Microbiological Processes

From the field to a market-ready product

1 Isolation of microorganisms

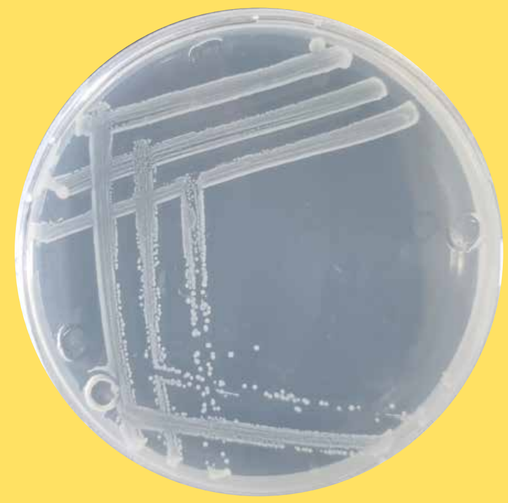
The plant-microorganism relationship:
- Parasitism
- Commensalism
- Symbiosis

Economically important microorganisms:
including endosymbiotic bacteria that form root nodules (nitrogen fixation); *Bacillus ssp.*, *Azospirillum ssp.* and other symbionts

In the laboratory, microorganisms are isolated from soil and water samples, as well as from plant tissue.

2 Separation and pure culture

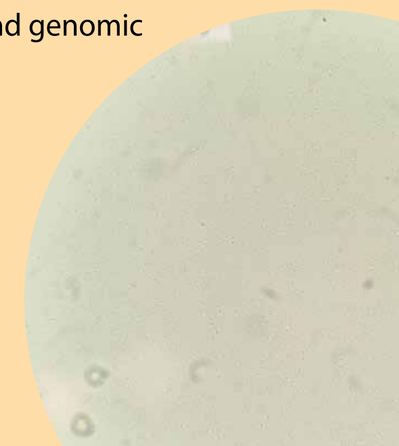
- **Culturing** in a sterile nutrient medium
- **Separation** to isolate genetically identical colonies of bacteria
- Transition to **strain maintenance**



3 Microbiological analyses

- **Microscopy:** determination of the number of cells, morphology and motility
- Chemotaxonomical **identification**
- Count of colony-forming units **(CFU/mL)**
- 16S rRNA sequencing and genomic analysis

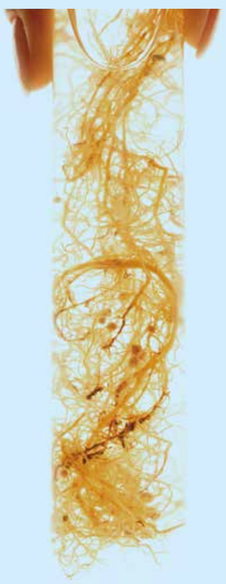
➜ Quality assurance/ -control and selection of strains with preferred traits



5 In vitro culturing

- Inoculations of sterile plants or seeds with bacteria
- Breeding in a specially selected growing medium
- Analysis of the plant's nodule formation, morphology and vitality

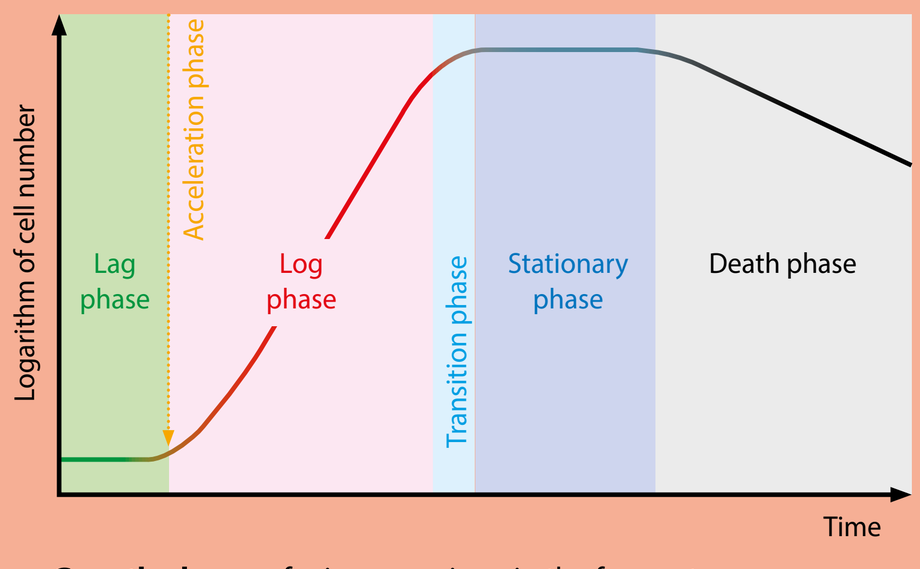
➜ Quality control



4 Fermentation

- **Liquid cultures** beginning with a small number of genetically identical cells
- Incubated in shake flasks and the fermenter for large-scale **propagation**

Ideal growth conditions for cells:
- Available nutrients
- Availability of O_2, N_2, CO_2
- Temperature
- Turbulence of liquid culture medium



Growth phases of microorganisms in the fermenter

Graphic adapted from: https://quizlet.com/de/417629364/vl-09-mikrobiologische-arbeitsmethoden-iimikro-bielles-wachstum-in-reinkultur-flash-cards/

Fermentation product – Result

RhizoFix®: Bacterial strains are optimised for each crop.
- *Bradyrhizobium japonicum*
- *Rhizobium fabae*
- *Rhizobium pisi*
- *Rhizobium leguminosarum*
- *Ensifer meliloti*