

# Marker assisted cold tolerance breeding in rice

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## Abstract

Spikelet fertility of rice decreases when rice plants are exposed to a low temperature, especially at the booting stage, due to the failure of microspore development. The sterile type of cold injury is a very serious problem at high latitudes and in uplands at low latitudes. Rice breeders have been making efforts to develop more cold-tolerant cultivars. In 1970's, two tropical *japonicas*, Silewah and Padi Labou Alumbis, were found to be cold-tolerant. Their cold tolerance was introduced into Japanese breeding lines by backcross breeding, and cold-tolerant cultivars, Norin-PL8 and Hokkai-PL9, were developed. Although Norin-PL8 and Hokkai-PL9 are more cold-tolerant than conventional cold-tolerant cultivars, they have unfavorable traits, such as long duration to heading and long culm. Despite continuous efforts to improve their agronomical traits, no commercial cultivar has been established by introducing cold tolerance of the two cultivars. We analyzed QTLs for cold tolerance of Norin-PL8 and Hokkai-PL9. The QTL for cold tolerance of Norin-PL8 was detected on the long arm of chromosome 4. We mapped the QTL using NILs and identified two closely linked QTLs, *Ctb1* and *Ctb2*. The QTL for cold tolerance of Hokkai-PL9 was detected on the short arm of chromosome 8 using F<sub>2</sub> and F<sub>7</sub> progenies between Hokkai-PL9 and a cold-sensitive cultivar, Hokkai287. We are introducing the QTLs into commercial cultivars using the DNA markers linked with the QTLs.

## Media summary

DNA markers associated with cold tolerance will be useful tools for the cold tolerance breeding of rice.

## Key Words

QTL, cold tolerance, DNA marker, rice

## Introduction

Rice is a cold-sensitive plant that has its origin in tropical or sub-tropical areas. Spikelet fertility decreases significantly by a low temperature at the booting stage (Satake and Hayase 1970). The sterile type of cold injury is a very serious problem both at high latitude (e.g., northern part of Japan) and in uplands at low

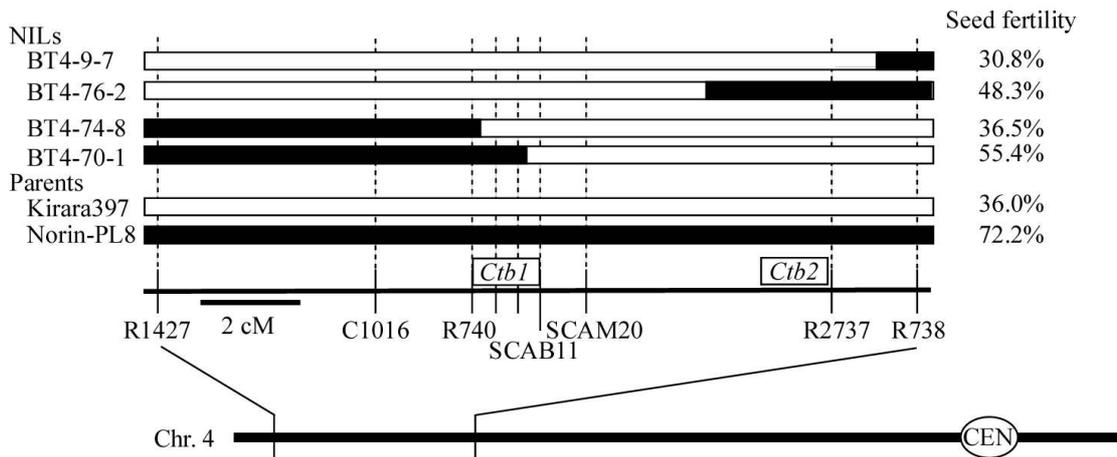
latitudes (e.g., Yunnan Province in China) (Dai et al. 2004). From 1974 to 1977, IRRI selected several cold tolerant varieties from 17,689 accessions in its germplasm bank. Satake and Toriyama (1979) tested their cold tolerance at the booting stage and showed that tropical *japonicas*, Silewah and Padi Labou Alumbis, are cold-tolerant. Their cold tolerance was introduced into Japanese breeding lines by backcross breeding, and cold tolerant cultivars, Norin-PL8 and Hokkai-PL9, were developed (Abe et al. 1989). In this study, we mapped the QTL for cold tolerance of Norin-PL8 and Hokkai-PL9. We developed DNA markers that can be used in cold tolerance breeding.

