Co-inoculated seedling biopriming with *Trichoderma* and Mycorrhiza on crop growth in tomato and brinjal

काशी हिन्दू विश्वविद्यालय

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To,
The Registrar
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Dear Sir,

I have the great pleasure in forwarding the thesis entitled **"Co-inoculated seedling biopriming with *Trichoderma* and Mycorrhiza on crop growth in tomato and brinjal"** submitted by **Mr. Mahesh Kumar Yadav, I.D. No. 18412SAC010,** in partial fulfilment of the requirements for the award of the degree of **Master of Science (Agriculture) in Soil Science and Agricultural Chemistry, Department of Soil Science & Agricultural Chemistry,** Institute of Agricultural Sciences, Banaras Hindu University, Varanasi.

I certify that the entire scheme of investigation reported herein, was planned and carried out by the candidate under my guidance. To the best of my knowledge and belief, the data presented in the thesis are genuine and original. No part of the work has been submitted for any degree or distinction.

Yours faithfully,

FORWARDED

(AMITAVA RAKSHIT)
Supervisor

BHU
capital of knowledge

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Date:

Place: Varanasi ***(Mahesh Kumar Yadav)***

LIST OF ABBREVIATIONS

%	Percent
°C	Degree centigrade
/	Per
µ	micro
AMF	Arbuscular mycorrhiza fungi
CEC	Cation Exchange Capacity
cm	Centimetre
dSm^{-1}	Deci siemen per meter
EC	Electrical Conductivity
et al.	And others
Fig.	Figure
g	Gram
ha	Hectare
i.e.	Id est (that is)
kg	Kilogram
L	Liter
M	Molar
mm	Millimeter
mg	Milligram
mL	Milliliter
MT	Million tons
ppm	Parts per million

pH	Puissance de Hydrogen
RDF	Recommended Dose of Fertilizers
SD	Standard Deviation
t	Tonnes
Viz.	namely

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Chapter-I

INTRODUCTION

The equilibrium between food demand and its supply has been disrupted by the strain of population growth in India. The population of India in 2019 is estimated at about 1.37 billion (World Population Review, 2019) and increasing at a rate of 1.1 per cent per year. According to the World Banks study in 2016, arable land per capita retains 0.12ha and continues to decrease every day as urbanization has decreased the amount of cultivable land. In the recent past, it led to more intensive cropping practices. Intensive agriculture favours higher application of inputs to achieve higher yield and economic returns per unit cultivable land. Green revolution in India since the 1960's has led to a major focus on the production of cereals. It may have served increasing demand but leads to severe negative effect on soil, i.e. cultivation of high yielding input responding cereals has resulted in depletion of organic matter and soil nutrients (John *et al*., 2001). Use of agro-chemicals in agriculture i.e. for plant protection, herbicide, plant growth promoter etc. has severe impact on environment through soil, water bodies, and air pollution.

India with a number of geographies (mountains, peninsula, tarai have different implications on agroecology areas, deltaic alluvium, plain lands, etc.) with varying quantities of rainfall and vegetation. The soil of these regions differs from place to place and from time to time resulting in various fertility constraints such as lower soil organic carbon content (except in mountain-based deltaic marshy lands and forest), mineral nutrient deficiency, soil erosion, etc. Deficiencies in mineral nutrients may differ from specific nutrient elements such as nitrogen (> 55%), zinc (about 45%), phosphorus (>35%), sulphur (>37%), boron (>30%), iron (13%) etc. This leads to greater application of fertilizer per unit area. Use of fertilizers has shown a higher yield, a higher level of food security and economic safety for farmers. Synthetic fertilizers (except for phosphatic fertilizers) are generally water soluble in nature and thereby after application they became readily available for either plant uptake or lost through volatilization or leaching. This not only lowers down the fertilizer use efficiencies but

also has tremendous environmental impacts. Moreover fertilizer use efficiency is decreasing day by day, more input cost of fertilizer has increased, allowing it to become more expensive over time. The N-P-K fertilizer application dosage has risen in the last five decades. Fertilizer consumption was less than 1 million tonnes before the mid-1960s. With the introduction of high-yielding variety seeds, there was acceleration in the growth of fertilizer consumption. It reached 12.73 million tonnes in 1991-92 as against 0.78 million tonnes in 1965-66. After the decontrol of P and K fertilizers the growth in consumption slowed. The highest consumption was recorded in 1999/2000 (18.07 million tonnes of nutrients). Since then, the growth in consumption has been erratic. In 2003/04, total nutrient consumption was 16.8 million tonnes. FAI (Fertilizers Association of India, 2012) reported that 3 countries namely China, India, and USA were consuming 69.01, 25.55, and 20.33 million tonnes of macronutrient fertilizers respectively. The manufacturing of synthetic fertilizers in various industries results in the release of hazardous by-products into ecosystem causing it to lose its equilibrium. Not only this, certain synthetic fertilizer contains plant- and animal-toxic heavy metals like cadmium, lead, arsenic etc. which warrants to fix threshold level or safety limits must be set for the application of fertilizer in soil. As a result, our agriculture has changed its direction from more land-based returns to demand-based quality goods in a sustainable manner where soil-health management is given a major focus (Sinha *et al.,* 2014). While organic farming has provided some quality produce needs, it wouldn't be enough to satisfy the demand of the large population. In fact, there is more pest infestation in organic farming. Thus crop production and management practices have acquired another dimension for applying microbiological agents that will not only boost the growth and development of the crop in the farm but also maintain the harmony of the ecosystem.

Vegetables are annual or perennial horticultural crops, with certain sections (roots, stalks, lowers, fruits, leaves, etc.) that can be consumed wholly or partially, cooked or raw. Vegetables are important for human nutrition in terms of bioactive nutrient molecules such as dietary fiber, vitamins and minerals, and non-nutritive phytochemicals (phenolic compounds, flavonoids, bioactive peptides, etc. In present scenario, there is a greater emphasis on 'nutritional security". Vegetables can be very

reasonable source of all kind of nutrients and minerals. Apart from nutritional benefits, the production of vegetables improves the economy of a country as these are very good source of income and employment. The contribution of vegetables remains highest (59 – 61%) in horticulture crop productions over the last five years. During 2019-20 the area under vegetables was 10.35 Million ha with a production of 77 Million Tonnes in India. For this period the total vegetable production was highest in case of West Bengal (281.13 MT) followed by Uttar Pradesh (261.94 MT).

The tomato is one of the most important "protective foods" both because of its special nutritive value and also because of its widespread production. It is the world's largest vegetable crop after potato and sweet potato, but it tops the list of canned vegetables. Tomatoes are used as soup, salad, pickles, ketchup, puree, sauces and in many other ways. Tomato is a major source of vitamins and minerals. It is widely used as salad vegetable. In India it is commonly referred as 'poor man's orange' (ascorbic acid 15 mg to 10-20 mg /100g edible portion).It is also rich in citric acid and mallic acid. Glutamic acid is an amino acid which is mostly present in tomato. Tomato contains many important minerals like Na, K, Ca, Mg, P, K, Fe, Zn, B. Tomato fruit is source of minerals such as copper (0.01-0.09 mg), manganese (0.09-0.13 mg) and zinc (0.1-0.17 mg kg-1 of fruit). The alkaloid present in tomato is called tamatin and the colored pigment is called lycopene. Lycopene content is high at 70^0 F or 21^0 C.

Brinjal is a stable vegetable high in nutritive value. It is rich in minerals is Ca, Mg, P, K and Fe. It is also a good source of vitamin A and C. Bitterness in brinjal is due to presence of glycoalkaloids. Glycoalkaloids content vary from 0.4 to 0.5 mg per 100 g of fresh weight.

Trichoderma a cosmopolitan and saprophytic fungus present in soil. As a fungus, *Trichoderma* can make efficient use of soil organic carbon and plant litters as a source of nourishment than other soil microbes. They can play various roles including reconstruction of soil structure through synthesis of sticky substances (polysaccharides), decomposition of organic matter, control of soil and seed borne diseases, recycling, solubilization and mobilization of plant nutrients, production of different plant growth promoting substances, alleviating biotic and abiotic stress

tolerance in plants and degradation of residues of synthetic agrochemicals in soil (Sivasakhti *et al.* 2014). Earlier, *Trichoderma* known only for its bio-control ability but it is now evident that *Trichoderma* can also increase the nutrient use efficiency for several nutrient elements. *Trichoderma* also increase 4ractice4 enzyme activity and maintain hormonal balance under unfavorable climatic conditions.

Trichoderma can be isolated from decaying plant material, forest soil and sometime in arable land. *Trichoderma* can culture easily. *Trichoderma* may be applied directly to the soil or inoculated with seed or seedlings to improve crop yields. The technique of treating hydrated seeds with biological agent such as *Trichoderma* is known as bio-priming. Arbuscular mycorrhizal fungi (AMF) on the other hand are symbiotic association between plants and the fungi that colonize on te roots and act as an aid in adapting and mitigating strategies for resolving stress.

Through analyzing the major challenges faced by Indian vegetables crop cultivation, *Trichoderma* and mycorrhiza should be included as an integral component in field trials. Most work has shown that the application of *Trichoderma* has improved the nutrient use efficiency, synthesized significant plant growth regulators and reduced salt stress, moisture stress, drought stress and other soil-borne pathogen induced biotic stresses posed on vegetables production.

Taking these facts into consideration, a pot experiment was planned on tomato & brinjal crops to assess the following goals:

- To examine the effect of co-inoculated seedling biopriming with *Trichoderma viridae* and mycorrhiza biopriming on growth of tomato & brinjal.
- To evaluate the effect of co-inoculated seedling biopriming on agronomic use efficiency in tomato & brinjal.

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Chapter-II

REVIEW OF LITERATURE

A brief review of pertinent research work relating to the objectives has been discussed in this chapter.

2.1 Nutritional aspects of vegetables

Vegetables are rich source of nutrients, important for human health. They are particularly important sources of micronutrient, vitamin-A, B6, C and E as well as folic acid, Fe and Mg. As vegetable contain many of the dietary factors like vitamins, minerals and amino acids they are considered as protective supplementary food. Potato, sweet potato Tapioca yam are the rich source of carbohydrate. Beans and Pea provides protein. Green leafy vegetables, palak, amaranthus, fenugreek, drumstick, carrot are rich in pro-vitamin-A. Vitamin-C present in tomato, Beans, sprouts etc. Follic acid found in spinach and other green leafy vegetables, legumes and contains Fe. Vegetables containing high amount of fibre are spinach (6.3%), broad beans (4.2%) and okra (3.1%). Follic acid found in abundance in spinach (123 mg/100g),other green leafy vegetables(48-80mg/100g) and beans(144mg/100g) which is requird for the multiplication and maturation of red cells. Its deficiency results in megaloblastic anaemia that is generally seen in infants and pregnant women. Vegetable consumption can avoid some serious chronic diseases like diabetes, cancer, obesity, metabolic syndrome, cardio-vascular diseases, as well as boost immunity these diseases (Taha and Funda *et al.,* 2018). Low vegetables intake in diet has been estimated to cause heart diseases in about 31% and stroke in 11% globally. Unbalanced diet with low vegetable intake and low consumption of complex carbohydrates and dietryfibre are estimated to cause 2.7 Million death every year, and were among the top 10 risk factors of mortality (World Health Report, 2007). Production of vegetables in India increased in last decade (2008 to 2017-2018) as 134.87 million MT to 182.03 million MT and per capita gross availability of vegetables (gms /day) 309.99 to 388.72 and per capita net

availability of vegetables from 217gms/day to 272gms/day(Horticultural Statistics at a Glance, 2018).

In present perspective, when we talk about nutritional security, vegetables can be a very reasonable source of all kind of nutrients and minerals.

2.2 Nutritional aspect of tomato and brinjal

The tomato is one of the most important **"**protective foods**"** both because of its special nutritive value and also because of its widespread production. It is the world's largest vegetable crop acerage after potato and sweet potato, but it tops the list of canned vegetables. Tomatos are used for soup, salad, pickles, ketchup, puree, sauces and in many other ways. Tomato is a major source of vitamins and minerals. It is widely used as salad vegetable. In England, it referred as "love of apple' or 'love apple'. In India it is commonly referred as 'poor man's orange' (Ascorbic acid 15 mg to 20 mg /100g edible portion).It is also rich in Citric acid and Mallic acid. Glutamic acid is an amino acid mostly present in tomato. Tomato contains many important minerals like Na, K, Ca, Mg, P, K, Fe, Zn, Boron. The alkaloid present in tomato is called tamatin and the coloured pigment is called lycopene. Lycopene content is high at 70^0 F or 21^0 C.

Tomato contains a number of phytochemicals of which lycopene is the most w ell known. Aditonally, there are other carotenoids (e.g. βcarotene, phytoene, phytoflue ne), phenolics (e.g. coumaric and chlorogenic acids, quercetin, rutine, and naringenin) , moderate amounts of antioxidant vitamin C (ascorbic acid), and a small amount of v itamin E (tocopherol). Red color of tomato fruit is due to lycopene. It is a powerful antioxidant; which neutralise free radicals, which may cause damage to cell components (e.g. DNA, protein, lipids). Lycopene is ranked as the most potent among the following antioxidants: lycopene > α-tocopherol> α-carotene > β-cryptoxanthin> β-carotene > lutein (Heber and Lu, 2002). Protein and dietary fibre are also present in tomato fruits, although the major constituent is water, comprising 94-95% of the fruit by weight (Davies & Hobson, 1981). Many scientific evidence are present for lycopene 's role in reducing prostate cancer risk. Lycopene can also help to reduce the risk of other cancers and cardiovascular diseases, and play an important role in eye health. Tomato not only consumed as a raw staple food due to their desirable nutritional

properties but they are also being increasingly used in many popular tomato products (Perez-Conesa *et al.,* 2009). More than 80% of ´ tomatoes grown are consumed in the form of processed products such as juice, soup, concentrate, dry-concentrate, sauce, salsa, puree, dry-tomato, ketchup, or paste (Kaur *et al.,* 2008). Tomato byproducts contain many biologically active substances which mostly go to waste despite being a promising source of dietary fibers, proteins, carotenoids, tocopherols, polyphenols, and other compounds (Vagi *et al.,* 2007; Lavelli and Torresani, 2011). Among these bioactive compounds, polyphenols, carotenoids, and vitamins have a broad range of physiological properties like anti-inflammatory, antiallergenic, antimicrobial, antithrombotic, cardioprotective, and antioxidant effects (Yang *et al.,* 2008a).

