# EVALUATION OF EFFECTIVE MICRO-ORGANISMS (EM) ON SOIL CHEMICAL PROPERTIES AND YIELD OF SELECTED VEGETABLES IN THE EASTERN CAPE, SOUTH AFRICA

BY

# LINDANI NCUBE

A dissertation submitted to the Faculty of Science and Agriculture in fulfilment of the requirements for the degree of Master of Science in Agriculture (Horticultural Science)

# **Supervisors:**

Mr M.O. Brutsch and Professor P.N.S. Mnkeni

Department of Agronomy, School of Agriculture and Agribusiness,

Faculty of Science and Agriculture, University of Fort Hare,

Alice 5700, South Africa

March 2008

# DECLARATION

I, Lindani Ncube declare that the work contained in this dissertation is entirely my own work with the exception of such quotations or references which have been attributed to their authors or sources and that all photographs are made by me except where I have acknowledged that another is the author.

# PREFACE

This dissertation is presented in the form of a common Introduction (Chapter 1) and Literature Review (Chapter 2) which introduce the reader to effective microorganisms (EM) in relation to the concept of sustainable agriculture and, for some, organic farming. This is followed by Chapter 3 which deals with two field trials where EM applications, with and without organic and inorganic amendments, were tested on tomatoes and butternuts. The two trials are followed by a similar greenhouse trial. Chapter 4 deals with a greenhouse trial with Swiss chard, also with similar treatments but with two harvests in order to monitor nutritional changes. In all the trials, treatment evaluations were done in terms of yield, quality, insect and disease control, and selected soil properties. The General Discussion and Conclusions follows (Chapter 5). Finally, the Appendices contain the statistical analyses of the experimental data. The purpose of presenting the dissertation in this form is to gain experience of presenting experimental data in the form of scientific papers.

#### ACKNOWLEDGEMENTS

First and foremost, I thank God whose divine love and protection has brought me to this level of education. It is with pleasure that I acknowledge the efforts of the many people who have contributed to the development of this dissertation. To start with, I thank Mr M.O. Brutsch and Professor P.N.S. Mnkeni for their hard work, tenacity and perseverance that helped to transform every piece of my work into this dissertation. I have received all the assistance that I wished for from both of my supervisors and I particularly valued their resolute and insightful guidance of my work that made me feel I was in safe hands. Many thanks to Doctors Fatunbi and Daudu, who gave me invaluable and specific guidance on some techniques used in this work. I am indebted to Siyasanga Mdana, Pamela Macingwane and Thanduxolo Nqagqweni for their assistance with the analytical part of this work. I also express my sincere gratitude to Mrs V. Bomali, Mr W. Seti and the farm workers, at the time, for their tireless effort in assisting me with the farm work. I also thank Mr Arnold Njainwe and Mr E.M. Gichangi for many useful suggestions they offered at various stages of my work.

Gratitude to many others – Fort Hare Seventh Day Adventist Church (SDASM), and the Alice community in the Eastern Cape of South Africa- who spent time with me over the years to rejuvenate my wellbeing throughout the preparation of this dissertation. While I am not listing you individually, I hope you will recognize your contribution when you see this piece of information. Special thanks to my friends, Rumbidzai Chidoori, Thabo Banda, Yvonne Reitsma, Lloyd Ganyani, Caroline Arisunta, Mfundo Maqhubela, Kifani Serge, Farai Murungu, Morris Fanadzo, Bothwell Musunda, Alen Manyevere, Faith Nkomo, Simphiwe Mhlontlo, Mackenzie Sikwela, Bethwell Moyo and Mary Mkhwananzi for the prayers and augmented moral support l often received from you. Guys, l will remember you for your unceasing encouragement.

I want to thank my mothers "Alice Nyoni", Ms M Mapaha and Josphine Mkhwananzi for being great understanding people who stood by me at all times. Special thanks to my uncle Siyabalandela Dube and my brother Senzani Thendele who assisted me so much financially.

I gratefully and deeply acknowledge the concern and prayers that gave me spiritual, moral and mental support from Memory, Agathar Mkhwananzi, Belita Mkhwananzi, Ndaba Ncube and the late brother Mathe.

Last, but not least, I am grateful to Miss Maria Smith (Faculty Manager), Miss C.M. Curthbert (School Manager), Mrs F. Maweza, Sisi Priscilla (Mrs P.N. Matakane), all my friends, students, lecturers and anybody else I could talk to and learn from as well as those who are still to come, there is so much to explore, to learn, to experience. I have learnt that I still have a lot to learn in life.

# ABSTRACT

Effective microorganisms (EM), a commercial concoction of microbes that includes yeasts, fungi, bacteria and actinomycetes, have been found to be effective in enhancing crop growth by a number of scholars. It is registered in South Africa, but it had not been thoroughly investigated. The present study investigated the effects of EM on growth, yield and quality of tomato (*Lycopersicon esculentum* Mill), butternut (*Curcurbita moschata*) and Swiss chard (*Beta vulgaris*), along with selected soil properties.

In field-grown tomato it was observed that the application of EM caused a significant increase in the number of fruits at seven weeks after transplanting. However, plants treated with EM alone, or EM in combination with other amendments, subsequently produced lower yields owing to an outbreak of early and late blights which affected them the most severely. Combined applications of EM with organic amendments improved plant N content and increased soil N content above initial levels. The application of compost resulted in soil N and P concentrations higher than those of the control presumably due to nutrients being slowly released from the compost material.

In a follow up greenhouse trial EM application had a negative effect on tomato leaf dry matter yield, number of leaves, number of trusses, fruit yield and number of fruits. The negative effects of EM were ascribed to N immobilization by the EM that could have resulted in reduced N availability to plants. The lower number of fruits associated with EM application resulted in improved average fruit weight of tomatoes grown in the greenhouse, possibly as a result of more assimilates being partitioned to the few fruits

formed. EM application also had a negative effect on field grown butternut as reflected by lower total yield, lower marketable yield and lower first grade yield. The results were attributed to immobilization of N induced by application of EM, and to the inability of EM to control pumpkin fly that attacked very young fruit, resulting in their failure to develop or resulting in the down grading of mature fruits.

The application of EM alone had a positive but non significant effect on the yields of both the first and second harvests of Swiss chard. However, when applied with compost or goat manure, a non significant negative effect on yield was observed. When applied with inorganic fertilizer, EM had no effect on yield but tended to increase the uptake of nitrogen by Swiss chard. Though goat manure had a narrower C: N ratio than compost, it did not result in greater EM effectiveness as had been hoped. However, goat manure had a more positive effect on soil properties than compost. It increased the N, P, and K contents of the soil and resulted in a narrower C: N ratio of the soil compared to compost. Generally, the results of the four trials conducted with three different crops indicated that EM had inconsistent effects on crop performance.

**Key words:** Butternut, tomato, Swiss chard, compost, effective microorganisms (EM), goat manure, mineral fertilizer, yield

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# LIST OF ABBREVIATIONS

EM	Effective microorganisms
M - EM	Multiplied effective microorganisms
EM – F.P.E	Effective microorganisms - fermented plant extract
EM – F.F	Effective microorganisms - fermented fish
ОМ	Organic materials Organic matter
FAO	Food and Agriculture Organisation
UNESCO	United Nations Educational, Scientific, and Cultural Organization

# **CHAPTER 1**

# **GENERAL INTRODUCTION**

The intensification of agricultural production is mostly done with the use of mineral fertilizers, planting of high-yielding cultivars and the use of agro-chemicals for crop protection. The FAO, for example, estimated in 1989 that about 50% of the increase in agricultural production in the world was due to use of chemical fertilizers (FAO, 1989). This approach is, however, increasingly proving to be unsustainable as it causes soil degradation and the cost of required inputs is often beyond the financial ability of smallholder peasants who constitute more than 80% of the food producers in the developing nations (Tittonell, Vanlauwe, Leffelaar, Shepherd & Giller, 2005).

There have been numerous attempts to develop alternative systems more suited to the needs of the tropical and subtropical smallholders. One such alternative system promotes the use of "effective microorganisms" (EM) to enhance crop growth. EM is a mixture of specially selected and cultured naturally occurring, beneficial microorganisms that have been studied and known to significantly improve soil quality and plant growth (Li & Ni, 1995). It contains selected species of microorganisms, including predominant populations of lactic acid bacteria and yeasts and smaller numbers of photosynthetic bacteria, actinomycetes and other types of organisms. All of these are claimed to be mutually compatible with one another and are able to coexist in liquid culture.

The concept of effective microorganisms (EM) was developed in 1971 by Professor Teruo Higa, University of the Ryukyus, Okinawa, Japan (Higa & Wididana, 1991). Research has shown that inoculation of the soil/plant ecosystem with EM cultures can improve soil quality, soil health, the growth, yield, and quality of crops (Higa & Parr, 1994). Daly & Steward (1999) also showed that application of EM to peas, sweet potato and onions increased yield by 31%, 23% and 29%, respectively.

Different brands of EM are currently being produced in about 40 countries across the globe using local microbial isolates. In South Africa EM products are produced and marketed by EMROSA (Pty) Ltd.

The use of EM is not yet widespread in South Africa, although there are some reports, mainly by EMROSA in their newsletters and on their website (www.emrosa.org.za) that some commercial farmers are already using the materials and they seem to find satisfaction with its effects. At the time the trials reported here were started these products were not officially registered for use on crops but it seems that as of 2006, exact date unknown, the products are registered and can be sold in shops (Anon., 2006). They apparently also conform with EUREP GAP organic requirements and "you can export your products anywhere in the world" (Anon., 2006). There has been only one scientific report of their use in the Eastern Cape and relatively few scientific reports worldwide (Mupondi, Mnkeni & Brutsch, 2006a, b).

As a result of a lack of rigorous research on the usefulness of EM in crop production, a need was felt to conduct an extensive evaluation of these products using commonly

grown vegetables in the Eastern Cape Province, South Africa. The main objective of the study was therefore to evaluate the usefulness of EM products using tomato (*Lycopersicon esculentum* Mill), butternut (*Curcurbita* spp) and Swiss chard (*Beta vulgaris*) which are commonly grown in the Eastern Cape Province, South Africa in field and greenhouse studies. The specific hypotheses are included under the different studies.

## **CHAPTER 2**

#### **GENERAL LITERATURE REVIEW**

# 2.1 Intensification in agriculture and sustainability

As a result of an increase in the global population, food shortages are expected to increase especially in developing countries due to a decline in arable land per capita (World Bank, 2000). However, production of food for human consumption, animal feed and industrial purposes has increased in the developed countries mainly because of an increase in crop yield and cropping intensity rather than because of expansion of arable land (Islam, 1995).

2.1.1 General overview of agricultural intensification

In order to cater for the rapidly growing world population, production of food and fibre has to be increased without increasing the land for production (Pretty, Thompson & Hinchchliffe, 1996). Despite the phenomenal growth in food production since the mid-1960s, some 800 million people still suffer from chronic hunger. This kind of a situation arises because of the uneven spread of natural and economic wealth between the developed and developing worlds, and also because modern production systems, the world over, have grossly overlooked the issues of environmental sustainability and social equity (Datta, 2002). One pressing issue worldwide is that the growth in food production is non-sustainable on various grounds, be it financial, economic, social, or environmental (Shah, 2006).

#### 2.1.2 Agricultural production in Africa

In Africa, agricultural production is still a major challenge with low yields being realized due to shortages of water, low levels of nutrients, and pest and disease infestations (Evans, 1998). Intensification in agriculture has seen a rise in the use of mineral fertilizers, use of high yielding cultivars and genetically modified crops and intensive use of agro-chemicals (Waddington & Hersey, 1997). However, available evidence has shown that intensification in agriculture is leading to a decline in soil fertility (Smaling, Nandwa & Janssen, 1997; Khor, 2004) especially in Africa. Nutrient depletion through soil erosion, leaching, low organic manure inputs and crop residue removal, coupled with improper application of mineral fertilizers, are the main causes of poor soil fertility in Africa (Carney, 1998).

#### 2.1.3 Constraints of intensifying agriculture in Africa

Although the use of mineral fertilizers has been adopted by the whole world, their use in Africa is limited by their high cost (Gardner, 1997) which is beyond the reach of communal farmers who constitute 80% of the food producers in Africa (Tittonnel *et al.*, 2005). A survey conducted in sub-Saharan Africa demonstrated that a shortage of finance was the main reason why alternative soil management techniques were not adopted by 64-70 % of the respondents (Harris *et al.*, 1998).

#### 2.1.4 Sustainable agricultural production

Sustainability is the most important aspect in all the proposed systems for alternative soil management. According to Hansen & Jones (1996), sustainable agriculture is "the ability of farming systems to continue into the future". This implies that sustainable agriculture means a "maintenance of the adaptive capacity of farming systems" (Park & Seaton, 1996), enabling the future generations to meet their food demands. Sustainable agriculture has multiple-dimensional characteristics that include economic, environmental and social aspects (Legg, 1999; Pretty & Hine, 2001). In sustainable agriculture, organic farming is being promoted due to the positive environmental, social and economic impacts (Legg & Viatte, 2001).

Organic farming is expanding rapidly worldwide. Agricultural production in organic systems depends mostly on the functions performed by soil microbial pools, particularly in nutrient supply (Smith *et al.*, 1993). Organic farming has been spreading at an annual rate of ca. 20% in the last decade (Lotter, 2003), covering over 24 million hectares worldwide (Willer & Yussefi, 2004), and has become a mainstream practice for some crops (Anon., 2004a).

Organic farming promotes soil structure formation (Reganold, Elliot & Unger, 1987; Pulleman *et al.*, 2003), enhances soil biodiversity (Doles, Zimmerman & Moore, 2001; Mäder *et al.*, 2002; Oehl *et al.*, 2004), alleviating environmental stresses (Horrigan, Lawrence & Walker, 2002; MacIlwain, 2004) and improving food quality and safety (Reganold *et al.*, 2001; Giles, 2004). Organic farming advocates the use of organic and biological inputs for controlling diseases and pests and for nutrient supply (Rigby & Cáceres, 2001; Watson *et al.*, 2002). Sustainability in organic farming depends largely on the build up of soil microbial pools that function as a transient nutrient sink and are responsible for releasing nutrients from organic matter for plant use (Smith & Paul, 1990; Dalal, 1998; Friedel, Gabel & Stahr, 2001). It has been shown that microbial biomass N contributes to the primary N source of potentially mineralizable N in soil (Myrold, 1987; Bonde, Schnürer & Rosswall, 1988). The use of EM is now promoted as a way of maximizing the returns of soil microbial pools.

#### 2.2 Effective microorganisms (EM)

EM is an abbreviation for effective microorganisms and refers to a cocktail of beneficial microorganisms that is used as a soil amendment (Woodward, 2003). EM contains selected species of microorganisms, including predominant populations of lactic acid bacteria and yeasts and smaller numbers of photosynthetic bacteria, actinomycetes and other types of organisms. All of these are claimed to be mutually compatible with one another and are able to coexist in liquid culture. Some microorganisms contained within EM are discussed in detail below.

#### 2.2.1 Lactic acid bacteria

Lactic acid bacteria produce lactic acid from sugars and other carbohydrates, developed by photosynthetic bacteria and yeast. Some foods and drinks such as yoghurt and pickles have been made with lactic acid bacteria for decades. However, lactic acid is a strong sterilizing compound, and suppresses harmful microorganisms and enhances decomposition of organic matter. Lactic acid bacteria promote the decomposition of materials such as lignin and cellulose and ferment these materials, thereby removing undesirable effects of un-decomposed organic matter (Prescott, Harley & Klein, 1996).

# 2.2.2 Yeasts

Yeasts synthesize antimicrobial and other useful substances required for plant growth from amino acids and sugars secreted by photosynthetic bacteria, organic matter and plant roots. The bioactive substances such as hormones and enzymes produced by yeasts promote active cell and root division. These secretions are also useful substrates for effective microbes such as lactic acid bacteria and actinomycetes (Prescot *et al.*, 2002).

### 2.2.3 Photosynthetic bacteria

The photosynthetic or phototropic bacteria are a group of independent, self-supporting microbes. These bacteria synthesize useful substances from secretions of roots, organic matter and/or harmful gases (eg. hydrogen sulfide), by using sunlight and the heat of soil as sources of energy. Useful substances developed by these microbes include amino acids, nucleic acids, bioactive substances and sugars, all of which promote plant growth and development. The metabolites developed by these microorganisms are absorbed directly into plants and act as substrates for increasing beneficial populations (Prescot *et al.*, 2002).

#### 2.2.4 Actinomycetes

Actinomycetes are part of the microorganisms which make up EM and they disturb the life cycle of insects thereby reducing the reproduction rate. Actinomycetes feed on the chitin produced by the larvae to become pupae, henceforth the metamorphosis is hindered. Actinomycetes are aerobic and can be cultivated easily on simple growing media and are gram positive (Prescot *et al.*, 2002).

#### 2.2.3 Brands of EM

The use of EM as a broad-based organic material for crop production is an innovative model for use in organic agriculture, which is growing in popularity worldwide. The use of various brands of EM has been found to improve the growth and quality of crops (Dally and Stewart, 1999) and the different brands of EM are discussed in detail in the succeeding sections.

# 2.2.3.1 Multiplied (M-) EM

Stock EM is multiplied to reduce cost. Multiplied - EM is a mixture of stock EM with molasses and water in a ratio of 1: 5: 94. After mixing the ingredients, the resultant solution is stored in an airtight container for between 10 to 14 days at temperatures between  $20^{\circ}$ C to  $25^{\circ}$ C until the pH is 3.7. During this time, it is speculated that the microbes enter a growth phase and multiply to reach a microbial population and composition similar to that of the stock EM. To maintain the quality of multiplied-EM,

it should be stored at temperatures ranging between  $10^{\circ}$ C and  $20^{\circ}$ C (Anon., 2004b; 2005).

### 2.2.3.2 EM - fermented plant extract (EM - F.P.E)

Table 2.1 shows ingredients used for producing EM - fermented plant extract.

Table 2.1: Ingredients require	d for fermented plant extract
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Ingredient	Quantity
Chopped fresh weeds	20 litres (not pressed down)
Chlorine free water	16 litres
Molasses (3%)	480 ml
Multiplied - EM (3%)	480 ml

Adapted from Anon. (2005).

Plants or weeds with repelling properties such as khakibos, syringa, clover, herbs and grass are used in the production of EM - fermented plant extract. During brewing, plants are chopped in their fresh state to lengths ranging from 2 to 5 cm which are then put into a sealable bucket. The multiplied - EM is then mixed with water and the resultant solution poured into the bucket containing the chopped plant material and the bucket is closed. After a period of 2 to 5 days, the fermentation of the plant constituent's starts and a lot of  $CO_2$  is generated and released through an air trap. The process continues until the pH drops below 3.7. At this point, the solution is filtered to remove the plant materials (Anon., 2004; 2005; Asia-Pacific Natural Agriculture Network, 1995). EM - F.P.E. acts as a repellent and disease suppressor and it also

contains amino acids, enzymes, hormones and vitamins. During the course of the growing season, EM - F.P.E. is either applied to the soil through irrigation or sprayed on the crop with a sticker (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

#### 2.2.3.3 EM 3-in-1

EM 3- in-1 is one of the strongest EM based insect repellents. It is produced in a similar way as EM - F.P.E. except for the ingredients used. In this case, fresh garlic, chili pepper, ginger (400 g of each, chopped), and black pepper (200 g powdered, 600 ml of Multiplied - EM and 18 litres of water are used (Anon., 2004b; 2005).