## Methods

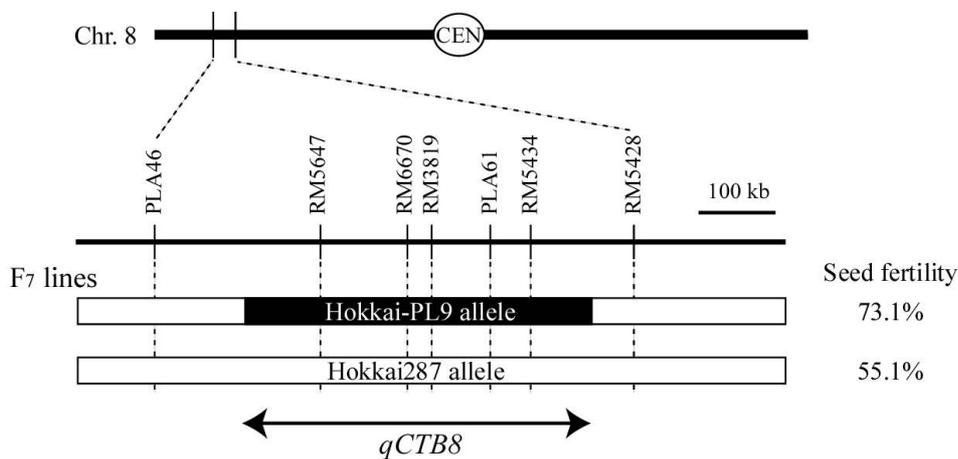
BC<sub>1</sub>F<sub>5</sub> lines between Norin-PL8 and a cold-sensitive cultivar, Kiara397, were used for the identification of the QTL for cold tolerance of Norin-PL8. Advanced backcrossed progenies developed by crossing Kirara397 recurrently were used to map the QTL. F<sub>2</sub> and F<sub>7</sub> populations between Hokkai-PL9 and a cold-sensitive cultivar, Hokkai287, were used for the identification of the QTL for cold tolerance of Hokkai-PL9. F<sub>7</sub> progenies derived from an F<sub>6</sub> plant that is heterozygous for the identified QTL was used to confirm the presence of the QTL. Cold tolerance was evaluated by the cool water irrigation method (Futsuhara and Toriyama 1964).

## Results

Ninety-two BC<sub>1</sub>F<sub>5</sub> lines between Norin-PL8 and Kirara397 were tested for cold tolerance and RFLP marker genotypes. The markers on the long arm of chromosome 4 were significantly associated with cold tolerance, indicating that the QTL for cold tolerance is located in the region. In order to map the QTL, one of the BC<sub>1</sub>F<sub>5</sub> lines was backcrossed 3 times to Kirara397, recombinants in the QTL region were selected, and NILs were developed. Marker genotypes and cold tolerance of the NILs indicated that there are at least 2 QTLs for cold tolerance, *Ctb1* and *Ctb2*, on the long arm of chromosome 4 (Figure 1). Fifty-nine F<sub>7</sub> lines between Hokkai-PL9 and Hokkai287 were tested for cold tolerance and SSR marker genotypes. The markers on the short arm of chromosome 8 were significantly associated with cold tolerance, indicating that the QTL for cold tolerance is located in the region. Interval mapping using 288 F<sub>2</sub> plants showed that the phenotypic variance explained (PVE) and additive effect (AE) of the QTL were 26.6% and 11.4%, respectively. We found an F<sub>6</sub> plant that is heterozygous for the QTL region and developed an F<sub>7</sub> population. Cold tolerance segregated in the F<sub>7</sub> population and the markers in the region were associated with cold tolerance significantly (Figure 2).



**Figure 1. Chromosomal location of the QTLs, *Ctb1* and *Ctb2*.**



**Figure 2. Chromosomal location of the QTL, *qCTB8*.**

## Conclusion

We identified 2 QTLs for cold tolerance of Norin-PL8 on the long arm of chromosome 4 and a QTL for cold tolerance of Hokka-PL9 on the short arm of chromosome 8. We are introducing the QTLs into commercial cultivars by using DNA markers.

## References

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