

# **Microbiologia del suolo**

Corso di Aggiornamento " **Obiettivo  
suolo**"

Sez. ANISN Firenze

# La biodiversità del suolo

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The Convention on Biological Diversity (CBD) defined the **soil biodiversity** as "*the variation in soil life, from genes to communities, and the ecological complexes of which they are part, that is from soil micro-habitats to landscapes*".

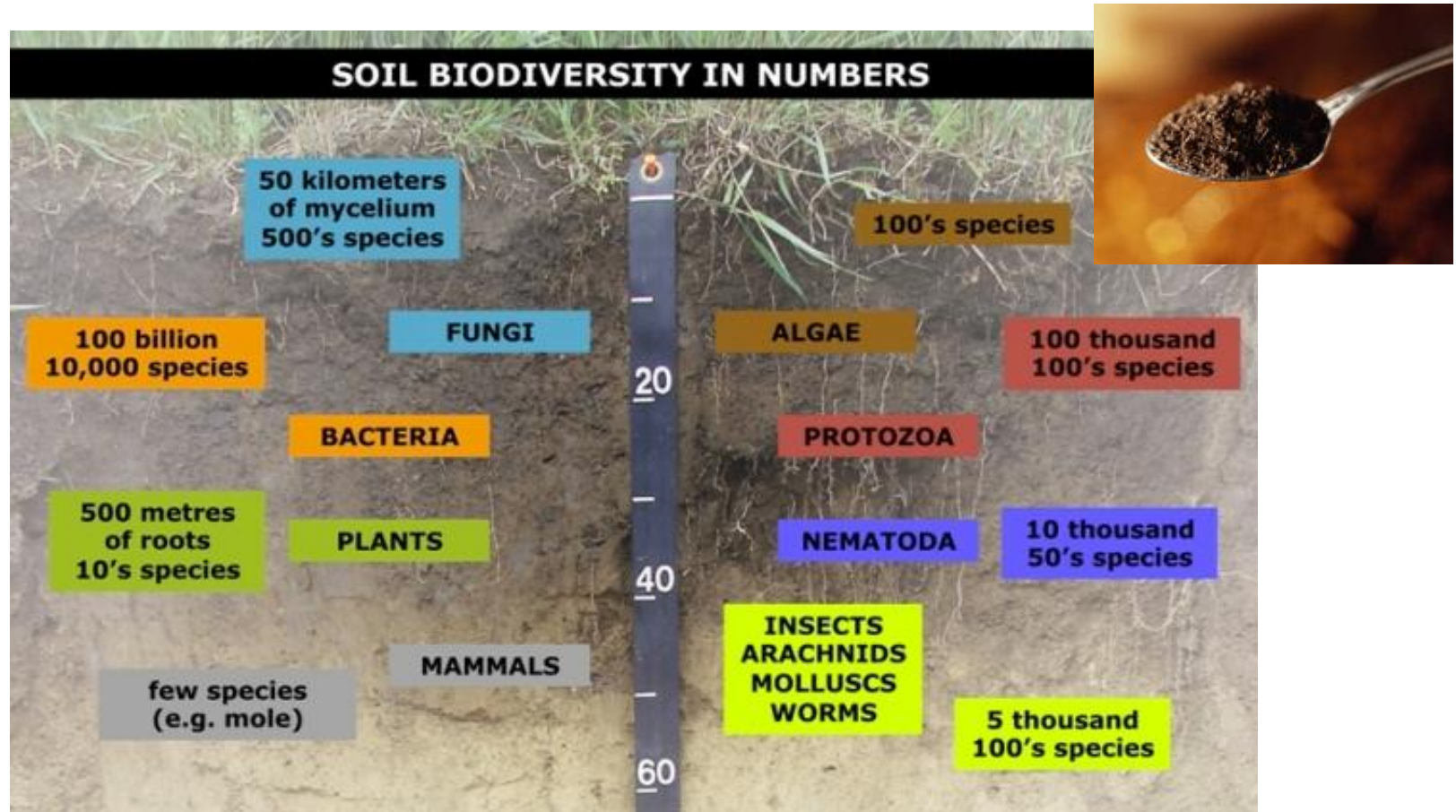
In other terms the **soil biodiversity represents the variety of life belowground**. The concept is conventionally used in a genetic sense and denotes the number of distinct species (richness) and their proportional abundance (evenness) present in a system, but may be extended to encompass phenotypic (expressed), functional, structural or trophic diversity. The total biomass belowground generally equals or exceeds that aboveground, whilst the **biodiversity in the soil always exceeds that on the associated surface by orders of magnitude, particularly at the microbial scale**.

# La biodiversità del suolo

Some numbers describe well the soil biodiversity:  
a **teaspoon** of soil (about one gram) may typically contain **one billion bacterial cells** (corresponding to about ten thousand different bacterial genomes), up to **one million individual fungi**, about **one million cells of protists**, and **several hundred of nematodes**.  
Beside microorganisms and microfauna, soil harbours different species of meso and macro/megafauna represented by arthropods, earthworms and mammals.



# La biodiversità del suolo



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The soil biota plays many fundamental roles in delivering key ecosystem **goods** and **services**.

