

BioResource Now!

Issue Number 6 January 2010



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Research and Bioresources No.8
Ongoing Column No.47

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Molecular Mechanism of Flood-adaptation in Rice



Some regions in South-east Asia, West Africa, and the Amazon River basin are prone to yearly, long-term, catastrophic flooding in the rainy season. Most plants do not grow under harsh conditions such as floods. Plants are immobile and thus are obliged to acquire novel characteristics in order to adapt to and endure severe environmental conditions. Floating rice, which grows in Southeast Asia, West Africa, and the Amazon River basin, has adapted to severe environmental conditions such as floods.

Common rice (approximately 1 m in height) may die if it is submerged under water. The height of floating rice is also approximately 1 m in shallow water; however, its stem elongates (internodal elongation) when the water level rapidly rises during a flood. As a result, the leaves stick out of the water surface and enable respiration. Therefore, stem elongation prevents the plant from being submerged under water and enables it to survive a flood (the rice stem is hollow and it functions like a snorkel for scuba diving)[Fig. 1].

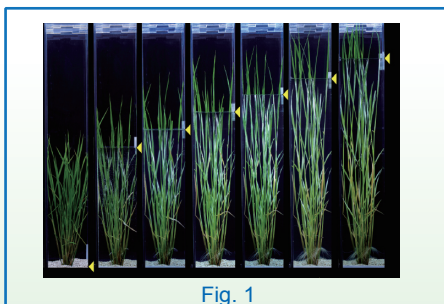


Fig. 1

The flood-tolerance capacity of floating rice, *i.e.*, its capacity for internodal elongation under deep water, is astonishing. This rice strain is known to rapidly elongate its stem in response to a rise in water level [Fig. 2]. Some floating rice strains can survive under water that is approximately 10 m deep.

In this manner, floating rice has evolved a strategy to ensure respiration under water and to overcome flooding. Few plants have been known to undergo drastic changes in size as floating rice does in response to a rapid environmental change. We aimed to elucidate the water-level-dependent internodal elongation mechanism of floating rice by identifying the genes that are responsible for this trait.

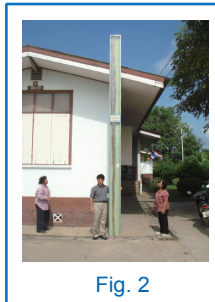


Fig. 2

As a first step to isolate the floating rice genes, we conducted a genetic analysis and identified that gene regions on chromosomes 1, 3, and 12 facilitate internodal elongation in response to a rise in water level. We then developed near-isogenic lines for each of the 3 identified gene regions and tested their internodal elongation capacity. All the newly developed strains exhibited the internodal elongation capacity; this finding confirmed that the 3 gene regions were indeed responsible for internodal elongation. In addition, we found that the gene region located on chromosome 12 showed the strongest effect on the internodal elongation among the 3 identified gene regions; therefore, we performed gene mapping of the target region on chromosome 12. We identified 2 genes, *Snorkel 1* and *Snorkel 2*, which are not expressed under normal cultivation conditions (*i.e.*, under shallow water), but their expression is significantly increased under a deep water condition. We also found that these genes specifically exist in floating rice but not in non-floating rice (cultivated rice), such as T65 and Nipponbare.



We introduced these genes into the T65, a non-floating rice strain, and found that the newly developed transgenic strain exhibited the internodal elongation capacity under a deep water condition. Furthermore, over-expression of the *Snorkel* genes in non-floating rice strains induced their internodal elongation even under a shallow water condition. Therefore, we concluded that *Snorkel 1* and *Snorkel 2* confer the floating capacity on rice.



Gene structure analysis of the isolated *Snorkel* gene revealed a nuclear transport signal and an AP2/ERF domain in the gene.

The genes harboring AP2/ERF domains are known to encode a transcription factor. On the basis of this information and the information regarding the amino acid sequence and its numbers, we presumed that the *Snorkel* gene encodes an ERF-type transcription factor (ethylene response factors; ERFs). This was also confirmed by a hormone treatment experiment in which the rice was treated with plant hormones, such as ethylene, gibberellins, abscisic acid, auxin, or cytokinin. We found that *Snorkel* expression was significantly induced only under ethylene treatment. We performed a binding assay to confirm the binding between EIN3 (ethylene insensitive 3), which is a factor in the ethylene signal transduction pathway that binds to the promoter of ERFs, and the promoter region of *Snorkel*. The results of this study clearly indicated that *Snorkel* was a factor involved in ethylene signal transduction.

It is considered that the widely consumed cultivated rice strain was domesticated from a certain wild rice strain, and has been cultivated for approximately 8,000 years. We analyzed the genes of this wild rice strain and found that it harbors both *Snorkel 1* and *Snorkel 2*. This finding implied that the wild rice strain was flood-tolerant.



