Available online at www.sciencedirect.com

ScienceDirect

Rice Science, 2018, 25(1): 19−31

Yield Traits and Associated Marker Segregation in Elite Introgression Lines Derived from *O. sativa* × *O. nivara*

G. HARITHA[1], B. P. M. SWAMY[1, 2], M. L. NAIK[1], B. JYOTHI[1], B. DIVYA[1], S. MALATHI[1], N. SARLA[1]

([1]*Indian Council of Agricultural Research, Indian Institute of Rice Research, Rajendranagar 500030, Hyderabad, India;* [2]*Plant Breeding, Genetics and Biotechnology Division, International Rice Research Institute, DAPO Box 7777, Metro Manila, the Philippines*)

Abstract: Introgression lines (ILs) derived from interspecific crosses are a source of new genetic variability. A total of 55 ILs derived from two crosses Swarna × *O. nivara* IRGC81848 (population A) and Swarna × *O. nivara* IRGC81832 (population B) were characterized for yield and yield-related traits/QTLs. Segregation of 103 simple sequence repeat (SSR) markers associated with yield-related QTLs was studied. Population A showed an average of 12.6% homozygous *O. nivara* alleles and population B showed 10.6%. Interestingly, three SSR markers, RM223, RM128 and RM517, showed conspicuous pattern of segregation. The distribution of parental alleles at three loci RM223, RM128 and RM517 linked to yield-related traits was unique. These markers flanked to several yield-related QTLs. RM223, flanking to *qyld8.3*, was heterozygous in almost all the 55 ILs except in IL10-3S and IL131S. RM128 on chromosome 1 and RM517 on chromosome 3 were mutually exclusive in 46 out of 55 ILs. These 46 ILs showed either of the marker allele RM128 or RM517 from *O. nivara* but not both. IL166S had both RM128 and RM517 from *O. nivara* and the other ILs showed homozygous Swarna allele at RM517 except IL65S. Population structure assigned the 55 ILs to three sub-populations based on their genomic diversity. IL65S, IL166S, IL248S, IL7K and IL250K showed high yields in multi-location trials, and IL248S was released for cultivation as DRRDhan 40.

Key words: introgression line; heterozygosity; *Oryza nivara*; wild rice; population structure; yield; simple sequence repeat

Wild species of rice have been identified as potential sources of important agronomic traits. The major objective of modern crop breeders is to recover the abundant genetic diversity in primary gene pool and introgress the favourable alleles into elite cultivars to overcome stress (Gur and Zamir, 2004; Sharma et al, 2013), for example, pests and diseases (Jena and Kim, 2010; Sarao et al, 2016), drought, salinity and cold (Koseki et al, 2010; Ndjiondjop et al, 2010; Yang et al, 2012). Many studies show that introgression from wild species enhances yield in the genetic background of elite varieties (Swamy et al, 2008; Bai et al, 2012). Recent studies show that introgression from non-AA genome species *Oryza minuta* (CCDD) and *O. australiensis* (EE) is also used in successful transfer of blast resistance genes into *O. sativa* (Jena and Khush, 2000; Fu et al, 2008). Introgression lines (ILs) are important tools which contain the segments from wild rice and used for primary mapping, fine mapping and positional cloning of important QTLs/genes. Development of these ILs (elite cultivar × wild) is achieved by several rounds of backcrossing with recurrent parent followed by marker-assisted selection (Guo and Ye, 2014; Hasan et al, 2015). The ILs are used for characterization of several agronomic traits (Tian et al, 2006; Rangel et al, 2008) and genotyping-by-sequencing (GBS) using simple sequence repeat (SSR) markers (Arbelaez et al, 2015). In the past, *O.*

Received: 10 July 2017; **Accepted:** 6 November 2017

Corresponding author: G. HARITHA (harithaguttikonda@gmail.com)

