Transcriptome Resources For Function Analysis and Genetic Enhancement of Rice

Agarwal P, Parida S, Kothari KS, Sharma G, Baranwal V[1], Kapoor S[1] and Tyagi A

National Institute of Plant Genome Research, Aruna Asaf Ali Marg, New Delhi-110067; and [1]Interdisciplinary Centre for Plant Genomics and Department of Plant Molecular Biology, University of Delhi, South Campus, New Delhi-110021, India. Email: akhilesh@genomeindia.org

Abstract

World food security depends on rice production to a large extent, particularly in the developing nations. It has been the target of major breeding and first crop genome sequencing efforts. There is a need to combine genomic resources with breeding to evolve new strategies for genetic enhancement of rice. Recently, several efforts have been made to determine differential expression of rice transcriptome at the level of development, stress, and heterosis to find out genes associated with such processes and related traits. Function of genes selected on the basis of transcriptome profiling has been demonstrated by using knock-out and over-expressing transgenics as well as gene mutants. Interestingly, such information can be used to search for diversity in target genes representing different alleles and utilized to develop gene-based markers. Association mapping of such genes with target traits can provide direct proof of the function of gene and its utility for deployment in breeding programs. It is hoped that such strategies would help desired genetic enhancement of rice.

Keywords: Development, gene function, genetic enhancement, heterosis, molecular mapping and breeding, rice, stress

Introduction

The monocot crop plant rice is a model system and has been the subject of genomic research at various levels. A study of the transcriptome of rice, in different organs, during development gives an insight into the regulatory aspects as well as various processes occurring during development and provides vital clues about genes, which can be exploited to improve related traits. A better insight can be gained by analyzing transcriptome variation amongst varieties with contrasting characters. These genes can further be used to enhance appropriate plant features by molecular breeding or genetic engineering. The protection of loss due to abiotic stresses like drought, salt and cold is another area to improve crop production. Hence, it becomes necessary to take into account this factor while considering various approaches for increasing the productivity of rice. Earlier, researchers used a gene-by-gene approach to decipher the functions of stress responsive genes. But, abiotic stress response is a complex phenomenon and involves a large number of genes and modulating a single gene may not impart the level of tolerance desired. The availability of the complete rice genome sequence has provided the means to investigate the expression profile of the whole genome under various stress conditions in order to identify stress-responsive genes for genetic modification. The utility of transcriptome resources for marker-assisted breeding is also being realized. Differentially expressed transcriptome or exome related markers can be utilized in individual or combined approaches of genetic and association mapping and genetical genomics, for rapid identification of functionally relevant molecular tags. Thus, the genes regulating the complex quantitative traits of agricultural importance have been elucidated and used in an attempt to improve the crop.

For long, Northern hybridization has been commonly used to detect the presence of a transcript. Reverse transcription polymerase chain reaction (RT-PCR) detects the expression of a gene by means of the presence of the corresponding cDNA. Before the advent of modern methods of transcriptome studies, analysis of transcriptomes by cataloging data on expressed sequence tags (ESTs) was the method of choice. ESTs are single-pass partial sequences of cDNA clones sequenced from one end. Large-scale EST sequencing has enabled the identification of expressed genes in many organisms under varying conditions. In different vegetative and reproductive tissues and developmental stages of diverse *indica*, *japonica* and wild rice genotypes and under normal growth and stress-induced

conditions, about 400,000 EST (NCBI GenBank, http://www.ncbi.nlm.nih.gov) and 60,045 full-length cDNA sequences (Kikuchi et al 2003; Lu et al 2008) have been generated. A large number of ESTs have been submitted in National Centre for Biotechnology Information (NCBI) in EST database (dbEST; http://www.ncbi.nlm.nih.gov/dbEST/). Progressing to relatively newer resources, serial analysis of gene expression (SAGE) emerged as an efficient tool for expression studies which could be be applied to quantitatively analyze the expression levels of a large number of genes simultaneously. Here, a 9-11 bp fragment (tag) from 3' end represents a transcript and the frequency of these tags is used as a measure to determine the abundance of a transcript in a sample. On similar lines, massively parallel signature sequencing (MPSS) developed as a high throughput tool to quantify gene expression on a genome-wide scale and is based on sequencing 17–20 nucleotides (a 'signature') adjacent to the 3′ most *Dpn*II site from millions of molecules in a sample. This depth provided a quantitative assessment of transcript abundance. An efficient and cost effective high throughput technique for transcriptome analysis is microarray where the transcriptome is labeled and the signal of its binding to the probe accounts for the presence or absence of transcripts. In recent years, next generation sequencing (NGS) technologies have opened up new vistas in this field. RNA-sequencing has enabled the detection of the complete transcriptome, which also includes hitherto undiscovered genes and their spliced versions (Lu et al 2010; Luo et al 2011).

In this article, we have attempted to bring together selected knowledge gained about transcriptome resources for rice development and abiotic stress response. This comprehensive information can be used for genetic enhancement of the rice crop not only in terms of improved yield-related characters but also imparting it the ability to withstand adverse environmental conditions, either by molecular breeding or transgenic approach.

Development related resources

As a step towards understanding rice development, transcriptome atlases covering many organs have been generated (Jiao et al 2009). Laser microdissection of 40 cell types at different developmental stages (various stages of germinating seeds, root development zones and leaf stages) from *japonica* rice followed by microarray revealed the expression of 80.8% of genes in at least one of the cell types. Each cell type had an individual transcriptome comprising of 26-52% genes examined. Around 7% genes were specific and a similar number was ubiquitous as well. Cell type-specific genes showed chromosomal clustering. The authors conclude that many genes with specific developmental roles include hormone-responsive and regulatory genes. As an example, they found the enrichment of a specific type of CCAAT box variant in a particular cell type in co-ordination with its corresponding NF-Y regulatory genes (Jiao et al 2009). In Nipponbare, the transcriptome of 48 different developmental tissues showed 731 specific genes. The analysis also revealed that the leaf transcriptome changes twice in accordance with the reproductive stages (Sato et al 2011a). Another atlas of reproductive development in Nipponbare, across 25 stages, brought to light the expression of many previously unknown genes. It was also shown that a group of genes got up regulated as anther development progressed while another group was down-regulated, indicating essential genes. Two anther-specific genes had male-sterile mutants, implying their role in the process (Fujita et al 2010). In *indica* rice, IR64, our microarray analysis across 19 stages of vegetative and reproductive development, also revealed the transcriptomic dynamics and genes essential for reproductive development (Sharma et al 2012). An atlas of 39 tissues throughout *indica* rice plant development, in varieties Zhenshan 97 and Minghui 63, revealed 5.2% tissue-specific genes. These varieties are parents of a well used, high yielding and highly adaptable hybrid Shanyou 63. Since this cross has been used to derive many QTLs contributing to positive plant characters, such as yield heterosis, bacterial blight resistance, grain size etc., the authors postulate that integration of such genetic information with expression data, will help to identify genes and pathways responsible for the superiority of the hybrid (Wang et al 2010). All the above mentioned transcriptome resources show the presence of transcription factors specifically expressed in a given tissue indicating their indispensable roles in development. Thus, a transcriptomic atlas, across a variety of developmental events, can deepen the understanding of important biological processes.

Apart from transcriptome databases covering various stages, those containing organ or tissue-specific data are also important. These give an idea about genes responsible for controlling the development of the organ under consideration and can be used for desirable manipulation of that organ. RNA-seq of *indica* cultivar 93-11 developing embryos at 3-5, 7 and 14 DAP indicated changes in expression profiles, including stage-specific and differentially expressed genes. Auxin controlled genes seemed to be important, as substantiated by embryo-defective mutants. Members of AP2, MADS box and homeobox transcription factor families emerged as significant ones (Xu et al 2012a). Microarray of *japonica* cultivars Zhongua 11 (cold-sensitive) and Hanfeng (cold-resistant) at various embryo/endosperm stages showed the down regulation

of essential seed regulatory genes on exposure to low temperature during early seed development, providing an explanation for slow growth. ABA-responsive genes and many transcription factor encoding genes, homologs of which are essential for *Arabidopsis* seed development emerged important (Xue et al 2012). Tissue covering the entire panicle developmental stages of *japonica* rice Shiokari was used for microarray to determine essential participating genes including transcription factors (Furutani et al 2006). In another *japonica* cultivar Zhonghua 10, microarray analysis revealed an array of genes responsible for anther development and pollen germination (Wei et al 2010). Microarray at four stages of anther development in IR64 has led to identification of specific genes and also of those with similar functions in other plants (Deveshwar et al 2011). Microarray of laser capture micro-dissected cells of Nipponbare developing anther revealed involvement of gibberellin and abscisic acid biosynthesis and signaling pathways as major players (Hirano et al 2008) along with scores of specific genes and essential *cis*-elements (Hobo et al 2008). A large number (33) of microarrays on laser microdissected anthers of Nipponbare rice at various stages of development were analyzed for co-expressing genes to elucidate the networking genes at meiosis and pollen wall synthesis, resulting in new putative participating genes (Aya et al 2011). Apart from reproductive development, RNA-seq has also been used for analyzing root development (Kyndt et al 2012).