The rice strains cultivated in the regions that experience frequent floods retain the floating capability, whereas most of the rice strains cultivated in other regions do not. We presume that it was unnecessary for the rice strains that were cultivated in the regions with infrequent floods to retain the *Snorkel* genes. Additionally, the rice strains harboring the genes become tall in the harvesting season even under the normal cultivation conditions and thus tend to fall over during heavy winds and unfavorable weather. Therefore, this phenotype might have been selectively eliminated in the past because it rendered the plants inconvenient for cultivation.



The water level in the river deltas of South-east Asia rises by several meters due to yearly flooding in the rainy season. Therefore, the people in this region cultivate floating rice during the floods because it is the only cereal that can be harvested in this season. However, the yield of floating rice is known to be extremely low. The development of a high-yielding, floating rice strain is hindered by the lack of progression in research aimed at improving flood-tolerance in rice. In conclusion, we elucidated that 3 chromosomal regions regulate the floating capacity of the floating rice. We also succeeded in introducing these 3 chromosome fragments into Japanese cultivars by hybridization.

As a result, the Japanese conventional cultivated rice started to exhibit stem elongation under a deep water condition. This achievement demonstrated that a normal rice strain could be transformed into a floating rice strain by introducing these 3 chromosome fragments into the original strain. Further, we have identified the genes responsible for high yield. Therefore we believe that an improved floating rice strain can be developed by introducing the genes that confer the floating capacity and a high-yield trait into the normal rice strain. In fact, we are currently developing a rice strain that is adapted to the flood-prone regions in South-east Asia. ■



Japanese-language Input Support Abroad

When we travel abroad on business or to attend conferences, we would occasionally like to send an email in Japanese or search the Internet by using keywords in Japanese. This will not be an issue if we have our own laptops at hand; however, it will be an issue if we only have access to computers in which only English alphabets can be input. What would you do if you find yourself in such a situation? Would you make an effort to type in English alphabets to describe Japanese words such as "Konkai ha Zenjutu no Joukyo de..." ?



In this issue, I would like to introduce 2 applications, Ajax IME and the handwriting recognition tool in Ajax, which are useful in the aforementioned situation. Ajax IME is a Web-based Japanese-language input support tool. The handwriting recognition tool in Ajax is an application that searches and inputs Kanjis that we know how to write but do not know how to pronounce. Please try these applications.

How to use Ajax IME

Ajax IME is compatible with major browsers such as Internet Explorer, Firefox, Safari, and Chrome; however, the compatibility view mode should be turned on in Internet Explorer 8.

- 01 Open the website, <http://ajaxime.chasen.org/>
- 02 Japanese-language input function can be turned on or off by pressing "Alt + o," "Ctrl + 9," or the "IME On/Off" button (Fig. 1).



Fig. 1. An example of converting "にほんご," Hiragana, to "日本語," Kanji.

- 03 When the form color changes from white to blue, type the desired Japanese words using English alphabets.
- 04 A conversion list containing Kanji will appear when you press the space bar.
- 05 Press the enter key or start typing the next word to confirm the desired Kanji.

How to use the handwriting recognition tool in Ajax



Figure: An example of handwriting recognition in Ajax showing a Kanji, "海," written using a mouse.

- 01 Open the website, <http://chasen.org/~taku/software/ajax/hwr/>
- 02 Draw a Kanji character inside the blue square by using a mouse. A list of candidate Kanji characters will then appear on the right.
- 03 Select the character of your choice. This character will be displayed on the form underneath.

(Gaku KIMURA)

Announcements (Details are available at <http://www.nbrp.jp/>)

The 3rd Rat Resource Research Workshop

Date and Time: January 29 (Fri) 13:00 - 17:30, 2010
Place: International Conference Hall I, Clock Tower Centennial Hall, Kyoto University

International Bioresource Symposium on *Drosophila*

Date: March 17(Wed), and March 18(Thu), 2010
Place: Enryakuji Kaikan Hotel, Enryakuji, Hieizan



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(The next month's issue will be "Hot News from Abroad.")

BioResource Information

- (NBRP) www.nbrp.jp/
- (SHIGEN) www.shigen.nig.ac.jp/
- (WGR) www.shigen.nig.ac.jp/wgr/
- (JGR) www.shigen.nig.ac.jp/wgr/jgr/jgrUrlList.jsp

Editor's Note

Did you like the story about the discovery of the Snorkel genes? This research attracted worldwide attention after its publication in *Nature* 460, 1026–1030 (2009). BBC News introduced the research results under the subject title "Snorkel rice could feed millions!" together with an animation on the website, <http://news.bbc.co.uk/2/hi/8208411.stm> (a commercial is occasionally broadcasted before the animation). Dr. Ashikari conducted sophisticated research that utilized genetic resources such as wild rice strains and chromosome segment substitution lines. This year, we will be introducing more interesting topics in the newly-designed "BioResource Now!" .(Y.Y.)