Brinjal is a stable vegetable high in nutritive value. It is rich in minerals is Ca, Mg, P, K and Fe. It is also a good source of vitamin A and C. Bitterness in brinjal is due to presence of glycoalkaloids. Glycoalkaloids content vary from 0.4 to 0.5 mg per 100 g of fresh weight. Purple variety has higher copper content and polyphenol oxidate activity whereas iron and catalase activity is the highest in the green cultivars. Amino acid content is higher in purple variety. Brinjal also used as cooked vegetables, pickle making and dehydration industry. Green leaves of brinjal plant are good appetizers, aphrodisiac and cardiotonic. It is also beneficial in vaata and kapha. In unani system roots are used to alleviate pain. Brinjal is a rich source of abundant nutrients which are desirable for proper body growth. Brinjal is assemblage of complete set of minerals, vitamins, nutritional fiber, protein, anti-oxidants, along with some phyto-chemicals that having scavenging activities (Noda *et al.,* 2000, Whitaker and Stommel. 2003). Major phytochemicals in brinjal are caffeic, chlorogenic (phenolic components) glucoside, delphinidin and nasunin (flavonoids) (Choudhury, 1976; Kwon *et al.,* 2004; Matsubara *et al.,* 2005; Bhasker and Kumar, 2015; Cassidy *et al.,* 2013). Studies shows that the brinjal extracts have superb healing effects on different disorders like burns, warts, inflammatory infections, gastritis, stomatitis and arthritis (Im *et al.,* 2016). Brinjal is producing various secondary metabolites along with some other compounds such as glycol-alkaloids, antioxidant compounds, and vitamins which carried a significant part in keeping good health. For example, a major phenolic compound chlorogenic acid (5-O-caffeoyl-quinic acid; CGA), found in fruit skin (Prohens *et al.,* 2013) which work as

an anti-obesity, anti-inflammatory, anti-diabetic agent and also have cardio-protective functions (Plazas *et al.,* 2013). Brinjal is the rich source of anthocyanin pigment, besides their coloring functions. It has been known that anthocyanin has significant role against diabetes, neuronal problems, cardiovascular disorders, and cancer as well. Purple colored Brinjal has a high amount of nasunin compound in their flesh helps against lipid peroxidation and ROS accumulation which occur due to a high level of iron in cells (Casati *et al.,* 2016). Anthocyanins present in the skin of brinjal rise serum antioxidant volume and support against heart illness and hyperlipidemia by decreasing LDL (low-density lipoprotein) oxidation. Fiber contents present in brinjal helps in digestion by removing toxins and harmful materials from our stomach thus by reducing stomach and colon cancer (Fraikue, 2016).

2.3 Cultivation of tomato and brinjal in india

The major tomato producing states in the country are Andhra Pradesh, Madhya Pradesh, Karnataka, Gujarat, Odisha, West Bengal, Chhattisgarh, Bihar, Telangana, Tamil Nadu, Uttar Pradesh, Maharashtra, Haryana and Himachal Pradesh. These states are account for about 90% of the total production of the country (Monthly Report: GOI, November, 2019).

The total global area under tomato is 46.16 lakh ha and the global production is to the tune of 1279.93 lakh tones. Tomato production is estimated to be 20.57 million tonne (increase of 8.2%) as compared to 19.01 million tonnes in 2018-19. The major constraints in tomato and brinjal cultivation as perceived by the growers are high cost of high yielding varieties. high cost of fertilizers and chemicals, lack of knowledge of disease resistant varieties, lack of knowledge about proper application methods of chemical fertilizers, lower price at harvesting time, unavailability of fertilizers in the local market at the time of sowing, minimum support price is not fixed by the government, lack of knowledge of seed treatment and lack of knowledge and skills about proper method of tomato production (Jat *et al.,* 2012). In addition to these Indian farmers are typically small and marginal. Small and marginal farmers accounted for 86.2% of total Indian farmers but own only 47.3% of total crop area (Bera, 2018). Thus it would not be technically feasible to grow more intensively at high production costs.

Organic farming which got momentum in recent years also has its limitation such as lower yield with comparison to conventional farming with less pest infestation and improvement in soil management practices. On average total tomato yield in the organic system represented 36.5% of the yield in the conventional system, while the marketable fruit yield was 36.0% (Bettiol *et al.,* 2004). Lockeretz *et al.,* 1984; Gliessman *et al.,* 1990; Creamer *et al.,* 1996 were also observed lower yield that compared organic with conventional system. In general NPK @ 75 to 150 kg: 60 kg: 60 kg per ha has been recommended for various tomato and brinjal varieties. Besides the above inorganic fertilizers combination of well decomposed FYM and groundnut cake is recommended. FYM is also advocated @ 20-25 tons per ha at the time of last ploughing and incorporated into soil. Regarding the inorganic chemical fertilizers half N, entire P and K should be applied as basal dose, half N is given in 2 to 3 splits. 30, 45, 60 days after sowing.

2.4 Incorporation of beneficial micro-organisms in crop cultivation

The use of agrochemicals in agricultural production system has adversely affected complex biogeochemical cycles in environment (Perrott *et al.,* 1992; Matson *et al.,* 1997; Steinshamn *et al.,* 2004). Indiscriminate use of these chemicals and their impact on soil microbial habitat not only lowered the environmental quality but also reduces the efficacy of fertilizers (Adesemoye and Kloepper, 2009). Fertilizer use has also risen significantly and is anticipated to rise in order to provide increased food demand With decrement of fertilizer use efficiency as well as nutrient use efficiency not only increased the cost of synthetic fertilizers but also increased the energy consumption in fertilizer production (Black, 2013).Higher fertilizers application results in leaching loss, volatilization loss, fixation and run-off loss of many nutrients, especially macro-nutrients such as nitrogen, phosphorus, potassium, sulphur etc. (Gyaneshwar *et al.,* 2002; Sutton *et al.,* 2013). Human health issues now a days became more serious due to environmental pollution created with the excessive use of chemical fertilizers in crop production (Tilman *et al.,* 2002).Current agricultural scientists have focused not only on improving package practices that would provide higher yield with lower investment, but also on minimizing the impact on ecosystem and animal health. Soil microbial biomass not only plays a key role in the cycling of nutrients, but also

provides improved soil and soil quality in the long term. Scientists have found that many micro-organisms can support crop growth by providing various growth factors and inducing stress tolerance in a vagarious environment (Dimpka *et al.,* 2009).

In agriculture, the function of microbes is mainly to increase the efficiency of plant nutrient up take and make more unavailable nutrients accessible (Egamberdiyeva, 2007).The soil beneficial microbial reservoir tends to hold some of the applied nutrients by immobilizing plant fertilizer nutrients. This reduces the loss of nutrients by various means and, following their death, the decomposition of their dead cells releases immobilized nutrients into the soil environment, making them more accessible to crop roots (Steiner *et al.,* 2007; Marschner*et al.,* 2011).For example, the use of P solubilizing microorganisms will solubilize and mineralize soil unavailable P into available P (Barea *et al.,* 2002; Zaidi and Khan, 2007; Zhang *et al.,* 2014) and thus increase the availability of P to plants (Kim *et al.,* 1997; Chen *et al.,* 2006). Some other findings of micro-organism mediated process are microorganism mediated nitrogen mineralization like ammonification (Wainwright and Pugh, 1973) and Nitrification (Stark and Firestone, 1995) and sulphur mineralization (Kertesz and Mirleau, 2004).Besides adding more of crop residues to soil plays important role in build-up of soil organic carbon which is primary source of food and energy for these microbes. Thus lower availability of organic carbon in soil can hamper the activity and growth of soil microbes (De Nobili *et al.,* 2001; Demoling *et al.,* 2007; Marschner *et al.,* 2011). Increasing applied nutrient use efficiency can only be accomplished by increasing soil microbial population by introducing organic carbon ionto the soil (Singh *et al.,* 2015; Shen *et al.,* 2016).This may result in lower use of fertilizers and may reduce production costs (Shaviv *et al.,* 1993; Chen, 2006). Microorganisms not only decreased the use of synthetic fertilizers but also decreased environmental contamination caused by the improper use of synthetic fertilizers (Adesemoye and Kloepper, 2009) and other agrochemicals (Chen *et al.,* 2015).

2.4.1 Arbuscular mycorrhiza

In 1885, Albert Bernhard Frank, a German botanist, gave the term “mycorrhiza.” It’s the Greek term that means “fungal roots”. Mycorrhiza is a specific

symbiotic relationship between fungi and roots of higher plants (vascular in nature) (Harrison, 1997).In this symbiotic relationship, plants and fungi mutually benefit each other through food exchange, shelter and growth-promoting substances, where plants supply photosynthates (carbohydrate) to the fungi group and then feed on fungi that increase the efficiency of mineral nutrient uptake, such as nitrogen (N), phosphorus (P), potassium (K), magnesium (Mg), iron (Fe), zinc (Zn), copper (Cu), and various growth promoting substances such as hormones (Ludwig-Müller, 2000; Chen *et al.,* 2009) and induce stress tolerance both biotic (Borowicz, 1997) and abiotic (Ruiz-Lozano *et al.,* 1995). This is an endophytic relationship.

Franciszek Kamieński studied mycorrhizal symbiosis for the first time in 1879-1882. If a fungus enters the roots of any host plant intracellularly creating a globular, swollen vesicle and arbusculus, the symbiosis known to us as Vesicular Arbuscular Mycorrhiza (VAM) but when the fungus colonizes extracellularly the root mantle of the host plant, it is known as ectomycorriza. Many studies have reported that some fungi have both types of mycorrhizal association in order to conduct this type of symbiosis. That is why the term VAM is used as a general definition. AM is the most common symbiotic association in forest soil. They form this symbiosis in a wide range of plants, including most important crops in agriculture such as maize, rice, cabbage, cauliflower, tomato, brinjal, chickpea, soybean, etc.

Nearly 150 fungal species are known to form AM in many plants and generally belong to Glomeromycota phylum, which has a high degree of diversity among them. In 2012, Bücking classified five orders for AM fungi, namely

- Archaeosporales (e.g. *Archaeosporatrappei*)
- Diversisporales (e.g. *Acaulosporalaevis*)
- Gigasporales (e.g. *Gigasporarosea*)
- Glomerales (e.g. *Glomusmosseae*)
- Paraglomerales (e.g. *Paraglomuslaccatum*)

Despite the fact that predominance of AM is noticeable in forests yet they are accounted for to be present in a wide variety of rhizospheres. Practice of AM application in agriculture got advantageous since nineteenth century. But mass

multiplication of AM fungi became difficult as any synthetic media was unable to supply all the growth factors required for multiplication of AM fungi (Hepper, 1984).

It has been reported that nearly 3 lakh higher plant species form root hair colonization with AM fungi accounting about 90% of higher plant species (Kendrick and Berch, 1985). Trappe in 1987, reported the existence of plant families forming mycorrhizal association is around 95%. In 2011, Bagyaraj presented a report showing that most of the agricultural crops form mycorrhizal symbiosis.

The significant effect of AM symbiosis can be categorized as follows:

- Establishment of micro sized root hairs (Mukherjee *et al.,* 2012; Samolski *et al.,* 2012)
- Nutrient uptake enhancement such as phosphorus (P), zinc (Zn), iron (Fe) etc. (Chen *et al.,* 2003; Smith *et al.,* 2004; Kafkas and Ortas, 2009)
- Improvement of soil structure (Caravaca *et al.,* 2006),
- Fasten nutrient recycling in soil (Toro *et al.,* 1997),
- Alleviating stress tolerance of plants against both biotic and abiotic stresses (Feng *et al.,* 2002).

Besides these beneficial effects, most important emphasis given on application of AM fungi is its P mobilizing action. Many scientists have shown the interaction in the mycorrhizosphere environment involves P absorption by plant roots and its recycling (Azcon *et al.,* 1976). According to Barea *et al.,* (2005) these interactions involve 3 processes, i.e.,

i. Hyphae of AM fungi act as junction between roots and rhizospheric soil helping roots to uptake more of insoluble (unavailable) from of soil.
ii. Reduce the leaching loss of solution P through their microbial cells by immobilization. But they're making it available for plant uptake after their death.
iii. AM fungi increase the root surface area and root volume, thus increases more contact with soil resulting in higher P uptake through soil exploration.

2.4.2 Trichoderma

Like AM fungi, *Trichoderma* is a common saprophytic fungus under the Ascomycota division. *Trichoderma*formes conidiophores in the sac like structure known as ascus. They can also be marked by an opportunistic, anti-virulent symbiont (Harman *et al.,* 2004; Umadevi *et al.,* 2017), establishing an endophytic relationship with the host's root cells (Bailey *et al.,* 2009). Therefore they are commonly present in many ecological conditions other than soil such as in the bark, plant litters, various organic substrates etc. most common habitat of these fungi other than soil are bark, plant litters, organic substrates etc. they are pleomorphic in nature means possess both sexual (teleomorphic) stage known as *Hypocrea*and asexual (anamorphic) stage which is called *Trichoderma* (Jaklitsch and Voglmayr, 2014).

Trichoderma was first described by Christiaan Hendrik Persoon in 1794 (Ojha, 2018). In addition to extensive application in multiple fields, starting from medical aspects to industrial use, *Trichoderma* species have been widely acknowledged as a bio-control agent and for providing plant growth promotingsubstances in agriculture. The use of *Trichoderma* in crop production has long been recognized. In short, *Trichoderma* helps to increase agricultural production in following way,

- Prevent the plant disease causing pathogen (Elad *et al.,* 1980),
- Increasing the efficiency of plant roots to absorb more mineral nutrients from soil (Singh *et al.,* 2014), especially low mobility nutrients such as P.
- Inducing plant stress tolerance (Mastouri *et al.,* 2010; Shukla *et al.,* 2012),
- Providing good amount of plant growth promoting substances (Harman, 2011) etc.
- More than 85 species of Trichoderma are recognised. Some commonly use species are *Trichoderma asperellum,T. viridae, T. hamatum, T. harzianum, T. reesei, T. virens*etc.

2.4.2.1 Mechanism of Trichoderma

In agricultural perspective, *Trichoderma* possesses various role of action in increasing yield of crops. As *Trichoderma* is very rapidly growing fungi, producing a large amount of biomass in soil or rhizosphere, they can serve as biocontrol agents, compete for nutrients and dominate the rhizosphere with the release of several

hydrolytic or cellulolytic enzymes, secondary metabolites toxic to pathogens (Mathivanan *et al.,* 2008).

2.4.2.1 Induction of micro-sized roots

Trichoderma spreads its hyphae after infecting and colonizing the root cortex cells, protruding out of the root cells enhancing water and nutrient uptake by the plants. This root hair infection is performed by host roots mediated by lectin signaling (Harman *et al.,* 2004). *Trichoderma*, when receive the signals, stretched out its appressoria like structure which is mediated by hydrophobins (Linder *et al.,* 2002). It encodes cysteine-rich cell-wall protein after touching the root mantle which helps to adhere very tightly to the roots so that the fungi are not washed out through any kind of unfavourable environment (Harman *et al.,* 2012). It then releases "swollenin" (Brotman *et al.,* 2008), a protein that disrupts and softens the crystalline cellulose structure in the root cell wall (Haraman *et al.,* 2004). After that, it secretes cell-wall degrading enzymes such as cellulose, amylase, lipase, etc. (Elad *et al*. , 1982) and penetrates into the cortex cells of the root where it releases swollenin again, which increases the root surface area thus increasing the coverage of root. After successful penetration, *Trichoderma* spreads its hyphae from root cells to soils (rhizosphere) that cover the host roots.These hyphae have been reported to act as secondary roots that exceed the soil cover by the primary roots of host plants (Kleifeld and Chet, 1992). Thus, further exploration of the soil volume can be accomplished, that can contribute to more nutrient uptake by plant roots.

