QTLs for Domestication-related and Agronomic Traits in Temperate Japonica Weedy Rice

Chang-Sik Oh1, Seung-Joon Lee1, Dong-Beom Yoon1, Jung-Pil Suh2, and Sang-Nag Ahn1*

1Department of Agronomy, College of Agriculture & Life Sciences, Chungnam National University, Daejeon 305-764, Korea
2National Institute of Crop Science, RDA, Suwon 441-100, Korea

Abstract: This study was conducted to identify the genetic basis of the domestication-related traits in weedy rice. An RIL population consisting of 80 lines was developed from a cross between the japonica weedy rice, Hapcheonaengmi 3 and the Tongil-type cultivar Milyang 23. The population was genotyped with 132 DNA markers, providing an average interval size of 11.0cM, and also evaluated for 20 traits related to domestication and agricultural performance. A total of 48 QTLs and two loci associated with qualitative variation for pericarp and base coloration were identified using single point and interval analysis. The number of QTLs per trait ranged from one to six. These 48 QTLs were located in 27 intervals distributed on 11 chromosomes except for chromosome 12. The results indicated that most domestication-related traits clustered in chromosomal blocks, and the positions of many of these clusters were consistent with those reported in previous studies. Phenotypic variation associated with each QTL ranged from 7.5 to 31.9%. For 10 (40%) of the QTLs identified for agricultural performance in this study, the Hapcheonaengmi 3-derived allele contributed a desirable agronomic effect despite the overall undesirable characteristics of the weedy phenotype. Favorable wild alleles were detected for days to heading, panicle exsertion, primary branch number, and leaf discoloration related with cold tolerance. When compared with previous studies involving interspecific crosses, it can be concluded that weedy rice is useful as a source of valuable alleles for rice improvement.

Key words: Rice (Oryza sativa L.), quantitative trait locus (QTLs), domestication, weedy rice

INTRODUCTION

Rice is often associated with weedy forms which are genetically related (Harlan et al. 1972). These weedy forms display intermediate characteristics between indica or japonica cultivars of O. sativa and a presumed wild progenitor, O. rufipogon (Oka 1988). Weedy rice usually shows a red pericarp, a high seed dispersal ability and a coloration in various organs including the hull, seed coat and pericarp (Suh & Ha 1994). Although the emergence of weedy forms remains unclear, the finding that weedy types of rice can occur where wild rice is not present, supports the possibility that weedy rice evolves through the degeneration of domesticated rice (Vaughan et al. 2003). The degeneration of domestication-related traits involves clusters of genes or quantitative trait loci (QTLs) (Bres-Patry et al. 2001). Weedy rice has become important resources for breeding and for studying the domestication process of rice (Bres-Patry et al. 2001, Suh et al. 1997). Several studies of their genetic characteristics have been reported; weedy rice strains also appear to be differentiated into indica and japonica types based on morphological and physiological traits, isozymes, restriction fragment length polymorphism (RFLP) and random amplified polymorphic DNA (RAPD) marker (Cho et al. 1995, Suh et al. 1997). Several reports indicated that weedy rice possesses useful genes conferring tolerance to various biotic and abiotic stresses (Suh & Ha 1994, Suh et al. 1999). In these respects, weedy rices are expected to play an important role in increasing the genetic diversity of cultivated rice (Suh et al. 1997, Suh et al. 1999).

Quantitative trait locus (QTL) mapping using molecular markers has made it possible to understand the genetic architecture of quantitative traits, including the number of loci underlying a trait, their chromosomal locations, their phenotypic effects, and the interactions among these genes ( Tanksley 1993). High-density genetic linkage maps comprised of simple sequence repeats (SSRs) markers are available in rice (Temnykh et al. 2000, 2001, McCouch et al. 2002). Rice has been widely used as a plant model for quantitative genetic research. In the past decade, a number of studies on Oryza species have identified numerous QTLs underlying various traits (for a summary, refer to
www.gramene.org, McCouch & Doerge 1995). The objectives of this study were to map genes and quantitative traits loci (QTLs) involved in the variation of weedy traits using an RIL population derived from a cross between the Koran japonica weedy rice, Hapcheonaengmi 3 as the donor and the Tongil-type rice, Milyang 23 as the recurrent.

