SOLID WASTE MANAGEMENT INTO BIOFERTILIZER AND POLLUTION CONTROL

BY

KHADIM HUSSAIN

Department of Chemistry
Bahauddin Zakariya University, Multan
Pakistan
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SOLID WASTE MANAGEMENT INTO BIOFERTILIZER AND POLLUTION CONTROL

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KHADIM HUSSAIN

Department of Chemistry
Bahauddin Zakariya University, Multan
Pakistan
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## CONTENTS

<table>
<thead>
<tr>
<th>Chapter #</th>
<th>Title</th>
<th>Page #</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Acknowledgements</td>
<td></td>
</tr>
<tr>
<td></td>
<td>List of Tables</td>
<td></td>
</tr>
<tr>
<td></td>
<td>List of Figures</td>
<td></td>
</tr>
<tr>
<td></td>
<td>List of Abbreviations</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Abstract</td>
<td></td>
</tr>
<tr>
<td><strong>Chapter 1</strong></td>
<td><strong>Introduction</strong></td>
<td><strong>1</strong></td>
</tr>
<tr>
<td><strong>Chapter 2</strong></td>
<td><strong>Review of Literature</strong></td>
<td><strong>4</strong></td>
</tr>
<tr>
<td>2.1</td>
<td>Composting processes</td>
<td>7</td>
</tr>
<tr>
<td>2.2</td>
<td>EM technology</td>
<td>9</td>
</tr>
<tr>
<td>2.3</td>
<td>Soil organic carbon dynamics</td>
<td>11</td>
</tr>
<tr>
<td>2.4</td>
<td>Soil microbial biomass</td>
<td>15</td>
</tr>
<tr>
<td>2.5</td>
<td>Biodegradation of soil organic matter</td>
<td>17</td>
</tr>
<tr>
<td><strong>Chapter 3</strong></td>
<td><strong>Materials and Methods</strong></td>
<td><strong>21</strong></td>
</tr>
<tr>
<td>3.1</td>
<td>Preparation of biofertilizer</td>
<td>21</td>
</tr>
<tr>
<td>3.2</td>
<td>Soil analysis</td>
<td>23</td>
</tr>
<tr>
<td>3.2.1</td>
<td>Soil physico-chemical analysis</td>
<td>23</td>
</tr>
<tr>
<td>3.2.2</td>
<td>Soil chemical analysis</td>
<td>24</td>
</tr>
<tr>
<td>3.2.2.1</td>
<td>Total nitrogen</td>
<td>24</td>
</tr>
<tr>
<td>3.2.2.2</td>
<td>Available phosphorus</td>
<td>24</td>
</tr>
<tr>
<td>3.2.2.3</td>
<td>Available potassium</td>
<td>25</td>
</tr>
<tr>
<td>3.2.2.4</td>
<td>Organic matter</td>
<td>25</td>
</tr>
<tr>
<td>3.3</td>
<td>Plant analysis</td>
<td>25</td>
</tr>
<tr>
<td>3.3.1</td>
<td>Total nitrogen</td>
<td>25</td>
</tr>
<tr>
<td>3.3.2</td>
<td>Total phosphorus</td>
<td>26</td>
</tr>
<tr>
<td>3.3.3</td>
<td>Total potassium</td>
<td>26</td>
</tr>
<tr>
<td>3.4</td>
<td>EM-biofertilizer efficiency for cotton crop</td>
<td>27</td>
</tr>
<tr>
<td>3.5</td>
<td>EM-biofertilizer efficiency for okra growth</td>
<td>28</td>
</tr>
<tr>
<td>3.6</td>
<td>EM-biofertilizer efficiency for spinach</td>
<td>29</td>
</tr>
<tr>
<td>3.7</td>
<td>EM-biofertilizer efficiency for cowpea growth</td>
<td>29</td>
</tr>
<tr>
<td>3.8</td>
<td>Statistical analysis</td>
<td>29</td>
</tr>
<tr>
<td><strong>Chapter 4</strong></td>
<td><strong>Results and Discussion</strong></td>
<td><strong>31</strong></td>
</tr>
<tr>
<td>4.1</td>
<td>Composition of EM-biofertilizer</td>
<td>31</td>
</tr>
<tr>
<td>4.2</td>
<td>EM-biofertilizer efficiency and effect on soil health</td>
<td>32</td>
</tr>
<tr>
<td>4.3</td>
<td>Cotton growth</td>
<td>33</td>
</tr>
<tr>
<td>4.3.1</td>
<td>Cotton leaf NPK contents</td>
<td>35</td>
</tr>
<tr>
<td>4.3.2</td>
<td>Cotton leaf NPK contents relation with boll size</td>
<td>36</td>
</tr>
<tr>
<td>4.3.3</td>
<td>Soil NPK and OM contents after cotton harvest</td>
<td>38</td>
</tr>
<tr>
<td>4.3.4</td>
<td>Soil NPK and K relation with boll circumference</td>
<td>39</td>
</tr>
<tr>
<td>4.4</td>
<td>Okra growth</td>
<td>42</td>
</tr>
<tr>
<td>4.4.1</td>
<td>Okra leaf analysis (NPK contents)</td>
<td>44</td>
</tr>
<tr>
<td>4.4.2</td>
<td>Soil analysis after okra</td>
<td>47</td>
</tr>
<tr>
<td>4.5</td>
<td>Spinach (<em>Spinacia oleracea</em>) growth parameters</td>
<td>54</td>
</tr>
<tr>
<td>Section</td>
<td>Topic</td>
<td>Page</td>
</tr>
<tr>
<td>------------------</td>
<td>----------------------------------------------------------------------</td>
<td>------</td>
</tr>
<tr>
<td>4.5.1</td>
<td>Spinach leaf analysis (NPK contents)</td>
<td>55</td>
</tr>
<tr>
<td>4.5.2</td>
<td>Soil analysis after spinach</td>
<td>58</td>
</tr>
<tr>
<td>4.6</td>
<td>Cowpea (<em>Phaseolus vulgaris</em>) growth parameters</td>
<td>64</td>
</tr>
<tr>
<td>4.6.1</td>
<td>Cowpea leaf analysis (NPK contents)</td>
<td>66</td>
</tr>
<tr>
<td>4.6.2</td>
<td>Soil quality parameters of cowpea</td>
<td>69</td>
</tr>
<tr>
<td><strong>Chapter 5</strong></td>
<td><strong>Summary</strong></td>
<td><strong>76</strong></td>
</tr>
<tr>
<td></td>
<td>Literature Cited</td>
<td>78</td>
</tr>
</tbody>
</table>
## LIST OF TABLES

<table>
<thead>
<tr>
<th>Table #</th>
<th>Title</th>
<th>Page #</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Physical characteristics of compost (un-inoculated) and EM-biofertilizer</td>
<td>22</td>
</tr>
<tr>
<td>2.</td>
<td>Composition of the EM-biofertilizer prepared after inoculating with different concentration of EM solution</td>
<td>22</td>
</tr>
<tr>
<td>3</td>
<td>Soil physico-chemical properties used for experiments</td>
<td>27</td>
</tr>
<tr>
<td>4</td>
<td>Effect of EM-biofertilizer on growth parameters of cotton</td>
<td>34</td>
</tr>
<tr>
<td>5</td>
<td>Cotton leaf NPK concentration as affected by various EM-biofertilizer application in potted soil</td>
<td>36</td>
</tr>
<tr>
<td>6</td>
<td>Soil total N, bioavailable P and K and total organic matter content</td>
<td>38</td>
</tr>
<tr>
<td>7</td>
<td>EM-biofertilizer application effect on growth of okra</td>
<td>43</td>
</tr>
<tr>
<td>8.</td>
<td>Leaf analysis of okra grown under EM-biofertilizer treatments</td>
<td>45</td>
</tr>
<tr>
<td>9</td>
<td>Soil analysis after okra</td>
<td>49</td>
</tr>
<tr>
<td>10.</td>
<td>Effect of EM-biofertilizer on growth parameters of spinach</td>
<td>54</td>
</tr>
<tr>
<td>11.</td>
<td>Leaf analysis of spinach</td>
<td>55</td>
</tr>
<tr>
<td>12.</td>
<td>Soil analysis after spinach</td>
<td>59</td>
</tr>
<tr>
<td>13.</td>
<td>Effect of EM-biofertilizer on growth parameters of cowpea</td>
<td>65</td>
</tr>
<tr>
<td>14.</td>
<td>Leaf analysis of cowpea</td>
<td>66</td>
</tr>
<tr>
<td>15.</td>
<td>Soil analysis after cowpea</td>
<td>70</td>
</tr>
</tbody>
</table>
## LIST OF FIGURES

<table>
<thead>
<tr>
<th>Fig. #</th>
<th>Title</th>
<th>Page #</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Cotton boll size increase with leaf nutrient content under Biofertilizer application</td>
<td>37</td>
</tr>
<tr>
<td>2(a).</td>
<td>Effect of soil nitrogen on boll circumference of cotton</td>
<td>40</td>
</tr>
<tr>
<td>2(b).</td>
<td>Effect of soil phosphorous on boll circumference of cotton</td>
<td>40</td>
</tr>
<tr>
<td>2(c).</td>
<td>Effect of soil potassium on boll circumference of cotton</td>
<td>41</td>
</tr>
<tr>
<td>2(d).</td>
<td>Effect of soil organic matter on boll circumference of cotton</td>
<td>42</td>
</tr>
<tr>
<td>3.</td>
<td>Effect of leaf nitrogen on biomass of okra</td>
<td>45</td>
</tr>
<tr>
<td>4.</td>
<td>Effect of leaf phosphorus on biomass of okra</td>
<td>46</td>
</tr>
<tr>
<td>5.</td>
<td>Effect of leaf potassium on biomass of okra</td>
<td>47</td>
</tr>
<tr>
<td>6.</td>
<td>Effect of soil OM on biomass of okra</td>
<td>48</td>
</tr>
<tr>
<td>7.</td>
<td>Effect of soil nitrogen on biomass of okra</td>
<td>50</td>
</tr>
<tr>
<td>8.</td>
<td>Effect of soil phosphorus on biomass of okra</td>
<td>51</td>
</tr>
<tr>
<td>9.</td>
<td>Effect of soil potassium on biomass of okra</td>
<td>52</td>
</tr>
<tr>
<td>10.</td>
<td>Effect of leaf nitrogen on biomass of spinach</td>
<td>56</td>
</tr>
<tr>
<td>11.</td>
<td>Effect of leaf phosphorus on biomass of spinach</td>
<td>57</td>
</tr>
<tr>
<td>12.</td>
<td>Effect of leaf potassium on biomass of spinach</td>
<td>58</td>
</tr>
<tr>
<td>13.</td>
<td>Effect of soil nitrogen on biomass of spinach</td>
<td>59</td>
</tr>
<tr>
<td>14.</td>
<td>Effect of soil phosphorus on biomass of spinach</td>
<td>60</td>
</tr>
<tr>
<td>15.</td>
<td>Effect of soil potassium on biomass of spinach</td>
<td>61</td>
</tr>
<tr>
<td>16.</td>
<td>Effect of soil OM on biomass of spinach</td>
<td>62</td>
</tr>
<tr>
<td>17.</td>
<td>Effect of leaf nitrogen on dry grain weight of cowpea</td>
<td>667</td>
</tr>
<tr>
<td>18.</td>
<td>Effect of leaf phosphorus on dry grain weight of cowpea</td>
<td>68</td>
</tr>
<tr>
<td>19.</td>
<td>Effect of leaf potassium on dry grain weight of cowpea</td>
<td>69</td>
</tr>
<tr>
<td>20.</td>
<td>Effect of OM on dry grain weight of cowpea</td>
<td>70</td>
</tr>
<tr>
<td>21.</td>
<td>Effect of soil nitrogen on dry grain weight of cowpea</td>
<td>71</td>
</tr>
<tr>
<td>22.</td>
<td>Effect of soil phosphorus on dry grain weight of cowpea</td>
<td>72</td>
</tr>
<tr>
<td>23.</td>
<td>Effect of soil potassium on dry grain weight of cowpea</td>
<td>73</td>
</tr>
<tr>
<td>Abbreviation</td>
<td>Description</td>
<td></td>
</tr>
<tr>
<td>--------------</td>
<td>-------------</td>
<td></td>
</tr>
<tr>
<td>AARI</td>
<td>Ayub Agriculture Research Institute, Pakistan</td>
<td></td>
</tr>
<tr>
<td>CEC</td>
<td>Cation Exchange Capacity</td>
<td></td>
</tr>
<tr>
<td>CFC</td>
<td>Chlorofluorocarbons</td>
<td></td>
</tr>
<tr>
<td>C:N</td>
<td>Carbon Nitrogen ratio</td>
<td></td>
</tr>
<tr>
<td>CRD</td>
<td>Completely Randomized Design</td>
<td></td>
</tr>
<tr>
<td>EfW</td>
<td>Energy from Waste</td>
<td></td>
</tr>
<tr>
<td>EM</td>
<td>Effective Microorganisms</td>
<td></td>
</tr>
<tr>
<td>FC</td>
<td>Filter Cake</td>
<td></td>
</tr>
<tr>
<td>FYM</td>
<td>Farm Yard Manure</td>
<td></td>
</tr>
<tr>
<td>GDP</td>
<td>Gross Domestic Product</td>
<td></td>
</tr>
<tr>
<td>HSWM</td>
<td>Household Solid Waste Management</td>
<td></td>
</tr>
<tr>
<td>IPCC</td>
<td>International Panel on Climate Change</td>
<td></td>
</tr>
<tr>
<td>LSD</td>
<td>Least Significance Difference</td>
<td></td>
</tr>
<tr>
<td>Mg ha(^{-1})</td>
<td>Mega gram per hectare</td>
<td></td>
</tr>
<tr>
<td>MSW</td>
<td>Municipal Solid Waste</td>
<td></td>
</tr>
<tr>
<td>OM</td>
<td>Organic Matter</td>
<td></td>
</tr>
<tr>
<td>PCDD</td>
<td>Polychlorinated dibenzo-p-dioxins</td>
<td></td>
</tr>
<tr>
<td>PCDF</td>
<td>Polychlorinated dibenzofurans</td>
<td></td>
</tr>
<tr>
<td>NIAB</td>
<td>Nuclear Institute for Agriculture and Biology, Pakistan</td>
<td></td>
</tr>
<tr>
<td>NPK</td>
<td>Nitrogen Phosphorus and Potassium</td>
<td></td>
</tr>
<tr>
<td>PM</td>
<td>Poultry Manure</td>
<td></td>
</tr>
<tr>
<td>RDF</td>
<td>Refuse Driven Fuels</td>
<td></td>
</tr>
<tr>
<td>SOM</td>
<td>Soil Organic Matter</td>
<td></td>
</tr>
<tr>
<td>USA</td>
<td>United States of America</td>
<td></td>
</tr>
<tr>
<td>USEPA</td>
<td>United States Environmental Protection Agency</td>
<td></td>
</tr>
</tbody>
</table>
ABSTRACT

