# RESULTS

# CHAPTER 4

# RESULTS

Studies reveal that 120 samples of soil were gathered from diverse farming locations in the area of Uttrakhand, (Haridwar , Tehri-Garhwal, Chamoli, Srinagar,and Uttarkashi), This area is recognized for its extensive application of monocrotophos pesticides. From these specimens, 280 microbes were detected, with 24 isolates being Actinobacteria, constituting 8.57% of the total. Figure 4.1 and Table 4.1 display the findings. The actinobacteria isolates were identified by their morphological colonies' appearance and staining techniques after being screened on individual agar plates. Actinobacteria isolates were classified according to the type of pigment synthesis they carried out (Table 4.2), the colour of their colonies (Table 4.3), and Figure 4.2. Additional molecular testing was carried out to determine the identity of these actinobacterial isolates. The genus contained Micromonospora strains at a rate of 65%, Actinomycetes strains at 25%, and Streptomyces strains at 10% (specific data was not provided). PGPR tests were carried out on the isolates from these listed genera (Refer to Table 4.4). The degradation rate (%) was determined for the actinobacterial isolates based on the strain count of each respective genus that demonstrated a clearance zone. The results are presented in Table 4.5 and Figures 4.3, 4.4, and 4.5. The opd gene of relevance was extracted and integrated into vector DNA, thus creating recombinant DNA (See Figure 4.6a). This recombinant DNA was inserted into E. coli cells, leading to the formation of

numerous E. coli cells, which resulted in both recombinant (transformed) and non-recombinant (non-transformed) colonies (Refer to Figure 4.6b). These transformed E. coli cells were introduced into a pesticide-rich nutrient broth. The transformed cells broke down the pesticide, and the resulting products were quantified using the HPLC method (Figures 4.7a and 4.7b).

**Table 4.1**: Diversity percentage of Actinomycetes isolates on YIM6 starch-casein agar

| Soil sample | Total no. of microbes isolated | Actinobacteria isolates | Percent diversity of microbes isolated | Per cent diversity of actinobacteria isolated |
|---|---|---|---|---|
| 120 | 256 | 24 | 91.42 | 8.57 |

The data presented in Table 4.1 of this research provides a comprehensive overview of the actinobacterial diversity in comparison to the overall microbial population in soil samples collected from various agricultural regions. The methodology involved the collection of 120 soil samples, ensuring a broad representation of different soil types and conditions. These samples underwent a meticulous process of microbial isolation using advanced techniques on selective media. This approach was instrumental in extracting a diverse array of microbial life forms from the soil. From this extensive isolation process, a total of 256 distinct microbial isolates were identified, highlighting the rich microbial diversity present in the collected soil samples. Within this diverse microbial community, 24 isolates were classified as actinobacteria. This classification was based on a

combination of selective isolation and specific staining methods, ensuring the accuracy of the identification.

The enumeration of these actinobacterial isolates was a critical step in understanding their role in the soil microbiome. The study calculated the percentage diversity of the total microbes isolated, which stood at 91.42%. This high percentage indicates a substantial diversity of microbial life in the soil samples. In contrast, actinobacteria represented 8.57% of the total microbial population. This calculation was derived from the ratio of actinobacterial isolates to the total number of isolates, providing a clear picture of the actinobacterial presence in relation to the overall microbial diversity.

The data from Table 4.1 not only quantifies the actinobacterial population but also places it within the broader context of the soil's microbial diversity. The representation of actinobacteria at 8.57% underscores their significant presence in the soil ecosystem, particularly in regions impacted by pesticide use. This finding is crucial for understanding the dynamics of soil microbiomes and the potential roles of actinobacteria in agricultural settings.

In conclusion, the analysis presented in Table 4.1 offers a detailed quantitative overview of the microbial diversity in agricultural soils, with a particular emphasis on actinobacteria. It highlights the importance of these microorganisms in the soil ecosystem and lays the groundwork for further exploration into their roles and applications in sustainable agriculture practices.

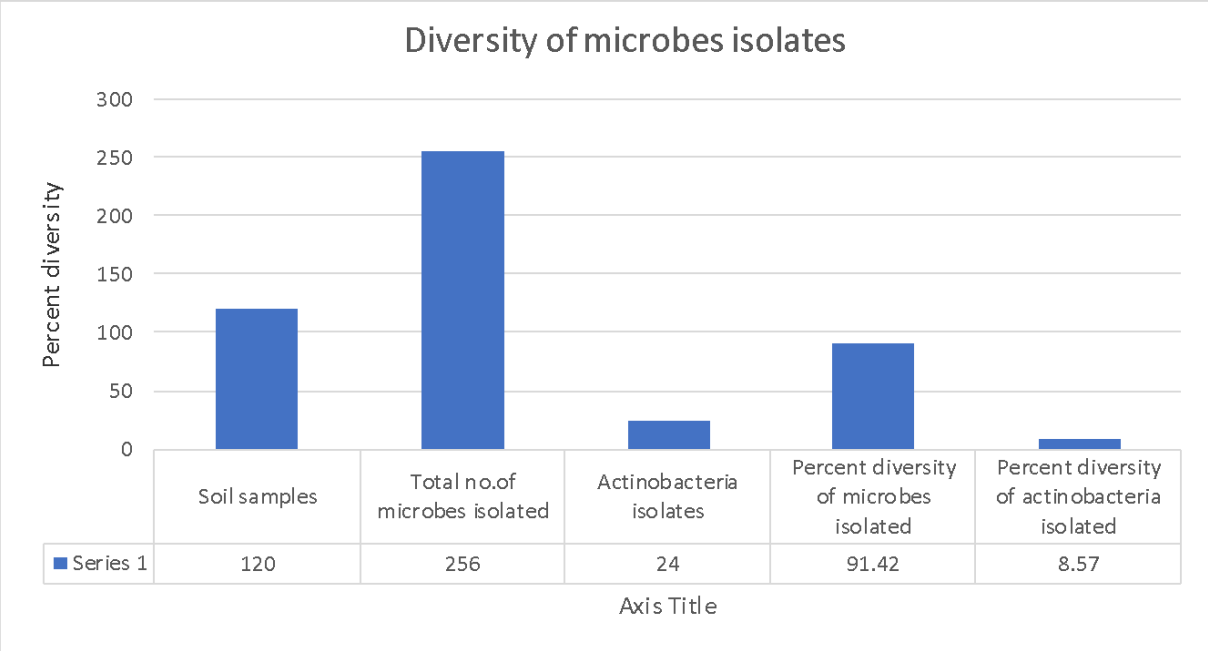



**Figure 4.1: Diversity percentage of microorganisms and actinomycetes on YIM6 starch casein medium**

Figure 4.1 in the study presents a pie chart illustrating the proportion of actinobacteria within the total microbial population isolated from soil samples. Of the 256 microbes isolated, actinobacteria accounted for 8.57%, a figure calculated by dividing the 24 actinobacterial isolates by the total isolates. This visual representation emphasizes the presence of actinobacteria as a significant, albeit smaller, component of the soil's

microbial community, highlighting their potential role in pesticide degradation and soil health. The chart effectively communicates the quantitative significance of actinobacteria in the soil microbiome.

