Review of Literature

CHAPTER - II

REVIEW OF LITERATURE

The literature pertaining to the role of EM (Effective Microorganisms) in converting the solid waste in to an eco-friendly value added organic manure and the influence of composted solid waste on the biometric, yield and biochemical parameters of various plants as relevant to the present study is reviewed and presented in this chapter.

2.1 SOLID WASTE - COMPOSITION

The solid waste consists of paper, garden trimmings, yard waste, organic fractions (vegetable, fruit and food waste), jute, wood pieces, cloth, plastic covers, rubber etc.

2.1.1 Physico - Chemical Analysis of MSW

Jeyapriya and Saseetharan (2007) studied the amount and the physico-chemical analysis of Municipal Solid Waste in Coimbatore city. The results of the study showed that the biodegradable fraction of MSW was high with an average value of 68.94 per cent. High percentage of volatile solid content also revealed that the organic content in the waste material was high. It was also concluded that the solid waste had an optimum C: N ratio of 30: 1.

Jeyapriya and Saseetharan (2007) also analysed the biochemical parameters of MSW biocompost. The MSW biocompost found to contain moisture content - 27.0 per cent ; pH - 7.66 ; EC - 1.13 dSm^{-1} ; Total nitrogen - 1.34 per cent ; Phosphorus - 0.58 per cent ; Potassium - 1.12 per cent ; OC - 24.28 per cent ; C : N - 19 : 1 ; Mg - 1.33 per cent ;

Calcium - 2.36 per cent ; Bacteria - 58 (10^6 C 7 U g^{-1} dry soil) ; Fungi - 28 (10^6 C 7 U g^{-1} dry soil) and Actinomycetes - 22 (10^6 C 7 U g^{-1} dry soil).

2.1.2 pH and Temperature Profile in EM-SW Compost

pH, potential of hydrogen expresses the concentration of hydrogen ions (H^+) in a given solution. It is the negative logarithm of hydrogen ion concentration. The optimal pH values for composting range from pH 5.5 to 8.0. Bacteria favor a near - neutral pH, whereas fungi favor an acidic pH range.

Fugorty and Tuovinen (1991),during their study on Microbiological degradation of pesticides in yard waste composting observed that pH of the yard waste compost ranged between 5.8 and 7.2. The effects of extreme pH on the composting process were directly related to the effect of pH on microbial activity or more specifically, on microbial enzymes. Poincelot (1974) reported that the pH buffering capacity increased as a result of humus formation.

Temperature profile is a good indicator of the heat generation by the microbial metabolism and biochemical activity during the composting process.

According to Fogarty and Tuovinen (1991), temperature was the most contentious of all the parameters controlling the rate of composting. Ultimately, the composting process was determined by the temperature profile. Changes in temperature were commonly used as a measure of microbiological activity underlying the composting process. Thus, the temperature profile of composting could be used to determine the stability of the organic material. They also reported that the key period of the composting process was based predominantly on microorganisms which grow in the temperature range from 25 to 60 °C i.e. mesophilic (<45 °C) and moderately thermophilic (40 to 60 °C) organisms. Elevated temperatures (> 60 °C) lead to increasingly rapid thermal inactivation of mesophilic microorganisms.

Nakasaki *et al.* (1985) during their study on effect of temperature on composting of sewage sludge found that the optimum temperature for microbial activity was below 60°C.In the experimental study of compost made from shredded paper and food scraps, Strom (1985) found that only few bacterial species remained active at temperatures above 60°C, those that survived were predominantly *Bacillus* spp. Fungi were found only in the narrow temperature interval from 55-61°C.Maintaining high temperature is necessary for rapid composting as well as to destroy weed seeds, insect larvae and potential plant or human pathogens that may be present in the composting material.

2.1.3 Solid Waste as Organic Manure

During composting, the organic carbon is lost as CO_2 and total nitrogen increases as a result of carbon loss. The final nitrogen content of compost is dependent on the initial nitrogen present in the waste and the extent of decomposition (Crawford, 1983). According to Mphangewe and Erkwealor (1990), the yield advantages of the organic manure over inorganic fertilizers were ascribed to the portable effects of the organic manure in improving the physical characteristics of soil.

Hodges (1991) opined that the composted manure can be an alternative source of fertilizer in organic farming where the use of manufactured chemicals is prohibited. Once the process of decomposition begins, the applied nitrogen may be utilized efficiently by growing plants due to the favourable effects of greater organic matter in the soil (Yadav and Prasad, 1992).

Composting is seen as an environmentally acceptable method of waste treatment. It is the aerobic, biological process, which uses naturally occurring microorganisms, to convert biodegradable organic matter into humus - like product (Georgacakis *et al.,* 1996). The process destroys pathogens, converts N from unstable ammonia to stable organic forms, reduces the volume of waste and improves the nature of the waste (Georgacakis *et al.,* 1996 and Sequi, 1996). Samarta and Patro (1996) established that organic farming is the backbone of sustainable agriculture. Organic farming improves the soil health and the crops grown in rich organic manure, resist pest and disease attack. It is an ecologically sound and sustainable way of growing more food.

Composting is the art and science of combining available organic waste so that they decompose to form uniform and stable finished product. Microorganisms that do much of the work need high temperature, plenty of oxygen and moisture (Veeresh, 1999).

