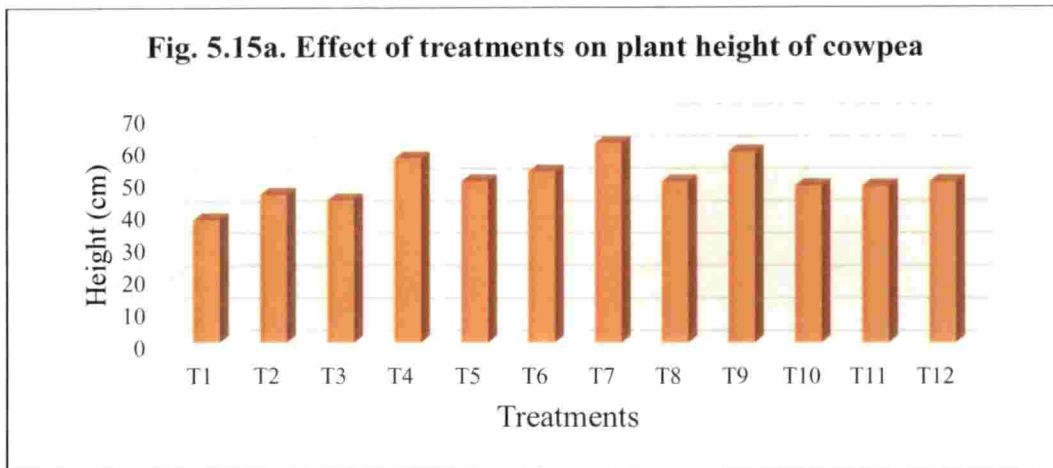
The perusal of data on yield and yield contributing characters showed treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) to be the optimum dose of magnesium to achieve maximum crop response. Fageria (2009) reported a quadratic response of the yield of dry bean and soybean to applied magnesium doses in Oxisols.

5.2.7. Critical nutrient level in soil and leaves of cowpea

Critical level of a nutrient in soil refers to the level below which crops readily respond to applied nutrient. According to White and Brown (2010) the critical concentration for sufficiency is defined as the concentration in the diagnostic tissue that allows a crop to achieve 90% of its maximum yield. The computed data for delineation of critical level of magnesium in soil and plant are presented in table 4.30. Critical limit of Mg was worked out by the method proposed by Cate and Nelson (1965). Scattered plot of soil magnesium and relative per cent yield revealed that critical level of magnesium in soil as 75 mg kg^{-1} (Fig. 4.1a). Similarly the plots of relative per cent yield and magnesium content in leaves identified critical level of magnesium in cowpea leaves at flowering as 0.38 % (Fig.41.b). According to White and Brown (2010), relative yield of 80-100% can be achieved in soils with cation exchange capacity less than 20 cmol (+) kg^{-1} if the critical level of available magnesium is 40-80 mg kg^{-1}. Critical level for available magnesium in soil was reported by Kasinath et al. (2014) to be 74 mg kg^{-1} and in leaf tissue of tomato to be 0.38% in Alfisols of Karnataka.

Summary

6. SUMMARY

Ultisols occupying more than fifty per cent of total geographical area of Kerala are characterized by low pH, low cation exchange capacity and low base saturation due to dominance of kaolinite, and oxides and hydrous oxides of iron and aluminium. The deficiency of magnesium is a common nutritional disorder in these soils due to leaching of bases under humid tropical conditions. Crops are found to respond to applied magnesium fertilizers with an increase in quantity and quality of produce.

The determination of critical level of nutrient in soil and plant helps to manage the nutrient deficiency and avoid crop loss. According to the *ad hoc* recommendations for management of secondary and micronutrients in Kerala, the critical level for sufficiency in soil is considered to be 120 mg kg^{-1} of exchangeable magnesium. This can seldom be attained under acidic soil environment. Hence, the management of magnesium deficiency in acid soils require the improvement in soil pH. Application of magnesium carbonate or dolomite can raise the soil pH apart from increasing the availability of magnesium in soil. The improvement in soil pH can have a positive influence on plant growth and population of soil micro flora.

Hence, the present study entitled "Optimization of soil environment and crop response for magnesium nutrition in Ultisol" was undertaken to evaluate the modifications of the acidic soil environment through added magnesium source and to validate the critical level of magnesium in soil and plant thereby optimizing magnesium nutrition for cowpea. Top soil (0-15 cm depth) representing Ultisols was collected from Water Management Research Unit, Vellanikkara to conduct an incubation study and a pot culture experiment. Initial characterization of physico-chemical properties and total population of Rhizobium/ Bradyrhizobium, free-living nitrogen fixing bacteria and total spore count of arbuscular mycorrhizal fungi was done using representative soil sample. The soil was very strongly acidic with pH of 4.70. Organic

carbon, available nitrogen and potassium were medium in status while phosphorus was high. The secondary and micronutrients except magnesium and boron were sufficient.