Peer review under responsibility of China National Rice Research Institute

http://dx.doi.org/10.1016/j.rsci.2017.11.001

rufipogon and *O. glumaepatula* of AA genome species have been used in the development of ILs in rice (Rangel et al, 2013). Recently, *O. minuta*, a tetraploid wild rice (BBCC), has been used in the construction of ILs (Guo et al, 2013). *O. nivara* is known as a source of important genes like cytoplasmic male sterility (Li et al, 2005), resistance to grassy stunt virus (Brar and Khush, 2004), bacterial leaf blight (Cheema et al, 2008) and brown planthopper (Madurangi et al, 2013; Sarao et al, 2016). Yield QTLs are mapped in BC_2F_2 population and in advanced backcross population using *O. nivara* as a donor parent and Swarna as a recipient parent (Kaladhar et al, 2008; Gaikwad et al, 2014; Swamy et al, 2014; Surapaneni et al, 2017). These populations were further advanced and the ILs were selected based on yield-related parameters for molecular characterization. The characterization of ILs with presence of yield enhancing QTL alleles can serve as pre-breeding materials for development of new high-yielding improved varieties or near-isogenic lines for fine mapping and gene discovery.

In the present study, we genotyped 55 ILs (BC_2F_6) using SSR markers linked to yield and yield-related QTLs reported in BC_2F_2 of the same cross. The aim of this study was to estimate the proportion of *O. nivara* alleles in the ILs and the presence of reported QTLs, and analyze the phenotypic variation, genetic diversity and population structure of ILs.

MATERIALS AND METHODS

Plant materials

Fifty-five BC_2F_6 ILs derived from two interspecific crosses were selected for yield and their morphological similarity with recurrent parent Swarna. The ILs designated as 'S-lines' (population A) were derived from the cross between an elite *indica* rice variety Swarna and *O. nivara* accession IRGC81848 from Uttar Pradesh, India. The ILs designated as 'K-lines' (population B) were derived from the cross between Swarna and *O. nivara* accession IRGC81832 from Bihar, India. The BC_2F_2 of these two crosses were used to map QTLs for yield and yield-related traits (Kaladhar et al, 2008; Swamy et al, 2011). The BC_2F_6 generation of ILs derived from these two interspecific crosses were used for this study (Table 1).

Phenotypic evaluation

The ILs were evaluated for grain yield and yield-related traits in a randomized block design with two replications containing 21 plants in 3 rows with 15 cm × 20 cm spacing at the research field of Indian Institute of Rice Research (IIRR), Hyderabad (17º19′ N, 78º29′ E), India, during the dry season of 2013. The traits were taken from five randomly selected plants from each replication, including plant height, number of tillers per plant (NT), number of productive tillers per plant (NP), days to 50% flowering (DFF), yield per plant, aboveground biomass, total dry matter and harvest index.

Statistical analysis

The mean values from each replication were subjected to statistical analysis using the software program Statistical Analysis for Agricultural Research STAR (STAR 2.0.1, http://bbi.irri.org). To determine the significant differences among ILs of both populations, analysis of variance (ANOVA) and descriptive statistics were performed for all the traits. Correlation coefficients were also estimated using Microsoft Excel 2007 and significant values were determined using Pearson coefficients at the 0.05, 0.01 and 0.001 levels.

DNA extraction and PCR amplification

The genomic DNA was extracted from young leaves using CTAB (Cetyl Trimethyl Ammonium Bromide) method according to Zheng et al (1995). The quality and quantity of isolated genomic DNA were measured through spectrophotometry using Nanodrop (ND-1000, Thermo Scientifics, USA). The DNA samples were diluted with Tris-EDTA (TE) buffer to get a concentration of 50 ng/μL.

A total of 103 SSR markers which were

Table 1. Introgression lines used.

Population	No. of lines	Introgression line
Population A Swarna/*O. nivara* IRGC81848	34	IL8S, IL10-1S, IL10-2S, IL10-3S, IL14S, IL14-3S, IL33S, IL53S, IL65S, IL70S, IL73S, IL75S, IL94S, IL96S, IL106S, IL126S, IL131S, IL137S, IL147S, IL150S, IL160S, IL164S, IL166S, IL175S, IL175-3S, IL175-5S, IL177S, IL205S, IL210S, IL212S, IL222S, IL230S, IL231S, IL248S
Population B Swarna/*O. nivara* IRGC81832	21	IL3K, IL7K, IL13K, IL14K, IL33K, IL94K, IL106K, IL131K, IL137K, IL144K, IL145K, IL149K, IL150K, IL230K, IL231K, IL242K, IL246K, IL250K, IL251K, IL254K, IL262K