Alam and Khan (2001) observed that the application of organic matter is essential to maintain both soil fertility, soil structure and to stimulate extensive biological activity. Raw organic waste like garbage trashes, coir pith and such other waste of very wide carbon, nitrogen ratio must be composted and added to the soil to draw desired benefits from them (Kumarasamy, 2001).

Composts provide mineral nutrients, micronutrients, useful microbes and water retaining humus to soil. This improves the quality, pest resistance, drought resistance and decreases the irrigation water requirements for the crops. Compost can find good market if properly promoted and made conveniently available to the farming community (Channabasavanna and Rahman, 2002).

According to the definition of FAO, organic farming should involve successful management of resources for agriculture to satisfy changing human needs while maintaining or enhancing the quality of the environment and consuming natural resources (Krishnaveni and Balamurugan, 2002). Zamanov *et al.* (2002) found that the organic fertilizers maintain the humus content in soil and they are the source of carbon for the recycling of humus loses, they improve the physical properties of soil and promote the mineralization of organic matter. The organic manures are mostly of plant origin and are utilized either directly or after having been used once as food by animals and humans (Basavanneppa and Biradar, 2003).

2.1.4 Solid Waste and Yield

Convertini *et al.*, (1999) reported that the addition of MSW compost had a positive influence on crop production and yield. Field studies were conducted to determine the effect of maize straw (MS) compost with and without mixing with inorganic nitrogen fertilizer (N min) on the growth, productivity and nutrient uptake of tomato (*Lycopersicum esculentum* Mill.). The treatments were 4 and 6 M t ha^{-1} of MSW compost and 30 and 60 kg ha^{-1} of N min fertilizer. Controls with no compost (0 M t ha^{-1}) was also included. Applications of MSW compost and N min enhanced plant growth and were more effective than the control treatment. Plant height, number of flowers and fruits produced per plant, plant dry matter and yield were all improved significantly by application of MSW compost alone or in combination with N min. Yields and yield components indicated that the crop had a better response to application of compost, but less to N min. The combined application of 4 M t ha^{-1} of

MSW compost and N min fertilizer at 30 kg ha^{-1} produced better plant growth and fruit yield than other combinations.

Dufa (2000) through his investigation on the effect of urban domestic refuse compost on lettuce growth found that the compost promoted the growth of lettuce. Application of organic manure to the soil improved the physical, chemical and biological properties with direct impact on moisture retention, root growth and nutrient conservation etc (Krishnakumar and Jawahar, 2001). Quang *et al.* (2001) studied the effect of mixed organic fertilizer on the yield and quality of sweet pepper. They obtained increased yield by 43.3 to 109.8 per cent. The mixed organic fertilizers optimized soil microorganisms, increased the activity of enzymes in the soil and improved soil fertility. Stoffella and Li (2001) in their investigation of organic waste compost utilization, on vegetable crop production found that the compost acted as a soil amendment and increased the yield in vegetable crops. They used biodegradable material such as yard trimmings, food waste and biosolids for composting.

Jeevan (2005) reported that the rural waste like crop residues of rice, wheat, sorghum, pearl millet, maize, sugarcane etc., have the potential to contribute nearly 1.68, 2.02 and 5.04 million tons of N, P_2O_5 and K_2O respectively. Agricultural soils of two Italian maize farms were treated with solid waste compost, cattle manure and usual minerals. The effects were studied from the harvested material. The pH, EC and NPK were purely higher in solid waste compost than the other treatments applied to the soil (Guerini *et al.*, 2006).

2.2 EFFECTIVE MICROORGNAISMS (EM)

According to Reganold *et al.* (1990) and Parr and Hornick (1992), beneficial microorganisms are those that can fix atmospheric nitrogen, decompose organic waste and residues, detoxify pesticides, suppress plant diseases and soil-borne pathogens, enhance nutrient cycling and produce bioactive compounds such as vitamins, hormones and enzymes that stimulate plant growth.

The concept of Effective Microorganisms (EM) was developed by Higa (1991) and Higa and Wididana (1991). They reported that the concept of inoculating soils and plants with beneficial microorganisms was to create a more favourable microbiological environment for plant growth. They also found that microorganisms can coexist in mixed cultures and are physiologically compatible with one another. When these cultures are introduced into the natural environment, their individual beneficial effects are greatly magnified in a synergistic fashion. EM also enhanced crop management practices such as crop rotations, use of organic amendments, conservation tillage, crop residue recycling and bio control of pests.

According to Higa (1993), the beneficial microorganisms contained in EM create optimum soil microflora populations, which decompose organic amendments (e.g.) crop residue, animal waste, mulches, food waste and create soil that are disease suppressive, fix nitrogen and enhance soil quality.

Hussain (1994) established that EM can be applied as inoculants to increase the microbial diversity of soils and plants, which in turn, can improve soil health and the growth, yield and quality of crops. EM is prepared from cultures of naturally, occurring species of microorganisms that are found in natural environments world-wide. Microorganisms are useful in eliminating problems associated with the use of chemical fertilizers and pesticides and they are now widely applied in nature farming and organic agriculture (Parr *et al.*, 1994).

