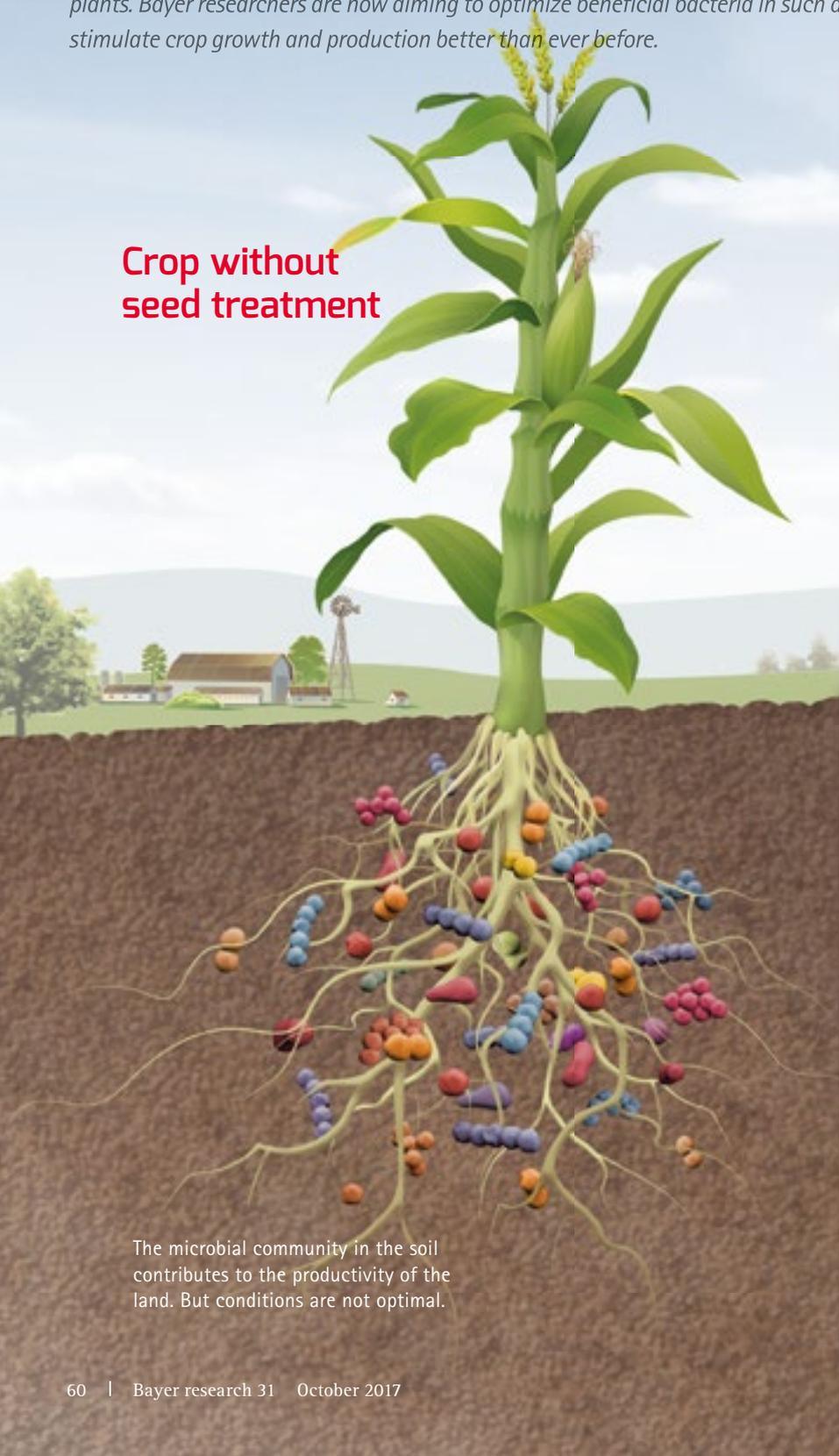


# Tiny microbes revolutionizing farming

For millions of years, plants and bacteria have coexisted in soil in a symbiosis from which both benefit. For example, microbes make essential elements such as nitrogen and phosphorus available in exchange for carbon provided by plants. Bayer researchers are now aiming to optimize beneficial bacteria in such a way that they will protect and stimulate crop growth and production better than ever before.