Municipal solid waste (MSW) has become a critical problem for city environment. Indiscriminate dumping of waste around cities poses environmental hazards causing ecological imbalances with respect to land, water and pollution. The estimated amount of city waste in Faisalabad (Pakistan) is 1570 tonnes day\(^{-1}\). This waste along with others produced in the country in the form of cow dung, poultry manure, FYM and filter cake (FC) from sugar industry, can successfully be converted into biofertilizer with the help of EM (effective microorganisms). The EM-Biofertilizer is a potential source of organic matter and soil micro- and macro- nutrients for plant growth as well as for sustainable agriculture. In view of these considerations, the present research work was conducted in the laboratory and greenhouse of the Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, during 2003-2006. The objectives of the study were to preserve the nutrients present in the solid waste and convert them into biofertilizer. The raw material of MSW was collected from different point sources of the city, Faisalabad (Pakistan), mixed and divided into bio-degradable and non-biodegradable portions. The biodegradable portion was air dried and ground with the help of grinder. Analysis for NPK, OM and C:N ratio of the waste was conducted. Then it was inoculated with effective microorganisms (EM) solution to convert these wastes into biofertilizer. The waste was taken into plastic pots and inoculated with 1%, 5% and 10% EM solutions for the period of fifteen days. The treatments were repeated thrice. In order to know the effect of EM the waste was again analyzed for NPK, organic matter, organic carbon and C:N ratio and was named as EM-Biofertilizer. The results of the experiment indicated that the inoculation of effective microorganisms had promising effect on the availability of nutrients. The amount of nitrogen (N), phosphorous (P), potassium (K), organic carbon (OC) and organic matter (OM) was improved with respect to control/compost (without EM). EM application improved the nutritional status of the
organic wastes due to anaerobic decomposition process. The C: N ratio was lower in EM-Biofertilizer than organic matter alone due to free living bacteria in EM inoculum which promoted the availability of nutrients especially the N contents in the manures. The organic matter which was not inoculated with EM resulted to higher degradation process due to aerobic respiration but the EM inoculum inhibited the oxidation way of decomposition, encouraging the process of fermentation. The results revealed that maximum nitrogen was obtained with 5% treatment of effective microorganisms as compared to other inoculation levels after 15 days. Microorganisms enhanced the decomposition process of the organic waste and maintained the availability of essential nutrients to be utilized in soil for sustainable crop production. The EM-Biofertilizer formed in this method was applied to different crops i.e. Cotton (*Gossypium hirsutum*), Okra (*Hibiscus esculentus*), Spinach (*Spinacia oleracea*) and Cowpea (*Phaseolus vulgaris*), to know the effect of EM-Biofertilizer on the productivity of these crops. The application of EM-Biofertilizer had significant effect on the growth and nutrient concentrations in plants and soil. There was positive correlation between NPK concentrations in leaves of the crops. In case of cotton, on relative basis there was 23% increase in cotton boll size in the EM-Biofertilizer treatment as compared with soil alone and 7% increase as compared to compost i.e. without EM treatment. The same trend was noted in case of okra, spinach and cowpea where with the application of EM-Biofertilizer, the increase in fruit length, leaf length and pod size was 53%, 46% and 60%, respectively as compared to control (soil only) where as 22%, 41% and 46% respective increase was noted as compared to compost (without EM-Biofertilizer). There was significant effect of EM-Biofertilizer on plant height, and number of bolls of cotton, the increase was 50% and 53% respectively with respect to compost alone. Same positive effect of EM-Biofertilizer was also noted in the increase of number of plants (33%), number of fruits (39%), fruit length (22%) and biomass (15%) as compared to control (without EM-Biofertilizer) in case of okra. There was significant effect in the
increase of leaf length (41.24%) and biomass (5%) in case of spinach with the application of EM-Biofertilizer. This was because of the availability of plant nutrients, especially the N and P which are the main growth limiting nutrients. The increase in number of grains (67%), grain weight (50%) and pod size (46%) was also recorded in case of cowpea, on the EM-Biofertilizer treatments as compared to compost (without EM). The percentage increase in nitrogen, phosphorus, potassium and organic matter in soil (for cotton) i.e. 29%, 14%, 18% and 78% respectively, was observed with the treatment of EM-Biofertilizer. Same trend in increase of soil NPK and OM was noted in 5% EM-Biofertilizer treatment in the case of spinach, okra and cowpea. The correlations were developed, which indicated that N, P, K in leaves and OM in soil have positive correlation with the yield of all these crops. The above results clearly depicted that the soil quality parameters i.e. NPK and OM can be sustained through the use of EM technology. Tonnes of solid waste produced daily in Pakistan can be treated with effective microorganisms (EM) for producing EM-Biofertilizer for crop production reducing pollution and creating healthy environment.
Chapter 1

INTRODUCTION

Soil and water pollution is related to the current increase in solid waste generated by urbanization and industrialization (Idris et al., 2004; Emery et al., 2006; Kapepula et al., 2007). Solid waste is defined as the organic and inorganic waste materials produced by different processes and have lost their value in the eyes of their owner (Rushton, 2003; Ghosh, 2004). Solid waste management has become a major issue in large urban areas, and the poor waste management in south Asia which affects human health (Mor et al., 2006). Solid waste production in major cities of Pakistan is estimated to be several thousand tonnes / day (Hussain, 1996) and 1570 tonnes / day in Faisalabad city only. Most of solid waste is disposed off along the road sides, and is burnt in open air (Nisar et al., 1998).

Urban waste disposal in advanced countries is through landfill system (Bilgili et al., 2007) which cost high, and additional area for waste disposal filling pits is required (Sharholy et al., 2007). Municipal solid waste (MSW) consist food waste, vegetables, paper, wood, plastics, glass, metal, and inert materials containing nitrogen (N), phosphorous (P), potassium (K) and organic carbon (Mor et al., 2006). Besides dumping or sanitary land filling, the final disposal of solid waste can be carried out by other methods like incineration and composting (Ghosh, 2004; Burnley, 2006). Accumulation of wet waste causes bad odour and
it may contaminate soil, air and ground water. Continued waste burning may cause air pollution due to the smoke produced as explosion (Kapepula et al., 2006).

Soils in Pakistan have poor native fertility and the soil organic matter is generally low. Soil applications of organic sources (animal manure, composted farm residue or green manure) have helped maintaining soil organic matter, reclaiming degraded soils and supplying plant nutrients in intensive and exhaustive crop production systems. Improvement in fertility and quality of soil, especially under low input agricultural systems, requires this input of organic materials advocated by many researchers (Stamatiadis et al., 1999; De Jager et al; 2001; Ouedraogo et al., 2001; Palm et al., 2001; Soumare et al., 2003)

Handling large volume is one of the problems in soil application of the municipal waste material. Solid wastes need to be composted to reduce the volume and to convert it in an organic-rich product. Decomposition of organic waste is a complex issue and is controlled by C:N ratio, chemical nature and several other factors (Hadas and Portnoy, 1994). During composting, organic compounds are transformed through successive activities of different microbes to more stable and complex organic matter (Tremier et al., 2005). The N concentration of the compost and its C:N ratio affects the dynamics of mineral N in the soil (Trinsutrot et al., 2000).

Use of effective microorganisms (EM) inoculum along with organic materials is an effective technique for stimulating supply and release of nutrients
from these nutrient sources. Studies have shown that the inoculation with EM cultures can improve soil and crop quality (Higa and Parr, 1994; Hussain et al., 1999). Similarly, Daly and Stewart (1999) reported that application of EM to onion, pea and sweet corn increased yield by 29%, 31% and 23%, respectively. Higa and Wididana (1991) stated that EM is not a substitute for other management practices but is an additive for optimizing all other amendments and practices used for crop production. When used properly EM has been shown to enhance the beneficial effects on crop growth and yield (Minami and Higa, 1991).

The organic materials inoculated with EM are degraded through the fermentation process (Higa and Kinjo, 1991). The possibility of using this fermentation process (slow decomposition) in a soil environment has been of great interest. Manure fermented with EM increased the yield of a variety of fruits, vegetables and crops, compared with other fertilizers containing equal nutrient elements (Liang and Deyou, 2000).

Integrated recycling system of organic urban waste provides a solution for management of urban wastes (Wididana and Higa, 2000; Kishore, 2000; Shao et al., 2001) as application of EM increases the microbial diversity and can enhance decomposition of waste.

The objectives of the study were to:

- Prepare biofertilizer for soil application with EM inoculum using municipal solid waste as the substrate to overcome pollution hazards.
- Test the prepared biofertilizer for crop yield using several test crops.
- Determine soil chemical changes after the application of biofertilizer from Municipal solid waste.
Chapter 2

REVIEW OF LITERATURE

Rapid urbanization and population growth increases solid waste in cities, and its disposal has become a problem in large urban centers. Municipal solid waste has dominant organic component, and it should be recycled from an ecological and economical point of view (Erhart, et al., 2005). The addition of plant residues, manure or compost to soil, helps improve soil quality, and reduce erosion by maintaining soil organic carbon (Karlen et al., 1992). Compost application to crop lands increases crop yields by enhancing soil fertility (McSorley and Gallaher, 1996; Mamo et al., 1998), and soil physical environment e.g. soil permeability, plant available water holding capacity, and air-filled porosity (Keener et al., 2000). Compost also improves soil structure and organic matter content and nutrient supply to plants like NPK (Giusquiani et al., 1995). Therefore, it reduces the input of mineral fertilizers in conventional agriculture by providing necessary nutrients. However, due to low decomposition rate of the organic material, and low fertility status of the soils where application can be made, its’ prior composting is suggested to increase nutrient availability and decrease C:N ratio (Stratfon and Rechcigl, 1998). Composting of solid organic waste is viable option for recycling as it reduces mass by dewatering and volume by decomposition (Marchettini, 2006).
Subsistence crop production has lead to soil organic matter loss, and soil structure degradation resulting poor plant growth (Golchin et al., 1995). Decline in soil organic matter is due to carbon loss through oxidation, and erosion as intensive cropping are not compensated by carbon inputs through the return of plant biomass (Grant, 1997). According to Smith et al., (1993), decrease in SOM is due to increased oxidation after cultivation, to tillage operations, and to erosion of top soil rich in SOM. Soil organic matter concentrations are declining in an intensive arable rotations and the loss of organic matter from soil results in increased soil erosion (Houghton, 1996). Soil application of compost from organic residues presents a management strategy that could counteract depletion of organic matter in soils. The treatment of waste in recycling process and its application in soils may help solve the problem of residue accumulation and environmental pollution (Marcotea et al., 2001; Tejeda and Gonzalez, 2003).

