Studies on sulfur oxidizing bacteria as potential biofertilizer

Thesis

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In

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6. ENGLISH SUMMARY

This dissertation explored the potential of sulfur-oxidizing bacteria (SOB) as promising biofertilizers to enhance onion growth, nutrient uptake, and yield under sustainable agricultural practices. The study addressed a critical challenge in agriculture: the low availability of sulfur and the heavy reliance on chemical fertilizers, which are costly and environmentally harmful. By isolating, characterizing, and applying SOB strains, this work provides compelling evidence for their dual role in sulfur oxidation and plant growth promotion. This investigation was focused on two different parts including:

Part I: (Isolation and definition SOB).

1. Twenty-three bacterial isolates were recovered from onion rhizosphere soils collected across Assiut Governorate. Their sulfur-oxidizing capacity was confirmed by a visible color shift in thiosulfate medium with bromocresol purple indicator, where 23 isolates induced medium acidification. This provided direct evidence of their metabolic activity in converting thiosulfate to sulfate.

2. Scanning electron microscopy revealed the adhesion of rod-shaped bacterial cells to sulfur granules in thiosulfate-enriched cultures, confirming their active role in sulfur metabolism compared to controls where such adhesion was absent.

3. A comprehensive evaluation of pH, sulfate concentration, optical density, and electrical conductivity highlighted substantial variation among isolates. Notably, **SOB12** demonstrated consistent sulfur-oxidizing activity across different nutritional settings whereas, **SOB15** recorded the highest sulfate accumulation (696.7 mg/L in one week under non-glucose conditions) and robust growth. These quantitative assays established SOB12 and SOB15 as the most efficient performers.

4. SOB12 was identified as *Bacillus spizizenii* and SOB15 as *Priestia aryabhattai* through 16S rRNA gene sequencing and phylogenetic analysis, supported by strong bootstrap values. Their morphological traits (Gram-positive, endospore-forming rods with distinct colony features) reinforced the molecular findings, ensuring reliable taxonomic placement.

5. Beyond sulfur oxidation, both isolates exhibited the ability to produce key phytohormones. SOB12 produced the highest levels of indole-3-acetic acid (141.9 µg/mL) and gibberellic acid (120.4 µg/mL), while SOB15 showed strong consistency across traits, thereby linking microbial metabolism directly to plant growth enhancement.

Part II: Application of SOB (pot experiment).

1. The biofertilizer potential of SOB12 and SOB15 was validated under greenhouse conditions with onion cultivar Giza 6. Plants were grown with graded sulfur levels (50%, 100%, 150%) either alone or combined with bacterial inoculation.

2. Sulfur application alone improved onion growth, but the greatest benefits were observed when sulfur was combined with bacterial inoculants. Dual inoculation with SOB12 and SOB15 under 50% sulfur doubled bulb dry weight compared to sulfur alone, while at 150% sulfur + SOB12 achieved the maximum fresh (183.79 g/plant) and dry bulb weight (22.49 g/plant) recorded in the study. These results highlight the synergistic effect between microbial inoculation and sulfur availability.

3. SOB15 consistently promoted shoot elongation, producing the tallest plants (87 cm under 50% sulfur, 82.6 cm under 150% sulfur). Bulb length and diameter were also enhanced, with co-inoculated treatments outperforming sulfur-only controls.

4. SOB inoculation enhanced soil acidification, lowering pH more effectively than sulfur alone, thereby improving nutrient availability in alkaline soils. It also boosted enzyme activities, particularly alkaline phosphatase and nitrogenase, demonstrating a broader impact on soil biological function

5. Inoculated plants showed stronger colonization by arbuscular mycorrhizal fungi, particularly with SOB15, suggesting that these bacteria support beneficial plant fungus interactions, further contributing to soil health and nutrient cycling.

6. SOB12 was particularly effective in increasing chlorophyll a, carotenoids, and total pigments, thereby boosting photosynthetic efficiency and biomass accumulation.

7. Inoculation significantly increased nitrogen, phosphorus, potassium, and sulfur uptake. The most pronounced effect was nitrogen accumulation with SOB15 under 150% sulfur (0.242 g/plant), representing more than a fourfold increase over the uninoculated control.

8. Treatments with SOB inoculation enriched onion bulbs with proteins, amino acids, carbohydrates, phenolics, and pyruvic acid. Notably, SOB15 with 50% sulfur yielded the highest amino acid concentration (486.17 mg/g DW), while dual inoculation with 100% sulfur maximized sulfur assimilation (0.167%). These findings confirm that SOB inoculation not only enhances yield but also improves bulb nutritional quality.

Conclusion and recommendations:

Taken together, this dissertation provides compelling evidence that *Bacillus spizizenii* (SOB12) and *Priestia aryabhattai* (SOB15) are multifunctional biofertilizer candidates. Their ability to oxidize sulfur, produce phytohormones, improve nutrient uptake, enhance photosynthesis, stimulate mycorrhizal colonization, and enrich bulb biochemical content underscores their broad agronomic potential.

The best overall results were observed in treatments combining SOB12 and SOB15 individually or in combination with 50% of the recommended sulfur dose. These treatments significantly enhanced plant height, leaf number, bulb weight, photosynthetic pigments, nutrient uptake (N, P, K, S), and biochemical constituents such as carbohydrates, amino acids, phenolics, and pyruvic acid. Notably, at 50% sulfur, co-inoculation (SOB12+SOB15) significantly increased bulb dry weight by 127% relative to 50% sulfur without inoculation; at 100% sulfur, SOB15 increased nitrogen uptake by 45% and bulb fresh weight by 65% versus sulfur alone; and at 150% sulfur, SOB15 raised total carbohydrate concentration by 146% compared with sulfur alone.

It is recommended that SOB12 and SOB15 be applied in onion cultivation, either individually or in combination with moderate sulfur doses, to maximize productivity while reducing chemical fertilizer dependence. Beyond onion, these isolates hold promise for broader application in crops cultivated on alkaline or sulfur-deficient soils. Future research should focus on scaling up field trials, developing commercial inoculum formulations, and integrating these microbial solutions into precision nutrient management strategies for sustainable agriculture.

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