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Comparative Analysis of Organic, Inorganic, and Plant Growth Promoting Bacteria Fertilizers on Pisum sativum Growth

Original Article

Sadaf Khalil[1], Beena Ghaffar[1], Mujeeba[1], Noor e Sehar[1] and Uqba Mehmood[2*]

ARTICLE INFORMATION

Corresponding Author:

- uqbaa@yahoo.com

Affiliations:

1. Department of Biological Sciences, Superior University, Lahore
2. Department of Microbiology and Molecular Genetics, Punjab University, Lahore

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ABSTRACT

The use of fertilizers has become a ubiquitous practice in agriculture, with farmers and agriculturists relying on them to Nurture soil fertility and optimize plant growth conditions. Fertilizers can be broadly grouped into three categories: organic fertilizers, inorganic fertilizers, and plant growth promoting bacteria. Organic fertilizers, such as livestock manure, agricultural waste and municipal sludge, offer a natural and sustainable alternative to synthetic fertilizers. Inorganic fertilizers, on the other hand, are based on chemical substances like nitrogen, phosphorus, calcium, and potassium. Plant growth promoting bacteria, including *Rhizobium, Azospirilium, Azobacter*, and *Cyanobacteria*, have also gained popularity for their ability to enhance plant growth.

This study aimed to conduct an analysis of these three fertilizer types to determine their effectiveness in promoting pea plant growth. To achieve this, pea plants were grown using each of the three fertilizers, as well as a control group without any fertilizer. The plants were then subjected to various tests, including soil pH, water holding capacity, fresh weight, dry weight, root length, and shoot length.

The results revealed that plants grown with organic fertilizers exhibited the most efficient growth patterns, followed by the control group, inorganic fertilizers, and plant growth promoting bacteria. Notably, the plant growth promoting bacteria showed the least growth among the four groups. These findings suggest that organic fertilizers may be a more effective and sustainable option for promoting plant growth, and highlight the need for further research into the optimal use of different fertilizer types.

Introduction:

Over the years use of fertilizers has increased extensively. In current times new categories and sub-categories of fertilizers have been explored. According to the basic classification, fertilizers are categorized into three major categories which includes organic fertilizers, inorganic fertilizers and plant growth promoting bacteria(PGPB).

Agricultural waste, livestock manure and municipal sludge, lie in organic fertilizers. While inorganic fertilizers includes chemical substances, for instance ammonia, urea, phosphorus and nitrogen etc. Third category which is called plant growth promoting bacteria(PGPB) is associated with different bacteria which are *Rhizobium, Azospirillium, Azobacter, and cynobacteria etc.*

Organic fertilizer is eco-friendly and maintains pH at which plants grow deliberately. The best way of using organic fertilizers is by composting. During composting, waste went through several stages or processes before finally being used. There are different types of compost. This process increase the microbial activity that can be helpful for plant growth(1). Animal dung or livestock manure contain different types of bacteria, fungal spores and some protozoa. In addition to microorganisms, they also contain several important inorganic nutrients. Organic acids present in organic fertilizer increase the efficiency of nutrients present along with them by acidify them. Organic fertilizer improves and maintains soil structure, moisture holding capacity and balance nutrients in soil. Manure application to subsoil increases transpiration activity of plant, this results in doubling of root. Addition of poultry livestock to other organic fertilizer increases some trace elements such as Cu, Fe, Zn, Mn, Co, Se, I. Major drawback of this is that it is difficult to store it for longer period of time due to its bad odor and risk of contamination of water and overall earth pollution(2). Long term use of this manure can lead to organic pollutant such as polycyclic aromatic hydrocarbons and organochlorines and heavy metals in soil that affect the plant growth(3).