# 2.2.3.4 EM - 5

EM - 5 is a mixture of EM1 (multiplied EM), molasses, vinegar, strong distillation alcohol (more than 30%) and water. After mixing the ingredients, the resultant mixture is fermented in a sealed container for more than 30 days until it produces no more fermentation gas (CO<sub>2</sub>). Natural herbs such as garlic and red pepper are usually added during the fermentation process. The resultant solution is applied to plants to prevent invasions of destructive insects as well as strengthening the natural immune system against diseases (Anon., 2004b; 2005).

EM - bokash is a mixture of multiplied - EM with fresh and quality organic materials like rice bran, wheat bran or fish meal. After the ingredients have been mixed, the resultant solution is kept for up to two weeks to ferment in sealed containers. The final product is used for:

• Accelerating the fermentation and anaerobic decomposition of organic waste materials when making compost.

• Adding to animal feed for improvements in general health and natural immunity (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

2.2.3.6 EM - fermented fish (EM - F.F.)

EM - F.F. releases nutrients like nitrogen and phosphorus slowly over a period of time, allowing plant growth from one season to the other. During preparation, the fish is crushed, making nutrients to be accessible to microorganisms. The product is then mixed with multiplied EM before spraying on plants (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

# 2.2.3.7 EM - fermented chicken manure

EM - fermented chicken manure is similar to EM - fermented fish as it also provides nitrogen and phosphorus to the plants. Chicken manure, extended EM and an equivalent of 1% of bokash are the ingredients used (Anon., 2004a; 2005).

EM - fermented kitchen garbage is produced by fermenting organic waste generated in the kitchens using multiplied - EM to produce a nutrient-rich fertilizer for plants. The method for its production is similar to that of EM - F.P.E (Anon., 2004b; 2005).

# 2.2.3.9 EM - X

This is a special version of the EM liquid which has been certified for human consumption. The beverage effective microorganisms - X (EM - X) is an antioxidant cocktail derived from fermentation of unpolished rice, papaya and sea weeds with grouped effective microorganisms of lactic acid bacteria, yeast and photosynthetic bacteria. It contains mixed-extracts of plants and effective microorganisms. EM - X contains over 40 minerals and antioxidants (such as flavonoids, kaempferol, panaxin, quercetin, lycopene, oryzanolum, ascorbic acid, tocopherol, ubiquinone) and other bioactive substances such as nucleotides, peptides and amino acids (such as nicotinamide mononucleotide, nicotinamide adenine dinucleotide, l-alanine and l-glutamine) (Sato *et al.*, 1997). If EM - X is taken as a daily dose over a period of time, it reduces free radicals in the body, greatly improving the immune system and serving to reduce the possibility of cancer cells being produced (Anon., 2004b; 2005).

### 2.2.4 Application of EM

According to Sangakkara (2004), EM is effective for crop production and is environmentally safe with different brands of EM being produced in about 40 countries across the globe using local microbial isolates. They find uses in different fields, ranging from crop agriculture, environmental management, animal production, and aquaculture. The different brands of EM are applied to the above mentioned environments in different ways and these are discussed fully in the succeeding sections.

#### 2.2.4.1 Inoculation of EM into the soil

Brands of EM can either be applied as a soil drench or be spread to plants during crop production. When inoculating into the soil, a 1: 500 dilution of multiplied - EM in water or EM - FKG (kitchen garbage) is used. When using EM - F.F (fermented fish) or EM - F.C.M (fermented chicken manure), a 1: 300 dilution is advisable. An equivalent of 2.5 tonnes of bokash or less is applied to soil per hectare. Dosages above 2.5 t ha<sup>-1</sup> are detrimental to the plants due to organic acids which can damage their roots. EM - bokash is usually applied between 10 to 14 days prior to planting and is placed at a distance of 10 cm to 15 cm away from roots (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

#### 2.2.4.2 Spraying EM on leaves

The spraying of EM on leaves of plants serves as a prophylactic spray mainly for disease and insect control. The spraying is often started earlier in the growing season and is conducted till the plants are harvested. A dilution of 1: 1000 of multiplied - EM, EM -F.P.E. or EM - 5, or a mixture of different EM derivatives, is advisable although a stronger dilution can also be used (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

#### 2.2.4.3 Soaking seeds in EM

Before planting, seeds are soaked in 0.1 % EM water with small seeds being soaked for about 30 minutes and big seeds for four to six hours. After soaking, the seeds are dried under shade to reduce the chances of them sticking together (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995) and planted in the field.

# 2.2.4.4 EM irrigation (fertigation)

Multiplied - EM or EM derivatives are frequently applied to soil through irrigation water. A ratio of 1: 1000-5000 of multiplied - EM or EM - F.P.E to water is used (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

#### 2.2.4.5 Insect control

EM also functions as a biological control measure in suppressing and controlling pests through the introduction of beneficial microorganisms to the plant environment. The odour emitted by EM repels harmful insects and serves as a prophylactic spray. EM-F.P.E and EM - 5 are insect repellant and they are not toxic to ladybirds, spiders, dragonflies, frogs etc. EM attracts fruit flies and affects mostly the females which later become sterile (Anon., 2004a; 2005). Pests and pathogens are suppressed or controlled through natural processes by increasing the competitive and antagonistic activities of the microorganisms in EM inoculants (Anon., 2004b; 2005; Asia-Pacific Natural Agriculture Network, 1995).

#### 2.2.5 Benefits of applying EM

The following are some of the beneficial influences of EM in agricultural production:

- Improvement of the physical, chemical and biological environments of the soil (increases the efficacy of organic matter as fertilizers) and suppression of soil-borne pathogens and pests,

improvement of germination of seeds, flowering, fruiting and ripening in plants.
enhancement of the photosynthetic capacity of crops and,

- increased crop yield.

As a result of the above-mentioned beneficial effects of EM, yields and quality of crops are enhanced (Asia-Pacific Natural Agriculture Network, 1995).

# 2.2.5.1 Effects of EM on organic matter

Organic manures are a source of multiple nutrients and can improve soil physical, chemical and biological characteristics. However, the effects of organic manures on crop yield are long term and not immediate, therefore farmers prefer using mineral fertilizers in their cropping systems. Addition of EM together with organic manures is thought to be an effective technique for stimulating supply and release of plant nutrients. Studies have shown that inoculating agro-ecosystems with EM can improve soil and crop quality (Higa and Parr, 1994; Hussain *et al.*, 1999). Following EM application into the soil, there is an increase in soil microorganisms that are beneficial for the growth of the plant that result in rapid mineralization of organic materials (Asia-Pacific Natural Agriculture Network,

1995). According to Khaliq, Kaleem & Hussain (2006), application of organic materials or EM alone did not significantly increase yield. However, their integrated use resulted in a 44% increase in yield over the control. Application of EM with mineral fertilizer in this case resulted in a slight increase in yield (14%) over the mineral fertilizer alone, demonstrating that EM is more effective when applied with organic manures. The relatively low response of mineral fertilizer compared to EM application was due to the fact that EM is made up of different microorganisms which can respond well only in the presence of sufficient organic matter. Aryal, Xu & Fujita (2003) showed that *Rhizobia* and arbuscular mycorrhizal (AM) inoculation of bean plants significantly increased pod yield in plots with organic matter supplements compared to chemically treated plots.

The relative effects of EM were further observed in plant leaf N concentration where its co-application with organic materials increased leaf N concentration by 38% relative to the control compared to 16% increase due to organic materials application alone (Khaliq, et *al.*, 2006). EM enhances the degradation and stimulates mineralization of organic materials, releasing plant nutrients into the soil (Hussain *et al.*, 1999). Application of EM into soil resulted in higher available phosphorus concentration 50 days after transplanting of tomato (Xu, 2000). However, 90 days after transplanting of tomato, both nitrogen and phosphorus concentration were low in EM treated soils and this was ascribed to more nutrients being taken up by plants that showed faster growth and subsequently higher yields. Piyadasa *et al.* (1995) studied the release of nitrogen and phosphorus from soils amended with organic matter over a 21 day incubation period at  $60^{0}$ C. Application of EM increased both inorganic nitrogen and phosphorus compared to the control.

Extensive studies have been conducted on the effects of EM especially when applied with bokash on plant growth, photosynthesis and yield as compared with mineral fertilizers (Fujita et al., 1997; Arshad, 2006). Fujita et al. (1997), found that plants treated with mineral fertilizer had higher dry matter yields during the early stages of growth but lower dry matter yields at the later stages compared to EM treated plants. Plants treated with EM and bokash maintained vigorous growth with greater root mass and activity and a higher rate of photosynthesis until harvest time compared to plants treated with mineral fertilizer. According to Yamanda et al. (1996), well developed roots in EM - bokash treated plants play an important role in maintaining a higher rate of growth and photosynthetic activity. Higher growth rates are due to sustained availability of nutrients from bokash through mineralization by EM microorganisms (Kato et al., 1997). There is a possibility that EM contains growth regulators that could stimulate root activity and delay senescence of plants (Yamada et al., 1996). Plant hormones like auxins, gibberellins and abscisic acid play important roles in root growth and development (Schneider & Wightman, 1974). In addition, bacteria, fungi and actinomycetes produce some bioactive substances that can enhance plant growth and metabolism (Arshad & Frankenberger, 1992). However, it is not yet clear how EM stimulates growth or plant metabolic processes. Some researchers have been speculating that the beneficial effects of EM may be due to their ability to biosynthesize antioxidants (Yamada and Xu, 2000)

## 2.2.5.3 Effects of EM on crop yield

Application of EM to soil also increases crop yield and quality due to an increase in plant nutrients and suppression of soil-borne pathogens (Asia-Pacific Natural Agriculture Network, 1995). In a study carried out by Daly & Stewart (1999), application of EM plus molasses caused a significant yield increase over the control and resulted in more first grade onions, peas and sweet corn.

# 2.3 Organic manures

Use of organic manures at agronomic rates for plant nutrient supply and for beneficial effects on physical properties is a traditional agricultural practice (Haynes and Naidu, 1998). Over the last decade the effects of organic manures on soil properties have received renewed attention due to an increased interest disposal of large amounts of waste being generated.

## 2.3.1 Effects of organic manures on physical and chemical properties of soil

Organic manure affects soil bulk density, soil stability and aggregation, pH, buffer capacity, cation exchange capacity, soil encrustation, water infiltration, soil penetrability, moisture content, drainage, tilth, aeration, temperature and nutrient supply and availability for plant growth (Woomer, 1993).

## 2.3.1.1 Soil bulk density

Addition of organic manures to soil increases both soil organic matter content and soil microbial pool activities which in turn improve the physical properties of soil (Sanchez *et al.*, 1989). A direct relationship between bulk density changes and water holding capacity as a function of net increases in soil organic C exist in the soil (Khaleel *et al.*, 1981). Addition of organic manures to soil decreases bulk density of the soil as a result of dilution of the denser mineral fraction of the soil. Organic manures are less dense and have increased pore sizes and numbers (Duggan & Wiles, 1976).

Addition of organic manures to soil induces formation of stable aggregates with the humic fractions reducing the plasticity, cohesion, and stickiness of clayey soils (Brady & Weil, 1999). Addition of organic manures to soils is followed by a lag phase after which the microbial biomass pool increases. An increase in the microbial biomass pool is accompanied by physical entanglement of fungal hyphae and the production of extra - cellular polysaccharides which link soil aggregates together and hence cause a rise in aggregate stability (Haynes & Naidu, 1998). Composted organic manures induce a slow and more steady increase in aggregate stability (Monnier, 1965) compared to fresh organic manures.

# 2.3.1.2 Soil pH and cation exchange capacity

Humus colloids hold cations like calcium, potassium, magnesium in exchangeable forms in such a way that they become available for plant use and are not leached by water. "Through its cation exchange capacity and acid and base functional groups, organic matter also provides much of the pH buffering capacity in soils" (Brady & Weil, 1999). Duggan & Wiles (1976) showed that where composted organic materials were incorporated to an acid soil there was an increase in soil pH with organic manure application. However, the same results were not obtained with neutral or basic soils (Gallardo-Lara & Nogales, 1987). Soils treated with organic manures also have a high CEC as shown by Follet, Murphy & Dohahue (1981). With a sandy soil, the CEC increased by a factor of 5 to 10 times as compared to a clay soil.

## 2.3.1.3 Soil water content and soil water holding capacity

Addition of organic manures to soil decreases surface crusting (Epstein, Taylor & Chaney, 1992), reduces displacement of soil particles by water raindrops (Mazurak, Chesnin & Tiarks, 1975), increases infiltration capacity and hydraulic activities (Cross and Fischbach, 1972) and therefore decreases the amount of runoff water (Hensler *et al.*, 1970), and reduces water lost through evaporation, ameliorates drainage and improves root penetration (Allison, 1973). Water holding capacity of soils is increased by additions of organic manures to soils and is controlled by the number, size and distribution of pores. An increase in water holding capacity at low tensions is due primarily to increased number of small pores. At higher tensions, almost all the pores are air filled and soil moisture content is determined by the surface area and thickness of water films on these surfaces (Khaleel *et al.*, 1981). Addition of organic manures to soil also increases the

specific surface area of soil and results in an increase in water holding capacity under higher tensions (Gupta, Dowdy & Larson, 1977).

#### 2.3.1.4 Soil nutrients

Nitrogen, phosphorus, sulfur and micronutrients are constituents of organic manures from which they are slowly released through mineralization. Quality of organic manures plays a crucial role in their mineralization. Therefore N release from organic materials is moderated by N mineralization and immobilization, which in turn is controlled by C: N, lignin: N, polyphenol: N and (lignin + polyphenol): N ratios, lignin and polyphenols and percent of N of the organic manure used (Mafongoya, Dzowela & Nair, 1997). The chemical composition of organic manures, especially C: N ratio, determines whether mineralization or immobilization processes will dominate in the early stages of decomposition. Release of inorganic N from organic manures to soil depends on the rate of decomposition of the material used and subsequent turnover of decomposed C and N in soil (Hadas & Portnoy, 1997). Mugwira (1984) demonstrated that the effectiveness of manure as a nutrient source depends on the time of its presence in the soil and the rate and type of manure used.

Addition of low quality organic inputs into soil over a long period can increase soil organic C build up, without necessarily increasing productivity. A cropping system study in India, for example, showed that wheat straw combined with urea substantially reduced yields, whereas a N equivalent amount of Sesbania green manure combined with urea

enhanced yields compared with a N equivalent amount of urea applied alone (Goyal *et al.*, 1992). According to Kamukondiwa & Bergstorm (1994), organic manures of low quality have low soluble C and N that are needed to enhance microbial pool activities in the soil and subsequently lead to a decrease in crop yield. Many crop residues and animal manures have a nitrogen content that ranges between 1.8 to 2.0% and can immobilize N temporally and are said to be of low quality.

Both composted and fresh organic manures have been used as a source of nutrients for production of several crops, with many degrees of success or failures. The outcome of applying organic manures to soil is a function of manure quality and soil properties. In South Africa, manure application rates of 5.5-11 t ha<sup>-1</sup> for field crops, and double that for crops with higher nutrient requirements, have been suggested (Malherbe, 1964). However, it is difficult to recommend the exact amount of organic manures needed for successful crop production as this varies with type of organic manure, soil type, crop requirements and prevailing environmental conditions. In the Eastern Cape Province of South Africa, small holder farmers have been reported to use manure at rates that range between 0.3 and 182 t ha<sup>-1</sup> (Yoganathan *et al.*, 1998). However, application rates range between 25 to 100 t ha<sup>-1</sup> of manure (Mkile, 2001). High doses of manure might be toxic to plants, animals and human beings (Meek, 1974; Donahue, 1977) and thus due caution must be exercised to keep its detrimental effects in check. According to Tester (1990), addition of 100 t ha<sup>-1</sup> compost per annum is enough to cause physical changes in the soil and increase yield.

A plethora of evidence suggests that a combination of organic manures with inorganic sources of minerals yield higher beneficial effects on production and development compared to their sole application. This may be due to the fact that organic manures do not contain all the required plant nutrients in adequate amounts. Inorganic fertilizers are leached easily and combining them with organic manures will combine their soluble minerals with the organic fraction. The nutrients will then be released slowly over time, in the form of sub-products of microbial pools and under action of organic acids (Allison, 1973).

2.3.2 Types of organic manures

Organic manures can be applied to soils as compost or in their fresh state. According to Cambardella *et al.* (2003), fresh organic materials contain higher inorganic N concentrations and have higher net N mineralization rates than composted manure. In a study by Paul and Beauchamp (1994), plants treated with fresh organic manures exhibited higher dry matter in the first growing season than composted manure.

#### 2.3.2.1 Compost

Composting is a process whereby organic materials are broken down, decomposed and stabilized by indigenous microorganisms under a moist, warm, aerobic environment, leading to the production of carbon dioxide, water, minerals and a stabilized organic matter while pathogenic microbes are destroyed by enzymatic combustion and the generated heat (Ouatmane *et al.*, 2000).

Composting is a process that offers an opportunity to recycle organic waste as a soil amendment. The benefits of composted organic waste to soil structure as well as in fertility and plant growth have been increasingly emphasized (Esse, Buerkert, Hiernaux & Assa, 2001). Application of undecomposed waste or non-stabilized compost to soil can lead to phytotoxicity and nutrient immobilization (Cambardella *et al.*, 2003).

## 2.3.2.2 Animal manures

In Africa, animal manure is applied to soil for fertility related issues and its benefits are well documented. Nutrient content in animal manure differs because of the variations in diet of the animals, collection and storage.

Manure and other waste products of livestock have been used as soil amendments for decades and were the only ways of enhancing soil productivity before mineral fertilizers were invented (Lupwayi et al., 2000). In Zimbabwe, application of animal manure is a common practice and the quality of manure as a plant nutrient source has been found to widely chemical composition (Tanner & Mugwira, 1984). vary in Goat, sheep, cattle and chicken manure are the common manures used in the southern African regions with cattle contributing two thirds of the total amount of manure found and the remainder is contributed by sheep and goats.

Nutrient composition of animal manures varies greatly between species (Serna & Pomares, 1991), animal nutrition, mineral particle content and storage conditions. However, in the Eastern Cape Province, the nutrients of animal manures range between 9.9–16.7 g N, 2.0–3.6 g P and 17.2–23.7 g K kg<sup>-1</sup> (Mkile, 2001), and are within the ranges reported for manures in the west African countries (Lupwayi *et al.*, 2000). However, the manures are low in P content which is a limiting factor.

The nutrient content of manures differs because of the variation in the animal diet that influences partitioning of N between faeces and urine. Feeding animals with a high quality diet results in N being excreted through urine and is lost through volatilization (Somda *et al.*, 1995). It is believed that feeds containing a sizeable amount of tannins increase the amount of N excreted in faeces. A study conducted by Mafongoya *et al.* (1997), showed that N in manures from animals fed with a diet rich in tannin is very resistant to mineralization in the soil. A study conducted in the Eastern Cape, South Africa by Mkile (2001), of nutrient composition of different manures, showed that cattle manure had the lowest N, P and K contents followed by sheep, and goat manure had the highest content.

