IMPACT OF SEWAGE WATER IRRIGATION ON SOIL MICROBIAL DIVERSITY AND CAULIFLOWER PRODUCTIVITY IN RAJNANDGAON DISTRICT (C.G.)

M.Sc. (Ag.) THESIS

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DEPARTMENT OF AGRICULTURAL MIROBIOLOGY

COLLEGE OF AGRICULTURE

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NAGESHWAR SAHU

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CERTIFICATE - I

This is to certify that this thesis entitled **"Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)"** submitted for degree of Master of Science, in the subject of **Agricultural Microbiology** to the Indira Gandhi Krishi Vishwavidyalaya, Raipur, is a bonafide research work carried out by **Nageshwar Sahu** under my guidance and supervision and that no part of this thesis has been submitted for any other degree. The assistance and help received during the course has been fully acknowledged.

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CERTIFICATE - II

This is to certify that this thesis entitled **"Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)"** submitted by **Nageshwar Sahu** to the Indira Gandhi Krishi Vishwavidyalaya, Raipur, in partial fulfillment of the requirements for the degree of **Master of Science in Agriculture** in the **Department of Agricultural Microbiology** has been approved by external evaluator and Student's Advisory Committee after oral examination, under the chairmanship of Head of the Department.

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LIST OF NOTATIONS / SYMBOLS

°C	Degree Celsius
%	Per cent
et al.	And others/ co-worker
+	Plus/Add
<	More than
>	Less than
=	Equal to
X	Multiplied by
μ	Mean value
e.g.	For example
—	Minus

ABREVIATION

ha.	Hectare
ppm	Parts per million
EC	Electrical conductivity
C	Carbon
CO_2	Carbon dioxide
DTPA	Diethylene Triamine Penta Acetic Acid
MBC	Microbial biomass carbon
Fig.	Figure
dSm^{-1}	Deci Siemens per metre
Mg/Kg	Milligrams per kilogram
Kg/ha	Kilogram per hectare
g/cm^3	gram per cubic centimetre
N	Nitrogen
P	Phosphorus
K	Potassium
Ca	Calcium
Mg	Magnesium
Zn	Zinc
Fe	Iron
Mn	Manganese
Cu	Copper
OC	Organic Carbon
pH	Negative logarithm of H^+ ion activity
NH_4-N	Ammonium
HCO_3	Bicarbonate
CO_3	Carbonate
Mg/h/g	Milligrams/hours/gram
L	Litre
ml	Milliliter
Tr	Trace
Ag	Silver

Hg	Mercury
Cd	Cadmium
Ni	Nickel
As	Arsenic
Pb	Lead
CV	Coefficient of Variation
S.D.	Standard Deviation

THESIS ABSTRACT

Title of thesis	:	**"Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)"**
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Date: 14/09/2020

Sign. Of Head of the Department

ABSTRACT

A field experiment-based study was conducted entitled **"Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)"** and all observation analyzed at Department of Soil Science and Agricultural Chemistry, Pt. Shiv Kumar Shastry College of Agriculture and Research Station, Rajnandgaon and Department of Agricultural Microbiology, Indira Gandhi Krishi Vishwavidyalaya, Raipur during the year 2019-20. The sewage and groundwater quality was tested and the result revealed that the groundwater is contaminated through leaching of sewage water irrigation. The soil samples were taken before and after sewage water irrigation from the cauliflower field. The impact of sewage water irrigation on physical, chemical and biological properties of soil was compared between the before and after sewage water irrigation. The pH, Electrical Conductivity (EC), Organic Carbon, available Nitrogen, Phosphorus, Potassium, Calcium, Magnesium, Zinc, Iron, Copper, Manganese and microbial activity *i.e.* soil microbial biomass carbon, basal soil respiration, total viable count of bacteria, *E. coli* population, *Pseudomonas species,* and *Azotobacter* is significantly increased after sewage water irrigation when compared with the before sewage water irrigation. However, the bulk density and *Rhizobium sp.* of the soil flooded with sewage water was decreased as compared to the bulk density and *Rhizobium sp.* of the soil before sewage water irrigation. The cauliflower yield was significantly increased when sewage water flooded field compared with the tube well water flooded field. The *E coli.* Contamination was higher in sewage water and groundwater that can cause health risk for the nearby communities, farmers and consumer of the farm products. Therefore, it may be concluded that in the scarcity of irrigation water, sewage and municipal wastewater can be used for irrigation after periodic monitoring of soil fertility and quality parameters of sewage and municipal wastewater or sewage water mixed with groundwater or alternate way of irrigation.

शोध सारांश

शोध शीर्षक	:	"राजनांदगांव जिले (C.G) में मृदा सूक्ष्मजीवों की विविधता और फूलगोभी उत्पादकता पर सीवेज जल सिंचाई के प्रभाव"
छात्र का नाम	:	नागेश्वर साहू
प्रदान की जाने वाली उपाधि	:	एम.एस.सी.कृषि सूक्ष्मजीवविज्ञान
मुख्य सलाहकार का नाम और पता	:	डॉ. रविंद्र सोनी, सहायक प्राध्यापक कृषि सूक्ष्मजीवविज्ञान विभाग, कृषि महाविद्यालय रायपुर
सह-सलाहकार का नाम	:	डॉ.ललित कुमार रामटेके , एसोसिएट प्रोफेसर मृदा विज्ञान एवं कृषि रसायन विभाग पं.शिव कुमार कृषि महाविद्यालय एवं अनुसन्धान केंद्र, राजनाँदगाँव
उपाधि प्रदान करने का वर्ष	:	2020

छात्र का हस्ताक्षर

प्रमुख सलाहकार के हस्ताक्षर

सह-सलाहकार के हस्ताक्षर

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शोध सारांश

शोध शीर्षक "राजनांदगांव जिले (C.G) में मृदा सूक्ष्मजीवों की विविधता और फूलगोभी उत्पादकता पर सीवेज जल सिंचाई के प्रभाव " को एक क्षेत्र पर प्रयोग-आधारित अध्ययन किया गया और सभी अवलोकन मृदा विज्ञान एवं कृषि रसायन विभाग,पं शिव कुमार शास्त्री कृषि महाविद्यालय एवं अनुसंधान केंद्र, राजनांदगांव और कृषि सूक्ष्म जीव विज्ञान विभाग,इंदिरा गांधी कृषि विश्व विद्यालय, वर्ष 2019-20 के दौरान रायपुर में विश्लेषण किया गया । सीवेज और भूजल की गुणवत्ता का परीक्षण किया गया और परिणाम से पता चला है कि सीवेज जल की सिंचाई से भूजल दूषित होता है। फूलगोभी प्रक्षेत्र में सीवेज जल सिंचाई से पहले और बाद में मिट्टी के नमूने लिए गए थे। मिट्टी के भौतिक, रासायनिक और जैविक गुणों पर सीवेज जल सिंचाई के प्रभाव की तुलना सीवेज जल सिंचाई के पहले और बाद में की गई थी। मृदा की पीएच, इलेक्ट्रिकल कंडक्टिविटी (ईसी), ऑर्गेनिक कार्बन, उपलब्ध नाइट्रोजन, फास्फोरस, पोटेशियम, कैल्शियम, मैग्नीशियम, जिंक, आयरन, कॉपर, मैंगनीज और माइक्रोबियल गतिविधि अर्थात मृदा माइक्रोबियल बायोमास कार्बन, बेसल मृदा श्वसन, बैक्टीरिया की कुल आबादी, ई कोली आबादी, स्यूडोमोनास प्रजातियां और एज़ोटोबैक्टर, सीवेज जल सिंचाई के पहले की तुलना में सीवेज जल सिंचाई के बाद काफी बढ़ जाती है। हालांकि, मृदा आभासी घनत्व और राइज़ोबियम की प्रजातियां सीवेज जल सिंचाई के बाद, सीवेज जल सिंचाई के पहले की तुलना में कम पाया गया । जब फूलगोभी प्रक्षेत्र को सीवेज जल तथा नलकूप जल से सिंचित किया गया और परिणाम से पता चला है कि सीवेज जल सिंचित क्षेत्र में फूलगोभी की उपज में काफी वृद्धि हुई । सीवेज जल और भूजल में ई.कोलाई प्रदूषण अधिक है, जो पास के समुदायों, किसानों और कृषि उत्पादों के उपभोक्ताओं के लिए स्वास्थ्य जोखिम पैदा कर सकता है। इसलिए यह निष्कर्ष निकाला जा सकता है कि सिंचाई के पानी की कमी में, सीवेज और नगरपालिका अपशिष्ट जल का उपयोग मिट्टी की उर्वरता की समय-समय पर निगरानी तथा सीवेज और नगरपालिका अपशिष्ट जल या सीवेज के साथ मिश्रित भूजल या सिंचाई के वैकल्पिक जल की गुणवत्ता मापदंडों की समय-समय पर निगरानी के बाद किया जा सकता है ।

CHAPTER - I

INTRODUCTION

Life is completely based on water and it is essential for plants and main components of protoplasm, containing up to ninety percent of their overall weight. Many Asian countries are expected to face severe water scarcity problems (Singh, 1999). In developing countries due to population growth causes scarcity of freshwater for irrigation in agriculture. Furthermore, the population of India is increasing at an alarming rate, the current population of India is 135.26 corers (Department of Economic and Social Affairs, Population Division) but water resource is limited and the requirement of water is increasing in more quantity for domestic, municipal and industrial purpose.

Water quality has a direct impact on soil properties, water management, and crop production for agricultural purposes (Shainberg and Oster, 1978).Therefore, sewage water or wastewater can be used in irrigation to fulfill the demand of agricultural sector. However, applying sewage to farm fields, and often industry effluent is a popular worldwide practice. At least 4–6 million hectares (ha.) area uses sewage water as a source of irrigation. (Jimenez and Asano, 2008; Keraita *et al.,* 2008, UNHSP, 2008).

Generally, sewage water used as an irrigation source for growing fodder crops and growing vegetables near the cities. It is one purpose because there is more market for vegetables in major cities such as radish, cauliflower, cabbage, brinjal, coriander, sweet corn, cowpea, and turnip etc. (Qadir& Gafoor,1997). Given the high prices in local commercial markets, vegetables are the most widely wastewater irrigated crops (Ensink *et al*., 2004).

Moreover, the application of domestic wastewater leads to increase in physicochemical soil attributes nutrient status and yield. Thus, it shows that improve in crop production with an increase in soil fertility status. The implementation of sewage water irrigation improves the yield of Rabi crops as oppose to well water irrigation; it also increases the overall soil content of N, P, K, organic carbon and microbial biomass carbon in agricultural soil. Increase in soil microbial biomass C and behaviors are likely to induce greater when wastewater is used for irrigation along with more rise in pollutant concentrations which assume to have adverse effects on soil microbial populations. The effects of long-term wastewater application on soil pH, Electrical Conductivity (EC), heavy metals are increased and changes the soil biological characteristics which is soil quality indicators, (Angin *et al.,* 2005).

Nonetheless, groundwater sources are free of pathogenic microbes because of the normal filtering capacity of the subsurface ecosystem and the extent to which a microbe needed to fly for entering the groundwater source. Contaminants which reach the groundwater might derive from lack of care, inadequate wastewater control, septic tank Contamination, subterranean holding tank or landfill leakage, food waste mismanagement disposal and because of which groundwater is polluted.

Furthermore, raw sewage water contains pathogenic bacteria, virus, protozoa, and certain parasitic helminths that can cause possible health risks to humans, animals, and plants. In wastewater-saturated soil, the fingers of the workers or children would be contaminated, so that some pathogens could be transmitted to their mouths and ingested. In this way, the volume of soil casually ingested was estimated as high as 100 mg per individual a day of exposure (*Haas et al.,* 1999, WHO 2001). Sewage or wastewater should contain 1 helminth egg per liter as environ. These helminthes are the human intestinal nematodes: *Ascaris lumbricoides* (human roundworm), *Trichuris trichiura* (human whipworm) and *Ancylostoma duodenale* and *Necator americanus* (human hookworm) (Feachem *et al.,* 1983). In addition, increased microbial activity may be attributed to the introduction of microbes by the drainage of wastewater carrying large microbial loads, including complete types of bacteria suggesting fecal pollution. The species of bacteria may have led to higher microbial respiration in soil, suggesting greater consistency of the soil (Sachidulal Raychaudhuri, Mausumi Raychaudhuri, Sachin Kanta Rautaray, Ashwani Kumar 2014).

Plant Growth-Promoting Rhizobacteria (PGPR) consist a large population of soil microorganisms, which, when develops into a host plant, brings about increment in the development of the host plant because of expanded up-take, mobility, and advancement of supplements or nutrient in the plant (Cakmakci *et al.,*2006). PGPR Bacteria colonize at plant roots that promote plant development and work as a biocontrol agent. Such microbes found in rhizoplane and the rhizosphere. Some PGPR affected when sewage water used as irrigation because sewage water contain high concentration of heavy metals.

Benefits of sewage water irrigation:-

- Prevention of surface water contamination that may develop by discharged of waste-water into rivers or lakes.
- Contribute to reasonable use of surface water, particularly in semi-arid and arid regions.

- Reduce the degree of groundwater pollution and the influx of seawater onto coastal areas.
- Sewage water is a good source of nutrient which ultimately reduces the cost of the chemical fertilizer requirement and also prevents the pollution of soil ecosystem caused by application of fertilizers.
- Prevention of soil erosion and soil conservation as the organic matter added by drainage runoff serves as a soil conditioner, growing its water-retaining ability.
- Desertification and reclaiming the desert, by irrigating and fertilizing the green belts.

It is scheduled to examine the "Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)" with the following objectives:-

1. To study the Physico-chemical and microbial properties of sewage water irrigated soil and sewage water.
2. Impact of sewage water irrigation on soil microbial population and their activity.

CHAPTER - II

REVIEW OF LITERATURE

The rapidly increasing population growth and the steady increase in water requirements for agricultural and industrial development have placed severe stress on the water resources available and the long term use of sewage water for irrigation highly affects soil properties. In this part, the work is undertaken about research in India and abroad has been summarized in the headings below:-

2.1 Impact of sewage water on soil

2.2 The properties of sewage water.

2.3 Impact of sewage water on soil microbial population

2.1: Impact of sewage water on soil

When soil was irrigated with sewage water as such or their dilution, soil hydraulic conductivity decreased significantly. The soil where Indian spinach was grown has significantly higher hydraulic conductivity values than those where carrots and cauliflower were grown (Kolakar *et al.*,1947).Sewage water helps in increasing N, P, and K contents in irrigated soil, (Lisoval *et al.* 1981). Further, it also influenced the biological characteristics of soil, especially including zinc and copper (Hundal and Sandhu, 1990).Further, waste effluent irrigation plus tubewell water substantially decreased soil pH and improved soil EC and organic matter (Hundal and Sandhu, 1990). Sen *et al., (*1997) reported that Nagpur's sewage effluent pH was alkaline and the effluent contained substantial quantities of basic cations. However, Ca was the dominant base followed by Na and Mg. However, the effect of irrigation by the paper industry effluent on soil properties was investigated. In an another report, it was observed that irrigation with industrial effluent, the pH, EC, and availability of N, P, K increased as compared to well water. The effluent had high total dissolved salts than well water. In specific, the abundance of Zn, Mn, and Cu in the surface layer (Roy *et al*., 2008).

Furthermore, the usage of an industrial solid waste comprising MnO_2 and neutralized with slaked lime, integrated elevated amounts of Al, K, Ca, Fe with lower rates of heavy metals (such as Cu, Zn, Co, and Ni) in two-year field trials in Greece. The pH was 7.9 and produced 14.12 percent total Mn (Karagi annidis, 2000).

Moreover, changes in plant nutrients after sewage water irrigation were observed by (Rattan *et al.,* 2001). They find that increase in the response of sewage water to crop yield, were in line with standard tube well water irrigation. At Sunhemp research station, Fresh domestic wastewater from AvasVikas colony, Pratapgarh with a high degree of contaminated water used for irrigation of crops such as wheat, black gram, mustard, and paddy (Pradhan *et al.,(* 2001).

In the case of agricultural effluents, Zn and Cr in the fertilizer industry effluent from Bharuch, the overall amount of different trace elements and heavy metals except Zn and Pb was beyond the irrigation quality limits (Patel *et al.,* 2004).The treated wastewater irrigated soils produce a marginally higher quantity of micronutrients (Fe, Mn, Zn, and Cu) relative to neighboring tube-fed irrigated soils. Further, the wastewater treated irrigated produced vegetables comparatively higher levels of micronutrients than the well-irrigated vegetables. Where the quantity of Zn, Fe, Cu, and Mn was higher in Okra, Cauliflower, Carrot, Radish, and large Beans, respectively, in their edible parts. Only treated wastewater irrigated and well-irrigated tube well, potato showed lower amounts of manganese than the deficiency mark (Saraswat *et al.,* 2005).

The impact on the surface water of industrial effluents attributed to the steel plant and other big factories at Rourkela in Orissa. Their scientific results on physico-chemical parameters showed that owing to urban and industrial toxic waste disposal the soil of some of the areas is polluted (Mishra *et al.,* 2005).The long-term use of wastewater irrigation shows the improvement in plant nutrient composition and improved soil salinity and reduced pH. Other positive improvements were also found including a rise in organic matter, the concentration of nitrogen and soil heavy metal. Regular use of wastewater increase N, P, K, Fe, Mn, Cu, Zn, B, Mo, and Cd content in cabbage and tomato (Angin *et al.,* 2005).

