ISSN (Online) 2287-3406
Journal of Life Science 2020 Vol. 30. No. 10. 844~850 DOI : https://doi.org/10.5352/JLS.2020.30.10.844

QTL Analysis of Rice Heading-related Genes Using Cheongcheong/Nagdong Doubled Haploid Genetic Map

Yoon-Hee Jang, Jae-Ryoung Park and Kyung-Min Kim*

School of Applied Biosciences, College of Agriculture and Life Science, Kyungpook National University, Daegu 41566, Korea

Received June 26, 2020 / Revised September 11, 2020 / Accepted October 6, 2020

Disaster-related extreme weather is rapidly increasing due to climate change. In Korea, typhoons accompanied by rainfall usually approach in August and September, causing great damage. The purpose of this study is to find a gene that regulates the heading date of rice in order to avoid loss of harvest from climate change and typhoons. Cheongcheong/Nagdong doubled haploid (CNDH) was used as the plant material to investigate the location of heading-related genes using QTL and sequence analysis by cloning the gene. In the distribution chart, the heading dates, culm lengths, panicle lengths, numbers of panicles, and 1,000-grain weights all have normal distributions. QTL analysis found 13 contigs on chromosome 8. One QTL, named qHd8, was detected on chromosome 8. The range at qHd8 was approximately 7.7 cM, with RM72 and RM404 markers near the peak. There were 13 contigs and 1 ORF. Protein sequence analysis showed that rice was similar to Os08g0341700, AtSFH13, and AtSFH7 proteins. Os08g0341700, which is involved in signal transduction, is similar to phosphatidylinositol transfer-like protein II, and complete information is not available, but it is believed to play a role in the phosphatidylinositol-specific signaling pathway related to Sec14P.

Key words : Heading date, QTL, rice, sequence analysis

Introduction

Rice (*Oryza sativa* L.) is one of the most important food crops in which more than half of the world's population is using stocks [4]. Recently, global warming and damage to typhoons have caused harvesting losses. If typhoon damage occurs during the flowering period of rice, it is highly likely that the ear will dry out or become discolored and infertile. This greatly affects the rice yield [7]. The heading is one of the important traits for rice to adapt to the cultivation area and season [11]. The Hd1 gene delays the heading of rice under long-day conditions. Ehd1, a rice-specific flowering regulator, plays an important role in the control pathway of rice heading [6]. A rice gene, OsPIPK1, encoding a 792-aa putative phosphatidylinositol 4-phosphate 5-kinase (PIPK) is involved in rice heading through regulation of floral induction genes, signaling and metabolic pathways. Homozygous transgenic plants showed 7-14 earlier heading than control plants, suggesting that OsPIPK1 negatively regulates floral initiation using antisense transgenic strategy to suppress the expression of OsPIPK1 [9]. Cheongcheong/ Nagdong double haploid (CNDH) is 120 strains of DH that were cultivated by anther culture in the Cheongcheong/ Nagdong F1 combination [3]. There is a CNDH genetic map with an average interval of 10.6cM between markers made using Mapmaker version 3.0 using 222 DNA markers [8]. Also, using CNDH as a material, many QTL studies such as quantity, white-backed plant hopper resistance, amylose content, and protein content have been conducted [2, 3]. In this study, we aimed to find the genes that pulled out the head to avoid the loss of crop yields due to typhoon and global warming. To find out the gene, QTL analysis was used. QTL analysis is used to predict the association between quantitative traits and marker alleles segregating in the population. Most QTL analyses in plants involved populations derived from pure lines [5, 9]. To find the gene location related to heading date QTL was performed [1]. And cloning was performed by selecting gene in the region with LOD value of 3.0 or more. Sequencing alignment is then performed to identify the genes and the results are reported.