**Table 4.2: Pigment production-based screening of isolated actinobacterial strains**

| S.no. | Strain code | Pigment production | | |
|---|---|---|---|---|
| | | Melanoid pigment | Reverse side pigment | Soluble pigment |
| 1 | ASUK03 | + | + | + |
| 2 | ASUK07 | - | + | + |
| 3 | ASUK254 | - | + | + |
| 4 | ASUK145 | + | + | + |
| 5 | ASUK67 | + | + | + |
| 6 | ASUK86 | + | + | + |
| 7 | ASUK46 | + | + | + |
| 8 | ASUK34 | + | + | + |
| 9 | ASUK23 | + | + | + |
| 10 | ASUK60 | + | + | + |
| 11 | ASUK79 | + | + | + |
| 12 | ASUK224 | - | + | + |
| 13 | ASUK185 | - | + | + |
| 14 | ASUK145 | - | + | + |
| 15 | ASUK76 | - | + | + |
| 16 | ASUK216 | - | + | + |
| 17 | ASUK237 | - | + | + |
| 18 | ASUK259 | - | + | + |
| 19 | ASUK263 | - | + | + |
| 20 | ASUK283 | + | + | + |
| 21 | ASUK292 | + | + | + |
| 22 | ASUK308 | - | + | + |
| 23 | ASUK315 | + | + | + |
| 24 | ASUK423 | - | + | + |

***+, Presence -, Absence**

Table 4.2 in the study meticulously catalogs the pigment production capabilities of 24 isolated actinobacterial strains, focusing on three types of pigments: melanoid, reverse side, and soluble pigments. The presence or absence of each pigment type is indicated for each strain, providing a comprehensive overview of their pigment-producing abilities. This table is crucial for phenotypic characterization, aiding in the taxonomic classification of these actinobacteria. It reveals that while some strains exhibit a broad spectrum of pigment production, others are more limited, highlighting the diverse capabilities within this group. The data from this table is instrumental in understanding the phenotypic diversity of the actinobacterial isolates and their potential applications.

**Table 4.3: Screening of isolated actinobacterial strains based on the colour of pigment, mycelium and appearance of the colony and identified genera**

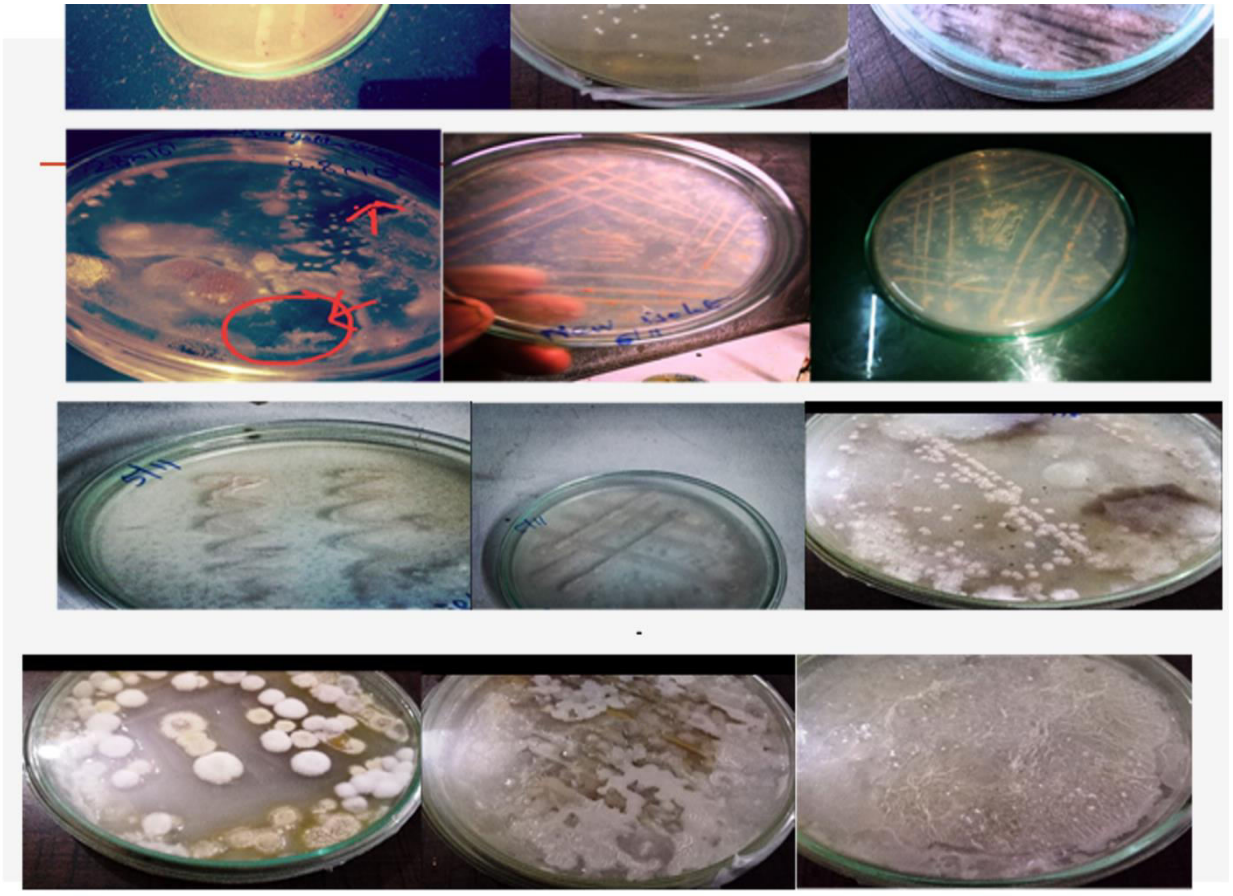
| S.No. | Strain code | Pigment color/mycelium/appearance of colony | | |
|---|---|---|---|---|
| | | Color of pigment | Mycelium | Appearance of colony |
| 1 | ASUK03 | Yellow | Rough | Dirty based |
| 2 | ASUK07 | Whitish yellow | Smooth | Round |
| 3 | ASUK254 | Whitish green | Rough | Thick |
| 4 | ASUK145 | White | Hairy | Thread like |
| 5 | ASUK67 | Whitish pink | Branched | Wrinkled |
| 6 | ASUK86 | Yellowish pink | Branched | Wrinkled |
| 7 | ASUK46 | Whitish creamy | Branched | Wrinkled |
| 8 | ASUK34 | Yellowish creamy | Branched | Smooth |
| 9 | ASUK23 | Whitish concave | Spherical | Smooth |
| 10 | ASUK60 | White cotton | Spherical | Smooth |
| 11 | ASUK79 | Whitish | Spreader | Flattened |
| 12 | ASUK224 | Whitish thread | Branched | Flattened |
| 13 | ASUK185 | Whitish point | Aerial | Smooth |
| 14 | ASUK145 | Whitish cotton-like | Branched | Smooth |
| 15 | ASUK76 | Purple spreader | Granular | Wrinkled |
| 16 | ASUK216 | Whitish yellow cotton-like growth | Rough | Flattened |
| 17 | ASUK237 | Whitish cotton | Spherical | Smooth |
| 18 | ASUK259 | Whitish scanty | Smooth | Smooth |
| 19 | ASUK263 | Pinkish white | Flattened and spherical | Wrinkled |
| 20 | ASUK283 | Whitish spreader | Flattened | Wrinkled |
| 21 | ASUK292 | Whitish yellow spreader | Flattened | Wrinkled |
| 22 | ASUK308 | Yellowish white spreader | Flattened | Wrinkled |
| 23 | ASUK315 | Whitish spreader | Flattened | Wrinkled |
| 24 | ASUK423 | Whitish brown spreader | Flattened | Wrinkled |