Sewage effluents usually contain 5.5, 3.6, 6.4, and 1.3 times more micronutrients such as Zn, Cu, Fe, Mn, Mo, and Ni than freshwater, respectively, were 1.42, 30, 74, 127, 159, and 85 μgL^{-1} (Ratan *et al.,* 2006).Further, the amount of DTPA- extractable Cu, Zn, Fe, and Mn in Jodhpur district, soil, getting tube well water was 0.25, 0.51, 2.30, and 3.30 mg/kg, respectively, and 1.36, 2.89, 12.14, and 12.40 mg/kg in urban effluent soil received (Singh & Bhati, 2005).When irrigated with or mixed with wastewater, the soil resulted in a rise in EC, usable N and P status and cation content of soil (Malla and Totawat, 2006).Continuous sewage water irrigation has increased the supply of Ca, Mg, Zn, Mn, and Fe in the land-use system. Although the detrimental effects of EC and ESP have been reported (Bhise *et al*., 2007).

Further, a research evaluates the impact of long-term sewage effluent treatment culminated in a decline in soil pH and an improvement in soil electrical conductivity in all three soils. Gross carbon from the surface and microbial biomass production rise across all soils in irrigated areas (Adeli *et al.,* 2008). In another long term experiment irrigation of waste and sludge water has raised the concentration of metals substantially in the soil and plants compared with the control map. Long term irrigation of effluent and sludge raised heavy metal contents in soil and plants and rendered this activity dangerous to be introduced by man, (Behbahaninia *et al.,* 2009).

One of the main sources of groundwater contamination is the surface impoundments used by municipalities and industries that dispose of wastewater to receiving bodies without treatment or used by farmers for agricultural purposes. The quality of groundwater is deteriorated due to higher electrical conductivity concentrations, total dissolved solids, and hardness as compared to WHO (Jaghrani, 2009).The sewage sludge modification also changed the soil chemical properties by minimizing pH and EC, organic carbon, gross N, and usable P in soil (Singh and Agrawal, 2010).

Furthermore, the groundwater quality in the Cheyyar basin, Tamilnadu found to be highly contaminated, mostly due to industrial effluents (Sathish Kumar and Ravichandran, 2011).The metals in landfill leachate (lead, cadmium, chromium, copper, and nickel) and heavy metals in wells downstream of a municipal solid waste landfill site in Mashhad City. The result shows the highest rates of heavy metals, the concentration of nickel in summer, and the concentration of lead in winter (Mansouri *et al.,* 2015).

However, pollutants are depending on the source from which the wastewaters originated, their chemical composition differed considerably (Becerra-Castro *et al.,* 2015). Ina nutshell the sewage water supplies essential nutrients and minerals like nitrogen (N), phosphorus (P), and potassium (K), it was considered a low price fertilizer due to the easy availability of sewage or wastewater nearby city residential farmers, it improves soil fertility and soil microbial activity. Wastewater irrigation may have effects at two different levels: change the soil physical-chemical properties and microbiological content, or introduce and lead to the accumulation of chemical and biological pollutants in soil.

2.2 The properties of sewage water

Drainage water quality often relies on locality eating patterns, population density, and manufacturing unit substance impurities that are applied to the sewage network. Sewage water in several Indian towns, including Cu, Zn , Mn, Fe, Cd, Pb, Ni and Cr ranged from

0.18- 6.30, 0.14-11.00, 4.10- 205.00, 0.10-37.80, 0.01-5.80, 0.03-40.00, 0.35-6.40 and 0.18-29.30 mgL^{-1} (Narwal and Kuhad, 2005).In Haryana state, wastewaters' composition was observed that pH, EC, TDS, and the BOD ranged from 7.1-8.30, 9-3.2 dSm^{-1}, 60-330 mgL^{-1}, and 176-345 mgL^{-1}, respectively. The soluble concentration of Cu, Zn, Fe, and Mn in these wastewaters ranged from 10-415,10-330,130-1887 and Tr.- 420 mgL^{-1} (Minhas *et al.,* 2005). In addition to this sewage waters from various places in Haryana find pH, EC, Organic carbon, TSS and BOD are varying from 1.1-1.8, 0.64-0.44 dSm^{-1}, 15-105 mgL^{-1},700-300 mgL^{-1} and 36-127 mgL^{-1}, respectively. In waste water, soluble amounts of N, P, K, Cu, Zn , Fe and Mn varied from 0.13-2.70, 0.88-17.25, 16.0-500, Tr-1.0, 1-6.8, Tr-56.5 and Tr-8.0 mgL^{-1}, respectively (Antil and Narwal, 2005).

Similarly a state-level study found that the majority of sewage waters were classified ideal for irrigation in Haryana state as it had EC 0.9-3.2 dSm^{-1}, SAR ratio 1.4-6.2 m $molL^{-1}$, and residual sodium carbonate 0-8.6 $meqL^{-1}$ (Dubey *et al.,* 2007).The treated wastewater handled includes 952 mgL^{-1} TDS, 29.4 mgL^{-1} NO_3-N ,15.5 mgL^{-1} PO_4, 33.3 mgL^{-1} K in addition to Cu 0.01 mgL^{-1},Zn 0.19 mgL^{-1}, Fe 0.87 mgL^{-1}, Mn 0.07 mgL^{-1}, Cd 0.02 mgL^{-1} and Pb 0.77 mgL^{-1} (Rusan *et al.,* 2007).

The groundwater quality in the Tamilnadu, Cheyyar basin is found to be heavily polluted, mainly due to industrial effluents (Sathish Kumar and Ravichandran, 2011). The pH of wastewater ranges from 7.3 to 7.8, the sewer water colour is yellowish-green or light brown or black attributable to organic matter. Septic drainage produces unacceptable odour, H_2S is the primary cause of contamination (Haseeb Jamal, 2017).

2.3 Impact of sewage water on soil microbial population

Residential wastewater is believed to be a mixture of any of all household wastewater, composed of black water, excreta, feces, associated sludge (kitchen and bathroom wastewater) from industrial enterprises. Organizations like hospital waste, sewage-polluted water contains various sewage micro-flora, several of which present a public health risk (Stuart and Gordon, Baross *et al.,* 1975). Some wastewater-borne bacteria and viruses *(enteroviruses, Norwalk-like viruses, coliphage)* were routinely found in (vertically) unconfined sandstone at depths of 60 meters and in enclosed sandstone at depths of 91 meters (Berge, 1978). It was reported that the *Escherichia coli* bacteria survive in organic soil was after manure over an 8-day duration (Tate, 1978).

The association between the amounts of bacteria used for spray irrigation in wastewater and the air was studied. Results revealed that the relative humidity and solar irradiation appeared to influence viable bacteria in the air; there was a strong association

between relative humidity and the number of aerosolized bacteria. On the other side, the association between solar irradiation and the degree of the bacteria was negative. Up to 10-fold, more aerosolized bacteria were detected during night irrigation than with day irrigation. Wind intensity did not play a major part in the aerosolized bacteria's survival (Teltsch and Katzenelson, 1978).

Furthermore, the main forms of transportation of pathogens and indicator species in manure receiving soil are by moving with the absorption of water and surface runoff and sediment and movement of the waste particles (Reddy *et al.,* 1981).However, smaller levels of microbial biomass and decreased microbial activity in soils from 1942 to 1961 when metal-contaminated wastewater sludge was received (Brookes *et al*. 1984).

Business solid waste may add pathogenic species to urban landfills, such as agricultural waste and chemical manufacturing waste. The proportion of fecal coliform to fecal *streptococci* contained in landfill waste, based on one study, indicates that pathogens in urban waste are primarily of non-human, warm-blooded animal origin (human feces have a far higher fecal coliforms density than other animals) (Pahren, 1987).The survival of many species under artificial saturated soil conditions and finding that the majority of the species studied, including *Escherichia coli*, lived at 10 °C for more than 100 days (Philip *et al.,* 1988).

The application of large quantities of sewage sludge over a long time may increase the concentration of pollutants with a negative impact on soil microflora (Balzer and Ahrens, 1990). The population of different microorganisms, dehydrogenase, and phosphate processes are greatly decreased by the usage of polluted irrigation water (Rao *et al.,* 1993). More metal-resistant microbes were contained in the polluted soil, but these microbes have little capacity to kill organic contaminants (Doelman *et al.,* 1994).

Moreover, the microbial population and respiration in soil receiving sewage sludge were stated that the higher rates of metal pollution resulted in a sustainable decrease in soil microbial biomass amounts (Flie Bach *et al.,* 1994). In addition, different management practices, like crop rotation, cultivation, organic modifications, fertilization, and plant residue management, greatly affect the level of microbial biomass (McGill *et al*., 1986; Van Lutzow and Ottow, 1994).

On the soil surface, the fecal bacteria have higher survival rates than the evaluation of the subsoil was threefold larger than the sandy soil (Zhai *et al.,* 1995).The pollution of solid waste with an animal and liquid excreta can involve numerous harmful microorganisms such as bacteria and viruses that are normally deposited off landfills and open garbage dumps

and nearby unlined drain sites (Fapetu *et al.*, 2000). The application rate of sewage sludge and metal alteration affected the density and action of microbial biomass and led to changes in the microbial population (Witter *et al*., 2002).

Wastewater affects the microbial population of the soil by increasing the levels of organic matter and micronutrients, thus providing substrates for microbial growth. Positive effects on soil microbial productivity and behavior were detected through uncontaminated treatment when contrasted with metal-polluted waste sludge (Brookes and McGarth, Perrucci, Selivanovskaya *et al*., 2002, Kao *et al*.). It was observed the sources of heavy bacterial levels in Jorhat city water are due to wastewater and groundwater runoff. Surface bodies of soil were found to be more contaminated with waste supplies (Thomas *et al*., 2003).

In Sivakasi, India, the physico-chemical and bacteriological conditions for drinking, borewells, and wastewater applications. Much of drinking and bore wells water's physico-chemical characteristics were below the allowable ranges. However, bacterial concentrations in water samples from both locations surpassed the WHO's suggested acceptable amount. At the downstream side of the river Thamirabarani at Ambassamuudram, the largest load of bacteria from fecal sources was found, which may be attributed to regular pollution by human, animal, and bird excreta. The occurrence of low coliform and streptococci of fecal origin on the upstream side may be attributed to persistent river flood, loss of organic nutrients and persistent water drainage, degradation of organic nutrients, and continuous usage of detergents, antiseptics, soap etc. (Umamaheshwari, 2004).

In past also, bacterial populations responded differently and were most sensitive to higher levels of Cu. The numbers of protista in the ground from uncultivated land were higher and appeared to be more susceptible to additional Cu than the number of such organisms in soil from cultivated soil (Du Plessis *et al*., 2005). The quantity of microbial biomass in metal-polluted soils were approximately half that observed in soils receiving uncontaminated organic manure or inorganic fertilizers from the experiment (Barajas, 2005).

In the metal-contaminated soils, *Pseudomonas fluorescence* concentrations were at least 1.5 times higher than in unpolluted soils as a proportion of total plate counts. Gram-negative counts in metal-contaminated soils were 62-68 percent greater than Gram-positive counts. (Abaye *et al.*, 2005).

The long-term impact of heavy metal exposition on the amount of aerobic culturable microbes, actinomycetes, fungi, and symbiotic nitrogen fixers. Contaminated soil samples displayed a pronounced reduction in overall cultivable numbers of the various microbial

species. Asymbiotic nitrogen fixation systems and heterotrophic bacteria were especially susceptible to long-term heavy metal pollution (Pampulha and Oliveira, 2006).

It has been shown that high concentrations of heavy metals have a detrimental impact on the composition, abundance, and behavior of microbial species in soil (Banerjee *et al*., Del Val *et al*., Liao and Xic, Wang *et al*., 2006). Further, raw sewage irrigated soil across Nagpur city of Nag river had lower rates of bacteria, *Azotobacter*, and fungi comparison to fresh well water irrigated natural soils, whereas, *Actinomycetes* were not affected suggesting their tolerance to harmful chemicals (Taywade and Prashad, 2008).

CHAPTER - III

MATERIAL AND METHOD

The present study entitles “Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G)” was carried out during Winter Season, 2019-20. A detailed description of the materials and the techniques used during the study is presented in this chapter:-

3.1: Location and Climate

3.1.1: Location of experimental site

A field experiment on Cauliflower was conducted in Paneka village near the Rajnandgaon city. Industrial effluents drain from the industrial area of Rajnandgaon whereas domestic wastewater comes from residential areas of Rajnandgaon. The field is located on the western side of the Pt. Shiv Kumar Shastri College of agriculture and Research station, Rajnandgaon. In the winter season, farmers have grown seasonal vegetables and irrigated with the wastewater flowing through the wastewater drain.

3.1.2: Geographical status of Rajnandgaon

Rajnandgaon is located at the Central Plane of India. The ground is bordered by Chotanagpur Plateau. Rajnandgaon has a highest farming area for agriculture. Also, the mineral is in abundance here due to the presence of mountains and rivers and thus ideal for the industry as well. The town of Rajnandgaon is located at 21.10 N 81.03° E. It has an average elevation of 307 meters above the mean sea level at the western part of Chhattisgarh plain. The town established on the banks of the Sheonath River.

3.1.3: The climate of Rajnandgaon

The town gets a good amount of rain. According to available data, Rajnandgaon gets approximately 1200 mm per annum rain during the rainy or monsoon season. Throughout the Monsoon the temperature drops to about 30 degrees. The Monsoon Season extends from the late June in the year until early October. The winter season starts from October. Throughout the winter season, the maximum temperature stays around 14ºCand the lowest may be as low as 5ºC. However, when the region experiences some rain during the winter, the temperature will fall much lower.

3.2: Experimental details

Experiment place - Paneka, Rajnandgaon

Crop - Cauliflower

Variety - Pusa Deepali

Planting method - Ridge and Furrow method

3.2.1: Field preparation

The field was prepared by two to three time ploughing by cultivators and manipulation of soil through rotavator. Levelling was done soon after ploughing.

3.2.2: Application of Fertilizer

The NPK is 120:60:60 kg/ha added. Phosphorus and potassium were added as basal, respectively, through di-ammonium phosphate and muriate of potash. When some portion of nitrogen transferred by DAP to the soil, the remainder of the nitrogen was added when basal by urea to the crop in two split dose.

3.2.3: Seed sowing in Nursery

Seed sowing was done on 02/09/2019 on the ridge nursery bed.

3.2.4: Transplanting

Transplanting was done on 29/09/2019 in ridge and furrow method.

3.2.5: Irrigation schedule

- First irrigation was done after transplanting.
- Second irrigation was done five days after transplanting.
- Third irrigation was done fifteen days after Second irrigation.
- Fourth irrigation was done one week after third irrigation.

3.3: Collection of samples

3.3.1: Soil sampling for Physico-chemical analysis

A composite soil sample from a depth of surface soil (0-15cm) and subsurface soil depth (15-30 cm) was collected from the site before sowing the crop and samples were collected from the sewage water irrigated fields from peri-urban areas of Rajnandgaon districts of Chhattisgarh State.

The collected samples are air dried under shade and ground with a wooden mortar and pestle to avoid contamination in the soil sample. After grinding, sieve the soil samples with a 2 mm stainless mesh sieve and then completely combine all the soil samples by spreading over sheet or paper. The need to extract one representative soil sample from this bulk soil sample using the partitioning process of quartering. About 250 g to 500 g of the soil sample is sufficient for analysis and stored the sample in dry and clean poly bags or cardboard boxes with proper labeling at 4ºC, for analysis of physicochemical properties and nutrients

3.3.1.1: Analysis of Physico-chemical parameter of soil

a) Soil pH:-Soil pH was determined by 1:2 soil-water suspension in glass electrode pH meter (Piper, 1967).

b) Soil EC (Electrical Conductivity):-Soil EC was determined by 1:2 soil-water extract or supernatant filtered liquid in EC meter (Jackson, 1973).

c) Organic Carbon:- Walkley and Black method (1934)used to determine the organic carbon content of soils.

d) Nitrogen:- The alkaline permanganate method (Subbiah and Asija, 1956) was used to estimate the nitrogen availability in soil samples.

e) Phosphorus:-The soil phosphorus content was obtained by using the Olsen method or Sodium Bicarbonate Extractable P method, given by (Olsen *et al*,1954).

f) Potassium:- Potassium was estimated by flame photometer with the help of a standard curve developed by Jackson(1967).

g) Calcium and Magnesium:- Exchangeable calcium and magnesium were estimated by using the Versanate titration method in a neutral normal ammonium acetate extract (Cheng and Bray, 1951).

h) Micronutrients (Available Metalic Ions (Fe, Mn, Cu. Zn):- The micronutrients were determined with the help of DTPA (Diethylenetriaminepenta acetic acid) extractant and Atomic Absorption Spectrophotometer. This method was given by Lindsay and Norvell, (1978).

Plate 1: soil sampling before sewage water irrigation

Plate 2: After sewage water irrigation

Plate 3: Determination of pH and EC of soil

Plate 4: Determination of available K, Ca and Mg of soil

3.3.2: Soil microbial analysis

The soil sample is taken from a depth of 0 to15 cm for determination of microbial biomass carbon and microbial activity, the soil was moistened to 60% water retention capacity and incubated at 30°C.