Then we have inorganic fertilizer, which includes different chemicals such as ammonia, ammonium, urea, potassium, magnesium etc. Plants absorb these nutrients directly from soil that helps them to grow at a faster rate. These inorganic fertilizers enhance plant growth by increasing enzymatic activity, rate of respiration, rate of photosynthesis, water balance and signaling pathway, and all this led to plant growth and increased crop yield. For an instance with the application of ammonia on soil, it results in temporarily elevated levels of pH. This happens when NH_3 combines with H^+ ions and removes them from soil and form NH_4. This removal of H^+ ion from leads to the elevated level of pH(4). This is how chemical fertilizers work. This fertilizer also provides protection to plants from insects that may harm plants. In addition to that there are some drawbacks of this fertilizer such as it can be harmful to the environment, can cause pest resistance and development of food safety decline. This fertilizer also lead to the escape of 50% nitrogen and Potassium to the atmosphere or water sources(5). They also cause greenhouse gas emission, eutrophication in aquatic system and salination in soil. Various researchers say use and release of phosphorus intensified greatly from past few years and planting is one of the major causes of it. Increased level of phosphorus cause contamination of groundwater and soil compaction(6).

Now heading toward PGPB which includes different kinds of bacteria such a *Rhizobium, Azobacter, Azospirilium, Cynobacteria.* etc. These bacteria fix nitrogen, promote plant growth, secure plants from infection and stress of abiotic through different mechanisms. Bacteria that create endophytic relationship with plants are playing more effective role regulating in plant growth. These fertilizers increase plant growth directly by forming auxins, gibberellins and cytokinin (these are plant growth regulating harmones) by the evoke of root metabolic activities and/or by providing biologically fixed nitrogen.

For example, *Cynobacteria* which is symbiotic nitrogen fixing bacteria, produces blue green algae that further fix nitrogen from soil to roots. With the help of fertilizers germination, root development, nutrient and water uptake are also improved by populating the rhizosphere and making the nutrients freely accessible.

These are also environment friendly. Growth regulating bacteria may also be able to lessen the negative effects of chemicals of agro ecosystems and fulfill the increased demand for food by the world's population(7, 8).

Therefore, by integrating the understanding of plant physiology with microbiological and technological considerations, the development of a stable bio formulation is feasible (9).

The aim of this study is to conduct a comparative analysis between Organic fertilizer, Inorganic fertilizer and Plant growth promoting bacteria (PGPB), to assess which one is more effective and the conditions suitable to be more functional. When they were grown, effectiveness analyzed. This study will help farmers to understand under what condition plant growth regulating bacteria works and how the activity of plant growth regulating bacteria can be enhanced.

MATERIALS AND METHODS

Experimental Design and Treatments

Pea plants (*Pisum sativum*) were grown using three categories of fertilizers: inorganic, organic, and biofertilizer. Four groups of pea plants were established:

Group A (organic fertilizer): Livestock manure and agricultural waste were used as organic fertilizer. Three pots containing 125g of soil each received 5g of organic fertilizer.

Group B (inorganic fertilizer): Urea was used as the inorganic fertilizer. Three pots containing 125g of soil each received 2.5g of inorganic fertilizer.

Group C (PGPB): Bacteria isolated from chickpea plants were used as Plant growth promoting bacteria. Three pots containing 125g of soil each were inoculated with the isolated bacteria, and then pea seeds were sown.

Group D (control group): Three pots containing 125g of soil each were grown without any fertilizer.

Each pot received 5 pea seeds. The experiment consisted of a total of 12 pots.

Isolation and culture of Bacteria

Root collection and surface sterilization: Roots of chickpea plants were collected and washed with 0.1% mercuric chloride ($HgCl_2$) solution to eliminate surface contaminants.

Grinding and suspension preparation: The sterilized roots were then ground into a fine paste using a mortar and pestle. The paste was transferred into clean test tubes containing 10ml of distilled water.

The isolated bacteria were then cultured on the prepared NA media plates, allowing for the growth and identification of the bacterial colonies.

Bacterial Inoculation and Seed Germination

The bacteria grown on the petri plates, to prepare bacterial suspension in 50 ml sterilized distilled water. Pea seeds were then soaked in this suspension for 2 hours to facilitate bacterial inoculation. Meanwhile, seeds for control and other groups were soaked in sterilized distilled water for an equal duration.

Seed Germination and Pot Preparation

After the soaking period, the inoculated and control seeds were sown into pots filled with soil. Two pots containing the inoculated seeds were labeled as Group C (PGPB treatment). The remaining seeds were sown into separate pots, which were labeled as Group A (organic fertilizer), Group B (inorganic fertilizer, urea), and Group D (control, no fertilizer).