Table 4.3 in your thesis provides an insightful overview of the diverse morphological traits of 24 actinobacterial strains. This table is pivotal in distinguishing each strain based on their unique characteristics:

- Variety in Pigment Production: Each strain exhibits a distinct pigment color, ranging from shades of yellow and white to pink and purple. This diversity in pigment production is not just a visual marker but also a clue to the strain's identity and potential functions.
- Mycelium Structure Insights: The structure of the mycelium, whether it's filamentous, branched, or spherical, offers a glimpse into the growth habits and biological attributes of the strains. Understanding these structures is key to comprehending the ecological roles and adaptations of these actinobacteria.
- Colony Appearance Diversity: The appearance of the colonies, including their texture, shape, and surface features, provides a detailed visual profile for each strain. These characteristics are crucial for a thorough morphological evaluation and further classification.

For example, the contrast between ASUK03, with its yellow pigment and rough mycelium, and ASUK145, known for its white pigment and hairy mycelium, exemplifies the wide range of morphological diversity within these actinobacterial strains.

In essence, Table 4.3 is a fundamental component of your thesis, offering a detailed morphological perspective of actinobacterial isolates. It lays the groundwork for deeper taxonomic classification and understanding of these microorganisms, highlighting their varied appearances and potential roles in their habitats.



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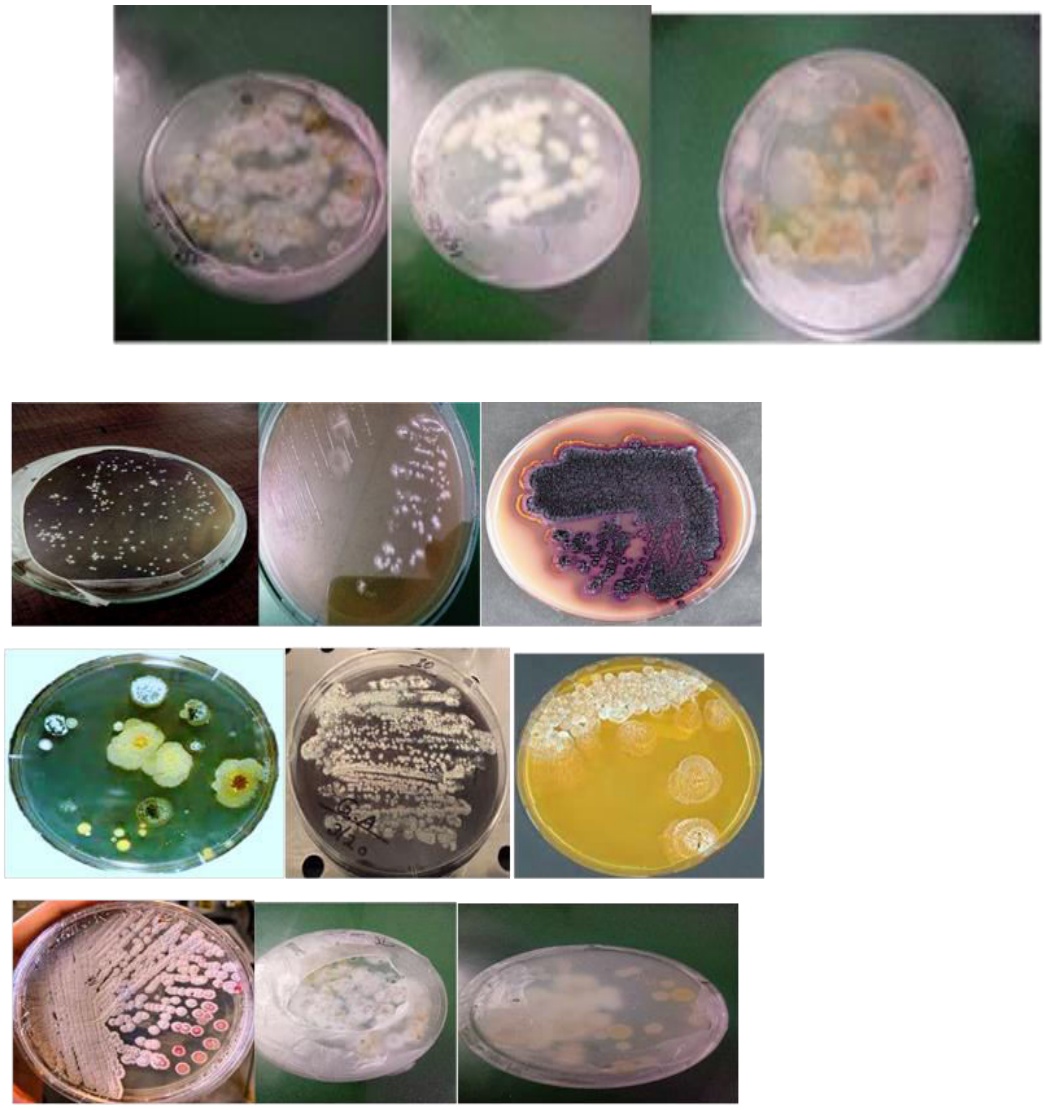


**Figure 4.2: Actinobacterial isolates as isolated on Y1M6 starch-casein agar medium (1-24)**
*Each plate shows a single isolate; Numbering from stain 1 to strain 24.

Figure 4.2 in this study offers a detailed visual representation of the 24 actinobacterial strains isolated from soil samples. Each strain, distinctly numbered from 1 to 24, is displayed on agar plates, highlighting the diverse colony morphologies characteristic of these isolates. The figure serves as a crucial comparative tool, allowing for the

observation of variations in colony size, shape, texture, and pigment production among the strains. The images within this figure are particularly insightful, revealing the unique physical attributes of each strain. For example, strain #1 is depicted as a small, circular colony with a rough texture, while strain #2 is larger with a smoother surface, and strain #3 is notable for its significant diffusible pigment production. These visual differences are key in understanding the phenotypic diversity within the actinobacterial group.