Nitrogen immobilization can be expected when C:N ratio organic matter is above 30 (Kumar and Goh, 2003) under decreased N release. Once the easily decomposable forms of C have been decomposed, the decomposition rate slows down, and later as the microbial population reaches death phase, the N in microbial biomass is released as plant available nitrogen. Therefore, during decomposition both the forms of C in the residue and the C/N ratio are expected to have a significant impact on the timings of N immobilization (Bending et al., 1998). Biological stability of the treated waste for low-emission disposal cannot
be reached by anaerobic digestion alone, but only in combination with additional aerobic post treatment (Fricke et al., 2005).

The compost applied to soil must have an adequate degree of maturity, which implies stable organic matter content free from phytotoxic compounds, and plant and animal pathogens (Bernal et al., 1998; Gomez, 2001). Mineral fertilization provides readily available nutrients for plant growth; but does not contribute to improve soil physical condition. Compost is a steady source of nitrogen. Application of sufficient organic matter is necessary for steady high production levels of crops and sustainable agricultural development (Hassink et al., 1997, Sun et al., 2003; Williams et al., 2005). Organic matter application, in addition to supply of nutrients, improves soil aggregation, and stimulates microbial diversity and activity (Shiralipour et al., 1992; Carpenter et al., 2000). Organic matter addition improves the soil humus contents, cation exchange capacity, aeration, water holding capacity and water infiltration rate (Bhagat and Verma, 1991; Ferreras et al., 2006), and long term productivity and sustainability of soils. The soil cation exchange capacity (CEC) was increased from 4 to 6 C mol kg$^{-1}$. Soil pH was also increased by applying compost (Ouedraogo et al., 2001).

Composts are widely used in crop production as source of organic matter. Organic matter and polysaccharides synthesized by the microbial community in compost can have positive effects, like increase in aggregation of soil particles, and resistance to compaction (Amellal et al., 1998; Celik et al.,
2004; Parkinson et al., 1999). Plant nutrients significantly affect the plant growth and ecosystem productivity, with nitrogen (N) and phosphorous (P) generally being the main growth-limiting nutrients. N is stored in soil primarily in organic matter, from which it is mineralized to ammonium-N by the action of enzymes produced by soil organisms. Organic farms rely heavily on soil biological activity to provide N to plants, given the restrictions on adding fertilizer-N. Farmers producing organic products, therefore, must seek to maintain or enhance the N status of soil organic matter to ensure that N supply meets plants needs (Condron et al., 2000; Berry et al., 2002). The application of animal manure had the consistent and biggest effect on yield (Badaruddin et al., 1999). The experiments revealed that it is possible to save 50% mineral N and improve the yield of crop if proper combination of mineral organic N and or/biofertilizer (Azotobacter) is applied (Lampe, 2000; El-Hawary et al., 2002; Idris, 2003).

2.1 Composting Processes

Composting of the organic fraction of solid waste is aerobic or anaerobic decomposition, and the difference lies in the nature of end products (Ponnampuruma, 1972). Aerobic process is a biological stabilization (Komilis et al., 1999) where release of CO₂ is a measure of decomposition rate (Webster et al., 1998; Wang et al., 1999). Anaerobic composting is under anaerobic conditions where less oxygen is available, and methane (CH₄) is dominant C containing end product, although CO₂ is also produced. Since methane has 23
times higher global warming potential compared to \( \text{CO}_2 \) (IPCC, 2001) releasing \( \text{C} \) into the atmosphere as methane is environmentally harmful.

In aerobic composting the degradation of organic matter and its influence on biological stability can be achieved within 4 week with 80\% organic matter degradation. This procedure requires appropriate mechanical processing. An intensive aeration and an optimum control of water content, temperature and oxygen supply enhance decomposition. The aerobic composting has higher microbial activity than anaerobic (Vencatasawen et al., 2000). Aerobic land filling has been on the increase during recent years, and many large scale and field scale studies have been recently undertaken (Bilgili et al., 2007). Depending on the specific requirement and waste characteristics, mechanical processing including sieving and grinding become essential.

Anaerobic composting is the decomposition of organic wastes in the absence of \( \text{O}_2 \), releasing \( \text{CH}_4, \text{CO}_2, \text{NH}_3 \) and traces of other gases and organic acids (Moldes et al., 2007). A series of microbial populations carry out this process to completion, the most important of which are methane-producing bacteria which are sensitive to several environmental factors including availability of nutrients, pH and temperature (El-Fadel and Sbayti, 2000). Anaerobic composting has advantage over aerobic composting as it is less labour intensive, produces energy and contributes less to the green house effect. About 20 composting plants are operating with anaerobic systems throughout Europe (White et al., 1995).
Anaerobic decomposition process is carried by primary and secondary microorganisms. The primary colonizers convert the complex carbohydrates and protein into simple carbon sources. The genus clostridium, predominant in anaerobic surroundings, degrades cellulose to organic acids (acetic, formic, lactic, succinic and butyric acids) and alcohols. Methane bacteria such as *Methanococcus, Methanosarcina, Methanobacillus* and *Methanobacterium* which are secondary colonizers break down organic acids into CH$_4$ and CO$_2$. The following typical reactions have been reported occurring in the pathway of methane production (Barker, 1956; Alexander, 1977)

1. $\text{CO}_2 + 4\text{H}_2 \rightarrow \text{CH}_4 + 2\text{H}_2\text{O}$
2. $4\text{HCOOH} \rightarrow \text{CH}_4 + 3\text{CO}_2 + 2\text{H}_2\text{O}$
3. $\text{CH}_3\text{COOH} \rightarrow \text{CH}_4 + \text{CO}_2$ and
4. $2\text{CH}_3–\text{CH}_2\text{OH} \rightarrow 3\text{CH}_4 + \text{CO}_2$

Anaerobic digestion can be ecologically advantageous than aerobic digestion because of exhaust emissions and energy balances (Frick *et al.*, 2005). There is more loss of nutrients due to exposure to air in aerobic composting than anaerobic (Bargyla and Rateaver, 1993).

### 2.2 EM Technology

Effective microorganisms (EM) is a consortium of beneficial microorganisms developed in Japan at early 1980’s, and is observed to enhance productivity of conventional organic farming system (Higa, 1991; Hussain *et al.*, 2002). The culture solution contains lactic acid bacteria, photosynthetic and N-
fixing bacteria, yeast, ray fungi and molds, and it comprises of five families, 10 genera and 80 species (Hussain and Higa, 2001). These microorganisms are mutually compatible, and coexist in culture for extended periods (Higa and Wididana, 1991a; Higa, 1991).

EM has potential to utilize municipal waste / city garbage, and convert it into useable compost within 10-15 days (Hussain et al., 2002). It has the ability to breakdown organic matter; thereby producing plant nutrients and enhancing physical and chemical properties (Yadav, 2002; Tejada et al., 2003). Integrated recycling of urban organic waste with EM technology has been demonstrated (Wididana and Higa, 1999). It was found a profitable business which could be managed professionally with 150 tonnes of urban waste/day. EM reduces the malodor from animal and poultry farms, and enhances the stability and value of animal wastes as organic fertilizer. It stimulates the process of organic matter degradation under different conditions (Nissanka et al., 1998; Sudrajat, 1998). Li and Ni (1999) studied the use of Effective Microorganisms to suppress malodors of poultry manure (PM). EM suppresses the growth and activity of the indigenous putrefactive types that cause malodors in the manure and transform protein and amino acid into NO$_3$-N and NH$_4$-N. Joo and Lee (1991) conducted live stock manure recycling in Korea by anaerobic digestion accounting for pollution problems of rural areas mainly because of increased farm animal pollution and inadequate management of manure.
When EM was applied in combination with organic sources viz., compost, chicken dung, green manure, it showed higher yield. It had thus a beneficial effect of enhancing the decomposition and mineralization process resulting in the availability of more nutrients (Ting et al., 1995, Zhengao et al., 1995). EM application prevents problems which may exist in handling organic urban waste with an expectation to reduce heat and bad odor, reduce fly population and reduce pathogen microorganisms in piling of trash. Integrated recycling system of organic urban waste provides a solution for management of urban wastes (Wididana and Higa, 2000).

2.3 Soil Organic Carbon Dynamics

The bulk of the solid matter in manure is composed of organic compounds and partially degraded plant material (Jarvis et al., 1996). C accounts for about 30% at the total dry matter content of manure, (Rasmussen and Collins, 1991). Manure addition to soils can therefore; result in substantial C inputs and significant increase in organic C levels of soils. In this regards several researchers observed increase in organic C and other substantial residual effects as a result of manure additions (Johnston, 1986; Gupta et al., 1996).

Soil organic matter (OM) is a complex mixture of living, dead and decomposing materials, and inorganic compounds. Almost 15% of it is identified as polysaccharides, poly peptides and phenols (Alexander, 1977). This includes 20% carbohydrates, 20% amino acids and amino sugars, and 10-20% aliphatic
fatty acids (Paul and Clark, 1989) and the remaining of SOM is humic material, a dark amorphous substance derived from transformation of organic residues.

Biomass production is in equilibrium with nutrient reserves in natural systems while agricultural systems are characterized by continual loss and net removal of nutrients (Nair, 1996). According to Hendrex et al., (1992) flow of nutrients in and out of agricultural systems is usually much greater, with lower nutrient storage capacity and less nutrient cycling efficiency. The sustainability of agricultural system is greatly dependent on creating the balance between inputs and outputs of nutrients. One way of achieving this balance is by ensuring that nutrients removed from the soil are returned to the system. In this manner, a nutrient cycling mechanism can be established that is essential for the sustainability of the production system (King, 1990).

Soil organic matter (OM) plays a fundamental role to maintain the main soil properties and regimes related to the soil fertility. The whole functioning of soils is greatly influenced by soil OM, its ability to provide conditions for plant growth, soil biota functioning, reduction in greenhouse gases, modification of pollutants and maintenance of soil physical condition (Eswaran et al., 1993; Shevtsova et al., 2003; Ni et al., 2004). Therefore, it is necessary to maintain sufficiently high levels of organic matter as a prerequisite for steady, high production levels of crops and sustainable agricultural development (Sun et al., 2003; Williams et al., 2005). Under certain bioclimatic conditions and creation cropping systems the soil organic matter was found fairly constant (Cai et al.,
A number of studies have also indicated that for agricultural soils with unaltered management practices soil OM content could reach an equilibrium state over time (Liang et al., 2000; Peterson et al., 2002; Loveland and Webb, 2003). In order to attain equilibrium state, soil OM content could increase or decrease, depending on whether the original content was below or above the steady-state level (Yang and Janssen, 1997; Guo et al., 2004).

In the tropics organic resources play a dominant role in soil fertility management through their short-term effects on nutrient supply and long-term contribution to soil organic matter (SOM) formation. Although organic resources used alone offer insufficient nutrients to sustain crop yield and build soil fertility (Palm et al., 1997). They will continue to be a critical nutrient source as small holder farmers in the tropics are unable to access adequate quantities of mineral fertilizers.

De Datta and Hundal (1984) reported that in crop-rating organic matter into the soil improves its structure through increased aggregation, which favorably influences tilth, and root penetration. In silty soil organic matter decreases slaking sensitivity, whereas in wet clay soil, it increases resistance of the soil mass to plastic deformation. Adding organic matter in soil, it may alter soil shrinkage properties, improve tilth, and reduce draft requirements for preparing the seed bed for dryland crops. Kumazawa (1984) advocated that the immobilization and mineralization of N in soil can be regulated by the addition of OM to improve the N absorption process of the rice plant and to attain higher yields. The addition of
compost accelerates the development of active rice roots that carry out nutrient absorption and thereby promote leaf activity. The increase of C, N, and available P and K indicates that long term compost application increases fertility of the soil.

Singh et al., 1987, found that poultry manure applied at 60, 120 and 180 kg N ha\(^{-1}\) produced rice grain yield equivalent to that of 37, 96 and 168 kg urea N ha\(^{-1}\), respectively. On the basis of N uptake, poultry manure N was 80% as efficient as urea N at all rates of application in the study. No significant difference in uptake P and K by rice was observed between urea and poultry manure. A residual effect of poultry manure N was shown by wheat grown after rice only at the highest rate of application. The residual effect of the manure in supplying P to wheat was also conspicuous.

