

Background

Rice sheath blight is a typical rice disease caused by *Rhizoctonia solani*, which is one of the three main diseases of rice in China.

- Difficult to detect through visual observation, so early detection is important.
- The existing detection methods are not suitable for farmers, so convenient field detection methods are necessary.
- At present, the treatments of rice sheath blight have certain limitations.

Design

In our blueprint, our comprehensive control system includes three steps: early detection of rice sheath blight, transmission blocking of *Rhizoctonia solani*, and biological control of *Rhizoctonia solani*.

- Step 1: early detection of rice sheath blight:
 - E-nose system -- routine field detection;
 - LAMP system -- accurate detection.
- Step 2: transmission blocking of *Rhizoctonia solani*:
 - Acrylic acid grafted starch -- wrap *Trichoderma atroviride* to aggregate them at the aquatic interface of rice.
 - The engineered *T. atroviride* --
 - overexpress *epi1* (encodes surface hydrophobic protein EPL1)
 - overexpress *prb1* (encodes protease PRB1)
 - express *sn1* (encodes antimicrobial peptide SN1);
- Step 3: biological control of *Rhizoctonia solani*:
 - RNAi -- interfere with the corresponding genes of key proteins PG and PMK1 in the infection of *R. solani*.
 - *E. coli* HT115(DE3) -- as a bacterial factory to produce a large number of RNAi small molecules.
 - R-body protein -- lysis the bacteria and release shRNA molecules.
 - Clay nanosheets -- wrap shRNA molecules to improve their stability, and they are supposed to be sprayed in field.
 - Considering safety, in order to prevent *Trichoderma atroviride* from escaping into the environment and causing adverse effects, we knocked out the light response gene *hda-2*, so that *Trichoderma atroviride* could not produce spores.



1. Detection of rice sheath blight



2. Transmission blocking of *R. solani*



3. Biological control of *R. solani*

Model

- Based on multiple regression analysis, the optimum concentration ratio for co-culture of *Trichoderma atroviride* and *Rhizoctonia solani* was predicted.
- Cellular automata was used to simulate the infection process of *Rhizoctonia solani* in rice.
- We designed a dynamical model to understand the inhibitory effect of RNAi on *Rhizoctonia solani*.
- We developed an E-nose system which was built based on cloud computing platform and constructed a combinatorial classifier to predict the condition of rice infection.

Outlook

Under the support of the national policy of "agriculture, rural areas and farmers" and agricultural modernization, we expect to combine our products with more modern agricultural technologies as part of modern agriculture so as to achieve intelligent and standardized agriculture. In the future, we want to apply the product to the rice blast and rice smut, which are also one of the three major rice diseases in China, and even expand its application to the detection and treatment of more hydroponic plant fungal diseases. Furthermore, we hope the mode of our project will be utilized in multiple application scenarios and make more contribution to agriculture.

Inspiration

What inspired us:

- China's Policy of "Agriculture, Countryside, and Farmer" and the trend of agricultural modernization.
- Several widely-used methods for detection and treatment of plant fungal diseases.
- Rice is the main food crop of our country while rice sheath blight is a severe problem in the rice planting industry.
- Information provided by rice growers: traditional methods are unable to fundamentally cure rice sheath blight, and if rice sheath blight is not effectively treated, the crop will be halved or even not harvested.

Human practices

When brainstorming in the early stage of the project, our team members learned about the status quo as well as its severity and the lack of effective remediation methods of rice sheath blight from our relatives who grew rice. Before starting to design our project, we conducted field research and interaction with relevant people in the rice farming. We learned the relevant knowledge of rice, saw the needs of farmers and public concerns through the research, and confirmed the feasibility of the project.

During the design and implementation of the project experiment, we communicated with many professors and postdocs to obtain suggestions on plasmid construction, rice planting, the conduct of experiment and so on. At the same time, we periodically feed back our progress to stakeholders, take their suggestions as an important basis for project improvement and iteration, and timely carry out experimental improvement and product improvement to make our products more user-friendly.

Finally, we plan to popularize rice sheath blight and our products to more rice growers, as well as establish a real-time communication platform with farmers as main users. In this process, we will obtain more feedback and conduct improvements. Simultaneously, we are looking forward to finding the potential of expanding the application of our products.

Hardware & Software

- LAMP system:
 - Store the reagents required by LAMP system in the form of dry powder, and add a certain amount of water when conducting LAMP amplification reaction;
 - Fluorochrome or lateral flow device (LFD) were used to detect whether the infected rice crop contains ITS sequence of *R. solani*.
- E-nose system:
 - Consists of hardware and software;
 - Monitor the volatile organic compounds (VOC) and growth environment of rice crops;
 - Feedback to users in APP.
- The spray for transmission blocking and biological control of *R. solani*:
 - *Trichoderma atroviride* preparations and shRNA preparations are manufactured into spray respectively.
 - Sprayed by UAVs in the infected field.

Highlight

- E-nose and LAMP-LFD devices have typical modern agricultural technology characteristics, realizing intelligent and standardized prevention and detection of rice sheath blight.
- The engineered *Trichoderma atroviride* wrapped in hydrophobic materials are capable of gathering at the aquatic interface of rice, effectively blocking the transmission of *Rhizoctonia solani*.
- We created a series of RNAi molecules combined with nanomaterials that are able to be stored for a longer time and have a more efficient delivery efficiency.