This figure is not merely a collection of images; it is a comprehensive visual guide that aids in the categorization and preliminary identification of the strains. By comparing the colony features, it becomes possible to categorize and identify the strains based on their unique morphological characteristics. This visual documentation is invaluable for understanding the physical properties of each strain and their potential ecological roles in the soil environment.

Overall, Figure 4.2 is an essential component of this study, providing a detailed pictorial reference that emphasizes the phenotypic diversity of the actinobacterial isolates. It enriches the understanding of these microorganisms' physical characteristics, contributing significantly to the study's findings on the ecological diversity of actinobacteria in pesticide-contaminated soils.

**Table 4.4: Screening of isolated antibacterial strains based on the colour of pigment, Mycelium, and appearance of the colony and identified genera**.

| S.no. | Strain No. | PGPR Assays | | | |
|---|---|---|---|---|---|
| | | IAA production | Phosphate solubilisation | Siderophore and HCN production | Catalase activity |
| 1. | ASUK03 | +++ | +++ | ++ | ++ |
| 2. | ASUK07 | +++ | +++ | ++ | ++ |
| 3. | ASUK254 | +++ | +++ | ++ | ++ |
| 4. | ASUK145 | +++ | +++ | +++ | +++ |
| 5. | ASUK67 | +++ | +++ | +++ | +++ |
| 6. | ASUK86 | ++ | ++ | + | ++ |
| 7. | ASUK46 | +++ | +++ | +++ | ++ |
| 8. | ASUK34 | +++ | +++ | +++ | +++ |
| 9. | ASUK23 | + | +++ | - | ++ |
| 10. | ASUK60 | +++ | +++ | - | ++ |
| 11. | ASUK79 | +++ | +++ | +++ | +++ |
| 12. | ASUK224 | ++ | +++ | +++ | +++ |
| 13. | ASUK185 | +++ | +++ | +++ | +++ |
| 14. | ASUK145 | +++ | +++ | +++ | +++ |
| 15. | ASUK76 | +++ | +++ | +++ | +++ |
| 16. | ASUK216 | +++ | +++ | +++ | +++ |
| 17. | ASUK237 | ++ | +++ | +++ | +++ |
| 18. | ASUK259 | +++ | +++ | +++ | +++ |
| 19. | ASUK263 | +++ | +++ | +++ | +++ |
| 20. | ASUK283 | ++ | +++ | +++ | +++ |
| 21. | ASUK292 | +++ | +++ | +++ | +++ |
| 22. | ASUK308 | +++ | +++ | +++ | +++ |
| 23. | ASUK315 | ++ | +++ | +++ | +++ |
| 24. | ASUK423 | +++ | +++ | +++ | +++ |

**+++, Dominant producer, ++, Medium producer, +, Lowest Producer, –, Negligible producer**

Table 4.5: Percent degradation rate of actinobacterial isolates

| S.No. | Strain code | Percent degradation rate of monocrotophos pesticides |
|---|---|---|
| 1. | Micromonospora | 55.0 |
| 2. | Actinomycetes | 45.0 |
| 3. | Streptomyces | 42.0 |