Physical growth parameters

After 15 days the physical parameters such as fresh weight, dry weight, root length and shoot length were noted.

Water holding capacity of Soil

20g soil from each pot collected in a piece of mousseline cloth. Then these cloths with soil were hang at some height over an empty pot, 20ml of water was passed through this soil. Left this soil for 40 to 45minutes. After 45minutes some of water absorbed by soil which is hung in cloth and the rest of the water will drop down to measuring cylinder. The water quantity absorbed by soil indicates the water holding capacity of the soil.

Soil pH Test

For pH test, soil was taken from all the pots separately. Mix the 8g of soil from each pot into 30ml of water. pH strips were put down into the mixture and measure pH according to different colors difference showed on pH strip.

RESULTS AND DISCUSSION

Fertilizers are natural or artificial substances containing essential chemical elements that promote healthy growth and productivity of plants. By enriching the natural fertility of soil or replenishing vital elements deprived by prior crops, fertilizers play a important part in optimizing yields and agricultural productivity.

Plants require seventeen crucial elements for growth and development. Three of these nutrients - carbon, hydrogen, and oxygen - are readily available from air and water. However, the other 14 elements must be obtained from the soil. In cases where the soil lacks these essential nutrients, supplementation with fertilizers or organic materials like compost may be necessary to support optimal plant growth.

Pea plants were cultivated separately with three distinct fertilizer treatments: chemical fertilizer, organic fertilizer, and plant growth-regulating bacteria. An additional control group was maintained without any fertilizer application. Upon maturity, various growth parameters were measured to assess significant differences among the treatments. The evaluated parameters included: Root length, Shoot length, Fresh weight, Dry weight, Soil pH, Water holding capacity of the soil

These measurements enabled a comprehensive comparison of the different fertilizers on pea plant growth and development.

Table:1 Root length, shoot length, fresh weight and dry weight of Pea Plant (mean values)

Sr.No	Growth regulating bacteria	Chemical fertilizer	Bio fertilizer	Control group
Shoot length(cm)	7.6	7.49	11.7	9.17
Root length(cm)	7.99	6.1	10.29	10.63
Fresh weight (g)	1.051	1.206	1.503	1.209
Dry weight(g)	0.756	0.904	0.9	0.95

This data (Table:1) elaborates to understand the effect of fertilizer along with comparison and how that growing material influences the activity of growing factors present within the plant. Measurements of the shoot length, root length, dry weight, fresh weight showed that plant was grown with Organic fertilizer with high shoot length followed by control group, growth regulating bacteria, and Inorganic fertilizer. Control group have high root length followed by organic fertilizer, inorganic fertilizer, and control group. Organic fertilizer has high fresh weight followed by control group, inorganic fertilizer and growth regulating bacteria.

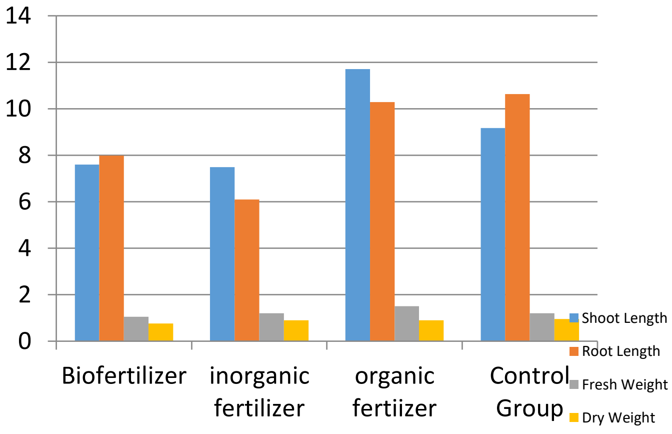

Figure:1 Comparative analysis of shoot length, root length, fresh weight and dry weight

The Comparative analysis of Pea plant grown by three different kinds of fertilizer (Chemical fertilizer, Organic fertilizer, Plant growth promoting bacteria (PGPB) and Control group)

High dry weight was observed in chemical fertilizer followed by plant growth promoting bacteria (PGPB), organic fertilizer and control group. Another parameter that define the growth of plants is soil and water holding content For this purpose water holding content, pH of soil were tested in order to understand how effective soil can be in plant growth.