Research has also moved in the direction of examining the transcriptome with the direct aim to understand improved quality traits in rice. Transcriptomes of various *japonica* cultivars - long grained and high yielding Cypress, low milling and long grained LaGrue, good eating quality Ilpumbeyo, low eating quality YR15965 and Nipponbare - from 6 days after planting seed were studied by massively parallel signature sequencing (MPSS) and RNA-seq. Each variety had certain specific genes, and its own set of differentially regulated ones. Starch metabolism and essential amino acid synthesis genes were not only up-regulated in Cypress but were also alternatively spliced. Seed-storage protein encoding genes were also up-regulated in Cypress and Ilpumbeyo. The up-regulated genes in both these varieties were found to have specific *cis*-elements in their promoter regions, which were proposed to be responsible for high milling and good eating grain qualities (Venu et al 2011). Similarly, the mature embryos of super-hybrid rice LYP9 and its parents, 93-11 and PA64, were used to construct cDNA libraries. Though stress and development-related genes were found in abundance in all the three varieties, there were certain genes that differentially expressed amongst them (Ge et al 2008).

As a next step towards analysis of microarray databases, co-expressing genes are now being determined, with the hypothesis that they may be involved in the same pathway. Based on their analysis of developmental tissues as mentioned above, the efforts are being made to compile the data in web accessible formats e.g. RiceXPro, to asses the expression of desired loci and co-expressed genes (Sato et al 2011b). Another publicly available data compilation is 'Rice oligonucleotide array database' which compares expression across 105 experiments on six platforms (http://www.ricearray.org/). The other such platforms are PlantArrayNet (http://arraynet.mju.ac.kr/arraynet/), gene co-expression network browser (http://www.clemson.edu/genenetwork/network.php) and ATTEDII (http://atted.jp/). A database of 136 Affymetrix® rice microarray datasets, covering development, stress and hormone treatments, has been used to elucidate 151 stably expressed genes (Narsai et al 2010). The sequence read archive (SRA; http://www.ncbi.nlm.nih.gov/sra) database of NCBI is a storehouse for next generation sequencing data, including RNA-seq from various rice organs. The integration of transcriptome data from multiple sources is advancement towards determination of common as well as variety-specific genes. With the availability of more such data, genes can subsequently be correlated with desired plant traits. Moreover, the elucidation of gene networks in a particular developmental process may eventually provide us with clues to engineer the requisite genes for genetic enhancement of the plant.

Once genes essential to development have been identified by expression, their role should be validated. *MADS29*, a seed-specific gene, was shown to be essential for early seed development. Silencing showed that MADS29 causes the PCD of maternal tissues after fertilization (Yin et al 2012). Rice prolamin box binding factor (RPBF) was isolated from EST clones, which have the conserved Dof sequence. *RPBF* is endosperm-specific and the protein regulates seed storage protein encoding genes (Yamamoto et al 2006). *HAZ1* was identified by screening a 3-day-old seed cDNA library. Though it shows ubiquitous expression, it is said to be a marker for embryonic radial axis differentiation (Ito et al 2004). *OsiEZ1*, identified by screening rice post-fertilization cDNA library, restored yeast telomeric silencing (Thakur et al 2003). Another gene identified by a cDNA library screen, *OsEBP-89*, is probably involved in seed maturation and shoot development (Yang et al 2002). *OsMYB2P-1*, shown to be expressed by phosphate starvation by microarray, was also found to be regulating root architecture (Dai et al 2012). These examples demonstrate that transcriptome resources can be used to search for genes of our interest, followed by determination of their function.

As can be seen, utilization of microarray data has begun at a global level wherein different datasets are being integrated or compared to find out genes really essential for a particular developmental process. The next step after the availability of such huge amounts of transcriptomic datasets is the functional validation of the genes designated to be important for development. The data will provide actual clues if the gene can be exploited for genetic enhancement of the plant.

Heterosis: Present understandings from high throughput studies

Heterosis refers to the better performance of the hybrid with respect to both the parents in a biological trait that would have survival advantage. Genetically, heterosis has been categorized as, 'mid-parent heterosis', 'heterobeltiosis' and 'standard heterosis', where comparison of hybrid performance for any trait is measured with respect to mid-parental value, the better parent and standard check variety growing in a particular area, respectively. First documented by Chales R Darwin (1876) while studying hybridization in vegetables, the phenomenon heterosis was revisited by GH Shull and CB Davenport (Davenport 1908; Shull 1908), who came up with 'overdominance' and 'dominance' models to explain the genetic basis of heterosis. The overdominance model advocated genetic advantage of a combination of different allele at single locus over the same alleles, while, the effects of recessive alleles were postulated to be masked by dominant alleles in case of dominance hypothesis. Later epistasis was also included to explain heterosis at genetic level (Powers 1945). Recently, with increasing understanding of the molecular components involved in epigenomic and small RNA mediated regulation of gene expression, epigenetic factors have also been shown to play vital role in manifestation of heterosis (Chen 2007; Eichten et al 2011). However, despite consistent efforts for over a century to incorporate physiological, genetic, biochemical, and molecular aspects to understand the underlying basis of heterosis, we have not been able to arrive at a common single explanation. In the following paragraphs, attempts have been made to compile recent views that reflect our current understanding of this long-standing riddle of heterosis.

Heterosis, as understood, is a multigenic complex quantitative trait that could be a cumulative effect of vegetative growth rate, biomass accumulation, flowering, number of reproductive features etc. (Lippman and Zamir 2007). Taking lead from the dominance and the overdominance hypotheses by Shull and Davenport, a number of investigations were undertaken to delineate the contribution of dominance and/or overdominance phenomenon during manifestation of heterosis. In 2003, Birchler and coworkers for the first time proposed to analyze transcriptome of a hybrid and its parental lines to establish correlation between genome wide fluctuations in transcript accumulation patterns and the hybrid vigor (Birchler et al 2003). The transcriptome level investigations were then extended to several hybrid:parent combinations in a number of species using SAGE, cDNA microarrays, high density expression arrays and next generation sequencing technologies (Bao et al 2005; Huang et al 2006b; Swanson-Wagner et al 2006; He et al 2010). Although these investigations varied in the both the developmental stage and tissues as well as the hybrid parent combinations used, they helped to formalize a unifying code for defining expression profiles in hybrids vis-à-vis their parental lines. Broadly, the expression profiles were divided into 'additive' and 'non-additive', respectively, if the genes exhibited mid-parental transcript accumulation level or significantly deviated from it in the hybrid. While some studies concluded additive mode as the major mode of gene action amongst differentially expressed genes (Hoecker et al 2008; Huang et al 2006a; Hui-Yong et al 2008; Swanson-Wagner et al 2006), others found predominance of the non-additive mode in younger and developing tissues (Fu et al 2011; Ge et al 2008; He et al 2010; Hoecker et al 2008; Huang et al 2006b).

Efforts have been made, and are also on going, to establish correlations between additive and non-additive modes of gene action and biomass/yield-linked metabolic and regulatory pathways. The differentially expressed genes have been associated with carbohydrate, lipid and energy metabolic pathways as well as with photosynthesis. Interestingly, a recent transcriptome level analysis of an *Arabidopsis* allotetraploid has demonstrated changes in two major clock components, *AtCCA*1 and *AtTOC*1, leading to positive entrainment of the circadian clock for high biomass production (Ni et al 2009). Further analyses of molecular processes leading to changes in the clock components have revealed involvement of variable histone methylation (H_3K_4) resulting in lowering of the *AtCCA*1 expression. Distribution and amplitude of epigenetic marks have been found to vary amongst different cultivars, genotypes, and ecotypes etc. and could be the potential cause of diversity in gene expression pattern.

In a recent study, attempt have been made to associate small RNA-mediated increase in the methylation levels at certain loci with the phenomenon of heterosis using an *Arabidopsis* hybrid (Groszmann et al 2011). In another study methylome profiling of a maize heterotic parental pair Mo17 and B73, led to the identification of approximately 700 differentially methylated regions

(Eichten et al 2011). In a similar study in *Arabidopsis* hybrid resulting from a cross between Ler and C24 ecotypes, large numbers of differentially methylated clusters were identified, which were further categorized into additive and non-additive methylation categories based on relative methylation amplitude in the hybrid and its parental lines. The methylation profiles of most of the loci were found to be additive, nevertheless, non-additivity was observed in ~18-22% of the loci. The heterotic potential of hybrid was compromised when treated with an agent that demethylates DNA or when production of functional small RNAs was abolished due to mutations in *Arabidopsis* RNA methyltransferase, *HUA ENHANCER1,* suggesting the role of RNA dependent DNA methylation (RdDM) pathway in the manifestation of heterosis (Shen et al 2012).