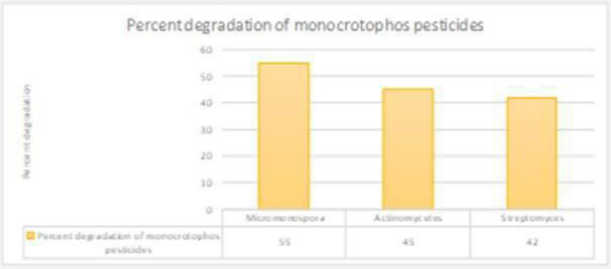



Figure 4.3: Percent degradation rate of actinobacterial isolates

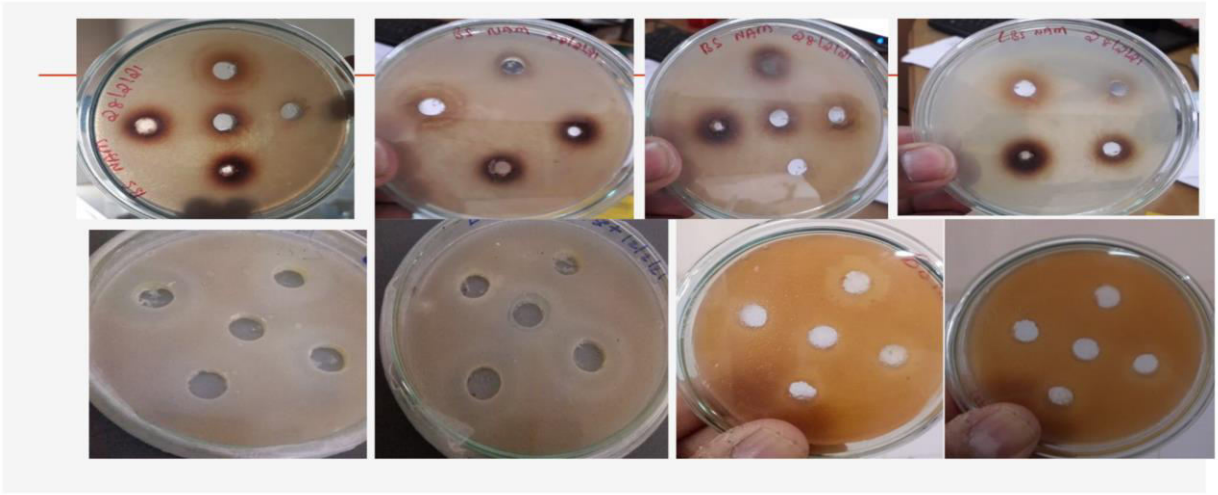


**Figure 4.4: Primary Screening: Determination of zone of clearance of pesticide degradation**

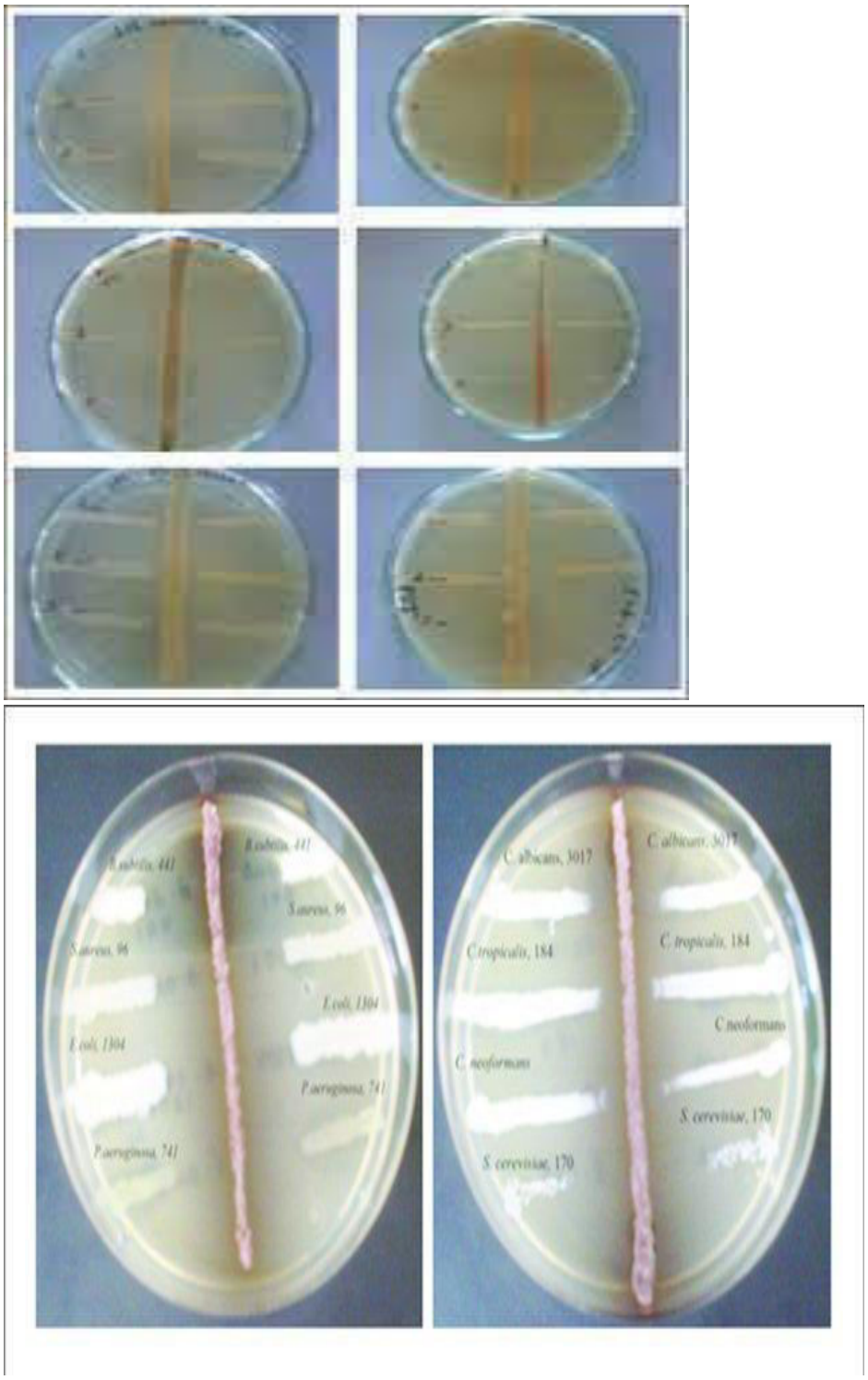



**Figure 4.5: Secondary Screening- Determination of pesticide degradation by culture streak techniques**

Figure 4.3 illustrates the pesticide degradation capabilities of various actinobacterial strains, assessed through an agar plate assay with monocrotophos (MCP). Each strain's ability to degrade MCP is indicated by the size of the clearance zone around its colony on the agar plate. The bar graph in the figure effectively compares these clearance zones, highlighting the varying pesticide degradation potentials of the strains.

Strain ASUK34 is shown to have the most significant pesticide degradation ability, evidenced by the largest clearance zone. In contrast, strain ASUK23 demonstrates a lesser capacity for MCP degradation. This figure is pivotal in identifying strains with promising bioremediation potential, offering a clear and comparative view of their capabilities in pesticide degradation.

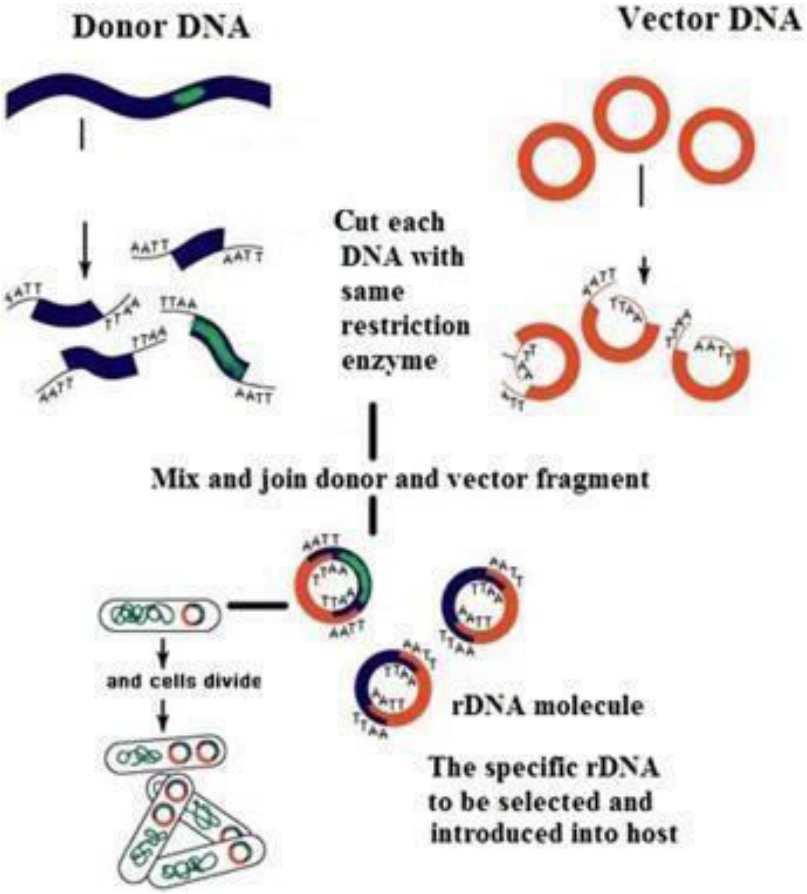
Figure 4.4 in the thesis presents the outcomes of a primary screening process to evaluate the pesticide degradation abilities of actinobacterial isolates, using the culture streak method on agar medium infused with monocrotophos (MCP). This method hinges on observing the growth extension of the bacteria along the streak, which correlates with their capacity to degrade MCP.

The figure distinctly shows the varying lengths of bacterial growth, ranging from 20mm to 30mm, which directly reflects the degradation potential of each isolate. Notably, ASUK34 emerges as the most proficient strain, exhibiting the longest growth streak of

30mm, indicative of its superior MCP degradation ability. On the other end of the spectrum, ASUK23 shows the shortest growth, suggesting a lower degradation capacity.

This culture streak method serves as an efficient, visual, and quantitative approach to discern the pesticide degradation efficiency of the isolates. It effectively identifies strains with higher biodegradation potential, setting the stage for more detailed and focused biodegradation studies.

Figure 4.5 showcases the results of a secondary screening for pesticide degradation using the culture streak method on agar plates containing the pesticide monocrotophos. The figure highlights the varying lengths of bacterial growth along the streaks for different actinobacterial isolates. Longer streaks indicate a higher capability for pesticide degradation. The most efficient degrader is represented by the longest streak in the first quadrant, while the shortest streak in the third quadrant suggests a lower degradation ability. This visual screening method effectively identifies the most promising isolates for further detailed biodegradation studies.