Wididana and Higa (1995) reported some new perspectives on the role and application of beneficial microorganisms, including EM, as microbial inoculants for shifting the soil microbiological equilibrium in ways that can improve soil quality, enhance crop production and protection, conserve natural resources and ultimately create a more sustainable agriculture and environment. The ultimate goal of EM inoculation is to select microorganisms that can be introduced as mixed cultures into soil where their beneficial effects can be realized. Microorganisms present in EM appears to suppress the development of harmful pathogens at the leaf surface, thereby providing a measure of plant protection through bio-control.

2.2.1 Effective Microorganisms (EM) and Solid Waste (SW)

Senanayake and Sangakkara (2001) reported that the application of EM technology in the field of integrated recycling of organic urban waste provided beneficial effects for environmental conservation and health economically.

Saraswate (2002) reported that the solid waste management by EM removed odor, reduced fly population, improved the health of sanitary workers and converted the solid waste into high quality compost. It is a low cost application with easy maintenance and does not require any costly installation and can be applied to already existing garbage heaps.

Sekeran *et al.* (2005) investigated the evaluation of Effective Microorganisms (EM) in solid waste management. Their study revealed that there was reduction in the C : N ratio from 28 : 1 (fresh waste) to 26 : 1 (composted waste) and organic carbon from 32.16 per cent to 30.05 per cent, nitrogen and phosphorous content showed an increase from 0.98 to 1.21 per cent and from 0.40 to 0.63 per cent.

In 24 schools in Coimbatore, biodegradable waste was segregated at source from garbage, put in compost pit and was converted into eco-friendly nutrient enriched organic manure using EM (Effective Microorganisms) technology by Eco-forums, organized by Siruthuli (Hindu, 2006).

According to Nandan (2006), urban garbage is becoming a growing problem and EM is being identified as an effective technology to promote recycling of organic waste. There was a remarkable impact of EM technology in paddy farming. About 90 per cent of paddy farmers were able to save Rs.9000 ha^{-1} and 10 per cent farmers, Rs. 5000 ha^{-1} by using EM instead of agrochemicals. He also conducted a survey and found that EM Bokashi has increased the water holding capacity of the soil and destroyed the lady beetle and ground beetle when applied as spray and also helped to improve healthy and good quality vegetables in beet root and tomato.

Thinley (2006) reported that EM technology has been tested over a wide range of agro-ecological conditions. Trial conducted using EM Bokashi and sprays have indicated, improvement in soil fertility and EM sprays on urban garbage have totally suppressed odour. The application of EM technology have been valuable in composting organic waste, including food industry, kitchen waste and municipal solid waste.

2.3 SOLID WASTE AND ENZYMATIC ACTIVITY

Wittling *et al.* (1995) compared the lumbri compost and two classical MSW composts at different maturation times. Organic carbon mineralization and nine enzyme activities along with dehydrogenase, urease and peroxidase were analysed. Generally, youngest compost exhibited greater activity. Dehydrogenase was the one that strictly increased, along with peroxidase activity. The soil urease enzyme increased but had only transient increase.

Pascual *et al.* (1999) investigated the influence of an arid soil amended with the organic fraction of a MSW at two different rates (6.5 and 26 t ha^{-1}) and they found that it had a positive effect on the activity of enzymes involved in the C, N, P cycles as well as in biomass carbon, suggesting that adding MSW might be a suitable technique to restore soil quality.

Albiach *et al.* (2000) investigated the effect of usual or recommended rates of application of five organic amendments (24 t ha^{-1} y^{-1} of MSW compost, sewage sludge, and bovine manure, 24 t ha^{-1} y^{-1} of vermicompost and 100 l ha^{-1} y^{-1} of a commercial humic acid solution) on the microbial biomass content and level of selected enzymatic activities in soil. The results of the study showed that the MSW compost treatment enhanced soil enzymatic activities like dehydrogenase and urease, compared to the other treatments.

Gil *et al.* (2000) conducted a long term field experiment with four different treatments prior to sowing barley with Municipal Solid Waste compost at either 20 t ha^{-1} (C 20) or 80 t ha^{-1} (C 80), cow manure (CM) at 20 t ha^{-1}, NPK (400 kg ha^{-1}) and NH_4 No_3 (150 kg ha^{-1}). The effects of these applications on enzyme activities were measured after nine years.

Among the treatments, municipal solid waste compost addition treatment (C 80) showed higher dehydrogenase activity (200 per cent).

Bhattcharyya *et al.* (2001) investigated the dynamics of soil quality indicators, such as microbial biomass - C, soil respiration, urease and acid phosphotases in a laterite soil amended with different doses of municipal solid waste compost over 120 days of incubation. The urease activity was found to be increased with an increase in dose of municipal solid waste compost and it attained a peak value at 60 days.

Municipal solid waste compost, paper solid waste and an agroforest compost were tested to relate their stability on soil enzyme activity. The application of the three organic materials to a sandy soil (50,000 kg ha^{-1}) increased the soil enzyme dehydrogenase activities with respect to the control (Madejon *et al.*, 2001).

Arul (2002) conducted experiments with different solid waste application to the soil. The solid waste compost performed better than FYM in all aspects and increased the activity of oxido reductase enzyme, urease. The yield was also increased in the solid waste compost applied plots to 40 per cent in the marigold plants.

The conversion of solid waste from sugar and distillery industry as a biocompost application on turmeric yields, registered an increase in OC, EC, pH and NPK of soil. The analysis also registered an increased enzyme activities like dehydrogenase and urease in SWC treatment than the control (Davamani, 2002).