The phenomenon heterosis has been investigated at various levels including functional genomics; proteomics, metabolomics and all these studies have helped in cataloging the differentially accumulated components. These components have been shown to affect basic metabolic and regulatory pathways like the clock, photosynthesis, carbohydrate and lipid metabolism and in some instances reproductive traits leading to higher yield in the hybrids. The stage is now set to undertake systems level analyses, which are able to assimilate the knowledge accumulated so far and take it to a level where the heterotic potential of a parental combination can be predicted even without making in single cross in the field.

Stress associated transcripts and their databases

A large collection of ESTs generated from drought stressed *indica* rice (Nagina 22) seedlings has been reported by Reddy et al (2002). Out of these, 334 ESTs possessed no expressional evidence from any public database, indicating that they expressed during drought conditions only. Also, a comparison with transcriptome data of other diverse plants such as, *Arabidopsis*, maize, rice and barley, identified 589 putative stress responsive genes shared by all these species (Gorantla et al 2007). In addition to this, many other studies have identified ESTs related to salt and low temperature stress (Shiozaki et al 2005; de los Reyes et al 2003; Sahi et al 2003). Matsumura et al (1999) used SAGE technique to study global gene expression in rice. Ten thousand one hundred and twenty two tags were derived from 5921 expressed genes from rice out of which only 23.1% matched the expressed genes from databases. Differentially expressed genes between anaerobically treated and untreated seedlings were also identified. Additionally, MPSS datasets are available for *Arabidopsis*, rice, grape and *Magnaporthe grisea* (rice blast fungus) (http://mpss.udel.edu). The rice database comprises of 20 MPSS libraries derived from different tissues, abiotic stress (cold, salt and drought) treated tissues and developmental stages.

Microarray has been used to study the change in expression patterns of genes in response to various stress conditions. Expression profiles of salt-tolerant (Pokkali) and salt-sensitive (IR29) rice varieties in response to high salinity were investigated with microarray having 1728 cDNAs from library of salt-stressed roots. Quick up- or down-regulation of transcripts in Pokkali within an hour of stress was proposed to be responsible for the tolerance while this response was delayed in IR29 (Kawasaki et al 2001). To identify rice genes that are up-regulated by cold, drought, salt and ABA, Cy3- and Cy5-labelled probes of plants treated with these stress conditions were hybridized with microarrays including 1700 independent cDNAs from drought, cold and salt treated libraries. Further, stress-inducible expression of candidate genes was confirmed by RNA gel-blot and 73 genes were identified out of which 58 were novel genes (Rabbani et al 2003). To gain insight into the expression patterns of stress-responsive genes in different organs during drought and salinity conditions, whole-genome expression analysis in rice shoot, flag leaf and panicle was done using 70-mer oligomer microarray covering 36,926 unique genes or gene models (Zhou et al 2007). A significant number of genes involved in transcription and cell signaling were found to be differentially expressed under both drought and salt stress in an organ-specific manner. To investigate the difference in response of two drought-tolerant (IR57311 and LC-93-4) and two drought-sensitive (Nipponbare and Taipei 309) rice cultivars under long-term drought stress, expression profiles were analyzed with 20K NSF oligonucleotide microarray. More genes were found to be differentially expressed in sensitive than in tolerant cultivars. Also, drought stress induced senescence processes were more pronounced in sensitive than in tolerant cultivars (Degenkolbe et al 2009).

Plant roots are the main organ for uptake of water and nutrients from soil and they can transduce water-deficit signals to the plant most efficiently. Therefore to analyze the root transcriptome under different levels of drought stress, two pairs of near-isogenic lines (NILs) with a common genetic background (IR64) but with contrasting drought tolerance were studied using Agilent 4 × 44 K oligoarray. As the level of stress increased, the number of differentially expressed genes in all NILs was increased suggesting that more genes were affected by increasing stress. The highly tolerant NIL showed the highest number of differentially regulated genes involved in cell growth, hormone biosynthesis, metabolism, signaling etc. (Moumeni et al

2011). The common link among different stresses such as drought, salt, extreme temperature, nutrient deprivation, UV-B radiation and air pollutants, is that they all produce an oxidative burst with damaging effects on cellular macromolecules such as lipids, enzymes and DNA. To investigate the oxidative stress response of *japonica* (Nipponbare) and *indica* (93-11) variety of rice, methyl viologen (MV) as a reactive oxygen species (ROS) generating agent was applied and it was observed that 93-11 seedlings exhibited leaf senescence with severe lesions under MV treatment compared to Nipponbare. Microarray analysis identified 1,062 probe sets to be differentially expressed between these two cultivars. These probe sets were analyzed by gene ontology (GO) and highlighted with enrichment GO terms related to toxin and oxidative stress responses as well as other responses (Liu et al 2010).

Recently, mRNA-Seq (an Illumina cDNA sequencing application) has been used to analyse the transcriptome of rice seedlings treated with salt stress. Thirty six base pair reads were mapped on the rice genomic sequence and 76.9% to 80.9% of the reads were mapped uniquely to the rice genome. After piling up of these short reads mapped on the genome, a series of programs (Bowtie, TopHat and Cufflinks) predicted 2,795 (shoot) and 3,082 (root) genes currently unannotated in the Rice Annotation Project database. Among the unannotated genes, 213 (shoot) and 436 (root) genes were differentially expressed in response to salinity stress. To predict the functions of un-annotated genes, BLASTX search against UniProt and RefSeq identified 995 (shoot) and 1,052 (root) transcripts which had ORFs similar to those encoding functional proteins (Mizuno et al 2010).

As can be seen, a huge amount of transcriptome data is now available for analyzing the stress responsive regulation of genes (Table 1). It is necessary to use these resources to validate functions of candidate genes involved in abiotic stress regulation. Work has already begun and a list of select genes that have been picked up from any transcriptome data and functionally characterized is given in Table 2. This will ultimately lead to construction of a network of stress regulatory pathways and will improve the understanding of the mechanism of stress tolerance, enabling us to eventually generate stress-tolerant plants.

Molecular mapping for genetic enhancement

The large-scale validation and high-throughput genotyping of transcript-derived genic markers, particularly encoding the known or candidate genes, in natural germplasm collections and mapping/mutant populations have been initiated recently in rice, using novel high-throughput and cost-effective next-generation sequencing and array-based genotyping technologies. The enormous transcript sequences generated for diverse rice genotypes have the potential to develop a large number of genic microsatellite and single nucleotide polymorphism (SNP) markers distributed over 12 chromosomes of rice genome. This has led to the identification of about 58,845 EST-derived and 13,637 unigene-derived microsatellite markers and over one million EST-derived genic SNP markers so far in rice (Varshney et al 2002; Parida et al 2006). This includes the known cloned or candidate rice genes which have been used to find homologous sequences amongst diverse rice genotypes (Caicedo et al 2007; Ebana et al 2010).

Many studies have also correlated the genome-wide single nucleotide polymorphism (SNP) with differential transcript profiling pattern based on the expression level polymorphism (ELP, West et al 2006) and single feature polymorphism (SFP, Kumar et al 2007). The next-generation whole genome transcriptome sequencing using Illumina Solexa Genome Analyzer has identified about 65,000 SNP loci between *indica* and *japonica* rice genotypes (Lu et al 2010). Of these, 262 SNP loci in the imprinted transcripts show differential expression specifically in the embryo and endosperm of Nipponbare and 93-11 (Luo et al 2011). Rice is rich in germplasm resources with wealth of trait diversity (Rafalski and Morgante, 2004) and thus has utility in analysis of allelic diversity of transcripts. At present about 110,000 cultivated and wild rice accessions representing diverse agro-climatic regions of the world including India are available at International Rice Germplasm Collection (IRGC) and International Rice Research Institute (IRRI). A detailed phenotypic characterization of these readily available genetic resources of rice for important agronomic traits including tolerance to biotic and abiotic stresses, yield, nutrition, and grain quality have been initiated. In recent years, efforts have also been made for constructing the core collections in rice (Zhang et al 2011) by identifying the largest amount of genetic diversity with a minimum number of accessions.

A larger set of advanced generation bi-parental and back-cross mapping population e.g. RILs (recombinant inbred lines), NILs (near isogenic lines) and DHs (double haploids) derived from the parental rice genotypes contrasting for yield and stress tolerance traits are available in rice. Besides, a total of about 66,891 EMS (ethyl methanesulfonate), fast neutron and gamma ray mutant lines of IR64 (Wu et al 2005) and Nipponbare (Till et al 2007) have been generated in rice to identify point mutations or SNP sites having functional relevance for trait regulation (http://tilling.ucdavis.edu; http://www.iris.irri.org).