## 2.4 General overview of the soils of the Eastern Cape and particularly at Fort Hare

Soils of the Eastern Cape are dominated by quartz, mica and kaolinite in the clay fraction (Mandiringana *et al.*, 2005). Quartz is the most dominant mineral in soil and is more concentrated in the coarser silt and sand fractions. The presence of quartz in large quantities contributes to the poor chemical and physical properties of the soils. However, soils from the University of Fort Hare Research Farm, where the field trials were conducted, developed from an alluvial parent material and are dominated by micas in the clay fraction, and have low amounts of quartz and kaolinite.

Soils of the Eastern Cape contain different amounts of feldspars and these minerals are present in substantial amounts in the silt fraction of soils from the University of Fort Hare Farm (Mandiringana *et al.*, 2005).

Soils in the Eastern Cape also contain low amounts of N and P and have the following nutrients in abundance: S, Mn, Zn, Cu, and B (Mandiringana *et al.*, 2005). All the soils are believed to be from Karoo sediments deposited during the late Carboniferous era (Visser, 1984).

## 2.5 Test crops

Tomato, butternut and Swiss chard were used as test crops as they are commonly grown in the Eastern Cape region by both commercial and small holder farmers.

# 2.5.1 Tomato

The tomato (*Lycopersicon esculentum* Mill) is one of the major vegetable crops cultivated in South Africa and is also grown commercially under irrigation in the Eastern Cape. The East London area is notable for hydroponic tomato production under shade cloth and in multispan tunnels, with decreasing traditional production in open fields.

The tomato is a high-value crop requiring considerable input in the form of capital and labour and requires good management. It has a relatively high fertilizer requirement in traditional production and requires a good spray programme to control a wide range of pests and pathogens that vary according to environmental factors, including the type of production system, whether in the open or under protection. Conditions at the University of Fort Hare (UFH) are very different from those near East London and from those prevailing in major tomato producing areas of Limpopo and Mpumalanga provinces, for example. At UFH tomatoes would be grown in the field or home garden during the frostfree spring, summer and early autumn periods. In East London they are grown yearround. In frost-free warm to hot subtropical areas of Limpopo and Mpumalanga provinces they would be grown any time except perhaps during the hottest summer months.

Prevailing weather conditions can have an important bearing on the incidence of tomato blights. However, regular preventive spray programmes need to be followed. Different tomato cultivars may have different degrees of susceptibility to important tomato diseases. From previous experience, common tomato diseases previously experienced at UFH have been early blight (*Alternaria solani*) and late blight (*Phytophthora infestans*), as well as spotted wilt virus. Pests were never a major problem although aphids, red spider and looper or American bollworm have periodically occurred (M. O. Brutsch, personal communication, 2007).

Early blight is caused by *Alternaria solani*. Affected plants develop small, dark coloured target spots on their leaves and stems especially on young seedlings in plant beds. Small, roundish spots are also produced on the fruit. The disease is favored by misty or rainy weather or nights with heavy dew (Nel *et al.*, 1999).

Late blight is caused by *Phytophthora infestans* and infected plants develop water-soaked or pale greenish spots on the leaves which turn brown or almost black. The disease also

attacks fruit and they develop small grayish green water-soaked areas that increase in size, turning brown and wrinkled. Cool wet nights with warm days provide optimum conditions for development of the disease (Nel *et al.*, 1999).

Spotted wilt is caused by a virus transmitted by thrips. Young leaves of infected plants develop numerous small, dark, circular spots and often show a bronzed appearance. The leaves sometimes turn dark and wither. The plants become severely stunted and ripe fruits show spots marked with concentric, circular bands of red and yellow (MacGillivray, 1953).

Some of the leading tomato and other vegetable growers in South Africa are apparently using EM. However, commercially available EM products supplied by EMROSA in South Africa were registered in 2006 only for sale in South Africa (Anon., 2006) and are still being tested. Initially, the trials reported here were being conducted with registration in mind but subsequently continued without the involvement of EMROSA, except that EM products were purchased from their local supplier.

# 2.5.2 Butternut

The butternut (*Cucurbita moschata* ) Duchesne ex Poiret) is a popular vegetable in South Africa. It is relatively easy to grow and stores well. Main problems could be pumpkin fly and powdery mildew. Tomato and butternut were the test crops initially recommended by EMROSA for EM tests at UFH.

2.5.3 Swiss chard

Swiss chard (Chenopodiaceae; *Beta vulgaris* subsp. *cycla*) also known as chard, leaf beet, or spinach beet is produced mainly for its large crisp leaves which are cooked as greens. Swiss chard is well suited to comparatively cool climates and has an optimum temperature that ranges between 18°C to 20°C. Continued exposure to temperatures less than 5°C induces seed production (bolting). Plants that are grown during hot weather conditions develop small leaves that are of inferior quality. During late summer, plants are subject to attack by leaf spot.

One of the advantages of using Swiss chard as a test crop, other than it having relatively few pests and diseases, is that leaves can be harvested as the crop grows thereby making it possible to monitor changes in response to different treatments as the crop is growing.

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# **CHAPTER 3**

# THE AGRONOMIC EFFECTIVENESS OF EFFECTIVE MICRO-ORGANISMS (EM), COMPOST AND MINERAL FERTILIZER ON TOMATO AND BUTTERNUT

## **3.1 Abstract**

Field and greenhouse experiments were conducted during the 2004-2005 summer season to evaluate the suitability of effective microorganism (EM) products to improve crop productivity and quality through enhanced soil microbial activities and pest and disease suppression. Tomato (Lycopersicon esculentum Mill) and butternut (Curcurbita *moschata*) were used as test crops. Experimental treatments were: control, effective microorganisms (EM), mineral fertilizer, EM + mineral fertilizer, compost, compost + EM, compost + mineral fertilizer and compost + mineral fertilizer + EM. With respect to the field-grown tomato experiment, application of EM caused a significant increase in the number of fruited tomato plants seven weeks after transplanting. However, application of EM alone or in combination with other amendments had a depressive effect on yield owing to an outbreak of early and late blights which affected the unsprayed (EM) treatments first, and also the most severely. Combined applications of EM with the amendments improved plant N content and increased soil N content above initial levels. Application of compost resulted in soil N and P concentrations higher than those of the control due to nutrients being slowly released from the compost material.

Results of the greenhouse study showed that EM had a negative effect on leaf dry matter yield, number of leaves formed, number of trusses formed, fruit yield and number of fruits formed. The negative effects of EM were ascribed to possible short term immobilization of N by the EM microorganisms that could have resulted in reduced N availability to plants. The lower number of fruits associated with EM application resulted in improved average fruit weight of tomatoes grown in the glasshouse as a result of more assimilates being partitioned to the few fruits formed. In the butternut trial, the application of EM also had a negative effect in that it depressed the total butternut yield, marketable yield and first grade yield. The results were attributed to immobilization of N induced by application of EM, and to the inability of EM to control pumpkin fly that attacked very young fruits, resulting in down grading of mature fruits.

# Key words: Butternut, compost, effective microorganisms (EM), mineral fertilizer, tomato

# **3.2 Introduction**

Production in agricultural systems depends largely on the action of the soil microbial biomass. Soil microbial biomass pools are an important component of the decomposer subsystem that regulates nutrient cycling, energy flow and plant and ecosystem productivity and form 2-3% of organic carbon. The soil microbial biomass pool responds quickly to changes in the soil environment (Pankhurst *et al.*, 1996).

Most agricultural practices affect soil quality by altering the physical, chemical and biological properties of soil and have led to a decrease in soil microbial populations (Valarini *et al.*, 2002). Addition of organic materials to soil, like compost, improves soil structure, serves as a source of nutrients, and strongly influences the soil microbial biomass and enhances soil enzyme activities (Albiach *et al.*, 2000). Addition of organic materials (OM) to soil is an agricultural practice for enhancing soil quality and a way of managing waste. Addition of OM into soils also encourages plant development and suppresses occurrence of soil-borne diseases (Erhart *et al.*, 1999; Cotxarrera *et al.*, 2002).

Increasing concern about the long-term productivity of agro-ecosystems has led to the need to develop management strategies that maintain and protect soil resources. It has also led to increased research efforts on the biological components of soil fertility dynamics (Smaling & Dixon, 2006). Agricultural scientists have been reviewing the concept of inoculating plants and soils with beneficial microorganisms as a way of creating an environment more conducive for plant growth (Asia-Pacific Natural Agriculture Network, 1995).

In the early 1990's the use of a microbial inoculum called effective micro-organisms (EM) together with organic materials was proposed and introduced to "nature farming systems" (Higa, 1994). EM inoculants are liquid microbial concoctions containing yeasts, actinomycetes, lactic acid and photosynthetic bacteria (Daly & Stewart, 1999). Most of the species in EM inoculants are heterotrophic and require organic sources of carbon and nitrogen for their nutrition. Therefore, EM inoculation has been more effective when applied in combination with organic materials to provide both carbon and nitrogen (Yamada & Xu, 2000). The microorganisms contained in the concoctions produce plant

hormones, beneficial bioactive substances and antioxidants while solubilizing nutrients (Higa & Parr, 1994).

Following the application of EM there is an increase in soil microorganisms that are beneficial for plant growth, resulting in more rapid mineralization of organic matter, suppression of soil-borne pathogens and increased crop yield and quality (Asia-Pacific Natural Network, 1995). Other studies have shown that inoculation of the agro-ecosystem with EM leads to an improvement in soil and crop quality in addition to higher crop yields (Higa & Parr, 1994; Li & Ni, 1995).

EM inoculants are produced and marketed in South Africa by EMROSA (Pty) Ltd. There is, however, only limited information on the effectiveness and use of EM in South Africa. Mupondi *et al.* (2006a, 2006b) found that co-composting of pine bark with EM had no effect on compost quality, but improved cabbage seedling growth.

The objectives of this study were therefore to evaluate (i) the effects of EM on the growth and yield of tomato and butternut in an Oakleaf soil in the Eastern Cape, (ii) the effects of co-application of EM with mineral fertilizer and compost, and (iii) the effects of EM application on selected soil properties. The hypotheses tested were (i) the application of EM increases growth and yields of tomato and butternut crops, (ii) co-application of EM with mineral fertilizer and compost has synergistic effects on butternut and tomato yield, and (iii) the application of EM improves soil chemical properties.

# 3.3 Materials and methods

# 3.3.1 Location and climate of the experimental site

The experiments were conducted on the Research Farm of the University of Fort Hare, Alice, Eastern Cape Province, South Africa during the 2004-2005 summer season. The farm is located at longitude 32<sup>0</sup>46' S and latitude 26<sup>0</sup>50' E at an altitude of about 535 m a.s.l. It has a warm temperate climate with an average annual rainfall of about 575 mm received mainly during the summer months. The soils are deep and alluvial, of the Oakleaf form (Oa), belonging to Jozini series, according to the South African system of soil classification (Soil Working Group, 1991). According to the soil map of the world compiled by FAO-UNESCO (1988), the soils are Eutric fluvisols (Fle). The soil had very low concentrations of total nitrogen, available phosphorus and organic C, but had high levels of micronutrients and exchangeable K (Table 3.1). The pH was 5.7 and suitable for growth of both tomato and butternut crops.

# 3.3.2 Effective microorganisms (EM)

Three experiments were conducted to evaluate the effectiveness of EM products supplied by EMROSA (Pty) Limited. The brands used in the trials included multiplied - EM, EM -F.P.E, EM 3- in-1 and EM - 5. The first was applied as a soil drench while the last three were applied as foliar pesticide mixtures. Multiplied - EM is a mixture of basic EM, molasses and water in a ratio of 1 : 1 : 20. EM - F.P.E stands for fermented plant extract and was prepared by mixing chopped fresh weeds, chlorine-free water, molasses (3 %) and multiplied - EM (3 %) in a ratio of 40 : 33 : 1 : 1. EM 3-in-1 is an insect repellent and was produced in a similar way as EM - F.P.E but using different ingredients. The ingredients used in this case were fresh garlic, chili pepper, ginger (400 g of each, chopped), black pepper (200 g powdered, 600 ml of multiplied - EM and 18 L of water. EM - 5 is a mixture of multiplied - EM, molasses, vinegar, strong distillation alcohol (more than 30 %) and water (Anon., 2004; 2005). All four brands of EM were used in EM - treated plots. Multiplied - EM was applied as a soil drench by dissolving EM in water in a ratio of 1 : 300 and the resultant solution applied at a rate of 200 L per experimental unit seven days before seedlings were transplanted. During the course of the experiment, multiplied - EM solution, in a ratio of 1 : 500, was applied to respective EM - treated plots at the rate of 50 L per week. Mixtures of EM - FPE, EM 3- in-1 and EM - 5 diluted with water in a ratio of 1 : 800 were sprayed to control diseases and pests in EM treated plots.

## 3.3.3 Compost

Just Nature compost was used for the field tomato experiment and an equivalent of 27 t ha<sup>-1</sup> (which supplied 332.1, 99.09, 88.56 kg ha<sup>-1</sup> of N, P, and K, respectively) was applied. The compost is made up of animal manure and other organic refuse. It is manufactured by ABAKOR Ltd and distributed by a number of marketers in South Africa. Some characteristics of the compost are shown in Table 3.2. Nature's Super Grow compost was used for the greenhouse tomato experiment and butternut field experiment at a rate of 27 t ha<sup>-1</sup> which supplied 54, 13.5 and 10 kg ha<sup>-1</sup> of N, P and K, respectively.

The compost is made up of pine bark and other organic refuse material and is manufactured by C.S.M at Brakkerfontein, Port Elizabeth. Some characteristics of the Nature's Super Grow compost are shown in Table 3.2.

The compost materials were analyzed for nutrient concentrations after the experiments were conducted, as described in section 3.4.2.

3.3.4 Experiment 1: Effects of the integrated use of EM, compost and mineral fertilizer on field-grown tomato

The objective of this experiment was to evaluate the effects of EM on the growth and yield of tomatoes (*Lycopersicon esculentum* Mill) grown under field conditions. The experiment was a randomized complete block design (RCBD) with six replicates. Treatments were: control, EM only (EM), recommended fertilizer (RF) (N 200: P 90 kg ha<sup>-1</sup>), EM + recommended fertilizer (EM + RF), compost only (Comp), compost + EM (Comp + EM), compost + recommended fertilizer (Comp + RF), compost + recommended fertilizer + EM (Comp + RF + EM). These amendments were applied to plots measuring 4.5 m x 5 m. The compost and the recommended fertilizer were applied in the top 5-7 cm of soil by mixing with a spade.

## 3.3.4.1 Agronomic practices

Tomato seeds (cv Hytec 36) for raising seedlings to be planted in EM-treated plots were soaked in 0.1 % multiplied - EM for 30 minutes. The other seeds were soaked for 30 minutes in distilled water only prior to sowing in Hygromix seedling mix (marketed by

Hygrotech, South Africa) in cavity trays in the greenhouse. After four weeks, the seedlings (10 to 15 cm high) were transplanted on pre-irrigated plots using a spacing of 50 cm within rows and 150 cm between rows. Plant growth was monitored regularly. Some plants were destroyed and or damaged by hail two weeks after transplanting. Copper Count - N {copper ammonium carbonate (SL)}, and Dithane M45 {Mancozeb (WP) applications were applied alternately and cutworm bait was applied in plots where EM was not applied. Treatment effects on plant growth were evaluated by counting the number of plants that had flowered five weeks after transplanting and plants with fruits at seven weeks after transplanting. Disease infestation was measured by counting the number of infested plants per plot. Plants affected by spotted wilt virus were uprooted and discarded to prevent the spread of the disease to unaffected plants (those without visible symptoms). The experiment was prematurely terminated at 10 weeks after transplanting due to severe infection by early and late blights. Yield was based on a single row, leaving out the border rows. Yield variables measured included the number of fruits, fruit set, total mass of fruits, and proportion of marketable fruits.

Characteristics	Value
pH (KCI)	5.7
Bulk density (gcm <sup>-3</sup> )	1.23
Total N (g kg <sup>-1</sup> )	0.9
Available P (mg kg <sup>-1</sup> )	59
Exchangeable K (mg kg <sup>-1</sup> )	441
Exchangeable Ca (mg kg <sup>-1</sup> )	1028
Exchangeable Mg (mg kg <sup>-1</sup> )	246
$Zn (mg kg^{-1})$	15.2
$Mn (mg kg^{-1})$	46
Organic C (g kg <sup>-1</sup> )	6.0
$Cu (mg kg^{-1})$	2.9

 Table 3.1 Selected properties of the experimental soil (upper 0-30 cm depth)

Table 3.2 Selected properties of the compost materials used

Characteristic	Just Nature	Nature's Super Grow
pH (H <sub>2</sub> O)	-	4.33
EC ( $\mu$ S cm <sup>-1</sup> )	-	2.37
Total N (g kg <sup>-1</sup> )	12.3	2.0
Total P (g kg <sup>-1</sup> )	3.67	0.5
Total K (g kg <sup>-1</sup> )	3.28	0.4
Polyphenol (g kg <sup>-1</sup> )	-	9.8
Total C (g kg <sup>-1</sup> )	215.8	193.3
C:N	17.5	96.65
C:P	58.8	386.6

# 3.3.4.2 Soil and leaf analysis

Soil and leaf samples were taken at harvest to assess treatment effects on soil and plant nutrient content. Leaf sampling was done by taking the third youngest fully expanded leaf from shoots (Jones *et al.*, 1971) of 10 plants from the experimental row of each experimental unit. The leaf dry matter was determined after oven drying to constant mass at 65 <sup>o</sup>C. The dried samples were ground in a hammer mill to pass through a 1 mm mesh sieve. The ground samples were digested with sulphuric acid, selenium powder and salicylic acid mixture for the determination of total P and K (Okalebo, Gathua and Woomer, 2002). Phosphorus was read on a colorimeter following colour development by the molybdenum blue method (Okalebo *et al.*, 2002). Potassium in digested samples was determined by flame photometry. Total nitrogen was determined using a LECO TruSpec C/N auto analyzer (LECO Corporation, 2003).

Soil samples taken after harvest were air dried for 2 weeks and ground to pass through a 2 mm mesh sieve. Soil pH and electrical conductivity (EC) were determined in water extracts as described by Okalebo *et al.* (2002). Samples were shaken in distilled water in a ratio of 1:2:5 on a reciprocal shaker for 10 minutes and left standing for 30 minutes, then shaken again for 2 minutes, after which pH was read using a WTW pH 526 meter, while EC was read on a WTW 330i conductivity meter. Total-N was determined using a LECO TruSpec C/N auto analyzer (LECO Corporation, 2003) and extractable P and K were determined following the Ambic-2 extraction method (Non-Affiliated Soil Analysis Work Committee, 1990).