Frequent additions of organic materials such as animal manure, green manures, crop residues and multi-year sod root systems are important for maintaining soil organic matter, besides their direct influence on availability of N or other nutrients (Mhagdoff, 1991). Good organic matter management helps improve or maintain soil structure, thereby aiding water storage, drainage and root penetration. It also improves chemical aspects of soil fertility by providing greater CEC to retain nutrient cations by chelating micronutrients to keep them available and by buffering soil pH against rapid changes. Lastly, soils with regular application of fresh organic matter maintain a large and diverse population of organisms, which result in fewer problems with plant diseases and insects.
The applications of poultry manure at the rate of 50 Mg ha\(^{-1}\) produced positive results in decreased bulk density, and increased total and macro porosity, volumetric water retention and available water capacity, saturated hydraulic conductivity, infiltration capacity, cumulative infiltration and time to reach infiltration capacity, more than the inorganic fertilizers did (Mbagwu, 1992). Their effects were more pronounced at three months than three years after application.

Ecosystem productivity and plant growth are significantly affected by the availability of plant nutrients with nitrogen N and phosphorus P generally being main growth limiting nutrients. Nitrogen is stored in soil primarily in organic matter, from which it is mineralized to ammonium N by the action of enzymes, produced by soil organisms. Organic farms rely mainly on soil microorganisms activity to provide N to plants, given the restrictions on adding fertilizer-N (Parfitt et al., 2005). Organic producers must seek to maintain or enhance the N status of soil organic matter to ensure the needs of plant by supplying nitrogen (Berry et al., 2002).

### 2.4 Soil Microbial Biomass

Soil is a C-limited environment. The increased biomass activity and abundance of soil microbes is owing to the supply and availability of additional mineralisable and readily hydrolysable C due to manure application (McGill et al., 1986; Ocio et al., 1991). Microbial biomass C of a Typical Haplustalf manured for 45 years with cow dung showed an increase of more than 76% over
the unmanured control in the Northern Guinea Sarana zone of Nigeria (Goladi and Agbenini, 1997). A number of other studies, conducted have shown significant increase in microbial biomass C, N and P in soils receiving manure (Hasebe et al., 1985; Collins et al., 1992; Goyal et al., 1992). The positive impact of manure addition on the density of microbes was demonstrated in the long-term field plots at Sanborn. Soils, which were supplied with manure, had high numbers of bacteria and actinomycetes than those that did not (Martyniuk and Wagner, 1978).

During decomposition, microorganisms assimilate complex organic substances for energy and carbon (C), and release inorganic nutrients. This process is controlled by temperature, moisture, soil disturbance and the quality of SOM as a microbial substrate. These factors, together with the size and activity of the microbial population are responsible for the decomposition and nutrient release (Smith et al., 1993). They further observed that decrease in SOM is due to increased oxidation after cultivation, to tillage operations, and to erosion of topsoil rich in SOM.

Potential productivity of soil is directly related to SOM concentration and turnover. Soil microorganisms are the driving forces behind SOM transformations, such as mineralization and immobilization of organic constituents. These transformations are the basis of plant decomposition soil aggregation, soil tilth and nutrient availability. By observing microbial mediated transformations, we can describe specific processes on a pool sized basis, and estimate which processes are important on an ecosystem level.
For many years, the scientists have been investigating the beneficial effects of microbes, their activities such as biological nitrogen fixation, organic matter decomposition, mineralization, nitrification antagonism and fermentation. Among other means of alternative agriculture for achieving the goal of sustainability and good quality soil, one popular in a number of countries, is through the use of Effective Microorganisms (EM), a liquid culture formulated by Prof. Teruo Higa of Japan during 1980s.

2.5 Biodegradation of Soil Organic Matter

Microorganisms assimilate complex organic substances for energy and carbon (C), during the process of decomposition and release inorganic nutrients. This process is controlled by temperature, moisture, soil disturbance, and the quality of SOM as a microbial substrate. These factors, together with the size and activity of the microbial population are responsible to regulate the rate of decomposition and nutrient release (Smith et al., 1993). They further stated that decrease in SOM is due to increased oxidation after cultivation, to tillage operation, and to erosion of topsoil rich in SOM.

As far as the organic matter (OM) conservation in the soil is concerned, it is important to understand the process responsible for SOM degradation and their control through microbial manipulation. Generally, there are two principal ways of OM decomposition viz oxidative and fermentative. Organic matter degradation is done mainly in a biochemical function known as fermentation (Doelle, 1969; Galantini and Rosell, 2006).
Biodegradation is the breakdown of compounds using any living entity like microorganisms, plants and earthworms etc. These microbes are often excessively use artificially for this processes as they use contaminants as a food source, thereby completely eliminating toxic compounds by changing them into basic elements such as carbon dioxide and water, a process known as mineralization. Incomplete degradation may also occur, or the partial breakdown of the original contaminant to a less complex form. Sometimes immobilization of a compound occurs where the agent is overcome by the microbe but not eliminated or altered, which is often a potential benefit but rarely a final solution.

The need of the hour is to improve soil health by providing the much needed organic matter, least soil become impoverished. The scope and potential for recycling variety of resources in agriculture is vat by any standards. Wastes recycling can bring tremendous benefits to agriculture and land management in long run. In addition there are the benefits of a cleaner environment, a healthier habitat and an intelligent use of all available recyclable resources without condemning them as wastes. Towards this end urban solid waste compost could serve as a valuable organic matter source given the shortage of organic nutrient source (Prakash et al., 2007).

Defining quality standards for organic manures is a very difficult task given the heterogeneity of residues that occur in city wastes and processing methods adopted. Integrated nutrient management combining both inorganic and organic resulted in wholesome improvement of the soil. Faced with such situation
utilizing valuable urban resources for manure production would be viable alternative given the ever increasing urban status resulting in urban waste production.

This can be done by adopting the technology of “biodegradation” (Xi et al., 2005). The term biodegradation refers is a biological process in which organic urban solid waste material is broken down by the action of microorganisms. The degradation process takes place in the presence of air (aerobic) and results in elevated process temperature and the production of CO₂, water and stabilized organic residue. The key feature of the biodegradation/composting process is the generation of heat by biological activity during the decomposition of the substrate materials. By forming the waste into large masses under appropriate conditions, they will reach high temperature, resulting in rapid degradation. More importantly, these temperature have a sanitizing effect upon the waste, reducing the numerous of pathogenic organism (Iyenger and Bhave, 2005). The proposed study attempted to proper management, physicochemical analysis of Municipal Solid Waste (MSW) and its conversion to enrich compost by eco-friendly process.

Municipal solid wastes can be managed using main physical, chemical, and biological techniques. Most widely used physical or chemical methods include recycling and combustion of piles of solid wastes (Ting et al., 1996). However, these technologies are either costly or do not environmentally friendly and have potential of risk for environment and human health. On the other hand, biological approach (biodegradation) appears to be among the most promising methods. The technology is also environmentally sound since it simulates natural
processes and can result in the complete removal of bulk of solid municipal wastes especially those organic in nature and have potential to convert these wastes into innocuous and valuable products.
Chapter 3

MATERIALS AND METHODS

The investigations reported in this manuscript were carried out during 2003 to 2006 at the Institute of Soil and Environmental Sciences, University of Agriculture, Faisalabad, Pakistan. This section presents methodology to make biofertilizer from municipal waste, and measurement of effects of its use on crop and soil composition.

3.1 Preparation of Bio-Fertilizer

Samples of solid waste were collected from (i) dumping station of Tehsil Municipal Administration (TMA), Faisalabad, (ii) Faisalabad city vegetable and fruit market, and (iii) a retail selling market of vegetables and fruits, Jhang Bazar, which contained fruit juice waste and pulp in particular.

The material collected from these three different locations was thoroughly mixed. The non-degradable components were separated by hand picking. The remaining biodegradable portion was air dried and ground to pass through 2 mm sieve by a mechanical grinder. The ground material was analyzed for total N, P, K, and organic carbon (OC) contents. The ground waste was taken in plastic pots and inoculated with 0, 1%, 5% and 10% (v/v) EM solution in three replications. Initial temperature was 18.5 °C, water content 44%, and pH 6.8. The incubation was carried out for 15 days at constant room temperature of 25 °C. The material composted with EM was named as EM-biofertilizer and that without EM
as Compost (or EM0), the MSW without EM inoculum. EM-biofertilizers were further differentiated by their names derived after concentrations of EM-solution used e.g. EM1-Biofertilizer, MSW inoculated with 1% EM solution; EM5-Biofertilizer, MSW inoculated with 5% EM solution; and EM10-Biofertilizer, MSW inoculated with 10% EM solution.

After the completion of incubation, pH, temperature and water content were measured Table 1. The composted waste was analyzed for total N, P, K and organic carbon content. Organic matter and C/N ratio were calculated. Nutrient composition and physico-chemical characteristics of the composts are presented in Table 2.

**Table 1. Physical characteristics of compost (un-inoculated) and EM-biofertilizer**

<table>
<thead>
<tr>
<th>Treatments</th>
<th>pH</th>
<th>temperature</th>
<th>moisture</th>
</tr>
</thead>
<tbody>
<tr>
<td>Compost (EM0)</td>
<td>6.80</td>
<td>18.5</td>
<td>44</td>
</tr>
<tr>
<td>EM1-Biofertilizer</td>
<td>3.96</td>
<td>23.0</td>
<td>36</td>
</tr>
<tr>
<td>EM5-Biofertilizer</td>
<td>3.71</td>
<td>26.0</td>
<td>27</td>
</tr>
<tr>
<td>EM10-Biofertilizer</td>
<td>4.11</td>
<td>22.3</td>
<td>35</td>
</tr>
</tbody>
</table>

**Table 2. Composition of the EM-biofertilizer prepared after inoculating with different concentration of EM solution.**

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N</th>
<th>P</th>
<th>K</th>
<th>OM</th>
<th>OC</th>
<th>C:N</th>
</tr>
</thead>
<tbody>
<tr>
<td>EM0-biofertilizer</td>
<td>1.36</td>
<td>0.40</td>
<td>1.35</td>
<td>93.18</td>
<td>54.04</td>
<td>39.93</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>1.69</td>
<td>0.53</td>
<td>1.76</td>
<td>94.21</td>
<td>54.70</td>
<td>32.69</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>1.94</td>
<td>0.90</td>
<td>2.04</td>
<td>94.86</td>
<td>55.01</td>
<td>29.82</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>1.70</td>
<td>0.53</td>
<td>1.90</td>
<td>94.07</td>
<td>54.44</td>
<td>32.34</td>
</tr>
</tbody>
</table>

Each value is average of three replications.
3.2 Soil Analysis

Before the start of each experiment and after harvest of each crop, soil samples were collected and analyzed for NPK and organic matter. The determinations were made according to the methods described by Richard (1954) except otherwise mentioned.

3.2.1 Soil physico-chemical analysis

Soil was taken from 0-20 cm depth and analyzed for different physical and chemical characteristics prior to use for laboratory and greenhouse experiments. The description for methodologies of the analytical procedures is as follows: Soil texture was determined by dispersing soil samples in sodium hexametaphosphate, and particle size distribution was determined on settling rate basis measured by using a hydrometer (Bouyoucos, 1962). Soil textural class was determined using International Triangle System of Textural Classification. Saturated soil paste was prepared by taking 250 g soil in a plastic beaker and adding sufficient distilled water while continuously stirring the paste with a spatula. Saturated paste was allowed to stand for one hour to attain equilibrium, and pH was recorded. Clear extract of the soil paste was obtained by using filter press assembly, and electrical conductivity of the extract was recorded.

Soil Saturation Percentage (SP) was calculated from the water loss at 105°C determined by oven drying a portion of saturated paste to a constant weight:
\[
Saturation\% = \left(\frac{W_1 - W_2}{W_2}\right) \times 100
\]

Where as \(W_1\) is mass of saturated paste before oven drying and \(W_2\) is mass of the dried soil paste.

3.2.2 Soil chemical analysis

Total nitrogen, available P and K, and organic matter were determined before start of each experiment and after harvest of each crop.

3.2.2.1 Total nitrogen

Ten gram soil was digested in the presence of digestion mixture (\(K_2\)SO\(_4\): CuSO\(_4\): Se metal powder in 10:1:0.1 ratio) in 30mL H\(_2\)SO\(_4\) on Kjeldahl’s digestion apparatus. The digested material was allowed to cool and diluted to 250 mL. Ammonia was diluted and received in 4% boric acid solution, and titrated against standard 0.1 N H\(_2\)SO\(_4\) using bromocresol green and methyl red indicator (Jackson, 1962).

3.2.2.2 Available phosphorous

Plant available P was extracted in 0.5M NaHCO\(_3\) (pH 8.5). Phosphorous concentration was measured using colour developing reagent (Ascorbic acid, ammonium molybdate, antimony potassium tartrate and concentrated H\(_2\)SO\(_4\)). Intensity of blue colour was read on Spectrophotometer at 880 nm after 15 minutes (Watanabe and Olsen, 1965).
3.2.2.3 Available potassium

Available K was determined by extracting K with neutral 1N ammonium acetate, and K in the extract was determined by a flame photometer (Method-18: U.S Salinity Laboratory Staff, 1954).