Table 1. Certain transcriptome resources available for analyzing stress responsive mechanisms

Transcriptome resource/ GEO* id	Sample description	Treatment	Reference
dbEST	Seedling at coleoptile, radicle and prophyll emergence stages (cv. CT6748-8-CA-17)	Germination under cold stress	de los Reyes et al (2003)
dbEST	1 week old seedling (indica cv. CSR 27, Pokkali, PB-1)	96 h salt stress	Sahi et al (2003)
dbEST	10 day old seedling (cv. Dee-Geo-Woo-Gen)	5 h salt stress treatment	Shiozaki et al (2005)
GSE14403	30 day old plant root (salt-tolerant FL478, Pokkali and IR63731, and salt-sensitive IR29)	3 day salt stress to 22 day old seedlings	Walia et al (2005)
GSE6600	14 day old rice seedling (indica cv. FR13A (tolerant) and IR24 (susceptible))	Salt stress for 15 min and 24 h	Data submitted by Kottapalli KR and Kikuchi S (2006)
dbEST	1 month old plant (indica cv. Nagina 22)	Drought treatment until leaf relative water content was 50-60%	Gorantla et al (2007)
GSE6362	14 day old rice seedling (indica cv. FR13A (tolerant) and IR24 (susceptible))	Submergence stress for 15 min and 24 h	Kottapalli et al (2007)
GSE6533	Shoot at 4 tiller stage, flag leaf and panicle at one-week before heading (indica cv. Minghui 63)	Drought and salt stress at different time points	Zhou et al (2007)
GSE6901	7 day old rice seedling (indica cv. IR64)	3 h drought, cold and salt stress	Ray et al (2007)
GSE6908	4 day old rice coleoptile (japonica cv.Nipponbare)	Germination under anoxia for 4 days	Lasanthi-Kudahettige et al (2007)
GSE4438	Rice crown and growing point tissue at panicle initiation stage (sensitive japonica, m103, tolerant japonica agami, sensitive indica IR29 and tolerant indica IR63731)	Salt stress	Walia et al (2007)
GSE4471	1 week old seedling root (cv. Azucena and Bala)	Arsenate treatment	Norton et al (2008)
E-MEXP-2267	Seedling development stage (cv. Amaroo)	Anaerobic germination up to 24 hr	Narsai et al (2009)
GSE14275	14 day old rice seedling (japonica cv. ZhongHua 11)	Heat stress at 420 Cfor 3 h	Hu et al (2009)
GSE16108	10 day old seedling (CSR27 and MI48 parental lines, tolerant bulk and susceptible bulk RILs)	24 h salt stress	Pandit et al (2010)
GSE20345	Developing caryopsis (cv. Nipponbare)	15 day high temperature stress	Yamakawa and Hakata (2010)
GSE20746	7 day old rice seedling shoot and root (cv. Nipponbare)	1 h salt treatment	Mizuno et al (2010)
GSE24048	Second youngest fully expanded leaf of 83 day old field grown plant (cv. Azucena and Bala)	24 day old drought stress starting from 59 days after sowing	Data submitted by Price AH, Jones HG, Norton GJ, Huang S and Xiong L (2010)
E-MEXP-2401	14-day old seedling (indica cv. Nagina 22 and IR64)	Drought stress treatment until leaf rolling was observed	Lenka et al (2011)
GSE26280	Leaves and roots at tillering stage and panicle elongation stage, leaves and young panicle at booting stage (DK151, a drought tolerant line from indica cv. IR64)	Drought stress treatment until leaf relative water content was 65%-75%	Wang et al (2011a)

*GEO = Gene expression omnibus

Table 2. Selected recent examples of genetic modification of rice using genes derived from transcriptome resource

Gene	Transcriptome resource used	Target organism	System*	Activity	Reference
OsiSAP1 (Stress associated protein)	Northern blotting	tobacco	OE	Abiotic stress tolerance	Mukhopadhyay et al (2004)
SNAC1 (NAC transcription factor)	Microarray	rice	OE	Improved drought and salinity tolerance	Hu et al (2006)
OsMYB3R-2 (MYB transcription factor)	Microarray	*Arabidopsis*	OE	Abiotic stress tolerance	Dai et al (2007)
OsMKP1 (mitogen-activated protein kinase (MAPK) phosphatases)	FL-cDNAs, EST	rice	M	Negative regulation of rice wound responses	Katou et al (2007)
OsUGE-1 (UDP-glucose 4-epimerase)	Microarray	*Arabidopsis*	OE	Abiotic stress tolerance	Liu et al (2007)
OsNAC6 (NAC transcription factor)	Microarray	rice	OE	Tolerance to dehydration and high salt stress; growth retardation and low reproductive yields	Nakashima et al (2007)
OsIRO2 (bHLH transcription factor)	Microarray	rice	R, OE	Positive regulator of Fe deficiency responses	Ogo et al (2007)
OsLEA3-1 (LEA protein)	Microarray	rice	OE	Tolerance to drought stress	Xiao et al (2007)
ZFP177 (Stress associated protein)	Microarray	tobacco	OE	Differential responses to various abiotic stresses	Huang et al (2008)
OsiSAP8 (Stress associated protein)	Microarray	tobacco, rice	OE	Abiotic stress tolerance	Kanneganti and Gupta (2008)
OsbZIP23 (bZIP transcription factor)	Microarray	rice	OE, M	ABA-dependent drought and salinity tolerance	Xiang et al (2008)
OsDHODH1 (dihydroorotate dehydrogenase)	Microarray, EST	rice, *E.coli*	R, OE	Tolerance to drought and salt	Liu et al (2009)
AP37 (APETELA2 transcription factor)	Microarray	rice	OE	Tolerance to drought stress	Oh et al (2009)
ONAC045 (NAC transcription factor)	Microarray	rice	OE	Tolerance to drought and salt stress	Zheng et al (2009)
OsABF2 (AREB transcription factor)	Microarray	rice	M	Abiotic stress tolerance, ABA signalling	Hossain et al (2010)
OsABF1 (AREB transcription factor)	Microarray	rice	M	Abiotic stress tolerance	Hossain et al (2010)
OsNAC10 (NAC transcription factor)	Microarray	rice	OE	Tolerance to drought stress	Jeong et al (2010)
ZFP179 (Zinc finger protein)	Microarray	rice	OE	Tolerance to salt stress	Sun et al (2010)
OsNAC5 (NAC transcription factor)	Microarray, northern blotting	rice	OE	abiotic stress tolerance	Takasaki et al (2010)
OsSDIR1 (RING E3-ligase)	Northern blotting	rice, *Arabidopsis*	OE	ABA sensitivity in Arabidopsis , drought tolerance in rice	Gao et al (2011)

Gene	Transcriptome resource used	Target organism	System*	Activity	Reference
OsRLCK253 (Receptor like cytoplasmic kinase)	Microarray	*Arabidopsis*	OE	Tolerance to water deficit and salt stress	Giri et al (2011)
OsMAPK33 (Mitogen activated protein kinase)	EST	rice	R, OE	Sensitivity to salt stress	Lee et al (2011)
OsbHLH148 (bHLH transcription factor)	Microarray	rice	OE	Tolerance to drought	Seo et al (2011)
OsMSR2 (Calmodulin like protein)	Microarray	*Arabidopsis*	OE	Tolerance to drought and salt, sensitivity to ABA	Xu et al (2011)
OsCPK12 (Calcium dependent protein kinase)	Microarray	rice	R, OE	Tolerance to salt, susceptible to blast fungus	Asano et al (2012)
OsCBSX4 (CBS domain containing protein)	Microarray	tobacco	OE	Tolerance to salt, oxidative, heavy metal stress	Singh et al (2012)
Osmyb4 (MYB transcription factor)	Microarray	barley	OE	Tolerance to frost, improved germination	Soltész et al (2012)
OsMYB2 (MYB transcription factor)	Microarray	rice	OE	Tolerance to cold, salt, dehydration	Yang et al (2012)

*OE = overexpression; R = RNAi silencing; M = mutant

Considering the importance of high-throughput and precision phenotyping for gene isolation and marker-assisted breeding, the automated modern high-throughput phenotyping and E-typing platforms have recently been developed for precise phenotyping and multi-environmental assays of a larger set of rice natural or mutant and mapping populations for complex yield component and abiotic stress tolerance traits, respectively (Clark et al 2011; Xu et al 2012b). The TILLING (targeting induced local lesions in genomes, Till et al (2007)) and EcoTILLING (Raghavan et al 2007; Till et al 2007) using the diverse natural rice germplasm collections and mutant population, respectively, has made it possible to mine novel functional allelic variants in candidate transcripts.