Ecosystems **goods** provided by soil biota are:

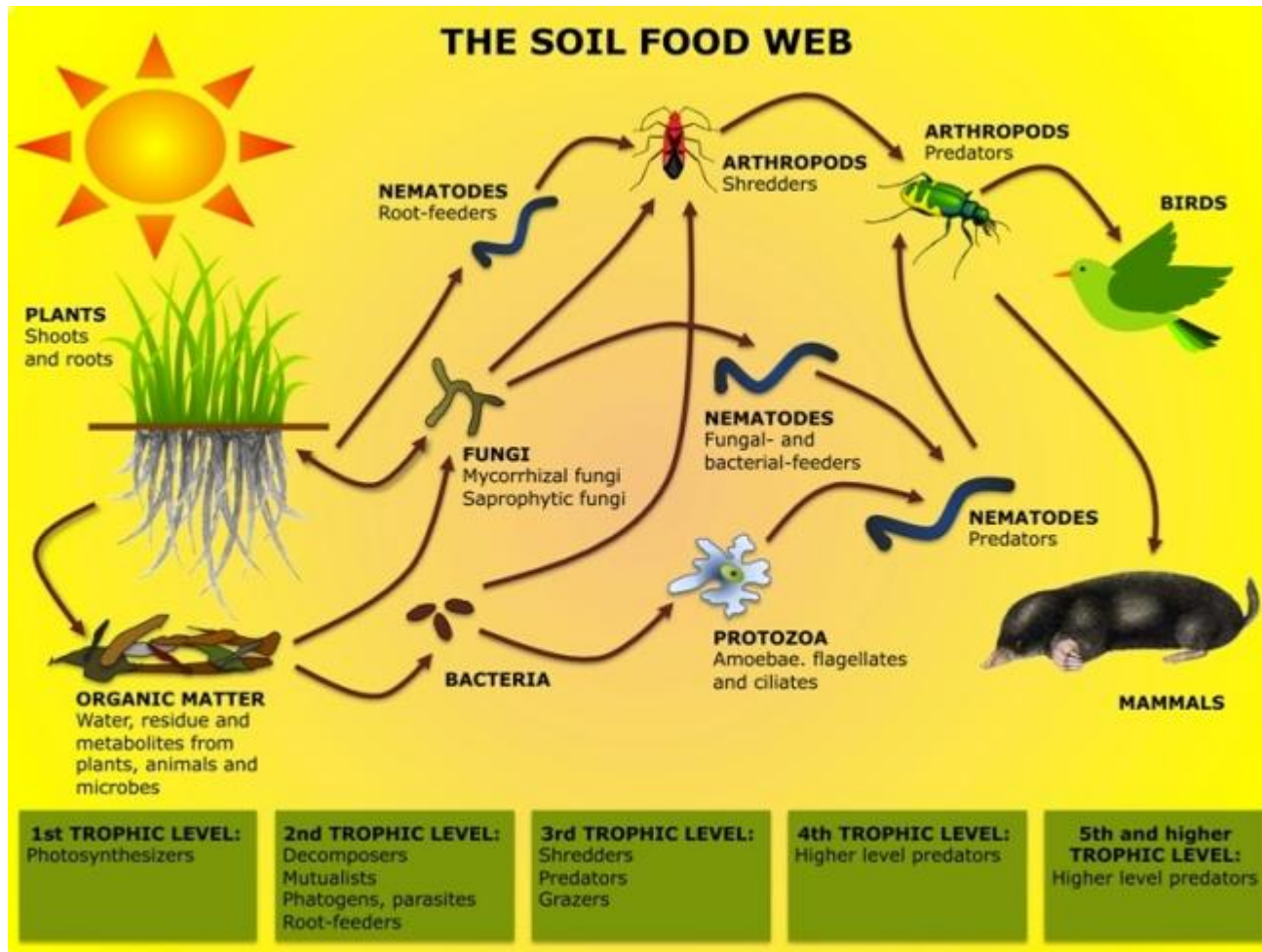
- ✓ food production;
- ✓ fibre production;
- ✓ fuel production;
- ✓ provision of clean water;
- ✓ provision of secondary compounds (e.g. pharmaceuticals and agrochemicals).

Ecosystems **services** provided by soil biota:

- ✓ driving nutrient cycling and regulation of water flow and storage;
- ✓ regulation of soil and sediment movement and biological regulation of other biota (including pests and diseases);
- ✓ soil structure maintenance;
- ✓ detoxification of xenobiotics and pollutants and regulation of atmospheric composition.