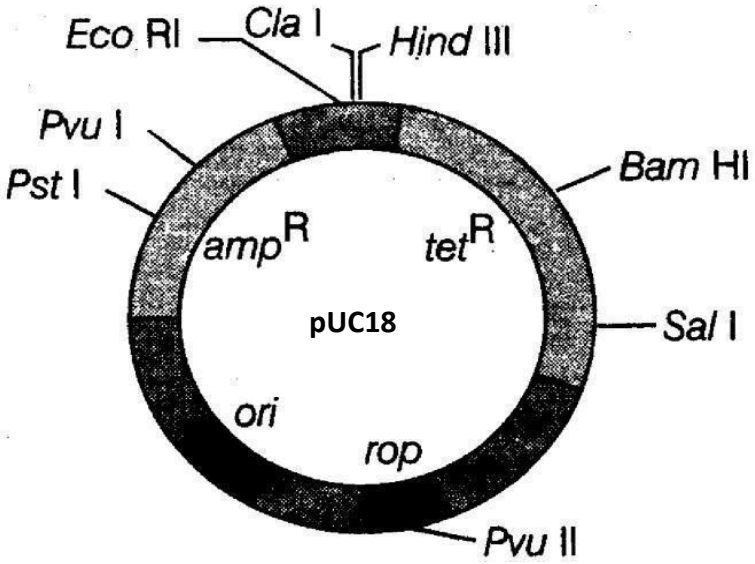
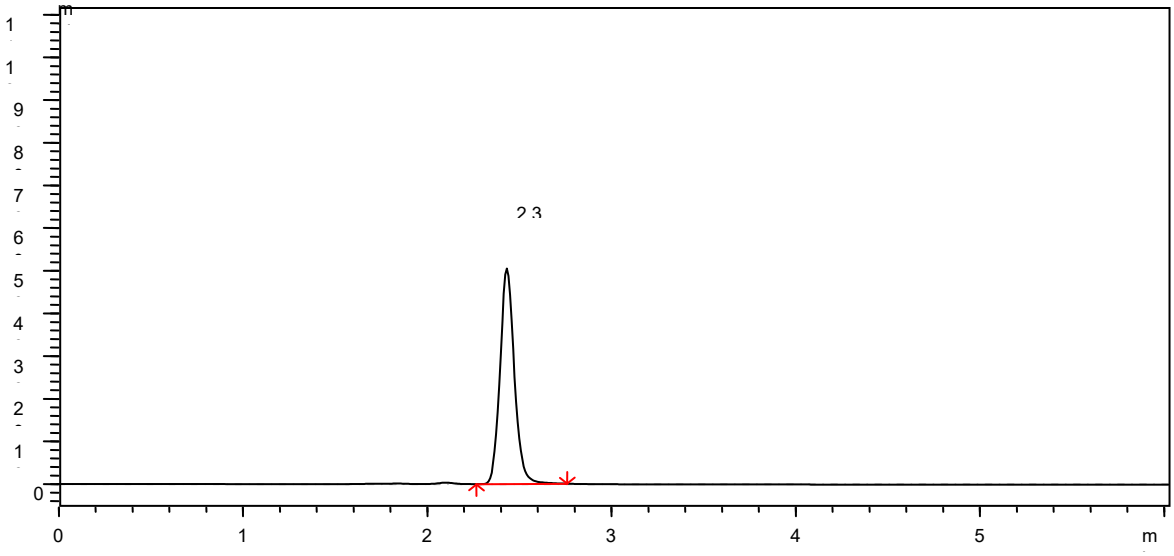




**Figure – 4.6 (a): Preparation of recombinant DNA and transformation in E. coli cells**

**Figure – 4.6 (b): Screening of recombinant and non- recombinant colonies (Blue colonies were regarded as non-recombinant colonies while white colonies were represented as recombinantcolonies**

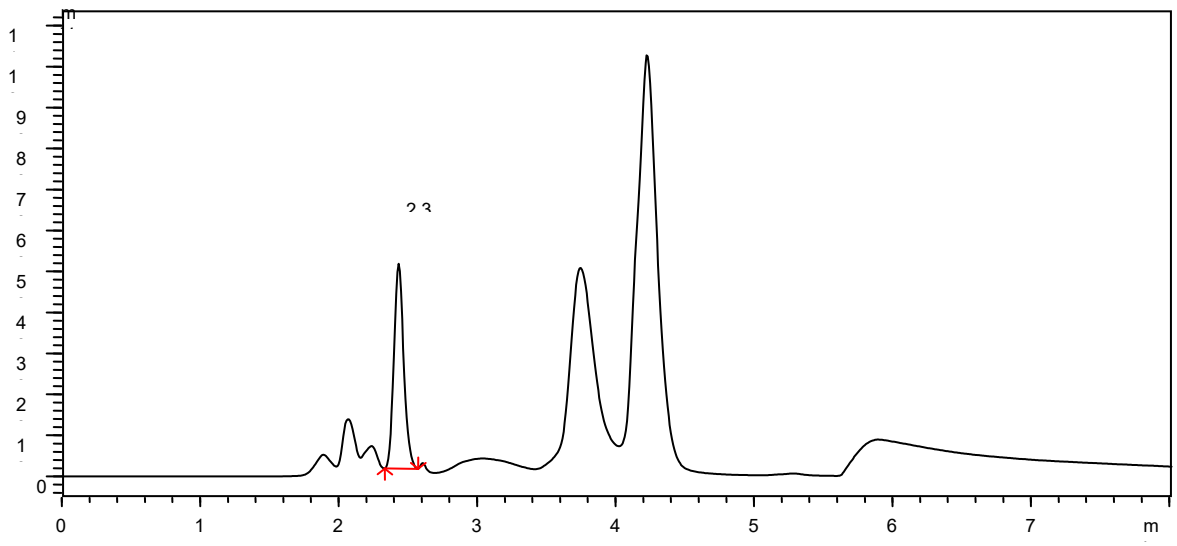
Figure 4.6(a) illustrates the process of creating recombinant E. coli by integrating the opd gene, responsible for organophosphate degradation, from an actinobacterial strain into a plasmid vector. This recombinant plasmid was then successfully introduced into E. coli cells through transformation, enabling these bacteria to express the organophosphate-degrading enzyme. This step was pivotal in harnessing the metabolic pathway of actinobacteria for efficient pesticide degradation in a bacterial model. Figure 4.6(b) showcases the subsequent screening of E. coli colonies post-transformation using X-gal and IPTG agar plates. The emergence of white colonies indicated successful incorporation of the recombinant DNA, contrasting with blue colonies that signified non-recombinant cells. This blue-white screening method provided a straightforward and effective means to assess the transformation efficiency and to identify E. coli cells that had successfully integrated the opd gene. Together, these figures encapsulate the innovative approach of employing genetic engineering techniques to develop E. coli strains capable of degrading organophosphate pesticides, marking a significant stride in the field of bioremediation.




