



## Biofertilizers: Types, Production Methods, and Applications in Sustainable Agriculture

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### Abstract:

Biofertilizers are environmentally sustainable agricultural inputs containing beneficial microorganisms that enhance soil fertility and plant growth by biological processes such as nitrogen fixation, phosphorus solubilization, and organic matter decomposition. Increasing environmental concerns and rising costs of chemical fertilizers have intensified interest in microbial-based nutrient management systems. This study reviews the classification, production methodologies, formulation techniques, and agricultural applications of biofertilizers. Key microorganisms including *Azotobacter*, *Acetobacter*, phosphate-solubilizing bacteria (PSB), plant growth-promoting rhizobacteria (PGPR), and Effective Microorganisms (EM) are discussed. Production involves mother culture preparation, fermentation, formulation, quality control, and packaging. Liquid biofertilizers demonstrate improved shelf life and thermal stability compared to carrier-based forms. Benefits include enhanced nutrient availability, improved crop yield, soil health restoration, and environmental protection. However, variability in field performance and storage constraints remain challenges. Strategic research and technological optimization can enhance the reliability and adoption of biofertilizers in sustainable agriculture.

**Keywords:** Biofertilizers, Nitrogen fixation, Phosphate solubilization, PGPR, Sustainable agriculture, Organic farming.

### Introduction:

#### 1. Background:

Modern agriculture relies heavily on chemical fertilizers to achieve high productivity. While these inputs have significantly increased food production, excessive application has resulted in soil degradation, nutrient imbalance, groundwater contamination, and environmental pollution. Sustainable agricultural systems emphasize the need for biological alternatives that reduce chemical dependency while maintaining productivity. Biofertilizers represent a viable solution by utilizing living microorganisms to improve nutrient availability in soil ecosystems.

#### 2. Definition of Biofertilizers:

Biofertilizers are formulations containing live microorganisms which, when applied to

seeds, soil, or plant surfaces, colonize the rhizosphere and enhance nutrient availability to plants through biological mechanisms such as nitrogen fixation and phosphorus solubilization.

Unlike organic fertilizers, which directly add organic matter, biofertilizers act through microbial metabolic activities.

#### 3. Objectives of the Study:

To classify major types of biofertilizers, to describe production and formulation methods, to evaluate agricultural applications and benefits, to discuss challenges and prospects.

#### Literature Review:

##### 1. Plant Growth-Promoting Rhizobacteria (PGPR):

Plant Growth-Promoting Rhizobacteria (PGPR) enhance plant growth through multiple

direct and indirect mechanisms, including biological nitrogen fixation, phosphorus and other nutrient solubilization, production of phytohormones such as auxins, gibberellins, and cytokinins, and suppression of plant pathogens through antibiotic production and induced systemic resistance. According to Vessey JK (2003), PGPR plays a crucial role in sustainable nutrient management systems by reducing dependence on chemical fertilizers and improving nutrient use efficiency. These beneficial microbes also improve root architecture, enhance stress tolerance under adverse environmental conditions, and contribute to long-term soil health and productivity. Their application in agriculture supports environmentally friendly and economically viable crop production systems.

## 2. Nitrogen-Fixing Biofertilizers:

**2.1 *Azotobacter*:** *Azotobacter* is a free-living aerobic nitrogen fixer capable of converting atmospheric nitrogen into ammonia. It forms cysts under adverse conditions and produces growth-promoting substances.

**2.2 *Acetobacter*:** *Acetobacter*, particularly *Acetobacter diazotrophicus*, thrives in sugar-rich crops such as sugarcane and fixes nitrogen efficiently under acidic soil conditions (pH 3–6).

## 3. Phosphate-Solubilizing Microorganisms:

Soils often contain abundant but insoluble phosphorus. Microorganisms such as: *Pseudomonas*, *Bacillus*, *Aspergillus* produce organic acids that convert insoluble phosphates into plant-available forms.

## 4. Effective Microorganisms (EM):

Effective Microorganisms technology consists of mixed beneficial microorganisms including lactic acid bacteria, yeasts, and photosynthetic bacteria. EM is used for composting, soil fertility improvement, and wastewater treatment.

## 5. Shelf-Life and Stability Studies:

Research indicates: Liquid formulations: Shelf life up to 2 years; tolerance up to 55°C, solid carrier-based formulations: Shelf life 3–6 months; temperature-sensitive.