A subset of these mined functional allelic variants has been correlated with traits of agricultural importance including drought (Yu et al 2012) and salinity (Negrao et al 2011) tolerance in rice. Remarkably, using the TILLING approach, about 6,000 mutated loci (novel SNPs and alleles) in the transcripts encoding the known or candidate genes have been identified from the MNU (methyl nitrosourea) and sodium azide induced selected mutants of Nipponbare at a genome-wide scale (Till et al 2007). The recent development of high-throughput array-based next-generation sequencing and marker genotyping technology such as automated fragment anlyzer, Illumina GoldenGate and Infinium assay and KASP (KBioScience Allele-Specific Polymorphism) profiling has expedited the evaluation of functional allelic variation or diversity level in the naturally occurring germplasm and mutant collections and thus could significantly improve the productivity and sustainability of rice agriculture.

The construction of high-resolution functional transcript maps, fine mapping of genes and QTLs and map-based cloning and positional cloning have traditionally proved to be the most powerful tools for gene isolation and dissection of the complex quantitative yield and stress tolerance traits in rice. Utilizing the whole genome transcriptome sequencing based high-throughput SNP genotyping assays in RILs, recently ultra-high-density linkage or transcript maps have been constructed in rice (Xie et al 2010; Wang et al 2011b) which successfully identified and mapped genes and QTLs at a high resolution scale in the transcript sequences regulating many important agronomic traits (Yu et al 2011). So far, about 125 genes for regulatory QTLs involved in important agronomic traits including grain yield and quality, stress tolerance and nutrient-use efficiency have been isolated and characterized using the positional cloning strategy based on fine-mapping of QTLs (Jiang et al 2011). However, such strategies are time consuming and provide difficulties in isolating causal genes of the QTLs, especially with minor effect. Recently, the integration of RIL population-based QTL mapping and microarray-based transcriptome profiling involving the parents and bulks of homozygous RILs is found to be a powerful approach to narrow down and pin-point the candidate genes underlying the QTLs of interest regulating the traits like seedling vigor (Yano et al 2012), grain number (Deshmukh et al 2010), and

salinity tolerance (Pandit et al 2010) in rice. The "genetical genomics" or "expression genetics" integrating the genetic or QTL mapping with transcript profiling has been demonstrated to be an effective approach for identification of candidate genes encoding transcripts and its regulatory sequences involved in expression of an individual trait in crop plants (Emilsson et al 2008). Using such and integrated approach, several recent studies have mapped the transcripts showing differential expression in the whole rice genome. Further, correlation of the transcript profiling with QTL mapping has been used to identify 'expression QTLs' (eQTLs) involved in the trait of interest. The candidate gene-based and genome-wide association mapping relies on the large-scale genotyping information of genic functional microsatellite and SNP markers. The recent discovery of low cost whole genome next-generation transcriptome sequencing and high-throughput genotyping technologies and novel advanced structural, functional and comparative genomic tools, have proven to be an effective approach. This has led to the identification of genes and alleles regulating complex quantitative yield and stress tolerance traits in rice (Zhao et al 2011; Li et al 2011). Specifically, the integrated approach of genome-wide and candidate gene-based association mapping and traditional bi-parental linkage mapping have been demonstrated as efficient approaches for precise identification of yield component trait related genes in rice (Mao et al 2010; Li et al 2011).

The natural genetic variation, particularly for yield component traits, scanned in a larger set of rice germplasm lines including landraces and wild species, has been transferred into the cultivated genetic background of rice for crop improvement. This has been achieved through introgression of favourable genes, QTLs or chromosomal segments using traditional approaches like introgression lines (ILs) and advanced-backcross QTL (AB-QTL) analysis, as well as modern methodologies such as association genetics and multi-parent advanced generation intercross (MAGIC) population (Tan et al 2008).

The trait-specific favourable genomic regions, superior functional genes or QTLs with major as well as minor effects and natural allelic variants once identified or validated, can be easily combined and pyramided in the selected rice genotypes of interest. This is done by the conventional breeding methods like marker-assisted selection (MAS) and marker-assisted back-crossing (MABC) or marker-assisted foreground and background selection. Also, novel approaches such as marker-assisted recurrent selection (MARS) and genomic or genome-wide (haplotype) selection for genetic enhancement in rice can be used. In summary, the transcriptome resources have utility in developing large-scale genic markers, establishing marker-trait linkages and rapid identification of favourable novel genes, QTLs and superior alleles, regulating the simpler qualitative as well as complex quantitative traits of agricultural importance. They can also be used for genetic enhancement through individual or integrated approach of positional cloning, association mapping and genetical genomics. Thus, this can expedite high-throughput marker-assisted breeding, eventually leading to development of superior high-yielding stress tolerant rice varieties with enhanced nutritional value.

Prospects

Analysis of transcriptome profiles is becoming cost effective and can be conducted at quantitative level with multiple genotypes unravelling hitherto unknown basis of diversity. This also needs to be extended to small RNAs and microRNAs leading to a better understanding of regulatory net-works. Epigenetic components involved in control of differential expression may provide additional useful targets for genetic enhancement. A combination of information on transcriptome, exome, promoters and epigenome with diversity analysis would provide useful specific targets in the genome for association with desired traits and their deployment for genetic enhancement by molecular breeding and transgenics approaches.

ACKNOWLEDGEMENT

Our work on rice genomics is supported by the Department of Biotechnology, Government of India.

REFERENCES

Asano T, Hayashi N, Kobayashi M, Aoki N, Miyao A, Mitsuhara I, Ichikawa H, Komatsu S, Hirochika H, Kikuchi S and Ohsugi R. 2012. A rice calcium-dependent protein kinase *OsCPK12* oppositely modulates salt-stress tolerance and blast disease resistance. *Plant J* 69**:** 26-36.

Aya K, Suzuki G, Suwabe K, Hobo T, Takahashi H, Shiono K, Yano K, Tsutsumi N, Nakazono M, Nagamura Y, Matsuoka M and Watanabe M. 2011. Comprehensive network analysis of anther-expressed genes in rice by the combination of 33 laser microdissection and 143 spatiotemporal microarrays. *PLoS One* 6: e26162.

Bao J, Lee S, Chen C, Zhang X, Zhang Y, Liu S, Clark T, Wang J, Cao M, Yang H, Wang S M and Yu J. 2005. Serial analysis of gene expression study of a hybrid rice strain (LYP9) and its parental cultivars. *Plant Physiol* 138: 1216-1231.

Birchler JA, Auger DL and Riddle NC. 2003. In search of the molecular basis of heterosis. *Plant Cell* 15: 2236-2239.

Caicedo AL, Williamson SH, Hernandez RD, Boyko A, Fledel-Alon A, York TL, Polato NR, Olsen KM, Nielsen R, McCouch SR, Bustamante CD and Purugganan MD. 2007. Genome-wide patterns of nucleotide polymorphism in domesticated rice. *PLoS Genet* 3**:** e163.

Chen ZJ. 2007. Genetic and epigenetic mechanisms for gene expression and phenotypic variation in plant polyploids. *Ann Rev Plant Biol* 58: 377-406.

Clark R, Maccurdy R, Jung J, Shaff J, McCouch SR, Aneshansley D and Kochian L. 2011. 3-Dimensional root phenotyping with a novel imaging and software platform. *Plant Physiol* 156**:** 455-465.

Dai X, Xu Y, Ma Q, Xu W, Wang T, Xue Y and Chong K. 2007. Overexpression of an R1R2R3 MYB gene, *OsMYB3R-2*, increases tolerance to freezing, drought, and salt stress in transgenic *Arabidopsis. Plant Physiol* 143**:** 1739-1751.

Dai X, Wang Y, Yang A and Zhang W H. 2012. OsMYB2P-1, an R2R3 MYB transcription factor, is involved in the regulation of phosphate-starvation responses and root architecture in rice. *Plant Physiol* 159: 169-183.

Darwin C. 1876. Difference between crossed and self-fertilised plants in constitutional vigour and in other respects, In: *The effects of cross and self fertilisation in the vegetable kingdom*. London. John Murray.

Davenport C B. 1908. Degeneration, albinism and inbreeding. *Science* 28: 454-455.

de los Reyes BG, Morsy M, Gibbons J, Varma TS, Antoine W, McGrath JM, Halgren R and Redus M. 2003. A snapshot of the low temperature stress transcriptome of developing rice seedlings (*Oryza sativa* L.) via ESTs from subtracted cDNA library. *Theor Appl Genet* 107**:** 1071-1082.