An incubation experiment was conducted to study the release pattern of magnesium from added source. Magnesium carbonate required to theoretically raise the available magnesium status in soil to 120 mg kg^{-1} was taken as the optimum dose and one level above (150%) and below (50%) the optimum dose was added to soil with and without the addition of recommended dose of calcium carbonate (250 kg ha^{-1}) and organic manure (20 t ha^{-1}). Soil pH, electrical conductivity (EC) and available magnesium was analysed at weekly intervals for four months of incubation.

A pot culture experiment to investigate the crop response to magnesium nutrition and to validate critical level of magnesium in soil and plant was conducted using cowpea variety Bhagyalakshmi as the test crop. The experiment consisted of twelve treatments with 4 replications *viz.*, absolute control (T_1), organic manure @ 20 t ha^{-1} (T_2), POP recommendation with calcium carbonate @ 250 kg ha^{-1} (T_3), POP recommendation with dolomite @ 400 kg ha^{-1} (T_4), T_3 + magnesium carbonate required to theoretically raise available magnesium in soil to 120 mg kg^{-1} (T_5) and treatments T_6 to T_{12} comprised of T_3+ graded doses of magnesium carbonate at 10% of T_5 (T_6), 20% of T_5 (T_7), 40% of T_5 (T_8), 60% of T_5 (T_9), 80% of T_5 (T_{10}), 125% of T_5 (T_{11}) and 150% of T_5 (T_{12}). Variations in physico-chemical properties of soil, microbial population in rhizosphere and nutrient content in stem, leaf and pods were analyzed during flowering and after crop harvest.

The salient findings of the study are summarized below

- The soil representing Ultisols collected for the experiment was very strongly acidic with pH 4.7.

- The status of organic carbon (1.32%), available nitrogen (476.67 kg ha^{-1}) and potassium (240.18 kg ha^{-1}) was medium in status while phosphorus was high (98.04 kg ha^{-1}).

- The status of available secondary and micronutrients except magnesium (64.53 mg kg^{-1}) and boron (0.24 mg kg^{-1}) were sufficient.

- Rhizobium population in soil was negligible whereas spore count of Arbuscular Mycorrhizal Fungi was found to be 21 per g^{-1} soil.

- The different fractions of magnesium in soil followed the order-Mineral Mg > exchangeable Mg > acid soluble Mg > organic-complexed Mg>water soluble Mg.

- The addition of different doses magnesium carbonate, calcium carbonate and vermicompost had significant influence on soil pH, EC and available magnesium throughout the period of incubation.

- Significant increase in soil pH was recorded after one week of incubation and the pH after sixteen weeks of incubation was higher than the initial pH.

- Electrical conductivity in soil was found to increase over the period of incubation indicating the release of basic cations from the added source.

- Available magnesium was significantly higher after eight weeks of incubation

- Significant variations were recorded in all the fractions of magnesium after the incubation period.

- The variations in acid soluble and mineral fraction between treatments substantiates the presence of magnesium carbonate as a solid phase in soil.

- The addition of graded doses of magnesium carbonate along with recommended dose of fertilizers produced significant variations in soil and crop.

- Soil pH and available magnesium in soil during flowering and after crop harvest increased with the increasing dose of magnesium carbonate added and the latter recorded higher pH and available magnesium.

- A significantly higher population of Rhizobium and free living nitrogen fixing bacteria were observed in the rhizosphere of T_7 that was supplied with 20% of quantity of magnesium carbonate required to raise available magnesium status to 120 mg kg^{-1}.

- Per cent root colonization with Arbuscular Mycorrhizal Fungi increased at higher doses of magnesium.

- Magnesium content in stem and leaf of cowpea during flowering and harvest increased with levels of added magnesium. But magnesium content in pods did not vary significantly.

- Crop uptake of nitrogen, phosphorus, potassium, iron and boron was significantly higher in T_6 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1} + $MgCO_3$ @10% of T_5).

- Crop uptake of calcium, magnesium, sulphur, manganese, zinc and copper significantly higher in T_7 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1} + $MgCO_3$ @20% of T_5)

- Highest number of nodules per plant was also recorded in T_7 that was supplied with 20 % of quantity of magnesium carbonate required to raise available magnesium status to 120 mg kg^{-1}.

- Chlorophyll a at flowering was significantly higher in T_7 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1} + $MgCO_3$ @20% of T_5)

- Significantly higher yield per plant of cowpea (79.33 g $plant^{-1}$) was recorded in T_7 (POP recommendation with $CaCO_3$ @ 250 kg ha^{-1} + $MgCO_3$ @20% of T_5)

- Scattered plot of relative per cent yield with available magnesium and magnesium content in leaves revealed the critical level of available magnesium in soil to be 75 mg kg^{-1} and that in leaf tissue at flowering to be 0.38 %.