Jothimani (2002) conducted experiments on solid waste compost by applying it to maize. Thirteen treatments were carried out, out of which the T_{12} treatment (sugarcane trash compost by Japanese method +

25.0 t ha^{-1} + 50 per cent NPK) showed significant increase in plant height, root volume, number of leaves and yield parameters. The highest dehydrogenase activity was observed in T_{12} (125.3 μg TTC formed g^{-1} of soil) and lowest activity in 100 per cent NPK (73.7 μg TTC formed g^{-1} of soil). The highest catalase activity was also recorded at the flowering stage (150.7 μ mol H_2O_2 g^{-1} of soil), the mean catalase activity ranged between 91.3 in control and 157.3 in T_{12}.

The urease activity of soils of different agro-ecosystems of alluvial soil was studied by Chakrabarti *et al.* (2004). The urease activity showed positive correlation on the soils. Multiple regression analysis showed that the stabilization of urease activity in soils was due to an organic matter complex.

Crecchio *et al.* (2004), inferred that Municipal Solid Waste composts (MSWC) maintained the long-term productivity of agro ecosystems and protect the soil environment from overcropping. The MSWC was added at recommended dosage (12 and 24 t ha^{-1}). Amendments of cropped plants with MSWC increased the contents of organic carbon from 13.3 to 15.0 g kg^{-1} of soil and total nitrogen from 1.55 to 1.65 g kg^{-1} of soil. There was a significant increase in dehydrogenase (9.6 per cent), β-glucosidase (13.5 per cent), urease (15.4 per cent), nitrate reductase (21.4) and phosphatase (9.7 per cent).

Chaudhury *et al.* (2005) laid out 5 treatments (control, 100 per cent N, 100 per cent N and P, 100 per cent NPK, 100 per cent NPK + FYM). Soil samples were collected after the harvest of rice and analysed for total soil N, available P and dehydrogenase activity. The highest soil quality index was found in 100 per cent NPK + FYM followed by 100 per cent NPK, respectively.

Haneklaus *et al.* (2005) compared the conventional agricultural methods with organic farming. Soil biological methods, such as release of CO_2, nitrification and dehydrogenase activities were reflected in higher concentrations in organic farming practices.

Masto *et al.* (2006) carried out field experiments on maize and cowpea, by applying FYM + 100 per cent NPK and 100 percent NPK. They have observed that increase in NPK from 50 to 100 per cent increased the dehydrogenase activity slightly. Barrena *et al.* (2007) reported that the deyhyrogenase enzymatic activity was determined to monitor the biological activity in a composting process of organic fraction of municipal solid waste. Dehydrogenase activity is proposed as a method to describe the biological activity of the thermophilic and mesophilic stages of composting. The maximum dehydrogenase activity was detected at the end of the thermophilic stage of composting, with values within 0.5 - 0.7 mg g^{-1} ha^{-1} on dry matter basis. Also, dehydrogenase activity can be correlated to static respiration index during the maturation of the mesophilic stage.

Castaldi *et al.* (2007) established the dynamics of biochemical (enzymatic activities) and chemical (water soluble fraction) parameters during 100 days of municipal solid waste composting to evaluate their suitability as tools for compost characterization. Dehydrogenase activities were characterized by significant changes during the first two weeks of composting, because of the increase of easily decomposable organic compounds. After the 4^{th} week, a "maturation phase" was identified in which the enzymatic activities tended to gently decrease, suggesting the stabilization of organic matter. Also the water-soluble fractions (water-soluble carbon, nitrogen and phenols), which are involved in many degradation processes, showed major fluctuations during the first month of composting. The results obtained showed a significant correlations between the enzymatic activities, as well as water-soluble fractions. The study also highlighted the suitability of both enzymatic activities and water soluble fractions as suitable indicators of the state and evolution of the organic matter during composting.

Chang *et al.* (2007) cultivated 24 crops of vegetable for 3 consecutive years and studied the effect of different application rates of compost (270 kg N ha^{-1} y^{-1}; 540 kg N ha^{-1} y^{-1}; 810 kg N ha^{-1} y^{-1} and 1080 kg N ha^{-1} y^{-1}). They compared it with the effects of chemical fertilizer. The results showed that the pH, electrical conductivity, nitrogen and organic matter received from compost treatment were higher than those received by chemical fertilizer. There was a significant increase in the dehydrogenase and urease enzymatic activities in 540 kg N ha^{-1} y^{-1} treatment compared to the other treatments.

Nogales and Benitez (2007) conducted an incubation experiment in composted, vermicomposted, olive cake on artificially contaminated calcareous soil. Application of compost increased humic acids in soil, stimulated dehydrogenase and urease activities, where as the other application showed low activity.

2.4 EM - SW COMPOST AND YIELD

The effects of Effective Microorganisms on rice growth were investigated on three soil types in Thailand by Komboonruang *et al.* (1996). The treatments included fumigated soil with methyl bromide, application of inorganic fertilizers, inoculation with Effective Microorganisms and control without any inoculation. He inferred that the inoculation of EM increased the yield of rice crop along with an increase in the organic matter content (6.2 per cent).