2.4.2.2 Protection against soil-borne diseases

Since 1930, endophytic fungi *Trichoderma* have been identified as a biocontrol agent against soil borne plant pathogens (Weindling, 1932). Mycoparasitism is a dominant mechanism in the management of soil borne pathogen (Haraman *et al.,* 2004). Trichoderma's mycoparastic capacity is very unique, as it is regulated by the pathway of mitogen activated protein kinase (MAPK), which is very common for all signaling pathways (Druzhinina *et al.,* 2011). MAPK mycoparasitis pathway starts when they sense any peptide molecules released during their growth and multiplication by pathogens.These peptide molecules get attached to G-protein receptors (Gpr1) or any other nitrogen sensing receptors on the surface of the *Trichoderma*hyphae. Now a signaling cascade is activated which, in turn, modulates certain transcriptional factors which encode genes for the biosynthesis of various secondary metabolites (chitinases,

pachybasins, glucanases, etc.) and cell-wall degrading enzymes (cellulose, lipase, etc.) which degrade and devour the cell wall of fungal pathogens.MAPK pathway involves genes that encode three MAPKs (enzymes) namely pathogenicity MAPK (TmkA) (Mukherjee *et al.,* 2003),cell integrity kinase (TmkB) and osmoregulatory MAPK (Hog1) (Druzhinina *et al.,* 2011). The Hog1 plays a major role in Trichoderma's self-defence system. A Hog1 is a protein that has an oxidative and osmotic stress tolerance effect, nullifying the production of harmful metabolites during attack on prey fungi (Druzhinina *et al.,* 2011).

2.4.2.3 Systemic resistance induction in plants

Trichoderma triggers plant defense against biotic stress (soil-borne pathogens attack) and abiotic stress (aberrant weather conditions, salinity stress, oxidative stress, etc.) (Abd-El-Kareem, 2007; Mastouri *et al.,* 2010). *Trichoderma*-inoculated seeds or plants exhibit two types of innate immune defense mechanisms to reduce biotic stress which are:

- Immunity triggered by Pathogen associated molecular pattern (PAMP)
- Effector triggered immunity (ETI) (Druzhinina*et al.,* 2011).

In this system, phenyl-alanine ammonia lyase is developed to improve lignification and the thickness of the cell wall resulting in inhibition of pathogens against puncturing the cell wall. *Trichoderma* fungal endopectinases release oligogalacturonides (Elad, 2000) which recognize PAMPs and other secondary molecules released during the early stage of infection. Plants release phenolic compounds in their cell walls after receiving signals, with simultaneous deposition of callose and cellulose (Hermosa *et al.,* 2012). This is how further 15ractice15es15 prevention is accomplished.

In case of oxidative and salinity stresses,, peroxidates, hydroperoxidelyase and other free radicles scavengers are generated by induction of signaling, pathways mediated by ethylene, jasmonic acid, and salicylic acid (Druzhinina *et al.,* 2011). They scavenge hydrogen peroxides (membrane lipid oxidizer), plant-free oxygen radicles, etc. (Singh *et al.,* 2011). They scavenge out the hydrogen peroxides (membrane lipid oxidizer), oxygen free radicles etc. from plant (Singh *et al.,* 2011).

2.4.2.4 Plant growth promotion

The *Trichoderma* fungi promote plant growth by the production of different hormones and their derivative compounds. Within the root cells they can produce a number of derivatives of Indole Acetic Acid (IAA) (Nieto-Jacobo *et al.,* 2017). Such derivatives activate auxin-dependent pericycle cells (Pelagio-Flores *et al.,* 2017), followed by primordial development and emergence, resulting in a more lateral root development (Garnica- Vergara *et al.,* 2016).Such auxin-like derivatives also reduce the Mitogen Activated Protein Kinase 6 (MAPK6), which is apparently inhibiting primary root growth and root hair formation (Martinez-Medina *et al.,* 2016). The *Trichoderma* fungus regulates ethylene within host plant. Ethylene antagonises the activity of auxins within the root cells of plants. *Trichoderma* produces ACC deaminase (ACCD) which cleaves 1-Amino Cyclopropane-1-Carboxylic Acid (ACC), the precursor to ethylene production, and produces ammonia and α-ketobutyrate (Viterbo *et al.,* 2010). Thus, inhibition of ethylene-auxin antagonism gets induced and more elongation of roots attained.

However, *Trichoderma*atroviride can balance the interactions between auxin and ethylene (Garnica- Vergara *et al.,* 2016) via the use of MAPKKK, also known as CTR1 (Harman and Shoresh, 2007), rather nullifying the ethylene effect. More root hairs are induced by this mechanism (Shoresh *et al.,* 2005; Contreras-Cornejo *et al.,* 2015).

2.5 Bio-priming

Biopriming is a process of seed treatment where any kind of biological agents (single or in consortium) can be inoculated after hydration with seeds.
Biopriming thus covers two key seed protection facts (Callan *et al.,* 1990):

I. physiological aspect of disease control (seed hydration),
II. biological aspect of disease control (inoculation with beneficial microbes).

Since it is a biological treatment, it can eliminate the use of synthetic chemicals to control crop disease. Not only it control soil borne pathogen, but it can also act as a dormancy breaker and induce germination, enhance vegetative growth and yield quality. Bacteria and fungi are generally used in biological treatment of seeds. In addition, some of these agents can establish better rhizosphere colonization

and reduce the competition for nutrients in soil with concomitant supply of several other additive effects to plants (Nancy *et al.,* 1997).

2.5.1 Factors affecting bio-priming

Biopriming is very, simple, easy and cost effective practice even for small and marginal farmers. In order to obtain better results from bio-priming, these following factors must be considered which settle down the outcome of biopriming (Ruan and Xue, 2002).

A) Aeration: Proper supply of O_2 is essential for cell rejuvenation.

B) Light: Some crops, also micro-organisms, proliferate well in light rather than dark.

C) Time: Length of seed soaking in osmotics and viability of spores taken into account for better priming effect.

D) Temperature: The temperature should not exceed 25^0C during inoculation because it increases the protrusion of radicals.

E) Method: Consideration of right salt solution to maintain optimum osmotic potential and right type of micro-organisms to inoculate particular type of crop species should satisfy the results.

2.5.2 Effects of bio-priming

Benefits of biopriming are enlisted (Gupta *et al.,* 2000; Bisen *et al.,* 2015*;* Rakshit *et al.,* 2015) below:

- Prevention of soil-borne pathogen-mediated diseases through food competition and the release of allelopathic chemicals (HCN).
- Induce plant growth through auxin production and regulation of the hormonal balance within plant cells.
- Greater tolerance to nutrient toxicity, salinity and drought.
- Improved plant nutrient uptake from soil by competition (Biofertilizer).
- Increase germination percentage thus reducing the germination time, leading to more uniform germination and strong seedling establishment.

- Reduce the adverse effects of synthetic agro-chemicals leading to more sustainable and environmentally friendly activities such as (Bioremediation).

Effect of *Trichoderma* and mycorrhiza biopriming on crops

Trichoderm species are genetically diverse and show specific traits (Harman *et al.,* 2004). It is harmless to apply in any crop regarding their mode of nutrition bcz. *Trichoderma* spp. in general are not associated with any harmful plant disease (Gams and Bissett, 2002). On the other side, they suppress the growth of many soil borne plant pathogenic microorganisms. When inoculated with crop seeds they help to alleviate stress, reduce the occurrence of plant diseases, and encourage good standing of crops and quality of yield. Mycorrhizal fungi can improve majority of nutritionally relevant crop's health mediated through enhanced nutrient acquisition based on the variabilities and extent of colonization. AM fungi support water and nutrient uptake of plants and may help the plants to achieve resistance to abiotic and biotic stress. The results of some scientists about *Trichoderma* biopriming are summarized in the following table;

Table 2.1: Effect of biopriming with *Trichoderma* (as single inoculum or in consortium) with mycorrhiza and other PGPR on different crops

Crops	***Trichoderma* and AMF inoculants (singly or in consortium)**	***Results***	**References**
Brinjal (*Solanum melongena*)	*Trichoderma harzianum*in consortium (BHU-51+BHU105+Pth)	Increase in germination rate (76.6%), shoot length (23.64%) and chlorophyll content (23.4%) and reduction in severity of *Sclerotiniasclerotiorum*rot	Singh and Singh (2012)
Tomato (*Lycopersicum esculentum*)	*Trichoderma harzianum*in consortium (BHU-51+BHU105)	Reduce Rhizoctoniasolani mediated root rot with concomitant increase in nutrient (N, P, Zn and Mn) uptake through release of organic acids (gluconic acid, citric acid and fumaric acid)	Singh *et al*. (2014)
Chilli (*Capsicum annuum*) var. PKM 1	*Trichoderma viride or Pseudomonas fluorescens* in each in single treatment	Boost germination and seedling vigour of chilli variety PKM 1	Ananthi*et al*. (2014)
Cucumber (*Cucumis sativus*)	*Trichoderma asperellum*	Improve micronutrient use efficiency in cucumber	Santiago *et al*. (2013)
Melon *(Cucumis melo)*	Arbuscular Mycorrhizal Fungi	Increase melon plant growth and nutrition particularly by solubilizing P to be up taken by roots	Martinez Medina *et al.,* (2011)
Tomato (*Lycopersicum esculentum*)	Arbuscular Mycorrhizal Fungi	Boost hormonal response that involves an altering in the levels of cytokinins, gibberellins, ethylene, abscisic acid, jasmonic acid and auxin.	Ferna´ndez *et al.,* (2014)
Tomato (Lycopersicum esculentum)	Arbuscular Mycorrhizal Fungi	Enhancement in regulated pathways of stress response mechanisms, their potential contribution to improved host stress tolerance	Rivero *et al.,* (2015)
Wheat (*Triticum aestivum*)	Arbuscular Mycorrhizal Fungi	Trigger metabolomic responses enhancing tolerance to water stress	Bernardo *et al.,* (2019)

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Chapter-III

MATERIALS AND METHODS

The present research trial entitled for "**Co-inoculated seedling biopriming with *Trichoderma* and Mycorrhiza on improves crop growth in tomato and brinjal"** involved a pot experiment followed by soil and plant sample analysis in laboratory of Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, U.P.

3.1 Description of the experimental site

3.1.1 Physiography of location

Varanasi is located at an elevation of 80.71 meters (264.8 ft.) in the center of the Ganges valley of North India, in the Eastern part of the state of Uttar Pradesh, along the left crescent-shaped bank of the river Ganges.

3.1.2 Climate

Varanasi experiences a humid subtropical climate with MDI (moisture deficit index) value ranges 20-40.commensment of south west monsoon start from mid of june, with peak in july- august and ceseson start in mid of july. Winter rains can also be seen. The annual precipitation is nearly 1100 mm. May and June are the hottest months with sometimes a maximum temperature above 43^0C. The cold period is between November and January and the minimum temperature may fall below 7^0C in January with a relative humidity of 63% which can rise to 85 percent during rainy season and drop to 41% during dry season. The details of the temperature, rainfall, relative humidity, sunshine and evaporation during the experimental period recorded week-wise by the meteorological observatory at Banaras Hindu University Agricultural Research Farm are presented in the table 3.1 below.

Table 3.1: WEEKLY METEOROLOGICAL DATA during the course of the pot experiment in 2019

Week No.	Month & Date	Rainfall mm	Temperature ^{0}C		R.H. %		Evaporation (mm)
			MAX	MIN	Morn.	Even.	
31	30-05	2.0	32.3	23.8	85	72	4.1
32	Aug 06-12	61	31.0	23.2	89	79	3.3
33	13-19	56.9	32.8	23.1	89	74	3.5
34	20-26	115.4	31.2	21.7	94	80	1.6
35	27-02	11.4	34.2	24.6	90	69	4.0
36	Sep 03-09	11.6	32.1	24.0	87	77	3.6
37	10-16	48.2	33.2	23.3	89	71	3.0
38	17-23	164.0	30.8	21.6	93	82	1.9
39	24-30	532.2	27.8	20.4	95	87	1.8
40	Oct 01-7	39.0	30.0	20.4	94	78	2.7
41	08-14	0.0	32.2	19.2	91	59	2.7
42	15-21	3.8	29.6	19.1	91	68	2.7
43	22-28	0.0	31.5	14.5	88	40	2.6
44	29-04	0.0	31.2	16.6	92	49	2.3
45	Nov 05-11	0.0	28.1	12.4	88	45	2.0
46	12-18	0.0	29.0	11.8	89	45	1.8
47	19-25	0.0	27.8	10.4	87	45	2.0
48	26-02	0.0	26.5	10.3	94	47	1.5
49	Dec 03-09	0.0	20.4	16.2	93	76	0.8
50	10-16	0.0	20.3	10.10	93	73	0.7
51	17-23	0.0	23.4	9.9	88	51	1.8
52	24-31	0.0	20.7	10.9	93	68	0.9

(Source: DEPARTMENT OF AGRONOMY, BHU, VARANASI)

3.2 Pot experiment

The experiment was carried out during September- December in 2019 in the net house of the Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi.

3.2.1 Experimental soil

Soil samples (0-18 cm) from Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi were collected to carry out the experiment. Farm soils are alluvium deposits of river Ganges. Higher silt and clay content with subsoil accumulation of exchangeable Ca^{2+} are characteristics of farm soil. Typically, illitic clay minerals are dominant with a higher potassium (K) content, but in these alluvium deposits a lower K value was reported. The soil was low in organic carbon content and Nitrogen content, and high in P content. The pH of the soil was slightly alkaline in nature.

3.2.2 Soil preparation and Pot filling

After collection of soil sample, the soil used in pot experiment were air dried ground and sieved through 2 mm sieve and filled in 10 kg earthen pot lined with polythene.