***Corresponding author**
Tel : +82-53-950-5711, Fax : +82-53-958-6880
E-mail : kkm@knu.ac.kr
This is an Open-Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/3.0) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Materials and Methods

Plant material and field experiment

The Cheongcheong/Nagdong doubled haploid group (CNDH) used for genetic mapping was developed by anther culture in the F1 combination, which resulted from crossing between 'Cheongcheong' and 'Nagdong'. Cheongcheong is a Tongil type rice variety with tolerance to brown plant hopper and high yield, derived from *Oryza nivara*, whereas Nagdong has a partial shattering layer in the pedicel and has been cultivated for more than 20 years. The CNDH group has been cultivated in paddy fields for three years since it was first developed in 2010. The present study is carried out at Kyungpook National University experimental field at Gunwi. The growth characteristics survey was carried out at Kyungpook National University experimental lab. The test material was sowed on April 29, and the planting density was 30×15 cm at one row per pedigree and one plant per hill, and the plant was transplanted from May 27 to May 28. The fertilization rate of $N\text{-}P_2O_5\text{-}K_2O$ was 9.0-4.5-5.7 kg/10 a. Other herbicides and insecticide spraying and disinfestation disinfection and pavement management followed the Rural Development Administration standard method. Observations were made from July to September in order to investigate the test material heading date.

Genetic mapping and QTL analysis

WinQTLcart 2.5 was used to analyze the QTL and genetic map with an average interval of 10.6 cM between markers created using Mapmaker version 3.0 using 222 DNA markers in the Molecular Breeding laboratory (Fig. 1). The program requires several factors such as the genetic distance between each marker, the name of every marker, the number of chromosomes, the genotype data, and the value of the target trait. Composite interval mapping (CIM) method was used, and LOD was set to 3.0 to perform QTL analysis.

Gene information analysis and gene search

The study used WinQTLcart version 2.5 to perform QTL analysis and used several factors such as marker name, chromosome number, genotype data, and target trait values. After locating the Marker with an LOD value of 3.0 or higher, we identified the genes that existed among the markers.