# La biodiversità del suolo



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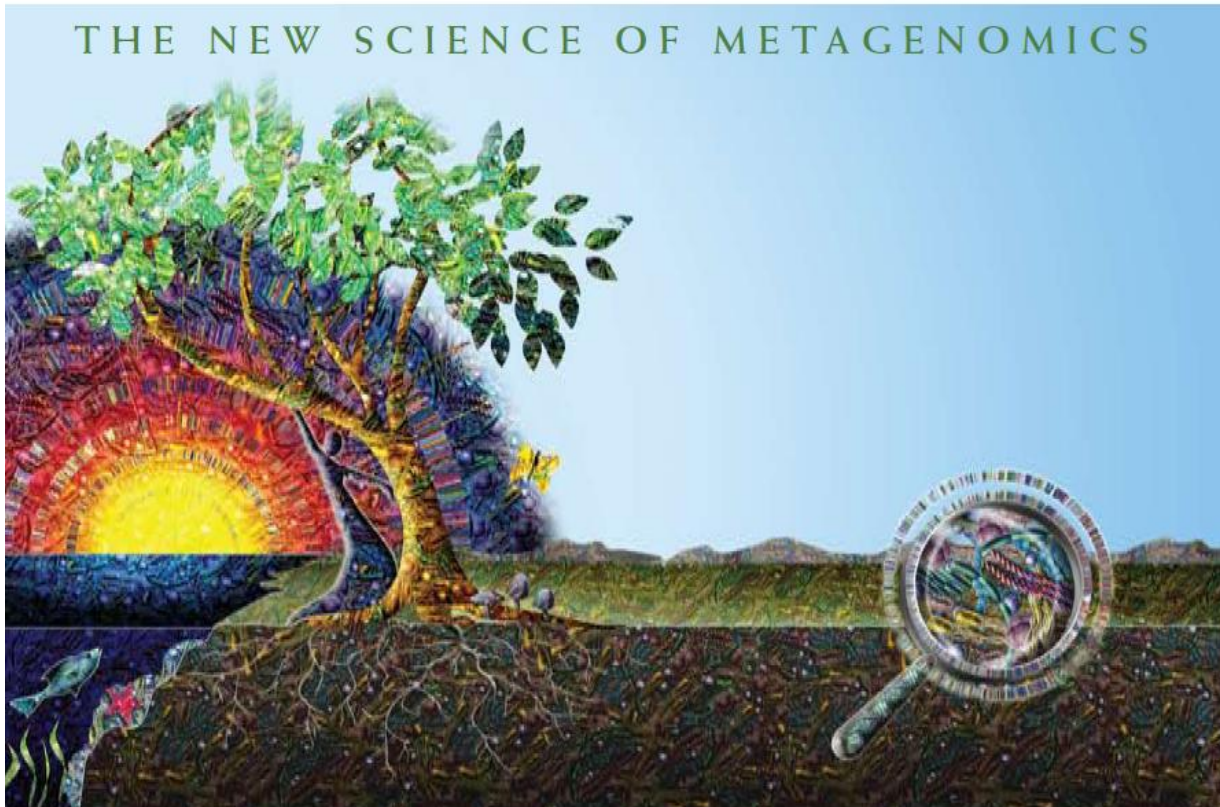
## Metagenomica

*Metagenomics transcends individual genes and genomes, enabling scientists to study the entire genetic makeup of a community as a whole. It is the science of microbial communities.*