3.2.3 Crop

Tomato	
Crop name	Tomato
Scientific name	(*Lycopersicon esculentum*)
Family	Solanaceae
Variety	Kashi Amul
Brinjal	
Crop name	Brinjal
Scientific name	*Solanum melongena L.*
Family	Solanaceae
Variety	Kashi Komal

3.2.3 Varietal characteristics

Kashi Amul has an average field potential of 50-60 t ha^{-1} the fruits mature in 85-90 days post transplanting. Kashi Komal is a hybrid variety, having semi upright plant growth (90-100 cm in height) with light green stem and leaves. The picking starts in 65-70 days after transplanting and gives average yield of 80 t ha^{-1}

3.2.4 Collection of microbial agents

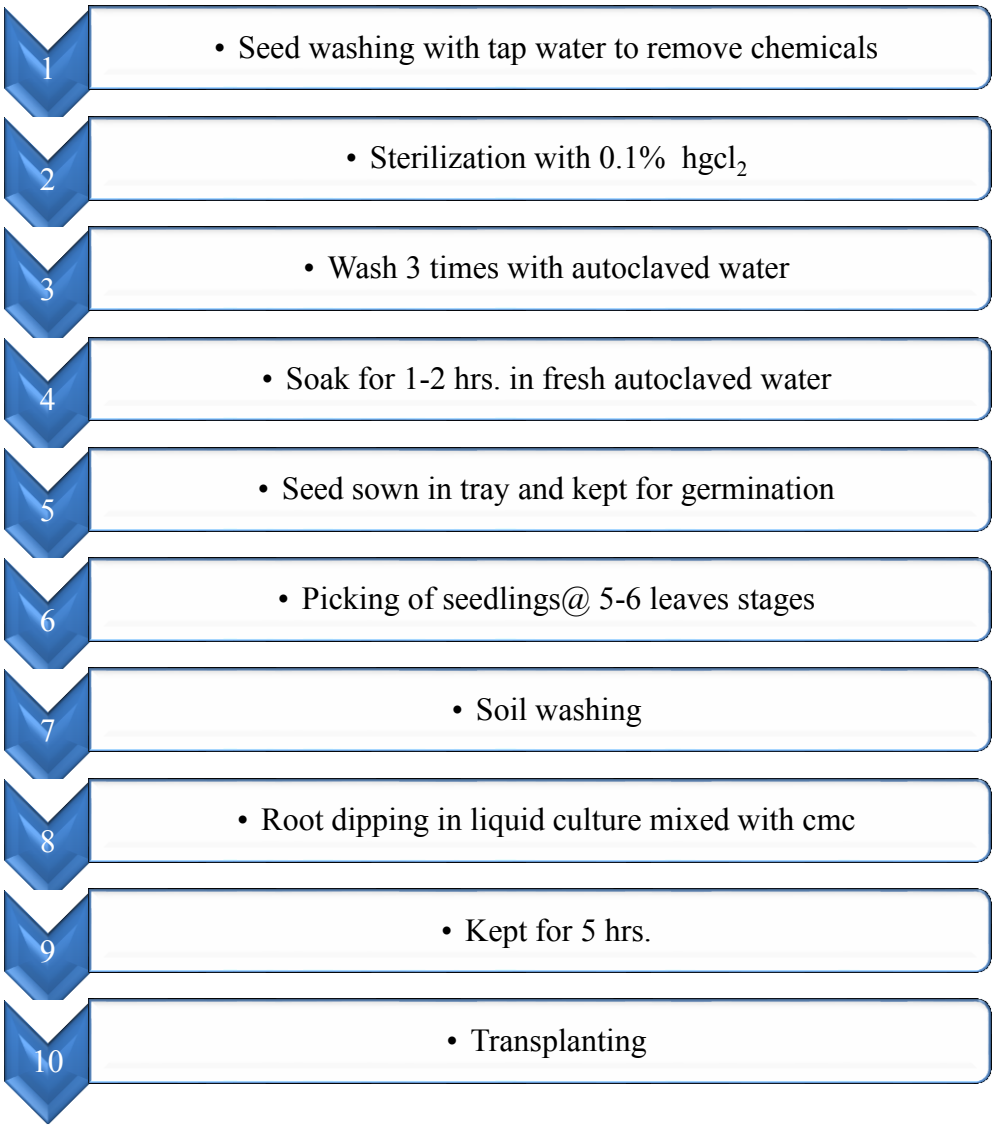
Trichoderma viridae spores (Ascomycetes fungi) are collected by growing them in PDA (Potato Dextrose Agar) media in the laboratory of the Department of Plant Pathology, Institute of Agricultural Sciences, University of Banaras Hindu, Varanasi, UP. Composition of PDA is shown in the following table.

Potato Dextrose Agar Composition

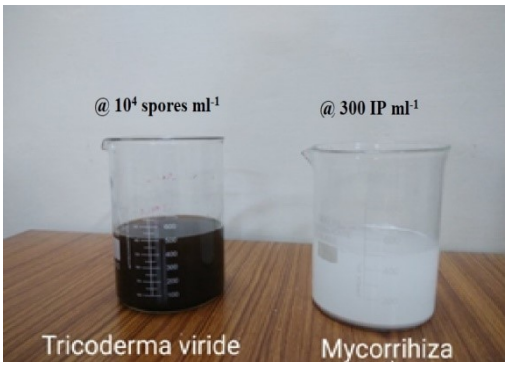
Ingredients	Amount (g/Lit)
Potato	200.00
Dextrose	20.00
Agar	15.00
pH (25^0C)	5.6

Mycorhizne fungi (trade name Cradle, Biostadt India Limited, Mumbai) in powder form was procured form Lucknow.

3.2.5 Procedure for Biopriming of seedling



Source: Sarkar *et al.,* 2019



Plates 1: Biopriming with *T. viridae* and Mycorrhiza in tomato and brinjal

3.3 Experimental Details

The pot experiment was conducted during mid-September to December 2019 in net house, Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi. Nutrients applied with varying doses of fertilizers. For N, both Di-ammonium phosphate (DAP) and Urea; for P, only DAP and for K, Murate of potash (MOP) were applied as recommended dose of N:P:K i.e.120:80:80 kg per ha. The experiment was performed in completely randomized block design (CRD).

3.3.1 Treatment details

Treatment No.	Doses of fertilizer (N:P:K) ($kgha^{-1}$)	Microbial primer
T_1	Control	-
T_2	75% RDF	*Trichoderma viridae* + Mycorrhiza
T_3	80% RDF	*Trichoderma viridae* + Mycorrhiza
T_4	85% RDF	*Trichoderma viridae* + Mycorrhiza
T_5	90% RDF	*Trichoderma viridae* + Mycorrhiza
T_6	RDF (120:80:80)	-

3.3.2 Planning and Layout

The total pots were divided into 3 (3) equal-size blocks. Each block was then divided into six (6) units leaving the bock effects. The total of fifty four (54) pots were then arranged accordingly and 3 replications of the 6 treatments were given. Each pot had two 2 plants in it.

Treatment:	06
Replication:	03
Harvest	03

Figure: Layout of Experiment

	Tomato						Brinjal					
30 DAT	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3
	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3
	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3
45 DAT	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3
	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3
	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3
60 DAT	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3	T_1R_1	T_1R_2	T_1R_3	T_3R_1	T_3R_2	T_3R_3
	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3	T_2R_1	T_2R_2	T_2R_3	T_4R_1	T_4R_2	T_4R_3
	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3	T_5R_1	T_5R_2	T_5R_3	T_6R_1	T_6R_2	T_6R_3

3.3.3 Growth stages and cultural practices

Date of fertilizer application	12.10.2019
Date of transplanting	15.10.2019
Plant population	2 plants/pot
Water management	Irrigation
Insect management	For red spider mite control @ 2 ml/l, Tissot for whitefly control @ 1 g/L of water
Weed management:	Manual weeding
Date of harvesting	15.12.19

Plate 2: Tomato and brinjal crop in net house

3.4 Analysis of Experimental soil

3.4.1 Soil Processing

The soils collected from the pot experiment were air-dried, crushed through wooden pistil-mortar and passed through 2 mm sieve both during pot filling and at various physiological growth stages of the crop as well as after the pot experiment was completed. Soils filled up in pots of 10 kg. After the experiment the initial soil samples and soils were tested to determine different physical and chemical properties.

3.4.2 Soil Physical properties

3.4.2.1 Bulk density

The initial soil samples were collected using core sampler in known volume cylinder. In electrical balance aluminum moisture boxes were weighted. After that the soils were collected in aluminium moisture boxes and weighed in electrical balance. The samples were dried on oven for 24 hours at 105^0C. After one day of drying in the oven, samples were taken out and electric balance weighed again. The bulk density was then calculated using the formula,

$$\text{Bulk density (Mg/m3)} = \frac{\text{(Box + Oven dried soil wt.) —Wt. of Empt Box}}{\text{Volume of core sampler}}$$

3.4.2.2 Particle density

The density of the particles was calculated by using Pycnometer. Six pycnometers were cleared, dried in the air and taken on electrical balance to measure its weight. The soil samples were filled in Pycnometer up to the brim and then tapped to settle it inside the Pycnometer. The process was repeated until the vacant space was filled totally. Then the weight of Pycnometer with soil was taken. After that soil was washed from it and water was filled up to the brim. Finally actual volume of Pycnometer was recorded using burette (Chopra and Kanwar, 1982).

3.4.2.3 Water Holding Capacity (W.H.C)

The water holding capacity of soil samples from the pot experiment was determined in the laboratory using the Keen-Rackzowski box (Chopra and Kanwar, 1982).

3.4.2.4 Mechanical analysis

Mechanical analysis of the soil was done using Bouyoucos hydrometer. Weighed 50 g of soil and poured 60 mL of 5% H_2O_2 (hydrogen peroxide) into a 1L beaker to remove organic matter.After that 400 mL of distilled water is added to dilute the suspension. The suspension was stirred for 10 minutes, then the whole system was placed into 1L measuring cylinder and shaken vigorously for 5 minutes with a plunger. Then the hydrometer was inserted in the suspension quickly. After stabilization of hydrometer, reading was taken after 4 minutes and 2 hours at surrounding temperature

25^0C. Those two readings correspondingly gave sand and silt material. The percentage of sand, silt and clay was then measured using reading and a correction factor, and the textural class was determined with the help of the textural triangle (Bouyoucos, 1962).

3.4.3 Soil Chemical Properties

3.4.3.1 Soil pH

Suspension of 1:2:5 soil-water was prepared by adding 25 mL of double distilled water in 10 g soil. This suspension was then used with the help of glass electrode pH meter for pH measurement (Jackson, 1973).

3.4.3.2 Electrical conductivity (EC)

1:2 soil-water suspension was prepared by adding 20 mL of double distilled water in 10 g soil. This suspension was allowed to settle down for some time in order to measure the EC of clear supernatant. The EC was measured with the help of EC meter. The unit of EC of soil is expressed in terms of ds/m (Jackson, 1973).

3.4.3.3 Cation Exchange Capacity (CEC)

CEC of initial soil sample was measured by leaching the soil with neutral NH_4OAc solution which saturate the soil with NH_4^+ ion, followed by removal of excess salt with alcohol. The exchanged NH_4^+ ion was distilled and evolved NH3 gas absorbed in a known volume of standard acid.To find out the CEC of initial soil samples (De Villiers and Jackson, 1967), the excess of standard acid is again titrated against a standard alkali.

3.4.3.4 Estimation of Organic Carbon content of soil

Organic carbon content of soil is determined using the Walkley and Black methods, in which oxidizable organic carbon content can be determined rather than total organic carbon content. Normally 1 g of soil (both initial and post-experimental soil) was used in 500 mL conical flasks where 10 mL 1 N $K_2Cr_2O_7$ (potassium dichromate) and 20 mL of laboratory standard concentrated H_2SO_4 (sulphuric acid) were added and gently swirled to mix them well. The flasks were then left in a dark chamber for 30 minutes to proceed the reaction. The flasks were taken out after half an hour, and 200 mL of distilled water was added to stop the further reaction. Then a pinch of NaF (sodium fluoride) was added in each conical flask and an indicator of 1 mL of DPA (Di-Phenyl Amine) was added which also gave a dull violet color of the Suspension. Titrated the conical flasks against 0.5 N FAS (Ferrous Ammonium

Sulphate) until the color of the suspension changed from violet to bright green. A blank titration had also been performed without soil. The following formula for estimation of organic carbon given by Walkley and Black in 1934,

$$\text{Organic C (\%)} = \frac{(B–T) \times 0.003 \times 100}{2 \times \text{wt. of soil}}$$

Where, B = Vol. of 0.5 N FAS solution used for Blank titration

T = Vol. of 0.5 N FAS solution used for Sample titration

3.4.3.5 Available Nitrogen content of Soil

Available Nitrogen (N) was tested with alkaline KMnO4 (Potassium permanganate) method (Subbiah and Asija, 1956). 20 g of soil sample (both initial and soil after experiment) was taken in Kjeldahl flask. 10 mL of water was added to wash out the adhering soil at sides and to moist the soil. Then 100 mL of 0.32 percent KMnO4 was added in conical flask with 5-7 glass beads and 2 mL of paraffin liquid to check the bumping during heating. Another 25 mL of 2 percent boric acid mixed indicator was taken in 250 mL conical flask and put below the receiver tube of Kjeldahl apparatus where 2.5 percent of NaOH (sodium hydroxide) was set previously. After placing the Kjeldahl flask in inlet tube of Kjeldahl apparatus and setting the apparatus with 100 mL 2.5 percent NaOH, the whole set is switched on for some time until 100 mL of bluish green distillate was collected.Then the apparatus was switched off and distillate was titrated with 0.02 N H_2SO_4 until the pink colour developed. By using the following formula,

$$\text{Available N (kg ha}^{-1}) = \frac{(S- B) \times 0.02 \times 14 \times 10^6 \times 2.24}{20 \times 1000}$$

Where, S = Sample reading;

B = Blank reading

3.4.3.6 Available phosphorus

Available P determination done by Olsen's method (1962). 2 g of soil samples were taken in a 100 mL conical flask, with a pinch of Darco G-60. $NaHCO_3$ (pH 8.5)

was then added to 40 mL 0.5 M and shaken in mechanical shaker for 30 minutes. After that suspension was filtered with Whatman filter paper No. 1 and 5 mL of filtrate was transferred in a 25 mL volumetric flask with 5 mL 1.5% ammonium molybdate reagent. The flask was shaken carefully to expel the CO_2 out. After completion of frothing, distilled water was used to wash down the sides and to make the volume up to 20 mL. Then 1 mL 40% $SnCl_2$ solution was added and volume was made up with distilled water up to the mark. The blue colour intensity was measured at 660 nm wave length using a red filter in a spectrophotometer. But firstly 5 standard reading was measured before the sample reading. Then the calculation follows,

Available phosphorus (kg ha^{-1}) in soil = (A × 5 × 2.24)/ (5 × 2) = A × 1.12

Where, A = Concentration of P (mg/kg) as read from the standard curve against the observed absorbance.