3.3.5 Experiment 2: Effects of the integrated use of EM, compost and mineral fertilizer on greenhouse-grown tomato

As a result of the premature termination of the field trial due to severe disease infestation, it was decided to repeat the experiment under green-house conditions. The number of treatments, soil and experimental design used were as described in Section 3.4 for the tomato field experiment except that treatments were replicated 10 times. Each replicate consisted of two tomato plants in 30 cm<sup>3</sup> pots containing 15 kg of soil. The agronomic practices were basically as described for the field experiment. Growth parameters measured included plant height, stem girth, number of leaves and trusses formed. As there were no significant observable signs of disease infection during the initial growth stages, it was not necessary to score disease incidence. However, occurrence of stem-end rot (a fruit physiological disorder) was observed and noted at harvest. Harvesting of mature fruits was done at 12 weeks after planting and yield was evaluated as number of fruits, total mass of fruits, average mass of fruit and proportion of marketable fruits. Leaf and stem biomass were also measured on a dry mass basis. Soil and leaf samples were taken at harvest time to assess treatment effects on soil and plant nutrient content. Leaf sampling was done by taking leaves from the fourth to the sixth clusters (Jones et al., 1971). The leaf dry matter was determined after oven drying to constant mass at 65°C. The dried samples were ground in a hammer mill to pass through a 1 mm mesh sieve and analyzed as described for the field experiment (Section 3.3.4.2).

3.3.6 Experiment 3: Effects of the integrated use of EM, compost and mineral fertilizer on field-grown butternut

The objective of this experiment was to evaluate the effects of EM on growth and yield of a butternut (*Curcurbita* sp) crop grown under field conditions. Treatments, experimental design and the soil were as described in Section 3.3.4 for the tomato field trial. There were four replicates, and each experimental unit measured 3 m by 5 m. To avoid having an excessively large experimental area, border rows were provided only for the perimeter plots. Application of EM, compost and mineral fertilizers was as described in Section 3.3.4 for the tomato field experiment.

# 3.3.6.1 Agronomic practices

Butternut seeds (cv Waltham) for the EM treatments were soaked in 0.1 % EM for 30 minutes and the other seeds were soaked in distilled water for 30 minutes prior to sowing directly in the field. Some planted seeds failed to germinate, necessitating replanting. This resulted in an uneven plant stand in some treatments, which was compounded by a hail storm, six weeks after planting that damaged and even destroyed some plants. Pumpkin fly control in plots not treated with EM was done once a week by alternating Trichlorfon 950 SP (Trichlorfon) and Topaz (Penconazole).

Disease and pest incidence and severity were assessed by counting the number of damaged leaves, the number of fruits that failed to develop due to attack by pumpkin fly and the number of fruits at harvest that had been affected by the fly. Yield of marketable

and unmarketable butternuts was evaluated by counting and weighing fruits. Unmarketable fruits were those damaged by pumpkin fly and those with prominent fruit cracks. Plant and soil samples were collected and analyzed as described in Section 3.3.4.2 for the field-grown tomatoes.

# 3.3.7 Data analysis

To eliminate the effects of an uneven plant stand resulting from removal of plants infected with spotted wilt virus, and damage from hail in the case of tomatoes planted in the field, or caused by unsatisfactory germination and by hail damage in the case of the butternut experiment, analysis of covariance was conducted on growth data, using plant population as the co-variant. The rest of the data was subjected to analysis of variance (ANOVA) using the SAS statistical package while means were separated using least significance differences (LSD) at the 0.05 level of significance.

## 3.4 Results

3.4.1 Effects of the integrated use of EM, compost and mineral fertilizer on field-grown tomato

# 3.4.1.1 Effects on number of fruited plants seven weeks after transplanting

There were no significant treatment effects on the number of flowered tomato plants, five weeks after transplanting. However, there were significant treatment effects on the number of fruited tomato plants, seven weeks after transplanting. EM application appeared to promote earliness of fruiting although the results were not statistically significant, possibly because of a high coefficient of variation (Table 3.3). For example, application of sole EM resulted in an increase of 83.3 % in the number of fruited plants relative to the control. When applied with compost a 62.1 % increase in the number of fruited plants was recorded relative to the compost treatment. Application of EM with mineral fertilizer resulted in a 52 % increase in the number of fruited plants relative to the mineral fertilizer treatment. Integrated use of EM, mineral fertilizer and compost resulted in a 51.6 % increase in the number of fruited plants compared to the compost and mineral fertilizer treatment.

# 3.4.1.2 Effects on tomato fruit yield

Treatment effects on tomato fruit yield are shown in Table 3.3. Only the reference fertilizer increased yield significantly relative to the control while sole application of EM or its application with compost, reference fertilizer or both, resulted in yield decreases. For example, the sole application of EM resulted in a 26.9 % decrease in fruit yield relative to the control while its application with compost resulted in a 23.2 % decrease in fruit yield relative to the compost treatment. The combination of EM and mineral fertilizer decreased fruit yield by 46 % relative to the fertilizer treatment and a 49.6 % decrease in fruit yield was observed relative to the compost + mineral fertilizer treatment when EM was co-applied with mineral fertilizer and compost.

Application of EM did not result in a significant reduction in pest and disease incidence and severity. All the plants were severely affected by the blight (Plate 3.1) that developed after a prolonged period of rainfall and overcast weather in February (Figure 3.1), and this necessitated the premature termination of the trial. Initially, plants in EM-treated plots looked better than the plants of the control treatment (Plate 3.2). As a result of the blight attack, the proportion of marketable tomato fruits was also severely lowered (Table 3.3). The unmarketable yield was accounted for mainly by excessively small fruits and fruit damaged by pests and diseases, in this case mainly American Bollworm and Late blight.

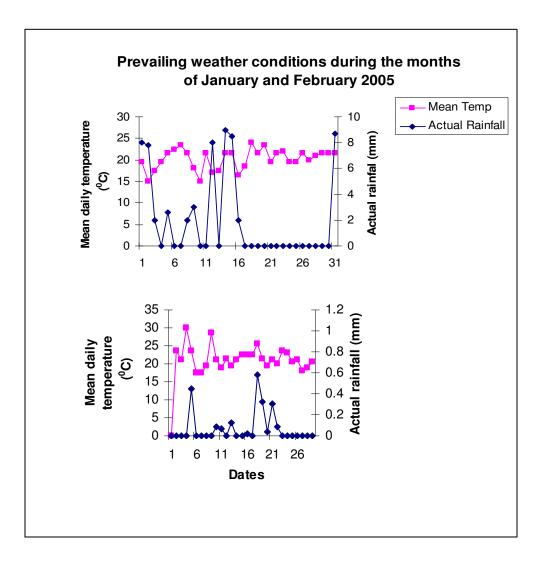


Figure 3.1 Prevailing temperatures and rainfall in January (above) and February (below) 2005



**Plate 3.1** Tomato plants affected by Late blight. Note that the EM - treated plants (foreground) were more affected by Late blight than those which were sprayed with fungicide (background).



Plate 3.2 Early growth of field-grown tomato. Note better growth at this stage with EM (right) than for control (left).

Table 3.3 Effects of EM, compost and recommended fertilizer combinations on the proportion of plants that had fruited seven weeks after transplanting of tomato, on total fruit yield (t ha<sup>-1</sup>) and on marketable yield (t ha<sup>-1</sup>) of field-grown tomato.

Treatment	No. of fruited	Yield	Marketable
	Plants ha <sup>-1</sup> $(10^{-3})$	$(t ha^{-1})$	Yield (t ha <sup>-1</sup> )
Control	2b	28.3abc	15.0ab
EM	4ab	20.7dc	8.1cd
RF	5ab	36.3a	15.5ab
EM+RF	7a	19.6cd	6.9d
Comp	3b	25.9bcd	14.3abc
Comp+EM	5ab	19.9cd	8.4cd
Comp+RF	3b	34.7ab	16.2a
Comp+RF+EM	5ab	17.5d	9.5bcd
CV (%)	55.0	29.9	42.1

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer, Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at P $\leq$  0.05 according to the LSD test.

Application of the different amendments, singly and in combination, significantly affected leaf N content (Table 3.4), which was expected considering the relatively low N and organic matter status of the soil (Table 3.1). Combined applications of EM with the amendments improved leaf N content compared to single application of the amendments. Leaf N content ranged from 36 g kg<sup>-1</sup> to 49.9 g kg<sup>-1</sup>. EM alone increased leaf N content by 38.6 % relative to the control. Application of EM + RF increased leaf N content by 15.1 % relative to the mineral fertilizer treatment. When applied with compost, a 21.6 % increase in leaf N content was observed relative to the compost treatment. Application of Comp + RF + EM resulted in a 16.3 % increase in leaf N content relative to the Comp + RF treatment. The highest leaf N content was observed in plots treated with EM + RF but the greatest effect of EM on leaf N content was attained with application of EM alone. The leaf P and K content were not significantly affected by any of the treatments (Table 3.4). The content of N and K in leaves was higher than the critical levels of 12 g kg<sup>-1</sup> for N and 3 g kg<sup>-1</sup> for K, respectively. Leaf P content, on the other hand, was lower than the critical level of 3 g kg<sup>-1</sup> for P reported by Foth and Ellis (1988).

Treatment	Leaf			Soil
	N	Р	K	N P K
	(g kg <sup>-1</sup> )	$(g kg^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$ $(g kg^{-1})$ $(g kg^{-1})$
Control	36.0c	0.8ab	22.3a	6.1d 0.5d 41.3b
EM	49.9a	0.8ab	23.0a	7.0cd 0.5cd 42.9b
RF	43.6abc	0.7ab	24.4a	6.8d 0.7cd 39.4b
EM+RF	50.2a	0.9a	25.3a	7.7bcd 0.9bc 35.0b
Comp	40.8bc	0.7ab	21.3a	9.8a 1.3a 35.9b
Comp+EM	49.6ab	0.8ab	25.4a	9.6ab 1.2ab 61.0a
Comp+RF	41.7abc	0.8ab	19.8a	10.3a 1.5a 45.1ab
Comp+RF	48.5ab	0.6b	22.1a	9.0abc 1.5a 42.0b
CV (%)	17.2	28.0	23.2	20.8 33.1 33.4

**Table 3.4** Effects of EM, compost and recommended fertilizer combinations on leaf andsoil N, P and K of field-grown tomato.

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer, Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at P $\leq$  0.05 according to the LSD test.

There were significant treatment effects on residual soil nutrient concentration. Residual soil N content ranged from 6.1 g kg<sup>-1</sup> to 10.3 g kg<sup>-1</sup>. Residual soil N in plots treated with sole EM, RF and EM + RF was not significantly different from the control (Table 3.4).

Application of compost alone or across treatments significantly increased soil N concentration relative to the control treatment. A similar trend was observed with available P. Soil extractable P ranged from 0.5 g kg<sup>-1</sup> to 1.5 g kg<sup>-1</sup>. Soil available K ranged from 35 g kg<sup>-1</sup> to 61 g kg<sup>-1</sup>. Addition of EM with compost did not result in higher levels of N and P being released. Application of Comp + EM increased available K relative to the control.

# 3.4.2 Effects of the integrated use of EM, compost and mineral fertilizer on greenhousegrown tomato

# 3.4.2.1 Effects on leaf dry matter yield

The objective of the greenhouse tomato trial (Plate 3.3) was to try and follow up the field experiment under a controlled environment. There were significant ( $P \le 0.05$ ) treatment effects on leaf dry matter yield. Application of EM and compost alone or their combined application did not increase leaf dry matter yield of tomato significantly over that of the control (Table 3.5). An 8.5 % decrease in leaf dry matter yield with sole EM application was observed relative to the control. When applied with compost, a 19.3 % decrease in leaf dry matter yield was observed relative to the compost treatment. The apparent depressive effect of EM was further observed when it was applied with recommended fertilizer whereby this treatment resulted in a 7.2 % decrease in leaf dry matter yield relative to recommended fertilizer treatment. Application of EM with mineral fertilizer and compost resulted in a decrease in leaf dry matter yield of 3.7 % relative to the mineral fertilizer and compost treatment. The results, therefore, demonstrated a definite negative trend whereby the application of EM singly or in combination with mineral fertilizer or compost depressed leaf dry matter yield.

#### 3.4.2.2 Effects on tomato fruit yield

The treatment effects on tomato fruit yield are shown in Table 3.5. Application of sole EM or combined with compost or mineral fertilizer had a negative effect on fruit yield. Application of EM alone in this study resulted in a 15.4 % decrease in yield over the control (Table 3.5). Similarly, application of EM with compost resulted in a 24.1 % decrease in fruit yield relative to the compost treatment and reduced fruit yield by 6.5 % when it was applied with mineral fertilizer relative to the mineral fertilizer treatment. When EM was combined with both mineral fertilizer and compost a 12.3 % decrease in fruit yield was recorded relative to the compost + mineral fertilizer treatment. Treatments that received a combination of compost + mineral fertilizer gave the highest fruit yield, with a 51 % fruit yield increase relative to the control treatment.

# 3.4.2.3 Effects on average fruit mass

Average fruit mass was significantly ( $P \le 0.05$ ) affected by some of the treatments. Although sole application of EM did not have a significant effect on average fruit mass, a positive trend was observed with its application. Sole application of EM resulted in an 11.6 % increase in average fruit mass relative to the control. When EM was applied with compost, a 9.9 % increase in average fruit mass was recorded relative to the compost treatment. On the other hand, application of EM with mineral fertilizer resulted in a 4.7 % increase in average fruit mass relative to the mineral fertilizer treatment. Application of EM + mineral fertilizer + compost resulted in an 11 % decrease in average fruit mass over the mineral fertilizer + compost treatment (Table 3.5).

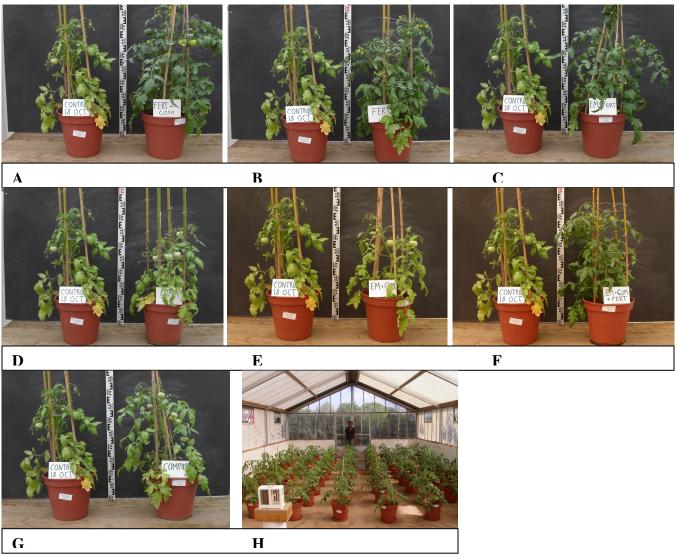


Plate 3.3 Tomato plants growing in the greenhouse: A = Control vs Fertilizer + Compost, B = Control vs Fertilizer, C = Control vs EM, D = Control vs EM + Fertilizer, E = Control vs EM + Compost, F = Control vs EM + Compost + Fertilizer, H = layout of the experiment in the greenhouse.

# Table 3.5 Effects of EM, compost and mineral fertilizer combinations on leaf dry matter yield (DMY), leaf number, number of trusses, fruit yield, fruits formed and on average fruit mass of greenhouse-grown tomato.

Treatment	Leaf DMY (g pot <sup>-1</sup> )	Leaf number pot <sup>-1</sup>	Number of Trusses pot <sup>-1</sup>	Fruit yield (g pot <sup>-1</sup> )	Fruits formed pot <sup>-1</sup>	Average fruit mass (g fruit <sup>-1</sup> )
Control	45.8cd	21c	11cd	418.9de	15c	42.1bc
EM	41.9d	17c	7e	354.4e	8cd	47.0ab
RF	57.3ab	39a	16a	563.4ab	20ab	29.8d
EM+RF	53.2abc	34ab	15ab	526.6bc	17b	31.2d
Comp	48.7bcd	20c	9de	470.0cd	10c	51.5ab
Comp+EM	39.3d	18c	8e	356.8e	7d	56.6a
Comp+RF	70.0a	32b	15ab	632.8a	20a	32.7cd
Comp+RF+EM	59.7a	33b	13bc	555.1ab	20a	29.1d
C.V (%)	16.8	22	20.7	18.3	20.2	27.2

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer,Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at P $\leq$  0.05 according to the LSD test.

# 3.4.2.4 Effects on plant and soil nutrient content

Leaf N, P and K content are shown in Table 3.6. The leaf N content ranged from 9 g kg<sup>-1</sup>

to 13.9 g kg<sup>-1</sup> and the content for most treatments was lower than the critical level of 12 g

kg<sup>-1</sup> for N cited by Foth and Ellis (1988). The low leaf N content was reflected in the yellowing of some plants (Plate 3.3). Application of EM with mineral fertilizer significantly increased leaf N content relative to the control and was the only treatment that resulted in leaf N content greater than the critical level. Application of compost + RF + EM improved leaf N content and N uptake and application of sole EM increased leaf N content but not its uptake. Application of EM singly or in combination with compost led to a decrease in leaf N content and plant N uptake.

The leaf P content ranged from  $1.3g \text{ kg}^{-1}$  to  $2 g \text{ kg}^{-1}$  and was much lower than the critical leaf P content of 3 g kg<sup>-1</sup> cited by Foth and Ellis (1988). Application of compost resulted in the highest leaf P content with application of EM + RF and Comp + RF + EM resulting in the lowest leaf P content

The leaf K content was higher than the critical level of 3 g kg<sup>-1</sup> cited by Foth and Ellis (1988). Soil nutrients were not significantly influenced by treatments (Table 3.6).

Treatment		Plant			Uptake			Soil		
	Ν	Р	K	N	Р	K	N	Р	K	
	$(g kg^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$	$(g pot^{-1})$	$(g pot^{-1})$	$(g \text{ pot}^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$	
Control	9.9b	2.1a	17.3abc	4.41dc	0.95bc	7.92bcd	3.8a	1.4b	18.4b	
EM	10.1b	2.1a	17.6abc	4.10dc	0.88bcd	7.29cd	3.5a	0.7e	18.8b	
RF	11.9b	1.8dc	15.4c	6.67ab	1.00ab	8.86bc	3.2a	1.4b	17.5b	
EM+RF	13.9a	1.4e	15.6c	7.30a	0.71d	8.21bcd	4.0a	0.9de	16.5b	
Comp	9.8b	2.4a	19.9ab	4.73dc	1.17a	9.65bc	3.5a	1.7a	23.0a	
Comp+EM	9.1b	2.4a	16.7abc	3.37d	0.95bc	6.34d	3.4a	1.0cd	24.6a	
Comp+RF	9.0b	1.7d	16.2bc	5.49bc	1.05ab	10.06ab	3.3a	1.8a	18.7b	
Comp+RF+EM	11.7ab	1.3e	20.6a	6.97ab	0.78cd	12.16a	3.4a	1.1c	16.9b	
CV (%)	30	15.6	18.8	27.5	17.5	23	52	18.5	15.1	

Table 3.6 Effects of EM, compost and mineral fertilizer combinations on plant and soil N, P and K of greenhouse-grown tomato

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer, Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at  $P \le 0.05$  according to the LSD test.

