A STEP CLOSER TO NEW NITROGEN-FIXING CROPS

The transfer of genes that allow nitrogen to be fixed from the atmosphere into PHOTOSYNTHESIZING CYANOBACTERIA could one day lead to similarly engineered crops.

Plants need nitrogen-rich soils to thrive, but there are a limited number of plants able to extract nitrogen gas from the atmosphere and fix it in a useful form in the soil. However, new kinds of crops capable of nitrogen fixation may soon be possible, after Japanese scientists successfully transferred nitrogen-fixing genes into a photosynthetic organism.

Nitrogen fixation is a process that chemically converts molecular nitrogen to ammonia, a nitrogen compound plants can use, explains Yuichi Fujita, a biochemist at Nagoya University, which is about 300 km west of Tokyo, Japan.

Much of the world's farming is currently supported by nitrogen-containing chemical fertilizers, but they are energy-intensive to produce and can lead to algal blooms when runoff pollutes waterbodies.

Many of the plants capable of nitrogen fixation are legumes, members of the bean family, but it would be game changing to have a wider variety of crops with this capacity, says Fujita.

His research is part of the COI-NEXT project, a bioplastic's project led by Kanazawa University and the Japan Science and Technology Agency. Fujita's research is focussed on the efficient production of crops that could be used as feedstock for bioplastics.

FRESH FIX

Legumes fix nitrogen through nodules that develop on their roots and host nitrogen-fixing bacteria, but conventional plant breeding, or even genetic engineering, hasn't yet succeeded at transferring the ability for nitrogen fixation via symbiosis with bacteria to other crops.

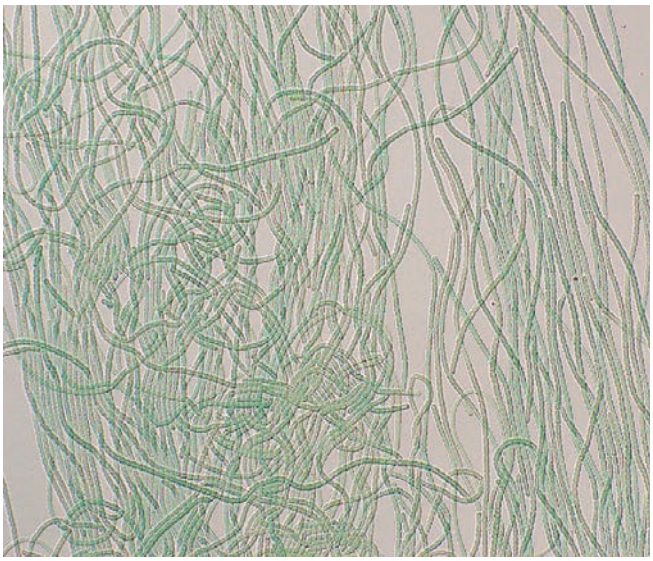
Fujita's team are instead exploring how to genetically engineer this ability, and have started with cyanobacteria, which can draw energy from the sun through photosynthesis, as plants can. Cyanobacteria are microorganisms that are either a single-cell or composed of single connected 'filamentous' cells. These microorganisms bear similarities to chloroplasts, the organelles responsible for photosynthesis within plant cells.

Passing the ability to fix nitrogen on to new crops has been challenging. This is largely because the process is driven by an enzyme called nitrogenase, which breaks down easily in the presence of molecular oxygen, a compound abundant in plant cells and the atmosphere.

"Since plants generate oxygen through photosynthesis, nitrogenase needs to be protected not only from atmospheric oxygen, but also from the oxygen produced within the plant cell," says Fujita.

As a first step, his team has engineered the genes for nitrogen fixation into certain cyanobacteria. This was an easier proposition than plants, because there were already cyanobacteria which produce nitrogenase.

The researchers identified 50 genes that code for nitrogenase and its closely related enzymes and proteins in a nitrogen-fixing cyanobacterium called *Leptolyngbya boryana*. They then introduced some of these genes into the genome of another strain of cyanobacteria which didn't have this ability, *Synechocystis* sp. PCC 6803, which subsequently was able to produce small amounts of the enzyme.



▲ **Nagoya University researchers have engineered nitrogen-fixing genes from a cyanobacterium (*Leptolyngbya boryana*, pictured) that already creates a key enzyme into one that does not.**

FUTURE FIXATIONS

The achievement is exciting, but such small quantities of nitrogenase can't "support nitrogen-fixing growth in cyanobacteria," says Fujita, "so we are trying to introduce other genes to enhance nitrogenase activity."

The team's cyanobacteria success is only a first step, and there are many hurdles to overcome before nitrogenase genes might be engineered into crops such as sugar beet, rice and maize.

Fujita says protecting nitrogenase from oxygen generated in plants cells might require engineering its production to only happen at night, while photosynthesis is paused; or for it to occur in the roots, where photosynthesis does not take place. ■

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