3.4.3.7 Available potassium

Available potassium content of soil was determined by Flame Photometer (1 N ammonium acetate extract) method (Jackson, 1973). Five gram soil was transferred in a 100 mL conical flask and 25 mL of 1 N ammonium acetate solution was added and it was shaken for 5 minutes. The suspension was then filtered through Whatman No. 1 filter paper and potassium concentration in the filtrate was measured using flame photometer. First standard reading was taken followed by sample reading. Then the available potassium was calculated by following,

Dilution factor = 25 / 5 = 5 times

Reading of the flame photometer for the test sample = R

Concentration of K in the sample from standard curve against the reading,

R = C

Available K (kg ha^{-1}) = C × 5 × 2.24 = C × 11.2

Physio-chemical properties of the experimental soil revealed the soil was sandy loam in texture with alluvial pH (7.5) with low in available N, medium in available P and low in available K (Table 3.4)

Table 3.4 Analysis of Physico-chemical properties of soil

Physio-chemical parameters	Values
Bulk Density (Mg m^{-3})	1.46
Particle Density (Mg m^{-3})	2.56
Water Holding Capacity (%)	41.2
Sand (%)	46.6
Silt (%)	31.46
Clay (%)	21.73
Soil Texture	Sandy loam
pHw (1:2.5)	7.5
EC (1:2)(ds m^{-1})	0.42
CEC (Cmol p+ kg^{-1})	32.24
Organic Carbon (%)	0.45
Available N (kg ha^{-1})	129.5
Available P (kg ha^{-1})	35.4
Available K (kg ha^{-1})	60.8

3.5 Study of plants

3.5.1 Growth parameters

3.5.1.1 Height of the plants

The height of plant was measured from the surface of soil to the tip of plant with the help of a meter scale at 30, 45 and 60 days after transplanting.

3.5.1.2 Dry weight of plants

After harvesting plant samples were kept in paper bags and dried in hot air oven at 60 ± 2°C till the constant weight.

3.5.1.3 Analysis of root

Root systems were separated from shoots and the fresh root biomass was weighed immediately.

Roots were carefully separated from soil by washing and flooding over sieves. After cleaning of any foreign material, roots were preserved in 20 per cent ethanol for measurement of root length by line interception method of Tennant (1975), using the formula:

$$RL= (11/14) \times NXG$$

Where, N is total numbers of intercepts of root with vertical and horizontal grid lines; G is grid square dimensions, cm; RL is root length, cm.

3.5.1.4 Estimation of chlorophyll content

The chlorophyll content was estimated at 60 days after sowing of wheat crop by following procedure of Arnon (1949). The leaf sample from sample plants were selected and 0.5 g weighed. The leaves were macerated with 80% acetone in a pestle and morter and then it was filtered by Whatman No. 1 filter paper and collected the supernatant. The volume of supernatant raised up to 50ml and absorbance was recorded at 663nm and 645nm spectrophotometer and from the absorbance (A) values. Total chlorophyll content was determined as follows. Using a

$$\text{Total Chlorophyll}= (20.2 \times A\ 645) + (8.02 \times A\ 663) \times V/W \times 1/1000$$

3.5.1.5 Yield/pot

Fruit yield/pot was recorded after maturity of fruits.

3.5.1.6 Harvesting

At maturity, the plants were harvested. Harvesting was done on 15th December, 2019.

3.6 Nutrient use efficiency

3.6.1 Agronomic use efficiency (AUE)

AUE is the outcome of the efficiency of nutrient recovery from applied nutrient and the physiological use efficiency (PUE X ANR). AUE can be increased by nutrient, crop, and soil management practices that alter PUE, ANR or both. Agronomic use efficiency of applied nutrient (kg yield increase per kg nutrient applied). AUE can be calculated by using the following formula,

AUE (kg/kg) = {(Yield of treatment kg – Yield of control in kg)/ Fertilizer nutrient applied in kg}

3.7 Statistical analysis

Statistical analysis of data was carried out using Microsoft Excel (version 2010). One way ANOVA was prepared along with F-test to calculate and compare treatment means and critical difference values in order to see whether the test is significant or not.

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Chapter-IV

RESULTS AND DISCUSSION

The present research work entitled "**Co-inoculated seedling biopriming with *Trichoderma* and Mycorrhiza on improves crop growth in tomato and brinjal**" has been carried out in the net house of Department of Soil Science and Agricultural Chemistry, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi during rabi season in the year of 2019-2020. This chapter elucidates results and discussions about variation in research parameters due to biopriming effect of *Trichoderma viridae* and mycorrhiza in isolation with varying doses of inorganic fertilizers on different morphological features (root length, plant height, dry weight, fruit yield and chlorophyll content), crop uptake of nutrients from soil and nutrient use efficiency of nitrogen, phosphorus and potassium. The data from the experiment on tomato and brinjal were analyzed statistically using CRD design to elaborate and explain the results.

4.1.1 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded doses of NPK fertilizers on plant height (cm) of Tomato

Plant's vegetative growth was affected significantly through *Trichoderma viridae* and mycorrhiza co-inoculated with results biopriming along with different levels of NPK fertilizers in the integrated crop and the management module are discussed in the following section

4.1.1 Plant height

Based on the data collected on different growth stages at 30 DAT (vegetative stage), 45 DAT (flowering stage) and 60 DAT (fruiting stage) different plant height values are presented in table 4.1.1 and depicted in figure 4.1.1.

At vegetative state (30 DAT), maximum plant height from the ground surface was attained at T_5 (29.66 cm; seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) which is at par with T_6 (27.5 cm; RDF) with the lowest value

attained at T_1 (19.66 cm; control). Treatments T_2 (20.83 cm; 75% RDF of N: P: K), T_3 (21.66 cm; seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) and T_4 (24.83 cm; seed biopriming with *T. viridae* + 85% RDF of NPK) are at par with each other and are significantly higher than T_1 (19.66 cm; control).

At flowering stage (45 DAT), maximum plant height from the ground surface was attained at T_6 (56 cm; RDF of N: P: K) which is at par with T_4 (51.66 cm; 80% RDF) and T_5 (51.66 cm; 90% RDF) with the lowest value attained at T_1 (32 cm; control). Treatment T_2 (44.66 cm; 75% RDF) and T_4 (49.33cm; 80% RDF) are at par with each other and are significantly higher than T_1 (32 cm; control).

Plate 3: Height of tomato crop at 45 Days after transplanting

At fruiting stage (60 DAT), maximum plant height from ground surface was observed at T_5 (81.5 cm; Seedling biopriming with *T. viridae* and mycorrhiza +90% RDF), which is at par with T_4 (77.6 cm; Seedling biopriming with *T. viridae* and mycorrhiza +85% RDF) and T_6 (72.6 cm; RDF) with the lowest value attained at T_1 (47.5 cm; control). Treatment T_2 (59 cm; Seedling biopriming with *T. viridae* and mycorrhiza +75 % RDF) and T_3 (67 cm; Seed biopriming with *T. viridae* and mycorrhiza +80% RDF) are at par with each other and are significantly higher than T_1 (47.5 cm; control). Highest value was observed in T_5 (81.5 cm) whereas lowest plant height was seen in T_1 (47.5 cm).

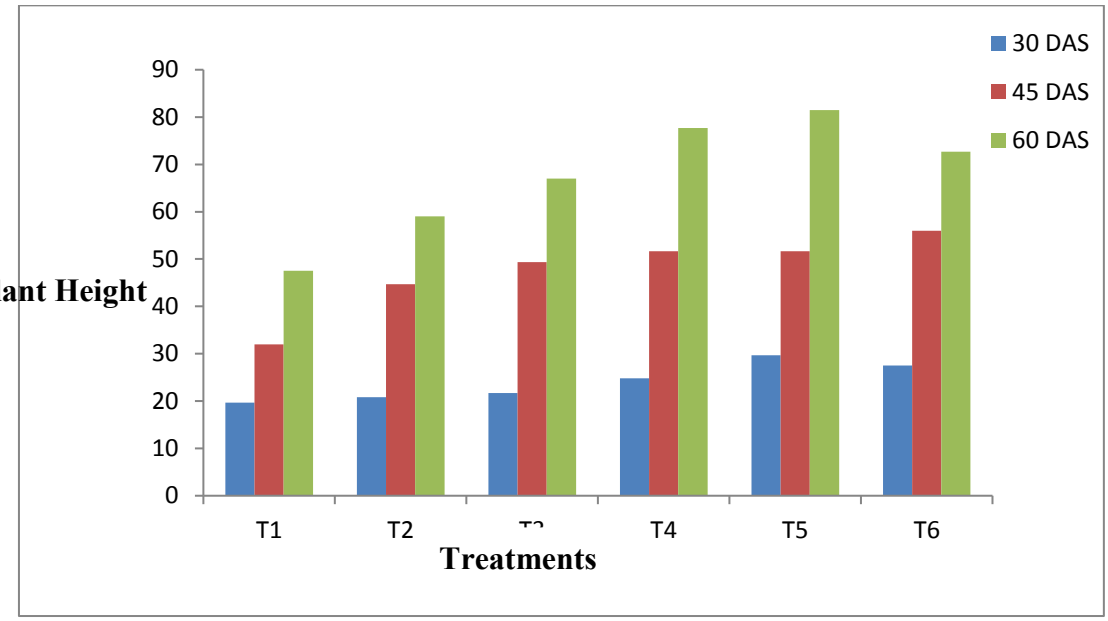
TABLE 4.1.1 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded doses of NPK fertilizers on plant height (cm) of tomato

TREATMENT	Mean value of plant height (cm)		
	30 DAT	**45 DAT**	**60 DAT**
T_1	19.66	32	47.5
T_2	20.83	44.66	59
T_3	21.66	49.33	67
T_4	24.83	51.66	77.66
T_5	29.66	51.66	81.5
T_6	27.5	56	72.66
SEm±	0.509	1.981	0.638
CD (P=0.05)	1.56	1.96	6.10

DAT: Days after transplanting

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.1.1 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded doses of NPK fertilizers on plant height (cm) of tomato



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

The increase in plant height due to the inclusion of *Trichoderma viridae* as a biopriming agent with graded N: P: K and recommended doses of fertilizers can be related to increased availability of nutrients for crops due to the extensive root network of the fungus, the development of secondary metabolites and the growth of hormones such as auxin (Harman *et al.,* 2004).In addition to increased photosynthetic activity, carbohydrate metabolism and greater nutrient absorption, also contributed in the enhancement Stewart and Hill (2014) stated that growth in plant growth can be attributed to the hormonal balance of growth hormone like Indole acetic acid (IAA), ethylene and GA by *Trichoderma* inoculants.

4.1.2 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded doses of NPK fertilizers on leaf chlorophyll content of tomato

4.1.2 Chlorophyll content

Chlorophyll content of different treatment was recorded at 30, 45 and 60 DAT using SPAD meter. Data are shown in table 4.1.2 and depicted in figure 4.1.2.

At vegetative stage (30 DAT), maximum chlorophyll content was recorded in T_6 (39.666; RDF of N:P:K) which was significantly higher than all other treatments whereas T_1 (21.666; control) showed least chlorophyll content which was significantly lower than other treatments. Each treatment differed significantly in respect to other treatments and general trend of decreasing chlorophyll content at 30 DAT followed the order T_6 (39.666; RDF of N:P:K) > T_5 (36.663; seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) > T_4 (36; seedling biopriming with *T. viridae* and mycorrhiza + 85% RDF of NPK) > T_3 (34; seedling biopriming with *T. viridae* and mycorrhiza + 80% RDF of NPK) > T_2 (28.333; seedling biopriming with *T. viridae* and mycorrhiza + 75% RDF of NPK) > T_1 (21.666; control).

Plate 4 : SPAD meter reading of tomato crop at 45 DAT

At flowering stage (50 DAT), maximum chlorophyll content was observed in T_6 (45.666) followed by T_5 (44.333), T_4 (43.333) and T_3 (41.666).T6 was significantly higher than all other treatments. Lowest value of chlorophyll content was recorded in T_1 (30). T_4 (43.333) and T_5 (44.333) are at par with each other but significantly higher

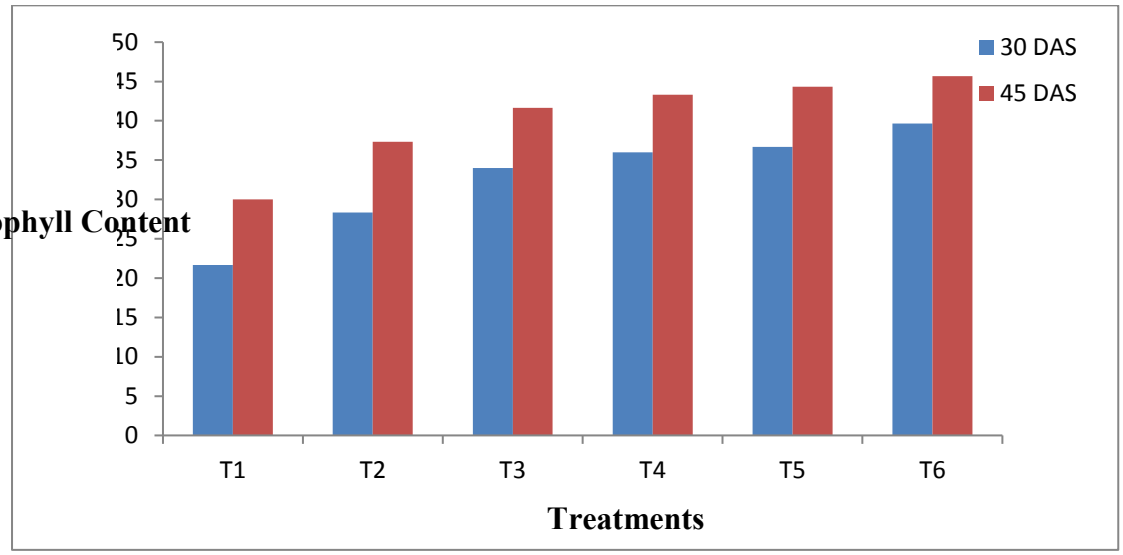
than T_2 (37.333) and T_1 (30) whereas T_2 (37.33) and T_1 (30) are at par with each other, indicating that the chlorophyll content did not vary significantly.

Table 4.1.2: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on leaf chlorophyll content of tomato

TREATMENT	Mean value of SPAD reading	
	30 DAT	45 DAT
T_1	21.66	30
T_2	28.33	37.33
T_3	34	41.66
T_4	36	43.33
T_5	36.66	44.33
T_6	39.66	45.66
SEm±	1.90	1.146
CD (P=0.05)	5.870	3.53

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.1.2 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded doses of NPK fertilizers leaf chlorophyll content of tomato



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

From the data collected from experiment, general view has shown that chlorophyll content from vegetative (30 DAT) to flowering (45 DAT) has increased. There was promising effect of *Trichoderma viridae* and mycorrhiza seedling biopriming with graded level of NPK fertilizers on chlorophyll content of leaves due to enhanced uptake of N, P and K through *Trichoderma* and mycorrhiza mediated extensive root-hyphae network. It has been reported that effect of amount of N on chlorophyll content was greater for full RDF than lower doses of N along with seedling biopriming with *Trichoderma viridae* and mycorrhiza.

4.1.3 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on root length (cm) of Tomato

4.1.3 Root length

Generally roots of tomato were confined to the immediate neighbourhood of the respective plant. Data of root length at vegetative stage (30 DAT), flowring stage (45DAT), fruiting stage (60 DAT) of tomato are given in the table 4.1.4 and depicted in figure 4.1.3 and 4.1.3

At vegetative stage (30 DAT), it was evident that T_6 (34.333 cm; RDF of NPK) showed maximum root length and it was at par with T_4 (32.33 cm; seedling bio-primig with *T. viridae* and mycorrhiza + 85% RDF of NPK) and T_5(33.33 cm; seedling biopriming with *T.viridae* and mycorrhiza + 90% of RDF).T_2 (28 cm; seedling biopriming with *T.viridae* and mycorrhiza +70% of RDF) and T_3 (31.5 cm; seedling biopriming with *T.viridae* and mycorrhiza + 80% of RDF) were at par with each other and did not significantly differ with each other. The lowest value of root length attained at T_1 (24.7 cm; control).