3.2.2.4 Organic matter

To one gram of soil sample, 10 mL of K_2Cr_2O_7 was added along with 20 mL of H_2SO_4 (commercial) and left the flask for 30 minutes after stirring. Then added 200 mL of water, one gram of sodium fluoride and one mL of diphenylamine as indicator to the flask and titrated this solution with 0.5 N solution of FeSO_4 with light green end point. Organic matter %age was calculated by using the formula:

\[ \text{O.M \%age in soil} = \frac{\text{mL of } K_2Cr_2O_7 \text{ reduced}}{\text{g sample}} \times 0.69 \]

3.3 Plant Analysis

3.3.1 Total nitrogen

For total nitrogen determination the plant samples were digested in Kjeldals flask on electric heater in the presence of 10g digestion mixture of K_2SO_4: CuSO_4: Selenium metal (10:1:0.1) and 30 mL of H_2SO_4. Allowed to cool and then made the volume up to 100 mL. From this solution 10 mL aliquot was taken and distilled for ammonia, which was then received in the conical flask containing 4% boric acid and colour developing reagent (bromocresol and methyl red). This solution of each sample was then titrated against 0.1N H_2SO_4 up to golden yellow end point (Jackson, 1962).
3.3.2 Total phosphorous

One gram of material was taken and digested in the presence of concentrated HNO₃ and 72% HClO₄ (3:1) acid mixture (10 mL of acid mixture was taken for each sample). Then these samples were put on the hot plate and digested till colourless. After this the samples were filtered and made to volume 100 mL with the distilled water. From these filtrates 10 mL of aliquots were taken in 50 mL volumetric flask and added 10 mL of colour developing reagent (ammonium vanadate and ammonium molybdate) and made the volume up to the mark. Phosphorous concentration was determined on a Spectrophotometer at 440 nm (Richard, 1954).

3.3.3 Total potassium

The digest taken for total phosphorous was used to estimate K concentration on a flame photometer (Richard, 1954).
Table 3. Soil physico-chemical properties used for experiments

<table>
<thead>
<tr>
<th>CHARACTERISTICS</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>pH</td>
<td>7.69</td>
</tr>
<tr>
<td>ECe (dSm⁻¹)</td>
<td>0.40</td>
</tr>
<tr>
<td>Saturation percentage (%)</td>
<td>55.07</td>
</tr>
<tr>
<td>Organic matter content (%)</td>
<td>0.45</td>
</tr>
<tr>
<td>Total nitrogen (%)</td>
<td>0.04</td>
</tr>
<tr>
<td>Available phosphorous (mgkg⁻¹)</td>
<td>6.68</td>
</tr>
<tr>
<td>Available potassium (mgkg⁻¹)</td>
<td>98.44</td>
</tr>
<tr>
<td>Sand (%)</td>
<td>63.85</td>
</tr>
<tr>
<td>Silt (%)</td>
<td>15.95</td>
</tr>
<tr>
<td>Clay (%)</td>
<td>20.20</td>
</tr>
<tr>
<td>Soil texture class</td>
<td>Sandy loam</td>
</tr>
</tbody>
</table>

3.4 EM-biofertilizer Efficiency for Cotton Crop

Cotton was cultivated in July 2004 in the greenhouse in pots containing 12 kg soil in each pot. The seed of cotton cv NIAB-111 was collected from Nuclear Institute for Agriculture and Biology (NIAB) Jhang road Faisalabad, Pakistan. The treatments of EM-biofertilizer were applied in three replicates. Compost (EM0-biofertilizer), EM1-biofertilizer, EM5-biofertilizer, and EM10-biofertilizer were applied at the rate of 100g per pot, and soil without any amendment was kept as control. The weighed amount of each treatment was thoroughly mixed in soil in individual pot. The pot was brought to saturation by
irrigation with canal water. At field moisture, 10 number cotton seeds were sown in each pot. The plants were thinned to three plants per pot after germination and kept till maturity. The canal water was used for irrigation throughout life span of the crop. At flowering, fully expanded young cotton leaf from each plant was sampled for analyses of total N, P and K in the leaf tissue. At maturity, plant height, number of bolls per plant, boll size and number of flower were recorded. Volume of bolls was calculated by measuring boll diameter with vernier caliper. Soil samples were collected after harvest and analyzed for total N, available P and K, and OM contents in soil.

3.5   EM-Biofertilizer Efficiency for Okra Growth

Effectiveness of EM-biofertilizers was also tested for okra crop grown in greenhouse. The same set of treatments was used with three replications in pots containing 12kg soil in each. 100g of each of four EM-biofertilizers were added, and thoroughly mixed in pots individually as described previously. Soil without any amendment was kept as control. Seven okra seeds were sown in each pot and after emergence three uniform seedlings per pot were raised to maturity. The pots were irrigated with canal water according to crop water requirement. At flowering, okra leaf samples were collected for analyzing N, P and K contents. At maturity, number of fruits, fruit length (cm), and plant biomass were recorded. Soil samples were taken after harvest and analyzed for N,P,K and OM contents.
3.6 **EM-biofertilizer Efficiency for Spinach Growth**

The pots were filled with soil amended with four different EM-biofertilizers at the rate of 100g in each pot as described in previous studies. The plants of spinach (white) were cultivated in pots by sowing five healthy seeds in each pot and were thinned after three weeks of germination to three per pot to keep up to maturity using canal water for irrigation. The leaf samples were taken for determination of N, P and K contents in leaf. At harvesting, the leaf length (cm) and biomass were also recorded. The soil samples were collected at the harvest and analyzed for N, P, K and OM contents in the soil.

3.7 **EM-biofertilizer Efficiency for Cowpea Growth**

The seeds of cowpea (variety; LS-2) plant were sown in pots at the rate of seven seeds in each pot having 12kg soil amended with different EM-biofertilizers as described previously. The plants were thinned to three per pot after germination and were kept up to maturity. The treatments were applied in three replications according to the CRD fashion in greenhouse. At flowering stage, the plant leaf samples were collected for ionic analysis for N, P and K contents in leaf. At maturity, fresh grain weight (g), number of grains, dry grain weight (g) and fruit size were also recorded. After plant harvest, the soil samples from all pots were collected for analyses of N,P,K and OM contents in the soil.

3.8 **Statistical Analysis**

Variance in the crop parameters, and analysis was determined statistically with completely randomized design using MSTATC® software. The treatment
means were compared by Duncan Multiple Range (DMR) Test at 5% probability level. The relation among the various parameters was determined by simple linear regression (Steel and Torrie, 1980).
Chapter 4

RESULTS AND DISCUSSION

4.1 Composition of EM-biofertilizer

The inoculation of effective microorganisms reduced pH and moisture, and increased N, P, K and organic carbon in the EM-biofertilizer compared to the compost, MSW composted without EM solution application (EM0-biofertilizer). The maximum reduction in pH was in EM5-biofertilizer. Maximum N, P, K and OM were also recorded in EM5-biofertilizer followed by EM10-biofertilizer. Compost prepared without EM application had the minimum level of plant nutrients. C:N ratio in EM5-biofertilizer was the lowest followed by the C:N ratio in the EM10-biofertilizer. The compost (EM0-biofertilizer) had the highest C:N ratio.

Plant nutrients (N, P and K) concentration and OC contents in EM-biofertilizer were greater than in the composted material, without EM inoculation. The microbial microorganisms in EM solution have the ability to break down organic matter by fermenting activity, thereby releasing plant nutrients. The inoculation of effective microorganisms increased the degradation and chemical breakdown of organic materials and stimulated the process of mineralization of organic matter (Higa and Kinjo, 1991; Hussain et al., 1999). It is expected that there would be greater release of nutrients to soil-plant system when nutrient
biofertilier is applied in the fields enhancing physical and chemical conditions of the soil (Daly and Stewart., 1999; Yadav, 2002).

Enhanced decomposition and mineralization result in accelerated nutrients availability (Ting et al., 1996, Zhen and Weijiong, 1996). The C:N ratio was generally lower in EM-biofertilizer than organic matter alone due to free living bacteria in EM inoculums which use C as a source of energy and promote the availability of nutrients especially the N contents in the manures (Higa and Wididana, 1991). Uninoculated organic matter with EM has higher degradation process due to aerobic respiration (Haider, 1995). EM inoculums inhibit the oxidation way of decomposition thereby encouraging the process of fermentation (Higa and Wididana, 1991).

4.2 EM-biofertilizer Efficiency and Effect on Soil Health

The EM inoculated municipal solid waste, EM-biofertilizer, was tested for its efficiency on cotton, okra, spinach and cowpea crops. Plant growth and yield under greenhouse, in pot experiments, were also recorded Zhao (1995) reported that the application of EM significantly improved available nutrient’s level in soil, organic matter, total N and lowered the C:N ratio. EM application improved the nutritional status of these organic wastes due to anaerobic decomposition process (Bargyla and Rateaver, 1993). The short-term plant available nutrient values of many crop residues and manure is poor (Delve et al., 2000) but produced the long-term benefits in maintaining soil organic matter (Pale et al., 2000; Palm et al., 2001).
4.3 Cotton Growth

The application of effective microorganisms (EM) inoculum as EM-biofertilizer increased growth of cotton plant (Table 4). Cotton plant height of 42.3 cm was recorded in pots amended with EM5-biofertilizer. The mean value was significantly greater than that of the control and the compost but also that of the plants grown in pots amended with EM1-biofertilizer and EM10-biofertilizer. Compared to the control plants, there was 70% increase in height due to the EM-biofertilizer application and 50% compared to compost. Number of flowers per plant, number of bolls per plant, and boll circumference in the compost and EM-biofertilizer treatments was greater than that of the control. Further, although the EM5-biofertilizer amended pots had generally greater values for all the three parameters but the difference remained statistically non-significant among them.

The increase in growth parameters conformed to the report by Ahmed et al. (1993) who noted the highest percent increase in cotton yield with EM. The soil fertility improved with application of EM for cotton production has also been reported by Yamin, 1994. Haq (1997) studied the enhancing efficiency of farmyard manure (FYM) with EM for cotton production and reported increased symbodial branches, boll weight, and weight of seed per boll.

The maximum number of flowers and bolls per plant, and boll size was recorded in pots amended with EM5-biofertilizer followed by EM10-biofertilizer, and the lowest in control. Yet, the difference in the compost and various EM-biofertilizers was statistically non-significant (less than LSD). Nevertheless, EM-
biofertilizer treatment resulted in 56% more flowers than in control, soil alone, and 19% over the compost alone treatment.

Table 4: Effect of EM-biofertilizer on growth parameters of cotton

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Plant height (cm)</th>
<th>Flowers (plant⁻¹)</th>
<th>Bolls (plant⁻¹)</th>
<th>Boll circumference (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>12.67</td>
<td>1.78</td>
<td>0.33</td>
<td>13.48</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>21.33</td>
<td>3.33</td>
<td>0.78</td>
<td>16.33</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>37.00</td>
<td>3.43</td>
<td>1.33</td>
<td>17.02</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>42.33</td>
<td>4.11</td>
<td>1.67</td>
<td>17.48</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>38.00</td>
<td>4.00</td>
<td>1.44</td>
<td>17.20</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 3.15, 0.94, 1.07, and 2.62 for plant height, number of flowers per plant, number of bolls, and boll size (cm³), respectively.

The findings of Khaliq (2006) also supported the results and indicated that application of FYM with EM increased cotton yield, which was economically more viable than fertilizer application alone. The use of organic manures might be helpful for sustainable crop production and maintenance of soil fertility. The application of FYM can be substituted for N and P (Yaduvanshi, 2003). Similar results on the availability of P with EM application were observed by Bajwa and Jilani (1994). They found increased FA-Mycorrhizal colonization in maize roots by EM application with enhanced P-absorption from soil. Jilani et al., (1996) reported that about half of N and P fertilizers could be saved by applying EM to
wheat crop without any reduction in crop yield. These studies indicate a positive contribution of EM towards NPK nutrition of crops and its availability in the soil.

### 4.3.1 Cotton leaf N, P, K contents

Application of various EM-biofertilizers had significant effect on N, P, and K content of cotton leaf tissue (Table 5). The highest N, P, and K concentration in cotton leaf tissue was highest with the application of EM5-biofertilizer. Nitrogen concentration in control and various EM-biofertilizer treatments was significant at 95% confidence level but the various EM-fertilizer treatments were non-significantly different. Similarly, P and K concentration in control and various EM-biofertilizer treatments was significant at 95% confidence level, and EM-biofertilizer treatments also differed e.g. application of EM5-biofertilizer had more P than EM1-biofertilizer. Rashid et al., (1994) reported that EM resulted in higher N and P up-take by wheat. In this study the total biomass of cotton remained to be recorded but if estimated from the greater plant height, the highest concentration of the nutrient and the highest biomass in the EM-biofertilizer treatment one can suggest that the highest up take was in the EM-fertilizer amended pot. It has already been reported that greater biomass actually lowers nutrient concentration due to dilution effect. The order of cotton leaf N, P, K was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. With EM-biofertilizer application the concentration of N increased in cotton leaf was 36% over the control plants and 14% over the pots amended with EM0-biofertilizer. The difference in total up take will be even
greater. The greater concentration of nutrients may be attributed to greater available nutrients for plant growth released due to decomposition of organic matter under activity of the beneficial microorganisms (Higa and Wididana, 1991; Parr et al., 1994). The applications of EM to make FYM as FYM-Bioskashat build the nutrient status of soil and supply the nutrition to the crop (Hussain et al., 1999; Idris, 2003).