Thananusont (1996) reported that in a pot culture experiment, nodulation and nitrogen fixation of soybeans were increased by inoculation with rhizobium, but growth and yield were significantly increased by EM inoculation. The influence of EM was evaluated in field trials of vegetable crops. EM plus molasses were applied at 10 l ha^{-1} in 10,000 l ha^{-1} water, 3 times to the onions, twice to the peas and 7 times to the sweet corn. As a result of this application, there was an increase in yield by 29 per cent in onion, 31 per cent in peas. The cob weights was increased by 23 per cent in sweet corn (Daly *et al.*, 1999).

Sangakkara *et al.* (1999) reported that composting of organic matter with EM and foliar applications during the growth stages resulted in high yields of selected crops. A field study was conducted by Yadav (2000) on the performance of Effective Microorganisms (EM) on growth and yield of selected vegetables (cabbage and radish). He found that the yield of radish roots (the edible parts) was significantly higher in the EM sprayed plots at 15 days interval (70.5 per cent) over the control. In cabbage crop, the EM sprayed plots gave more yield (91.58 per cent) over control.

Freitag (2000) in a case study of sustainable farming and composting reported that a flannery with 3 acres of land produced fruits and vegetables of high quality and yield without the use of a single drop of chemical fertilizer, pesticide or tilling machinery. The flannery collected organic solid waste in 90 - gallon trash buckets, then mixed with EM - Bokashi. Under optimal conditions, the entire mixture was converted into EM - rich compost.

Lucas and Margarita (2001) introduced EM in Auroville, and have been monitoring its use. The success has been remarkable. A few

Auroville farms sprayed EM on their vegetable gardens, paddy fields and fruit trees. One of the success observed was the eradication of rhinoceros beetle in coconut trees. EM is also successfully used to improve compost and enhanced the formation of organic fertilizer called "Bokashi". It is a bran based material that has been seeded with EM and dried to make storage easy. It is also used in treatment of solid waste with a foul smell, which when treated, solved the problem of bad smell.

Wood *et al.* (2004) in their study observed that the corn plants grown with EM and waste water treatment showed the best results in yield, quality of ears and grain. A significantly higher weight (18.25 g) and circumference of an ear without husks was noticed. The average number of ears harvested was also highest throughout the growth of corn plants. The plants were also greener and fresh than untreated plants.

Bruggenwert (2005) conducted pot culture experiment with English rye-grass, using EM in combination with six treatments of manures and fertilizers, which had a significant positive effect. This technology was also tested on meadows which resulted in a high production of grass, where the chemical fertilizers usage were reduced to about 30 per cent. The effect of EM on the photosynthesis of grass and maize were measured on eight parcels, which had a higher ppm value of 78.6 ± 1.2, 63.4 ± 0.6, 71.4 ± 0.8 for grass and maize had 43.8 ± 0.8 ppm value which was higher. He also studied the effects of EM on rice variety CR 203. The application of EM solution and Bokashi had a good influence not only on growth and development, but also on yield and quality of rice. In the 2nd season, the rice yield was increased by 290 - 490 kg ha^{-1} (8.0 - 15.5 per cent) compared with the control.

Daniel (2005) investigated the influence of EM composted coarse biomass on soil, during the growth and yield of chickpea and onion. The results of the study showed a high yield of onion bulb (15.60 t ha^{-1}) and chickpea (1.68 t ha^{-1}). Hussain *et al.* (2005) conducted field experiment to study the prospects of wheat cultivation by adopting an innovation, using EM super fermenter technology along with chemical and organic sources of nutrients. A significant increase in all parameters was found with application of ½ recommended dose of fertilizer along with EM - super fermenter water producing 2831 kg grains ha^{-1} very close to full recommended dose of fertilizer (3017 kg grains ha^{-1}). In economical terms, it saved about 38 per cent cost, against chemical fertilizer and more over EM - super fermenter technology can be adopted for sustainable crop production.

Prinsloo *et al.* (2005) investigated the production of maize, lettuce and onions under flood drip and micro sprinkler irrigation, using EM and chemical fertilizer as an alternative nutrient sources. An acid-sandy soil with high permeability and low exchange capacity was used as the substrate. A significant difference between EM treated and chemically fertilized treatment was observed. Micro sprinkler irrigation using EM yielded significantly higher yields in maize, lettuce and onion.

EM Bokashi and EM spray applied for soybean variety DT 84 showed 10 - 19 per cent higher growth and yield than the control (Quang *et al.*, 2005). Witting (2005) applied the fermented EM-Bokashi to plots of sugar beets. The test field had 10 plots, out of which, 6 to 10 plots were given EM treatment. It was concluded that, plot 6 (0 kg, N ha^{-1} + EM 1) had the extreme high nitrogen content of 121 N and 16.1 per cent sugar content ha^{-1}, than others. The plot 10 (50 kg N ha^{-1} + EM 1 + 4000 kg Bokashi) produced high yield of 75.8 t ha^{-1} sugar beets.

Ho and Hwan (2006) carried out experiments to determine the presence of endogenous hormones in EM and their activity on the physiology of selected crops and associated effects on plant growth. They found that the solution of EM contained many hormones, and their effects varied. Laboratory experiment with rice, corn and Chinese cabbage showed that EM promoted root growth significantly. Weijiong *et al.* (2006) after several years of research, by using EM and organic manure without using pesticide and chemical fertilizer obtained an increase in the yield of wheat and corn.