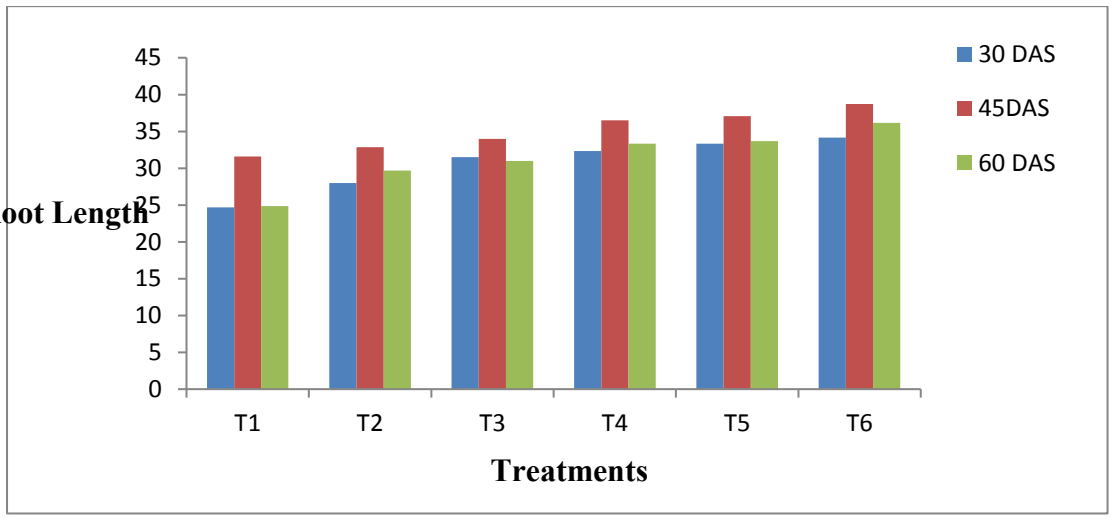
Plate 5: Root Length of tomato Crop at 60 days after transplanting

Table 4.1.3: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on root length (cm)

TREATMENT	Mean value of Root length (gm)		
	30 DAT	45 DAT	60 DAT
T_1	24.7	31.6	24.83
T_2	28	32.83	29.66
T_3	31.5	34	31
T_4	32.33	36.5	33.33
T_5	33.33	37.06	33.66
T_6	34.16	38.73	36.16
SEm±	0.833	0.228	0.509
CD (P=0.05)	2.567	0.704	1.568

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.1.3: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on root length (cm)



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Increase in root size by *Trichoderma* can be concluded due to production of indole acetic acid (IAA) derivatives along with ethylene destruction into α-ketobutyrate and ammonium (Harman *et al.,* 2004) and production of other secondary metabolites. Results are in agreement with Singh *et al*. (2003) who found out that application of *Trichoderma* increases deep tap-root system which in fact helps plant to uptake more nutrients along with water.

4.1.4 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizer on plant dry weight (g plant^{-1}) of tomato

4.1.4 Plant Dry Matter

At peak vegetative stage (30 DAT) maximum dry weight was recorded in T_6 (2.5 g plant^{-1}; RDF;seedling biopriming with *Trichoderma viridae* and mycorrhiza + 90% RDF of NPK) which was at par with T_5 (2.47 g plant^{-1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 90% of RDF). Lowest value was attained at T_1 (0.82 g plant^{-1}; control).T_2 (1.15 g plant-^{1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 75% RDF of NPK) and T_3 (1.6 g plant^{-1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 80% of RDF) did not differ significantly. Also T_4 (2.4 g plant^{-1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 85% RDF of NPK) did not differ significantly than T_5 (2.47 g plant^{-1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 90% of RDF).

At flowering stage (45 DAT), maximum plant dry weight was produced by T_6 (3.13 g plant-^{1}) and lowest dry weight was attained at T_1 (1.3 g plant-^{1}). T_3 (2.33 g plant-^{1}), T_4 (2.96 g plant-^{1}) and T_5 (7.7 g plant-^{1}) are at par with each other. T_2 (1.9 g/plant) was at par with T_3 (2.33 /plant) and did not differ significantly with each other.

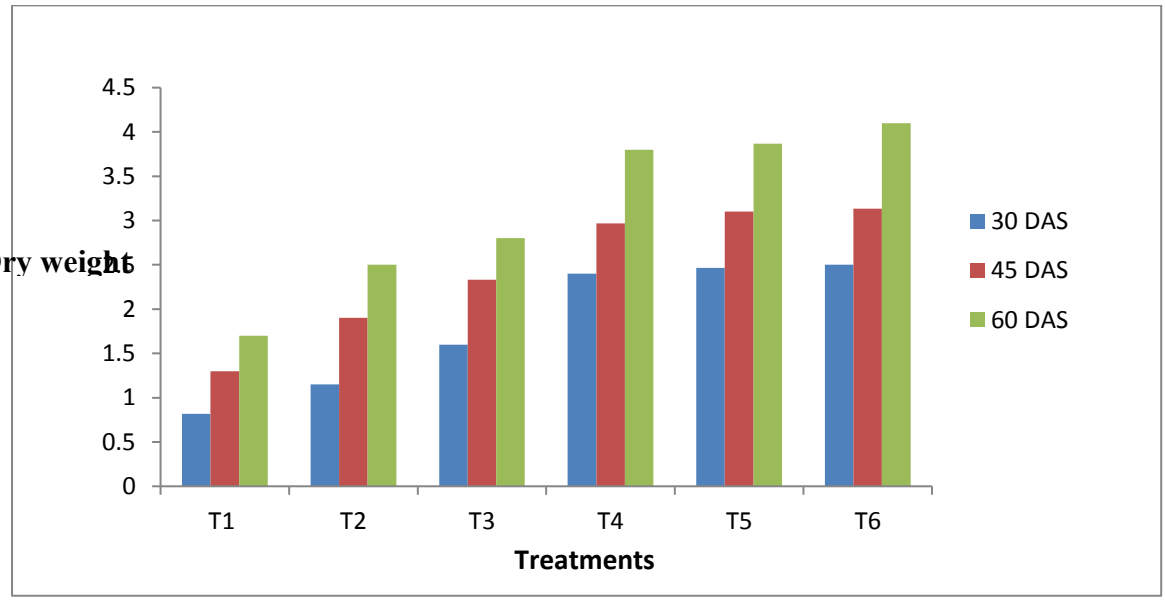
At fruiting stage (60 DAT), maximum dry weight of plant was attained at T_6 (4.1 g plant^{-1}) whereas lowest dry weight was attained at T_1 (1.7 g plant^{-1}). Again T2 (2.5 g plant^{-1}), T_3 (2.8 g plant-1) do not varied significantly among each other but they are significantly lower than T_4 (3.8 g plant^{-1}) and T_5 (3.87 g plant^{-1}) which was at par with T_6 (4.1 g plant^{-1}).

Table 4.1.4: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on plant dry weight (g plant^{-1}) of tomato

TREATMENT	Mean value of DRY WEIGHT (gm)		
	30 DAT	45 DAT	60 DAT
T_1	0.82	1.3	1.70
T_2	1.15	1.9	2.50
T_3	1.60	2.33	2.80
T_4	2.40	2.96	3.80
T_5	2.47	3.1	3.87
T_6	2.50	3.13	4.10
SEm±	0.0703	0.0824	0.0638
CD (P=0.05)	0.216	0.254	0.196

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.1.4: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on plant dry weight (g plant^{-1}) of tomato



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

From the results, it was evident that tomato plants bio-primed with *Trichoderma* in combination with graded levels of NPK fertilizers showed increased dry matter production due to biopriming mediated enhanced nutrient uptake and production of various growth promoting secondary metabolites and growth hormones (Harman *et al.,* 2004). Mycorrhiza production by *Trichoderma* results in modification of tomato root system in bio-primed seeds (T2, T3, T4 and T5) with hyphal extention which increases higher nutrient uptake causing more plant biomass production (Parihar *et al*.,2019). Similar results were found in works done by Srinath *et al*. (2003) on banana dry matter production, Aseri *et al.* (2008) on pomegranate and Sharma *et al*. (2012) on wheat crop.

4.1.5 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on fruit yield (g pot^{-1}) of tomato

4.1.5 FRUIT YIELD

Data on tomato fruit yield (g pot^{-1}) is presented in table 4.1.5 and depicted in figure 4.1.5. and 4.1.5.

After harvest at 60 DAT, highest yield value was attained at T_6 (63.66 g pot-^{1}; RDF of NPK) followed by T_5 (58.33 g pot^{-1};seedling biopriming with *Trichoderma viridae* and mycorrhiza + 90% of RDF), T_4 (55 g pot^{-1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 85% RDF of NPK), T_3 (41 g pot-^{1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 80% RDF of NPK), T_2 (36 g pot-^{1}; seedling biopriming with *Trichoderma viridae* and mycorrhiza + 75% RDF of NPK) and T_1 (28.33 g pot-^{1}; control). T_6 (63.66 g pot-^{1}) and T_5 (58.33 g pot-^{1}) are at par with each other and T_5 (58.33 g pot-^{1}), & T_4 (55 g pot-^{1}) did not differ significantly with each other.

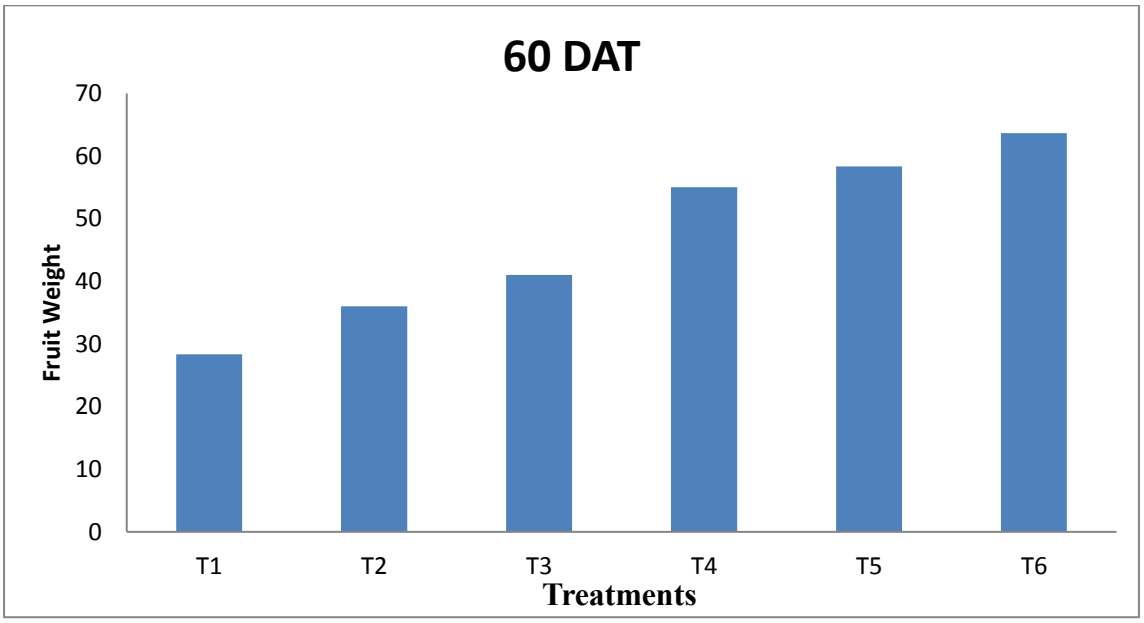
It can be clearly concluded that maximum yield was attained full RDF of NPK followed by *Trichoderma viridae* seed biopriming along with 90% NPK application. Full RDF application without *Trichoderma* seed biopriming had resulted in considerable yield but it differed significantly from other treatments bio-primed with *Trichoderma viridae* with graded levels of NPK fertilizers except for T_2. Similar results were found by Egberongbe *et al.,* (2010) and Entesari *et al.,* (2013) on soybean. Study on fruit yield of cucumber and chilli was reported to have promising result by Saeed *et al*. (2015) and Thilagar *et al.* (2016) respectively.

Table 4.1.5 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on fruit weight (gm)

TREATMENT	Fruit weight of tomato (gm)
	60 DAT
T_1	28.3
T_2	36
T_3	41
T_4	55
T_5	58.3
T_6	63.7
SEm±	0.881
CD (P=0.05)	2.717

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.1.5. Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on fruit weight (gm)



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

It can be clearly concluded that maximum yield was attained at RDF followed by *Trichoderma viridae* and mycorrhiza seedling biopriming along with 90% of RDF application. Full RDF application without *Trichoderma viridae* and mycorrhiza seedling biopriming had resulted in considerable yield but it did not differed significantly from other treatments bio-primed with *Trichoderma viridae* and mycorrhiza seedling biopriming with graded levels of NPK fertilizers except for T6. Similar results were reported by Egberongbe *et al.,* (2010) and Entesari *et al.,* (2013) on soybean. Study on fruit yield of cucumber and chilli was reported to have promising result following PGPR and fertilizer combination by Saeed *et al*. (2015) and Thilagar *et al.* (2016) respectively.

4.2 BRINJAL

4.2.1 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on plant height (cm) of brinjal

4.2.1 Plant height

Based on the data collected on different growth stages at 30 DAT (vegetative stage) , 45 DAT (flowering stage)and 60 DAT (fruiting stage) different plant height values are presented in table 4.1.1 and depicted in figure 4.1.1.

At vegetative stage (30 DAT), maximum plant height from the ground surface was attained at T_5 (29.66 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF) which is at par with T6(27.5 cm; RDF) with the lowest value attained at T_1 (19.66 cm; control). Treatments T_2 (20.83 cm; Seedling biopriming with *T. viridae* and mycorrhiza 75% of RDF), T_3 (21.66 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) and T_4 (24.83 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF) and are significantly higher than T1 (19.66 cm; control) .