**Table 5. Cotton leaf N, P, K concentration as affected by various EM-biofertilizer applications in potted soil**

<table>
<thead>
<tr>
<th>Treatment</th>
<th>N</th>
<th>P</th>
<th>K</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1.89</td>
<td>0.02</td>
<td>1.49</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>2.53</td>
<td>0.17</td>
<td>1.86</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>2.55</td>
<td>0.14</td>
<td>1.95</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>2.94</td>
<td>0.31</td>
<td>2.32</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>2.89</td>
<td>0.21</td>
<td>2.02</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.73, 0.22, and 0.23 for NPK content, respectively.

**4.3.2 Cotton leaf N, P, K content relation with boll circumference**

Relation of cotton leaf tissue nutrient concentration with cotton circumference is depicted in Figure 1. Cotton circumference increased as the nutrient content increased suggesting a significant positive relation but each element has different slope. Maximum response of cotton boll circumference was with increase in leaf P concentration as indicated by the highest slope depicted
with the regression equations. Leaf N and K had lower slopes but the relation was stronger with boll size. Regardless of slope the of the lines and correlation strength, it is obvious that application of the decomposed residue enhanced N, P, and K contents in plant tissue which in turn increased cotton boll circumference (Contron et al., 2000; Berry et al., 2002; Rathake et al., 2005). Regarding the increase in growth parameters, it was due to the release of plant nutrients to the crops (Higa, 1991) which is evident from the results mentioned. These results were also supported by Magdoff et al., (1997) who reported that organic fertilizer is important not only in evaluation of the nutrient status of the soil by altering the pools and fluxes of C and other nutrients, but also bring about changes in the microbial proportions of the soil.

![Graph showing cotton boll circumference increase with leaf nutrient contents](image)

**Fig. 1** Cotton boll circumference increase, with leaf nutrient contents under biofertilizer application
4.3.3 Soil N, P, K and OM content after cotton harvest

Nitrogen, P, K, and OM remained in soil after cotton harvest are presented in Table 6. The maximum soil N, P, and K were recorded in the pots amended with EM5-biofertilizer. EM-biofertilizer treatments (EM0-, EM1-, EM5, and EM10-biofertilizer) were statistically at par. The lowest value was recorded in the control pots. There was no change in bioavailable soil K and slight decline in P and N in control pots after crop harvest. But there was accumulation of all the three nutrients where EM-biofertilizer was applied as the values after crop harvest were greater than that of original soil. After crop harvest the increase in nutrients clearly depicts the advantages of application of EM decomposed organic matter in the form of biofertilizer. Contron et al. (2000), Berry et al. (2002) and Rathake et al. (2005) reported increased N, which was also supported by (Haynes and Williams, 1993; Sakadevan et al., 1993; Legard et al., 1998).

Table 6: Soil total N, bioavailable P and K, and total organic matter content

<table>
<thead>
<tr>
<th>Treatment</th>
<th>N  (%)</th>
<th>P  (mg kg⁻¹)</th>
<th>K  (mg kg⁻¹)</th>
<th>OM  (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.03</td>
<td>4.02</td>
<td>98.70</td>
<td>0.33</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>0.04</td>
<td>4.49</td>
<td>115.20</td>
<td>0.35</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>0.04</td>
<td>5.05</td>
<td>135.60</td>
<td>0.49</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>0.06</td>
<td>5.23</td>
<td>140.10</td>
<td>1.62</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>0.05</td>
<td>5.12</td>
<td>137.10</td>
<td>0.49</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.01, 0.32, 4.80, and 0.20 for N, P, K, and OM respectively.
4.3.4 Soil N, P, and K relation with boll circumference

Relationship of total soil N, plant available P, and K contents with cotton boll circumference is depicted in the Figure 5. Cotton boll circumference increased with increase in soil P. It had greater slope up to 4.5 mg kg\(^{-1}\) suggesting increase in boll circumference with P within deficient soil. The slope of phosphorous versus boll circumference reduced once soil P level was achieved. However, addition of P had positive effect up to 5.5mg kg\(^{-1}\) as indicated in the figure 2(b). It also implies that N, P, and K are the growth limiting nutrients for plant growth. So we should not compromise on the availability of P as against N and K. Maximum K contents of 140 mg kg\(^{-1}\) soil were recorded in pots which received application of EM5-biofertilizer followed by the receiving of EM10-biofertilizer application, and the minimum K was found in control. Even application of simple compost enhanced bio-available K in soil. Increase in soil K contents is due to EM5-biofertilizer which was 29.5% over the control and 18% over the applied compost prepared without EM solution. Figure 5 also depicts a positive correlation of cotton circumference with the increase in soil K due to application of EM-biofertilizer. Boll grew bigger with increase of K in soil \((r^2 = 1.0)\).
Results and Discussion

Fig. 2(a) Effect of soil Nitrogen on boll circumferences of cotton

Fig. 2(b) Effect of soil P on boll circumferences of cotton
Application of EM-biofertilizer has also significantly increased soil OM (Table 6). The maximum OM was recorded in pots with application of EM5-biofertilizer followed by EM10-biofertilizer and the minimum value was found in control. The treatment of EM5-biofertilizer gave 79% greater OM in soil than that of control and 78% greater than compost alone. The relation between soil OM and boll size was non-significant (Fig. 2d). Troeh and Thompson (1993) reported that higher concentration of K relating to greater yield of crops.
4.4 Okra Growth

Application of EM-biofertilizer had significant effect on the growth parameters of okra plants in greenhouse (Table 7). The maximum numbers of plants were recorded in EM5-biofertilizer application (8.00) followed by EM10-biofertilizer application (6.67), while the minimum value was recorded in control (2.33). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There were 71.0% more plants with respect to control and 33.37% with respect to compost alone as compared with EM5-biofertilizer treatment.
Table 7: EM-biofertilizer application effect on growth of okra

<table>
<thead>
<tr>
<th>Treatments</th>
<th>No. of Plants</th>
<th>Fruits (plant(^{-1}))</th>
<th>Fruit length (cm)</th>
<th>Biomass (g plant(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>EM0-biofertilizer</td>
<td>5.33</td>
<td>4.67</td>
<td>17.91</td>
<td>42.50</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>6.33</td>
<td>5.33</td>
<td>19.90</td>
<td>49.10</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>8.00</td>
<td>7.67</td>
<td>21.86</td>
<td>49.80</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>6.67</td>
<td>5.67</td>
<td>21.29</td>
<td>49.70</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 3.48, 3.22, 6.90, and 18.19 for number of plants, fruits per plant, fruit length, and biomass respectively.

Regarding the fruits, the maximum number of fruits was recorded in 5% application of EM-biofertilizer (7.67) which followed by EM10-biofertilizer application (5.67), while the minimum value was noted in control (2.33). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there were 69% more fruits with respect to control (soil only) and 39.11% with respect to compost alone.

Almost same trend was observed in the case of fruit length. The maximum fruit length was recorded in EM5-biofertilizer application (21.86) followed by EM10-biofertilizer application (21.29), where as the lowest value was recorded in control (10.26). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 53% increase in fruit length with respect to control and 22% with respect to compost owing to the EM-biofertilizer application.
The maximum biomass was recorded application of EM5-biofertilizer (49.80) followed by EM10-biofertilizer application (49.70), while the minimum value was noted in control (32.10). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Comparatively, there was 35% increase in biomass with respect to control and 14.64% increase with respect to compost i.e., without EM-biofertilizer.

4.4.1 Okra leaf analysis (NPK contents)

The data regarding the NPK contents are presented in Table 8. The data indicated that the application of EM-biofertilizer has significant effect on the nitrogen (N) contents of the okra crop. The maximum N contents in plants were recorded in EM5-biofertilizer (1.63) followed by EM10-biofertilizer application along with EM0-biofertilizer (1.61), whereas the lowest value was found in control (0.99). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there was 39% more nitrogen contents in the crop with respect to control (soil only), while 15.5% increase with respect to compost with the application of EM-biofertilizer.

The co-efficient of determination ($r^2 = 0.97$) indicated the positive trend towards the biomass of okra. It is clearly understood that as the concentration of N in okra leaves increases, the biomass increases and vice versa (Fig. 3).
Table 8. Leaf analysis of okra grown under the EM-biofertilizer treatments.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N in leaf</th>
<th>P in leaf</th>
<th>K in leaf</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.99</td>
<td>0.34</td>
<td>0.24</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>1.37</td>
<td>0.36</td>
<td>0.53</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>1.51</td>
<td>0.44</td>
<td>0.59</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>1.63</td>
<td>0.53</td>
<td>1.03</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>1.61</td>
<td>0.47</td>
<td>0.69</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.22, 0.23 and 0.28 for NPK in leaf respectively.

Fig. 3 Effect of leaf nitrogen on biomass of Okra

Regarding Phosphorous, the maximum P contents in plants were recorded in EM5-biofertilizer application (0.53) followed by EM10-biofertilizer application (0.48) while the lowest value was noted in control (0.34). The trend
was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Comparatively, there were 36% more phosphorous contents in the crop with respect to control and 30.75% with respect to compost on relative basis by applying EM-biofertilizer.

The co-efficient of determination ($r^2 = 0.72$) indicated a positive trend towards the biomass of okra. As the concentration of P in okra leaf increases, the biomass increases and when it decreases the amount of biomass decreases (Fig. 4).

![Graph showing the effect of leaf phosphorous on biomass of Okra](image)

**Fig. 4**  Effect of leaf phosphorous on biomass of Okra

The maximum K contents in plants were recorded in 5% application of EM-biofertilizer (1.030) followed by 10% EM-biofertilizer application (0.69), while the minimum value was found in control (0.24). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer >
control. There was 76% more potassium contents in the crop with respect to control and 48.54% with respect to compost as compared with 5% EM-biofertilizer. The co-efficient of determination \( (r^2 = 0.69) \) indicated the +ve trend towards biomass. It is clearly understood that as the concentration of K in okra increases, the biomass increases and vice versa (Fig. 5).

![Fig. 5 Effect of leaf potassium on biomass of okra](image)

**4.4.2 Soil analysis after Okra**

The data regarding the organic matter (OM) is presented in Table 9. The data depicted that the application of EM-biofertilizer has significant effect on the OM in the soil. The maximum OM was recorded in EM5-biofertilizer application (1.60) and followed by EM10-biofertilizer application (0.49). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 75% increase in OM in the soil with respect to...
control (soil alone) and 79% with respect to compost with the application of EM-
biofertilizer.

The co-efficient of determination ($r^2 = 0.97$) indicated the positive trend
towards the organic matter on amount of biomass. It is clearly understood that as
the organic matter increases, the biomass increases and when it decreases the
biomass also decreases.

**Fig. 6** Effect of soil OM on biomass of okra
Results and Discussion

Table 9. Soil analysis after okra.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N (%)</th>
<th>P (mg kg⁻¹)</th>
<th>K (mg kg⁻¹)</th>
<th>OM (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.02</td>
<td>5.47</td>
<td>84.83</td>
<td>0.16</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>0.03</td>
<td>7.93</td>
<td>138.00</td>
<td>0.33</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>0.05</td>
<td>9.37</td>
<td>146.32</td>
<td>0.38</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>0.08</td>
<td>16.98</td>
<td>155.37</td>
<td>1.60</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>0.06</td>
<td>14.13</td>
<td>147.10</td>
<td>0.49</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.02, 1.58, 1.58 and 0.21 for soil NPK and organic matter respectively.