3.4.3 Effects of the integrated use of EM, compost and mineral fertilizer on field - grown butternut

# 3.4.3.1 Effects on fruit yield and yield components

Application of EM and compost alone, or their combination, did not significantly affect total fruit yield of butternut over the control treatment (Table 3.7). However, the application of EM alone and in combination with compost or mineral fertilizer had a consistent but non-significant depressive effect on total fruit yield, marketable yield and yield termed as first grade. Application of EM alone resulted in a 15.6 %, 12.6 % and 18.1 % reduction in total fruit yield, marketable yield and first grade yield, respectively, relative to the control treatment. When EM was applied with mineral fertilizer, a 42 %, 50.5 % and 57.8 % decrease in total fruit yield, marketable yield and first grade yield, respectively, was observed relative to the recommended fertilizer treatment. When EM was applied with compost, decreases of 8.2 %, 16.8 % and 5.2 % in total fruit yield, marketable yield and first grade yield, respectively, were observed relative to the compost treatment. Similarly, the application of EM with mineral fertilizer and compost resulted in 42.8 %, 55.2 % and 57.4 % decreases in total fruit yield, marketable yield and first grade yield, respectively, relative to a combination of compost + mineral fertilizer treatment. The highest total fruit yield, marketable yield and first grade yield were observed in plots treated with mineral fertilizer + compost, where yield was boosted by 52.3 %, 83.9 % and 108.7 %, respectively, relative to the control treatment.

Treatments	Total yield	Marketable	First-grade
	$(t ha^{-1})$	yield (t ha <sup>-1</sup> )	$(t ha^{-1})$
Control	25.6bc	19.9bc	12.7bc
EM	21.6c	17.4c	10.4c
RF	34.8ab	30.9ab	24.4ab
EM+RF	20.2c	15.3c	10.3c
Comp	21.9c	19.6bc	11.6c
Comp+EM	20.1c	16.3c	11.0c
Comp+RF	39.0a	36.6 a	26.5a
Comp+RF+EM	22.3c	16.4c	11.3c
CV (%)	32.5	37	53.4

**Table 3.7** Effects of EM, compost and mineral fertilizer combinations on fruit yield of field-grown butternut.

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer,Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at P $\leq$  0.05 according to the LSD test.

# 3.4.3.2 Effects on plant and soil nutrient content

Individual factors (EM, compost and mineral fertilizer) did not have any significant effect

on leaf and soil N, P and K (Table 3.8) content.

Treatment	Leaf				Soil	
	N	Р	K	N	Р	K
	(g kg <sup>-1</sup> )	(g kg <sup>-1</sup> )	$(g kg^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$	$(g kg^{-1})$
Control	15.6a	1.4a	16.8a	2.4a	0.5a	34.5a
EM	16.3a	1.4a	18.3a	3.7a	0.6a	42.3a
RF	15.7a	1.6a	20.1a	3.8a	0.7a	36.4a
EM+RF	17.1a	1.6a	22.4a	3.2a	0.9a	33.4a
Comp	16.4a	1.6a	18.9a	3.1a	0.7a	36.0a
Comp+EM	15.6a	1.4a	18.6a	3.2a	0.6a	39.4a
Comp+RF	14.7a	1.5a	19.6a	3.4a	0.9a	39.6a
Comp+RF+EM	16.1a	1.5a	18 8a	2.5a	0.9a	40.7a
C.V (%)	11.0	13.3	13.5	30.6	21.1	15.0

**Table 3.8** Effects of EM, compost and recommended fertilizer combinations on leaf and soil N, P and K of field-grown butternut

EM: effective microorganisms, RF: recommended fertilizer, EM + RF: effective microorganisms and recommended fertilizer, Comp: compost, Comp + EM: compost and effective microorganisms, Comp + RF: compost and recommended fertilizer,Comp + RF + EM: compost, recommended fertilizer and effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at P $\leq$  0.05 according to the LSD test.

# **3.5 Discussion**

3.5.1 Effects of the integrated use of EM, compost and mineral fertilizer on field-grown tomato

#### 3.5.1.1 Effects on number of fruited plants seven weeks after transplanting

The results obtained in this study showed that EM had a positive effect on fruiting of tomato plants, with the highest number of fruited plants being observed where EM was applied with mineral fertilizer. This suggests the possible existence of some synergistic activities between mineral fertilizer and EM that could lead to improved fruiting in tomato. Treatments with combined applications of EM and chemical fertilizer had a significantly higher number of fruited plants compared to treatments with combined applications of compost and chemical fertilizer, compost alone or control. The higher number of fruited plants associated with combined applications of EM was possibly due to the production of plant growth regulators by microorganisms associated with the EM amendment, as suggested by Arshad & Frankenberger (1992). Following application of EM into the soil, there is an increase in soil microbial biomass which increases the rate of symbiotic biological nitrogen fixation through increases in Azotobacter bacteria (Hussain et al., 1994). Further, it is speculated that following application of EM, there is an increase in the rate of photosynthesis in plants through increased utilization efficiency of solar energy from the actual utilization of less than 3 % to between 10-20 % (Lenghari, undated). Similar results were obtained by Xu, Wang & Mridha (2000) where application of EM increased fruit yield and plant growth of a tomato crop.

#### 3.5.1.2 Effects on tomato fruit yield

Although application of EM had a positive effect on fruiting of tomato plants, its application alone or in combination with compost or fertilizer appeared to have had a negative effect on tomato fruit yield. The apparent depressive effects of EM on tomato fruit yield could have been a result of the severe blight infestation on the tomato crop which started in the EM-treated plots before rapidly spreading to the other treatments. In plots treated with EM, only EM - FPE, EM - 5 and EM - 3 in 1 were used to try to control diseases and pests. The EM residues (molasses rich in C and N) on the leaves of treated plants could have served as a good substrate for microorganisms, some of them pathogenic, like those causing tomato blight.

Early and late blights are caused by *Alternaria solani* and *Phytophthora infestans*, respectively, which either absorb their food from the surrounding water or soil, or may invade the body of another organism to feed (De Graaf & Hamann, 2001). The application of EM was totally ineffective in controlling the blights. It is possible that in other instances where EM has been found to have positive effects on tomato, the weather may not have been favorable for blight attack. In the Eastern Cape, where this experiment was carried out, the weather is at times very conducive to the development of blight and the results clearly indicate that EM may not be effective in controlling it. Nicholson (personal communications, 2006) is of the opinion that with tomatoes in the Eastern Cape coast and midlands, EM alone will not prevent outbreaks of late blight when weather conditions are favorable for its development.

Early blight development is favored by warm temperatures and high humidity conditions, but can occur in drier situations where heavy dews occur. Early blight disease is widespread in most areas where potatoes or tomatoes are grown. Late blight development is favored by high humidity and fairly low temperatures. At the beginning of the season, plants might develop quite satisfactorily for a time and only one or two lesions might be noticed. Under favorable conditions spores are formed on these primary lesions and bring about infections over a large area (Mercure, 1998). In South Africa, early blight is found in all provinces and is a limiting factor in production in late summer (Van der Waals, Undated) and poses a constant threat to production in coastal regions and the midlands (Denner, undated).

These results suggest that one could use EM initially to stimulate better flowering and fruiting but that if conditions are conducive to the development of fungal diseases, such as early and late blight, then registered fungicides should be utilized following the initial use of EM.

Although application of EM had a negative effect on tomato fruit yield, its application had a positive effect on leaf N and soil N content at harvest time. Both single and combined applications of EM and amendments increased soil N above initial levels as well as over the control (Tables 3.1 and 3.4). These results could be attributed to the effect of EM stimulating mineralization of organic matter, with subsequent release of more nutrients into the soil-plant system (Higa & Kinjo, 1991; Daly & Stewart, 1999). It is further suggested that nitrogen did not limit tomato yield and that the observed

negative effect of EM on field grown tomato yield was, as noted earlier, due to the blight attack which EM was ineffective in controlling.

# 3.5.2 Effects of the integrated use of EM, compost and mineral fertilizer on greenhousegrown tomato

Results of the greenhouse tomato experiment set up as a follow-up to the field study revealed trends similar to those observed in the field study. The application of sole EM or in combination with compost or mineral fertilizer had a negative effect on leaf dry matter yield, number of leaves, number of fruit trusses and tomato fruit yield. In the field study, the negative effect of EM application on tomato growth and yield was attributed to its inability to control early and late blights that affected the crop during the growing season. The greenhouse tomato crop was, however, not affected by early or late blight so disease infestation could not be the cause for the observed negative effect of EM on tomato growth in the greenhouse. It is possible that the inoculated effective microorganisms proliferated very fast in the soil, thriving on the native and added nutrients in the soil and resulted in their temporary immobilization. Therefore, it is speculated that introduction of EM microbes into the soils could have set in short-term competition between the microbes and the plants for nutrients such as nitrogen in the limited pot soil volumes whose net effect was reduced plant growth. This was not observed in the field possibly because plants were exploiting an unlimited soil volume. The suspected nutrient immobilization could also have been exacerbated by the introduction of carbon through molasses while applying EM to the soil. This could have stimulated indigenous microbial biomass pool activities in soil (Daly & Stewart, 1999), causing N and P immobilization and reduced plant growth (Bååth *et al.*, 1978; Ritz & Griffiths, 1987). This speculation is supported by the low N uptake observed in plots treated with EM (Table 3.6).

The combined application of EM with compost, as recommended by EM promoters, is scientifically sound, as the compost is expected to serve as a source of labile C and nutrients for proliferation of the microorganisms. However, results obtained from this study showed a negative effect of combined application of EM + compost. This observation could possibly be due to N immobilization by the soil microbial biomass pools as the total N content of the compost material was below the critical level of 11.5 g kg<sup>-1</sup> suggested by Bartholomew (1965). Addition of organic materials with a total N content less than 11.5 g kg<sup>-1</sup> can initiate N immobilization in the soil (Bartholomew, 1965). The suspected nutrient immobilization can also be explained in terms of C: N ratio and C: P ratio of the compost material. The optimum C: N ratio for speedy decomposition of organic material and subsequent N mineralization is reported to be less than 30 (Brady & Weil, 1999). In terms of C: P ratio, Rustad & Cronan (1988) suggested that the critical C: P ratio of organic materials above which nutrient immobilization can occur ranges between 350 and 480. The C: N ratio of the compost material used was 96.7. This value was far above the optimum level suggesting that addition of compost could have caused N immobilization, reducing plant-available N. However, the C: P ratio of the compost material was within the suggested range, ruling out the possibility of P immobilization.

3.5.3 Effects of the integrated use of EM, compost and mineral fertilizer on field - grown butternut

The results for the butternut field trial were affected by uneven plant stands owing to poor seed germination and hailstorm damage to those that germinated. Nevertheless, they also revealed a consistent negative effect of EM application alone or in combination with compost or fertilizer on total fruit yield, marketable yield and yield termed as first grade (Table 3.7). The observed depressive effects of EM on fruit yield of butternut could be attributed to the inability of the EM to control pumpkin fly that attacked small developing fruits, often leading to their lack of further development. Fruits that managed to develop had ugly scars that reduced their market value and were graded as unmarketable (Plate 3.4). The depressive effects of EM could not be linked to N immobilization as the application of EM had a positive effect on soil N (Table 3.8).



Plate 3.4 Unmarketable butternuts with pronounced pumpkin fly damage and cracks

# **3.6 Conclusions**

The results of this initial study were inconclusive with respect to the effectiveness of EM on crop growth. Results of the tomato field experiment suggested that EM application could potentially increase the yields of tomato as it significantly increased the proportion of fruited plants in the field though this did not translate into positive yield increases presumably due to the inability of EM to control early and late blight. However, the addition of EM also depressed the yield of greenhouse tomato which was attributed to possible initial nutrient immobilization as blight infestation did not occur in the glasshouse. These mixed findings suggest the need for a more systematic study to provide a better understanding of the mechanisms by which EM influences plant growth.

The use of compost in the greenhouse tomato experiment and in the butternut field experiment did not have the desired effect as the compost may have induced N immobilization in soil due to its wide C: N ratio. The resultant N immobilization reduced the yield of both the tomato and butternut crop. The effect of organic material on EM effectiveness was, therefore, explored further in a separate study (Chapter 4) in which goat manure with a narrow C: N ratio was used.

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#### **CHAPTER 4**

# EFFECTS OF CO-APPLICATION OF GOAT MANURE, COMPOST AND MINERAL FERTILIZER WITH EFFECTIVE MICRO-ORGANISMS (EM) ON YIELD OF SWISS CHARD AND ON SELECTED SOIL PROPERTIES

# 4.1 Abstract

A greenhouse experiment was conducted to assess the effects of EM on single and combined applications of mineral fertilizer, compost and goat manure on Swiss chard (*Beta vulgaris*) growth and on selected soil properties. The crop was harvested after eight weeks and allowed to re-grow for another eight weeks to determine the residual effects of amendments, after which soil samples were collected for analysis. Treatments included: control, EM, reference fertilizer, reference fertilizer + EM, compost, compost + EM, 1/2 reference fertilizer + compost, 1/2 reference fertilizer + compost + EM, goat manure, goat manure + EM, 1/2 reference fertilizer + goat manure, 1/2 reference fertilizer + goat manure + EM. The yield obtained from the first harvest was higher than the yield obtained from the second harvest, except for the goat manure treatments as a result of initial N removal in the soil (first harvest). Improvement in yield was observed for the second harvest where goat manure had been applied and this was ascribed to improved nutrient availability due to the extended incubation of goat manure in soil. The application of EM alone had a positive but non significant effect on the yields of both the first and second harvests of Swiss chard. However, when applied with compost or goat manure, a non significant negative effect on yield was observed. When applied with inorganic fertilizer, EM had no effect on yield but tended to increase the uptake of nitrogen by Swiss chard. Though goat manure had a narrower C: N ratio than compost, it did not result in greater EM effectiveness as had been hoped. However, goat manure had a more positive effect on soil properties than compost and resulted in higher yields than compost. It increased the N, P, and K contents of the soil and resulted in a narrower C: N ratio of the soil compared to compost. The application of fertilizer alone more than doubled the yields but, more significantly, yields were not compromised when half the recommended fertilizer was applied with either goat manure or compost.

Key words: Compost, EM, goat manure, mineral fertilizer, Swiss chard (Beta vulgaris)

# 4.2 Introduction

Addition of effective microorganisms (EM) to agricultural soils as amendments, especially for disease control and maintenance of healthy resilient soils, has been reported by a number of scholars (Higa & Parr, 1994; Daly & Stewart, 1999). EM refers to a microbial culture of a naturally occurring assortment of beneficial microorganisms such as photosynthetic bacteria, lactic acid bacteria, yeast, actinomycetes and fermenting fungi (Higa & Parr, 1994; Daly & Stewart, 1999) that coexist together. Effective microorganisms are applied as an inoculant to increase the microbial biomass diversity of soils through rapid proliferation of its constituents (Asia-Pacific Natural Network, 1995). The concept of EM and its practical application was developed by Professor Teruo Higa, a horticulturalist at the University of Ryukyus in Okinawa, Japan (Higa, 1994).

EM is widely reported to improve crop quality, growth and yield through effective mineralization of soil organic matter (Piyadasa *et al.*, 1995). In general, EM is applied with a carbon and energy source "molasses" for the micro-organisms (Daly & Stewart, 1999). Integrated use of EM with organic amendments is believed to be an effective technique for enhancing nutrient release and supply from the sources. The mechanism of EM activities for rapid nutrient release from organic amendment involves rapid proliferation of its "effective and beneficial" microorganism content within the soil system.

Some studies have shown that inoculation of soils with EM can improve soil and crop quality (Higa & Parr, 1994; Hussain *et al.*, 1999). Research and field testing of EM has been conducted in the Asia Pacific region (Sangakkara & Higa, 1992; Myint, 1994; Sangakkara, 1994) and in New Zealand (Daly & Stewart, 1999) where its application to onions, peas and sweet corn increased yields by 29%, 31% and 23%, respectively. Xu *et al.* (2000) reported that inoculation of bokash and chicken manure with EM increased photosynthesis and fruit yield of tomato plants. Khaliq, Abbas & Hussain (2006) reported that integrated use of compost with EM resulted in a 44% increase in seed cotton over the control treatment. Similarly, Valarini *et al.* (2002) reported that application of EM with 50 t ha<sup>-1</sup> of animal manure and 30 t ha<sup>-1</sup> of a combination of various green crop residues and weeds separately, increased the production of polysaccharides and alkaline phosphatase and esterase enzymes.

Whilst EM have often shown to be effective in improving plant and soil quality, an earlier study conducted at the University of Fort Hare (Chapter 3 of this dissertation) did not detect a substantial contribution to crop yield by the recommended application of EM in combination with commercial compost. This was attributed to the low quality C constituents in the compost used, which is typical of most matured compost. Composted organic wastes are low in soluble C as most of it is utilized by microorganisms during the composting process (Groenestein & van Faassen., 1996). Therefore mature compost such as the one used in the earlier study may not be able to effectively support proliferation of the decomposer community, including EM.

The objective of this study was to evaluate single and integrated application of EM with fresh and composted organic sources of nutrients on the yield of Swiss chard grown in an Oakleaf soil in pots. The hypothesis tested was that EM has a greater effect on yield of Swiss chard when applied with fresh organic manures than when applied with compost.

## 4.3 Materials and methods

#### 4.3.1 Soil characteristics

The soil used was collected from the A horizon of an Oakleaf form (Oa) soil belonging to the Jozini series (Soil Working Group, 1991) at the University of Fort Hare Research Farm. According to the world soil map compiled by FAO-UNESCO (1988), the soils are Eutric fluvisols (Fle). The soil had a low concentration of total nitrogen, available phosphorus and organic C, but had high levels of micronutrients and exchangeable K, and a pH of 6.1 (Table 4.1).

#### 4.3.2 Goat manure and compost

The goat manure used was collected from the University of Fort Hare Farm and an equivalent of 30 t ha<sup>-1</sup> (which supplied 657, 30, 135 kg ha<sup>-1</sup> of N, P, and K, respectively) was applied. Selected characteristics of the goat manure are shown in Table 4.1.

Nature's Super Grow compost was used for the greenhouse Swiss chard experiment and an equivalent of 30 t ha<sup>-1</sup> (which supplied 60, 15 and 12 kg ha<sup>-1</sup> of N, P, and K, respectively) was applied. Selected properties of Nature's Super Grow are described in section 3.3.3 of this dissertation and Table 4.1 shows some of its characteristics. Both goat manure and the compost materials were analyzed for nutrient concentrations after the experiments were conducted as described in section 4.3.5 of this dissertation.

#### 4.3.3 Treatments and experimental design

The experiment was a randomized complete block design (RCBD) with four replicates. Treatments were : control, EM alone (EM), recommended fertilizer (RF) (N 150: P 90 kg ha<sup>-1</sup>), recommended fertilizer + EM (RF + EM), compost alone (Comp), compost + EM (Comp + EM), 1/2 recommended fertilizer + compost (1/2 RF + Comp), 1/2 recommended fertilizer + EM (1/2 RF + Comp + EM), goat manure alone (GM), goat manure + EM (GM + EM), 1/2 recommended fertilizer + goat manure (1/2 RF + GM) and 1/2 recommended fertilizer + goat manure + EM (1/2 RF + GM + EM).