3.3.2.1: Microbiological studies

Microbiological studies were conducted using the methodology of spread plate technique, utilizing respective media for different groups of organisms. Following media used for growing different groups of microorganisms.

a). Total Bacterial population

The bacterial population was estimated by spread plate count technique using Nutrient Agar Media.

b). *E. coli* population

E. coli population was estimated by spread plate count technique using MacConkey Agar Media.

c). *Rhizobium sp.* population

Rhizobium sp. population was estimated by spread plate count technique using YEMA Media.

d). *Pseudomonas species* population

Pseudomonas species population was estimated by spread plate count technique using King's Medium B Base.

e). *Azotobacter species* population

The *Azotobacter* species population was estimated by spread plate count technique using Jensen's Medium. This medium is recommended for the detection and cultivation of nitrogen-fixing bacteria.

3.3.2.2: Estimation of Colony Forming Units

One gram of soil sample taken and blended it with 9 mL of sterilized water in a clean and sterilized test tube to dilute 10^{-1} (1:10).After shaking 1-millilitre suspension taken and diluted up to 9 millilitres of distilled water in a test tube and make 10^{-2} dilution (1:100). Likewise, make eight more serial dilutions to get 10^{-8} dilution.

Calculate the number of colony unit per gram of the sample as the following formula:-

$$\textbf{CFU/g.} = \frac{\textbf{Average no.of colonies}}{\textbf{Dryweightofsoilsample}} \times \textbf{dilution factor}$$

Where, Dilution factor = reciprocal of dilution.

3.3.2.3: Basal soil respiration

This research was carried out to understand the rate of respiration of microflora present in the soil rhizosphere. Basal respiration of soil was determined by calculating levels of CO_2 evolution (Anderson, 1982).

Calculation:-

$$\textbf{Soil respiration (mg of } CO_2\textbf{/h/100g soil)} = (\mathbf{B - V})\ \mathbf{NE/hours\ of\ incubation}$$

Where, B-Volume of acid (ml) needed for the blank, V-Volume of acid (ml) needed for the NaOH exposed to soil, N-Normality of acid, E- Equivalent weight, i.e. 22.

3.3.2.4: Microbial biomass carbon (MBC)

Chloroform fumigation approach suggested by Jenkinson and Powlson (1976) determination for microbial biomass carbon.

Calculation:-

$$\textbf{Microbial biomass carbon (µg/g soil)} = (\mathbf{ECF - ECNF})/\mathbf{0.25}$$

Where, ECF = Extractable C in the fumigated soil, ECNF = Extractable C in the non-fumigated soil, 0.25 = K_{EC} represents the efficiency of extraction of microbial bio mass carbon.

3.3.3: Sewage and ground water sampling

During the sewage water sample extraction, sampling bottles are rinsed two times, but the bottles carry a dechlorinating agent. Samples from three separate sampling sites were obtained with a gap of two to three meters. Containers were loaded with the desired container volume of about 1 per cent to enable thermal expansion.

3.3.3.1: Analysis of sewage and ground water

a) pH:- pH was determined by pH meter.

b) EC:- Electrical Conductivity was measured by the EC meter.

c) Carbonate and bicarbonate:- Measured by titrating a specific amount of H_2SO_4 with normal wastewater, first in the presence of phenolphthalein and then with methyl red indicator (Richards, 1954).

d) Calcium:- Calcium was determined by titration with standard 0.01 N EDTA and pH-12 obtained with the support of NaOH using ammonium parpurate as an indicator when the solution's pink colour changed to purple (Jackson, 1973).

e) Magnesium:- Magnesium was determined by titration with 0.01N EDTA by buffering the pH 10 solution with NH_4, C-NH_4OH buffer using Erichrome black T indicator. The result was noted by changing the colour of an initial red wine to the final blue sky (Jackson, 1973).

f) Potassium:- Potassium was estimated by flame photometer with the help of a standard curve outlined by (Jackson, 1967).

g) Micronutrients (Available Metallic Ions (Fe, Mn, Cu. Zn):- The micronutrients were determined with the help of Atomic Absorption Spectrophotometer.

3.3.3.2: Biological properties of sewage water

a). Total Bacterial population

The bacterial population was estimated by spread plate count technique using Nutrient Agar Media.

b). *E. coli* population

E. coli population was estimated by spread plate count technique using MacConkey Agar Media.

Plate 5: Different media used for growing different groups of microorganisms.

Plate 6: Determination of basal soil respiration

Plate 7: Determination of microbial biomass carbon.

Plate 8: Pouring of soil and water samples on different media

3.4: Statistical analysis

The "t" test for the paired sample was used to determine the impact of sewage water irrigation on microbial diversity and cauliflower productivity. For the statistical analysis of data Microsoft Excel window version, 7.0 were used.

3.4.1: "t" test for paired sample

The Paired T-Test used to compare the means of two groups of observations. This test can be used in making observations on the same sample before and after an event.

Formula:

The paired t-test statistics value can be calculated by using the following formula:

$$t = \frac{m}{s/\sqrt{n}}$$

Where, m is the mean differences, n is the sample size (i.e., size of d), s is the standard deviation of d

We can compute the p-value corresponding to the absolute value of the t-test statistics (|t|) for the degrees of freedom (df): df = n-1.

3.4.2: Mean

The mean is found by adding the numbers in a data set and dividing by how many numbers there are.

$$\bar{X} = \sum x / N$$

Here, ∑ represents the summation, X represents scores; N represents a number of scores.

3.4.3: Standard Deviation

The standard deviation is an estimation of the amount of variation or dispersion of a data set. The formula for the sample standard deviation is:-

$$S = \sqrt{\frac{1}{N-1}} \sum_{I=1}^{N} (xi - \bar{x})$$

Where, ($x_1, x_2, \ldots x^n$) are the observed values of the sample items, $\bar{x}$ is the mean value of these observations and *N* is the number of observations in the sample.

3.4.4: Coefficient of variation:

The coefficient of variation (CV), also called a relative standard deviation (RSD). The coefficient of variation (CV) is a statistical analysis of the dispersion of data points in a data series around the mean.

Formula:

$$CV = \frac{\sigma}{\mu} * 100$$

Where, σ = Standard Deviation, μ = Mean.

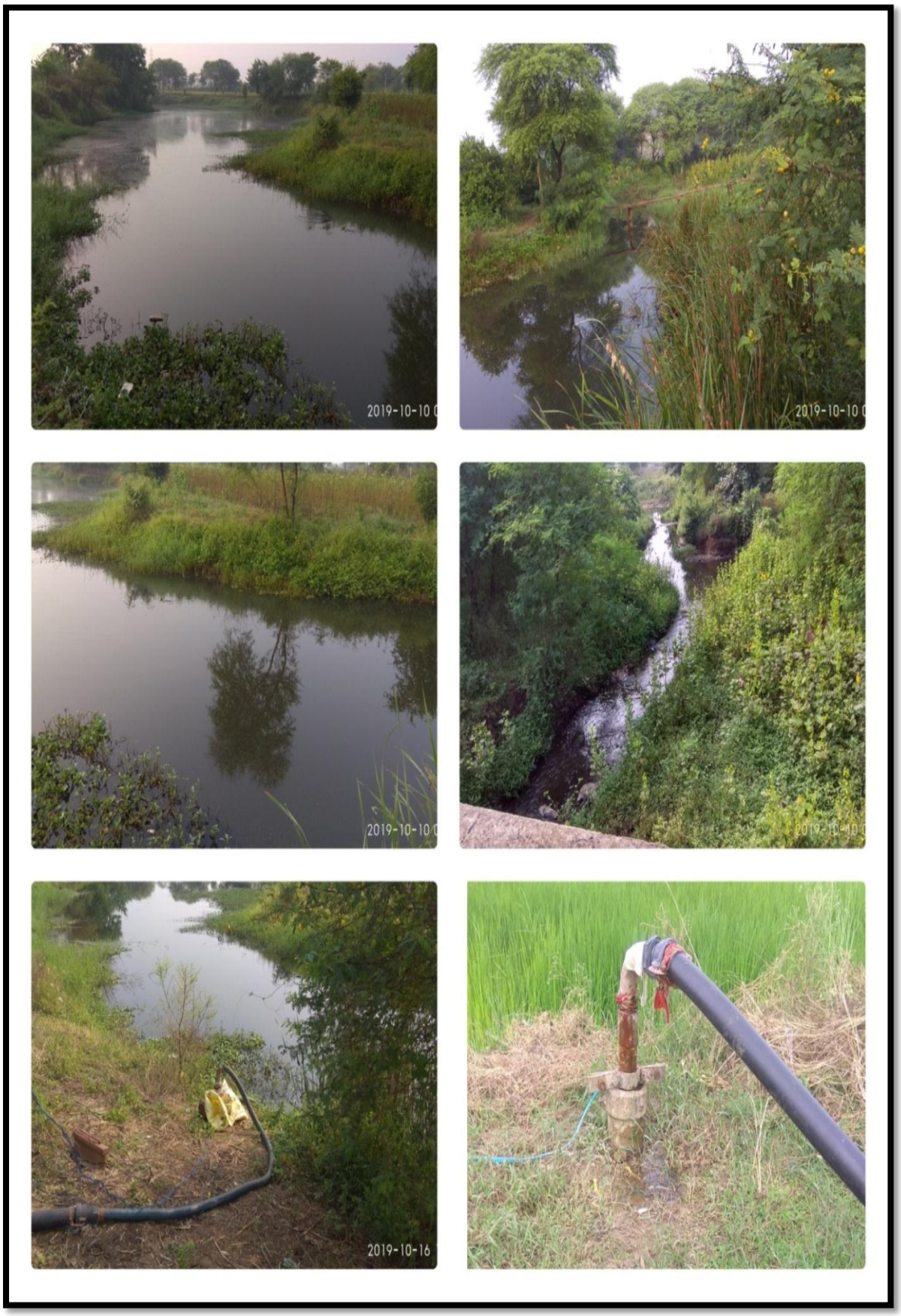

Plate 9: Source of irrigation *e.g.*- sewage and Tube-well water

CHAPTER – IV
RESULTS AND DISCUSSION

The results obtained during the investigation have been described in this chapter. The data obtained during the observation has been summarized with table and illustrated by figures with statistical interpretation. The chapter has been described with the following headings:

4.1 Microbial properties of sewage water and Tube-well water.
4.2 To study the Physico-chemical properties of sewage water and Tube-well water.
4.3 To study the Physico-chemical properties of sewage water irrigated soil.
4.4 Impact of sewage water irrigation on soil microbial population and their activity.
4.5 Dynamics of microbial population pre and post-exposure to sewage water irrigation.
4.6 Effect on PGPR population pre and post-exposure to sewage water irrigation.
4.7 The effect of sewage water irrigation on cauliflower yield

4.1: Microbial properties of sewage water and Tube-well water

The presence of *Escherichia coli* in water gives proof of fecal contamination of human or animal. In the examination zone, it is found that the non-sterilized groundwater has the high bacterial burden and no connection is found between total viable count and *E. coli*. Thus, *E. coli* is not generally present as a segment of bacterial burden and bacteria may have originated from environmental sources, for example - soil ecosystem, (Pronk *et al.*, 2007)

The population of *E. coli* is higher compared to the standard limit given by BIS (1995) and WHO (2004). Clark *et al.* (2003) likewise propose that the high natural substance and the clayey ideal for growth and bacterial development. The continuous use of *E. coli* contaminated water for drinking or irrigation purpose may have dangers for health risks, for example, urinary tract infections, respiratory diseases, abscesses and skin-twisted diseases, to neighborhood residents of this location, particularly youngsters and Children's.

Table 4.1 Microbial activity in sewage and Tube-well water

Microorganism	Sewage water	Tube-well water
Total viable Count	$2.92*10^8$	$3.5*10^5$
E. coli	$2.83*10^8$	$2.9*10^5$

Table 4.2 Contamination status of water-based on *E. coli*

E. coli CFU/ml	Water pollution status-WHO 2004	Water pollution status-BIS 1995
10,000	Heavily polluted	Polluted
1000	Polluted	Polluted
100	Slightly polluted	Polluted
10	Satisfactory	Acceptable
3 or less	Potable	Excellent

Bureau of Indian Standards (BIS 1995) and world Health Organization (WHO 2004).

4.2: To study the Physico-chemical properties of sewage water and Tube-well water

The qualities of municipal sewage effluent and tube-well water were assessed for irrigation concerning their pH, EC and concentrations of nutrients. Results indicated that all water samples were alkaline. The pH of the municipal sewage effluent (pH 7.7) was higher compared to the tube-well water (pH 7.5), whereas the effluent's salt content (EC 1.2 dSm^{-1}) was considerably higher compared to the tube-well waters (0.98 dSm^{-1}).

Table 4.3 Physico-chemical properties of sewage water and tube- well water:-

Serial no.	Parameter	Unit	Sewage water	Tube-well water	Standard Limit for irrigation
1	Temperature	ºC	27.4	27.4	------
2	pH	-------	7.7	7.5	6.5-8.4
3	EC	dSm^{-1}	1.2	0.98	<3
4	NH_4-N	mgL^{-1}	6.6	1.5	30
5	PO_4-p	mgL^{-1}	Not Detection	Not Detection	2
6	K	mgL^{-1}	20	25	0.2
7	Ca	mgL^{-1}	245	86	400
8	Mg	mgL^{-1}	149	57	60
9	CO_3	mgL^{-1}	Nil	Nil	6
10	HCO_3	mgL^{-1}	285	532	600
11	Zn	mgL^{-1}	0.58	0.34	2
12	Cu	mgL^{-1}	0.3	0.2	0.1
13	Fe	mgL^{-1}	2.16	0.92	5
14	Mn	mgL^{-1}	0.6	0.42	0.2

This indicated that the sewage effluent was saline in nature. The EC of sewage water is 1.2 dSm^{-1} which may cause saltiness of the soil and restricted to the plant development (Antil, 2012). Comparable results were supported by Yadav *et al.,* (2003), Abril (2005), Hussain (2005) and Sial *et al.,* (2006). The consequence of the chemical properties examination of the sewage water that was utilized for the irrigation system. The sewage water was compared with FAO norms for irrigation, there were clear contrasts in many estimated parameters. The estimations of K, Mg, Zn and Cu in sewage water were high; compared with the FAO esteems (1992). These results were in concurrence with Bharose *et al.,* (2013).

In the current investigation, a portion of the substantial or heavy metals which likewise come under micronutrients like Zn, Cu, Fe, Mn is fundamental for plant development (Wintz *et al.,* 2002). The concentration of Fe, Mn, Zn, and Cu in the present examination was higher in sewage water when compared with tube-well water and the sewage water contain a good amount of micronutrient and help in overcoming the lack of expensive synthetic fertilizer (Kharache *et al.,* 2011). The sewage has frequently high estimate of temperature, pH, hardness, alkalinity, NH_4-N and cations like potassium, calcium and magnesium, Ghafoor *et al.,* (1995). Same outcomes were accounted for by Singh *et al.* (2010).

4.3: To study the Physico-chemical properties of sewage water irrigated soil

The physico-chemical and Microbiological properties of soil are significantly differing as compared to before sewage water irrigation and after sewage water irrigated soil. The static tools “t” test for paired sample used for comparison. The following results below in the table 4.4.

4.3.1: Effect of sewage water irrigation on soil bulk density

The data in the given Table 4.5, it was confirmed that the bulk density of the soil irrigated with sewage water was significantly decreased at ($P<0.01$) when compared with a bulk density of the before sewage water irrigation. Subsequently, this was the explanation that the bulk density affected by organic carbon of the, soil which builds the porosity and at least decreased the bulk density of the soil. Comparable outcomes have been reported by Mathan (1994), Kharche *et al.* (2011) and Subramani *et al.* (2014).

Table 4.4 The effect of sewage water irrigation on different Physico-chemical and biological properties of soil

Soil properties	Unit	P(T<=t) two-tail value	Statical P level
Bulk Density	g/cm^3	0.0032**	<0.01
pH	-----	0.0423*	<0.05
EC	dSm^{-1}	0.011*	<0.05
OC	%	0.0023**	<0.01
N	Kg/ha.	0.0036**	<0.01
P	Kg/ha.	0.0448*	<0.05
K	Kg/ha.	0.0379*	<0.05
Ca	Kg/ha.	0.0238*	<0.05
Mg	Kg/ha.	0.0332*	<0.05
Fe	ppm	0.0045**	<0.01
Mn	ppm	9.9E-06**	<0.01
Cu	ppm	0.0202*	<0.05
Zn	ppm	5.03E-05**	<0.01
Soil basal respiration	-----	0.0036**	<0.01
Microbial biomass carbon	-----	0.0027**	<0.01
Total bacterial population	-----	0.0002**	<0.01
E. coli population	-----	0.0018**	<0.01
Rhizobium population	-----	0.0006**	<0.01
Pseudomonas population	-----	0.01196**	<0.01
Azotobacter	-----	0.0308*	<0.05
Cauliflower Yield	In gram	3.212E-07**	<0.01

* Significant at P = 0.05 level; ** Significant at P = 0.01 level

4.3.2: Effect of sewage water irrigation on soil pH

The pH results are in table 4.6. The irrigation with sewage water is significant at ($P<0.05$) expanded soil pH compared to before sewage water irrigation. Comparative outcomes were seen by (Rusan 2007, Rattan *et al.* 2005), who studied that the long term effect of sewage and wastewater effluents on soil properties.