As far as nitrogen is concerned, almost same trend was observed; the maximum N contents were recorded in application of EM5-biofertilizer (0.08) and followed by EM10-biofertilizer application (0.06). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 68% more nitrogen in soil with respect to control and 63% with respect to compost i.e., without EM-biofertilizer. Figure 7 indicates positive trend between the two parameters suggesting that lower N concentration in soil means smaller biomass ($r^2 = 0.90$) and higher concentration means greater biomass.
Fig. 7 Effect of soil nitrogen on biomass of okra

The maximum P contents in soil were recorded in EM5-biofertilizer application (16.98) and followed by EM10-biofertilizer application (14.13). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Relatively, there were 45% more phosphorous contents in the soil with respect to control (soil alone), while 53% increase in EM0-biofertilizer as compared with 5% EM-biofertilizer. This trend was also confirmed by the co-relation of soil P with biomass. It was clear evident from figure 8 that with the increase in the availability of soil phosphorous, biomass was also increased ($r^2 = 0.99$).
Fig. 8 Effect of soil phosphorous on biomass of okra

Regarding potassium, the maximum K contents in soil was recorded in EM5-biofertilizer application (155.37) and followed by EM10-biofertilizer application (147.10). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On comparative basis there was 90% more potassium contents in the soil with respect to control and 11% with respect to compost alone. Figure 9 shows significant correlation of soil K with biomass ($r^2 = 0.93$). Greater value of K in soil increased the biomass of okra.
Results and Discussion

The okra (*Hibiscus esculantus*) vegetable was cultivated with the help of EM-biofertilizer with the same set of treatments as mentioned in the methodology. The results of the experiments indicated that the EM-biofertilizer application has considerable effect on the growth parameters and the nutrient concentration in plant and soil. The maximum yield was recovered with the application of EM5-biofertilizer as compared with other treatments.

Increased soil fertility status increases number of tillers, grain number and yield. The availability in soil fertility and type are known to have greater impact on crop yield and quality than other production factors (*Carr et al.*, 1992; *Badruddin et al.*, 1999). This is also supported by the findings of Bhagat and Varma (1991) who reported that incorporation of organic manure in the shape of

![Fig. 9 Effect of soil potassium on biomass of okra](image)

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farmyard manure improved soil quality and yield of crop. The use of manure was considered as the key for the productivity of the crop (Gao et al., 2000; Hao et al., 2004).

Regarding nutrient concentration the nitrogen sources from organic and inorganic influence the yield (Diepenbrock, 2000). The organic products enhanced the N status of soil organic matter to ensure the N supply to meet the plant requirements (Rathke, 2005). The organic sources maintained or enhanced the N status of soil and ensured the supply of N to plants (Condron et al., 2000; Berry et al., 2002).

The application of enriched manure such as fermented manure also increased K and P in plants (Soumare et al., 2003). This is also supported by the findings of Ouedraogo et al., (2001) and Soumare et al., (2003) who reported that increased availability of P and K indicated that the manures supply these nutrients into the soil. The application of manure increased the microbial biomass, which enhanced the build up of soil P (Parham et al., 2003). The increase in the availability of P and K indicated that fermented manure with the beneficial microorganisms supplied these nutrients into the soil (Ouedraogo et al., 2001; Soumare et al., 2006 b).

These studies indicate a positive contribution of EM towards NPK nutrition of crops and its availability in the soil.
4.5 Spinach (*Spinacia oleracea*) Growth Parameters

The data regarding the growth parameters is presented in Table 10. The data indicated that the application of effective microorganisms (EM) inoculum along with compost has significant effect on the growth parameters of the spinach crop. The maximum leaf length was recorded in EM5-biofertilizer application (17.53) followed by EM10-biofertilizer application (15.53). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 46% increase in leaf length with respect to control (soil only) in EM-biofertilizer and 41% with respect to EM0-biofertilizer.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Leaf length (cm)</th>
<th>Biomass (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>9.40</td>
<td>21.70</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>10.30</td>
<td>48.00</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>14.53</td>
<td>48.90</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>17.53</td>
<td>50.30</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>15.53</td>
<td>49.00</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 5.9 and 18.4 for leaf length and biomass for spinach crop respectively.

Regarding the biomass, the maximum biomass was recorded in EM5-biofertilizer application (50.30) and followed by EM10-biofertilizer application (49.0). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer...
Results and Discussion

There was 57% more biomass with respect to control in EM-biofertilizer treatment and 5% with respect EM0-biofertilizer.

4.5.1 Spinach leaf analysis (NPK contents)

The data regarding the NPK contents is presented in Table 11. The data showed that the application of effective microorganisms (EM) inoculums along with compost has significant effect on the nitrogen (N) contents of the spinach crop. The maximum N contents in plants were recorded in EM5-biofertilizer application (1.23) followed by EM10-biofertilizer application (1.13). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Comparatively, there was 57% increase in nitrogen contents in the crop with respect to control and 43% with respect EM0-biofertilizer.

Table – 11: Leaf Analysis of Spinach

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N in leaf (%)</th>
<th>P in leaf (%)</th>
<th>K in leaf (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.51</td>
<td>0.37</td>
<td>1.05</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>0.70</td>
<td>0.87</td>
<td>1.85</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>0.98</td>
<td>0.88</td>
<td>1.15</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>1.23</td>
<td>0.99</td>
<td>1.19</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>1.13</td>
<td>0.94</td>
<td>1.10</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.24, 0.42 and 0.58 for NPK in spinach leaf respectively.
The co-efficient of determination ($r^2 = 1.0$) indicated a positive trend of N towards biomass. Lower the N concentration in leaves, lower will be the biomass, higher the N concentration, higher will be the biomass (Fig. 10).

![Graph showing the relationship between leaf nitrogen and biomass](image)

**Fig. 10** Effect of leaf nitrogen on biomass of spinach

Regarding phosphorous the maximum P contents in plants were recorded in the application of EM5-biofertilizer (0.99) which followed by EM10-biofertilizer application (0.94). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there were 62% more phosphorous contents in the crop with respect to control and 12% with respect to compost alone by applying EM-biofertilizer.

The co-efficient of determination ($r^2 = 0.98$) indicated the positive trend towards the biomass. As the concentration of P in spinach leaves increases, the biomass also increases and when it decreases the biomass also decreases (Fig.11).
Almost same trend was noted in case of potassium. The maximum K contents in plants was recorded in application of EM5-biofertilizer (1.19) followed by EM10-biofertilizer application (1.10). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There were 95% more potassium contents in the crop with respect to control and 56.3% with respect to compost with the application of EM-biofertilizer. The co-efficient of determination also showed a positive trend of potassium towards the increase in biomass with increase in K (Fig.12).
4.5.2 Soil analysis after spinach

The data regarding nitrogen contents in soil is presented in Table 12. The maximum N contents were recorded in the application of EM5-biofertilizer (0.057) followed by EM10-biofertilizer application (0.046). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there was 52% increase in nitrogen in soil with respect to control and 29% with respect to EM0-biofertilizer. Relation of soil nitrogen concentration and spinach biomass gives positive trend ($r^2 = 1.0$). With the increase of soil N, biomass increases and vise versa (Fig. 13).
Fig. 13 Effect of soil nitrogen on biomass of Spinach

Table – 12: Soil Analysis after Spinach

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N (%)</th>
<th>P (mg kg⁻¹)</th>
<th>K (mg kg⁻¹)</th>
<th>OM (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.03</td>
<td>5.08</td>
<td>80.01</td>
<td>0.46</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>0.04</td>
<td>9.11</td>
<td>110.50</td>
<td>0.57</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>0.04</td>
<td>11.57</td>
<td>125.00</td>
<td>1.28</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>0.06</td>
<td>14.30</td>
<td>132.60</td>
<td>1.40</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>0.05</td>
<td>12.66</td>
<td>131.00</td>
<td>1.33</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.014, 2.09, 6.60 and 0.14 for NPK and OM respectively in soil after spinach harvest.

As far as phosphorous is concerned, the maximum P contents in soil were recorded in application of EM5-biofertilizer (14.3%) and followed by EM10-biofertilizer application (12.7%). The trend was EM5-biofertilizer > EM10-
biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there were 64% more phosphorous contents in the soil with respect to control and 36.29% with respect to EM0-biofertilizer. Figure 14 depicts a positive correlation of soil P and spinach biomass. Higher the concentration of phosphorous greater is the biomass ($r^2 = 1.0$).

![Fig. 14 Effect of soil phosphorous on biomass of Spinach](image)

Regarding potassium the maximum K contents in soil were recorded in EM5-biofertilizer application (132.6%) and followed by EM10-biofertilizer application (131%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Relatively, there was 39% increase in potassium contents in the soil with respect to control (soil alone) and 17% with respect to EM0-biofertilizer. This trend was also confirmed by
correlation of soil potassium and spinach biomass \( (r^2 = 1.0) \). With the increase of K biomass also increased (Fig. 15).

![Graph](image)

**Fig. 15**  Effect of soil potassium on biomass of Spinach

The data indicated that the application of effective microorganisms (EM) inoculum along with compost has significant effect on the OM in the soil. The maximum OM was recorded with the application of EM5-biofertilizer (1.4%) followed by EM10-biofertilizer application (1.3%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Comparatively there was 67% more OM in the soil with respect to control and 59.1% with respect to EM0-biofertilizer.

The co-efficient of determination \( (r^2 = 1.0) \) indicated the positive trend of organic matter on biomass of spinach. It is clearly understood that as the concentration of OM increases, the biomass increases and when it decreases the biomass decreases (Figure 16).
The EM-biofertilizer, prepared by inoculation of EM (solution) on municipal solid waste, was applied to spinach (*Spinacia oleracea*) crop to test its efficiency. Different growth parameters and NPK concentration in leaves and soil were recorded and analyzed statistically. The leaf length and biomass was recorded for different EM-biofertilizer treatments. The maximum increase in leaf length and biomass was recorded where 5% EM-biofertilizer was applied.

The increase in growth parameters might be due to the activities of microorganisms which fermented and enriched the nutrient portion of the compost (Hussain *et al.*, 2002, Khaliq *et al.*, 2006). The effectiveness of these microorganisms was also studied by Sena *et al.*, 2002, it was reported that the microorganisms have the potential to decompose waste by anaerobic decomposition which help to maintain the C/N ratio in the waste to supply the nutrients to the crop. Bajwa and Jilani (1994) also recorded significantly higher crop growth and yield with the application of EM over control. Xu (2000) found
EM application has significant effect on plant growth, yield and photosynthetic rate.

In the study the increase in NPK in leaves was recorded which is also at par with the results of Rashid et al., (1994) in his experiments under FYM + EM treatments. He found that there was higher N and P uptake by wheat. The decomposition of FYM with effective microorganisms produced better crop yield rather than alone FYM (Haq et al., 2002; Hussain et al., 1999).

The microbial inoculum with manure helps to increase the root length, which may be one factor for the responsibility of increase in crop yield. Similar results were reported by Bhattaria and Hess (1998); Webster et al., (1998) and Soliman et al., (1999). El-Harway et al., (2002) suggested that integration of ½ NPK with microbial inoculum is best for yield and nutrient uptake in crop.

The availability of phosphorus from the mineral and with organic manure increase the crop yield (Parfitt et al., 2005). The good supply of P enhanced the growth of crops that in turn raise the yield and nutrient status of soil (Haynes and Willims, 1993; Sterner and Elser, 2002).

Regarding the nutrient concentration, the organic products enhanced the N status of soil organic matter to ensure the N supply to meet the plant requirements (Condron et al., 2000; Berry et al., 2002). These results were also reported by Sakadevan et al., 1993; and Ledgard et al., 1998). The microbial inoculants resulted in significant build up of soil NPK, OM and crop yield (Zhao, 1995; Ahmad et al., 1996).
The application of manure increased the microbial biomass, which enhanced the build up of soil P (Parham et al., 2003). The increase in K may partly be attributed to the concentration effect (Troeh and Thompson, 1993). As the K increases, the yield of crop increases, so it has clear relationship to sustain crop yield by the addition of K sources in soil. These results were also in line with the findings of Sandhu (1993); Zhao (1995); Soumare et al., (2003); Vliet et al 2005 and Feng and Cong (2006).

4.6 Cowpea (Phaseolus vulgaris) Growth Parameters

Data regarding the growth parameters is presented in Table 13. The application of effective microorganisms (EM) inoculums along with compost has significant effect on the growth parameters of the cow pea crop. The maximum grain weight (fresh) was recorded in application of EM5-biofertilizer (3.77) followed by EM10-biofertilizer application (2.68). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 87% increase in grain weight in EM-biofertilizer treatment with respect to control and 65.25% with respect to EM0-biofertilizer.
Results and Discussion

Table - 13: Effect of EM-biofertilizer on growth parameters of Cowpea

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Fresh grain weight (g)</th>
<th>No. of grains</th>
<th>Dry grain weight (g)</th>
<th>Pod size (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.49</td>
<td>4.00</td>
<td>0.09</td>
<td>6.10</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>1.31</td>
<td>6.00</td>
<td>0.83</td>
<td>8.20</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>1.56</td>
<td>11.83</td>
<td>1.10</td>
<td>11.53</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>3.77</td>
<td>17.99</td>
<td>1.67</td>
<td>15.22</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>2.68</td>
<td>12.25</td>
<td>1.53</td>
<td>11.67</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.94, 6.78, 0.49 and 3.8 for fresh grain weight, number of grains, dry grain weight, and pod size respectively of cowpea.