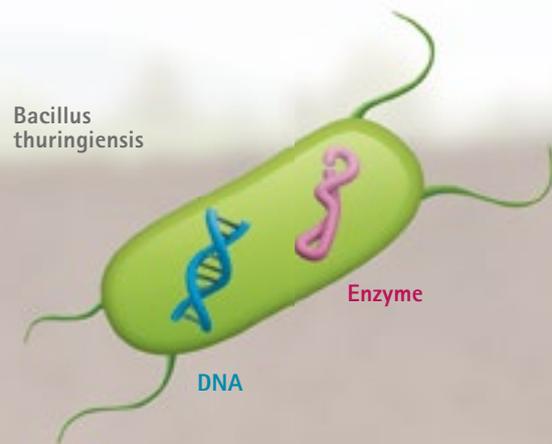
**Crop without seed treatment**



The microbial community in the soil contributes to the productivity of the land. But conditions are not optimal.

**Seed treatment**

- 1** Genetically enhanced bacterial DNA leads to stabilized enzymes.



- 2** The enzymes are more stable in the dormant spore form of the bacteria. Spores are made by certain bacteria under adverse conditions. Bayer researchers are harnessing this mechanism.



Spore formation

## Crop with seed treatment

The productivity of a field depends to a large extent on the soil quality, in which microbes play a major role (left). Bayer scientists make use of this fact: they apply bacteria in a special coat to the seeds (center) and deliver them directly to the root zone (right). Here the micro-organisms assist in nutrient availability, and, following an enhanced uptake, can help increase yields of the crop.



4

Seeds coated with bacterial spores are planted.

3



The stabilized enzymes are anchored to the surface of the spores.

Plant seeds are coated with bacterial spores.

5

The stabilized enzymes on the spores help create optimum growth conditions for crops in the soil.

Today, "biologicals" – products derived from natural materials such as plants, bacteria, fungi or minerals – contribute to sustainable agriculture. They provide benefits to farmers by protecting plants against pests and diseases and making them more productive. Bayer researchers are now able to provide new solutions to growers by optimizing beneficial bacteria. "Farmers have always planted their crops in the soil that offers them good conditions for growth. Today, our understanding of the relationship between soil bacteria and plants has increased with new research and advancements of technologies," explains Dr. Damian Curtis, head of Microbial Genetic Systems in Bayer's Crop Science Division.

The scientists at Bayer's site in West Sacramento, California, are aiming to stimulate peak crop performance by means of directed application of optimized bacteria. One way these tiny beneficial organisms are introduced to the fields is as a seed treatment on the seed that is being sown. "There are incredibly large numbers of different soil bacteria, and the root microbiome has a highly complex composition," says Dr. Bjorn Traag, group head of Microbiology. Selecting the right strains – different members of a species of bacteria that can have extremely variable properties – is a very early and crucial step in the work of the bacterial geneticists. "We take a look at the physical and biochemical characteristics – or phenotype – of the microbes and their genes, the units of the heritable functions present on the DNA of living organisms," explains Traag.

### Bacterial DNA can be changed in a very targeted manner

Once the researchers have identified a strain of bacteria with activities of interest from their collection of naturally occurring bacteria sourced from the soil, they optimize its genetic material to further improve the relevant characteristics of the strain. "There are methods to optimize the DNA of a strain that are based on random variation of the genetic blueprint in the cells, but also methods which allow us to make changes in a very targeted manner," explains Traag. The Bayer scientists are working with both methods depending on the specific agricultural need.

When using random methods, the team utilizes different techniques that cause small changes to the microbe's genome and then evaluate thousands or tens of thousands of microbes derived from the original strain. Some of the "offspring" will have improved phenotypes that the researchers can investigate using a variety of different measuring tools. These "screens" have been used to optimize microbes for a variety of applications for many years.

