No. B-2E

PJ: Development of Next-Generation CO2-Fixing Plant Through the Gene Optimization, Distant Hybrid, and Microbial Symbiosis

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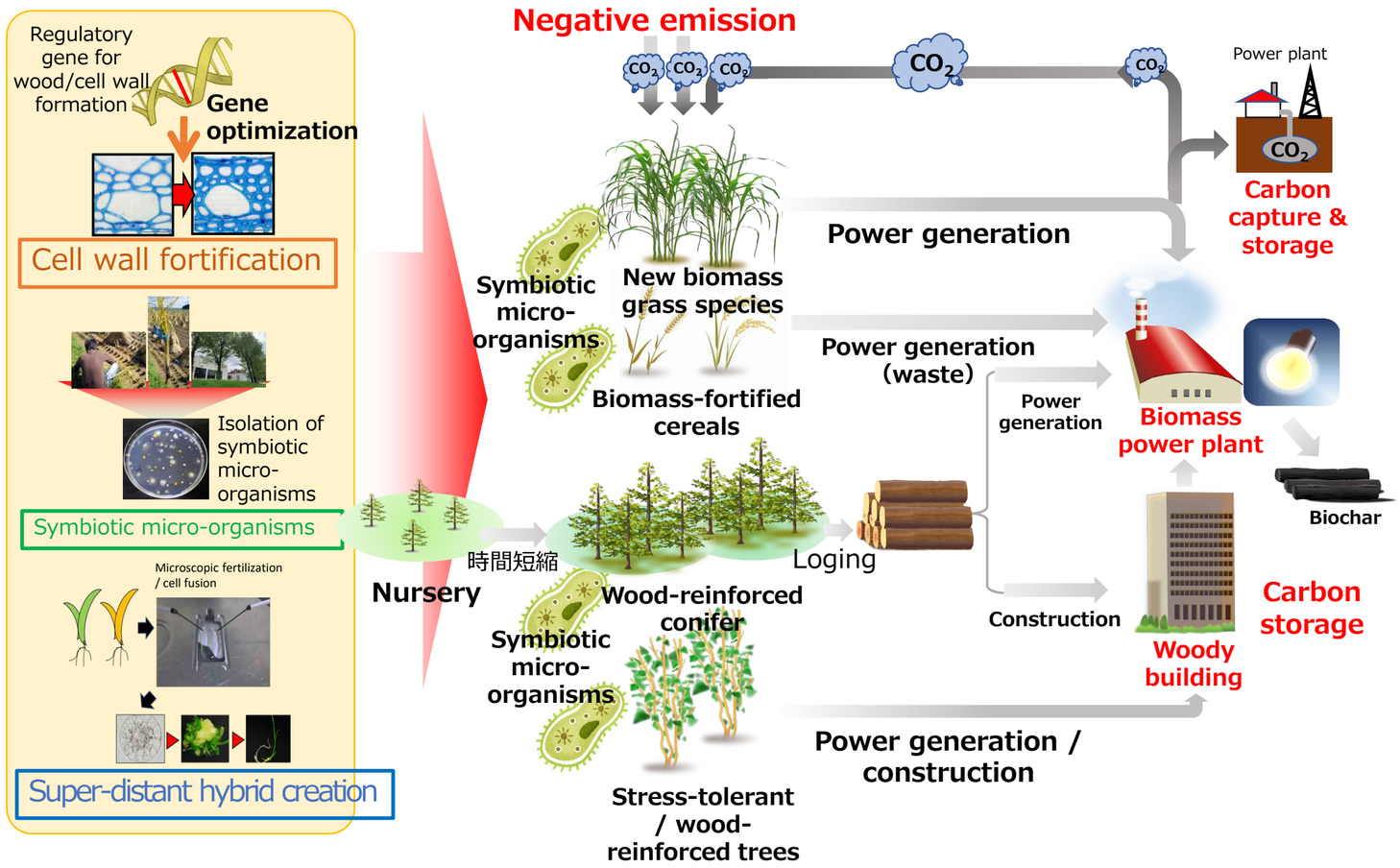
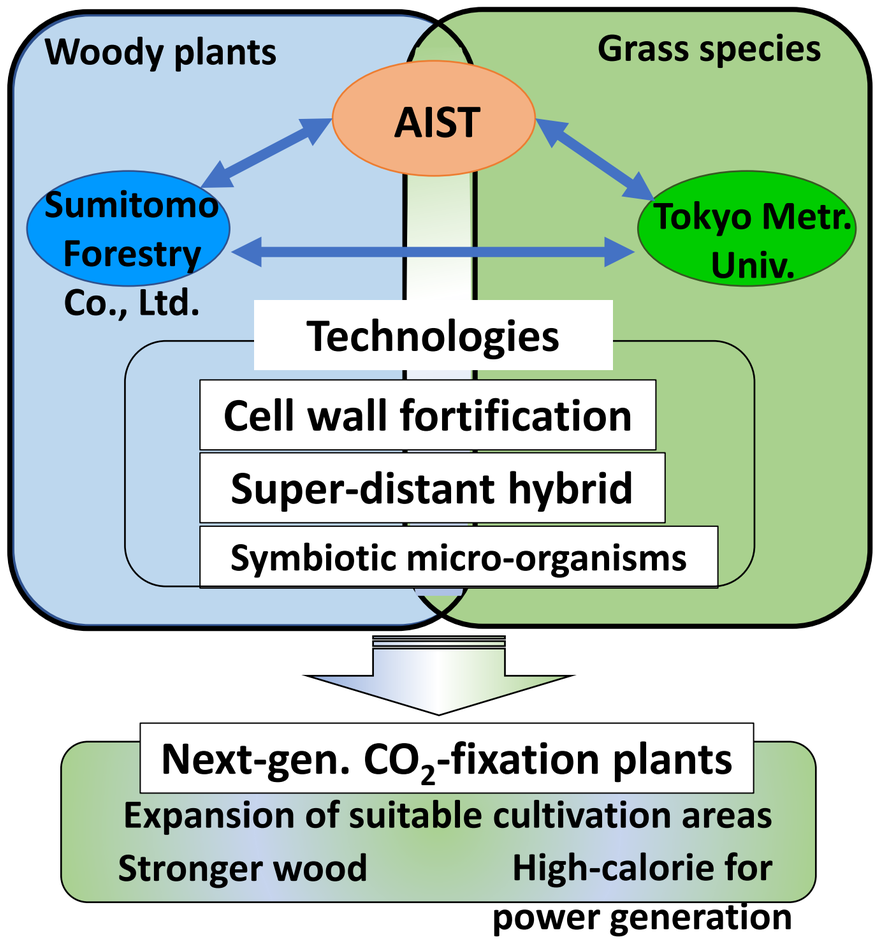
1. Abstract