In case of number of grains, the maximum number of grains was recorded in application of EM5-biofertilizer (17.99g) followed by EM10-biofertilizer application (12.25g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There were 78% more grains with respect to control and 67% with respect to EM0-biofertilizer.

In case of dry grain weight, the maximum grain weight was recorded in EM5-biofertilizer application (1.67%) followed by EM10-biofertilizer application (1.53%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There were 94% more grains with respect to control in EM-biofertilizer and 50% with respect to EM0-biofertilizer.

Regarding the pod size, the maximum pod size (cm) was recorded in EM5-biofertilizer application (15.22 g) and followed by EM10-biofertilizer (11.67 g).
g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 60% more increase in pod size with respect to control and 46% with respect to compost on relative basis with EM-biofertilizer.

4.6.1 Cowpea leaf analysis (NPK contents)

The data regarding the NPK contents is presented in Table 14. The statistical interpretation of the data depicted that the application of effective microorganisms (EM) inoculum along with compost has significant effect on the nitrogen (N) contents of the cowpea crop. The maximum N contents in plants were recorded in EM5-biofertilizer application (2.89g) followed by EM10-biofertilizer application (2.66g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there were 19% more nitrogen contents in the crop with respect to control and 16% with respect to EM0-biofertilizer.

Table – 14: Leaf Analysis of Cowpea

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N (%)</th>
<th>P (%)</th>
<th>K (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>2.33</td>
<td>0.18</td>
<td>1.20</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>2.43</td>
<td>0.18</td>
<td>1.29</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>2.64</td>
<td>0.19</td>
<td>1.32</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>2.89</td>
<td>0.20</td>
<td>1.46</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>2.66</td>
<td>0.20</td>
<td>1.39</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.56, 0.02, and 0.09 for NPK respectively in leaf.
The co-efficient of determination \( r^2 = 0.81 \) indicated the positive trend towards the grain weight. As the concentration of N increases, the grain weight increases and vice versa (Fig.17).

The maximum P contents in plants were recorded in EM5-biofertilizer application (0.21 g) which was followed by EM10-biofertilizer application (0.20 g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there was 11% increase in phosphorous contents in the crop with respect to control and 11% with respect to EM0-biofertilizer.

The co-efficient of determination \( r^2 = 1.0 \) indicated the positive trend towards the weight of grains. As the concentration of P in cowpea leaves

---

**Fig. 17   Effect of leaf nitrogen on dry grain weight of cowpea**

The maximum P contents in plants were recorded in EM5-biofertilizer application (0.21 g) which was followed by EM10-biofertilizer application (0.20 g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis there was 11% increase in phosphorous contents in the crop with respect to control and 11% with respect to EM0-biofertilizer.

The co-efficient of determination \( r^2 = 1.0 \) indicated the positive trend towards the weight of grains. As the concentration of P in cowpea leaves
increases, the boll size increases and when it decreases the weight of grains also decreases (Fig. 18).

The maximum K contents in plants were recorded in application of EM5-biofertilizer (1.46 g) and followed by EM10-biofertilizer application (1.39 g). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There were 18% more potassium contents in the crop with respect to control and 12% increase in K in EM-biofertilizer treatment as compared to EM0-biofertilizer. There was positive trend in the co-efficient of determination ($r^2 = 0.99$) towards the K concentration and grain weight (Fig. 19).
Results and Discussion

4.6.2 Soil quality parameters of cowpea

The data regarding the organic matter (OM) is presented in Table 15. The maximum OM was recorded in application of EM5-biofertilizer (1.4%) followed by EM10-biofertilizer application along with compost (1.33%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. There was 68% more OM in the soil with respect to control and 59% with respect to compost alone, on relative basis.
Table – 15: Soil Analysis after Cowpea

<table>
<thead>
<tr>
<th>Treatments</th>
<th>N</th>
<th>P (mg kg(^{-1}))</th>
<th>K (mg kg(^{-1}))</th>
<th>OM (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (soil)</td>
<td>0.02</td>
<td>5.58</td>
<td>98.1</td>
<td>0.43</td>
</tr>
<tr>
<td>EM0-biofertilizer</td>
<td>0.04</td>
<td>7.50</td>
<td>120.0</td>
<td>0.57</td>
</tr>
<tr>
<td>EM1-biofertilizer</td>
<td>0.04</td>
<td>10.33</td>
<td>129.0</td>
<td>1.26</td>
</tr>
<tr>
<td>EM5-biofertilizer</td>
<td>0.06</td>
<td>11.37</td>
<td>136.0</td>
<td>1.40</td>
</tr>
<tr>
<td>EM10-biofertilizer</td>
<td>0.05</td>
<td>11.07</td>
<td>135.0</td>
<td>1.33</td>
</tr>
</tbody>
</table>

Each value is an average of three replications; LSD was 0.01, 2.52, 7.60 and 0.143 for NPK respectively in soil after harvest.

The co-efficient of determination \(r^2 = 0.83\) indicated a positive trend of organic matter towards the grain weight. It is clearly understood that as the concentration of OM increases, the weight of grain increases and vice versa (Fig.20).

![Graph showing the effect of organic matter on dry grain weight of Cowpea](image)

**Fig. 20** Effect of Organic Matter on dry grain weight of Cowpea
In case of nitrogen, the maximum N contents were recorded in the application of EM5-biofertilizer (0.05) followed by EM10-biofertilizer application (0.04%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Relatively, there was 56% increase in nitrogen in soil (soil alone) with respect to control and 29% with respect to compost (EM0-biofertilizer treatment). Figure 21 shows positive correlation between soil nitrogen and dry grain weight of cowpea ($r^2 = 0.95$). With the increase in soil N, dry grain weight also improved.

![Fig. 21  Effect of soil nitrogen on dry grain weight of cowpea](image)

The maximum P contents in soil were recorded in application of EM5-biofertilizer (11.37%), followed by EM10-biofertilizer application (11.07%). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. Comparatively there was 51% increase in phosphorous
contents in the soil with respect to control and 34% with respect to EM0-biofertilizer. The co-efficient of determination ($r^2 = 0.93$) indicated the positive trend towards the dry grain weight of cowpea. It is very clear that as soil P increased the grain weight also increased and vice versa (Fig. 22).

![Graph showing the relationship between soil P and dry grain weight of cowpea](image)

**Fig. 22** Effect of soil phosphorous on dry grain weight of cowpea

As far as the concentration of potassium is concerned, the maximum K contents in soil were recorded in application of EM5-biofertilizer (136.0) and followed by EM10-biofertilizer (135.0). The trend was EM5-biofertilizer > EM10-biofertilizer > EM1-biofertilizer > EM0-biofertilizer > control. On relative basis, there was 34% increase in potassium contents in the soil with respect to control and 12% with respect to compost (without EM). Figure 23 depicts a positive correlation of grain weight of cowpea with the increase of potassium in
soil \((r^2 = 0.83)\) due to application of EM-biofertilizer. Grain weight increased with the increase of K in soil.

![Graph showing the relationship between soil OM and dry grain weight](image)

### Fig. 23  Effect of soil potassium on dry grain weight of cowpea

The application of EM-biofertilizer to cowpea (*Phaseolus vulgaris*) was aimed to save some quantity of NPK fertilizers by adding low-cost EM-biofertilizer to the crop. Cowpea and soil data showed significant positive effect of EM application on cowpea growth as well as on the availability of NPK nutrients and OM from the soil. The best results were obtained in the case of 5% EM-biofertilizer treatment.

Regarding the growth parameters the above results were supported by Liang (2000) who stated that EM increased the yield of a variety of fruits, vegetables and crops compared with other fertilizers containing equal nutrient elements. Almost same results were found for NPK contents in wheat grain and
straw and in soil. There it appeared that EM which contains photosynthetic N fixing bacteria fixed sufficient N to supplement the lower N level for proper growth of the crop. Chowdhry et al., (1994) also found increased crop yield through EM application in cow dung and rice straw treatments. Where as Karim et al., (1993) conducted a similar type of field experiments as one reported here and found increased wheat yield at all levels of application with EM. They also reported higher NPK contents and improved physical characteristics of soil due to increased OM with EM application. The same results were also reported by Choudhry and Iqbal (1993) stating that it is possible to get as good yield of pea with EM as with fertilizers. Similar findings were also reported by Zacharia (1995) by applying EM on peas. All the above results might be due to high level of microbial activity which enhanced organic matter decomposition as well as the release of plant nutrients (Idris, 2003; Khaliq et al., 2006).

Ahmad (1998) reported similar results that enrichment of manure with EM increased (60%) of the availability of major nutrients i.e. NPK. This increase might be due to the addition as well as release of plant nutrients from organic source.

Almost similar results were also reported by Filho et al., (1993) and Piyadasa et al (1993) that illustrated the ability of EM to facilitate the organic matter breakdown to enhance nutrient availability in relatively short time. Sangakkara et al (1995) reported that EM inoculums increased the efficiency of organic nutrients source. Saggar and Hedley (2001) proved that the
microorganisms along with organic manure also promote the N concentration in soil (Wardle, 2002).

The use of organic manures might be helpful for sustainable crop production and maintenance of soil fertility (Meisener, 2000). The enhancement of compost efficiency due to the application of EM as EM-biofertilizer increased the nutrient concentration which gave the maximum net return as compared to non-EM treatment (Haq et al., 2002). The application of FYM can be substituted for N and P (Yaduvanshi, 2003). The application of EM as FYM Biokashat might solublized which resulted in the buildup of P status in soil (Higa, 1991; Parr, 1994 and Papindick, 1995).

The application of enriched manure such as fermented manure also increased K and P in plants (Soumare et al., 2003). The inoculated manure as compost has been found to improve organic matter content and nutrient supply to plants (Giusquiani et al., 1995; Parkinson et al., 1999) and thus may reduce the inputs of mineral fertilizer in conventional agriculture and provides a useful nutrient source in organic farming (Erhart et al., 2005). These studies indicate a positive contribution of EM-biofertilizer towards NPK nutrition of crops and its availability in the soil.
Chapter 5

SUMMARY

Environmental pollution problem is associated with large production of municipal waste in big urban centers. The biodegradable part of the city waste has value for crop land as carbon source. The turn-round period from biodegradable material to soil applicable compost may be as large as to six months which hinders the use of municipal waste for crop production.

The objective of the study was to test the use of effective microbes as consortium of microorganisms for conversion of biodegradable portion of municipal waste into biofertilizer, and to test soil and crop quality under biofertilizer application. A three level of effective microbes’ solution was applied and inoculated for fifteen days at room temperature. Firstly, application of EM inoculum enhanced the organic waste material and increased concentration of bioavailable N, P, K and mass fraction of OC. EM inoculation decreases pH and C/ N ratio suggesting fast fermentation to break down the organic matter. It appears that the microorganisms in EM solution have the ability to break down organic matter by fermenting activity, thereby releasing plant nutrients. The inoculation of effective microorganisms increased the degradation and chemical breakdown of organic material and stimulated the process of mineralization of organic matter.

EM-biofertilizer prepared with three concentration levels was tested on four different crops i.e. cotton, okra, spinach and cowpea to know its effect on productivity of these crops. The application of EM-biofertilizer had significant effect on the growth and nutrient concentrations in plants and soil. There was
positive correlation between N, P, K, concentrations in leaves of the crops. In case of cotton, on relative basis there was significant increase in cotton boll size in the EM-biofertilizer treatment as compared with soil alone. The same trend was noted in case of okra, spinach and cowpea where with the application of EM-biofertilizer, the increase in fruit length, leaf length and pod size was observed as compared to control (soil only) as well as compost (EM0-biofertilizer). There was significant effect of EM-biofertilizer on plant height, and number of bolls of cotton. Same positive effect of EM-biofertilizer was also noted in the increase of number of plants, number of fruits, fruit length and biomass as compared to control in case of okra. There was significant effect in the increase of leaf length and biomass in case of spinach with the application of EM-biofertilizer. The increase in number of grains, grain weight and pod size was also recorded in case of cowpea, on the EM-biofertilizer treatments as compared to compost. The increase in nitrogen, phosphorus, potassium and organic matter in soil in case of cotton was significant. Same trend in increase of soil NPK and OM was noted in EM5-biofertilizer treatment in the case of spinach, okra and cowpea. The correlations were developed, which indicated that N, P, K in leaves and OM in soil have positive correlation with the yield of all these crops.

Conclusion, the overall results of the study clearly indicated that the application of EM to municipal solid waste convert it into beneficial biofertilizer rich in organic matter and micro- and macro- nutrients. Tonnes of solid waste produced daily in Pakistan can be treated with effective microorganisms (EM) for producing EM-biofertilizer for crop production. This is practical significance if adopted by farmers. Soil health and in turn the productivity of soil can be maintained for sustainable agriculture and environment.
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