**Table 4.1** Selected properties of the experimental soil (upper 0-30 cm depth), goatmanure and compost.

Characteristics	Soil	Goat	Nature's Super	
		manure	Grow compost	
pH(1:2.5 soil: water)	6.1	8.0	4.33	
EC ( $\mu$ Scm <sup>-1</sup> )	90.0	2.2	2.37	
Total N (g kg <sup>-1</sup> )	0.6	21.9	2.0	
Available P (g kg <sup>-1</sup> )	0.4	-	-	
Total P (g kg <sup>-1</sup> )	-	1.0	0.5	
Exchangeable K (g kg <sup>-1</sup> )				
Total K (g kg <sup>-1</sup> )	5.0	4.5	0.4	
Organic C (g kg <sup>-1</sup> )	6	-	-	
Total C (g kg <sup>-1</sup> )	9.4	426.3	193.3	
C:N	16.5	19.5	96.65	
C:P	-	426.3	386.6	

The EM products used in this study were obtained from EMROSA (Pty) Ltd and included multiplied - EM, EM - F.P.E, EM 3-in-1 and EM -5. The multiplied - EM was applied as a soil drench and the rest as foliar applied pesticide mixtures. All four brands of EM were

used in EM - treated plots. Multiplied - EM applied as a soil drench was dissolved in water in a ratio of 1: 300 and applied to raise the soil moisture to approximately field capacity seven days before seedlings were transplanted. During the course of the experiment, multiplied - EM solution, in a ratio of 1 : 500, was applied to respective EM - treated pots to maintain the soil moisture at approximately field capacity. Mixtures of EM - FPE, EM 3-in-1 and EM - 5 diluted with water in a ratio of 1 : 800 were sprayed once to control aphids in EM - treated pots, eight weeks after transplanting.

# 4.3.4 Agronomic practices

Swiss chard seeds (cv Lucullus) for raising seedlings to be planted in EM – treated pots were soaked in 0.1 % multiplied - EM for 30 minutes. The other seeds were soaked for 30 minutes in distilled water only prior to sowing in Hygromix seedling mix (marketed by Hygrotech, South Africa) in cavity trays in the greenhouse. After four weeks, the seedlings were transplanted into pre - irrigated pots. Two plants were transplanted in each 30 cm pot, containing 15 kg of soil. After eight weeks of growth, the crop was harvested. It was then allowed to re-grow for another eight weeks before the final harvest. Swiss chard was chosen as a test crop as it is one of the main vegetables consumed in the Eastern Cape region and has relatively few diseases and pests. It also allows more than one harvest of the leaves.

#### 4.3.5 Soil and leaf analysis

Soil and leaf samples were taken at harvest to assess the treatment effects on soil and plant nutrient content. Leaf sampling was done by taking the youngest mature leaves from the top of the plant (Jones *et al.*, 1971). The leaf dry matter was determined after oven drying to constant mass at 65 <sup>0</sup> C. The samples were ground in a hammer mill to pass through a 1 mm mesh sieve. The ground samples were digested with sulphuric acid, selenium powder and salicylic acid mixture for the determination of total P and K (Okalebo, Gathua & Woomer, 2002). Phosphorus was read on a colorimeter following colour development by the molybdenum blue method (Okalebo *et al.*, 2002). Potassium in digested samples was determined by flame photometry. Total nitrogen was determined using a LECO TruSpec C/N auto analyzer (LECO Corporation, 2003).

Soil samples taken after harvest were air dried for 2 weeks and then ground to pass through a 2 mm mesh sieve. Soil pH and electrical conductivity (EC) were determined in water extracts as described by Okalebo *et al.* (2002). Samples were shaken in distilled water in a ratio of 1: 2.5 on a reciprocal shaker for 10 minutes and left standing for 30 minutes, then shaken again for 2 minutes after which pH was read using a WTW pH 526 meter, while EC was read on a WTW 330i conductivity meter. Total-N and C were determined using a LECO TruSpec C/N auto analyzer (LECO Corporation, 2003) and extractable P and K were determined following the Ambic - 2 extraction method (Non-Affiliated Soil Analysis Work Committee, 1990).

The data obtained were subjected to analysis of variance (ANOVA) using the SAS statistical package while means were separated using least significance differences (LSD) at the 0.05 level of significance.

# 4.4 Results

4.4.1 Effects of EM, goat manure, compost and mineral fertilizer on yield of Swiss chard

Application of amendments influenced yield of Swiss chard during the first and second harvests (Table 4.2, Plate 4.1 and Plate 4.2). In general, yield obtained during the second harvest was lower than yield obtained for the first harvest, possibly due to declining soil nutrient content as a result of nutrient removal by the initial growth. The application of EM, compost, goat manure alone or in combination, did not significantly increase yield over the control for the first harvest. However, applying sole EM improved yield by 7.3 % and 11.4 % for the first and second harvests, respectively, relative to the control treatment and an increase of 8.85% in total yield was observed.

An improvement in yield, relative to the control treatment (Table 4.2), was observed for the second harvest where goat manure with or without EM was applied. A 132.5 % increase in yield for the second harvest was observed with goat manure application relative to the control treatment. It is noteworthy that yield obtained from the control and treatments other than goat manure declined for the second harvest (Table 4.2). The application of compost had no effect on yield but its combined application with EM depressed the yield of the first harvest, second harvest and total yield by 7.9 %, 8.7 % and 8.2 %, respectively, relative to the compost treatment.

The application of the reference fertilizer caused a significant increase in yield for both the first and second harvests (Table 4.2). The application of EM with the reference fertilizer had no significant effect on yield relative to the reference fertilizer treatment.

The application of half the reference fertilizer with compost and EM resulted in a yield that was equivalent to that obtained with the application of the reference fertilizer. Applying half the reference fertilizer with compost and EM improved yield of the first and second harvests by 11.9 % and 14.3 %, respectively, relative to half the reference fertilizer with compost treatment, with an overall of 8.3 % increase in the total yield. The combination of half the reference fertilizer with goat manure resulted in a yield that was not statistically different from that obtained with the reference fertilizer during the first and second harvests. Addition of half the reference fertilizer with goat manure and EM had no effect on yield relative to the half reference fertilizer with goat manure treatment.

Treatments	First	Second	Total yield
	harvest	harvest	
	DMY	DMY	DMY
	$(g \text{ pot}^{-1})$	$(g \text{ pot}^{-1})$	(g pot <sup>-1</sup> )
Control	14.3b	7.7e	22.0d
EM	15.4b	8.6e	24.0d
RF	30.0a	14.6bc	44.6a
RF + EM	30.2a	13.1cd	43.3a
Comp	15.2b	8.5e	23.6d
Comp + EM	14.0b	7.8e	21.7d
1/2RF + Comp	27.2a	8.7e	35.9b
1/2RF + Comp + EM	30.4a	9.9e	40.3ab
GM	11.1b	17.9ab	29.0c
GM + EM	10.2b	18.8a	29.0c
1/2RF + GM	27.5a	15.6abc	43.1a
1/2RF + GM + EM	29.0a	12.2cd	41.2a
C.V (%)	18.9	19.7	10.1

**Table 4.2** Effects of integrated use of organic and inorganic sources of nutrients with

 effective microorganisms (EM) on Swiss chard dry matter yield (DMY).

EM; effective microorganisms, RF; reference fertilizer, RF+EM; reference fertilizer + EM, Comp; compost alone, Comp + EM; Compt + effective microorganisms, 1/2RF + Comp; half reference fertilizer + compost , 1/2RF + Comp + EM; half reference fertilizer + compost + effective microorganisms, GM; goat manure alone , GM + EM; goat manure + effective microorganisms, 1/2RF + GM; half reference fertilizer + goat manure, 1/2RF + GM + EM; goat manure + half reference fertilizer + effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at  $P \le 0.05$  according to the LSD test

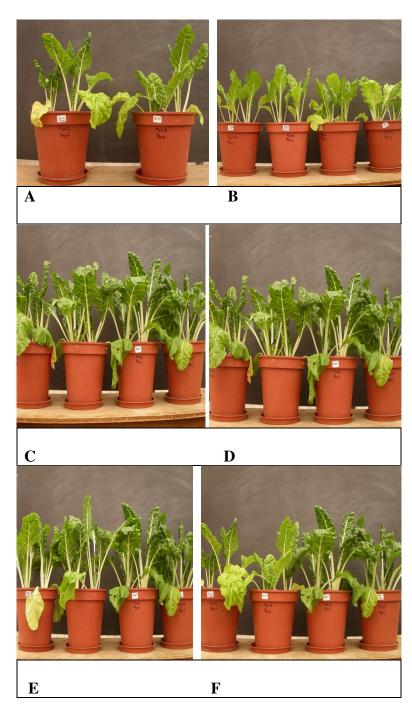


Plate 4.1 Swiss chard plants growing in the greenhouse: A = Control and EM, B = ½ Reference Fertilizer + GM + EM, GM + EM and Comp + EM, C = GM, GM + EM, Reference Fertilizer and Reference Fertilizer + EM, D = ½ Reference Fertilizer + Comp, ½ Reference Fertilizer + GM, Reference Fertilizer and Control, E = ½ Reference Fertilizer + GM, GM, Reference Fertilizer and Control, F = Control, EM, Reference Fertilizer and Reference Fertilizer + EM

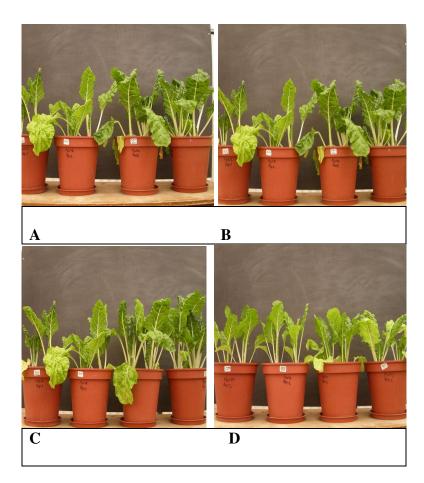


Plate 4.2 Swiss chard plants growing in the greenhouse: A = Control, EM, <sup>1</sup>/<sub>2</sub> Reference
Fertilizer + Comp and Comp, B = Control, EM, <sup>1</sup>/<sub>2</sub> Reference Fertilizer + Comp,
<sup>1</sup>/<sub>2</sub> Reference Fertilizer + Comp + EM, C = Control, EM, GM and Comp, D =
GM, GM + EM, Comp, Comp + EM

4.4.2 Effects of EM, goat manure, compost and mineral fertilizer on leaf nutrient content

There were significant ( $P \le 0.05$ ) treatment effects on leaf N content for both harvests (Table 4.3). Leaf N content ranged from 1.34 g kg<sup>-1</sup> to 3.79 g kg<sup>-1</sup> for the first harvest and ranged from 1.12 to 1.73 g kg<sup>-1</sup> for the second cropping. Leaf N content of the second harvest was lower than leaf N content of the first harvest possibly due to declining soil N content as a result of N removal by the initial leaf growth (first harvest). Application of

EM, compost and goat manure and their combination did not significantly affect the leaf N content of the first harvest although leaf N content in these treatments was lower than that of the control treatment. The application of reference fertilizer significantly increased leaf N content of both the first and the second harvests over the control. Addition of EM with the reference fertilizer resulted in a 25.5 % and 26.3 % increase in leaf N content of the first and second harvests relative to the reference fertilizer treatment. The highest leaf N concentration was observed with the application of the reference fertilizer with EM for both the first and second harvests. A non-significant positive trend was observed with EM application across treatments, except with the compost treatment.

The application of half the reference fertilizer with compost resulted in a significant 18 % increase in leaf N content relative to the control for the first harvest. A non-significant increase of 3.3 % in leaf N content was observed with the application of EM with half the reference fertilizer relative to half the reference fertilizer with compost. For the second harvest, application of half the reference fertilizer with compost resulted in leaf N content that was lower than that of the control treatment although this was not significantly different. Addition of half the reference fertilizer with goat manure resulted in leaf N content that was lower than that of the control for both harvests, with addition of EM making no significant increase.

A similar trend was observed with leaf N uptake with the highest leaf N uptake being observed with reference fertilizer with EM treatment for both harvests. Due to the close relationship between N and protein, a similar trend was observed with crude protein for both harvests.

Leaf P content of the first harvest ranged from 0.06 g kg<sup>-1</sup> to 0.14 g kg<sup>-1</sup> and from 0.10 g kg<sup>-1</sup> to 0.25 g kg<sup>-1</sup> for the second harvest. Leaf P content increased remarkably with the second harvest, changing from a depressed state in the first harvest to a state where significant increases in leaf P were observed. The application of different amendments had no effect on leaf P content in the first harvest but in the second harvest where goat manure was applied, leaf P was significantly (P $\leq$  0.05) higher than the leaf P content of the control treatment (Table 4.3). The application of half the reference fertilizer with compost and half the reference fertilizer with compost and half the reference fertilizer with compost and EM had leaf P content equivalent to that of the control for the second harvest. Application of goat manure without or with EM resulted in leaf P content that was significantly lower than that of the control. Both single and combined applications of EM and amendments had no effect on leaf K content of both harvests.

Treatments N (g kg <sup>1</sup> )		First harvest					Second harvest				
	N uptake (mg pot <sup>-1</sup> )	Crude protein (g kg <sup>-1</sup> )	$\frac{P}{(g kg^{-1})}$	K (g kg <sup>-1</sup> )	N (g kg <sup>-1</sup> )	N uptake (mg pot <sup>-1</sup> )	Crude protein (g kg <sup>-1</sup> )	$\frac{P}{(g kg^{-1})}$	$\frac{K}{(g kg^{-1})}$		
Control	2.34bc	477.8ecd	0.59bc	0.07bc	1.24ab	1.46bac	150.06ba	0.15bc	0.25a	0.89ab	
EM	1.34c	200.0e	0.33c	0.09abc	1.15ab	1.52ba	223.35a	0.08c	0.22ab	0.80b	
RF	3.02ab	876.7ba	0.75ba	0.06c	1.05b	1.37bdc	140.92ba	0.19ba	0.17bc	0.84ab	
RF+ EM	3.79a	1120.2a	0.95a	0.06c	1.04b	1.73a	177.83ba	0.24a	0.21ab	0.90ab	
Comp	1.59c	238.9ed	0.40c	0.08abc	1.15ab	1.26bdc	132.14ba	0.10c	0.24a	0.90ab	
Comp + EM	1.39c	192.5e	0.35c	0.07bc	1.31ab	1.29bdc	152.36ba	0.09c	0.22ab	1.04ab	
1/2RF+ Com	2.76ab	757.0bc	0.69ba	0.06c	1.31ab	1.30bdc	203.14ba	0.17ba	0.25a	0.79b	
1/2RF+Com +EM	2.85ab	728.4bc	0.71ba	0.13ab	1.21ab	1.39bdc	171.67ba	0.18ba	0.25a	0.77b	
GM	1.63c	180.8e	0.41c	0.10abc	1.25ab	1.12d	157.34ba	0.10c	0.10d	1.11a	
GM+EM	1.99bc	199.8e	0.50bc	0.14a	1.27ab	1.33bdc	120.18b	0.12bc	0.11cd	0.91ab	
1/2RF+GM	1.67c	482.3ecd	0.42c	0.07bc	1.21ab	1.26bdc	158.46ba	0.10c	0.19ab	1.03ab	
1/2RF+GM +EM	1.95bc	560.8bcd	0.49bc	0.06c	1.39a	1.23dc	131.27ba	0.12bc	0.21ab	0.91ab	
CV (%)	32.64	47.92	32.64	47.53	18.09	14.21	38.64	32.64	23.02	22.10	

**Table 4.3** Effects of integrated use of organic and inorganic sources of nutrients with effective microorganisms (EM) on leaf Ncontent, N uptake, crude protein, P, and K by Swiss chard plants.

EM; effective microorganisms, RF; reference fertilizer, RF+EM; reference fertilizer + EM, Comp; compost alone, Comp + EM; Compt + effective microorganisms, 1/2RF + Comp; half reference fertilizer + compost + effective microorganisms, GM; goat manure alone , GM + EM; goat manure + effective microorganisms, 1/2RF + GM; half reference fertilizer + goat manure, 1/2RF + GM + EM; goat manure + half reference fertilizer + effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at  $P \le 0.05$  according to the LSD test.

4.4.3 Effects of EM, goat manure, compost and mineral fertilizer on selected soil properties

Post-cropping soil pH was significantly ( $P \le 0.05$ ) affected by different soil amendments (Table 4.4). The application of the reference fertilizer significantly depressed post cropping pH which declined to levels that were moderately acidic (Table 4.4), with the addition of EM resulting in a non-significant slight decrease. A similar trend was observed with the application of half reference fertilizer with compost and half reference fertilizer with compost and half reference fertilizer with compost and EM. The application of EM and sole compost or in combination resulted in post-cropping pH values which were comparable to the control treatment. By contrast, the application of sole goat manure or with half the reference fertilizer, without or with EM, significantly increased post-cropping soil pH relative to the control treatment. The highest post-cropping pH was observed with sole application of goat manure.

Addition of sole EM and sole compost resulted in EC values similar to that of the control. The highest EC value was observed where the reference fertilizer was applied. A similar pattern was observed with application of sole goat manure or in combination with half the reference fertilizer, with or without EM.

There were significant ( $P \le 0.05$ ) treatment effects on residual soil N concentration (Table 4.4) although the values did not differ much from the initial concentration (Table 4.1). Soil N levels associated with the reference fertilizer, half the reference fertilizer with

compost and EM, goat manure, goat manure with EM, half the reference fertilizer with goat manure and half the reference fertilizer with goat manure and EM, were significantly higher than that of the control treatment suggesting that the plants did not exhaust N from these added amendments (Table 4.4). The increase in N observed in the above-mentioned treatments could be attributed to nutrients being slowly released through mineralization from these organic materials. A similar trend was observed with soil C and the greatest amounts of soil N and C were observed in soils treated with goat manure and EM together. The C: N ratio of the soil decreased with application of different amendments but was not statistically different from that of the control. The C: N ratio provides information on the capacity of the soil to store and recycle nutrients (Goyal *et al.*, 1999). The observed decrease in soil C: N ratio, indicated a build - up of the N pool in the soil.

Application of different amendments decreased extractable soil P to below initial levels. Soil P levels associated with sole EM, compost, and their combination, were similar to that of the control and the leaf P concentration of these amendments was lower or equivalent to that of the control. The highest extractable P was observed with the application of reference fertilizer suggesting that the crop had not exhausted the soil P from the fertilizer applied.

Soil residual K in plots treated with goat manure, goat manure with EM and half the reference fertilizer with goat manure, and EM, were significantly higher than that of the control treatment. The highest soil residual K was observed where goat manure was applied with EM, although the manure used had a relatively low concentration of K

(Table 1). According to Bornman *et al.* (1989), kraal manure has, on average, about 2% K, which is far higher than the 0.5 % contained in the goat manure used. General residual soil K in all the amendments exceeded the critical level of 80-120 mg kg<sup>-1</sup> suggested by Bornman *et al.* (1989). Results from this study are consistent with what was reported by Laker (1976), that most South African soils are not deficient in K.