Degenkolbe T, Do PT, Zuther E, Repsilber D, Walther D, Hincha DK and Kohl KI. 2009. Expression profiling of rice cultivars differing in their tolerance to long-term drought stress. *Plant Mol Biol* 69**:** 133-153.

Deshmukh R, Singh A, Jain N, Anand S, Gacche R, Singh A, Gaikwad K, Sharma T, Mohapatra T and Singh N. 2010. Identification of candidate genes for grain number in rice (*Oryza sativa* L.). *Funct Integr Genomics* 10**:** 339-347.

Deveshwar P, Bovill WD, Sharma R, Able JA and Kapoor S. 2011. Analysis of anther transcriptomes to identify genes contributing to meiosis and male gametophyte development in rice. *BMC Plant Biol* 11: 78.

Ebana K, Yonemaru JI, Fukuoka S, Iwata H, Kanamori H, Namiki N, Nagasaki H and Yano M. 2010. Genetic structure revealed by a whole-genome single-nucleotide polymorphism survey of diverse accessions of cultivated Asian rice (*Oryza sativa* L.). *Breed Sci* 60**:** 390-397.

Eichten SR, Swanson-Wagner RA, Schnable JC, Waters AJ, Hermanson PJ, Liu S, Yeh C-T, Jia Y, Gendler K, Freeling M, Schnable PS, Vaughn MW and Springer NM. 2011. Heritable epigenetic variation among maize inbreds. *PLoS Genet* 7: e1002372.

Emilsson V, Thorleifsson G, Zhang B, Leonardson AS, Zink F, Zhu J, Carlson S, Helgason A, Walters GB, Gunnarsdottir S, Mouy M, Steinthorsdottir V, Eiriksdottir GH, Bjornsdottir G, Reynisdottir I, Gudbjartsson D, Helgadottir A, Jonasdottir A, Jonasdottir A, Styrkarsdottir U, Gretarsdottir S, Magnusson KP, Stefansson H, Fossdal R, Kristjansson K, Gislason HG, Stefansson T, Leifsson BG, Thorsteinsdottir U, Lamb JR, Gulcher JR, Reitman ML, Kong A, Schadt EE and Stefansson K. 2008. Genetics of gene expression and its effect on disease. *Nature* 452**:** 423-428.

Fu Z, Jin X, Ding D, Li Y and Tang J. 2011. Proteomic analysis of heterosis during maize seed germination. *Proteomics* 11:1462-72.

Fujita M, Horiuchi Y, Ueda Y, Mizuta Y, Kubo T, Yano K, Yamaki S, Tsuda K, Nagata T, Niihama M, Kato H, Kikuchi S, Hamada K, Mochizuki T, Ishimizu T, Iwai H, Tsutsumi N and Kurata, N. 2010. Rice expression atlas in reproductive development. *Plant Cell Physiol* 51: 2060-2081.

Furutani I, Sukegawa S and Kyozuka J. 2006. Genome-wide analysis of spatial and temporal gene expression in rice panicle development. *Plant J* 46: 503-511.

Gao T, Wu Y, Zhang Y, Liu L, Ning Y, Wang D, Tong H, Chen S, Chu C and Xie Q. 2011. *OsSDIR1* overexpression greatly improves drought tolerance in transgenic rice. *Plant Mol Biol* 76**:** 145-156.

Ge X, Chen W, Song S, Wang W, Hu S and Yu J. 2008. Transcriptomic profiling of mature embryo from an elite super-hybrid rice LYP9 and its parental lines. *BMC Plant Biol* 8: 114.

Giri J, Vij S, Dansana PK and Tyagi AK. 2011. Rice A20/AN1 zinc-finger containing stress-associated proteins (SAP1/11) and a receptor-like cytoplasmic kinase (OsRLCK253) interact via A20 zinc-finger and confer abiotic stress tolerance in transgenic *Arabidopsis* plants. *New Phytol* 191**:** 721-732.

Gorantla M, Babu PR, Lachagari VB, Reddy AM, Wusirika R, Bennetzen JL and Reddy AR. 2007. Identification of stress-responsive genes in an *indica* rice (*Oryza sativa* L.) using ESTs generated from drought-stressed seedlings. *J Exp Bot* 58**:** 253-265.

Groszmann M, Greaves I K, Albertyn Z I, Scofield G N, Peacock W J and Dennis E S. 2011. Changes in 24-nt siRNA levels in *Arabidopsis* hybrids suggest an epigenetic contribution to hybrid vigor. *Proc Natl Acad Sci USA* 108: 2617-2622.

He G, Zhu X, Elling A A, Chen L, Wang X, Guo L, Liang M, He H, Zhang H, Chen F, Qi Y, Che, R and Deng X W. 2010. Global epigenetic and transcriptional trends among two rice subspecies and their reciprocal hybrids. *Plant Cell* 22: 17-33.

Hirano, K, Aya, K, Hobo, T, Sakakibara, H, Kojima, M, Shim, R.A, Hasegawa, Y, Ueguchi-Tanaka, M and Matsuoka, M. 2008. Comprehensive transcriptome analysis of phytohormone biosynthesis and signaling genes in microspore/pollen and tapetum of rice. *Plant Cell Physiol* 49: 1429-1450.

Hobo T, Suwabe K, Aya K, Suzuki G, Yano K, Ishimizu T, Fujita M, Kikuchi S, Hamada K, Miyano M, Fujioka T, Kaneko F, Kazama T, Mizuta Y, Takahashi H, Shiono K, Nakazono M, Tsutsumi N, Nagamura Y, Kurata N, Watanabe M and Matsuoka M. 2008. Various spatiotemporal expression profiles of anther-expressed genes in rice. *Plant Cell Physiol* 49: 1417-1428.

Hoecker N, Lamkemeyer T, Sarholz B, Paschold A, Fladerer C, Madlung J, Wurster K, Stahl M, Piepho H-P, Nordheim A and Hochholdinger F. 2008. Analysis of nonadditive protein accumulation in young primary roots of a maize (*Zea mays* L.) F_1-hybrid compared to its parental inbred lines. *Proteomics* 8: 3882-3894.

Hossain MA, Cho JI, Han M, Ahn CH, Jeon JS, An G and Park PB. 2010. The ABRE-binding bZIP transcription factor OsABF2 is a positive regulator of abiotic stress and ABA signaling in rice. *J Plant Physiol* 167**:** 1512-1520.

Hu H, Dai M, Yao J, Xiao B, Li X, Zhang Q and Xiong L. 2006. Overexpressing a NAM, ATAF, and CUC (NAC) transcription factor enhances drought resistance and salt tolerance in rice. *Proc Natl Acad Sci USA* 103**:** 12987-12992.

Hu W, Hu G and Han B. 2009. Genome-wide survey and expression profiling of heat shock proteins and heat shock factors revealed overlapped and stress specific response under abiotic stresses in rice. *Plant Sci* 176**:** 583-590**.**

Huang J, Wang MM, Jiang Y, Bao YM, Huang X, Sun H, Xu DQ, Lan HX and Zhang HS. 2008. Expression analysis of rice A20/AN1-type zinc finger genes and characterization of *ZFP177* that contributes to temperature stress tolerance. *Gene* 420**:** 135-144.

Huang Y, Li L, Chen Y, Li X, Xu C, Wang S and Zhang Q. 2006a. Comparative analysis of gene expression at early seedling stage between a rice hybrid and its parents using a cDNA microarray of 9198 uni-sequences. *Sci China C Life Sci* 49: 519-529.

Huang Y, Zhang L, Zhang J, Yuan D, Xu C, Li X, Zhou D, Wang S and Zhang Q. 2006b. Heterosis and polymorphisms of gene expression in an elite rice hybrid as revealed by a microarray analysis of 9198 unique ESTs. *Plant Mol Biol* 62: 579-591.

Hui-Yong Z, Hang H, Liang-Bi C, Lei L, Man-Zhong L, Xiang-Feng W, Xi-Gang L, Guang-Ming H, Run-Sheng C, Li-Geng M and XingWang D. 2008. A genome-wide transcription analysis reveals a close correlation of promoter indel polymorphism and heterotic gene expression in rice hybrids. *Mol Plant* 1: 720-731.

Ito Y, Chujo A, Eiguchi M and Kurata N. 2004. Radial axis differentiation in a globular embryo is marked by *HAZ1*, a PHD-finger homeobox gene of rice. *Gene* 331: 9-15.

Jeong JS, Kim YS, Baek KH, Jung H, Ha SH, Do Choi Y, Kim M, Reuzeau C and Kim JK. 2010. Root-specific expression of *OsNAC10* improves drought tolerance and grain yield in rice under field drought conditions *Plant Physiol* 153**:** 185-197.

Jiang Y, Cai Z, Xie W, Long T, Yu H and Zhang Q. 2011. Rice functional genomics research: Progress and implications for crop genetic improvement. *Biotechnol Adv* (first online) DOI:10.1016/j.biotechadv.2011.08.013.