The preservance of nutrients in soil depends on various factors, including:

1. Soil composition: The proportion of sand, silt, and clay particles, such as loam, loamy sand, and silt loam, affects nutrient availability.
2. Organic content: The amount of organic particles, like humus and decaying plant material, influences nutrient cycling and availability.
3. Soil pH: The acidity or alkalinity of the soil, measured as pH, impacts the solubility and access of essential elements for plant uptake.

Understanding these factors is crucial for optimizing soil fertility and nutrient management.

Soil pH refers to the measure acidity or alkalinity of soil. Optimal soil pH is crucial, as extreme levels can disrupt chemical reactions, nutrient availability, and biological activity.

For most fruits and vegetables, a slightly acidic to neutral soil pH (5.5-7.0) supports healthy growth and development. This optimal pH range enables plants to efficiently absorb essential nutrients from the soil.

While most fruits and vegetables thrive in slightly acidic to neutral soils (pH 5.5-7.0), some plants have unique requirements. For instance, blueberries necessitate a more acidic environment, with an optimal pH range of 4.2-5.2.

Fortunately, soil pH can be adjusted using various amendments. To increase soil pH, lime (ground limestone) can be applied. Conversely, elemental sulfur can be used to lower soil pH, making it more acidic.

Plant that was grown with Organic fertilizer has highest pH of 9 in comparison to others followed by plant growth promoting bacteria (PGPB), plant grown with inorganic fertilizer has pH of 8.7, control group has pH of 8 and at last that contain soil containing Biofertilizer(PGPB) has pH 7.

Soil Water Holding Capacity (WHC) is a crucial concept for farms to grasp, as it directly impacts crop production. In simple terms, WHC refers to the amount of water that a specific soil type can retain for crop use.

Water Holding Capacity (WHC) refers to the ability of a soil to physically retain water against gravity. This occurs through cohesion, where soil particles attract and hold water molecules.

Soil texture significantly influences WHC. For instance:

- Sandy soils: Low WHC due to large pores and low cohesion between particles.
- Silt loam soils: Higher WHC, as smaller pores and increased cohesion enable better water retention.

Understanding WHC is essential for optimizing irrigation, reducing water loss, and promoting healthy plant growth.

Soil	Available water content in mm water depth per m soil depth (mm/m)
sand	25 to 100
loam	100 to 175
clay	175 to 250

Now heading toward water holding capacity control group absorb most of the water that is 16ml followed Organic fertilizer absorb 13ml Inorganic fertilizer absorb 9ml plant growth promoting bacteria (PGPB) absorb 8ml.

Farmers often encounter several obstacles that impact crop yields and agricultural productivity. Some of the most prevalent difficulties include; Irrigation limitations: Inadequate water supply or inefficient irrigation systems; Increased desiccation: Sandy soils exacerbate water loss due to exposure and free drainage; Water stress: Insufficient moisture availability, leading to reduced crop growth and productivity. Addressing these challenges requires careful planning, effective water management strategies, and soil conservation practices to ensure sustainable agricultural production (10). In greenhouses, growers face unique challenges related to growth sources. To prevent soil pore clogging caused by capillary barriers, they often rely on high-permeability mediums. However, these mediums, such as sand: Have naturaly low plant-available water content (PAWC)- Contain low nutrient levels. To address these limitations, researchers have explored the fertilizers utilization and addition of other additives, enhance PAWC in growth sources. This area of study has gained significant attention in recent years, even for mediums with high PAWC, aiming to optimize plant growth and productivity in controlled environments (11) Utilizing additives that enhance Plant-Available Water Content (PAWC) in soil and growth mediums has been shown to: Improve seed germination rates, Enhance plant growth and development, Increase plant survival rates. Notably, these benefits can be achieved without requiring large volumes of growth medium, making this approach an efficient and sustainable solution for optimized plant production (11, 12). Enhancing Plant-Available Water Content (PAWC) can have additional benefits, including: Reduced nutrient leaching in sandy soils, particularly for ammonium (NH_4^+) and other mobile nutrients- Lower fertilizer application requirements, resulting in cost savings and minimized environmental risks associated with excessive fertilizer use. By optimizing PAWC, growers can promote sustainable nutrient management practices, mitigate environmental impacts, and maintain healthy, productive crops(12).