Treatment							
	pH (1:2.5 soil:water)	EC (µs cm <sup>-1</sup> )	$\frac{N}{(g kg^{-1})}$	Total C (g kg <sup>-1</sup> )	C:N	$\frac{P}{(g kg^{-1})}$	$\frac{K}{(g kg^{-1})}$
Control	5.7c	9.7c	0.5e	7.4c	13.2abc	0.4g	2.6d
EM	5.7c	10.6c	0.6cde	7.7c	14.8a	0.4g	3.3dc
RF	5.2e	48.6a	0.8ab	7.8c	14.8a	1.1a	2.8d
RF+EM	5.1e	44.4a	0.6cde	8.1bc	12.7abc	1.0ab	2.0d
Comp	5.5cd	10.1c	0.6cde	8.3bc	13.6abc	0.4g	3.1d
Comp+EM	5.5dc	10.1c	0.5e	7.7c	14.1ab	0.4g	3.0d
1/2RF+Comp	5.2e	28.7b	0.7bcde	8.6bc	11.8bc	0.8dc	2.8d
1/2RF+Comp +EM	5.3de	27.3b	0.8ab	10.2a	11.4c	0.7de	2.0d
GM	6.5a	32.2b	0.8ab	10.3a	12.1bc	0.6ef	5.2abc
GM+EM	6.2b	22.8b	0.9a	10.4a	11.4c	0.5gf	6.3a
1/2RF+GM	6.1b	31.1b	0.7bcde	9.4ab	14.2ba	0.8dc	3.9bcd
1/2RF+GM +EM	6.0b	45.1a	0.8ab	10.0a	13.4abc	0.9bc	5.5ab
CV (%)	3.4	29.3	16.77	11.46	13.2	15.3	40.9

Table 4.4 The effects of integrated use of organic and inorganic sources of nutrients with effective microorganisms (EM) on selected

EM; effective microorganisms, RF; reference fertilizer, RF+EM; reference fertilizer + EM, Comp; compost alone, Comp + EM; Compt + effective microorganisms, 1/2RF + Comp; half reference fertilizer + compost + effective microorganisms, GM; goat manure alone , GM + EM; goat manure + effective microorganisms, 1/2RF + GM; half reference fertilizer + goat manure, 1/2RF + GM + EM; goat manure + half reference fertilizer + effective microorganisms

\*\*Means in each column followed by the same letter are not significantly different from each other at  $P \le 0.05$  according to the LSD test

soil properties after harvest of Swiss chard.

## 4.5 Discussion

4.5.1 Effects of EM, goat manure, compost and mineral fertilizer on yield of Swiss chard

Results obtained in this study show that yield declined during the second cropping and this could be attributed to a decline in nutrient content as a result of removal by the first crop. The removal of nutrient by the first crop is confirmed by higher levels of leaf N content and subsequent N uptake by the first crop compared to the second crop. In terms of P, leaf P content increased during the second cropping and this ruled out the possibility of P limiting plant growth during the second cropping.

The results obtained indicate that the application of EM, compost and goat manure and their combination did not cause a significant increase in yield of the first crop. However, during the second cropping, yield in the goat manure treatment with or without EM increased significantly over the control treatment. The positive effects of incorporating goat manure with or without EM observed during the second cropping suggests that soil production was better maintained under goat manure treatment possibly as a result of nutrients being slowly released over a period of time as suggested by Cooke (1972). This is because nutrients contained in manure are primarily organic and must be mineralized before they can be used by plants. These results are in agreement with those of Tanner and Mugwira (1984) in which application of manures to soil resulted in an increase in nutrient uptake by the second crop rather than the immediate crop. The implication of

these results is that farmers should take measures to ensure that nutrients in organic material become available before plants begin their rapid development.

The apparent depressive effects on yield associated with compost application were also observed with the greenhouse-grown tomato and field-grown butternut experiments. This observation could be ascribed to N immobilization by the soil microorganisms as described in Chapter 3, section 3.9.1.2 of this dissertation. The differences in the C: N ratio of compost and goat manure explains their contrasting results. The highest yield was attained in pots treated with the reference fertilizer during the first cropping possibly due to the immediate release of nutrients from the added reference fertilizer. However, a decline in yield was observed during the second cropping as a result of nutrient removal from the soil by the first crop.

Although application of sole EM did not cause a significant increase in yield, a positive trend was observed relative to the control treatment. The increase in yield resulting from EM application could have been a result of mineralization of nutrients. It is speculated that following application of EM to soil, the effective microorganisms proliferate rapidly enhancing the degradation and chemical breakdown of OM and stimulate mineralization (Higa & Kinjo, 1991; Hussain *et al.*, 1999). Nutrients are then released into the soil-plant system (Daly & Stewart, 1999). However, the positive effects of EM in this case were not pronounced possibly because plants were exploiting a limited soil volume. A positive trend was also observed with the application of EM singularly or across treatments on leaf N, content meaning that EM can contribute meaningfully to crop nutrition.

Application of half the reference fertilizer with either compost or goat manure resulted in yield that was not statistically different to that obtained with the reference fertilizer. These results suggest that it is possible to substitute half the recommended fertilizer with goat manure because of greater nutrient supply to the soil.

# 4.5.2 Effects of EM, goat manure, compost and mineral fertilizer on selected soil properties

The incorporation of goat manure into the soil increased post-cropping soil pH whilst the application of the reference fertilizer decreased post-cropping soil pH. These results indicate that goat manure has a liming effect whereas the mineral fertilizer had acidifying effects. The liming effects of goat manure can be of great value in areas like the Eastern Cape region, South Africa, parts of which have critically low soil pH (Mandiringana *et al.*, 2005). Similar results were obtained by Mhlontlo *et al.* (2007), where the application of sheep kraal manure at rates greater than 2.5 t ha<sup>-1</sup> resulted in higher pH values compared to the control and mineral fertilizer treatments.

The application of compost and goat manure and their combination with EM resulted in an increase in soil N suggesting that the plants did not exhaust N from these amendments. The increase in N observed could possibly be due to the slow release of nutrients through mineralization from these organic materials. The application of goat manure with EM resulted in the highest soil N and C contents. This suggested that EM increased the mineralization of goat manure applied to soil.

# 4.6 Conclusions

The results of this study indicated that, as with the other crops investigated and reported on earlier, EM application had inconsistent effects on Swiss chard yields. The use of goat manure which had a narrower C: N ratio than the compost used in the earlier studies did not result in improved EM effectiveness indicating that the observed ineffectiveness of EM was not related to the quality of the organic material used.

The results of this study, however, demonstrated the benefits of combined application of organic amendments with half the recommended fertilizer over the separate full application of inorganic fertilizer or organic amendment. If adopted, this approach may reduce the variable cost for farmers with a supply of organic amendments as they will purchase only the half of the recommended fertilizer.

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# **CHAPTER 5**

# GENERAL DISCUSSION AND CONCLUSIONS

The use of various brands of EM have been found to improve growth and quality of crops (Daly & Stewart, 1999) through the rapid proliferation of their constituent more beneficial microorganisms and subsequent suppression of soil-borne pathogenic organisms, reducing the incidence of pests and diseases. They also have the ability to effectively mineralize soil organic matter and consequently improve nutrient availability (Piyadasa *et al.*, 1995), which is the mechanism through which EM could benefit soil health and crop growth. The beneficial effects of EM for improving crop production through the promotion of germination, flowering, fruiting and ripening in plants (Asia-Pacific Natural Agriculture Network, 1995) have been widely reported (Sangakkara, undated). However, some trials have not shown consistent beneficial effects of EM on yield (Rukhsana, Arshad, & Nusrat, 1999), suggesting that the results are not reproducible.

Our studies, which were aimed at establishing the usefulness of this product, consisted of four trials involving three popular crops in South Africa, namely tomato, butternut and Swiss chard. The studies encompassed the evaluation of the effects of EM on the growth and yield of tomato and butternut in an Oakleaf soil in the Eastern Cape; and the effect of single and integrated application of EM with inorganic and organic sources of nutrients in an Oakleaf soil on Swiss chard (*Beta vulgaris*) yield. Chapter 5 discusses the findings of these studies and explores research gaps and aspects that need further research.

As described in Chapter 3, the application of EM initially caused a significant increase in the number of fruited tomato plants with the greatest number of fruited plants being observed in the EM + reference fertilizer treatment. The increase in the number of fruited tomato plants in the field could have been as a result of production of plant growth regulators by microorganisms associated with the EM amendment. These results are consistent with what was reported by the Asia-Pacific Natural Agriculture Network (1995), that the application of EM to soils promotes fruiting in plants.

However, the application of EM alone or in combination with compost was ineffective in increasing fruit yield of tomato. The apparent depressive effects of EM observed could have been a result of the severe blight infestation on the tomato crop. Evidence of this phenomenon was provided by measurements of unmarketable yield which comprised mostly of fruits affected by blight. It is, therefore, concluded that EM is an ineffective amendment on its own for tomato production in areas that experience blight.

Although EM depressed tomato yield it improved plant N content of field-grown tomato compared to single application of the amendments. The greatest effect of EM on plant N content was attained with the application of EM alone. It is speculated that after the application of EM into soil, the microorganisms present proliferate rapidly, and stimulate organic matter mineralization followed by subsequent release of more nutrients into the soil-plant system. The apparent increase in leaf N associated with the application of EM observed is in agreement with results obtained by Khaliq *et al.* (2006). These results

indicated that the observed depressive effects of EM on tomato yield were not due to N immobilization but due to blight infestation as stated earlier.

The demonstration that the application of EM can increase tomato leaf N content under field conditions suggests the possibility of beneficially integrating EM to increase productivity under resource poor farming where, or when blight is not prevalent and thereby contributing to improved plant nutrition. The scenario above discussed is expected to result in substantial benefits to resource poor farmers where inputs are limiting.

After observing the failure of EM to control both early and late blight under prevailing conditions, the experiment in Chapter 3, Section 3.5 of this dissertation was designed as a follow up and was conducted under a controlled environment. The experimental design and the soil used were as for the tomato grown under field conditions. The experiment was conducted to specifically explore the effect of EM on tomato yield and leaf nutrient uptake.

Results obtained in this study did not show yield benefits accruing from the application of EM alone or in combination with fertilizers. In actual fact, a negative trend was observed with EM application on leaf dry matter, number of leaves, number of trusses, fruit yield and number of fruit formed. The negative effects of EM could have been due to soil N immobilization that led to reduced crop growth and yield. This assertion is supported by the results on N uptake which was low in plots treated with EM. Further, our experiment

clearly showed that the application of EM in combination with chemical fertilizer resulted in a significant increase in the uptake of N. The results suggested that EM may not lead to optimum nutrient uptake under nutrient-limiting conditions, implying that the use of EM in areas with low fertility may result in low yield due to N immobilization. The effects of EM on nutrient supply in treated soil were further illustrated by low concentrations of nutrients in the soil after harvesting of tomato plants.

The subdued impact of the application of compost with a wide C: N ratio is reflected by the low yields obtained. It is widely acknowledged that the application of organic amendments with a wide C: N ratio to soils induces initial N immobilization. The significant depressive effects of compost on yield observed consistently in these two experiments and on subsequent studies (Chapter 4 of this dissertation) are similar to what was observed by Soumare *et al.* (2002). However, our findings from this experiment also show that the application of compost with the reference fertilizer and EM improved leaf N content and N uptake.

In the case of the butternut trial, the application of EM and its subsequent combination had a consistent depressive effect on total fruit yield, marketable yield and yield termed as first grade. The observed depressive effects of EM on fruit yield of butternut are described in Chapter 3, section 3.9.3 of this dissertation.

Evidence obtained in the greenhouse-grown tomato and field-grown butternut consistently showed the negative effects of EM on the yield of these crops when applied

with compost. We, therefore, investigated in Chapter 4, whether combined application of EM with goat manure which had a narrower C: N ratio could increase yield and enhance soil quality comparable to combined applications of EM with compost. In this case, Swiss chard was used as a test crop. Results presented in this chapter clearly show that yield obtained during the first cropping was higher than yield obtained during the second cropping except for the goat manure treatment with or without EM. The improvement in yield observed with incorporation of goat manure during the second cropping was attributed to improved nutrient availability due to the extended incubation of goat manure in soil. The application of EM alone had a positive but non-significant effect on the yields of both the first and second harvests of Swiss chard. However, when EM was applied with compost or goat manure, a non-significant negative effect on yield was observed. When applied with inorganic fertilizer, EM had no effect on yield but tended to increase the uptake of nitrogen by Swiss chard. Though goat manure had a narrower C: N ratio than compost, it did not result in greater EM effectiveness as had been hoped. However, goat manure had a more positive effect on soil properties than compost and resulted in higher yields than compost. It increased the N, P, and K contents of the soil and resulted in a narrower C: N ratio of the soil compared to compost.

The application of reference fertilizer resulted in a significant increase in yield during the first cropping which declined during the second cropping. The significant increase in yield observed during the first cropping could have been due to the immediate availability of nutrients from the added reference fertilizer. It is widely acknowledged that nutrients in mineral fertilizers are readily available for plant uptake but their

sustainability is low. The application of fertilizer alone more than doubled the yields in both the first and second harvests but, more significantly, yields were not compromised when half the recommended fertilizer was applied with either goat manure or compost. The application of half the reference fertilizer with compost and EM treatment resulted in yield that was equivalent to that obtained with the application of the reference fertilizer. A similar trend was observed with the combined application of half the reference fertilizer with goat manure. These results as suggested in Chapter 4, section 4.5.1 of this dissertation suggest that it is possible to substitute half the recommended fertilizer with organic materials because of greater nutrient supply to the soil.

On the basis of these findings, it is reasonable to suggest that the application of goat manure maintains soil better, with nutrients being slowly released. In addition, it is well established that the application of organic materials like goat manure to soil, improves the soil physical structure with microbial biomass and soil enzyme activities responding to soil management practices as compared to total soil organic matter (Dick, 1992; Doran *et al.*, 1996). A decrease in soil C: N ratio was observed with addition of organic material, indicating build-up of the N pool in the soil.

While some researchers (Daly & Stewart, 1999 ; Xu *et al.*, 2000; Khaliq *et al.*, 2006) have shown the beneficial effects of EM in increasing crop yield, results from our experiments did not show a clear effect, indicating that the effect of EM is inconsistent possibly due to factors such as, (i) fluctuating environmental conditions, (ii) variable conditions of fermentation as each user has to brew his/her own EM from a stock

solution, (iii) differences in practical application of the technology that depends on the resources available, (iv) different packaging and storage environment, and (v) a lack of practical application technology that can suit areas with different soils and weather regimes.

As mentioned earlier on, the use of EM is yet to be widespread in South Africa, although some commercial farmers are already using the product and are finding merits in its use. A participatory study with interested farmers is recommended to (i) investigate suitable local methods for brewing different brands of EM in South Africa for both small scale and large scale farmers, (ii) identify ideal conditions for EM storage, (iii) determine application rates that are suitable for different crops, and (iv) determine ideal application time and frequency of application of EM. In addition to the participatory study, more intensive and systematic on-farm trials among the commercial farmers in South Africa are required to provide a better understanding of the usefulness of EM in increasing crop and soil quality. Also the cost-to-benefit implication of such practice should be ascertained.

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# APPENDICES

**Appendix 1**: Number of flowered plants, five weeks after transplanting of tomato in the field.

#### ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	4629629.62	925925.92	0.35	0.8765
Treatment	7	12111111.08	1730158.73	0.66	0.7033
Error	35	91666666.40	2619047.6		
Total	47	108407407.10			
-			2619047.6		

Grand mea	n = 500	Grand sum = 577 333.30
CV =	13.46 %	LSD(0.05) = 2187

**Appendix 2**: Number of fruited plants, seven weeks after transplanting of tomato in the field.

Source	Degree of freedom	Sum of square	Mean of square	F value	Probability
Replication	5	1025.19	205.04	0.68	0.64
Treatment	7	5739.40	819.91	2.72	0.02
Error	34	10247.82	301.41		
Total	46	17017.12			

Grand mean	= 138.90	Grand sum =	198 666.70
CV =	49.11 %	LSD(0.05) =	23.76

Appendix 3: Total fruit yield of field grown tomato

## **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	171.05	34.21	0.60	0.70
		2172 (7	210.20	5.42	0.0002
Treatment	/	2172.67	310.38	5.42	0.0003
Error	35	2005.86	57.31		
Total	47	4349.59			

Grand me	an = 0.96	Grand sum = 1217.173
CV =	29.90 %	LSD(0.05) = 10.23

Appendix 4: Marketable yield of field grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	248.99	49.80	2.04	0.0972
The second second	7	(22.05	00.00	2 ( 1	0.0047
Treatment	/	622.05	88.86	3.64	0.0047
Error	35	855.05	24.43		
Total	47	1726.10			

Grand mean	= 0.46	Grand sum = 563.33
CV =	42.11 %	LSD (0.05) =6.68

Appendix 5: Unmarketable yield of field grown tomato

## **ANOVA Table**

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	151.7966491	30.3593298	0.79	0.5613
-					
Treatment	7	514.9142510	73.5591787	1.92	0.0951
Error	35	1338.115832	38.231881		
Total	47	2004.826732			

Grand mean $= 0.50$	Grand sum = 668.29
CV = 44.41 %	LSD(0.05) = 8.35

Appendix 6: Leaf N content of field grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	15.88783630	3.17756726	5.31	0.0010
Treatment	7	11.83063097	1.69009014	2.82	0.0193
Error	35	20.96071961	0.59887770		
Total	47	48.67918688			

Grand me	an = 4.40	Grand sum = $211.30$
CV =	17.18%	LSD(0.05) = 0.907

Appendix 7: Leaf P content of field grown tomato

#### **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	0.01283044	0.00256609	5.83	0.0005
-					
Treatment	7	0.00426011	0.00060859	1.38	0.2436
Error	35	0.01541242	0.00044035		
Total	47	0.03250298			

Grand me	ean = 0.07	Grand sum = $3.41$
CV =	28.03 %	LSD(0.05) = 0.0246

Appendix 8: Leaf K content of field grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	0.68056893	0.13611379	0.48	0.7889
-					
Treatment	7	1.69417227	0.24202461	0.85	0.5522
Error	35	9.92991241	0.28371178		
Total	47	12.30465360			

Grand me	an = 2.29	Grand sum = 110.13
CV =	23.22 %	LSD(0.05) = 0.62

Appendix 9: Soil nitrogen after harvest of field grown tomato

## **ANOVA Table**

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	0.13950950	0.02790190	0.94	0.4678
Treatment	7	1.02701900	0.14671700	4.94	0.0006
Error	35	1.03971150	0.02970604		
Total	47	2.20624000			

Grand mear	n = 0.83	Grand sum = 39.77
CV =	20.80 %	LSD(0.05) = 0.202

Appendix 10: Soil phosphorus after harvest of field grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	0.00646903	0.00129381	1.17	0.3434
-					
Treatment	7	0.07187531	0.01026790	9.29	<.0001
Error	35	0.03870162	0.00110576		
Total	47	0.11704596			

Grand mear	n = 1.00	Grand sum =48.21
CV =	33.11	LSD(0.05) = 0.04

Appendix 11: Soil potassium after harvest of field grown tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	5	5.39857052	1.07971410	0.53	0.7522
Treatment	7	27.62592970	3.94656139	1.94	0.0931
Error	35	71.3415322	2.0383295		
Total	47	104.3660325			