# La biodiversità del suolo

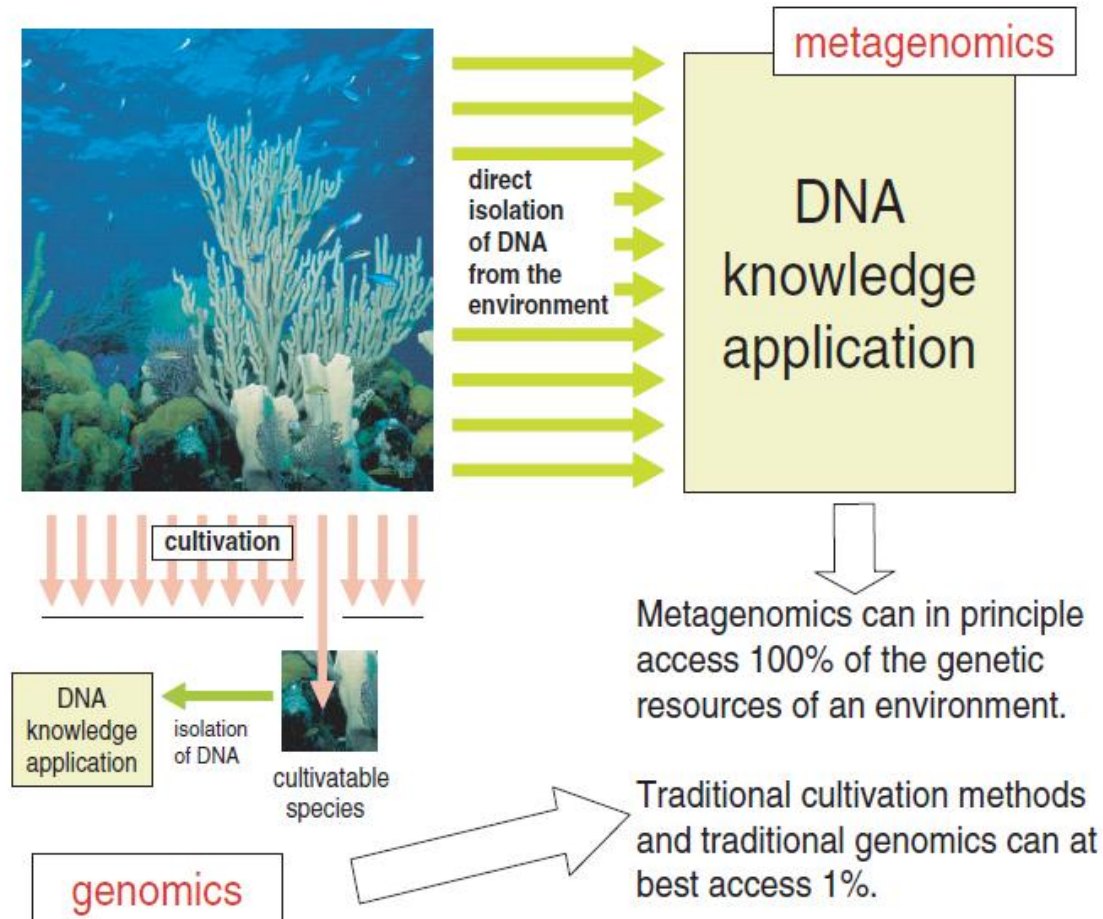
## UNDERSTANDING OUR MICROBIAL PLANET

THE NEW SCIENCE OF METAGENOMICS





# La biodiversità del suolo

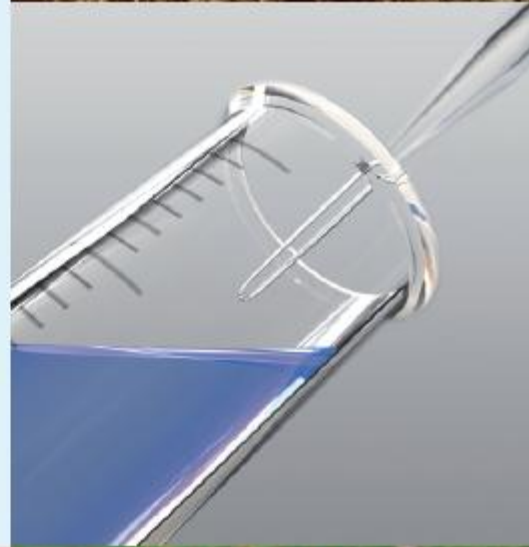


# La biodiversità del suolo

## Illuminating biology

Metagenomics could answer some fundamental biological questions. Microbial communities are composed of thousands or millions of different but interdependent individuals; some are closely related enough to be considered the same species, whereas others have few genes in common. Genetic material in these communities is often passed from one species to another, which poses questions such as: what is a genome; what is a species; how diverse is life; how do microbial communities react to change; and how do microorganisms evolve?

Metagenomics is uniquely suited for identifying genes involved in competition or cooperation. Such genes are almost impossible to identify outside of the community context, but metagenomic analysis can yield informative insights. Investigations of communication among bacteria, for example, have found that subinhibitory concentrations of many antibiotics induce quorum sensing. Using metagenomics to screen for signalling and inhibitory compounds might therefore yield molecules that are quorum-sensing inducers as well as antibiotics. Metagenomics can also support community-wide assessments of metabolic and geochemical functions.



# La biodiversità del suolo

## Cross-disciplinary applications

Metagenomics makes possible insights that could help to address some of the most complex medical, environmental, agricultural and economic challenges in today's world.

*Medicine:* understanding how the microbial communities that inhabit our bodies affect human health could lead to new strategies for diagnosing, treating and preventing diseases.

*Earth sciences:* exploring how microbial communities in the soil and ocean affect the atmosphere and environmental conditions could help us understand, predict and address global changes.

*Alternative energy:* harnessing the power of microbial communities might result in sustainable and eco-friendly bioenergy sources.

*Environmental remediation:* metagenomics could aid the development of microorganism-based tools for monitoring environmental damage and cleaning up oil spills, groundwater, sewage, nuclear waste and other hazards.

*Biotechnology:* taking advantage of the functions of microbial communities might lead to the development of new functional food and health products.

*Agriculture:* understanding the roles of beneficial microorganisms living in, on and around domesticated plants and animals could enable detection of diseases in crops and livestock, and aid the development of improved farming practices.

*Biodefence and microbial forensics:* studying the DNA and biochemical fingerprints of microbial communities helps to monitor pathogens, create more effective vaccines and therapeutics against bioterror agents, and reconstruct attacks that involve microorganisms.