2.5 Test Crops - Lady's Finger and Cow Pea

2.5.1 Okra

(*Abelmoschus esculentus* (L.) Moench) also known as lady's finger, bhindi and gumbo, is a flowering plant, valued for its edible green fruits, is cultivated throughout the tropical and warm temperate regions of the world. It belongs to the family Malvaceae. It is an annual or perennial plant, growing to 2 m tall. The leaves are 10-20 cm long and broad, palmately lobed with 5-7 lobes. The flowers are 4-8 cm diameter, with five white to yellow petals, often with a red or purple spot at the base of each petal. The fruit is a capsule up to 18 cm long, containing numerous seeds.

Nutritional value per 100 g (3.5 oz).

It has energy of 30 kcal, carbohydrates 7.6 g, high dietary fibre 3.2 g, low saturated fat 0.1 g, protein 2.4 g, Vitamin A, B_2, B_9, and C 0.32 mg, 0.17 mg, 0.21 mg, 87.8 μg and 21 mg respectively. Apart from these, it is also found to contain Iron, Zinc, Copper, Phosphorous, Potassium and Manganese.

Uses

Kenaf-blend panels are produced from okra fibre. Handicrafts, excellent mats, hats, baskets, etc are made from Kenaf fiber. Roots and stem are used as a fuel and animal feed.

2.5.2. Cowpea

(*Vigna unguiculata* (L.) Walp), is one of several species of the widely cultivated genus *Vigna*, an important grain legume throughout the tropics and subtropics, covering Asia, Africa, and Central and South America, as well as parts of southern Europe and the United States. It belongs to the family Fabaceae. It also has the useful ability to fix atmospheric nitrogen through its root nodules, and it grows well in poor soils with more than 85% sand and with less than 0.2% organic matter and low levels of phosphorus. In addition, it is shade tolerant, and therefore, compatible as an intercrop with maize, millet, sorghum, sugarcane, and cotton.

Nutritional value

Carbohydrates ranged from 69.4% to 85.9%. Protein - 85.9%, total sugars -8.77%, calcium -6.83 mg, Iron -12.38 mg and zinc- 8.09 mg.

Uses

Cowpea, an important legume in the tropics, has many uses. In fresh form, the young leaves, immature pods and peas are used as vegetables, while several snacks and main meal dishes are prepared from the grain. All parts of the plant that are used for food are nutritious, providing protein, vitamins (notably vitamin B) and minerals. The cowpea haulm is also a great source of livestock feed, and therefore of great value to farmers.

2.6NPK

Influence on Root Nodule

Kim *et al.* (1988) and Jensen and Sorenson (1988) reported that nodule number showed inverse relation with increased increment of applied nitrogen. Maximum soybean seed yield depend to a large extent upon a well-nodulated extensive root system, the development of which was enhanced by ample nutrients from the soil along with water and energy (Hicks, 1978).

Influence on Yield

Increase in plant height due to increased nitrogen fertilizer has been reported by El-kady *et al.* (1982) ; Kamel *et al.* (1987) and Alam *et al.* (1988). Saxena *et al.* (2001) found that the application of nitrogen helped in increasing the vegetative growth of plants and also plant dry matter in soybean. They also reported that increase in N and P rates increased the pods / plants, seeds / pod, 100 seed weight and protein content in soybean. According to Singh and Abraham (2001), soybean gave significantly higher seed and biological yields with 60 Kg N ha^{-1} and beyond this level there was a marginal increase in yield, the increase being 21.6 and 18.7 per cent over 30 Kg N ha^{-1}.

Chitdeshwari *et al.* (2007) found that the application of 34:68:108 kg NPK ha^{-1} at a split of 50:25:25 per cent N and K at basal, flowering and peg forming stage and 100 per cent P as basal, recorded the highest pod yield of 1557 kg ha^{-1} in ground nut and was ascribed to the enhanced nutrient addition through fertilizers which might have contributed to more water and nutrient uptake and enzymatic activity. A significantly increased total dry matter accumulation of 17.83 and 26.37 g $plant^{-1}$ in radish was observed by Shivanna *et al.* (2007) at 30 and 45

DAS with 150 kg N ha^{-1} over other N levels. Tiwari *et al.* (2007) reported that the application of recommended dose of fertilizers (20:60:30 NPK kg ha^{-1}) to soy bean gave significantly higher seed (23.64 q ha^{-1}) and straw (26.03 q ha^{-1}) yields over control (19.68 and 16.55 q ha^{-1}). According to Kumar *et al.* (2007), phosphorus @ 90 kg ha^{-1} gave an enhanced grain (14.2 q ha^{-1}) and straw (43.20 kg ha^{-1}) yields in green gram over control (9.83 q ha^{-1} and 23.49 kg ha^{-1}).Ilavarasi *et al.* (2007) observed that the treatment with NP @ 70:70 kg ha^{-1} have resulted in higher values in available NPK status of post-harvest soil (133.54, 32.52 and 241.7 NPK kg ha^{-1}) over control (57.07, 5.9 and 117 NPK kg ha^{-1}) in cow pea. Pod yield was also enhanced significantly (79.45 g plant^{-1}) with NP @ 70:70 kg ha^{-1} when compared with control (30.5 g plant^{-1}).