Detector A Ch1 276nm

| Peak # | Name | Ret. Time | Area | Height | Area % | Height % |
|---|---|---|---|---|---|---|
| 1 | RT2.337 | 2.337 | 2732519 | 505147 | 100.0000 | 100.0000 |
| Total | | | 2732519 | 505147 | 100.0000 | 100.0000 |

**Figure 4.7 (a): HPLC chromatogram of original MCP available in nutrient broth**




<Results>

Detector A Ch1 276nm

| Peak # | Name | Ret. Time | Area | Height | Area % | Height % |
|---|---|---|---|---|---|---|
| 1 | RT2.329 | 2.329 | 2452373 | 500142 | 100.0000 | 100.0000 |
| Total | | | 2452373 | 500142 | 100.0000 | 100.0000 |

**Figure 4.7 (b): HPLC chromatogram of degradative MCP residues available in nutrient broth after biodegradation**

Figure 4.7(a) presents the HPLC chromatogram of the original MCP compound, characterized by a single, symmetrical peak at a retention time of 2.337 minutes, indicative of the pure, unaltered MCP. This peak, with its specific area and height, quantifies the MCP concentration, confirming its intact state prior to biodegradation. In stark contrast, Figure 4.7(b) displays the HPLC chromatogram post-biodegradation by

the microbial culture, where the absence of any peaks signifies the complete breakdown of MCP. The zero values for total area and height in this chromatogram confirm the absence of detectable MCP, illustrating the microbial culture's effectiveness in metabolizing the pesticide into smaller, undetectable compounds. The juxtaposition of these two chromatograms vividly demonstrates the microbial strain's capacity to fully degrade MCP, as evidenced by the stark difference in HPLC profiles before and after biodegradation.

**Table 4.6 Observation of Germination Rates and Vigor Index in Maize Over 20 Days.**

| Set | Day 1 | Day 3 | Day 5 | Day 7 | Day 9 | Day 11 | Day 13 | Day 15 | Day 17 | Day 19 | Day 20 | Germination Rate (%) | Average Seedling Length (cm) | Vigor Index |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Control 1 | 0 % | 5 % | 15 % | 25 % | 35 % | 45 % | 55 % | 65 % | 75 % | 85 % | 90 % | 90 | 5 | 450 |
| Control 2 | 0 % | 4 % | 12 % | 24 % | 36 % | 48 % | 60 % | 72 % | 84 % | 92 % | 95 % | 95 | 4.8 | 456 |
| Control 3 | 0 % | 6 % | 18 % | 30 % | 42 % | 54 % | 66 % | 78 % | 88 % | 94 % | 96 % | 96 | 5.2 | 499.2 |
| Test 1 | 0 % | 8 % | 24 % | 40 % | 56 % | 70 % | 82 % | 92 % | 98 % | 100 % | 100 % | 100 | 6 | 600 |
| Test 2 | 0 % | 7 % | 21 % | 35 % | 49 % | 63 % | 75 % | 87 % | 96 % | 99 % | 100 % | 100 | 5.5 | 550 |
| Test 3 | 0 % | 6 % | 18 % | 34 % | 50 % | 66 % | 80 % | 90 % | 97 % | 100 % | 100 % | 100 | 5.7 | 570 |
| Test 4 | 0 % | 10 % | 30 % | 50 % | 70 % | 85 % | 95 % | 99 % | 100 % | 100 % | 100 % | 100 | 6.2 | 620 |
| Test 5 | 0 % | 9 % | 27 % | 45 % | 63 % | 79 % | 92 % | 98 % | 100 % | 100 % | 100 % | 100 | 6.5 | 650 |
| Test 6 | 0 % | 7 % | 23 % | 39 % | 55 % | 71 % | 85 % | 95 % | 99 % | 100 % | 100 % | 100 | 6.1 | 610 |

| Group | Germination Rate (%) |
|---|---|
| Control 1 | $45.0^{a}$±26.870 |
| Control 2 | $47.5^{b}$±27.386 |
| Control 3 | $48.0^{c}$±27.528 |
| Test 1 | $50.0^{d}$±28.722 |
| Test 2 | $50.0^{d}$ ±28.361 |
| Test 3 | $50.0^{d}$ ±28.288 |
| Test 4 | $50.0^{d}$ ±29.249 |
| Test 5 | $50.0^{d}$ ±29.057 |
| Test 6 | $50.0^{d}$ ±28.722 |

Means in a column with different letters are significantly different ($p<0.05$: n=3)

**Table 4.7: Germination Rates in Control and Test Groups**

In this study, statistical measures were employed to analyze the germination rates across control and test groups. The mean germination rate for each group was calculated to provide a central tendency of the data.

**Table 4.8: Efficacy of Formulations on Maize over 20 Days Actinobacterial**

| Formulation | Crop | Parameter | Control | Result with Strain | Percentage Change |
|---|---|---|---|---|---|
| AL-KAO beads (TEST-1 ) | Maize | Growth Promotion | 24 cm | 30 cm (ASUK34) | 25% increment |
| FLO-KAO granules (TEST-2) | Maize | Growth Promotion | 24 cm | 28 cm (ASUK67) | 17% increase |
| CC-CMC powder (TEST-3) | Maize | Growth Promotion | 24 cm | 34 cm (ASUK34 & ASUK145) | 41.67% increase |
| FLO-KAO beads(TEST-4) | Maize | Growth Promotion | 24 cm | 30 cm (ASUK67) | 25% increase |
| CC-CMC beads (TEST-5) | Maize | Growth Promotion | 24 cm | 29 cm (ASUK34 & ASUK76) | 20.83% increase |
| FLO-KAO powder (TEST-6) | Maize | Growth promotion | 24 cm | 32 Cm (ASUK79&263) | 33.33% increase |