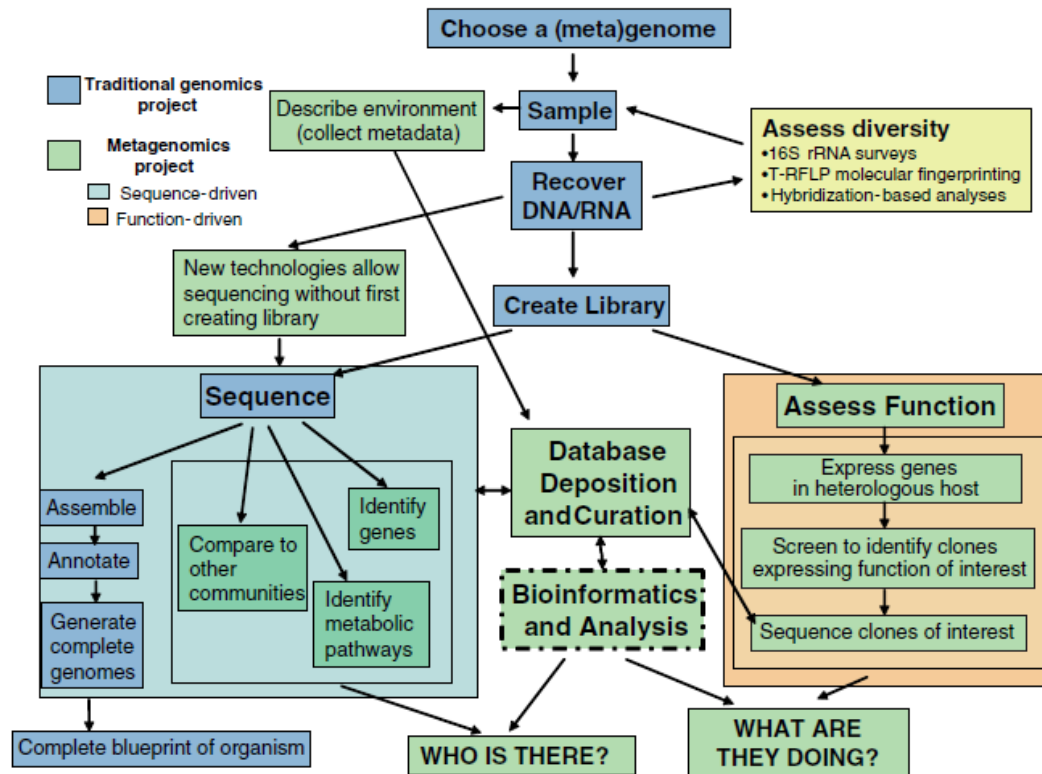




# La biodiversità del suolo

DESIGNING A SUCCESSFUL METAGENOMICS PROJECT

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# La biodiversità del suolo

## OUR MICROBIAL PLANET

MICROBES—life forms too tiny to see—play a surprisingly large role in life on Earth. Microbes are everywhere, and they do a lot of good for human health and our planet. In fact, disease-causing microbes make up only a very tiny fraction of the millions of types of microbes. Microbes...

Think microbes are bad guys? Think again.

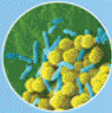


**Keep us healthy.** Amazingly, only about 1 out of 10 cells in the human body is actually a human cell; most of the cells in our bodies are microbes! Some of the microbes living in our bodies actually help us fight disease-causing microbes by competing against them for space. This mutually beneficial relationship helps to protect us from getting diseases while giving the "good" microbes a place to live.

**Make air breathable.** Without microbes, we wouldn't have oxygen to breathe. This is because many microbes are photosynthetic—like plants, they harvest their energy from the sun, releasing oxygen into the air. Billions of years ago, photosynthetic microbes gradually added oxygen to Earth's atmosphere, making it possible for larger forms of life—including humans—to live.



**Provide sources of new medicines.** Hundreds of medicines available today were derived from chemicals first found in microbes. Microbes naturally produce an amazing variety of chemicals, which scientists can use to create new medicines.



**Help us digest food.** Many of the foods we eat would be indigestible without the 10-100 trillion microbes living within our guts. Microbes also play a major role in creating many of the foods we love, such as cheese, yogurt and bread.



**Keep our environment clean.**

Because of their special adaptations, some microbes can help clean up gasoline leaks, oil spills, sewage, nuclear waste, and many other types of pollution.



**Support and protect crops.**

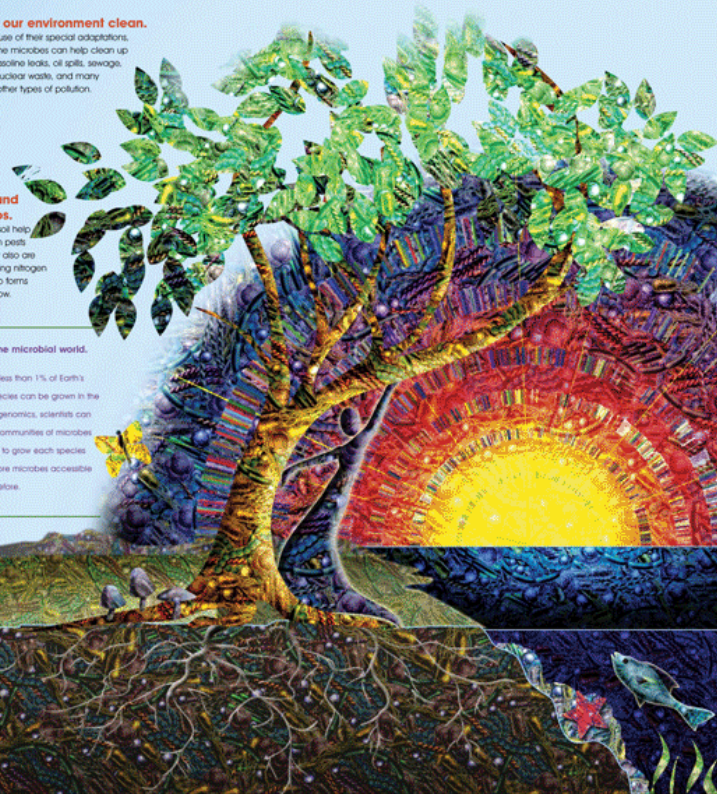
Microbes living in soil help protect plants from pests and diseases. They also are essential for converting nitrogen and other nutrients into forms that plants can use to grow.

The science of metagenomics is shedding new light on the microbial world.



Scientists estimate that less than 1% of Earth's millions of microbial species can be grown in the laboratory. Using metagenomics, scientists can now study how whole communities of microbes function without having to grow each species separately—making more microbes accessible to science than ever before.

Living in a microbial world...



Visit [www.nationalacademies.org/microbes](http://www.nationalacademies.org/microbes) to learn more!

This poster was derived from the National Research Council report *The New Science of Metagenomics: Revealing the Secrets of Our Microbial Planet* (2012). Reports from the National Academies are available from the National Academies Press, 500 Fifth St., NW, Washington, DC 20001; 800-424-6342. <http://www.nao.edu>. Reports are available online in a PDF searchables format.