Table 4.5 Effect of sewage water irrigation on soil bulk density

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	1.31	1.24
2	1.39	1.21
3	1.42	1.29
4	1.35	1.26
5	1.29	1.22
6	1.27	1.21
Mean	1.33	1.23
S.D	0.05	0.03
CV%	3.75	2.43

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.6 Effect of sewage water irrigation on soil pH

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	6.9	7.4
2	7.2	7.5
3	7.3	7.6
4	7.4	7.2
5	7.1	7.6
6	7.2	7.5
Mean	7.17	7.46
S.D	0.17	0.15
CV%	2.36	2.01

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

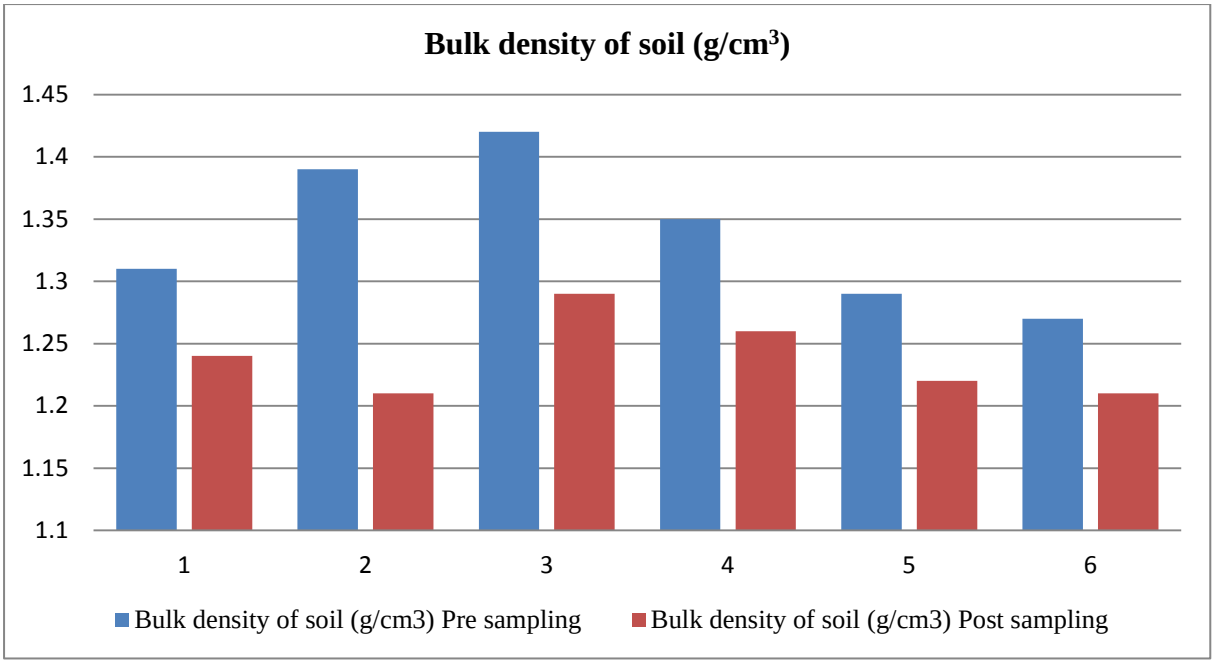

Fig.1: Effect of sewage water irrigation on soil bulk density

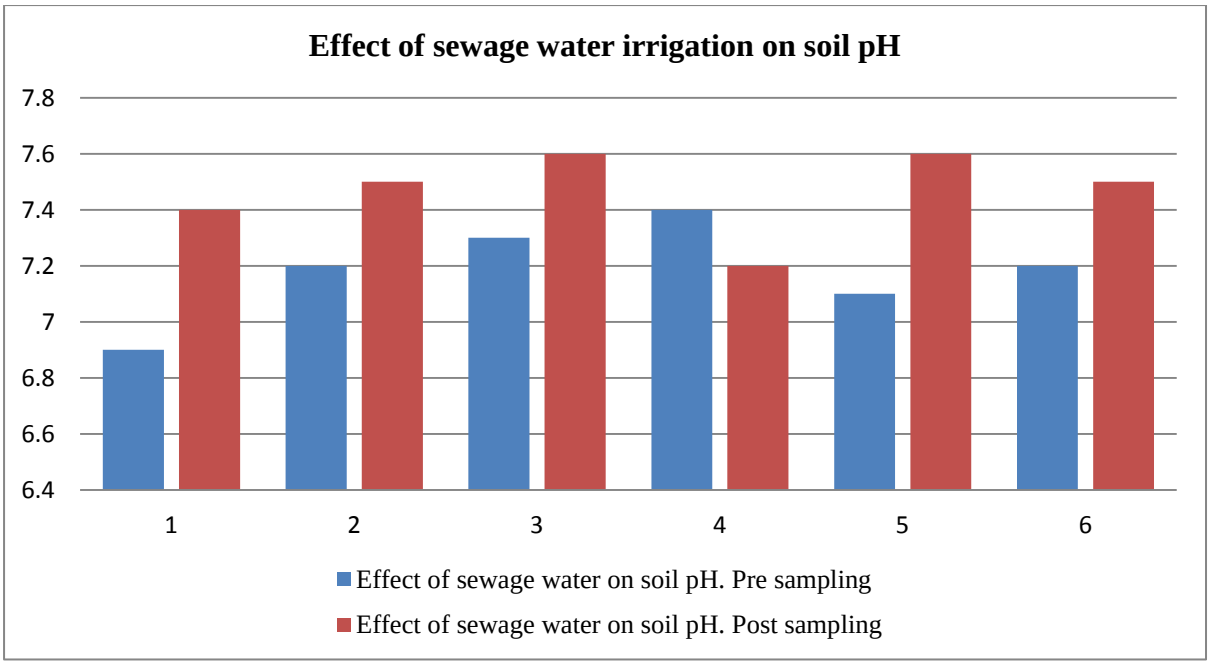

Fig.2: Effect of sewage water irrigation on soil pH.

The soil irrigation with wastewater may cause at first a diminishing of soil pH and after a long term used sewage water for irrigation cause an increase of soil pH due to the saline nature of sewage water.

4.3.3: Effect of sewage water irrigation on soil EC

The irrigation with sewage water is significantly increased at ($P<0.05$) of soil EC compared to before sewage water irrigation. The electrical conductivity of soils was increased due to irrigation with wastewater because of the highest electrical conductivity in sewage water (Table 4.7 and Fig.3). This is in line with the reported of Rusan *et al.* (2007), Jahantigh (2008). The presence of high amount of cations, for example, Na and K in wastewater promoted an expansion of EC.

4.3.4: Effect of sewage water irrigation on soil OC

The organic carbon in soils irrigated with sewage water is exceptionally significant at ($P<0.01$) when contrasted with organic carbon of the before sewage water irrigation. The higher quantity of organic carbon present in sewage water due to the municipal waste effluents. The organic carbon in the sewage water irrigated soil was seen higher which improved the infiltration rate, CEC and diminished the bulk density. Every one of these properties helped in improving the harvest profitability of the crop. Comparable outcomes were reported for by Datta *et al.* (2000), Yadav *et al.* (2002), Rattan *et al. (*2005), Gwenzi and Munondo (2008), Kesba *et al.* (2010), Verma *et al. (*2013) and Subramani *et al.* (2014).

4.3.5: Effect of sewage water irrigation on available macronutrients (N, P and K) of soil

Nitrogen, Phosphorus and Potassium were higher in the soils irrigated with sewage water in contrast with available N, P and K in the soil before sewage water irrigation. The expansion in N substance of soils flooded with sewage water because sewage water contained more nitrogen, which expands the available nitrogen in the soil. The P-value of available nitrogen of the soil flooded with sewage water of the current examination was significant at ($P<0.01$). In like manner, the available phosphorus and potassium substance of the sewage water applied soils were significant at ($P<0.05$) and ($P<0.05$) when compared with the before of sewage water. The phosphorus and potassium were higher due to higher P and K substance in applied sewage water. Comparable outcomes were reported for by Datta *et al.* (2000), Yadav *et. al.*, (2002), Rattan *et al.* (2005) and (Gwenzi and munondo, 2008).

Table 4.7 Effect of sewage water irrigation on soil EC

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	0.35	0.61
2	0.53	0.54
3	0.33	0.52
4	0.39	0.52
5	0.41	0.53
6	0.38	0.68
Mean	0.39	0.56
S.D	0.07	0.06
CV%	17.94	10.71

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.8 Effect of sewage water irrigation on soil OC

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	0.36	0.51
2	0.37	0.47
3	0.33	0.68
4	0.33	0.53
5	0.41	0.58
6	0.38	0.61
Mean	0.36	0.56
S.D	0.03	0.07
CV%	8.33	12.5

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

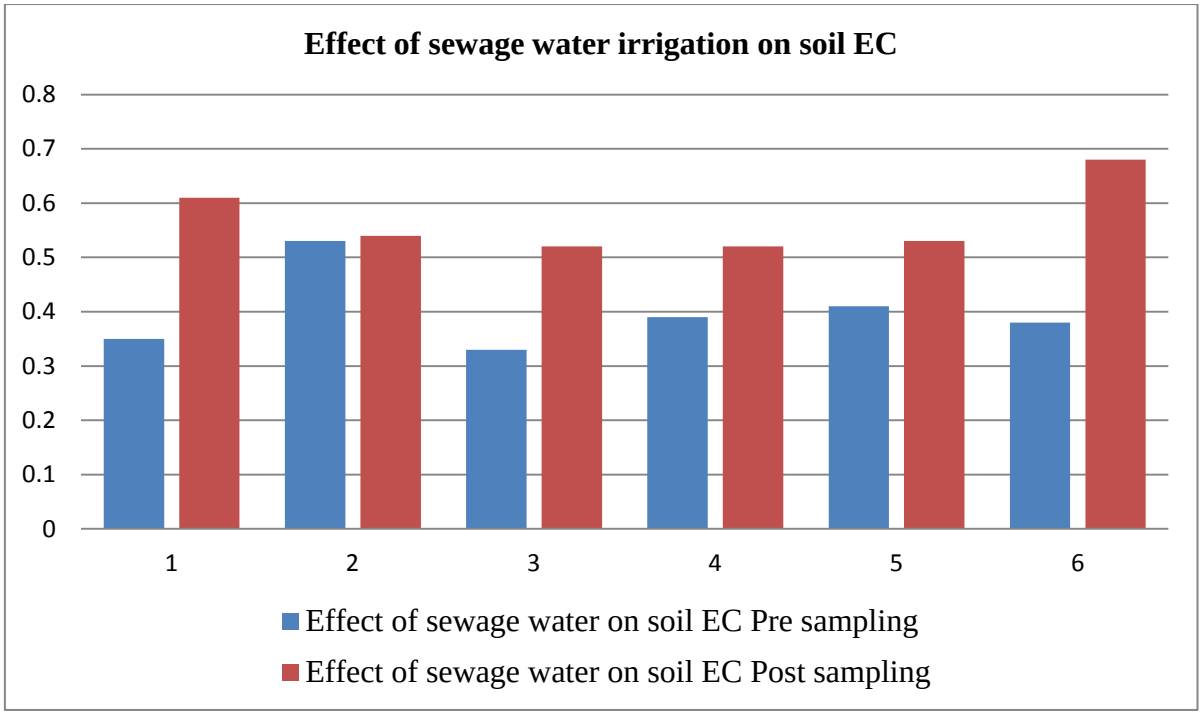

Fig.3: Effect of sewage water irrigation on soil EC.

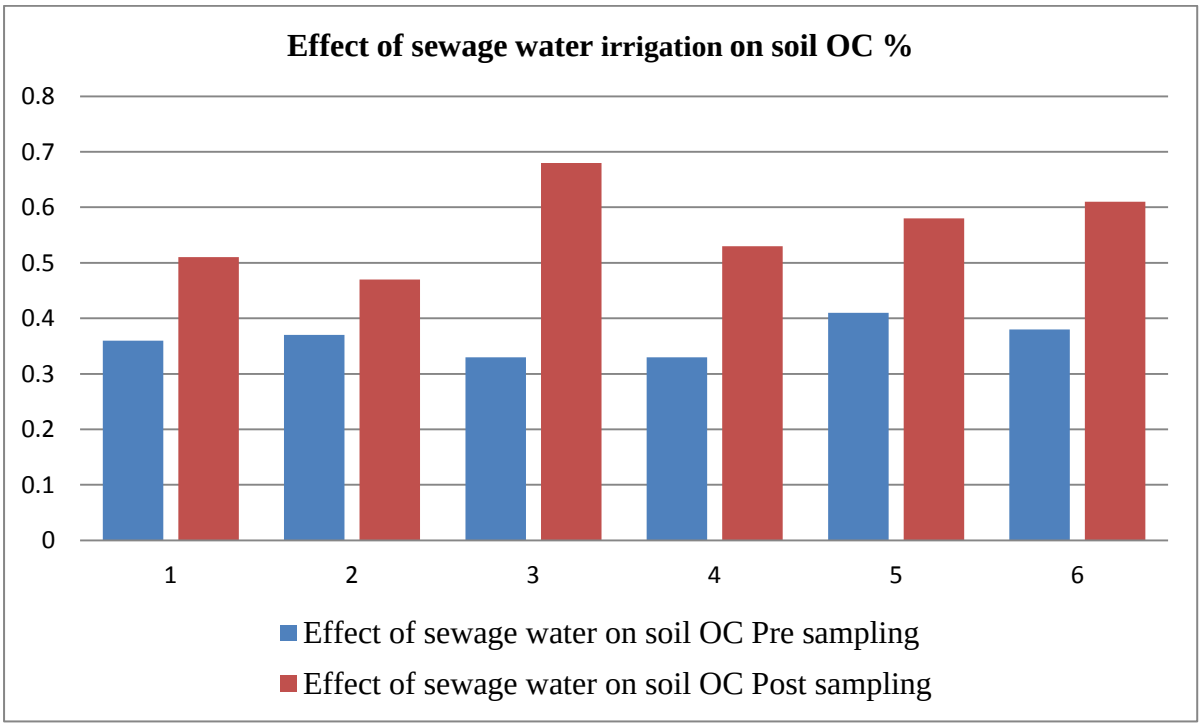

Fig.4: Effect of sewage water irrigation on soil OC

Table 4.9 The effect of sewage water irrigation on available nitrogen in the soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	255	331
2	212	362.5
3	190.5	217.5
4	137.5	300
5	167	266.5
6	152.5	261
Mean	185.75	289.75
S.D	43.11	52.31
CV%	23.2	18.05

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.10 The effect of sewage water irrigation on available Phosphorus

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	25.5	28.5
2	30	38
3	28	29
4	23.5	33
5	30.5	31
6	33.5	36
Mean	28.5	32.5
S.D	3.61	3.82
CV%	12.66	11.75