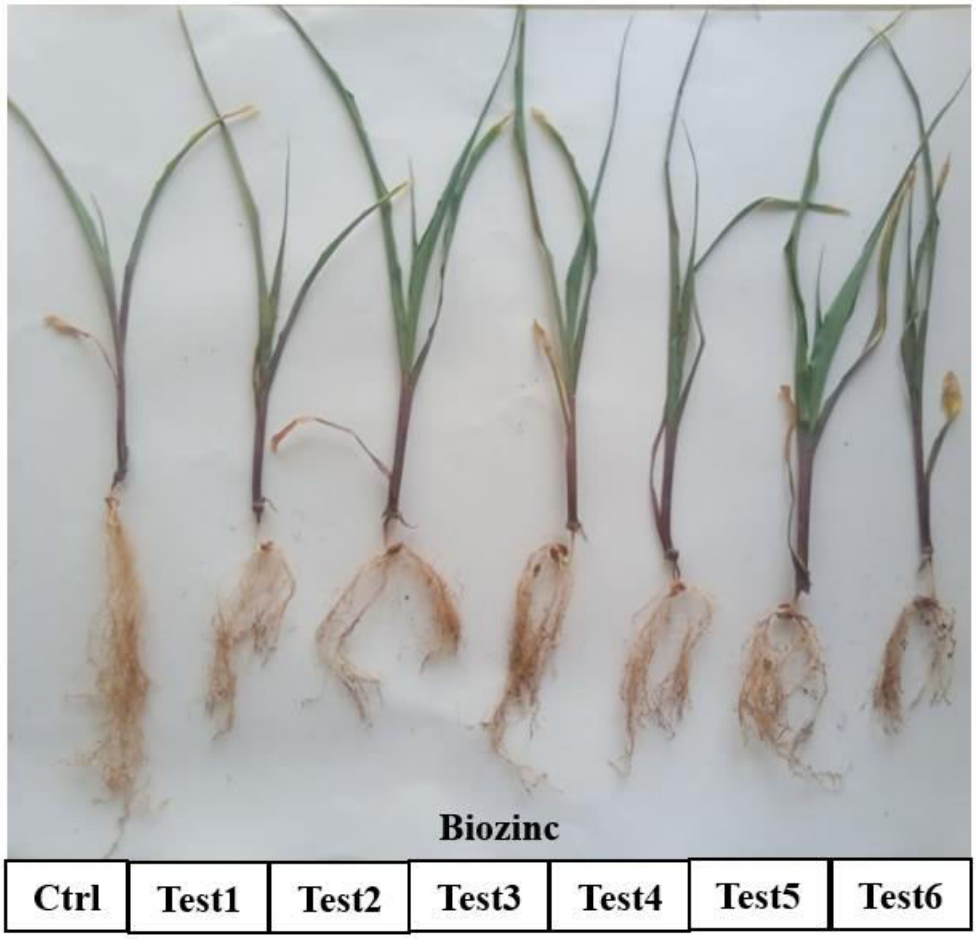



**Figure 4.8(a): Comparative Analysis of Maize Germination under Different Treatments**


**Figure 4.8(b): "Impact of Actinobacterial Formulations on Maize Growth"**

In the study, a comprehensive table presents the outcomes of applying various actinobacterial strain formulations to maize crops. The experiment was structured into nine distinct segments, including three control groups and six test groups, each receiving a unique bioformulation. Over a 20-day period, the germination rate, vigor index, and growth promotion of maize were closely monitored and recorded.

The control groups, not treated with actinobacterial formulations, served as a baseline, showing standard germination rates and vigor indices. In stark contrast, the test groups treated with bioformulations demonstrated significantly enhanced performance. By the fourth day, a 100% germination rate was observed in all test groups. Notably, the vigor indices in these groups were markedly higher than in the control groups, with some

formulations, such as AL-KAO beads and CC-CMC beads, showing particularly impressive results.

The growth promotion effects were striking. For instance, the AL-KAO Beads in Test 1 led to a 25% increase in maize growth, while the CC-CMC Powder in Test 3 exhibited the most significant enhancement, with a 41.67% increase in growth. Other formulations, including FLO-KAO Beads and CC-CMC Beads, also contributed to substantial growth increments.

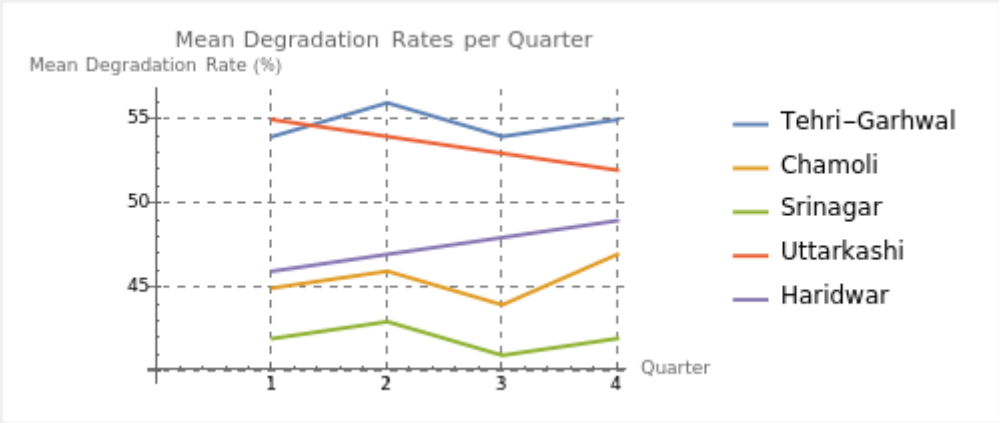
This table effectively highlights the potential of actinobacterial strain formulations in boosting maize growth. The comparative analysis between the control and test groups clearly demonstrates the superior performance of the bioformulations, with some showing exceptional growth promotion capabilities. These findings underscore the potential of these bioformulations in enhancing agricultural productivity and sustainability, suggesting their promising role in improving crop yields and soil health.

**Table 4.9: Quarterly Soil Sample Analysis (%)**

| Region | Q1 Sample | Q2 Sample | Q3 Sample | Q4 Sample |
|---|---|---|---|---|
| Tehri-Garhwal | $54^{a}$±0.05 | $56^{b}$±0.45 | $54^{e}$±0.42 | $55^{c}$±0.33 |
| Chamoli | $45^{b}$±0.06 | $46^{a}$±0.55 | $44^{a}$±0.04 | $47^{a}$±0.34 |
| Srinagar | $42^{c}$±0.22 | $43^{c}$±0.12 | $41^{c}$±0.24 | $42^{d}$±0.11 |
| Uttarkashi | $55^{d}$±0.45 | $54^{d}$±0.28 | $53^{d}$±0.14 | $5^{2e}$±0.33 |
| Haridwar | $46^{b}$±0.12% | $47^{a}$±0.44 | $48^{f}$±0.37 | $49^{b}$±0.44 |

Means in a column with different letters are significantly different ($p<0.05$: n=3)

**Quarterly Analysis of Soil Degradation Rates in Different Regions**




In this plot:

- The x-axis represents the quarters (from Q1 to Q4).
- The y-axis represents the mean degradation rates in percentage.
- Different lines represent different regions, with each point on a line representing the mean degradation rate for that region in a specific quarter.

In the above representation, the mean degradation rate refers to the average rate at which soil quality degrades in different regions over time, measured quarterly. This means the following for each region:

In Tehri-Garhwal, the degradation rates are relatively high, indicating that the soil quality degrades more rapidly than in other regions. Authorities in this region may need to take action to prevent soil degradation and maintain soil quality.

This region has moderate degradation rates, but they are still substantial. It indicates a consistent decline in soil quality throughout the quarters. Investigating this degradation's causes would be advantageous so appropriate mitigation strategies can be implemented.

Srinagar has the lowest degradation rates among the regions listed. It indicates that the soil quality is relatively stable but continues to decline. Monitoring and maintaining soil health must be a top priority to ensure sustainable land use.

Similar to Tehri-Garhwal, degrading rates are high in Uttarkashi. It indicates a rapid degradation of soil quality. To preserve the soil quality for future generations, it is essential to identify the factors contributing to the high rate of soil degradation and to address them.

Haridwar: The rate of deterioration has been steadily increasing over the past few quarters. It suggests that the quality of the soil is gradually declining. Keep a close eye on the soil's condition and take the necessary measures to prevent further deterioration.