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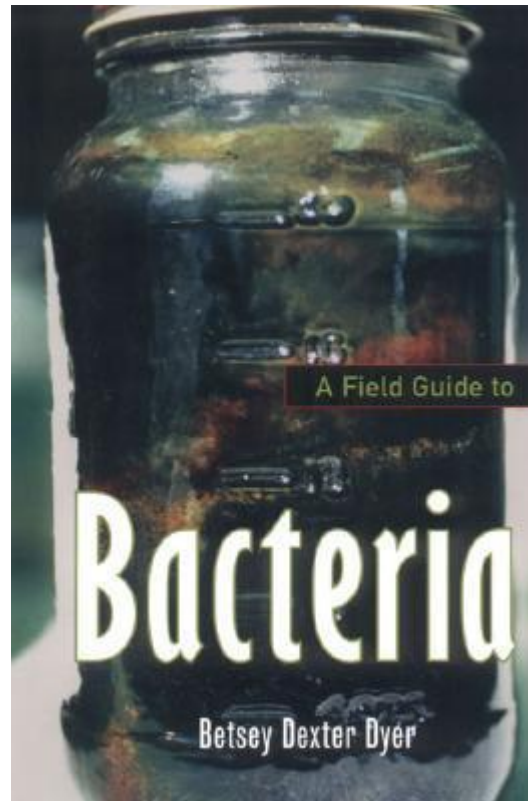


# La biodiversità del suolo

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# A Field Guide to Bacteria

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by Betsey Dexter Dyer

# Microrganismi e piante

## I rizobi (azotofissatori)

Rhizobia are soil bacteria that fix nitrogen (diazotrophs) after becoming established inside root nodules of legumes (Fabaceae). Rhizobia require a plant host; they cannot independently fix nitrogen.



# Microrganismi e piante

## Fissazione biologica dell'azoto

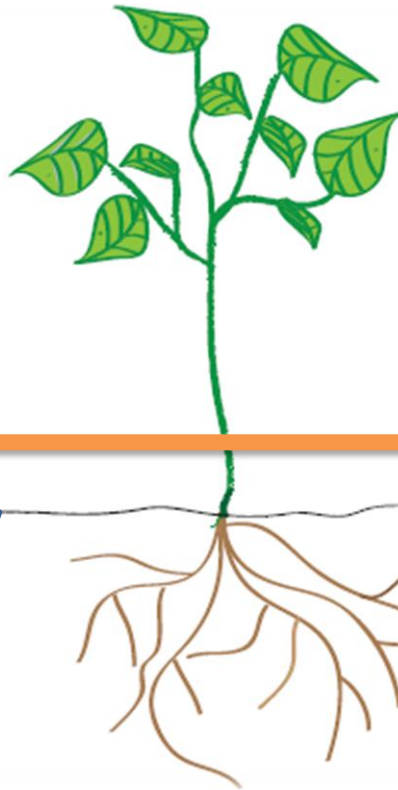


# Gli azotofissatori simbiotici

**Rizosfera**

$\text{NH}_3$ , protezione da  
parassiti e patogeni

Fonti di carbonio



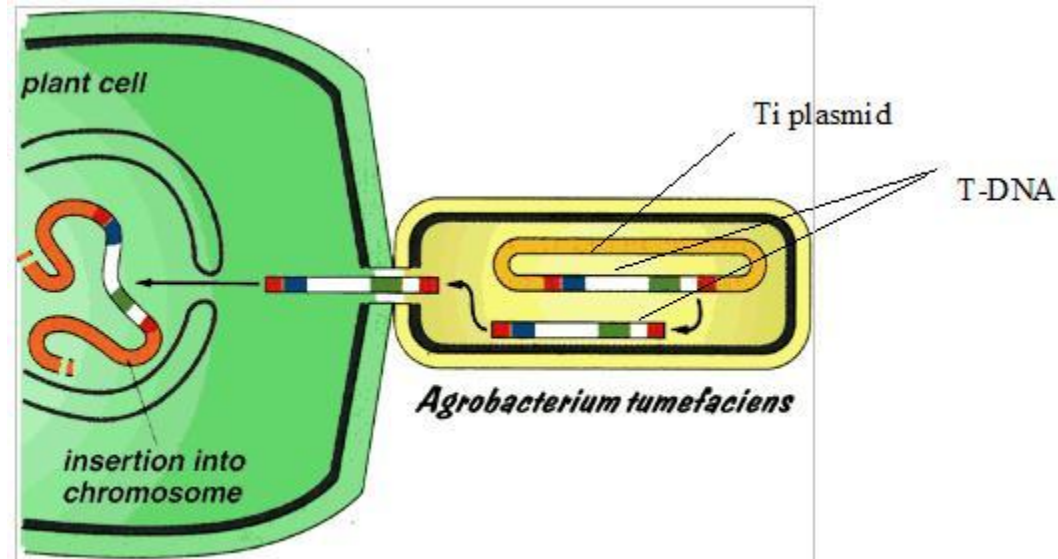


# Microrganismi e piante

## L'agrobatterio (l'ingegnere genetico naturale)

L'*Agrobacterium tumefaciens* è un batterio capace di infettare le piante attraverso la trasmissione di un segmento di DNA, definito T-DNA, che penetra all'interno delle cellule vegetali integrandosi nel loro genoma.

È capace di infettare principalmente le dicotiledoni e causare in esse una crescita paragonabile a quella tumorale, una patologia nota con il nome di galla del colletto.