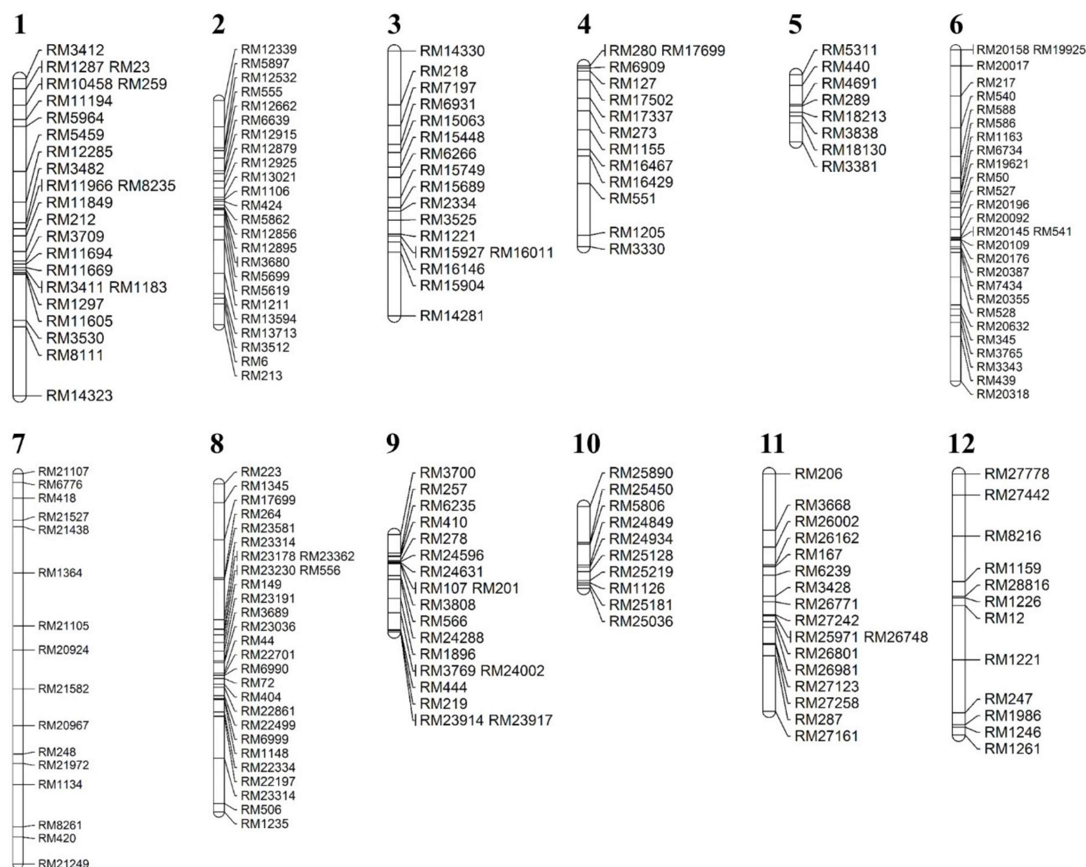

Fig. 1. Genetic map of CNDH. Genetic map with an average interval of 10.6 cM between markers created using Mapmaker version 3.0 using 222 DNA markers in the Molecular Breeding laboratory.

We searched quantitative trait loci related to heading date and downloaded gene sequences in Gene Bank.

DNA extraction

DNA extraction and PCR were performed to determine whether the genes found in the markers identified through QTL analysis were related to heading date. DNA extraction was carried out using DNeasy Tissue kit. The genomic DNA of Cheongcheong and Nagdong were extracted by CTAB (Cetyltrimethyl ammonium bromide) from fresh DHL and parental leaves cultured in a greenhouse at 25℃ for 3 weeks. Inside 2 µl microtubes, 20.2 g of leaves and iron balls were frozen for one minute with sample liquid nitrogen. The sample was pulverized for 30 seconds at a vibration frequency of 20 per second using a Tissue Layer (QIAGEN, Cat. No. 85220) and added 2x CTAB buffer (2% CTAB, 0.1 M Tris, pH 8.0, 1.4 M NaCl, 1% PVP) in tube. After vortexing, the tubes were incubated in a water bath at 65℃ for 20 minutes and then vortexed at room temperature for 20 minutes after addition of 750 µl of PCI (phenol: chloroform: isoamylalcohol = 25: 24: 1). Then, centrifugation was performed at 14,000 rpm for 10 minutes, and 500 µl of the supernatant was added to a new microtube. 350 µl of isopropanol was added to the separated supernatant, and the mixture was stirred for 5 minutes and reacted at -72℃ for 2 hr. After dissolving the reaction product at room temperature, the tube was centrifuged at 14,000 rpm for 10 minutes, and the white pellet precipitated on the bottom of the tube was washed twice with 70% ethanol and dried at room temperature. Finally, DNA was dissolved in 20 µl of distilled water and the DNA concentration of each sample was adjusted to 20 ng/µl. The extracted DNA was subjected to PCR using markers identified in QTL analysis. PCR amplification was carried out using a QIAxcel (QIAGEN, Germany) electrophoresis analyzer for DNA amplification at 94℃ for 5 minutes, 94℃ for 30 seconds, 61.5℃ for 30 seconds, 72℃ for 1 minute, and 72℃ for 10 minutes, Respectively.

PCR

The PCR amplification was carried out by adding 4 µl of 20 to 30 ng/µl template DNA, 2 µl of each primer, 0.75 µl of dNTP, 4.8 ml of Ex Taq polymerase, 10× Ex buffer and 34.25. For PCR amplification, 4 µl of 20 or 30 ng/µl template DNA, 2 µl of each primer, 0.75 µl of dNTP, 4.8 ml of Ex Taq polymerase, 10× Ex buffer and 34.25 nuclease water was added. Anneal at 56℃ for 30 seconds and at 72℃ for 30 seconds. The reaction was annealed at 72℃ (initial temperature 61.5℃) for 30 seconds while being annealed at 96℃ for 5 minutes, 96℃ for 30 seconds, and 34℃ for 30 seconds. PCR was performed on a GeneAmp PCR System 2700 (Applied Biosystems, USA) or MyGenie 96 Thermal Block (Bioneer, Korea). The primers were: Forward 5′- ATG CAC ATT ACT ACA GTT GAC CGC-3′, Reverse Transcription 5′-TCA GCA TAC CAC ACC ATC GAC ATC-3′ and 5′-TCA GCA TAC CAC ACC ATC GAC-3′ (Table 1). After PCR, electrophoresis was performed on 0.8% agarose gel.