## Materials and Methods:

### 1. Classification of Biofertilizers:

**Table 1.** Types of Biofertilizers and Their Functions

S. No.	Type	Microorganisms	Primary Function
1	Nitrogen-fixing	<i>Azospirillum</i> , <i>Rhizobium</i> , <i>Azotobacter</i> , <i>Acetobacter</i>	Atmospheric N <sub>2</sub> fixation
2	Phosphate-solubilizing	<i>Bacillus</i> , <i>Pseudomonas</i>	Solubilization of insoluble P
3	Phosphate-mobilizing	<i>Mycorrhiza</i>	Enhanced P uptake
4	PGPR	<i>Pseudomonas sp.</i>	Growth promotion and disease suppression
5	Decomposer cultures	Mixed microbes	Organic waste degradation

### 2. Production Process:

#### 2.1 Mother Culture Preparation:

- Pure strains inoculated in 200–1000 ml flasks.
- Media examples: Jensen's medium (*Azotobacter*), LGI medium (*Acetobacter*), Pikovskaya's medium

(PSB), Incubation at 28–30°C for 3–4 days, Shaking at 120 RPM, Quality check: pH, turbidity, contamination.

**2.2 Fermentation:** Fermenter sterilized at 121°C for 30 minutes,

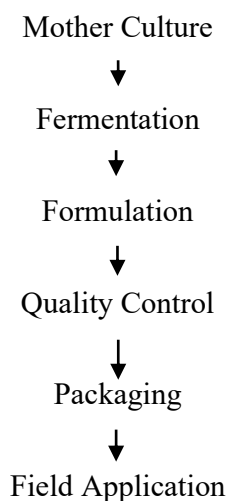
- Inoculated with 10 L starter culture, incubated 3–4 days under controlled

aeration, Cell count adjusted to  $\geq 1 \times 10^9$

CFU/ml.



**Figure 1.** Inoculum media of *Azotobacter sp.* and *Acetobacter sp.*



**Figure 2.** General Production Flowchart of Biofertilizers

**2.3 Formulation and Stabilization:** Additives used: Glycerol (protectant), sucrose (energy source), adhesives for seed coating, curing time: 24–48 hours.

**2.4 Packaging and Labeling:** Packed in sterilized plastic containers (1–20 L).

Label includes product name, target crops, viable cell count, batch number, manufacturing and expiry date and storage instructions.



**Figure 3.** Packaging and Labeling of Liquid Biofertilizer in the Bottle

**2.5 Preparation of EM Solution:** 1 L EM.1 culture, 1 kg jaggery, 20 L chlorine-free water, fermentation: 5–7 days at 20–40°C and final pH  $\approx$  3.5

#### 2.6 Application Methods:

- **Soil Application:** Apply 1–5 L/ha diluted with water.
- **Seed Treatment:** Seeds coated before sowing.
- **Compost Enrichment:** Layer organic waste with culture; compost 90–120 days.
- **Waste Management:** Applied to organic waste heaps or spent wash for decomposition.

## Results and Discussion:

**1. Nitrogen Fixation Efficiency:** *Azotobacter* fixes approximately 10–20 kg N/ha/year, *acetobacter* supports high nitrogen-demand crops such as sugarcane, reduced requirement of synthetic nitrogen fertilizers observed.

**2. Phosphate Solubilization Activity:** PSB forms clear halo zones on Pikovskaya's agar, indicating phosphate solubilization through organic acid production.

### 3. Comparison of Liquid and Solid Biofertilizers:

**Table 2.** Comparison Between Liquid and Carrier-Based Biofertilizers

Parameter	Liquid	Solid
Shelf Life	Up to 2 years	3–6 months
Temperature Tolerance	Up to 55°C	Low
CFU Stability	High	Declines rapidly
Application Efficiency	Higher	Moderate

**4. Environmental Benefits are** Reduced Chemical Fertilizer Use, Improved soil microbial diversity, Lower greenhouse gas emissions, Enhanced nutrient cycling.

**5. Constraints:** Inconsistent field results, Soil-type dependency, Limited farmer awareness, Storage sensitivity, Research supports multi-strain consortia for synergistic effects.

## Conclusion:

Biofertilizers provide a sustainable alternative to chemical fertilizers by improving nutrient availability through biological mechanisms. Nitrogen-fixing bacteria such as *Azotobacter* and *Acetobacter*, phosphate-solubilizing bacteria, and EM consortia significantly enhance soil fertility and crop productivity. Liquid formulations offer superior shelf life and thermal stability. Although challenges remain, improved production technology, strain selection, and farmer education

can expand adoption in sustainable agricultural systems.