In some cases it is more efficient, or even necessary, to use targeted techniques for strain improvement to create products providing benefits to growers. One example is being employed by scientists led by Dr. Damian Curtis and uses the microbial spore as a carrier of agriculturally active proteins. "Our objective is to stabilize these biomolecules via targeted methods," says Curtis. Proteins and enzymes can improve plant health, increase yields, and protect plants against pests or diseases but they are nor-



Improved attributes: Dr. Jennifer Riggs is holding corn seeds with a special coating. The green color is used to distinguish coated products from untreated ones and from other products with different attributes.

mally unstable or easily degraded in agricultural settings. Bayer and Elemental Enzymes, a U.S. start-up venture, have applied an approach that makes the enzyme more stable to degradation processes.

Elemental Enzymes are collaborating with Bayer, combining their proprietary technology for enzyme stabilization to genetically enhance bacteria with Bayer's decades of experience in agriculture. The scientists are using bacteria from the *Bacillus* genus and have introduced a gene consisting of DNA from two closely related *Bacillus* species – the optimized microbe is referred to as "intrageneric."

### Enzymes are stabilized to make crops more productive

The bacteria is utilized in two ways: first, to express the enzyme that improves the availability of nutrients in the soil, and second, to act as their carrier in the spore form. "You can picture the surface of some spores as being like a forest. Between the

taller trees, there are some smaller ones embedded and protected. Those are our enzymes, which are surrounded by protective proteins," says Curtis. In this way, the Bayer researchers stabilize bacterial enzymes that make crops more productive. Targeted strain improvement techniques are common practice in other industries such as pharmaceuticals, textile manufacturing, and enzyme production for producing vitamins, laundry detergents, biofuels, and cosmetics.

Once the Bayer scientists and their counterparts at Elemental Enzymes find an interesting protein, determine that it can be carried by a bacterial spore, and that the combination delivers a meaningful biological effect, a lot of work still remains in developing a product. Human and environmental safety is a key priority: all strains undergo a safety evaluation beginning at the laboratory stage, prior to field trials and during the course of development. If the enzymes together with the spores are intended to be used as a seed treatment product, it will be applied externally to the seeds. This seed treatment can also be combined with chemical crop protection agents to ward off pests such as nematodes, insects, and fungi. It is necessary that the seed treatment is formulated in such a way that all of the active substances can reach the plant and ensure the best-possible microenvironment. Combining chemical crop protection agents, proteins, and living organisms such as bacterial spores takes a



Dr. Damian Curtis  
Microbial Genetic Systems  
Team Lead

*"Our objective is to stabilize proteins via targeted strain improvement tools."*

very broad set of scientific skills. "Microorganisms and macromolecules such as enzymes are considerably more sensitive to ambient conditions than chemical products," explains Dr. Milind Singh, Principal Scientist in SeedGrowth Formulation Technology at Bayer.

Years of research go into finding the optimum composition of all formulation ingredients, and being able to deliver a final product that is safe to humans and the environment. For the customer, the most important aspect – besides the efficacy – is the shelf-life of the product. "We know that we currently achieve a minimum stability of 18-24 months with certain biological



Combining chemistry and biology: Dr. Milind Singh is searching for the perfect composition of all formulation ingredients to achieve the best seed treatment.

formulations. For enzyme-based products, being such a new technology, we have not had time to assess longer-term storage stability. So far, we have achieved 12 months in final packaging and stability on-seed, which is substantial," says Singh.

"Through lab, greenhouse and field testing, we ensure that the biological seed treatment remains stable for just as long as the seed itself remains viable," says Dr. Jennifer Riggs, Product Development Manager at Bayer. The scientists then consider the product to be ready for the market.

### Bacterial treatment can give crops a crucial advantage

Over the last 10 years, through testing seed treatments on corn, the right combination of crop protection agents and microbes has resulted in significantly increased yields of up to 10 percent. Similar benefits in yield have been achieved in the production of soybeans, cotton, and cereal grains. The selected bacterial treatment can give the crops a crucial advantage in a critical time window for establishing yield. The use of optimized microbes could be an important step to help feed the growing world population, as the right microbes can make naturally resource-limited land more productive.

"The potential of which soil microbes can contribute to optimizing crop production is nowhere close to being exhausted," says Riggs. The innovative approaches utilized by Bayer's scientists to optimize bacteria are just the next step to bring new products to growers. For researchers, soil remains a fascinating reservoir, packed with tiny microbes with a big impact on sustainable agriculture. ■