Gel extraction

The DNA samples were separated by 0.8% agarose gel into target sizes and extracted. The separated gel was purified using a QIAEX II gel extraction kit. Briefly, 200 mg agarose gel was placed in a 1.5 µl tube. 900 µl of QX I and 10 µl of QIAEX II were added, followed by vortexing for 30 seconds. The reaction was allowed to proceed for 10 minutes in a water bath at 50℃ and vortexing was performed every 2 minutes. After the reaction, centrifugation was performed for 1 minute (13,000 rpm) and the supernatant was discarded. 500 µl of QXI was added, vortexed and centrifuged for 1 minute. The supernatant was discarded and 500 µl of PE buffer (containing ethanol) was added, followed by vortexing and centrifugation for 1 minute. Discard the supernatant as before. Then open the lid of the tube and wait for 10-15 minutes. Add 20 µl of free water and vortex until DNA is dissolved. After the lid is closed, the reaction is allowed to proceed for 5 minutes, followed by centrifugation for 1 minute. Since the DNA is contained in the supernatant, the supernatant is extracted and stored in the tube. Add 3 volumes of QX1 buffer to the tube. Add of 30 µl of QIEX II, vortex for 30 seconds. Incubate at 50℃ for 10 minutes to dissolve the agarose and bind the DNA. Centrifuge the sam-

Table 1. Designed primer for PCR

Primer	Sequence (5′- 3′)	Size (bp)	GC %	Tm (℃)
Forward_1	ATG CAC ATT ACT ACA GTT GAC CGC	24	45.8	61.0
Reverse_1	TCA GCA TAC CAC ACC ATC GAC ATC	24	50.0	62.7
Forward_2	ATG CAC ATT ACT ACA GTT GAC CGC	24	45.8	61.0
Reverse_2	TCA GCA TAC CAC ACC ATC GAC	21	52.4	59.8

ple for 30 seconds and carefully remove the supernatant. The pellet is washed with 500 µl Buffer QX1. Air dry for 10 minutes until the pellet turns white, add 20 µl of nuclease-free water at room temperature for 5 minutes, centrifuge for 30 seconds, and then transfer the supernatant to a new 1.5 µl tube.

PGEM-T easy vector ligation

Ligation is the process of inserting a gene into a cell and cutting the cell DNA to connect it with the target DNA fragment. To prepare the sample, make 3 µl of target DNA, 5 µl of 2X ligation buffer, 1 µl of PGEM-T easy (vector) and 1 µl of T4 DNA ligase, A total of 4 samples were made as a sample. For positive control, 2 µl of Control DNA, 5 µl of 2X ligation buffer, 1 µl of PGEM-T easy (vector), 1 µl of T4 DNA ligase and 1 µl of ddH_2O were used. For background control, 5 µl of 2X ligation buffer, 1 µl of PGEM-T easy (vector), 1 µl of T4 DNA ligase, and 3 µl of ddH_2O were used. After 1 hr of reaction at room temperature, 50 µl of component cell (*E. Coli*) was added and the reaction proceeded in ice for 20 minutes. Then, the reaction is performed in a water bath at 42℃ for 45 seconds, and the target DNA is introduced into *E. coli*. After 45 seconds, re-react for 2 minutes in ice, add 950 µl of LB medium, and put in Incubate for 90 minutes at 37℃. Incubate and centrifuge for 10 minutes. Add 40 µl of X-gal and 50 µl of IPTG to the solid medium. The centrifuged recombinant gene is incubated at 37℃ for 16-24 hr in Incubate in solid medium. If the experiment is successful, there is white and blue colony in the sample, white colony in the positive control, and white and blue colony in the background control. There is only a blue colony in the negative control. In the sample, insert the colony containing the white recombinant DNA into a 5 ml LB solution and incubate for 16 hr at 37℃ on Incubate.