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## References

- Adesemoye, A. O., & Kloepper, J. W. (2009). Plant–microbes interactions in enhanced fertilizer-use efficiency. *Applied Microbiology and Biotechnology*, 85(1), 1–12. <https://doi.org/10.1007/s00253-009-2196-0>
- Ahemad, M., & Kibret, M. (2014). Mechanisms and applications of plant growth promoting rhizobacteria: Current perspective. *Journal of King Saud University – Science*, 26(1), 1–20. <https://doi.org/10.1016/j.jksus.2013.05.001>
- Backer, R., Rokem, J. S., Ilangumaran, G., Lamont, J., Praslickova, D., Ricci, E., Subramanian, S., & Smith, D. L. (2018). Plant growth-promoting rhizobacteria: Context, mechanisms of action, and road to commercialization of biostimulants for sustainable agriculture. *Frontiers in Plant Science*, 9, 1473. <https://doi.org/10.3389/fpls.2018.01473>
- Bashan, Y., de-Bashan, L. E., Prabhu, S. R., & Hernandez, J. P. (2014). Advances in plant growth-promoting bacterial inoculant technology: Formulations and practical perspectives (1998–2013). *Plant and Soil*, 378(1–2), 1–33. <https://doi.org/10.1007/s11104-013-1956-x>
- Bhardwaj, D., Ansari, M. W., Sahoo, R. K., & Tuteja, N. (2014). Biofertilizers function as key players in sustainable agriculture by

- improving soil fertility, plant tolerance, and crop productivity. *Microbial Cell Factories*, 13(1), 66. <https://doi.org/10.1186/1475-2859-13-66>
6. Chaudhary, P., Sharma, A., Mishra, A., & Jha, B. (2022). Advances in biofertilizer research for sustainable agriculture: Current trends and future perspectives. *Journal of Applied Microbiology*, 132(5), 3214–3232. <https://doi.org/10.1111/jam.15474>
  7. Glick, B. R. (2012). Plant growth-promoting bacteria: Mechanisms and applications. *Scientifica*, 2012, 963401. <https://doi.org/10.6064/2012/963401>
  8. Kumar, A., Patel, J. S., Meena, V. S., & Srivastava, R. (2021). Recent advances of PGPR-based approaches for stress tolerance in plants for sustainable agriculture. *Biocatalysis and Agricultural Biotechnology*, 31, 101913. <https://doi.org/10.1016/j.bcab.2020.101913>
  9. Lugtenberg, B., & Kamilova, F. (2009). Plant-growth-promoting rhizobacteria. *Annual Review of Microbiology*, 63, 541–556. <https://doi.org/10.1146/annurev.micro.62.081307.162918>
  10. Mahanty, T., Bhattacharjee, S., Goswami, M., Bhattacharyya, P., Das, B., Ghosh, A., & Tribedi, P. (2017). Biofertilizers: A potential approach for sustainable agriculture development. *Environmental Science and Pollution Research*, 24(4), 3315–3335. <https://doi.org/10.1007/s11356-016-8104-0>
  11. Mitter, E. K., Tosi, M., Obregón, D., Dunfield, K. E., & Germida, J. J. (2021). Rethinking crop nutrition in times of modern microbiology: Innovative biofertilizer technologies. *Frontiers in Sustainable Food Systems*, 5, 606815. <https://doi.org/10.3389/fsufs.2021.606815>
  12. Singh, D., Geat, N., Rajawat, M. V. S., & Prasanna, R. (2022). Biofertilizers and biostimulants for sustainable agriculture: Mechanisms and applications. *Agronomy*, 12(3), 672. <https://doi.org/10.3390/agronomy12030672>
  13. Vessey, J. K. (2003). Plant growth promoting rhizobacteria as biofertilizers. *Plant and Soil*, 255(2), 571–586. <https://doi.org/10.1023/A:1026037216893>
  14. Yadav, A. N., Verma, P., Kumar, V., Sangwan, P., Mishra, S., Panjiar, N., Gupta, V. K., & Saxena, A. K. (2021). Advances in biofertilizer technology for sustainable agriculture. *Plant and Soil*, 461(1–2), 1–26. <https://doi.org/10.1007/s11104-020-04757-9>
  15. Zahir, Z. A., Arshad, M., & Frankenberger, W. T. (2004). Plant growth promoting rhizobacteria: Applications and perspectives in agriculture. *Advances in Agronomy*, 81, 97–168. [https://doi.org/10.1016/S0065-2113\(03\)81003-9](https://doi.org/10.1016/S0065-2113(03)81003-9)