



Isolation And Identification of Beneficiary Microorganism Study with Fungi

Alok Kumar Mishra, Faculty, Department of Botany, Govt. College, Mehgaon, Bhind (M.P.)

Abstract

Through symbiotic and asymbiotic interactions, beneficial microorganisms promote plant growth and development directly or indirectly according to their unique abilities. These microorganisms produce plant growth regulators, including ACC-deaminase and siderophore, those that release organic acids, N₂-fixing bacteria and phosphate solubilizing bacteria, and those that increase acquisition of nitrogen and phosphorus under stressed and nonstressed conditions. Beneficial microorganisms can also affect the physiological and molecular events of plants. In this study, we discuss how beneficial microorganisms contribute to salt stress tolerance through physiological and molecular events in plants and how those mechanisms work.

Keywords: symbiotic and asymbiotic interactions, beneficial microorganisms

Introduction

Beneficial microorganisms include bacteria, fungus, and yeast that can be isolated from fruits or soil, and these exert an antagonist action against different pathogens. Besides soil, other environments where antagonist microorganisms can be isolated exists, such as the ocean (Hernández Montiel et al., 2018). The action mode against fungus species is attributed to different mechanisms such as antibiosis, parasitism, colonization sites competence and nutrients, lithic enzymes, and induction resistance (Hernández Montiel et al., 2018).

On blueberry fruits, bacteria have been used to its postharvest control mentioned by Kurniawan, Wilson, Mohamed, and Avis (2018). These authors isolated *Bacillus* and *Pseudomonas* to inhibit *Botrytis cinerea* and *Alternaria alternata*, showing that bacteria produce peptide origin compounds with antimicrobial properties as part of its action mechanism, inhibiting its *in vitro* growth with values of 42 and 27% for *Botrytis cinerea* and *Alternaria alternata*, respectively.

Trichoderma is considered the most important antagonist specie to control the postharvest diseases of blueberries due to its capacity to inhibit diseases in fruits caused by a fungus (Romero-Arenas et al., 2017). Furthermore, yeasts such as *Debaryomyces hansenii*, *Meyerozyma caribbica*, *Rhodotorula minuta*, among others have also been studied since they do not produce antibiotics, are not toxic, and do not pollute the environment, being its main action mechanisms the antibiosis as well the production of volatile compounds and the resistance induction (Hernández Montiel et al., 2018).

Materials and Methods

Soil Sampling

To isolate potent bacteria, surface soil samples (0–15 cm) were collected from wastewater irrigated agricultural soils of major industrial area. Five random sub-samples collection with the help of a wooden core borer from each sampling site within a radius of 500. The soil samples keep on, ice and store at Gravel and stones remove from the soils and samples. The physicochemical analysis of soil samples was carried

Isolation of Bacteria

Bacteria isolation from soil using a serial dilution method. Briefly, 10 g of soil was added to 90 mL of 1% normal saline solution in a 250 mL flask. The flask was then shaken for 20 min on a rotary shaker. One mL of the suspension was taken out of the flask and added to 9 mL of normal saline solution, and serial dilutions were made up to 10⁻⁹ dilution. One mL of each dilution was spread on the nutrient agar and MacConkey agar media plates and were incubated for up to 3 days at 28 ± 2 °C to observe bacterial growth [20]. Bacterial colonies were studied for their colony morphology such as size, shape, margins, elevations, texture and opacity. Single colonies were re-streaked on to fresh LB agar plates and incubated under similar conditions. The process was repeated three times to purify the colonies. Bacterial colonies were preserved at -20 °C in 50% sterile glycerol solution for future use.



Minimum inhibitory concentration (MIC) of above-mentioned heavy metals for bacterial isolates with at least one tested plant growth promoting trait was determined with the plate dilution method [28]. Various concentrations of heavy metals ranging from 0.1 in LB agar inoculated with 18 h old bacterial culture. Inoc incubated at 28 ± 2 °C for 48 h.

Isolation of Microorganisms

Microorganisms occur in natural environment like soil. They are mixed with several other forms of life. Many microbes are pathogenic. They cause a number of diseases with a variety of symptoms, depending on how they interact with the patient. The isolation and growth of suspected microbe in pure culture is essential for the identification and control the infectious agent. The primary culture from natural source will normally be a mixed culture containing microbes of different kinds. But in laboratory, the various species may be isolated from one another. A culture which contains just one species of microorganism is called a pure culture. The process of obtaining a pure culture by separating one species of microbe from a mixture of other species, is known as isolation of the organisms.

Observation and result

Composting is an effective way to introduce beneficial microbes to the soil. By composting organic matter, such as plant residues and animal manure, farmers can create a rich source of nutrients and beneficial microbes. Compost can also help reduce soil erosion and runoff risk, promoting environmental sustainability. Cover cropping is another effective way to promote beneficial microbes on farms. Farmers can improve soil structure, reduce erosion, and promote nutrient cycling by planting cover crops like legumes or grasses. Cover crops can also help to suppress weeds and reduce the need for synthetic herbicides. When cover crops are terminated and left on the soil surface, they can provide a source of organic matter and beneficial microbes for the soil. Crop rotations can also help to promote beneficial microbes on farms. Farmers can reduce the risk of soil-borne diseases by rotating crops and improving soil structure. Different crops have different nutrient requirements and root exudates, which can promote the growth of different beneficial microbes. Farmers can promote a diverse range of beneficial microbes by rotating crops and reducing the risk of nutrient depletion and disease.

Some specific examples of beneficial microbes for the farm include the nitrogen-fixing bacteria *Rhizobium*, which can improve the productivity of legume crops, and the mycorrhizal fungi *Glomus intraradices*, which can improve nutrient uptake in many crop species.