Plasmid DNA

The *E. coli* cultured in the LB medium is centrifuged to remove the LB medium. Dissolve the pellet in 250 µl of P1 buffer (for DNA separation and purification). Add 250 µl of P2 buffer and mix thoroughly by inverting 4-6 times. If you used LyseBlue reagent, the solution turns blue. Add 350 µl of N_3 buffer and mix 4-6 times by inverting. If you used LyseBlue reagent, the solution turns colorless. Then centrifuge at 13,000 rpm for 10 minutes. The supernatant obtained after centrifugation is washed with 500 µl of PB buffer in a QIAprep spin column and centrifuged for 1 minute. Discard the filtrate. Add 750 µl of PE buffer and centrifuge for 1 minute. Discard the filtrate and centrifuge for an additional 1 min to remove the bottom of the filtrate and QIAprep spin column. Place the filter section of the QIAprep spin column in a 1.5 ml microcentrifuge tube to dissolve the DNA. Add 40 µl of distilled water to the QIAprep spin column, incubate for 15 minutes, and centrifuge for 1 minute. Thereafter, the filtrate in the e-tube contains plasmid DNA.

Results

When the number of days from sowing to the heading date was expressed, the result of examining the heading date of Cheongcheong/Nagdong doubled haploid (CNDH) showed that the mother plant, Cheongcheong and Nagdong, were 105 and 104, respectively, similar to CNDH, and showed a continuous variation close to the normal distribution (Fig. 2). As a result of investigating the growth characteristics, In the case of culm length, the CNDH group (69.5±13.4 cm) was lower than that of Cheongcheong (74.2±15.7 cm) and higher than Nagdong (64.6±13.5 cm). In the case of panicle length, the CNDH group (21.1±2.4 cm) showed a similar pattern in both Cheongcheong (21.3±1.4) and Nagdong (19.3±1.8). In the case of panicle number, the CNDH group (11.2±2.7) showed a similar pattern in both

Table 2. The plant traits of Cheongcheong/Nagdong doubled haploid (CNDH) lines

Plant traits	Parents			CNDH population	
	Cheongcheong	Nagdong	Mean	Range	Mean
Heading date	105.3±2.9[a]	104.6±5.2	105.0±4.2	86-141	103.1±10.1
Culm length (cm)	74.2±15.7	64.6±13.5	67.6±14.9	43.7-111.2	69.5±13.4
Panicle length (cm)	21.3±1.4	19.3±1.8	19.9±1.9	15.7-27.9	21.1±2.4
Number of panicle	11.1±2.5	10.7±2.7	11±2.6	5.0-19.0	11.2±2.7
1,000 grain weights (g)	28.5±1.2	25.6±8.7	27.2±9.2	8.2-31.8	25.0±3.4

[a]The data are presented in mean ± standard deviation.