*A. tumefaciens* è un proteobatterio della famiglia delle Rhizobiaceae, la stessa cui appartengono molti batteri azotofissatori simbiotici delle piante. A differenza di questi ultimi, tuttavia, *A. tumefaciens* si comporta da parassita; in questi termini, ha una notevole importanza in campo economico, in grado di arrecare danni a coltivazioni quali quelle della vite.

# Microrganismi e piante

L'agrobatterio (l'ingegnere genetico naturale)

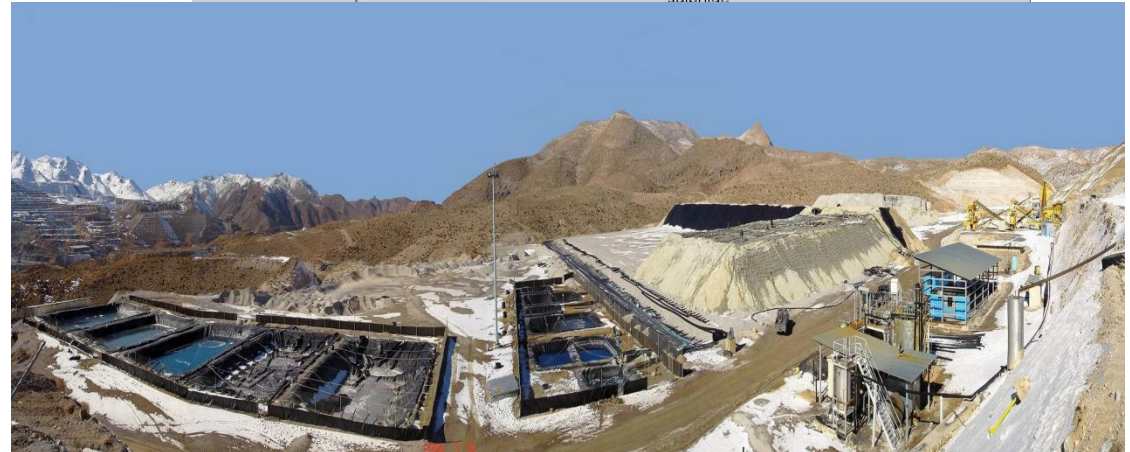
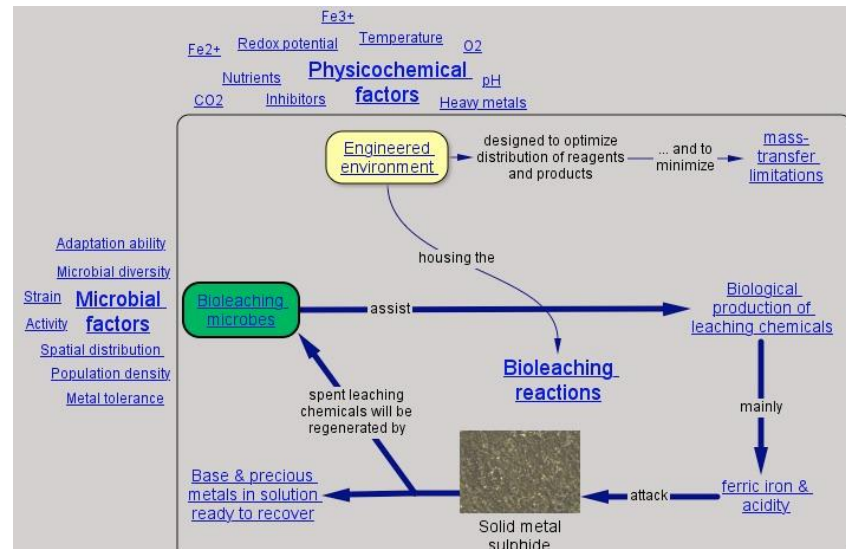




# Microrganismi e rocce

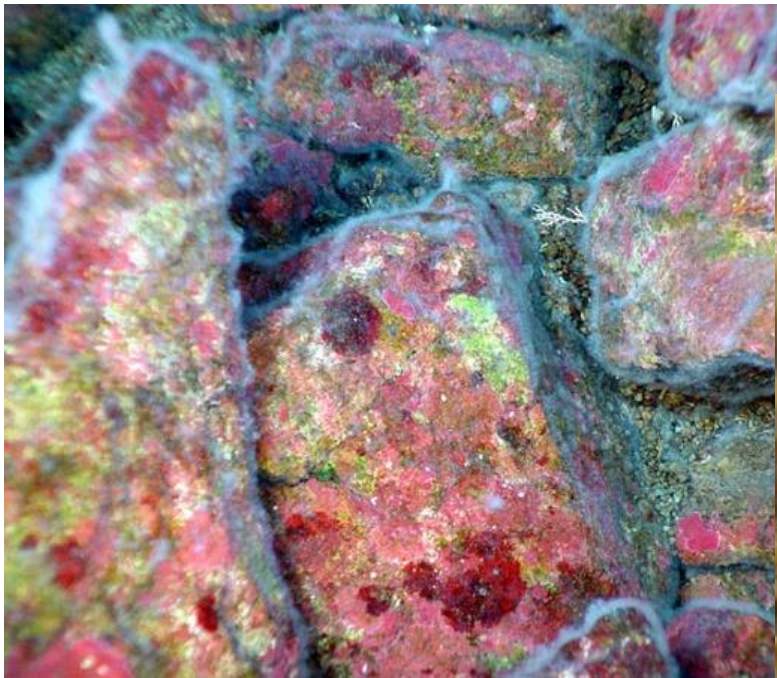
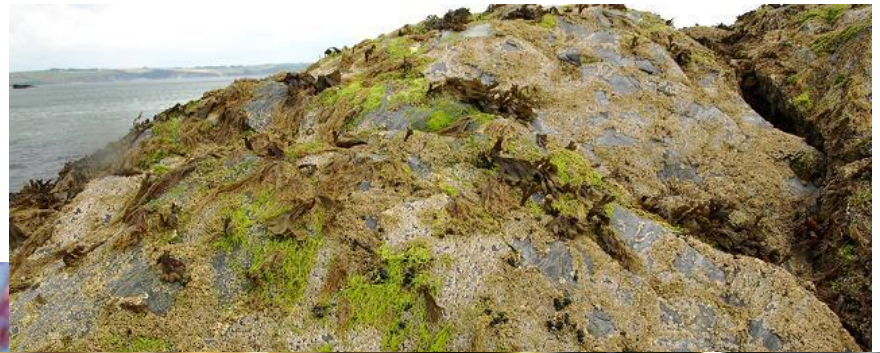
## Batteri “mangia rocce”