Grand mean	n = 4.82	Grand sum = $0.10$
CV =	33.36	LSD(0.05) = 1.67

Appendix 12: Stem dry matter yield of glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	136.2846604	15.1427400	0.36	0.9476
_					
Treatment	7	142.7659491	20.3951356	0.48	0.8408
Error	39	1645.885575	42.202194		
Total	55	1936.220484			

Grand mean	1 = 23.40	Grand sum = 1123.29
CV =	27.8 %	LSD(0.05) = 8.33

Appendix 13: Leaf dry matter yield of glasshouse grown tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	322.510023	35.834447	1.24	0.2974
Treatment	7	3074.449143	439.207020	15.25	<.0001
Error	39	1122.915663	28.792709		
Total	55	4685.542743			

Grand mean	n = 26.81	Grand sum = 1447.89
CV =	20.01 %	LSD(0.05) = 6.879

Appendix 14: Total dry matter yield of glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	596.579576	66.286620	0.94	0.5056
-					
Treatment	7	3294.237068	470.605295	6.65	<.0001
Error	39	2761.759117	70.814336		
Total	55	6759.710998			

Grand mea	n = 50.21	Grand sum = 2812.03
CV =	16.76 %	LSD(0.05) = 10.79

Appendix 15: Total number of leaves produced in glasshouse tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	508.644939	56.516104	1.65	0.1221
Treatment	7	5268.696130	752.670876	21.91	<.0001
Error	62	2129.543949	34.347483		
Total	78	7867.772152			

Grand mea	n = 26.66	Grand sum = 2132.66
CV =	21.98 %	LSD(0.05) = 6.113

Appendix 16: Total number of trusses formed in the glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	55.6727851	6.1858650	1.06	0.4012
-					
Treatment	7	847.0128645	121.0018378	20.83	<.0001
Error	62	360.227215	5.810116		
Total	78	1274.354430			

Grand mea	an = 11.63	Grand sum = $930.63$
CV =	20.72 %	LSD(0.05) = 2.514

Appendix 17: Total fruit yield of glasshouse grown tomato

#### ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	98191.6946	10910.1883	1.40	0.2085
Treatment	7	700618.4324	100088.3475	12.83	<.0001
Error	62	483756.828	7802.529		
Total	78	1293213.823			

Grand mean =	= 483.06	Grand sum = 38644.84
CV =	18.29 %	LSD(0.05) = 92.13

Appendix 18: Total number of fruit formed in the glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	49.959156	5.551017	0.72	0.6912
-					
Treatment	7	2204.543283	314.934755	40.68	<.0001
Error	62	479.940844	7.740981		
Total	78	2765.341772			

Grand mea	n = 13.78	Grand sum = $1102.79$
CV =	20.18 %	LSD(0.05) = 2.902

Appendix 19: Average fruit weight of glasshouse grown tomato

## **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	778.181831	86.464648	0.73	0.6780
Treatment	7	8067.677960	1152.525423	9.76	<.0001
Error	62	7323.04437	118.11362		
Total	78	16291.86299			

Grand mean	n = 40.02	Grand sum = 3202.22
CV =	27.15 %	LSD(0.05) = 11.34

Appendix 20: Fruit N content of glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	2	1.31035299	0.65517649	0.48	0.6279
Treatment	7	10.70867172	1.52981025	1.12	0.4019
Error	14	19.05977511	1.36141251		
Total	23	31.07879982			

Grand mean $= 1.66$		Grand sum = $132.54$	
CV =	70.42 %	LSD(0.05) = 2.0433	

Appendix 21: Fruit P content of glasshouse grown tomato

## **ANOVA Table**

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	2	0.01310353	0.00655176	0.48	0.6279
Treatment	7	0.10708672	0.01529810	1.12	0.4019
Error	14	0.19059775	0.01361413		
Total	23	0.31078800			

Grand mean = $0.166$		Grand sum = $13.25$
CV =	70.42 %	LSD(0.05) = 0.2043

Appendix 22: Fruit K content of glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	2	2.97467401	1.48733700	10.96	0.0014
Treatment	7	1.99635908	0.28519415	2.10	0.1123
Error	14	1.90041533	0.13574395		
Total	23	6.87144842			

Grand mean =	= 2.93	Grand sum = $234.78$
CV =	12.55 %	LSD(0.05) = 0.6452

Appendix 23: Leaf N content of glasshouse grown tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability	
	freedom	square	square			
Replication	9	2.01087701	0.22343078	2.20	0.0436	
_						
Treatment	7	1.36499915	0.19499988	1.92	0.0930	
Error	39	3.96775328	0.10173726			
Total	55	7.28031470				
Grand mean =	= 1.06		Grand sum =	84.45		
CV =	30.21 %		LSD(0.05) =	0.4089		

Appendix 24: Leaf P content of glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	0.00983316	0.00109257	1.18	0.3342
_					
Treatment	7	0.07703525	0.01100504	11.89	<.0001
Error	39	0.03609347	0.00092547		
Total	55	0.12610957			

Grand mean :	= 0.20	Grand sum = $15.63$
CV =	15.57 %	LSD(0.05) = 0.03900

Appendix 25: Leaf K content of glasshouse grown tomato

## **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	0.91353442	0.10150382	0.96	0.4859
Treatment	7	1.44751913	0.20678845	1.96	0.0862
Error	39	4.11879694	0.10561018		
Total	55	6.58441240			

Grand mean =	= 1.73	Grand sum = $138.36$
CV =	18.79 %	LSD(0.05) = 0.4166

Appendix 26: Leaf N uptake by the glasshouse grown tomato plants

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	3674.921560	408.324618	1.97	0.0697
Treatment	7	9320.531099	1331.504443	6.43	<.0001
Error	39	8077.42206	207.11339		
Total	55	21458.81766			

Grand mean =	= 52.27	Grand sum = $4181.52$
CV =	27.53 %	LSD(0.05) = 18.45

Appendix 27: P uptake by the glasshouse grown tomato plants

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	28.26806670	3.14089630	1.13	0.3634
Treatment	7	78.64499219	11.23499888	4.05	0.0020
Error	39	108.0622149	2.7708260		
Total	55	234.1711055			

Grand mean	= 9.50	Grand sum = 759.82
CV =	17.53 %	LSD(0.05) = 2.134

Appendix 28: K uptake by the glasshouse grown tomato plants

Source	Degree of	Sum of		F value	Probability
	freedom	square	square		
Replication	9	1353.01011	150.33446	0.38	0.9359
_					
Treatment	7	12909.88704	1844.26958	4.71	0.0007
Error	39	15284.62076	391.91335		
Total	55	29679.00075			

Grand mean	= 86.03	Grand sum = 6882.53
CV =	23.01%	LSD(0.05) = 25.38

Appendix 29: Soil N after harvesting of the glasshouse grown tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	0.54294206	0.06032690	1.81	0.0848
Treatment	7	0.05779816	0.00825688	0.25	0.9711
Error	61	2.03425374	0.03334842		
Total	77	2.62770111			

Grand mean	= 0.35	Grand sum = $28.07$
CV =	52.04 %	LSD(0.05) = 0.1918

Appendix 30: Soil P after harvesting of the glasshouse grown tomato

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	0.00236456	0.00026273	0.49	0.8762
Treatment	7	0.09577269	0.01368181	25.49	<.0001
Error	61	0.03274830	0.00053686		
Total	77	0.13165844			

Grand mean	n = 0.13	Grand sum = $10.00$
CV =	18.53 %	LSD(0.05) = 0.02434

Appendix 31: Soil K after harvesting of the glasshouse grown tomato

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	9	4.25203545	0.47244838	5.59	<.0001
-					
Treatment	7	5.99197020	0.85599574	10.13	<.0001
Error	61	5.15525197	0.08451233		
Total	77	15.39388668			

Grand mean	= 1.92	Grand sum = $153.90$
CV =	15.11 %	LSD(0.05) = 0.3054

Appendix 32: Total fruit yield of field grown butternut

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	378.223889	126.074630	1.81	0.1759
_					
Treatment	7	1461.159444	208.737063	3.00	0.0239
Error	21	1461.211667	69.581508		
Total	31	3300.595000			

Grand mean = $25.70$		Grand sum = 796.57
CV =	32.46 %	LSD(0.05) = 22.3000000

Appendix 33: Marketable yield of field grown butternut

## **ANOVA Table**

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	353.264445	117.754815	1.85	0.1697
-					
Treatment	7	1717.891111	245.413016	3.85	0.0076
Error	21	1339.028889	63.763280		
Total	31	3410.184445			

Grand mean	n = 21.7	Grand sum = $672.7$
CV =	37.04 %	LSD(0.05) = 16.4000000

Appendix 34: First grade yield of field grown butternut

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	316.121666	105.373889	1.64	0.2100
Treatment	7	1236.608333	176.658333	2.75	0.0340
Error	21	1347.918333	64.186587		
Total	31	2900.648333			

Grand mean $= 14.63$		Grand sum = $453.50$
CV =	53.4 %	LSD(0.05) = 11.33333333

Appendix 35: Leaf N content of field grown butternut

# ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.66942869	0.22314290	7.37	0.0016
•					
Treatment	7	0.12132383	0.01733198	0.57	0.7699
Error	20	0.60576804	0.03028840		
Total					

Grand mean	= 1.59	Grand sum = $49.17$
CV =	10.97 %	LSD(0.05) = 0.262

Appendix 36: Leaf P content of field grown butternut

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00348213	0.00116071	2.90	0.0601
-					
Treatment	7	0.00254052	0.00036293	0.91	0.5199
Error	20	0.00799233	0.00039962		
Total	30	0.01393498			

Grand mea	an = 0.15	Grand sum = $4.67$
CV =	13.28 %	LSD(0.05) = 0.0301

Appendix 37: Leaf K content of field grown butternut

#### **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	1.27152972	0.42384324	6.37	0.0033
Treatment	7	0.62574275	0.08939182	1.34	0.2816
Error	20	1.32975139	0.06648757		
Total	30	3.19391766			

Grand mean $= 1.90$		Grand sum = $59.07$	
CV =	13.53%	LSD(0.05) = 0.39	

Appendix 38: Soil N content after harvest of the butternut crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.09132021	0.03044007	3.21	0.0449
Treatment	7	0.07140819	0.01020117	1.08	0.4137
Error	20	0.18948388	0.00947419		
Total	30	0.35301942			

Grand mean	= 0.32	Grand sum = $9.85$
CV =	30.63 %	LSD(0.05) = 0.15

Appendix 39: Soil P content after harvest of the butternut crop

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00374581	0.00124860	5.42	0.0068
Treatment	7	0.00634207	0.00090601	3.93	0.0075
Error	20	0.00461114	0.00023056		
Total	30	0.01447279			

Grand mean :	= 0.07	Grand sum = $2.23$
CV =	21.09	LSD(0.05) = 0.02

Appendix 40: Soil K content after harvest of the butternut crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	2.75158984	0.91719661	2.85	0.0633
Treatment	7	2.63442877	0.37634697	1.17	0.3631
Error	20	6.43808742	0.32190437		
Total	30	11.43432756			

Grand mean $= 3.81$		Grand sum = $118.05$
CV =	14.89913	LSD(0.05) = 0.8541

Appendix 41: Dry matter yield of Swiss chard first crop

## **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	15.806225	5.268742	0.33	0.8038
Treatment	11	3078.550425	279.868220	17.52	<.0001
Error	33	527.265675	15.977748		
Total	47	3621.622325			

Grand mean = $21.20$		Grand sum = 10.17.54	
CV =	18.8 %	LSD(0.05) = 3.32	

Appendix 42: Dry matter yield of Swiss chard second crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	10.7055729	3.5685243	0.64	0.5934
Treatment	11	708.7735229	64.4339566	11.59	<.0001
Error	33	183.4117021	5.5579304		
Total	47	902.8907979			

Grand mean :	= 11.93	Grand sum = $573.05$
CV =	19.75 %	LSD(0.05) = 1.96

Appendix 43: Total dry matter of Swiss chard crop

# ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	15.171323	5.057108	0.45	0.7176
Treatment	11	3675.021823	334.092893	29.86	<.0001
Error	33	369.171402	11.187012		
Total	47	4059.364548			

Grand mean	= 33.14	Grand sum = 1590.59
CV =	10.09 %	LSD(0.05) = 2.78

Appendix 44: Swiss chard N content after harvest for the first crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.19417494	0.06472498	1.75	0.1788
Treatment	11	1.09810690	0.09982790	2.69	0.0154
Error	30	1.11221903	0.03707397		
Total	44	2.39671819			

Grand me	<b>an</b> = 2.15	<b>Grand sum =</b> 103.22
CV =	32.64 %	LSD(0.05) = 0.29

Appendix 45: Leaf N uptake by Swiss chard first crop

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	6626.685	2208.895	0.04	0.9888
Treatment	11	4185006.765	380455.160	7.03	<.0001
Error	30	1623889.962	54129.665		
Total	44	5826589.632			

Grand mean =	485.51	Grand sum = 23304.45
CV =	47.92 %	<b>LSD (0.05) =</b> 354.16

Appendix 46: Swiss chard N content after harvest for the second crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.19417494	0.06472498	1.75	0.1788
Treatment	11	1.09810690	0.09982790	2.69	0.0154
Error	30	1.11221903	0.03707397		
Total	44	2.39671819			

<b>Grand mean =</b> 1.35		<b>Grand sum =</b> 65.02
CV =	14.21399	<b>LSD (0.05) =</b> 0.2931

Appendix 47: Leaf N uptake by Swiss chard second crop

# ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	5090.42718	1696.80906	0.45	0.7172
Treatment	11	38799.39530	3527.21775	0.94	0.5166
Error	30	112415.6013	3747.1867		
Total	44	155439.5530			

Grand mean =	= 158.41	<b>Grand sum =</b> 7603.62
CV =	38.64 %	LSD(0.05) = 93.182

Appendix 48: leaf P content after harvest for the first crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00119621	0.00039874	0.25	0.8598
Treatment	11	0.03052753	0.00277523	1.75	0.1054
Error	33	0.05236581	0.00158684		
Total	47	0.08408955			

Grand mean = 0.08		<b>Grand sum =</b> 4.02
CV =	47.54 %	LSD (0.05) = 0.0573

Appendix 49: Swiss chard P content of the second crop

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00766110	0.00255370	1.20	0.3263
Treatment	11	0.11909663	0.01082697	5.07	0.0001
Error	33	0.07044250	0.00213462		
Total					

Grand mean =	0.20	<b>Grand sum =</b> 9.63
CV =	23.03 %	LSD(0.05) = 0.0665

Appendix 50: Swiss chard K content of the first crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.35657354	0.11885785	2.46	0.0804
-					
Treatment	11	0.48262800	0.04387527	0.91	0.5447
Error	33	1.59696765	0.04839296		
Total	47	2.43616919			

<b>Grand mean =</b> 1.22		<b>Grand sum = 58.36</b>	
CV =	18.09 %	LSD(0.05) = 0.3165	

Appendix 51: Swiss chard K content of the second crop

## ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.78789722	0.26263241	6.55	0.0013
Treatment	11	0.48654826	0.04423166	1.10	0.3893
Error	33	1.32313629	0.04009504		
Total	47	2.59758178			

Grand me	<b>an =</b> 0.91	<b>Grand sum =</b> 43.50
$\mathbf{CV} =$	22.10 %	LSD(0.05) = 0.2881

Appendix 52: Swiss chard crude protein content of the first crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.01409847	0.00469949	0.15	0.9272
Treatment	11	1.52946035	0.13904185	4.52	0.0005
Error	30	0.92384991	0.03079500		
Total	44	2.46475975			

Grand mean :	= 0.54	<b>Grand sum =</b> 25.80
CV =	32.64266	LSD(0.05) = 0.2671

Appendix 53: Swiss chard crude protein content of the second crop

# ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00088115	0.00029372	0.15	0.9272
Treatment	11	0.09559127	0.00869012	4.	0.000552
Error	30	0.05774062	0.00192469		
Total	44	0.15404748			

Grand mea	n = 0.13	<b>Grand sum =</b> 6.45
CV =	32.64266	LSD(0.05) = 0.0668

Appendix 54: Soil N content after harvest of Swiss chard crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00162657	0.00054219	4.00	0.0155
-					
Treatment	11	0.00681258	0.00061933	4.57	0.0003
Error	33	0.00447046	0.00013547		
Total	47	0.01290961			

Grand mean = 0.07		<b>Grand sum =</b> 3.33	
CV =	16.77 %	LSD(0.05) = 0.02	

Appendix 55: Soil P content after harvest of Swiss chard crop

# ANOVA Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.00054643	0.00018214	1.73	0.1794
Treatment	11	0.02733500	0.00248500	23.63	<.0001
Error	33	0.00346966	0.00010514		
Total	47	0.03135108			

Grand mean = 0.07		<b>Grand sum =</b> 3.21
CV =	15.33 %	LSD(0.05) = 0.01

Appendix 56: Soil K content after harvest of Swiss chard crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.01600108	0.00533369	0.26	0.8559
-					
Treatment	11	0.88771185	0.08070108	3.89	0.0012
Error	33	0.68526346	0.02076556		
Total	47	1.58897639			

Grand mean =	0.35	<b>Grand sum =</b> 16.90
CV =	40.92237	LSD (0.05) = 0.2073

Appendix 57: Soil C content after harvest of Swiss chard crop

# **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.02316350	0.00772117	0.75	0.5294
-					
Treatment	11	0.62960717	0.05723702	5.57	<.0001
Error	33	0.33909600	0.01027564		
Total	47	0.99186667			

Grand mean	= 0.88	<b>Grand sum =</b> 42.45
CV =	11.46 %	LSD(0.05) = 0.0842

Appendix 58: C: N of the soil after harvest of Swiss chard crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	60.06784050	20.02261350	6.63	0.0013
_					
Treatment	11	67.52616392	6.13874217	2.03	0.0572
Error	33	99.7305345	3.0221374		
Total	47	227.3245390			

Grand mean =	= 13.12	<b>Grand sum =</b> 629.96
CV =	13.25 %	LSD(0.05) = 2.5009

Appendix 59: Soil pH after harvest of Swiss chard crop

## **ANOVA** Table

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	0.08910833	0.02970278	0.79	0.5072
Treatment	11	8.25137500	0.75012500	19.99	<.0001
Error	33	1.23804167	0.03751641		
Total	47	9.57852500			

Grand mean =	5.67	<b>Grand sum =</b> 272.1
CV =	3.41 %	LSD(0.05) = 0.2786

Appendix 60: Soil Ec after harvest of Swiss chard crop

Source	Degree of	Sum of	Mean of	F value	Probability
	freedom	square	square		
Replication	3	255.854850	85.284950	1.39	0.2625
Treatment		9190.145600	835.467782	13.64	<.0001
	11				
Error	33	2021.75585	61.26533		
Total	47	11467.75630			

Grand mean	= 26.71	<b>Grand sum =</b> 1281.96	
CV =	29.30719	LSD(0.05) = 11.26	