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

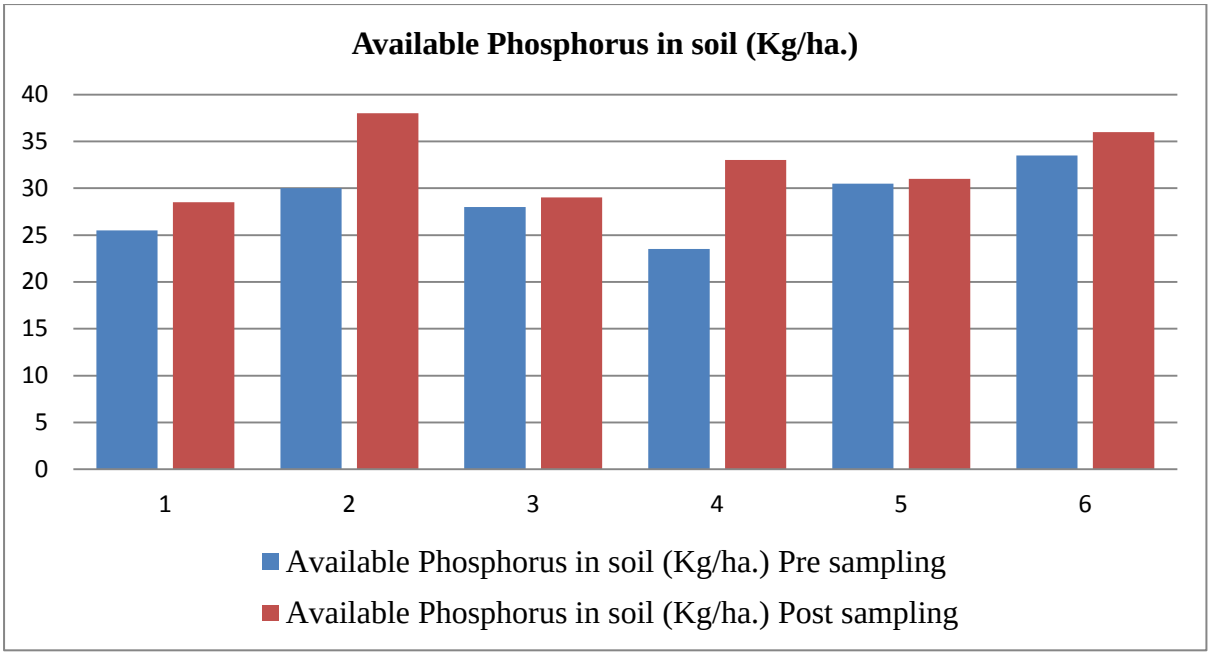

Fig.6: The effect of sewage water irrigation on available Phosphorus of soil

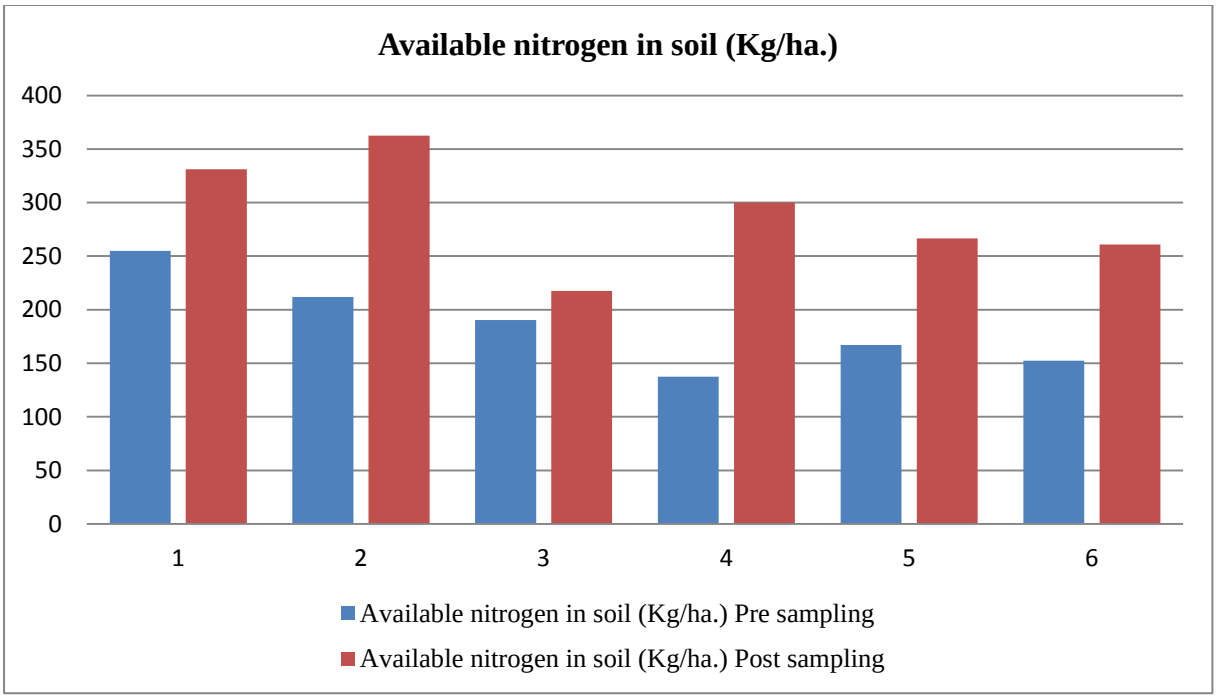

Fig.5: The effect of sewage water irrigation on available nitrogen in the soil

4.3.6: Effect of sewage water irrigation on available cations like Calcium and Magnesium of soil

The cations like calcium and magnesium were higher in the soils irrigated with sewage water compared with available cations in the soil before sewage water irrigation. The expansion in calcium substance of soils irrigated with sewage water because sewage water contained more calcium, which expands the available calcium in the soil. The P-value of available calcium in the soil flooded with sewage water of the current study was significant at ($P<0.05$). In like manner, the available magnesium substance of the sewage water applied soils were significant at ($P<0.05$) when compared with the before of sewage water irrigation. The magnesium was higher due to higher magnesium substance in applied sewage water. Comparable outcomes were reported by Gloaguen *et al.* (2007), Krishna and Govil (2008) and Kumar *et al.* (2011).

Table 4.11 The effect of sewage water irrigation on available Potassium

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	615.0	784.0
2	692.0	734.0
3	599.0	577.5
4	522.0	568.0
5	548.0	668.5
6	512.0	660.5
Mean	581.3	665.4
S.D	68.01	84.87
CV%	11.69	12.75

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.12 The effect of sewage water irrigation on available Calcium of soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	620	700
2	722.5	807.5
3	623	645
4	561.5	597.5
5	532	546.5
6	541.5	557
Mean	600.08	642.25
S.D	71.36	99.08
CV%	11.89	15.42

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.13 The effect of sewage water irrigation on available Magnesium of soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	313.5	447.5
2	357.5	401
3	315	397
4	330.5	380.5
5	323.5	328.5
6	316	335
Mean	326	381.5
S.D	16.7	44.6
CV%	5.12	11.69

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

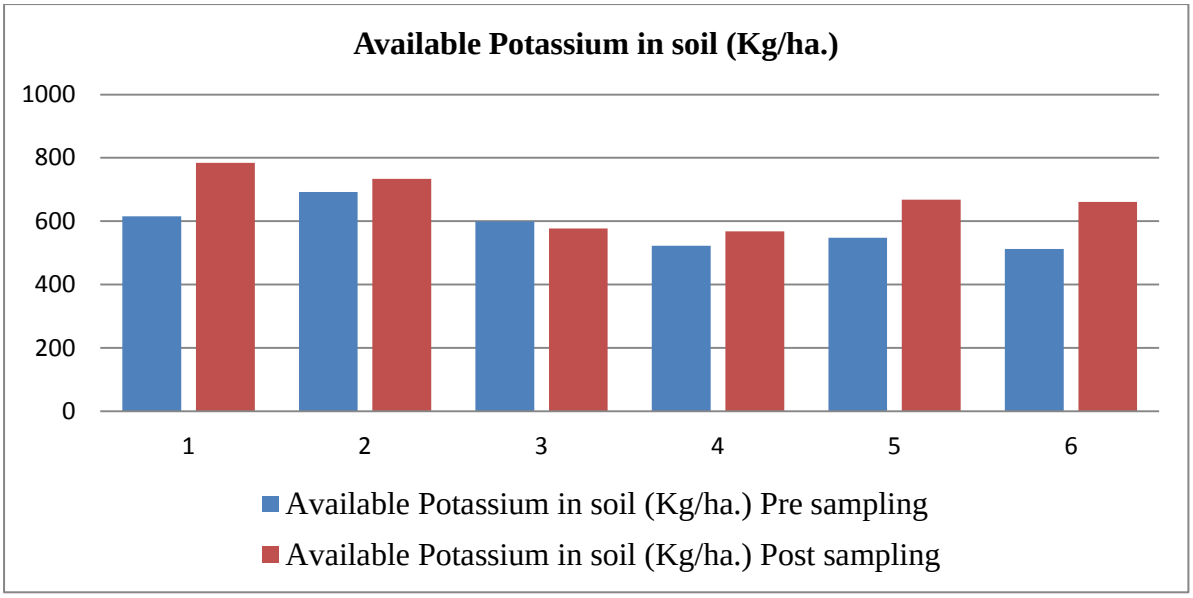

Fig.7: The effect of sewage water irrigation on available Potassium of soil

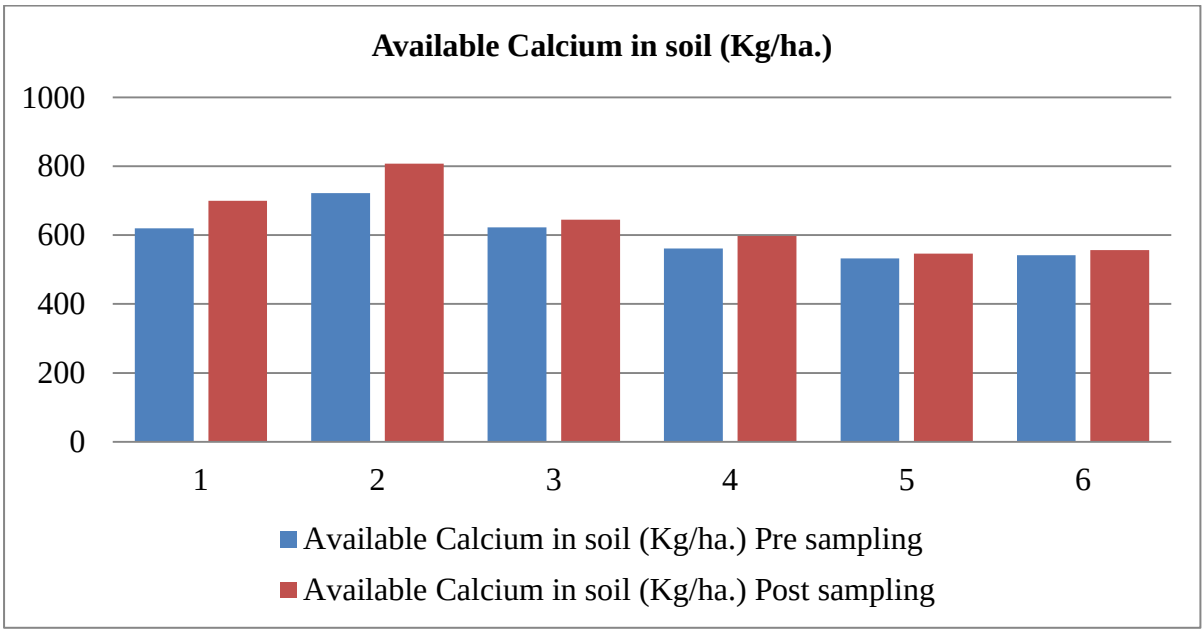

Fig.8: The effect of sewage water irrigation on available Calcium of soil

4.3.6: DTPA extractable micronutrients (Zn, Cu, Fe and Mn)

The DTPA extractable microelements like Zn, Cu, Fe and Mn of the highest in soils flooded with sewage water in the current investigation has significant at ($P<0.01$), ($P<0.05$), ($P<0.01$) and ($P<0.01$) compared with the before sewage water irrigation. The higher concentration of micronutrient in the soil makes toxicity for plant development.

Table 4.14 The effect of sewage water irrigation on available Zink

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	0.33	2.81
2	0.34	3.04
3	0.28	1.94
4	0.18	3.07
5	0.18	2.95
6	0.26	3.3
Mean	0.26	2.85
S.D	0.06	0.47
CV%	23.07	16.49

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.15 The effect of sewage water irrigation on the Copper concentration of soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	1.29	1.59
2	1.04	2.61
3	0.56	1.64
4	0.79	1.55
5	1.03	1.17
6	0.92	1.43
Mean	0.93	1.66
S.D	0.24	0.49
CV%	25.8	29.51

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

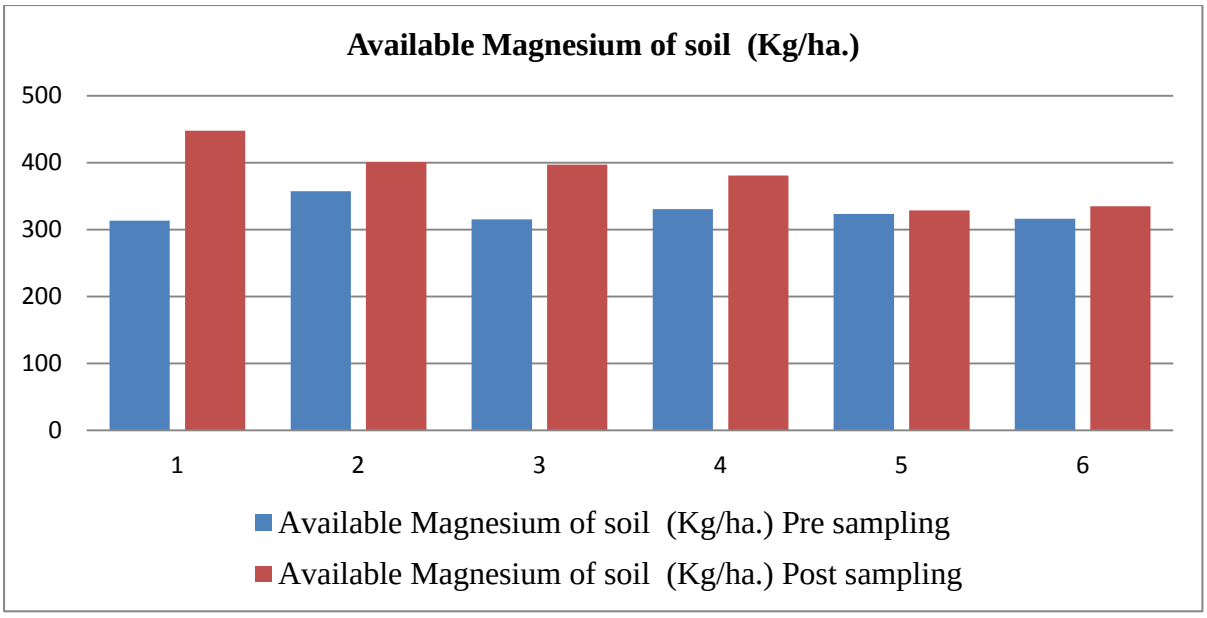

Fig.9: The effect of sewage water irrigation on available Magnesium of soil

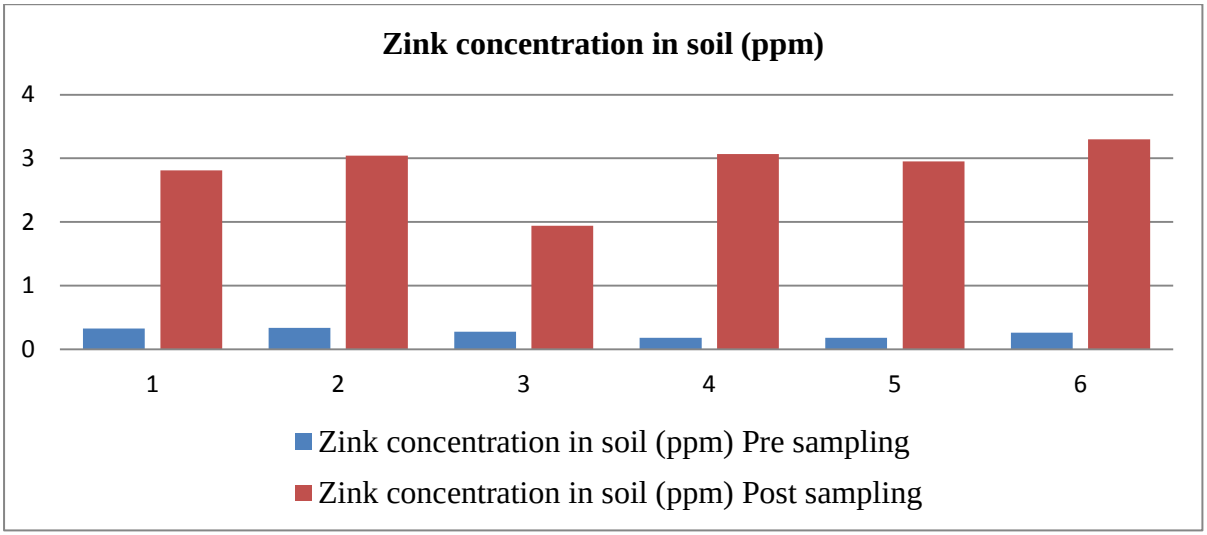

Fig.10: The effect of sewage water irrigation on Zink concentration of soil

The sewage water contained all these DTPA extractable microelements presences in higher concentration. The higher concentration of micronutrients in the soil due to sewage water applied as a source of irrigation. Comparable outcomes were accounted for by Datta *et al.* (2000), Rattan *et al.* (2001), Patel *et al.* (2004), Rattan *et al.* (2005) and Suhag *et al.* (2008), Oman et al. (2010), Prakash *et al.* (2010), Kharche *et al*. (2011) and Zan *et al.* (2013).