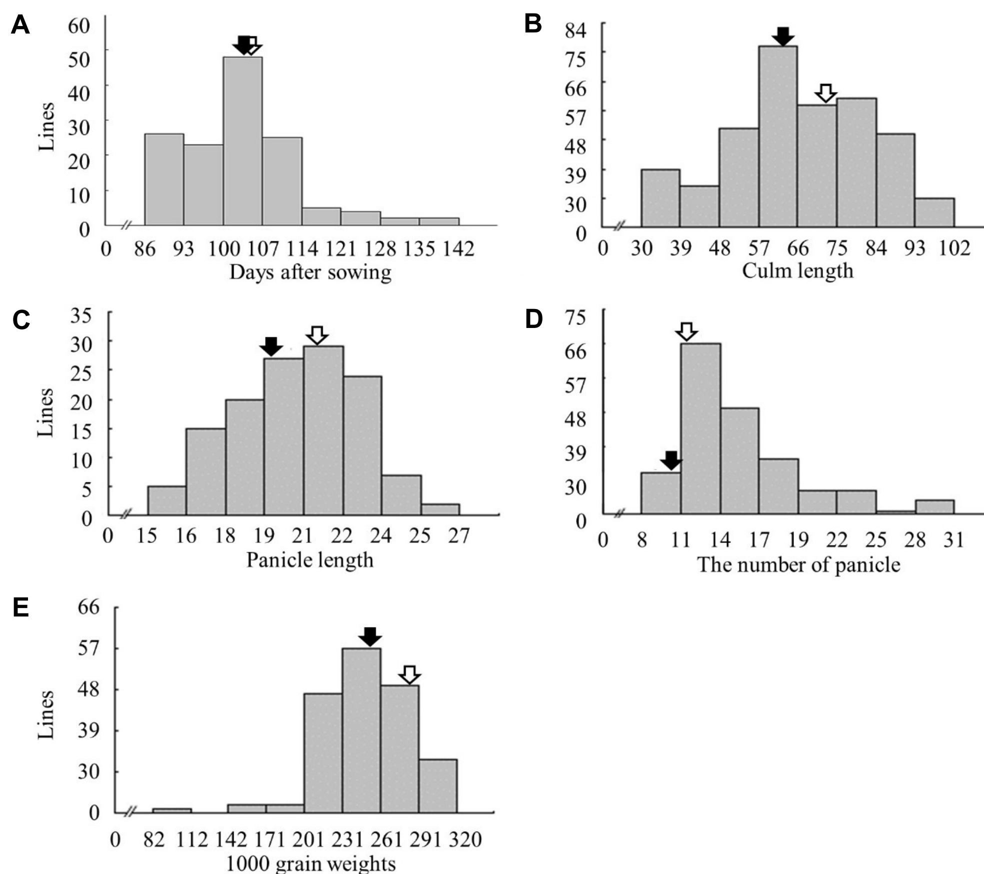

Fig. 2. Trait analysis of parents and population. The frequency distribution for growth survey in CNDH population. The black arrow represents Nagdong, the white arrow represents Cheongcheong. (A) Days after sowing, Nagdong; 104.6 days, Cheongcheong; 105.3 days, (B) Culm length, Nagdong; 64.6 cm, Cheongcheong; 74.2 cm, (C) Panicle length, Nagdong; 19.3 cm, Cheongcheong; 21.3 cm, (D) The number of panicle, Nagdong; 10.7, Cheongcheong; 11.1, (E) 1,000 grain weights, Nagdong; 25.6 g, Cheongcheong; 28.5 g.

Cheongcheong (11.1±2.5) and Nagdong (10.7±2.7). In the case of thousand grain weights, the CNDH group (25.0±3.4 g) was lower than Cheoncheong (28.5±1.2 g), but it was similar to Nagdong (25.6±8.7 g). In QTL analysis, one QTL was detected on chromosome 8 and was named qHd8. qHD8 was located between the RM72 and RM404 markers, and he range at qHd8 was approximately 7.7 cM. The LOD value was 3.93 and the R2 value was 0.14% (Table 3). There were 13 Contig and 1 ORF. The gene named PSP, a gene for signaling related protein, can be seen that it is related to growth reported by Yanying et al., 2008[12]. Gene cloning was performed using the ORF found in QTL. After PCR, it was confirmed that the gene was cloned into a plasmid vector in 24 of 30 samples. The gene length is 125 bp which is the same as the ORF length. Protein sequence analysis showed that PSP-like proteins were similar to OsO8g0341700, AtSFH13 and AtSFH7 proteins (Fig. 3). Os08g0341700, which is involved in signal transduction, is similar to phosphatidylinositol transfer-like protein II, and complete information has not been released. However, it is estimated to play a role of phosphatidylinositol-specific signaling pathway related to Sec14P.