Further studies are to be conducted on the efficacy of magnesium carbonate or dolomite under field conditions to optimize crop nutrition. The

determination of critical level of magnesium in soil and plant has to be done for annual and perennial crops. Critical level determination is to be done for soils with varying texture.

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OPTIMIZATION OF SOIL ENVIRONMENT AND CROP RESPONSE FOR MAGNESIUM NUTRITION IN ULTISOL

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ABSTRACT OF THE THESIS

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Abstract

Ultisols constitute the major soil type occupying more than fifty per cent of the total geographical area of Kerala. They are characterized by low pH, low cation exchange capacity and low base saturation due to dominance of kaolinite and oxides and hydrous oxides of iron and aluminium. The deficiency of magnesium is a common nutritional disorder in these soils due to leaching of bases under humid tropical conditions. Hence, the present investigation entitled "Optimization of soil environment and crop response for magnesium nutrition in Ultisol" was undertaken at Radiotracer Laboratory, College of Horticulture, Vellanikkara during 2017-2019. The objectives of the study were: (i) to evaluate the modifications of the acidic soil environment through added magnesium source and (ii) to validate the critical level of magnesium in soil and plant thereby optimizing magnesium nutrition for cowpea.

Top soil (0-15 cm depth) representing Ultisols was collected from Water Management Research Unit, Vellanikkara to conduct an incubation study and a pot culture experiment. Initial characterization of physico-chemical properties and total population of Rhizobium/ BradyRhizobium, free-living nitrogen fixing bacteria and total spore count of arbuscular mycorrhizal fungi was done using representative soil sample. The incubation experiment was conducted to study the release pattern of magnesium from added source. Magnesium carbonate required to theoretically raise the available magnesium status in soil to 120 mg kg^{-1} was taken as the optimum dose and one level above (150%) and below (50%) the optimum dose was added to soil with and without the addition of recommended dose of calcium carbonate (250 kg ha^{-1}) and organic manure (20 t ha^{-1}).

Soil pH, electrical conductivity (EC) and available magnesium was analysed at weekly intervals for four months of incubation. The increase in soil pH, EC and available magnesium was concomitant to the added doses of magnesium carbonate. Though significantly higher soil pH was recorded after one week of incubation, pH in

all the treatments at the end of the experiment was higher than the initial value. This indicated the persistence of magnesium carbonate throughout the incubation period. Available magnesium was significantly higher after eight weeks of incubation whereas electrical conductivity in soil was found to increase over the period of incubation. The fractionation of soil magnesium after incubation period showed all fractions to be significantly influenced by the treatnment. The variations in acid soluble and mineral fraction between treatments substantiates the presence of magnesium carbonate as a solid phase in soil.

A pot culture experiment to investigate the crop response to magnesium nutrition and to validate critical level of magnesium in soil and plant was conducted using cowpea variety Bhagyalakshmi as the test crop. The experiment consisted of twelve treatments with 4 replications *viz.*, absolute control (T_1), organic manure @ 20 t ha^{-1} (T_2), POP recommendation with calcium carbonate @ 250 kg ha^{-1} (T_3), POP recommendation with dolomite @ 400 kg ha^{-1} (T_4), T_3 + magnesium carbonate required to theoretically raise available magnesium in soil to 120 mg kg^{-1} (T_5) and treatments T_6 to T_{12} comprised of T_3+ graded doses of magnesium carbonate at 10% of T_5 (T_6), 20% of T_5 (T_7), 40% of T_5 (T_8), 60% of T_5 (T_9), 80% of T_5 (T_{10}), 125% of T_5 (T_{11}) and 150% of T_5 (T_{12}).

Physico-chemical properties of soil, microbial population in rhizosphere and nutrient content in stem, leaf and pods were analyzed during flowering and after crop harvest. An increase in soil pH and available magnesium content in soil could be achieved at both crop stages in response to added doses of magnesium carbonate. A significantly higher population of Rhizobium and freeliving nitrogen fixers were observed in rhizosphere soil of T_7 that was supplied with 20% optimum dose of magnesium. Though the magnesium content in stem and leaf of cowpea increased with the graded doses of magnesium, the data on yield per plant (74.33 g plant^{-1}) showed

treatment T_7 (T_3+ $MgCO_3$ @ 20% of T_5) to be the optimum level with maximum crop response.

Scattered plot of relative per cent yield with available magnesium and magnesium content in leaves revealed the critical level of available magnesium in soil to be 75 mg kg^{-1} and that in leaf tissue at flowering to be 0.38 %. Further studies are to be conducted on the efficacy of magnesium carbonate or dolomite under field conditions to optimize crop nutrition.

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164