Table 4.16 The effect of sewage water on Iron concentration of soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	3.92	6.5
2	3.99	8.95
3	3.09	4.71
4	2.58	5.18
5	3.31	5.35
6	3.62	5.08
Mean	3.41	5.96
S.D	0.53	0.53
CV%	15.54	26.51

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Table 4.17 The effect of sewage water irrigation on Manganese concentration of soil

Sample no.	Pre sampling Average of surface and subsurface soil	Post sampling Average of surface and subsurface soil
1	14.16	33.06
2	14.59	29.29
3	13.00	28.68
4	13.15	28.65
5	14.39	31.75
6	14.51	27.16
Mean	13.96	29.76
S.D	0.7	2.2
CV%	5.01	7.39

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

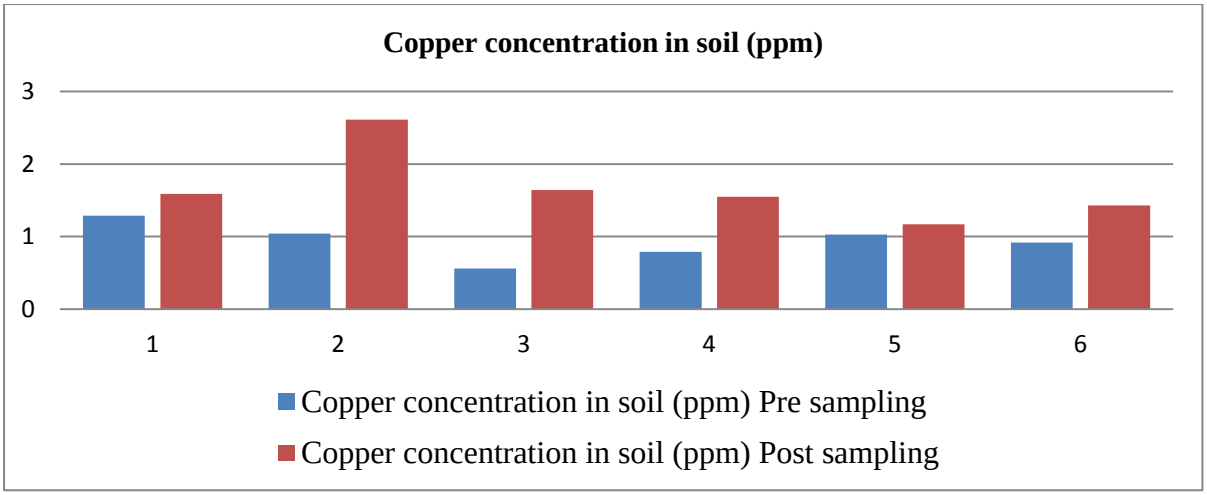

Fig.11: The effect of sewage water irrigation on the Copper concentration of soil

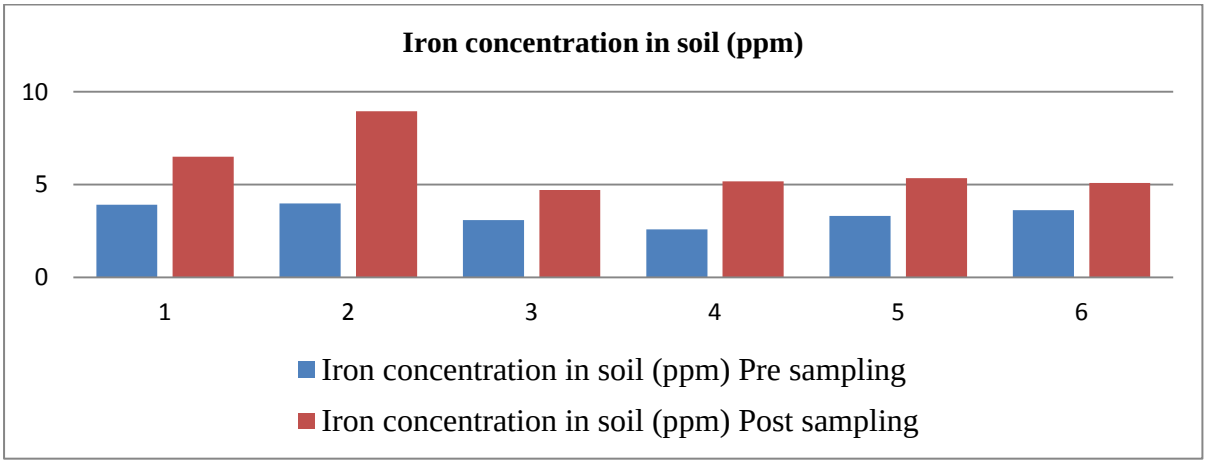

Fig.12: The effect of sewage water irrigation on Iron concentration of soil

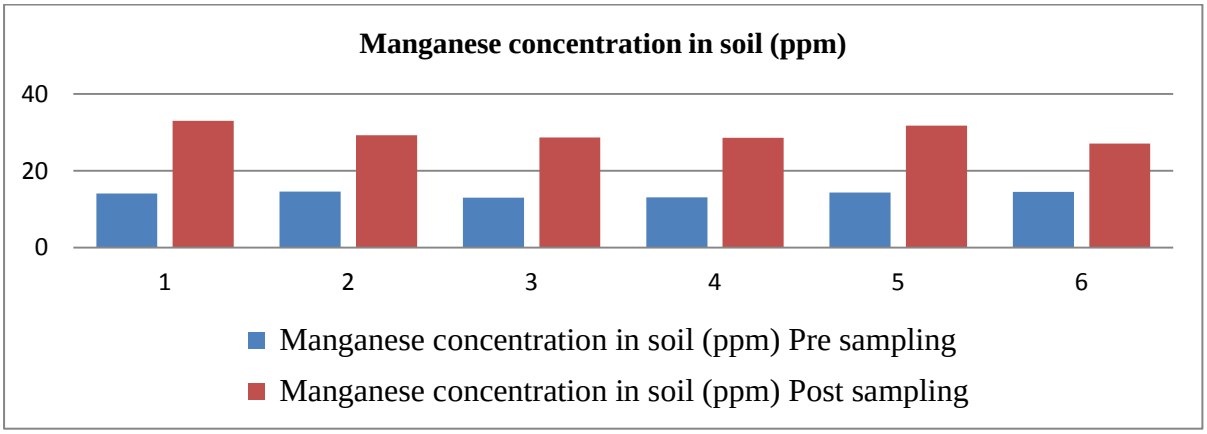

Fig.13: The effect of sewage water irrigation on Manganese concentration of soil

4.4: Impact of sewage water irrigation on microbial population and activity of soil

4.4.1: Microbial biomass carbon

The Microbial Biomass Carbon (MBC) is significantly expanded at ($P<0.01$) in sewage water flooded soil compared with before sewage water irrigation. This is a source or sinks of accessible nutrients and it assumes a basic job in nutrients transformation in earthly environments (Singh *et al.,* 1989). The quantity of the microbial biomass may influence by organic matter addition in soil, here upon the soil microbial action completely influenced by soil environmental system (Smith *et al.,* 1993). In the current examination, MBC was estimated higher in the soils flooded with sewage water when compared with the soils flooded with the non-sewage water. Higher MBC in sewage applied soil was because of higher soil organic substance of these soils which rests into the higher microbial mass. The microbial biomass carbon value ranged from 165.5 mg kg^{-1} (before sewage water irrigation) to 213.66 mg kg^{-1} (field irrigated with sewage water). The microbial biomass carbon was higher in soil flooded with treated wastewater than the before sewage water irrigation. This outcome affirmed that found in the writing that the bacterial population was commonly present in the soil compartment wealthy in organic matter (Palese *et al.* 2009). The MBC diminished with the increasing soil depth.

Table 4.18 Effect of sewage water irrigation on microbial biomass carbon of soil

Microbial biomass carbon (mg CO_2/kg of soil)		
Sample no.	Pre sampling	Post sampling
1	182	246
2	192	228
3	135	195
4	147	212
5	164	218
6	173	183
Mean	165.5	213.6
S.D.	21.47	22.63
CV%	12.97	10.59

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

Comparative outcomes were accounted for by Antil *et al.* (2004), Zan *et al.* (2013), Kharche *et al.* (2011), Oman *et al.* (2010), Suhag *et al.* (2008), Rattan *et al.* (2005), Rattan *et al.* (2001) and Datta *et al.* (2000).

4.4.2: Effect of sewage water on Basal soil respiration

Soil basal respiration is the balanced rate of respiration in soil, which begins from the mineralization of organic matter (Pell *et al.*, 2006) and is assessed either based on CO_2 enhancement or O_2 up-take (Dilly and Zyakun, 2008). The Soil basal respiration is significantly increased at ($P<0.01$) in sewage water flooded soil appeared differently before sewage water irrigation. The mean estimation of the soil basal respiration extended between 0.513 (Pre sampling) and 0.692 (Post sampling) mg C-CO_2/hr./100 gram of soil. The higher basal respiration of soil because presences of high organic carbon in sewage water irrigated soil. Similar results reported by (Cacio Luiz Boechat *et al.*, 2012). Reports from Cleveland *et al.* (2007) demonstrated that the expanded reaction in soil microbial action was related to easily accessible C sources in the soil.

Table 4.19 Effect of sewage water on Basal soil respiration

Basal soil respiration (mg C-CO_2/h/100gram of soil)		
Sample no.	Pre sampling	Post sampling
1	0.55	0.715
2	0.605	0.855
3	0.495	0.55
4	0.385	0.66
5	0.605	0.715
6	0.44	0.66
Mean	0.513	0.692
S.D.	0.08	0.09
CV%	15.59	13

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

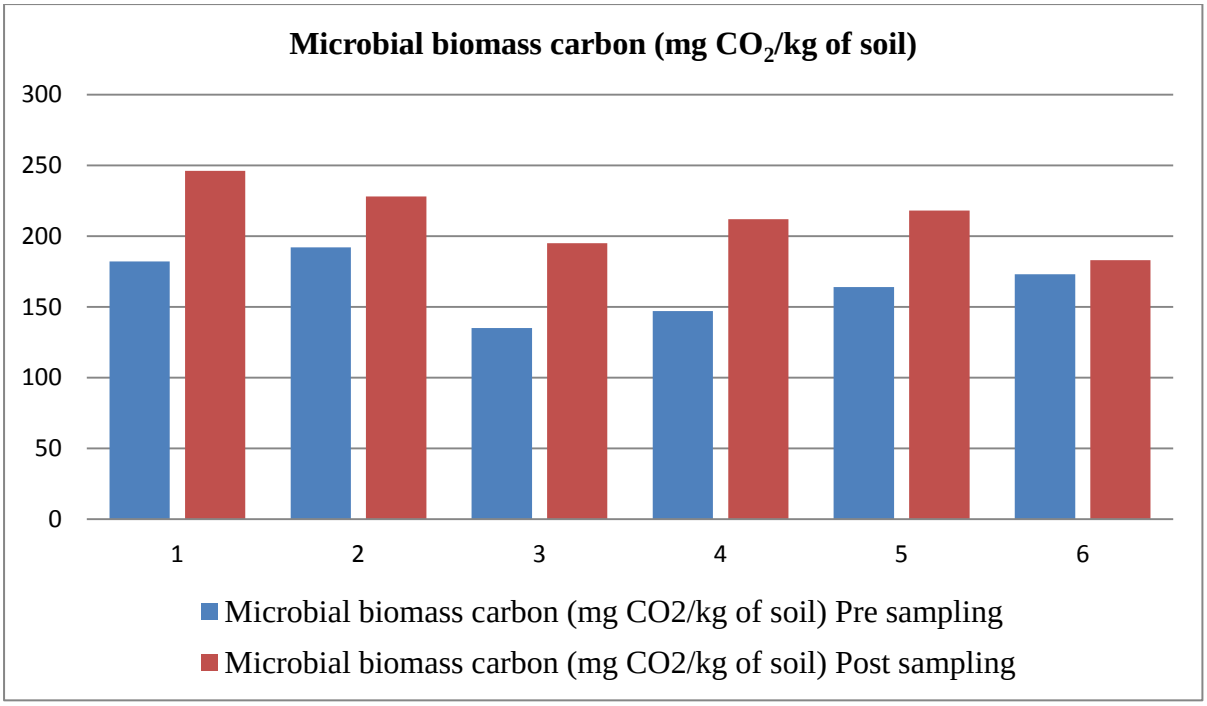

Fig.14: Effect of sewage water irrigation on microbial biomass carbon of soil

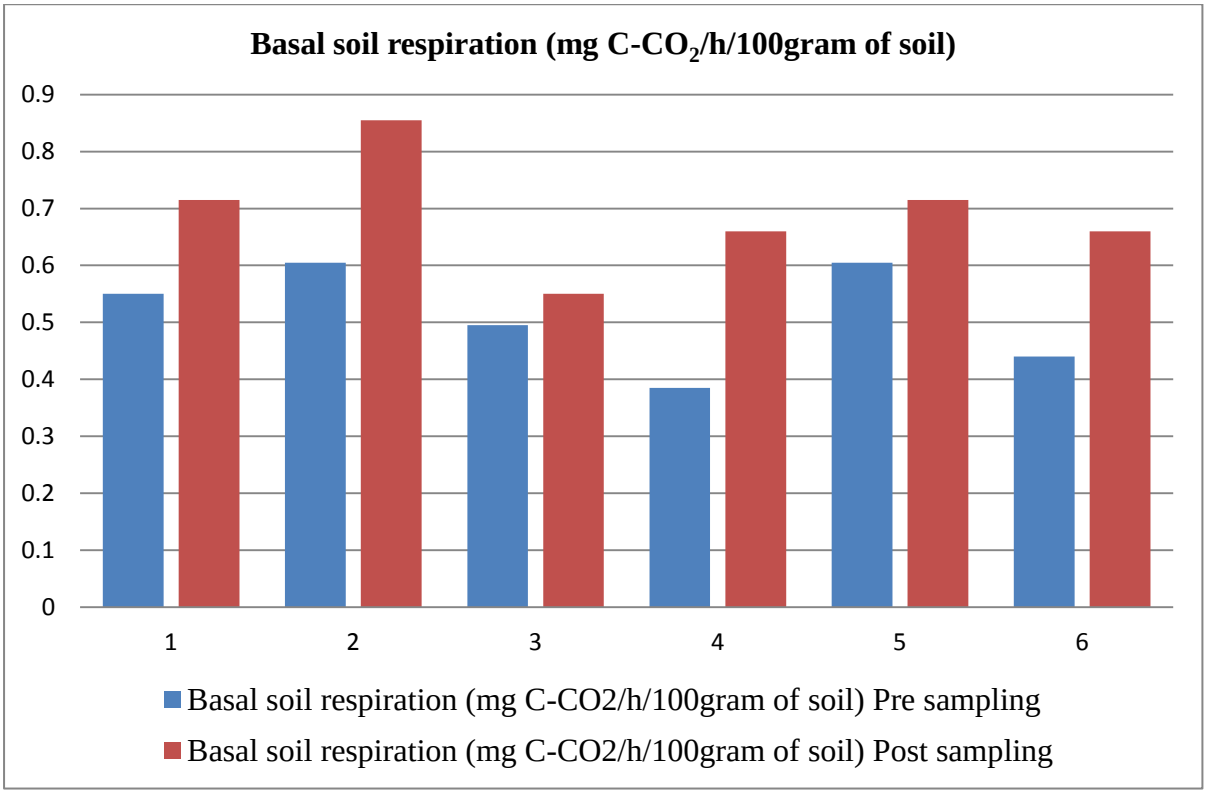

Fig.15: Effect of sewage water on Basal soil respiration

4.5: Dynamics of microbial population pre and post-exposure to sewage water irrigation

4.5.1: Total Viable Count of bacteria

In sewage irrigated soil, the number of microorganisms was expanded significantly at ($P<0.01$) as compared with pre sampling. The mean value of total viable bacteria in sewage water irrigated soil was 8.38 and 7.90 before sewage water irrigation. The microbial population in sewage- flooded soils was higher for bacteria, fungi and actinomycetes when compared with the pre sampling, Kharche *et al.* (2011). This might be higher because of the suspended organic material added to the soil through sewage which works as a source of vitality for a microbial population. Comparative outcomes were seen by (Seaker and Sopper, 1988, Joshi and Yadav, 2003).

Table 4.20 Effect of sewage water irrigation on Total Viable Count of bacteria

Sample no.	The Total Viable Count of the bacterial population on Nutrient agar Media			
	Pre sampling		Post Sampling	
	cfu/gram	Log10 Value	cfu/gram	Log10 Value
1	$1.37*10^8$	8.13	$2.95*10^8$	8.46
2	$1.15*10^8$	8.06	$2.73*10^8$	8.43
3	$8.7*10^7$	7.75	$2.46*10^8$	8.39
4	$7.8*10^7$	7.89	$1.98*10^8$	8.29
5	$6.1*10^7$	7.78	$2.42*10^8$	8.38
6	$6.7*10^7$	7.82	$2.33*10^8$	8.36
Mean	7.79		8.38	
S.D.	0.15		0.05	
CV%	1.95		0.59	

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

4.5.2: Effect of sewage water irrigation on *E. coli.* Population:-

Wastewaters describe as the fundamental wellspring of water contamination in various pieces of the world, for example, Egypt (Yehia and Sabae, 2011); Poland (Niewolak, 2000); Nigeria (Akaninwor *et al.*, 2007) and Brazil (Gunkel *et al.*, 2007). Some researchers have inspected the vehicle of microorganisms in the soil after application of sewage water, (Schaub and Sorber, 1977, Smith *et al.*, 1985). Yongabi *et al.* (2011) were detailed by

introducing that contaminated water contains a high quantity of microorganism. The fecal coliform microbes demonstrate that a waterway contaminated through sewage and the presence of other pathogenic microbes. The irrigation with sewage water was significantly expanded at ($P<0.01$) fecal coliform population of soil when compared with before sewage water irrigation.

The soils are highly polluted because of the long term used of sewage water for irrigation. The various harmful microorganisms produced when irrigation with contaminated water irrigation like contaminated untreated sewage or fertilizer. Some strains of *E. coli* cause sickness in people, including the fever, diarrhea, abdominal pain and vomiting. The *E. coli 0157:H7* strain is one of the strains, which deliver a poison known as Shiga. It is one of the most impressive poisons, and it can cause intestinal contamination, (Yvette Brazier 2017).