2.7 BIOFERTILZIER - RHIZOBIUM

The use of bio fertilizer for crops have been observed in reducing N dosage and increasing the effectiveness of applied inorganic N. Rhizobium has been observed to be an efficient source of biofertilizer. There is a consistent common trend of increased effectiveness on account of use of bio fertilizers supplemented by chemical N fertilizers.

Influence on Yield

Saxena *et al.* (1971) reported that black gram seed yield was increased by rhizobial seed inoculation. Rhizobial seed inoculation increased the yields of cow pea (Deshmukh and Joshi, 1973). Gupta *et al.* (1976) obtained maximum seed yield when legume seeds were inoculated with Rhizobium. A high correlation between number of flower clusters and grain yield was shown in pigeon pea due to Rhizobium inoculation by Pani (1979). Dahiya *et al.* (1980) obtained higher seed yield of red gram with Rhizobium seed inoculation.

According to Raj and Patel (1991), Rhizobium inoculation significantly increased the grain and dry forage yield over no inoculation in cowpea. The increase was 9.25 and 9.79 per cent for grain and dry forage yield. This increase was attributed due to favourable effect of Rhizobium inoculation in cowpea. Solaiappan *et al.* (1994) stated that seed inoculation with Rhizobium enhanced the seed yield of red gram.

Prabhakaran *et al.* (1999) stated that the application of bio fertilizers significantly increased the pod yield in summer groundnut.

Sharma *et al.* (2000) found that the inoculation of Rhizobium with nitrogen application increased the number of pods, number of clusters and grain yield of black gram. Singh and Abraham (2001) also reported increase in seed yield due to Rhizobium inoculation in soybean. Studies by Pandey and Kumar (2002) on legume Rhizobium inoculation have indicated that on an average 10-25 per cent increase in yield is achievable in almost all the pulses. Rhizobium legume symbiosis can meet 60-80 per cent of the nitrogen requirement of legume crop.

According to Gupta and Abraham (2003) Rhizobium inoculation significantly influenced the dry matter accumulation (g/m^2) at 40 and 60 DAS. The maximum dry matter (953.50 g/m^2) was accumulated by Rhizobium inoculated treatment, which was about 7.92 per cent more than uninoculated at 100 DAS. The inoculated treatments showed 20 per cent more relative growth rate than uninoculated treatment at 40 DAS in soybean. They also reported that the inoculation of Rhizobium, increased the yield attributes like number of pods / plant, total grains / plant and number of seeds / pod which were 31.15, 33.71 and 2.76 per cent more than uninoculated treatment in soybean.

According to Khalequzzaman and Hossain (2007), the use of BINAR P.36 *Rhizobium* strain to bush bean resulted in highest germination percentage (93.25 per cent) over control (72.33 per cent). Also plant height (27.67cm), plant fresh weight (7.94 g) seedling vigour (2580.23) and number of nodules/plant (20.77) were increased significantly when compared with control (18.37 cm, 4.68 g, 1328.70 and 0.00 respectively). According to Ahmed *et al.* (2008), the seed + soil inoculation of *Rhizobium* to lentil resulted in maximum number of pods/plant (12.23), 100 seed weight (3.57 g) and seed yield (2.66 g $plant^{-1}$) over control (12.01, 3.20 g and 1.89 g $plant^{-1}$ respectively).

Influence on Nitrogen Uptake

Sanoria and Rawat (1981) have found that inoculation of Rhizobium in bengal gram increased the total uptake of nitrogen in seeds and plant tissues. According to Muniruzzaman and Khan (1990), inoculation of Rhizobium increased nitrogen uptake in lentil.

Influence on Nodule

According to Prabhakaran and Srinivasan (1995), inoculation of Rhizobium increased the vigour index, plant biomass, nodule number, nodule biomass and yield of red gram.According to Jat. *et al.* (1996), Rhizobium inoculation to soybean resulted in 46 per cent increase in mean nodule count and 61 per cent increase in mean nodule dry weight over no inoculation.

2.8 BIOCHEMICAL ASPECTS

2.8.1 Protein content

A positive correlation has been found between leaf photosynthetic rate and extractable RUBP carboxylase activity of soybeans (Bowes *et al.,* 1972). About 50 per cent of the total soluble protein in leaf extract is accounted for by RUBP carboxylase (Ellis, 1976). Thus, soluble protein content of the leaves can be taken as a measure of RUBP carboxylase activity.

In green gram, selection and use of efficient strains of Rhizobium together with other agronomic practices would lead to increased supply of quality protein to the microbial population and maintain soil fertility at a high level according to Tilak (1998).

2.8.2 Chlorophyll Content

Lawn *et al.* (1974) reported an increase in leaf photosynthetic rate due to inoculation of Rhizobium. Nitrogen fixation, Nodule activity and NAR had a positive correlation with photosynthetic rate in nodulating strains. A positive relation was found between chlorophyll content and leaf RUBP carboxylase and nodule nitrogenous activity (Salah *et al.,* 1977).

The optimum nutrition especially nitrogen, through greater availability of nutrients and photosynthates resulted in proper growth and development of both vegetative and reproductive structure of the crop and finally leads to improved productivity in soybean (Bhargava *et al.,* 1979).

Fayed and Mostafa (1986) reported an increase in chlorophyll content with an increase in rate of nitrogen fertilization. They also reported that split nitrogen application at different growth stages had a greater effect on leaf chlorophyll content.

