## New study shows CRISPR can be applied to produce biofortified rice

A team of California plant scientists has taken a CRISPR-Cas9 approach to develop more nutritious varieties of rice.

Their research, published today in <u>Nature Communications</u>, demonstrated that CRISPR-Cas9 can be successfully used to biofortify rice with carotenoid, a precursor to the essential nutrient vitamin A. Previous research has demonstrated that biofortification of rice can help <u>prevent blindness</u>, <u>weakened immune</u> <u>systems and other health problems</u> associated with vitamin A deficiency, which is especially prevalent among children in developing nations.

The study was led by Prof. Pamela Ronald and Oliver Dong, a post-doctoral fellow in the Ronald laboratory at the University of California, Davis, and members of the <u>Innovative Genomics Institute</u> (IGI) in Berkeley. Other collaborators included scientists from the Department of Energy Joint Genome Institute and the Joint Bioenergy Institute, both in Northern California.

The research team used CRISPR-Cas9 to insert marker-free DNA fragments, or cassettes, into the rice genome at two targeted locations. Though other CRISPR researchers had previously achieved targeted insertions in plants, they used relatively small fragments of DNA, which restricted the amount of genetic information that could be introduced into the genome. The Ronald team inserted a 5.2 kb cassette that was more than twice the size of previous similar targeted insertions, resulting in carotenoid-enriched rice with a golden-colored grain.

Previous biofortification work in rice relied on conventional agrobacterium- or particle bombardment-based plant transformation, which integrates transgenes at random locations in the plant genome. This can disrupt gene function, sometimes resulting in reduced yield.

In this new approach, Ronald and her team identified genomic "safe harbors" that could accommodate insertion of the carotenoid cassette without disrupting desirable agronomic traits. They also demonstrated the lack of off-target mutations in the carotenoid- enriched plants, as evidenced by whole-genome sequencing.

The results suggest that CRISPR-Cas9 genome editing offers a promising strategy for making genetic improvements in rice — a staple food for more than half the world's population — and other crops. The study also suggests that targeted gene insertion could facilitate stacking multiple genes with desired traits in a specific location in the genome — a process that is currently challenging using conventional plant breeding.

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