Discussion

Disaster-related extreme weather is rapidly increasing

Table 3. The chromosomal location of QTL associated with heading date in CNDH population

Locus	LOD	R^2	Additive effect	Marker	Allele source
qHd8	3.93	0.14	8.62	RM72-RM404	Cheongcheong

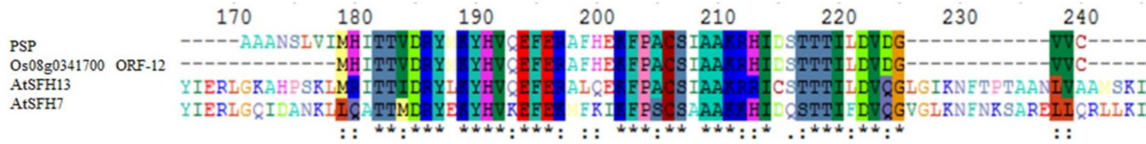

Fig. 3. Similar protein sequences were analyzed at NCBI and the same amino acids were labeled.

due to unusual weather caused by climate change such as global warming. Usually in Korea, typhoons accompanied by rainfall approach August to September, causing great damage. In general, some breeze during the growing period of rice is known to give beneficial conditions to the physiological action of rice, but excessive strong wind takes moisture from plants in addition to mechanical injury to the rice, resulting in physiological disturbances such as wilting of leaves, discoloration and infertility of the glume. Causes In particular, if the wind hits strongly during the heading stage, the whole ear will dry and die, which greatly affects the quantity [2]. If the amount of discoloration increases due to the wind caused by typhoon, this directly affects the ripening rate and the perfect kernel rate, and as a result, it acts as a major factor in the reduction of yield. Therefore, in order to reduce typhoon damage, it is important to control it so that it does not heading during a typhoon [10]. The average of heading date of the CNDH line (103.1±10) was relatively short compared to the average of the heading date of the CNDH line in 2017(107.1±7.8), but this seems to be due to differences in weather by year [3]. The locations of the Cheongcheong and Nagdong, the mother plants of the CNDH line, were indicated by black and white arrows in the distribution chart, and the characteristics of the heading date, culm length, panicle length, the number of panicle and 1,000 grain weights all have a normal distribution. To identify the QTLs, WinQTLCart Cartographer 2.5 was used. An LOD value of 3.0 was used as a threshold for obtaining QTL at $p<0.05$. The QTL locations identified in this study were compared to previously reported QTLs affecting rice heading. There have been many QTL studies on rice heading date. Zhang et al., 1997 [14] reported that the main QTL, which regulates the yield, heading date, and plant height of rice, is located on chromosome 8. Yano et al., 1997[13] performed QTL analysis using more than 850 markers. It was reported that two QTLs with a large effects were present on chromosomes 6 and 7, respectively, and three QTLs with minor effects were located on each chromosome 6, 7 and 8. Li et al., 1995[7] reported that chromosomes 3 and 8 had one QTL associated with the heading date. In this study, it was confirmed that one QTL related to the water dispenser was located on chromosome 8.

Acknowledgement

This work was supported by a grant from the New breeding technologies development Program (Project No. PJ0147 93012020), Rural Development Administration, Republic of Korea

The Conflict of Interest Statement

The authors declare that they have no conflicts of interest with the contents of this article.

References

1. Bres-Patry, C., Lorieux, M., Clement, G., Bangratz, M. and Ghesquière, A. 2001. Heredity and genetic mapping of domestication-related traits in a temperate japonica weedy rice. *Theor. Appl. Genet*. **102**, 118-126.
2. Bruno, E., Choi, Y. S., Chung, I. K. and Kim, K. M. 2017. QTLs and analysis of the candidate gene for amylose, protein, and moisture content in rice (*Oryza sativa* L.). *3 Biotech.* **7**, 40.
3. Huang, J., Ghosh, R. and Bankaitis, V. A. 2016. Sec14-like phosphatidylinositol transfer proteins and the biological landscape of phosphoinositide signaling in plants. *Biochim. Biophys. Acta.* **1861**, 1352-1364.
4. Jang, Y. H., Park, J. R. and Kim, K. M. 2019. Characterization of heading- and yield-related gene loci in the Cheongcheong/ Nagdong doubled haploid line using rice QTLs. *Kor. J. Crop Sci.* **64**, 1-17
5. Kang, S., Shon, J., Kim, H., Kim, S., Choi, J., Park, J. and Yang, W. 2018. Analysis of genetic variation in pre-harvest sprouting at different cumulative temperatures after heading of rice. *Kor. J. Crop Sci.* **63**, 8-17.
6. Li, Z., Pinson, S. R. M., Stansel, J. W. and Park, W. D. 1995. Identification of quantitative trait loci (QTLs) for heading date and plant height in cultivated rice (*Oryza sativa* L.). *Theor. Appl. Genet*. **91**, 374-381.
7. Lin, H. X., Yamamoto, T., Sasaki, T. and Yano, M. 2000. Characterization and detection of epistatic interactions of 3 QTLs, Hd1, Hd2, and Hd3, controlling heading date in rice using nearly isogenic lines. *Theor. Appl. Genet.* **101**, 1021-