Plate 6: Height of brinjal crop at 45 DAT

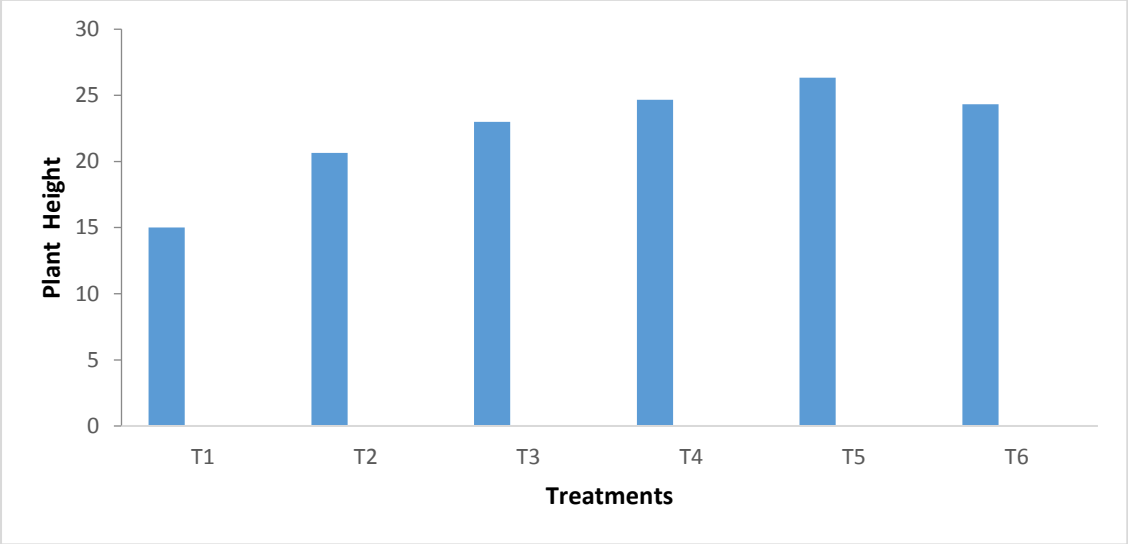
At flowering stage (45 DAT), maximum plant height from the ground surface was attained at T_6 (56 cm; RDF of NPK) which is at par with T_4 (51.66 cm; 80% RDF) and T_5 (51.66 cm; 90% RDF) with the lowest value attained at T_1 (32 cm; control). Treatment T_2 (44.66 cm; 75% RDF) and T_4 (49.33cm; 80% RDF) are at par with each other and are significantly higher than T_1 (32 cm; control).

4.2.1 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on plant height of brinjal (cm)

TREATMENT	Mean value of plant height (cm)
	45 DAT
T_1	15
T_2	20.66
T_3	23
T_4	24.66
T_5	26.33
T_6	24.33
SEm±	0.9229
CD (P=0.05)	2.843

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.2.1Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on plant height of brinjal (cm)



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

The increase in plant height due to the inclusion of *T. viridae* and mycorrhiza *Trichoderma* viridae as a biopriming agent with graded N: P: K doses of fertilizers can be related to increased availability of nutrients for crops due to the extensive root network of the fungus, the development of secondary metabolites and the growth of hormones such as auxin (Harman *et al.,* 2004).In addition to increased photosynthetic activity, carbohydrate metabolism and greater nutrient absorption contribute to this increase in plant height.

4.2.2 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on leaf chlorophyll content of brinjal

At vegetative stage (30 DAT), maximum chlorophyll content was recorded in T_5 (44.33; 90% RDF of N: P: K) which was significantly higher than all other treatments whereas T_1 (30.33; control) showed least chlorophyll content which was significantly lower than other treatments. Each treatment differed significantly in respect to other treatments and general trend of decreasing chlorophyll content at 30 DAT followed the order T_5 (44.33; Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF) >T_6 (42;RDF of NPK) > T_4 (42.33 Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF) > T_3 (40.33; Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) > T_2 (36.66; Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF) > T_1 (30.33; control).

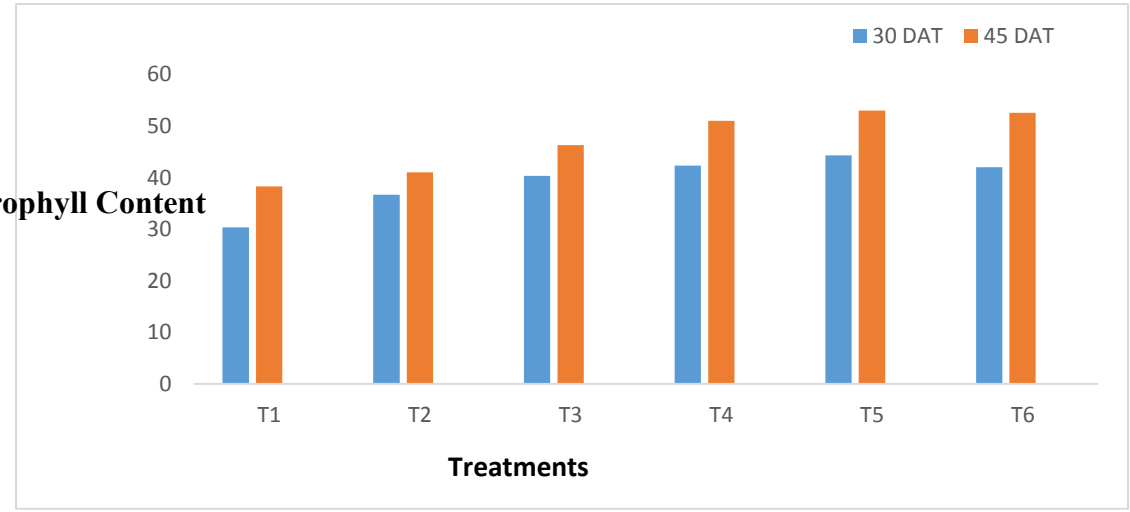
At flowering stage (45 DAT), maximum chlorophyll content was observed in T_5 (53) followed by T_6 (52.16), T_4 (51) and T_3 (46.333). T_5 was significantly higher than all other treatments. Lowest value of chlorophyll content was recorded in T_1 (38.33). T_4 (51) and T_5 (53) are at par with each other but significantly higher than T_2 (41) and T_1 (38.33) whereas T_2 (41) and T_1 (38.33) are at par with each other, indicating that the chlorophyll content did not vary significantly.

Table 4.2.2 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on leaf chlorophyll content of brinjal

TREATMENTS	Mean value of SPAD reading	
	30 DAT	45 DAT
T_1	30.33	38.33
T_2	36.66	41
T_3	40.33	46.33
T_4	42.33	51
T_5	44.33	53
T_6	42	52.6
SEm±	0.6938	0.6948
CD (P=0.05)	2.138	2.1409

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.2.2 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on leaf chlorophyll content of brinjal



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

From the data presented above it is pertinent, that chlorophyll content from vegetative (30 DAT) to flowering (45DAT) has increased. Because of increased uptake of N, P, and K through *Trichoderma* mediated extended root-hyphae network, there was a positive effect of *Trichoderma viridae* seed biopriming with graded levels of NPK fertilisers on chlorophyll content of leaves. It has been shown that lower dosages of N combined with seed biopriming with *Trichoderma viridae* had a stronger effect on chlorophyll content than full RDF of NPK.

4.2.3 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on root length (cm) of brinjal

4.2.3 Root length

Generally roots of brinjal were also confined to the immediate neighbourhood of the respective plant like tomato. Data of root length at vegetative stage (30 DAT), flowring stage (45 DAT),fruiting stage (60 DAT) of brinjal are given in the table 4.2.5 and depicted in figure 4.1.5.1 and 4.2.5.2.

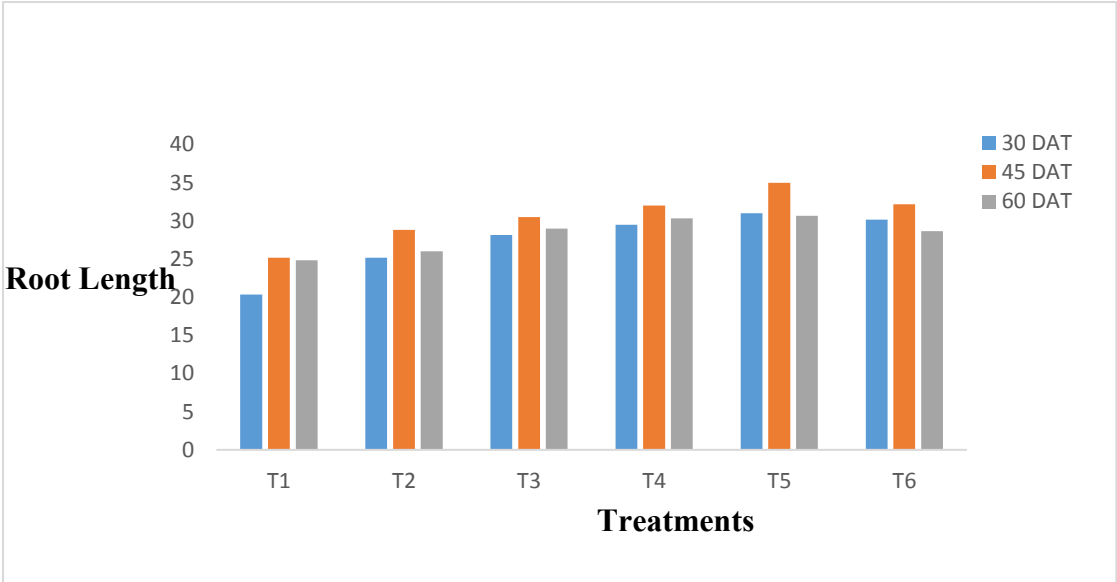
At vegetative stage (30 DAT), it was evident that T_5 (31 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) showed maximum root length and it was at par with T_6 (30.1 cm; Seedling biopriming with *T. viridae* and mycorrhiza + RDF of NPK) and T_4 (29.5 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF). T_2 (25.1 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF) and T_3 (28.1 cm; Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) were at par with each other and did not significantly differ with each other. The lowest value of root length attained at T_1 (20.3 cm; control).

Table 4.2.3 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on root length (cm) of brinjal

TREATMENT	Mean value of Root length (cm)		
	30 DAT	45 DAT	60 DAT
T_1	20.36	25.16	24.83
T_2	25.16	28.83	26
T_3	28.16	30.5	29
T_4	29.5	32	30.33
T_5	31	35	30.66
T_6	30.16	32.16	28.66
SEm±	0.4946	0.5892	0.4614
CD (P=0.05)	1.524	1.8156	1.4219

(T1: Control; T2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.2.3 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on root length (cm) of brinjal



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Plate 7: Root length of brinjal at 60 days after transplanting

The secretion of indole acetic acid (IAA) derivatives, as well as the breakdown of ethylene into ketobutyrate and ammonium and the production of other secondary metabolites, can be attributed to *Trichoderma*'s increase in root growth. Singh *et al* (2003) explain that application of *Trichoderma* improves the depth of the tap-root system, allowing the plant to absorb more nutrients and water. Co-inoculation with mycorrhiza further helps in greats exploration of soil volumes and increases the surface area as well as root length.

4.2.4 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on plant dry weight (g plant^{-1}) of brinjal

Dry weight of Brinjal

Data obtained on plant dry weight (g plant^{-1}) from different treatments of the experiment are shown in table and depicted in figure

At peak vegetative state (30 DAT) maximum dry weight was recorded in T_5 (7.0 g plant^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) which superior that with T_6 (6.2 g plant^{-1}; RDF of NPK). Lowest value was attained at T_1 (2.2 g plant^{-1}; control). T_2 (3.25 g plant^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 75% RDF of NPK) and T_3 (4.5 g plant^{-1}; Seedling biopriming with *T.*

viridae and mycorrhiza + 80% RDF of NPK) did not differ significantly. Also T_4 (5.8 g plant^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 85% RDF of NPK) was less than T_5 and T_6.

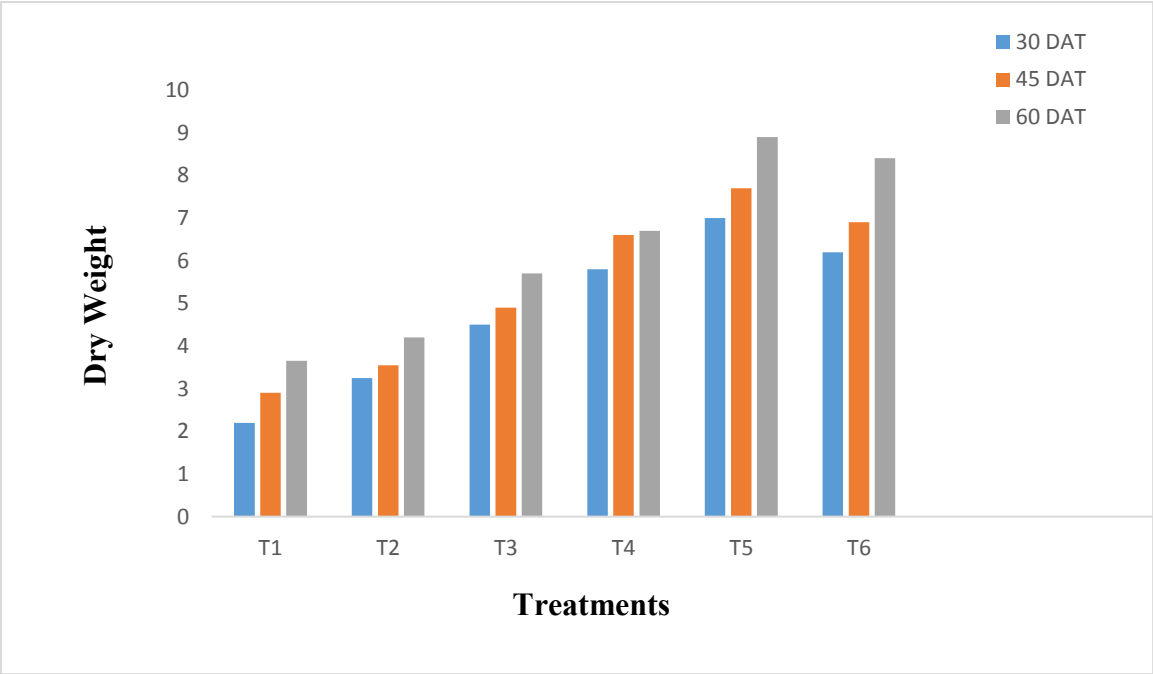
At flowering stage (45 DAT), maximum plant dry weight was poroduced by T_5 (7.7 g plant^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) and lowest dry weight was attained at T1(2.9 g plant^{-1})..further at fruiting stage (60 DAT), maximum dry weight of plant was attained at T_5 (8.9 g plant^{-1}) whereas lowest dry weight was attained at T_1 (3.65 g plant^{-1}). Again T_2 (4.2 g plant^{-1}), T_3 (5.7 g plant^{-1}) do not varied significantly among each other but they are significantly lower than T_4 (6.7 g plant^{-1}) and T_6 (8.4 g plant^{-1}) which was at par with T5 (8.9 g plant^{-1}).