Bioleaching is the extraction of metals from their ores through the use of living organisms. This is much cleaner than the traditional heap leaching using cyanide. Bioleaching is one of several applications within biohydrometallurgy and several methods are used to recover copper, zinc, lead, arsenic, antimony, nickel, molybdenum, gold, silver, and cobalt.



# Microrganismi e rocce

Croste di cianobatteri





# Microrganismi e rocce

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Croste di cianobatteri



Stromatoliti





# Microrganismi e rocce

## Batteri e monumenti

- Cianobatteri
- Produzione di solfato da  $H_2S$  e trasformazione del marmo in gesso



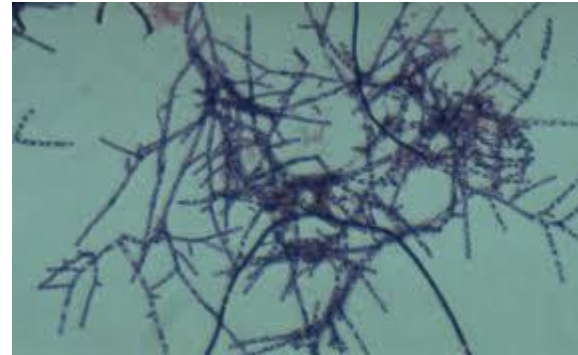
# Microrganismi e suolo

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Many times after a rain, there is a distinctive odor in the air — a sort of musky smell. This pleasant fragrance is most common in rains that follow a dry spell. If you are a gardener, you may find this smell similar to the smell you sense when you turn over your soil. Good organic soils contain bacteria. One bacterium that is abundant in damp warm soils is actinomycete.

**Actinomycetes** are a key ingredient in the decomposition of organic materials in the soil. These bacteria thrive when the soil is moist. When the soil dries out, the actinomycetes produce tiny spores. These spores are part of their reproduction cycle. Rain kicks up these spores when the raindrops hit the ground and make them airborne. Air movements then disperse the spores and carry some of the spores to our nose, where we detect a pleasant aroma. Actinomycetes are very common, which is why you experience the pleasing after-the-rain smell in many locations. Winds can transport odors from areas upwind of you. We also can experience bad after-the-rain smells, especially in urban regions. Rainwater tends to be slightly acidic and can break apart organic materials on the ground. The rain can release minerals in the soil that can also react with any chemicals on roadways, such as gasoline and oil, leading to unpleasant smells. These chemical smells are more noticeable with rains that follow a dry spell, as the chemicals have not been diluted or washed away.

# Microrganismi e suolo



# Analisi della microflora batterica

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## Materiali occorrenti

- Piastre di terreno di coltura agarizzato (LB, TY, TSB, ecc.)
- Provette per diluizioni (es. 1.5 ml)
- Provette da 50 ml
- Spatola sterile
- Anse per piastrare
- Pipette automatiche
- Puntali per pipette automatiche
- Soluzione di 10 mM  $\text{MgSO}_4$

# Analisi della microflora batterica

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## Procedura

1. Prelevare in maniera sterile con un cucchiaino circa 1 g di suolo
2. Mettere il suolo in una provetta da 50 ml e aggiungere 10 ml di 10 mM  $\text{MgSO}_4$  (o altra soluzione salina).
3. Porre la provetta con il suolo e la soluzione salina in agitazione per circa 30'.
4. Lasciar decantare le particelle più grosse per 20'.
5. Prelevare 100  $\mu\text{l}$  di soprantante e preparare delle diluizioni seriali (es.  $10^{-2}$ ,  $10^{-4}$ ,  $10^{-6}$ ).
6. Piastrare aliquote di ciascuna diluizione in piastre di terreno di coltura agarizzato.
7. Lasciare ad incubare a  $30^\circ\text{C}$  per 2 giorni



# Analisi della microflora batterica

## Analisi risultati

- ✓ Stimare il titolo batterico (conta delle colonie su piastra)
- ✓ Determinare la biodiversità (a livello morfologico):
  - quanti morfotipi ci sono?
  - quanto sono rappresentati i morfotipi?

Calcolare gli indici di biodiversità:

1. Indice di Shannon

$$H = - \sum_{i=1}^x p_i \ln p_i$$

2. Richness (n. taxa presenti)

3. Evenness

$$E_H = H / H_{\max} = H / \ln S$$