Jiao Y, Tausta S L, Gandotra N, Sun N, Liu T, Clay N K, Ceserani T, Chen M, Ma L, Holford M, Zhang H Y, Zhao H, Deng X W and Nelson T. 2009. A transcriptome atlas of rice cell types uncovers cellular, functional and developmental hierarchies. *Nat Genet* 41: 258-263.

Kanneganti V and Gupta AK. 2008. Overexpression of *OsiSAP8*, a member of stress associated protein (SAP) gene family of rice confers tolerance to salt, drought and cold stress in transgenic tobacco and rice. *Plant Mol Biol* 66**:** 445-462.

Katou S, Kuroda K, Seo S, Yanagawa Y, Tsuge T, Yamazaki M, Miyao A, Hirochika H and Ohashi Y. 2007. A calmodulin-binding mitogen-activated protein kinase phosphatase is induced by wounding and regulates the activities of stress-related mitogen-activated protein kinases in rice. *Plant Cell Physiol* 48**:** 332-344.

Kawasaki S, Borchert C, Deyholos M, Wang H, Brazille S, Kawai K, Galbraith D and Bohnert HJ. 2001. Gene expression profiles during the initial phase of salt stress in rice. *Plant Cell* 13**:** 889-905.

Kikuchi S, Satoh K, Nagata T, Kawagashira N, Doi K, Kishimoto N, Yazaki J, Ishikawa M, Yamada H, Ooka H, Hotta I, Kojima K, Namiki T,

Ohneda E, Yahagi W, Suzuki K, Li CJ, Ohtsuki K, Shishiki T; Foundation of Advancement of International Science Genome Sequencing & Analysis Group, Otomo Y, Murakami K, Iida Y, Sugano S, Fujimura T, Suzuki Y, Tsunoda Y, Kurosaki T, Kodama T, Masuda H, Kobayashi M, Xie Q, Lu M, Narikawa R, Sugiyama A, Mizuno K, Yokomizo S, Niikura J, Ikeda R, Ishibiki J, Kawamata M, Yoshimura A, Miura J, Kusumegi T, Oka M, Ryu R, Ueda M, Matsubara K; RIKEN, Kawai J, Carninci P, Adachi J, Aizawa K, Arakawa T, Fukuda S, Hara A, Hashizume W, Hayatsu N, Imotani K, Ishii Y, Itoh M, Kagawa I, Kondo S, Konno H, Miyazaki A, Osato N, Ota Y, Saito R, Sasaki D, Sato K, Shibata K, Shinagawa A, Shiraki T, Yoshino M, Hayashizaki Y and Yasunishi A, Rice Full-Length cDNA Consortium. 2003. Collection, mapping, and annotation of over 28,000 cDNA clones from *japonica* rice. *Science* 301**:** 376-379.

Kottapalli KR, Satoh K, Rakwal R, Shibato J, Doi K, Nagata T and Kikuchi S. 2007. Combining in silico mapping and arraying: an approach to identifying common candidate genes for submergence tolerance and resistance to bacterial leaf blight in rice. *Mol Cells* 24**:** 394-408.

Kumar R, Qiu J, Joshi T, Valliyodan B, Xu D and Nguyen HT. 2007. Single feature polymorphism discovery in rice. *PLoS ONE* 2**:** e284.

Kyndt, T, Denil, S, Haegeman, A, Trooskens, G, De Meyer, T, Van Criekinge, W and Gheysen, G. 2012. Transcriptome analysis of rice mature root tissue and root tips in early development by massive parallel sequencing. *J Exp Bot* 63: 2141-2157.

Lasanthi-Kudahettige R, Magneschi L, Loreti E, Gonzali S, Licausi F, Novi G, Beretta O, Vitulli F, Alpi A and Perata P. 2007. Transcript profiling of the anoxic rice coleoptile. *Plant Physiol* 144**:** 218-231.

Lee SK, Kim BG, Kwon TR, Jeong MJ, Park SR, Lee JW, Byun MO, Kwon HB, Matthews BF, Hong CB and Park SC. 2011. Overexpression of the mitogen-activated protein kinase gene *OsMAPK33* enhances sensitivity to salt stress in rice (*Oryza sativa* L.). *J Biosci* 36**:** 139-151.

Lenka SK, Katiyar A, Chinnusamy V and Bansal KC. 2011. Comparative analysis of drought-responsive transcriptome in *indica* rice genotypes with contrasting drought tolerance. *Plant Biotechnol J* 9**:** 315-327.

Li Y, Fan C, Xing Y, Jiang Y, Luo L, Sun L, Shao D, Xu C, Li X, Xiao J, He Y and Zhang Q. 2011. Natural variation in *GS5* plays an important role in regulating grain size and yield in rice. *Nat Genet* 43**:** 1266-1269.

Lippman, Z and Zamir, D. 2007. Heterosis: revisiting the magic. *Trends Genet* 23: 60-66.

Liu D, Lu Z, Mao Z and Liu S. 2009. Enhanced thermotolerance of *E coli* by expressed *OsHsp90* from rice (*Oryza sativa* L.). *Curr Microbiol* 58**:** 129-133.

Liu F, Xu W, Wei Q, Zhang Z, Xing Z, Tan L, Di C, Yao D, Wang C, Tan Y, Yan H, Ling Y, Sun C, Xue Y and Su Z. 2010. Gene expression profiles deciphering rice phenotypic variation between Nipponbare (*Japonica*) and 93-11 (*Indica*) during oxidative stress. *PLoS One* 5**:** e8632.

Liu HL, Dai XY, Xu YY and Chong K. 2007. Over-expression of *OsUGE-1* altered raffinose level and tolerance to abiotic stress but not morphology in *Arabidopsis*. *J Plant Physiol* 164**:** 1384-1390.

Lu T, Huang X, Zhu C, Huang T, Zhao Q, Xie K, Xiong L, Zhang Q and Han B. 2008. RICD: a rice *indica* cDNA database resource for rice functional genomics. *BMC Plant Biol* 8**:** 118.

Lu T, Lu G, Fan D, Zhu C, Li W, Zhao Q, Feng Q, Zhao Y, Guo Y, Li W, Huang X and Han B. 2010. Function annotation of the rice transcriptome at single-nucleotide resolution by RNA-seq genome. *Genome Res* 20**:** 1238-1249.

Luo M, Taylor JM, Spriggs A, Zhang H, Wu X, Russell S, Singh M and Koltunow A. 2011. A genome-wide survey of imprinted genes in rice seeds reveals imprinting primarily occurs in the endosperm. *PLoS Genet* 7:e1002125.

Mao H, Sun S, Yao J, Wang C, Yu S, Xu C, Li X and Zhang Q. 2010. Linking differential domain functions of the GS3 protein to natural variation of grain size in rice. *Proc Natl Acad Sci USA* 107**:** 19579-19584.

Matsumura H, Nirasawa S and Terauchi R. 1999. Technical advance: transcript profiling in rice (*Oryza sativa* L.) seedlings using serial analysis of gene expression (SAGE). *Plant J* 20**:** 719-726.

Mizuno H, Kawahara Y, Sakai H, Kanamori H, Wakimoto H, Yamagata H, Oono Y, Wu J, Ikawa H, Itoh T and Matsumoto T. 2010. Massive parallel sequencing of mRNA in identification of unannotated salinity stress-inducible transcripts in rice (*Oryza sativa* L.). *BMC Genomics* 11**:** 683.

Moumeni A, Satoh K, Kondoh H, Asano T, Hosaka A, Venuprasad R, Serraj R, Kumar A, Leung H and Kikuchi S. 2011. Comparative analysis of root transcriptome profiles of two pairs of drought-tolerant and susceptible rice near-isogenic lines under different drought stress. *BMC Plant Biol* 11**:** 174.

Mukhopadhyay A, Vij S and Tyagi AK. 2004. Overexpression of a zinc-finger protein gene from rice confers tolerance to cold, dehydration, and salt stress in transgenic tobacco. *Proc Natl Acad Sci USA* 101**:** 6309-6314.

Nakashima K, Tran LS, Van Nguyen D, Fujita M, Maruyama K, Todaka D, Ito Y, Hayashi N, Shinozaki K and Yamaguchi-Shinozaki K. 2007. Functional analysis of a NAC-type transcription factor *OsNAC6* involved in abiotic and biotic stress-responsive gene expression in rice. *Plant J* 51**:** 617-630.

Narsai R, Howell KA, Carroll A, Ivanova A, Millar AH and Whelan J. 2009. Defining core metabolic and transcriptomic responses to oxygen availability in rice embryos and young seedlings. *Plant Physiol* 151**:** 306-322.