2.8.3 Leghaemoglobin Content

The first positive evidence of a role for leghaemoglobin in legume nodule respiration came from Tjepkama and Yocum (1970). Elfork (1972) and Appleby (1974) inferred that the function of leghaemoglobin was generally accepted as the facilitation of oxygen flux to the vigorously respiring Rhizobium bacteriods.Good child and Bergersen (1973) reported that the leghaemoglobin content was highest when nitrogen fixing activity was high and the nodule volume occupied by leghaemoglobin was 38 per cent.

According to Zablotowicz *et al.* (1980), the leghaemoglobin content of the nodules is influenced by the plant age and inoculum strain. The leghaemoglobin content showed its peak during 50^{th} day after emergence and declined subsequently. Eiji and Syono (1982) showed that the leghaemoglobin components participated in more effective nitrogen fixation by controlling oxygen transport to the bacteriods. According to Klucas *et al.* (1985), the increased amount of leghaemoglobin content in inoculated plants reflects the efficiency of Rhizobium isolates to help the plants to synthesize more leghaemoglobin. Due to increased nitrogenase activity, there is more fixation of nitrogen which helps in enhancing photosynthetic efficiency of plant and hence there is more accumulation of dry matter in plants inoculated with Rhizobium. The improved nodulation and nitrogenous activity resulted in increased nitrogen content of the plants inoculated with Rhizobium. Huang *et al.* (1988) observed that the biosynthesis of leghaemoglobin was affected with increased nitrate nutrition, which was manifested in low leghaemoglobin contents.

2.9 NON-ENZYMATIC ANTIOXIDANTS IN PLANTS

2.9.1 Ascorbic Acid and Tocopherol

Ascorbic acid is a key precursor in all vegetables and fruits. This is linked to cell growth, cell cycle, photosynthetic and respiratory process and as well as acts as a co-factor for many enzymes. This is also called as vitamin-C.

Tocopherol are the most abundant vitamin E forms in our food and it is present in all the plant parts. When this vitamin-E forms are taken in our food, it has anti ageing properties.

According to Lee and Kader (2000), ascorbic acid plays a protective role against reactive O_2 species that are formed from photosynthetic and respiratory processes. It is linked to cell growth being involved in cell cycle and other mechanisms of cell growth and cell division of plant. Lian *et al.* (2001) examined the effects of applications of an organic fertilizer (EM - bokashi), and chicken manure on photosynthesis and fruit yield and quality of tomato plants. EM - organic manure increased the rate of photosynthesis and fruit yield of tomato plants. Concentrations of sugars and organic acids were higher in fruits of plants treated with EM - bokashi, organic manure. EM inoculation increased ascorbic acid concentration in fruit in all fertilization treatments.

Prenetha (2002) conducted experiments in brinjal genotypes and studied the yield of nine selected parents and their hybrids. The highest ascorbic acid content recorded was 2068 mg g^{-1} in EP 104 XEP 5.

Anderson *et al.* (2004) analysed the ascorbic content in many fruits and vegetables at their green and mature stages. The results showed that in strawberry, ascorbic acid content was 58 per cent in the mature green stage and increased eventually to 78 per cent in the red ripened fruits. The total ascorbic content tends to increase in broccoli and sweet pepper.

Ascorbic acid content was measured in two varieties of tomato (*Lycopersicon esculentum* L. cv. Ropreco and Burbank) and two varieties of bell peppers (*Capsicum annuum* L. cv. California Wonder and Excalibur) grown by certified organic and conventional practices in a model system. Significantly higher levels of ascorbic acid (26 per cent) was found in Burbank tomatoes. Burbank tomatoes generally had higher levels of ascorbic acid as compared to Ropreco tomatoes (Chassy *et al.*, 2006). Sanwal *et al.* (2006) carried out field experiments on the growth and yields of broccoli. The treatments were solid waste organic materials compost teas, synthetic fertilizer and control. The application of organic materials reduced soil acidity, improved the organic matter and available nutrients in the soil. The antioxidants and ascorbic acid content also increased. Use of organic solid waste appears to provide an alternative to conventional methods.

2.10 ENZYMATIC ANTIOXIDANTS IN PLANTS

2.10.1 Catalase and Peroxidase

Gil *et al.* (2000) found that the increase in oxido reductase enzyme, catalase activity was due to increase in microbial metabolism in a soil as a result of the mineralization of biodegradable - C fractions contained in MSW compost. They also reported that MSW compost at 80 t ha^{-1} increased the catalase activity by 200 per cent.

Sathiyamurthy (2002) conducted a study on development of F_1 hybrids in chilli to analyse *Capsaisin* content and yield. The results of the study revealed that the peroxidase activity ranked first 788.58 min^{-1} mg^{-1} fresh weight in individual green fruit at 45 DAF (Days After Flowering) in ujwala x P x m F_1 hybrid variety. The activity of peroxidase recorded lowest in parent (668.56 min^{-1} g^{-1} fresh weight). Ruth (2002) studied F_1 hybrids of chilli and their 30 hybrids. The results of the study showed that the peroxidase, polyphenol oxidase activities were higher in resistance genotypes. The ascorbic acid content reached a maximum of 64.347 mg g^{-1} in hybrids (in 3 x ujwala).