Figure:2 Pea plant (*Pisum sativum*) growing in four groups: Group A (organic fertilizer), Group B (inorganic fertilizer, urea), Group C (biofertilizer (PGPB) treatment) and Group D (control, no fertilizer).

In comparative analysis at this point growth measuring parameters shows mix results at some points Biofertilizer was more effective than others and at some extents Inorganic fertilizer was more effective same case happen to organic fertilizer and control group. None of them seem to be fulfilling all the growth requirements (Figure: 1,2). As our focus was plant growth promoting bacteria (PGPB), which do not have impressive role in plant growth this may be because the PGPB used was taken from chickpea plant where it showed impressive growth. But for Pea plant (*Pisum sativum*) it was not suitable enough and in previous studies as the fresh weight of plant is lower than others it means plant do not hold much water. This support the idea that it won't be able to survive longer during drought conditions. Because for the survival of plant during drought one must contain high amount of water in addition to other nutrients(13). Lower water content also effects the transportation of nutrients from roots to shoot and shoot to stem. It was also observed that plants which are water deficit are more prone to electrolyte leakage, and levels of hydrogen peroxide, superoxide were elevated at considerable amount. This will lead to plant growth and its overall yield(13). Another parameter which was under consideration was soil pH, As soil pH test was performed which says growth regulating bacteria has low pH in comparison to others, soil pH of growth regulating bacteria was a neutral, this means when pH of soil is neutral than plant won't be able to grab enough nutrients from soil this could definitely affect plant's overall growth (14). Now heading toward organic fertilizer and control group, there' growth was considerably high in comparison to others. Organic fertilizer shoot length is high than all of other three groups and root length of control group is high of all other three group. This is due to the use of dead leaves of plant and animal waste which contain high amount of methane, nitrogen, and some other growth regulating bacteria such as some bacillus spores, Cornynebacterium , Lactobacterium and some fungal species such as Aspergillus and Trichodermain addition to fungal spores and bacterial spores some protozoa are also present(14). Aspergillus and Trichoderma are known to play a key role in growth of bacteria by enhancing and promoting nutrient absorption, some synthesis of phytohormone which are auxin, gibbrellin and cytokinine etc. They also prevent plant from invasion of insects and diseases, they also promote the synthesis of some enzymes such as lytic enzyme thus preventing plant from pathogenic fungal attacks. Trichoderma also enhance shoot length, stem length, and left a remarkable effect on both fresh weight and dry weight which are of great importance(15). These fungal spores also recycle carbon and nitrogen directly from environment(16). With some elements These bacterial and fungal spores found in animal waste also provide metals which are also important to note in plant growth, these metals include iron, zinc etc Heading toward plant waste which contain high amount of nutrient including some carbohydrates some salts such sodium, potassium, chloride, sulfate, calcium, and magnesium salts, and some vitamins such as vitamin A, vitamin C, vitamin E etc(17). This plant waste also contain phytohormones and secondary metabolites such as flavnoidsetc and some antioxidants(18, 19).

CONCLUSIONS

When selecting a fertilizer, it's essential to consider two key factors:

1. Soil test results: Understand the soil's nutrient content, pH, and other properties to identify deficiencies and limitations.
2. Plant needs: Consider the specific nutrient requirements of your crops, including the type and quantity of nutrients needed,

By taking a data-driven approach, a fertilizer can be selected that optimizes plant growth, minimizes waste, and reduces environmental impact. It is suggested that PGPB can be more effective if some other microorganisms such as fungal spores were also added in combination. Organic fertilizer contains multiple microorganisms and some various nutrients, which enables it, to exhibits relatively better plant growth results.

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CONFLICT OF INTEREST

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DATA SHARING STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request

JBAHS web address: www.jbahs.pk
Email address: editor.jbajhs@superior.edu.pk