Table 4.21 Effect of sewage water irrigation on *E. coli.* Population

Sample no.	*E. coli.* population on MacConkey Agar media			
	Pre sampling		Post Sampling	
	cfu/ gram	Log 10 Value	cfu/ gram	Log 10 Value
1	$1.07*10^9$	9.02	$2.50*10^9$	9.39
2	$1.73*10^9$	9.23	$2.67*10^9$	9.42
3	$1.39*10^9$	9.14	$2.30*10^9$	9.36
4	$1.86*10^9$	9.26	$2.59*10^9$	9.41
5	$1.23*10^9$	9.08	$2.25*10^9$	9.35
6	$1.61*10^9$	9.2	$2.12*10^9$	9.32
Mean	9.15		9.37	
S.D.	0.09		0.03	
CV%	0.98		0.32	

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

4.6 :Effect on PGPR population pre and post-exposure to sewage water irrigation

After investigation of population density of PGPRs separated from the soil irrigated with effluent water of wastewater treatment plants. It is seen that number of advantageous micro fauna were significantly diminished like – *Rhizobium* population of soil as compared with the before sewage water irrigation.

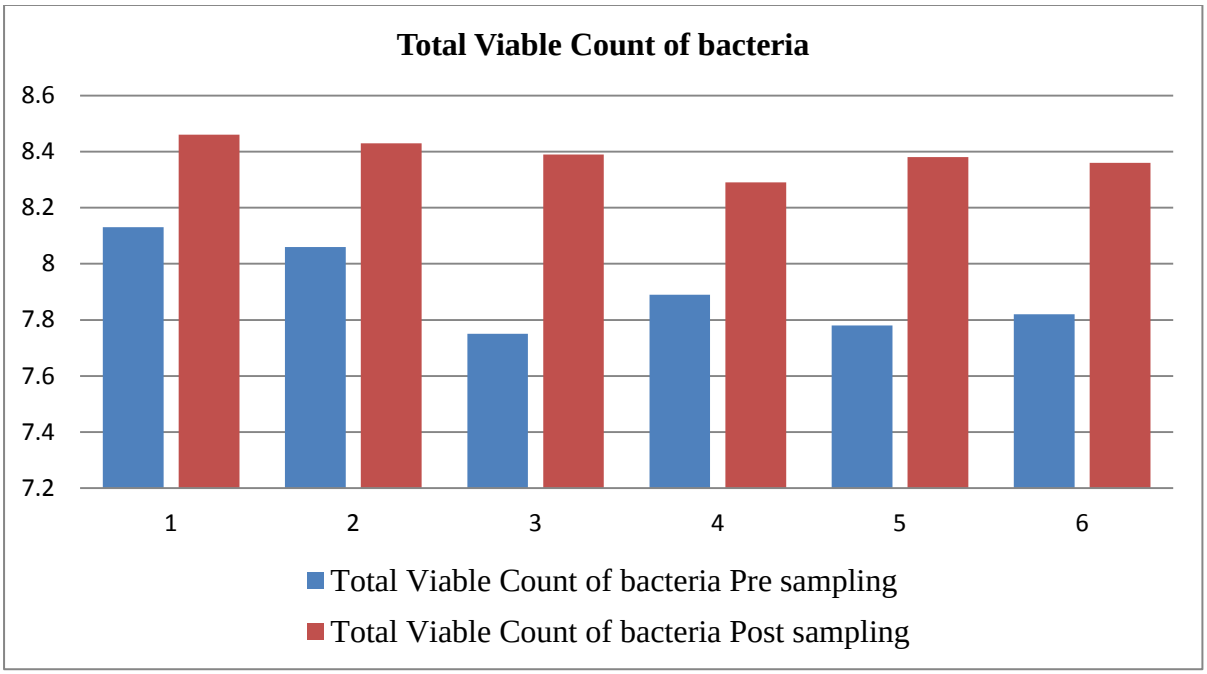

Fig.16: Effect of sewage water irrigation on Total Viable Count of bacteria

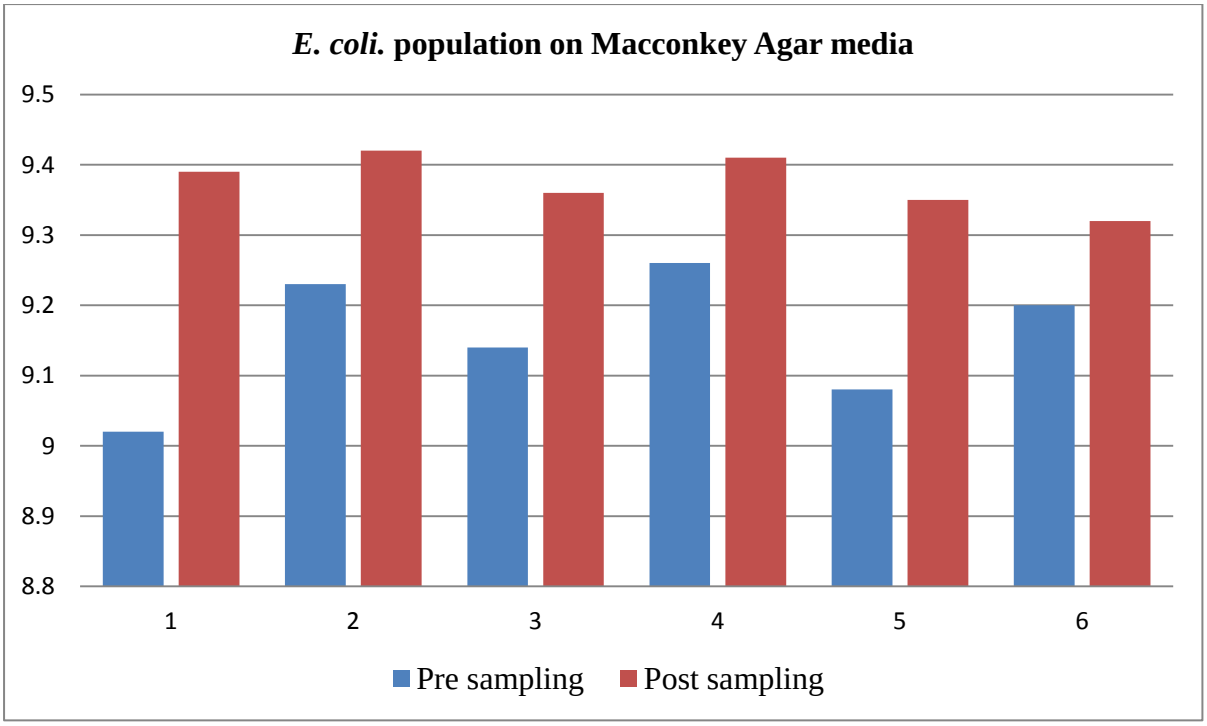

Fig.17: Effect of sewage water irrigation on *E. coli.* Population

Plate 10: Total Viable Count of bacteria on Nutrient Agar Media

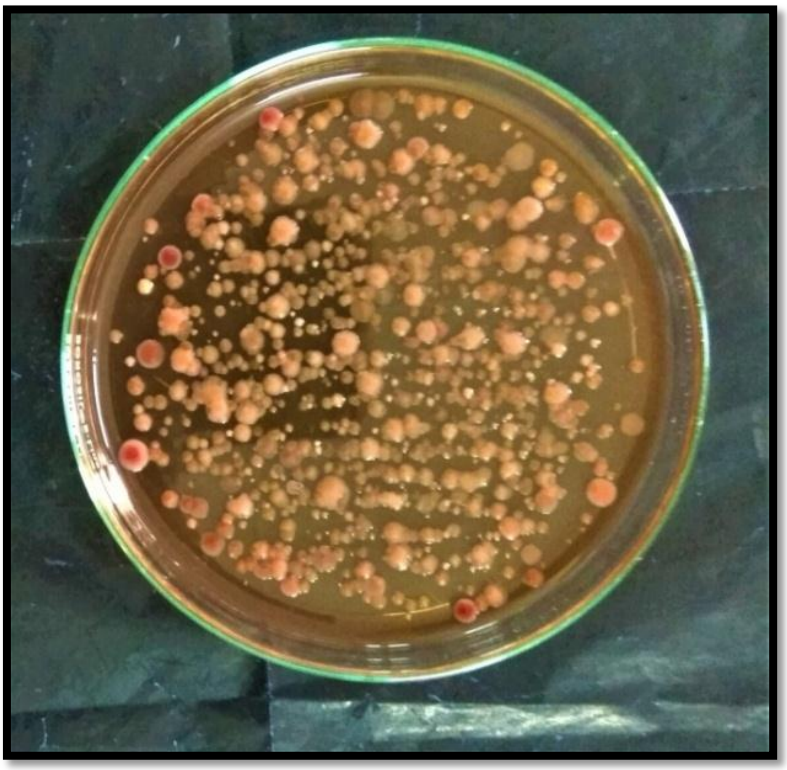

Plate 11: *E. coli.* Population on MacConkey Agar Media

Plate 12: Population of *Rhizobium species* on Yeast Extract Mannitol Agar Media

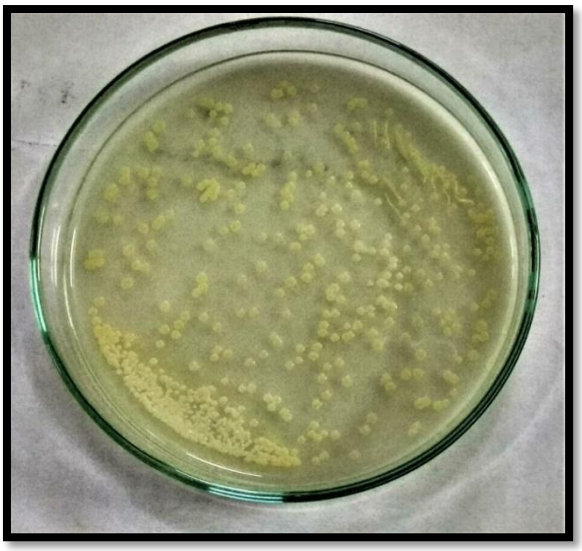

Plate 13: Population of *Pseudomonas species* on ‘kings B’ Agar Media

Plate 14: Population of *Azotobacter species* on “Jenson N free” Agar Media

The heavy metal contaminants like Ag, Hg, Pb, Zn, As, Cd, Cr, Na, K and Cu harms for PGPRs and other helpful microorganisms in the contaminated soil. The presence of Ag nano particle decreased the CFU of all PGPR organisms. This line is similar with the reported of Revathi *et al.,* (2011).

4.6.1: Effect of sewage water irrigation on *Rhizobium sp.*

The irrigation with sewage water is significantly decreased in ($P<0.01$) *Rhizobium* population of soil as compared to before sewage water irrigation. The *Rhizobium* population of soils was decreased due to irrigation with wastewater because of the high concentration of heavy metals in sewage water. Comparative outcomes were seen by (Castro *et al.* 1997; Chaudhary *et al.* 2004; Chaudri *et al.* 2000; Lakzian *et al.* 2002; Smith 1997) who studied the effects of heavy metals on growth, development, morphology and physiology of different strains of *R. leguminosarum*. Continuous introduction of heavy metals changes the suitable bacterial cells into a non-reasonable structure, (Paton *et al.* 1997; Hirsch *et al.* (1993). Decrease in bacterial cells of *Rhizobium sp.* have been accounted with the expanding concentration of heavy metals, for example, Cu, Zn and Pb, either solely or in mixes, (Stan *et al.,* 2011).

Table 4.22 Effect of sewage water irrigation on *Rhizobium sp.*

Sample no.	The population of *Rhizobium sp.* on YEMA media			
	Pre sampling		Post Sampling	
	cfu/gram	Log10 Value	cfu/gram	Log10 Value
1	$8.9*10^6$	6.94	$7.6*10^6$	6.88
2	$8.5*10^6$	6.92	$6.7*10^6$	6.82
3	$6.3*10^6$	6.79	$4.5*10^6$	6.65
4	$7.3*10^6$	6.86	$5.9*10^6$	6.77
5	$8.5*10^6$	6.92	$6.7*10^6$	6.82
6	$6.4*10^6$	6.8	$5.6*10^6$	6.74
Mean	6.87		6.78	
S.D.	0.06		0.07	
CV%	0.87		1.03	

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

4.6.2: Effect of sewage water irrigation on *Pseudomonas species*

A few types of *Pseudomonas species* found in the environment, for example, plant growth promoters (Sullivan *et al.,* 1992), plant pathogenic (Schroth *et al.,* 1991) and xenobiotic degraders (Ridgway *et al*, 1990). The results indicated that urban wastewater contains a high amount of microorganism and the endurance capability of different PGPR to grow heavy metals is varied. The *Pseudomonas sp.* can multiply better and had the most extreme CFU. In the availability of city wastewater, the noticeable increment in CFU of *Pseudomonas sp.* might be the presence of organic matter in the urban wastewater which filled as C/N source for the PGPR. Comparable results were accounted by Yongabi *et al.* (2011).

Table 4.23 Effect of sewage water irrigation on *Pseudomonas sp.*

	The population of *pseudomonas spp.* on "Kings B" Agar Media			
Sample no.	Pre sampling		Post Sampling	
	cfu/ gram	Log 10 Value	cfu/ gram	Log 10 Value
1	$4.5*10^5$	5.65	$4.8*10^5$	5.68
2	$3.0*10^5$	5.54	$4.2*10^5$	5.62
3	$3.2*10^5$	5.5	$3.9*10^5$	5.59
4	$2.8*10^5$	5.44	$5.2*10^5$	5.71
5	$4.2*10^5$	5.62	$6.2*10^5$	5.79
6	$2.6*10^5$	5.41	$4.1*10^5$	5.61
Mean	5.52		5.66	
S.D.	0.09		0.07	
CV%	1.63		1.23	

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

4.6.3: Effect of sewage water irrigation on *Azotobacter species*

The quantity of *Azotobacter* in sewage irrigated soils was expanded as compared with the before sewage water irrigation, shown significantly by statistical analysis ($P<0.05$). The free-living nitrogen-fixing microorganisms are first expanded and afterwards diminished after sewage water irrigation (Shang *et al.,* 2007). The increasing of *Azotobacter* population in sewage water irrigated soil due to high organic matter present in sewage water. This line is similar to reported for by (Zhang *et al.,* 2007).

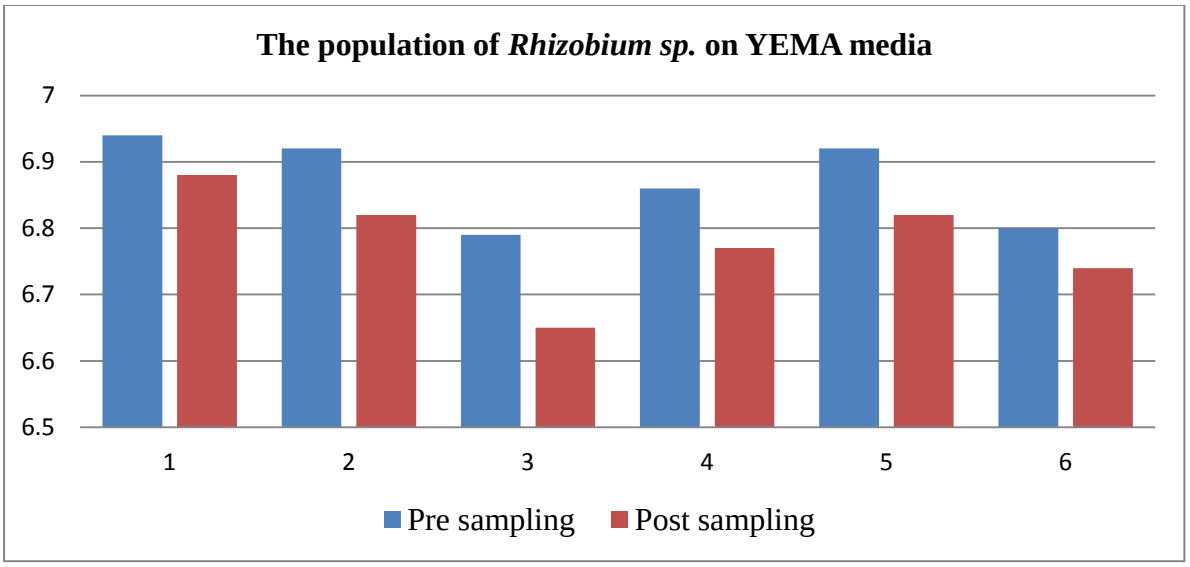

Fig.18: Effect of sewage water irrigation on *Rhizobium sp.*

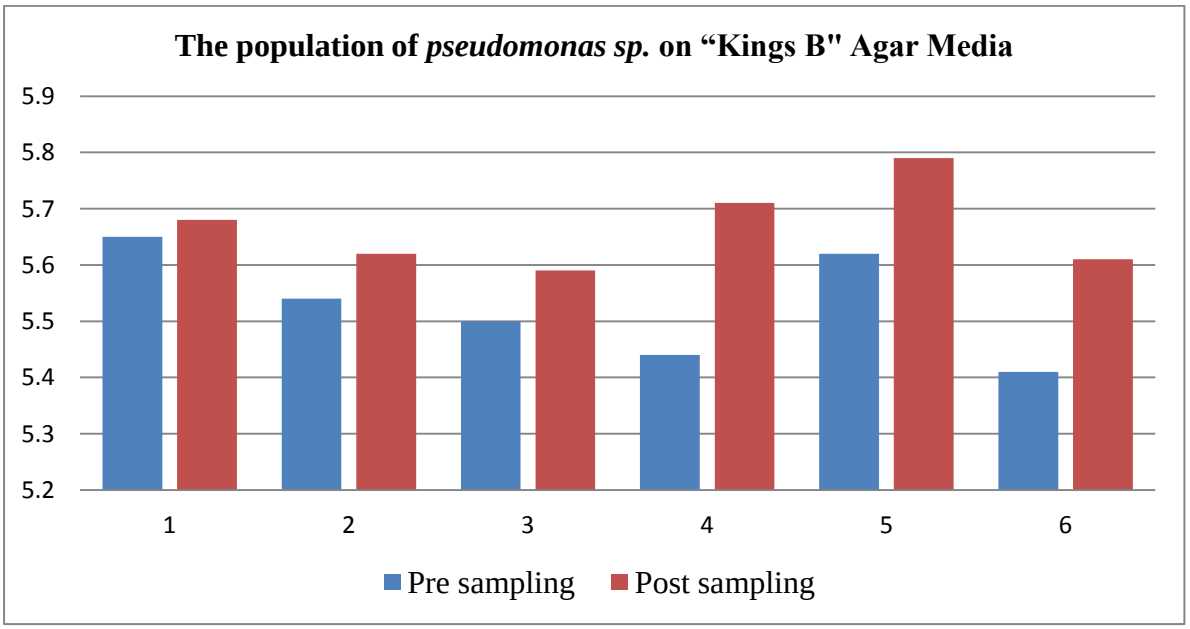

Fig.19: Effect of sewage water irrigation on *Pseudomonas species*

Table 4.24 Effect of sewage water irrigation on *Azotobacter sp.*

Sample no.	The population of *Azotobacter spp*. on "Jenson N free" Agar Media			
	Pre sampling		Post Sampling	
	cfu/ gram	Log10 Value	cfu/ gram	Log10 Value
1	$8.0*10^4$	4.9	$9.3*10^4$	4.96
2	$5.0*10^4$	4.69	$8.6*10^4$	4.93
3	$4.6*10^4$	4.66	$6.1*10^4$	4.78
4	$5.6*10^4$	4.74	$7.5*10^4$	4.87
5	$6.9*10^4$	4.83	$7.3*10^4$	4.86
6	$5.9*10^4$	4.77	$6.3*10^4$	4.79
Mean	4.76		4.86	
S.D.	0.08		0.07	
CV%	1.68		1.44	

Pre sampling = before sewage water irrigation, Post sampling = after sewage water irrigation

4.7: The effect of sewage water irrigation on cauliflower yield

The irrigation with sewage water is significantly increased at ($P<0.01$) of cauliflower yield compared to tube-well water irrigation. The cauliflower yield was increased due to irrigation with sewage wastewater because of the highest nutrient concentration found in sewage water. This is in line with the reported of (Sheikh *et al.*, 2012).