1028.
8. Lee, G. H., Kang, I. K. and Kim, K. M. 2016. Mapping of novel QTL regulating grain shattering using doubled haploid population in rice (*Oryza sativa* L.). *Int. J. Genomics* **2016**, 2128010.
9. Ma, H., Xu, S. P., Luo, D., Xu, Z. H. and Xue, H. W. 2004. *OsPIPK1*, a rice phosphatidylinositol monophosphate kinase, regulates rice heading by modifying the expression of floral induction genes. *Plant Mol. Biol.* **54,** 295-310.
10. Park, J. K., Kim, B. S., Jung, W. S., Kim, E. B. and Lee, D. G. 2006. Changes in statistical characteristics of typhoon affecting the Korean peninsula. *Atmosphere* **16**, 1-17.
11. Park, S. J., Kim, S. L., Lee, S., Je, B. I., Piao, H. L., Park, S. H. and Colasanti, J. 2008. Rice Indeterminate 1 (*OsId1*) is necessary for the expression of *Ehd1* (Early heading date 1) regardless of photoperiod. *Plant J.* **56**, 1018-1029.
12. Qu, Y., Mu, P., Zhang, H., Chen, C. Y., Gao, Y., Tian, Y. and Li, Z. 2008. Mapping QTLs of root morphological traits at different growth stages in rice. *Genetica* **133**, 187-200.
13. Yano, M., Harushima, Y., Nagamura, Y., Kurata, N., Minobe, Y. and Sasaki, T. 1997. Identification of quantitative trait loci controlling heading date in rice using a high-density linkage map. *Theor. Appl. Genet.* **95**, 1025-1032.
14. Zhang, Y., Luo, L., Xu, C., Zhang, Q. and Xing, Y. 2006. Quantitative trait loci for panicle size, heading date and plant height co-segregating in trait-performance derived near-isogenic lines of rice (*Oryza sativa*). *Theor. Appl. Genet.* **113**, 361-368.

초록 : 청청/낙동 배가반수체 유전자 지도를 이용한 쌀의 출수기 관련 양적형질유전자좌(QTL) 분석

장윤희 · 박재령 · 김경민*
(경북대학교 농업생명과학대학 응용생명과학부)

본 연구는 지구온난화와 태풍에 의해 수확기의 손실을 막기 위해 벼의 출수기를 당기는 유전자를 찾는 것을 목표로 한다. 청청/낙동 배가반수체(CNDH)와 모본인 청청, 부본인 낙동을 재료로 사용하여 QTL을 이용해 출수기 관련 유전자의 위치를 조사하고 gene을 cloning하여 염기서열을 분석하였다. 분석결과 염색체 8번에 13개의 contig가 있었고 그 중 출수기와 관련된 1개의 ORF가 존재했다. 단백질 서열을 분석한 결과 벼의 Os08g0341700, 그리고 AtSFH13, AtSFH7 단백질과 유사한 것으로 보인다. 신호전달과 관계가 있는 Os08g0341700은 phosphatidylinositol transfer-like protein II와 유사하며 아직 완전한 information은 밝혀지지 않았다. 하지만 Sec14P와 연관이 있으며 세포 생장 등에 관여하는 phosphatidylinositol 특이적 신호전달 경로의 역할을 할 것으로 추정 중이다.