Table 4.2.4 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on plant dry weight (g plant^{-1}) of brinjal

TREATMENT	Mean value of dry weight (g)		
	30 DAT	45 DAT	60 DAT
T_1	2.2	2.9	3.65
T_2	3.25	3.55	4.2
T_3	4.5	4.9	5.7
T_4	5.8	6.6	6.7
T_5	7	7.7	8.9
T_6	6.2	6.9	8.4
SEm±	0.03849	0.04906	0.226
CD (P=0.05)	0.1186	0.15118	0.0736

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.2.4 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on plant dry weight (g plant^{-1}) of brinjal



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

From the above results, it is evident that brinjal plants bio-primed with *Trichoderma* and mycorrhiza in combination with graded levels of NPK fertilizers showed increased dry matter production due to biopriming mediated enhanced nutrient uptake and production of various growth promoting secondary metabolites and growth hormones (Harman *et al.,* 2004). Mycorrhiza biopriming results in modification of tomato root system in bio-primed seeds (T_2, T_3, T_4 and T_5) with hyphal extention which increases higher nutrient uptake causing more plant biomass production (Parihar *et al.,* 2019).

4.2.5 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on fruit yield (g pot^{-1}) of brinjal

Data on brinjal fruit yield (g pot^{-1}) is presented in table 4.2.5 and depicted in figure 4.2.5

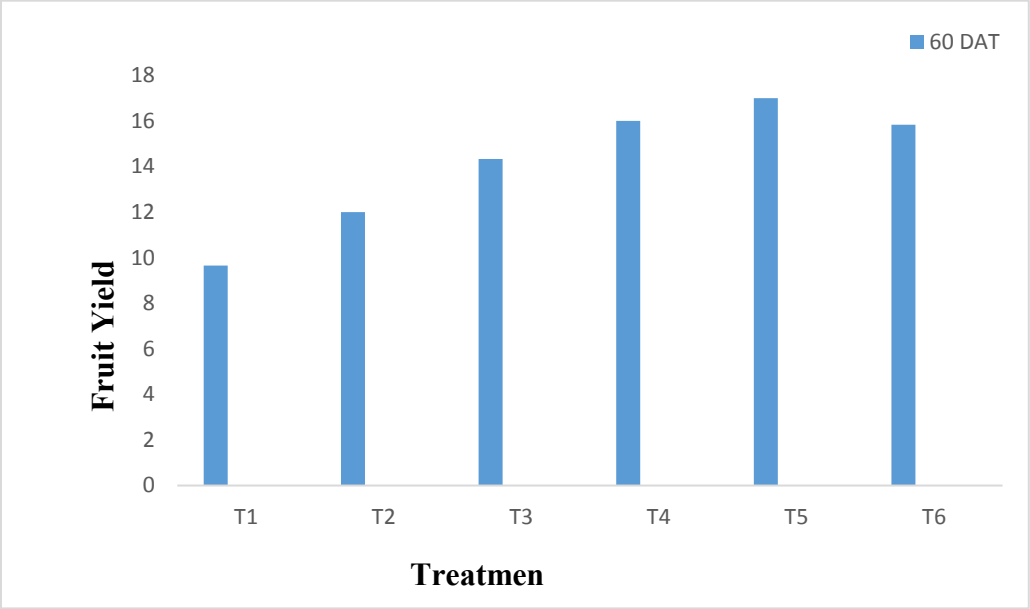
After harvest at 60 DAT, highest fruit yield value was attained at T_5 (17 g pot^{-1}, Seedling biopriming with *T. viridae* and mycorrhiza + 90% RDF of NPK) followed by T_6 (15.83 g pot^{-1}; RDF of NPK), T_4 (16 g pot^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 85% RDF of NPK), T_3 (14.33 g pot^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 80% RDF of NPK), T_2 (12 g pot^{-1}; Seedling biopriming with *T. viridae* and mycorrhiza + 75% RDF of NPK) and T_1 (9.66 g pot^{-1}; control). T_6 (15.83 g pot^{-1}) and T_5 (17 g pot^{-1}) are at par with each other and T_5 (17 g pot^{-1}) and T_4 (16 g pot^{-1}) did not differ significantly with each other.

Table 4.2.5 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on fruit yield (g pot^{-1}) of brinjal

TREATMENT	Fruit weight of brinjal
	60 DAT
T_1	9.66
T_2	12
T_3	14.33
T_4	16
T_5	17
T_6	15.83
SEm±	0.5892
CD (P=0.05)	1.815

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.2.5 Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with different doses of NPK fertilizers on fruit yield (g pot^{-1}) of brinjal



(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

It can be clearly depicted here that maximum yield was attained at *Trichoderma viridae* and mycorrhiza seedling biopriming along with 90% NPK application. Full RDF application without *Trichoderma* seed biopriming had resulted in comparable yield but it did not achieve maximum potential yield. Similar results were found by Egberongbe *et al.,* (2010) and Entesari *et al.,* (2013) on soybean. Study on fruit yield of cucumber and chilli was reported to have promising result by Saeed *et al*. (2015) and Thilagar *et al.* (2016) respectively.

4.3 Agronomic use efficiency

Agronomic use efficiency is the product of the efficiency of nutrient recovery from applied nutrient and the efficiency with which the plant used each additional unit of nutrient acquired. Data pertaining to agronomic use efficiency was recorded at 60 DAT and presented in (Table-4.3.1 and Figure-4.3.1) depicted that agronomic use efficiency was maximum recorded in tomato (4.5 kg kg^{-1} for N, 8.47 kg kg^{-1} for P and 26.5 kg kg^{-1} for K) followed by brinjal (1.19 kgkg^{-1} for N, 2.23 kg kg^{-1} and 7.06 kg $kg^{-1}$for K). Agronomic use efficiency varied between in tomato 0.97 to 4.5, 1.83 to 8.47 and 5.81 to 26.52 kg kg^{-1} for N, P and K, respectively where as in binjal it varied between 0.29 to1.19, 0.55 to 2.23 and 1.76 to 7.06 kg kg^{-1} for N, P and K under alluvial soil respectively in different treatment combinations of T_2 to T_6. Significantly lower agronomic use efficiency was recorded with T_2 (0.97 for N, 1.83 for P and 5.81 for K; Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF) in Tomato, besides this in brinjal crop lower values of AUE was recorded with T_2 (0.29 for N, 0.55 for P and 1.76 for K; Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF) in alluvial soil. Effect of soil with different combination of fertilizer and *Trichoderma viridae* and mycorrhiza interaction effect in soil found to be non-significant *viz* T_6 (4.50 kg kg^{-1} ; RDF) followed by T_5 (3.82 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF), T_4 (3.40 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF), T_3 (1.61 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) and T_2 (0.97 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF) for tomato crop. For brinjal the maximum AUE of N is in order of T_6 (1.19 kg kg^{-1} ; RDF) followed by T_5 (0.29 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF), T_4 (0.80 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF), T_3 (0.59 kg kg^{-1} ; Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF) and T_2 (0.29 kg kg^{-1} ;Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF). AUE of P and K also follow the same trend of order $T_6 > T_5 > T_4 > T_3 > T_2$.Results indicates that seedling biopriming along with Trichoderma viridae and mycorrhiza improves agronomic use efficieny of tomato and brinjal crop by more nutrient uptake from soil through root prolification and boosting metabolism of plant.

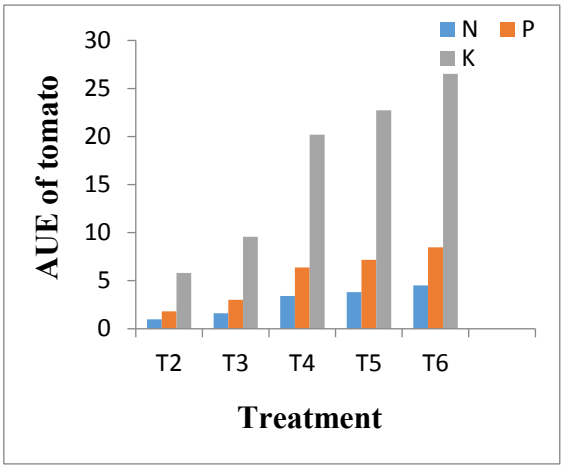
Table 4.3.1: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on agronomic use efficiency (kg kg^{-1}) of nitrogen, phosphorus and potassium in tomato and brinjal

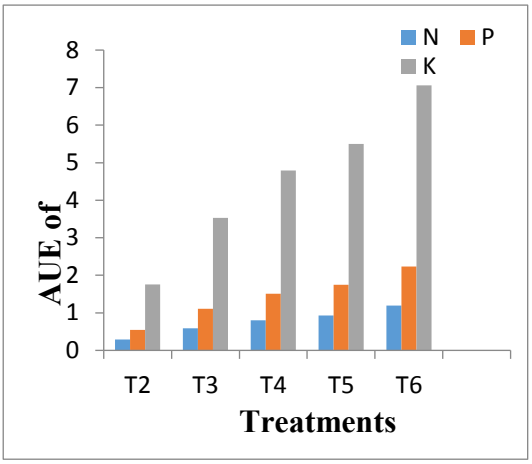
TREATMENT	Tomato			Brinjal		
	N	P	K	N	P	K
T_2	0.97	1.83	5.81	0.29	0.55	1.76
T_3	1.61	3.03	9.57	0.59	1.11	3.53
T_4	3.40	6.39	20.20	0.80	1.51	4.79
T_5	3.82	7.19	22.72	0.93	1.75	5.50
T_6	4.50	8.47	26.52	1.19	2.23	7.06
SEm±	1.202	1.809	5.697	0.254	0.508	1.413
CD (P=0.05)	3.704	5.575	17.555	0.784	1.567	4.35

(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1})

Figure 4.3.1: Effect of seedling biopriming with *Trichoderma viridae* and mycorrhiza along with graded levels of NPK fertilizers on

agronomic use efficiency (kg kg^{-1}) of nitrogen, phosphorus and potassium in tomato and brinjal





(T_1: Control; T_2: Seedling biopriming with *T. viridae* and mycorrhiza + 75% of RDF; T_3: Seedling biopriming with *T. viridae* and mycorrhiza + 80% of RDF; T_4: Seedling biopriming with *T. viridae* and mycorrhiza + 85% of RDF; T_5: Seedling biopriming with *T. viridae* and mycorrhiza + 90% of RDF; T_6: RDF @ 120:80:80 kg ha^{-1}).

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Chapter-V

SUMMARY AND CONCLUSIONS

Nutrient Use Efficiency is an important ecological measure as it integrates a variety of physiological processes in how nutrients taken up by plants are generally used for the production of biomass. Intensive cropping and exhaustive nature of present cropping system in varied agro ecology have led to the depletion of soil organic carbon content and inherent soil fertility resulting in a serious threat to the sustainability of these production systems.

Nowadays seedling biopriming with efficient microorganisms on growth, development, nutrient uptake of crop, nutrient use efficiency and nutrient management practice have given a new dimension to modern agriculture.

The present investigation on tomato and brinjal was carried out in pots in the net house of the Department of Soil science and Agricultural Chemistry, Institute of Agriculture Sciences, Banaras Hindu University, Varanasi, India situated at an altitude of 80.71 mts above mean sea level and at 25°18' North latitude and 80°36 East longitudes.

The experiments were carried out in orders Inceptisol in pots. Seedling biopriming with *Trichoderma viridae* and mycorrhiza along with gradual application of NPK fertilizer were applied. The design of the plot was FCRD. A number of observations pertaining to growth, and development were taken at different growth stages of tomato and barley and yield parameters and yield were recorded at harvest of crop. For determining the significance of difference between treatment means and to draw conclusions, the data obtained by the various observations at different stages of crop growth were subjected to statistical analysis by adopting method of "Analysis of variance" for factorial CRD.

The experiment on "**Co-inoculated seedling biopriming with *Trichoderma* and Mycorrhiza on crop growth in tomato and brinjal**'' was conducted in order to

evaluate biopriming effect on tomato and brinjal. With the collection of data from the experiment and analysis of data statistically, encouraging results were obtained. Based on these results the experimental findings were summarized and concluded in the following section:

- Better plant establishment was observed in case of both tomato and brinjal treated with *Trichoderma viride* and mycorrhiza along with graded levels of recommended fertilizer doses (RDF) than control and in case of brinjal combination of seedling biopriming and 90% full recommended doses of fertilizers emerged as a suitable option compared to full dependence of inorganic fertilizers.

- Root length and root biomass of tomato plants treated with *Trichoderma virid ae* and mycorrhiza have increased substantially thus, biopriming increases root density alter root architecture colonisation in the rhizosphere.

- From the experiment carried out in pots showed that chlorophyll content of tomato and brinjal leaves is not associated with seedling biopriming withf *Trichoderma viridae* and mycorrhiza and maximum chlorophyll content was recorded higher in full RDF.

- Plant height (above ground part) was higher in case of seedling biopriming with *Trichoderma viride* and mycorrhiza along with 90% of RDF followed by full recommended dose of fertilization.

- Dry weight was higher in case of tomato plants in full recommended dose of fertilization in pots contrary to brinjal where, the maximum dry weight was obtained at seedling biopriming *Trichoderma viride* and mycorrhiza along with 90% of RDF.

- Agronomic use efficiency (AUE) of N, P and K were maximum in bio-primed seeds than untreated seeds. Tomato plants having biopriming with varying doses of N, P and K along with *Trichoderma viride* and mycorrhiza seed biopriming results in a better synergistic relation of microbes for nutrients acquisition.

- Yield (fruit weight per plant) was highest with RDF in tomato crop. On the other hand in brinjal, fruit yield was highest with seedling bio-primed with *Trichoderma viridae* and mycorrhiza + 90% of RDF which can be attributed to biopriming which facilitate uptake of nutrients and plants beneficial growth factors than untreated plants resulting in higher yield of brinjal plants treated with *Trichoderma viride* and mycorrhiza. Even with reduced fertilizer doses.

- Comparing the two crops further it was revealed that tomato crop was more agronomically efficient with reference to unit addition of N, P and K. At recommended dosage of fertilizer, tomato was 3.7 times efficient than brinjal. Seedling biopriming coupled with 10% replacement of recommended fertilization enhances the efficiency to 4.1 times, thus implicating the role of bio-primers in improving agronomic use efficiency.

- Seedling biopriming with inorganic fertilization in integrated crop management module of tomato and brinjal not only increased the growth, nutrient use efficiency and yield rather showed apart from better crop stand with supplementation in the fertilizer scheduling. This result can benefit farmers to reduce the input cost without compromising with crop productivity directly and can control environmental pollution due to higher use of agrochemical application.

- The study has also revealed that inclusion of AM fungi with T*r*ichoderma in vegetables like tomato and brinjal crop can enhance the nutrient uptake in plants from soil and sustain higher yield even after application of reduced amount fertilizer nutrients. Thus higher returns per unit input application is achieved than conventional system through combined application of *Trichoderma* and mycorrhiza which has resulted in higher yield in an eco-friendly manner.

- Moreover the experiment must not be misinterpreted with fertilizer replacement rather integrated management of synthetic fertilizer along with application of beneficial microbes.

- The whole study indicated that biopriming can perform best supplemental role only in combination with inorganic nutrient supply.
- Thus study will be further upscaled to field level to assess the real time impact of *Trichoderma* and AMF priming in growth promotion and against different stresses.

Thus Biopriming may play a significant role in LEISA (Low External Input Sustainable Agriculture) and in doubling farmer's income.

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