Table 4.25: Comparison of cauliflower yield between fields irrigated with tube-well water and sewage water

Sample no.	Field irrigated with tube-well water	Field irrigated with sewage water
	Weight of curd (Gram)	Weight of curd (Gram)
1	490	540
2	443	625
3	425	554
4	448	575
5	408	605
6	428	518
7	380	530
8	432	570
9	465	598
10	473	515
Mean	439.2	563
S.D	32.21	36.13
CV%	7.33	6.41

(Used two sample t-test for independent group)

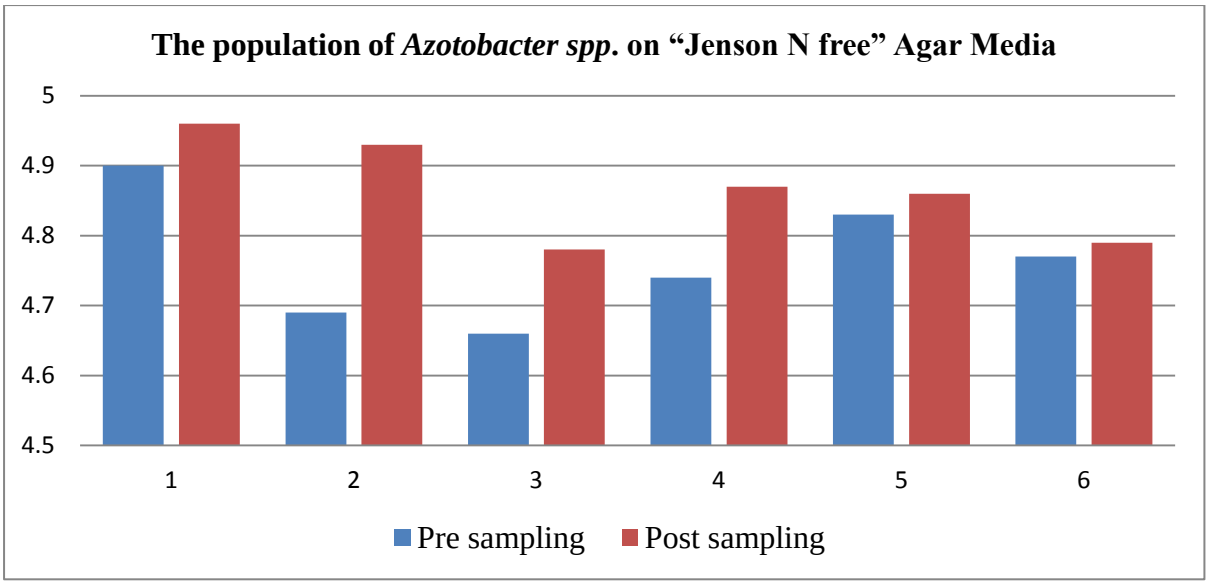

Fig.20: Effect of sewage water irrigation on *Azotobacter sp.*

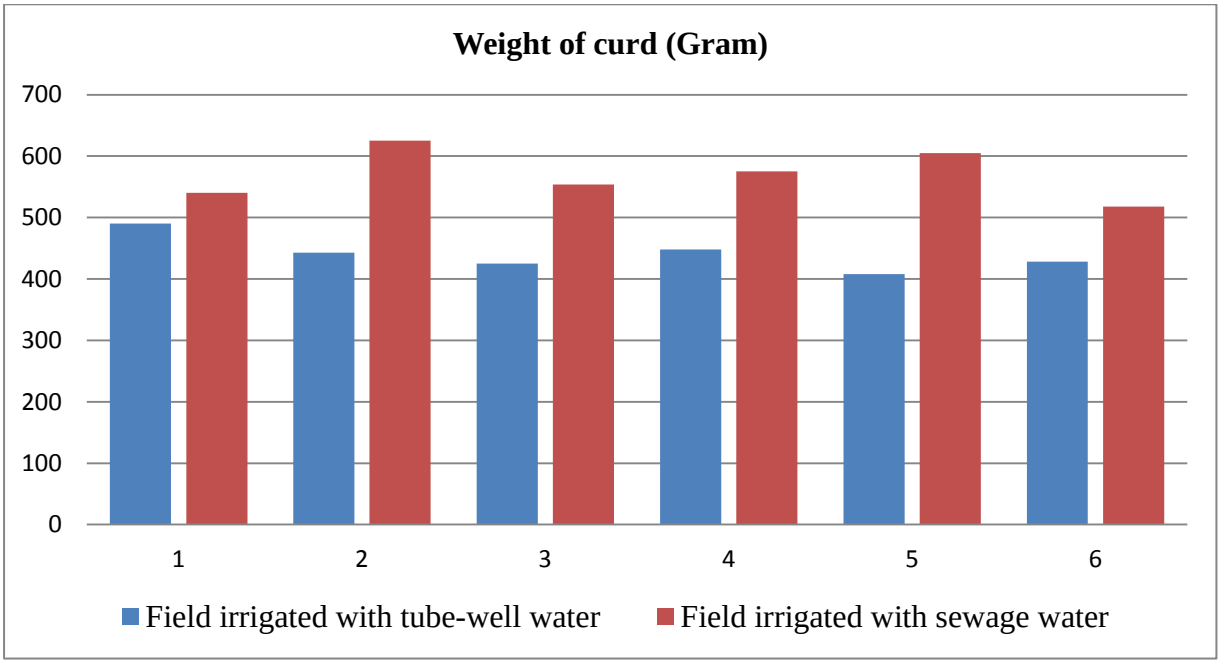

Fig.21: Comparison of cauliflower yield between fields irrigated with tube-well water and sewage water

CHAPTER – V

SUMMARY AND CONCLUSIONS

The present investigation **"Impact of sewage water irrigation on soil microbial diversity and Cauliflower productivity in Rajnandgaon District (C.G.)"** was conducted at farmer's field and all observations analyzed at Pt. Shiv Kumar Shastry College of Agriculture and Research Station, Rajnandgoan, Department of Soil Science and Agricultural Chemistry and Department of Agricultural Microbiology, Indira Gandhi Krishi Vishwavidyalaya, Raipur during the year 2019-20. The investigation was done to study the impact of sewage water irrigation on soil qualities. The 't' test for a paired sample selected as a statistical tool for comparison between before sewage water irrigation and after sewage water irrigation of soil properties.

The important findings of the present investigation are summarized below:-

I. The concentration of heavy metals *e.g.* - Zn, Mn, Fe and Cu higher in sewage water compare to FAO standard parameters.

II. The *E. coli.* Population compared higher in sewage water and groundwater compare to standard parameter given by BIS and WHO.

III. The bulk density of soil significantly decreased after sewage water irrigation compared to before sewage water application.

IV. The chemical properties of soil like - pH, Electrical Conductivity (EC), Organic Carbon, Nitrogen, Phosphorus, Potassium, Calcium, Magnesium, Zinc, Iron, Copper and Manganese are significantly increased after sewage water irrigation.

V. The soil microbial biomass carbon and basal soil respiration significantly increased when field irrigated with the sewage water.

VI. After sewage water irrigation, the Total Viable Count of bacteria (TVC) and Fecal coliforms are unbelievably increased.

VII. Some PGPR of soil *e.g.* - *Rhizobium sp.* significantly decreased Whereas, *Pseudomonas sp.* and *Azotobacter sp.* is significantly increased.

CONCLUSIONS

- The results revealed that use of sewage water for irrigation in Village - Paneka, District – Rajnandgaon (C.G) was found to improve the physical, chemical, biological properties (except *Rhizobium sp.)* of the soil and Cauliflower yield without any treatment and all the heavy metals like- Zn, Mn, Fe and Cu were found in high concentration and harming the soil *Rhizobium sp*..
- The outcomes from the site under investigation where sewage and municipal wastewater is being utilized for about long period explicated the advancement of soil with organic carbon and nutrients status of the soil. We have used this water as a form of fertilizer due to the high level of N, P, K and high micro-nutrients.
- Hence, the potential use of sewage and municipal wastewater can successfully increase water resource for irrigation and may help with expanding agricultural production. But sometimes it gives an adverse effect on crop and beneficial microbes due to the high level of toxic heavy metals. When the sewage and municipal wastewater are used in agriculture, certain chemicals and virulent *E. coli.* may cause hazards to human health from exposure through different sources.
- So, the best possible management of sewage and municipal wastewater for irrigation and intermittent observing of soil fertility and quality parameters are needed to ensure prosperous, safe and long period reuse of wastewater water irrigation. Like this use of sewage water is mixed with groundwater or alternate way of irrigation.

Suggestion for future work:-

The finding of this study provides some suggestions depending on which further line of work can be taken up.

- Experiments may be done by the isolation of PGPR microorganism for bioremediation in polluted soil.
- The effect of soil pH and EC on PGPR microorganism.
- To study the correlation between heavy metals and PGPR microorganism.

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APPENDIX - A: Different Types of Culture Media used for growing Different types of microorganism.

a). Nutrient agar Media used for Growing total bacterial population

Composition **Ingredients**	**Gms/litre**
Peptone	10.000
Meat extract B[#]	10.000
Sodium chloride	5.000
Agar	12.000
pH after sterilization	7.3±0.1

*Formula adjusted, standardized to suit performance parameters

[#]Equivalent to Beef extract

b). Culture media for *E. coli*.

Composition **Ingredients**	**Gms/litre**
Peptones (meat and casein)	3.000
Pancreatic digest of gelatin	17.000
Lactose monohydrate	10.000
Bile salts	1.500
Sodium chloride	5.000
Crystal violet	0.001
Neutral red	0.030
Agar	13.500
pH after sterilization(at 25ºC)	7.2±0.2

*Formula adjusted, standardized to suit performance parameters

c). Culture Media for *Rhizobium sp.*

Composition **Ingredients**	**Gms/litre**
Yeast extract	1.000
Mannitol	10.000
Dipotassium phosphate	0.500
Magnesium sulphate	0.200
Sodium chloride	0.100
Congo red	0.025
Agar	20.000
Final pH (at 25°C)	6.8±0.2

*Formula adjusted, standardized to suit performance parameters

d). Culture Media *Pseudomonas species*

Composition **Ingredients**	**Gms/litre**
Protease peptone	20.000
Dipotassium hydrogen phosphate	1.500
Magnesium sulphate heptahydrate	1.500
Agar	20.000
Final pH (at 25°C)	7.2±0.2

*Formula adjusted, standardized to suit performance parameters

e). Culture Media for *Azotobacter species*

Composition **Ingredients**	**Gms/litre**
Sucros	20.000
Dipotassium phosphate	1.000
Magnesium sulphate	0.500
Sodium chloride	0.500
Ferrous sulphate	0.100
Sodium molybdate	0.005

Calcium carbonate	2.000
Agar	15.000

*Formula adjusted, standardized to suit performance parameters

APPENDIX – B. Weekly meteorological data during production period.

Wk. No.	Date	Max. Temp.(°C)	Min. Temp.(°C)	Rainfall (mm)	Rainy days	Relative Humidity (%) Max.	Min.
1.	**Sep 03-09**	29.5	24.9	239	6	94	87
2.	**Oct-16**	30.7	25	9.2	1	92	74
3.	**17-23**	33	25.5	2.4	0	88	63
4.	**24-30**	30.2	24.2	178.1	5	91	74
5.	**Oct 01-07**	32	24.3	1.8	0	90	64
6.	**Aug-14**	31.3	23.6	1.2	0	91	64
7.	**15-21**	30.9	21.8	51	3	92	64
8.	**22-28**	28.1	22.2	27.6	1	92	71
9.	**29-Apr**	31.4	22.2	0	0	92	51
10.	**Nov 05-11**	30.6	23	81.6	4	91	65
11.	**Dec-18**	29.6	15.5	0	0	90	38
12.	**19-25**	30.2	15.2	0	0	89	38
13.	**26-Feb**	29.7	16.3	0	0	90	43
14.	**Dec 03-09**	28	13.3	0	0	84	34
15.	**Oct-16**	29.5	15.3	0	0	91	48
16.	**17-23**	26.7	14.1	0.8	0	88	42
17.	**24-31**	26.1	11.9	0	0	81	35

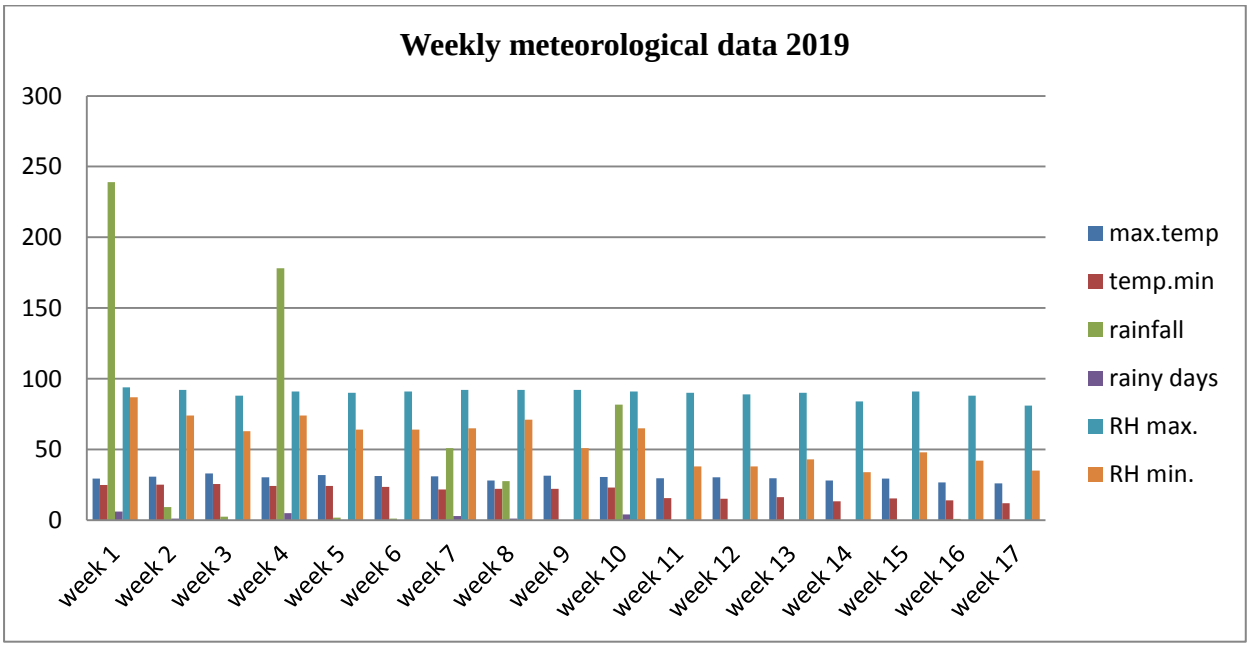

Fig. **- Weekly meteorological data 2019**

CURRICULUM VITAE

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