In order to promote beneficial microbes on their farms, farmers can take a variety of steps. They can implement composting programs, plant cover crops, and implement crop rotations. Farmers can also use biofertilizer, microbial inoculants or microbial amendments to introduce beneficial microbes to the soil. By promoting beneficial microbes, farmers can improve soil health, promote healthy plants and animals, and promote environmental sustainability on their farms.

The bacteria of the genera *Rhizobium*, *Bradyrhizobium*, *Mesorhizobium*, *Sinorhizobium* and *Azorhizobium* collectively known as rhizobia, in symbiotic association with leguminous plants reduce atmospheric nitrogen. The rhizobial colonies appear raised, wet, shining, translucent or opaque with smooth margin on yeast extract mannitol agar (YEMA) medium. The legume-rhizobia symbiosis culminates in the formation nitrogen fixing root or stem nodules.

Conclusions

The beneficial microorganisms play many significant roles in many fields, especially in medicine, agriculture, and industry. They belong to groups like archaea, bacteria, actinomycetes, and fungi. They are not only involved in plant growth and development but also improve plant health by alleviating abiotic (acidic, alkaline, salinity, drought, temperatures, and pressure) as well as biotic stress (attack by pathogens). These functions are performed by several mechanisms such as N₂ fixation, mineral solubilization like potassium,

phosphorus, and zinc, production of antagonistic compounds, siderophores, PGPRs like auxin and gibberellins. These microbes are also applicable as biofertilizers and biopesticides. So this article provides detailed information about the different functions and strategies used by plant PGPMs under difficult or stressful conditions to cure plant health and their growth and development. There is a need for further studies on the molecular pattern of plant microbe interaction and genetic expression of genes involved in this mechanism. New techniques like nanotechnology may be helpful in the field of biofertilizer formulation.

REFERENCE

Anwar, S.; Nawaz, M.F.; Gul, S.; Rizwan, M.; Ali, S.; Kareem, A. Uptake and distribution of minerals and heavy metals in commonly grown leafy vegetable species irrigated with sewage water. *Environ. Monit. Assess.* 2016, 188, 541.

Ayangbenro, A.S.; Babalola, O.O. A new strategy for heavy metal polluted environments: A review of microbial biosorbents. *Int. J. Environ. Res. Public Health* 2017, 14, 94.

Agarwal, A. and Tripathi, H.S. 1999. Biological and chemical control of botrytis gray mold of chickpea. *Journal of Mycology and Plant Pathology*, 29: 52–56.

Ahmed, M. and Kibret, M. 2014. Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. *Journal of King Saud University*, 26: 1–20. Ahmad, J.S. and Baker, R. 1988. Rhizosphere competence of *Trichoderma harzianum*. *Phytopathology*, 77:182–189.

Akhtar, M.S. and Siddiqui, Z. A. 2010. Effect of AM fungi on plant growth and root rot diseases of chickpea. *American-Eurasian Journal of Agriculture and Environmental Science*, 8:544–549.

Alström, S. 1991. Induction of disease resistance in common bean susceptible to halo blight bacterial pathogen after seed bacterization with rhizosphere pseudomonads. *Journal of General and Applied Microbiology*, 37:495–501.

Altschul, S.F., Gish, W., Miller, W., Myers, E.W., and Lipman, D.J. 1990. Basic local alignment search tool. *Journal of Molecular Biology*, 215:403–410

Anderson, T.H and Domsch, K.H. 1989. Ratios of microbial biomass carbon to total organic carbon in arable soils. *Soil Biology and Biochemistry*, 21:471–479.

Aneja, K.R. 2003. In (eds) Staining methods: Gram staining of bacteria. In New Age (eds) Experiments in Microbiology Plant pathology and Biotechnology, New Age International (P) Limited Publishers, pp 102–106.

Araújo, J.M. 1998. Strategies for selective isolation of actinomycetes. In: Melo IS, Azevedo JL, (eds) Microbial Ecology, Jaguariuna: EMBRAPA – CNPMA, 351–367.

Awasthi, C.P., Abidi, A.B., and Chowdhury, A.R. 1991. Studies on the nutritional quality of different varieties of chickpea. *Indian Journal of Agricultural Research*, 25: 21–26.

Bailey, B.A., Bae, H., Strem, M.D., Roberts, D.P., Thomas, S.E., Crozier, J., Samuels, G.J., Choi, I.Y. and Holmes, K.A. 2006. Fungal and plant gene expression during the colonization of cacao seedlings by endophytic isolates of four *Trichoderma* species. *Planta*, 224:1449–1464.

Bais, H.P., Weir, T.L., Perry, L.G., Gilroy, S. and Vivanco, J.M. 2006. The role of root exudates in rhizosphere interactions with plants and other organisms. *Annual Review of Plant Biology* 57:233–266.

Bazzicalupo, M., and Fani, R. 1995. The use of RAPD for generating specific DNA probes for microorganisms. In: Clap, J.P. (ed.) *Methods in Molecular Biology, Species Diagnostic Protocols: PCR and other Nucleic Acid Methods*; Humana Press Inc, Totowa NJ, 122–124.

Bhattacharya, A., Chandra, S. and Barik, S. 2009. Lipase and protease producing microbes from the environment of sugar beet field. *Industrial Journal of Agricultural Biochemistry* 22:26–30.

Bhosale, H.J. and Kadam, T.A. 2015. Generic diversity and a comparative account on plant growth promoting characteristics of actinomycetes in roots and rhizosphere of *Saccharum officinarum*. *International Journal of Current Microbiology and Applied Science* 4:230–244.