Generation & Deployment of Genome-Edited, Nitrogen-use-Efficient Rice Varieties

OBJECTIFS

Project objectives:

1. Proof of concept for GE-assisted variety development for a complex trait of agronomic interest such as NUE
2. Analyze the societal and institutional feasibility (regulations, environmental and social risks, impacts) of the deployment of GE-assisted varieties for a food crop such as rice, in the context of the emergence of regulatory structures for biosecurity in a developing country, Madagascar
3. Strengthen the collective capacities through information exchange and training of Malagasy biosafety regulation bodies and other stakeholders to assess the risks and opportunities associated with the deployment of new crop varieties improved through GE technology and to make decision

Research questions

1. Optimize CRISPR/Cas9 technology in rice for simultaneous KO and base substitution, including the investigation of a non-integrative approaches,
2. Establish relationships between our main target gene BT2/OsBT, plant N status, nitrate transporter genes such as NRT1.1/OsNRT1.1Bs, OsCCA1& OsELF4 clock genes, and TGA transcription factors between Arabidopsis and rice plant models,
3. Decipher the eco-physiological components associated with NUE for improved rice varieties developed with GE,
4. Determine the conditions for the social acceptability of the proposed GE-new varieties with regards to potential socio-economic risks and impacts for farmers and other stakeholders of the local rice value chain,
5. Address the needs for capacity building or institutional adaptations to implement national biosafety legislation.

ACTIONS

Development of rice lines improved for NUE using genome editing CRISPR / Cas9 of rice varieties with better NUE

- Preparation of the next generation of methods and target genes for GE-assisted improvement of rice for NUE.
- Participatory analysis of institutional impacts to characterize, assess and manage the social and environmental risks linked to the dissemination of genome-edited rice varieties in Madagascar
- Agronomic evaluation and ecophysiological characterization of the performance of GE-assisted rice lines
- I377Identification and implementation of training actions (students' thesis, professional and academic training, participatory workshops) to strengthen the research and decision-making capacities of stakeholders

RESULTATS

Main results

On the biological level

1. Demonstration of the feasibility to use genome editing technology for developing rice varieties with improved NUE

- Feasibility of the evaluation of CRISPR-CAS9 constructs in rice by transformation of protoplasts and regeneration of fertile plants, which could be multiplied and tested under laboratory and field controlled conditions for their NUE potential;
- Development of a system able to produce mutations by deletions in rice;
- Feasibility of target insertion/deletion of few bases in rice genome leading to target gene knock-out (KO) by frameshift introduction or single base substitutions (base edited);
- Demonstration at field level in Colombia (CIAT) that some of the base edited lines obtained from an upland rice cultivar Chhomrong Dhan (CHD) have a positive effect on NUE and yield: increases in yield were of about 15% in both low N and high N fertilization treatments (Figure 1).

2. New knowledge on the regulation pathways for NO3 transport and uptake:

- In Arabidopsis thaliana, BTs genes are not involved in the NRT1.1 signaling pathway for the regulation of NRT2 by nitrates, but results obtained in 2019 and 2020 show that HRS1 and HHO1 genes are part of this signaling pathway and involved in the repression by nitrate of NRT2s and root nitrate uptake;
- preliminary results indicate that, unlike what we observed in Arabidopsis, the repression by nitrate of NRT2s does not exist in rice and thus the mutation of NRT1.1B has no impact;
- Overexpression of BT reduces plant growth and NUE under limiting N while overexpression of CCA1 reduces growth and NUE under non-limiting N (need confirmation with on-going experiment).

On the socio-economic level :

- The sociotechnical transition model (Greels and Schot, 2007) constitutes a relevant theoretical framework for the analysis of the determinants of varietal innovation based on genome editing;
- the project actions generated a collective improvement in the capacity of the different stakeholders to understand the conditions of use of GE varieties;
- the project has allowed to structure interactions between civil society, public authorities and research in the joint development of a methodological framework including the participation of stakeholders in the context of improving public regulatory systems;
- A main recommendation concerns the need to involve stakeholders in the risk assessment methods that must compare the benefits expected from GE varieties with other possible variety and agronomic options usable by farmers;