**MATERIALS AND METHODS**

**Plant materials**
A population of 80 recombinant inbred lines (RILs) was derived from a cross between “Milyang 23” and “Hapcheonaengmi 3”. A Korean Tongil-type cultivar, Milyang 23 (Oryza sativa ssp. indica), was used as the female parent and also served as the recurrent parent. A Korean japonica weedy rice, Hapcheonaengmi 3 is photoperiod insensitive, and has fully exserted panicles (Suh et al. 1999). F1 plants were backcrossed to Milyang 23 to produce BC1F1 plants. These plants were grown in the field and selfed for five generations via single seed descent.

**Field trial and trait evaluation**
Eighty BC1F6 lines were grown in the field at the Chungnam National University, Daejeon, Korea during the summer of 2003. Two-row plots with 30 plants per row were planted by 30 cm × 15 cm distance in a completely randomized block design with two replications. Twenty domestication-related and agronomic traits were evaluated in the BC1F6 families. Two qualitative traits, pericarp coloration (pc) and base coloration (bc), were included and evaluated according to the absence or presence of the red color. Other traits were quantitatively inherited and evaluated based on commonly used methods in rice genetics and breeding. Days to heading (dth) was evaluated as the number of days from seeding until 50% of the panicles of the 30 plants were heading. For culm length, panicle length, panicles per plant and seedling height, 15 plants per line in the middle were measured and the average of the measurements was used as the phenotype of each line. Culm length (cl) was measured in centimeters from the soil surface to the neck of tallest panicle. Panicle length (pl) was measured in centimeters from panicle neck to the panicle tip. Panicle number (pn) was calculated as the number of panicles per plant. Seedling height (sdh) was measured as the number of centimeters from the soil to the tip of tallest leaf before transplanting. The panicle exertion were evaluated on ten plants per line. Panicle exertion (pe) was measured as the number of centimeters from the top of the flag leaf sheath to the panicle neck. Six representative panicles per line were bagged after panicle emergence to avoid seed shattering and evaluated for spikelets per panicle, percent seed set, awn length, primary branches per panicle and secondary branches per panicle. Spikelets per panicle (svp) was calculated as the average number of spikelets per panicle. Percent seed set (pss) was calculated as a percentage: the number of filled grains per panicle was divided by the number of spikelets per panicle and percentages for each panicle were averaged. Awn (awn) was evaluated categorically using 2 classes (1 = non, 3 = presence). Tiller angle (ta) was scored visually at peak tillering stage based on the standard scale of 1-9 scores in which 1 indicated an angle of zero degree with the stem, and 9 indicating an angle larger than 90 degrees with the stem. Primary branches per panicle (pb) were evaluated by counting the average number of primary branches per panicle. Secondary branches per panicle (sb) were similarly counted as the average number of secondary branches per panicle. Seed shattering (sh) was recorded on a 1-to-9 scale where 1 = non, 3 = low level, 5 = average, 7 = high level, and 9 = shattered by hand gripping. Seed dormancy (sd) for each RIL was evaluated as the germination percentage. Thirty days after harvesting, 30 intact seeds per line were placed in Petri dishes and germinated in the dark at 30°C. The number of germinated seeds after 10 days was counted. Grain length (gl), grain width (gw), grain thickness (gt) and length/width ratio of grain (hw) were measured on 30 grains per line using the vernier caliper. Means for the two replications were calculated for each trait and used in data analysis.

**SSR analysis and linkage map construction**
DNA was extracted from young fresh leaves in bulk of BC1F6 plants as described in Causse et al. (1994). A total of 257 SSR markers were used in polymorphic survey between the parents. The polymorphic markers between the parents were used for genotype analysis of the 80 BC1F6 lines. SSR analysis was performed according to the method described in Panaud et al. (1996), with the following modification in the PCR profile: 94°C for 5 min., followed by 35 cycles of 94°C for 1 min., 55°C for 1 min., and 72°C for 1 min., and lastly 5 min. at 72°C. The linkage map was constructed with 2 morphological, 131 SSR and 1 telomeric marker, TEL-1a (Yang et al. 2003), and the order and distance between markers were based on two previously developed SSR maps (Temenykh et al. 2001, McCouch et al. 2002).