In order to fix CO_2 at low cost and on a large scale, the development of plants (and technologies to support their growth) that can be grown in a wide area, fix more CO_2, and produce higher-performance products (woody biomass) is desired. In this research project, we are going to create next-generation CO_2–fixation woody plants and grass species with enhanced CO_2 absorbing capacity. For the development in woody plants, we will mainly apply gene optimization (genome editing) technology to enhance wood formation. In grass species, we will employ super-distant hybrid creation and the gene optimization technologies. In both cases, optimization of symbiotic micro-organisms will be applied in the early growth stage. By combining these elemental technologies, we will establish next-generation CO_2 fixation plants and their utilization strategies.

2. Graphical abstract



3. Team building

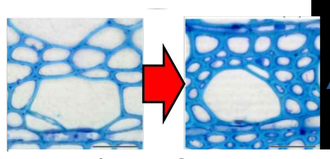


4. Three major technologies

4 – 1. Gene optimizatio

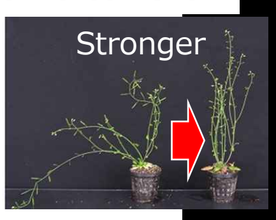
◆**Three strategies**
Based on gene edit technology

1. Wood reinforcement by NST hyperactivation

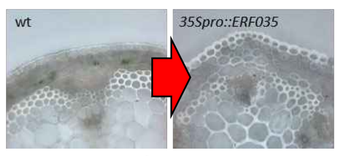


Wood reinforcement

2. Increased strength
   - ➢ Beneficial trait in addition to higher wood productivity



3. Primary cell wall enhancement (only for grass)



PCW enhancement

NST transcription factor

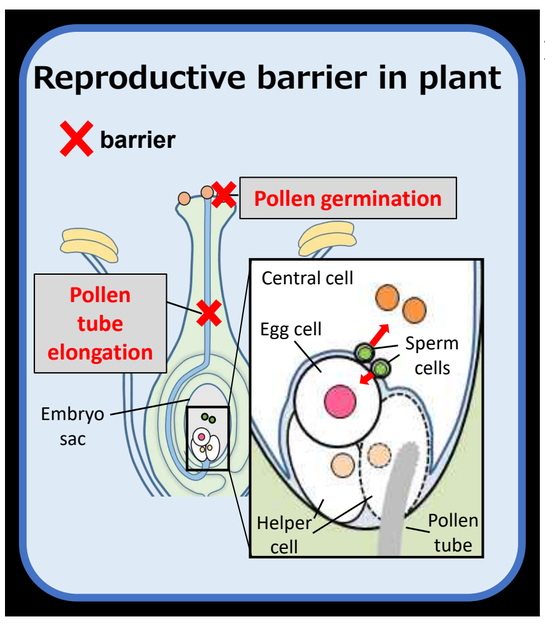
Master regulator of wood / secondary cell wall formation. Loss of function lost wood formation. Gain of function can fortify wood formation

WT NST knockout

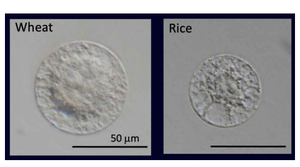
Takata et al., *Tree Physiol.* **39**, 514-525 (2019)

4 – 2. Super-distant hybrid

◆**Super-distant hybrid creation technology**
Technology to overcome reproductive barrier between different species through "microinsemination" or "cell fusion".

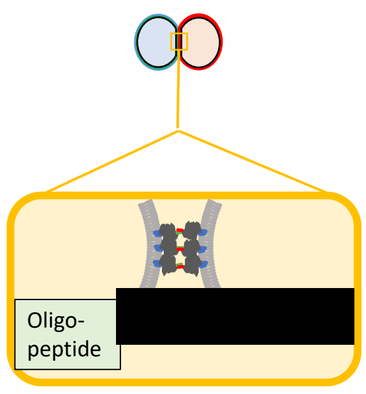


1. Microinseminatoin 2. Promotion of cell fusion



Gamate isolation

Electric gamate fusion



Promotion of cell-cell adhesion and fusion by peptide-PEG lipids

➢ Towards higher biomass production and/or expansion of suitable cultivation areas

4 – 3. Symbiotic micro-organisms

◆**Exploration of symbiotic micro-organisms**