Narsai R, Ivanova A, Ng S and Whelan J. 2010. Defining reference genes in *Oryza sativa* using organ, development, biotic and abiotic transcriptome datasets. *BMC Plant Biol* 10: 56.

Negrao S, Almadanim C, Pires I, McNally KL and Oliveira MM. 2011. Use of EcoTILLING to identify natural allelic variants of rice candidate genes involved in salinity tolerance. *Plant Genet Resour* 9**:** 300.

Ni, Z, Kim, E.-D, Ha, M, Lackey, E, Liu, J, Zhang, Y, Sun, Q and Chen, Z J. 2009. Altered circadian rhythms regulate growth vigour in hybrids and allopolyploids. *Nature* 457: 327-331.

Norton GJ, Lou-Hing DE, Meharg AA and Price AH. 2008. Rice-arsenate interactions in hydroponics: whole genome transcriptional analysis. *J Exp Bot* 59**:** 2267-2276.

Ogo Y, Itai RN, Nakanishi H, Kobayashi T, Takahashi M, Mori S and Nishizawa NK. 2007. The rice bHLH protein OsIRO2 is an essential regulator of the genes involved in Fe uptake under Fe-deficient conditions. *Plant J* 51**:** 366-377.

Oh SJ, Kim YS, Kwon CW, Park HK, Jeong JS and Kim JK. 2009. Overexpression of the transcription factor *AP37* in rice improves grain yield under drought conditions. *Plant Physiol* 150**:** 1368-1379.

Pandit A, Rai V, Bal S, Sinha S, Kumar V, Chauhan M, Gautam RK, Singh R, Sharma PC, Singh AK, Gaikwad K, Sharma TR, Mohapatra T and Singh NK. 2010. Combining QTL mapping and transcriptome profiling of bulked RILs for identification of functional polymorphism for salt tolerance genes in rice (*Oryza sativa* L.). *Mol Genet Genomics* 284**:** 121-136.

Parida SK, Rajkumar KA, Dalal V, Singh NK and Mohapatra T. 2006. Unigene derived microsatellite markers for the cereal genomes. *Theor Appl Genet* 112**:** 808-817.

Powers, L. 1945. Relative yields of inbred lines and F_1 hybrids of tomato. *Bot Gaz* 106: 247-268.

Rabbani MA, Maruyama K, Abe H, Khan MA, Katsura K, Ito Y, Yoshiwara K, Seki M, Shinozaki K and Yamaguchi-Shinozaki K. 2003. Monitoring expression profiles of rice genes under cold, drought, and high-salinity stresses and abscisic acid application using cDNA microarray and RNA gel-blot analyses. *Plant Physiol* 133**:** 1755-1767.

Rafalski A and Morgante M. 2004. Corn and humans: recombination and linkage disequilibrium in two genomes of similar size. *Trends Genet* 20**:** 103-111.

Raghavan C, Naredo MEB, Wang HH, Atienza G, Liu B, Qiu FL, McNally KL and Leung H. 2007. Rapid method for detecting SNPs on agarose gels and its application in candidate gene mapping. *Mol Breed* 19**:** 87-101.

Ray S, Agarwal P, Arora R, Kapoor S and Tyagi AK. 2007. Expression analysis of calcium-dependent protein kinase gene family during reproductive development and abiotic stress conditions in rice (*Oryza sativa* L ssp *indica*). *Mol Genet Genomics* 278**:** 493-505.

Reddy AR, Ramakrishna W, Sekhar AC, Ithal N, Babu PR, Bonaldo MF, Soares MB and Bennetzen JL. 2002. Novel genes are enriched in normalized cDNA libraries from drought-stressed seedlings of rice (*Oryza sativa* L subsp *indica* cv Nagina 22). *Genome* 45**:** 204-211.

Sahi C, Agarwal M, Reddy MK, Sopory SK and Grover A. 2003. Isolation and expression analysis of salt stress-associated ESTs from contrasting rice cultivars using a PCR-based subtraction method. *Theor Appl Genet* 106**:** 620-628.

Sato Y, Antonio B, Namiki N, Motoyama R, Sugimoto K, Takehisa H, Minami H, Kamatsuki K, Kusaba M, Hirochika H and Nagamura Y. 2011a. Field transcriptome revealed critical developmental and physiological transitions involved in the expression of growth potential in *japonica* rice. *BMC Plant Biol* 11: 10.

Sato Y, Antonio B.A, Namiki N, Takehisa H, Minami H, Kamatsuki K, Sugimoto K, Shimizu Y, Hirochika H, and Nagamura Y. 2011b. RiceXPro: a platform for monitoring gene expression in *japonica* rice grown under natural field conditions. *Nucleic Acids Res* 39: D1141-1148.

Seo JS, Joo J, Kim MJ, Kim YK, Nahm BH, Song SI, Cheong JJ, Lee JS, Kim JK and Choi YD. 2011. OsbHLH148, a basic helix-loop-helix protein, interacts with OsJAZ proteins in a jasmonate signaling pathway leading to drought tolerance in rice. *Plant J* 65**:** 907-921.

Sharma R, Agarwal P, Ray S, Deveshwar P, Sharma P, Sharma N, Nijhawan A, Jain M, Singh A.K,

Singh V.P, Khurana J.P, Tyagi A.K and Kapoor S. 2012. Expression dynamics of metabolic and regulatory components across stages of panicle and seed development in *indica* rice. *Funct Integr Genomics* 12**:** 229-248.

Shen H, He H, Li J, Chen W, Wang X, Guo L, Peng Z, He G, Zhong S, Qi Y, Terzaghi W and Deng X W. 2012. Genome-wide analysis of DNA methylation and gene expression changes in two *Arabidopsis* ecotypes and their reciprocal hybrids. *Plant Cell* 24: 875–892.

Shiozaki N, Yamada M and Yoshiba Y. 2005. Analysis of salt-stress-inducible ESTs isolated by PCR-subtraction in salt-tolerant rice. *Theor Appl Genet* 110**:** 1177-1186.

Shull GH. 1908. The composition of a field of maize. *J Heredity* 4: 296-301.

Singh AK, Kumar R, Pareek A, Sopory SK and Singla-Pareek SL. 2012. Overexpression of rice CBS domain containing protein improves salinity, oxidative, and heavy metal tolerance in transgenic tobacco. *Mol Biotechnol* doi: 10.1007/s12033-011-9487-2.

Soltesz A, Vagujfalvi A, Rizza F, Kerepesi I, Galiba G, Cattivelli L, Coraggio I and Crosatti C. 2012. The rice *Osmyb4* gene enhances tolerance to frost and improves germination under unfavourable conditions in transgenic barley plants. *J Appl Genet* 53**:** 133-143.

Sun SJ, Guo SQ, Yang X, Bao YM, Tang HJ, Sun H, Huang J and Zhang HS. 2010. Functional analysis of a novel Cys2/His2-type zinc finger protein involved in salt tolerance in rice. *J Exp Bot* 61**:** 2807-2818.

Swanson-Wagner R A, Jia Y, DeCook R, Borsuk L A, Nettleton D and Schnable PS. 2006. All possible modes of gene action are observed in a global comparison of gene expression in a maize F1 hybrid and its inbred parents. *Proc Natl Acad Sci USA* 103: 6805-6810.

Takasaki H, Maruyama K, Kidokoro S, Ito Y, Fujita Y, Shinozaki K, Yamaguchi-Shinozaki K and Nakashima K. 2010. The abiotic stress-responsive NAC-type transcription factor OsNAC5 regulates stress-inducible genes and stress tolerance in rice. *Mol Genet Genomics* 284**:** 173-183.

Tan L, Zhang P, Liu F, Wang G, Ye S, Zhu Z, Fu Y, Cai H and Sun C. 2008. Quantitative trait loci underlying domestication- and yield-related traits in an *Oryza sativa* x *Oryza rufipogon* advanced backcross population. *Genome* 51**:** 692-704.

Thakur J.K., Malik M.R., Bhatt V., Reddy M.K., Sopory S.K., Tyagi A.K. and Khurana J.P. 2003.. A POLYCOMB group gene of rice (*Oryza sativa* L. subspecies indica), *OsiEZ1*, codes for a nuclear-localized protein expressed preferentially in young seedlings and during reproductive development. *Gene* 314: 1-13.

Till BJ, Cooper J, Tai TH, Colowit P, Greene EA, Henikoff S and Comai L. 2007. Mutant resources in rice discovery of chemically induced mutations in rice by TILLING. *BMC Plant Biol* 7**:** 19.

Varshney RK, Thiel T, Stein N, Langridge P and Graner A. 2002. *In-silico* analysis on frequency and distribution of microsatellites in ESTs of some cereal species. Cell Mol Bio Lett **7:** 537-546.

Venu R, Sreerekha M, Nobuta K, Belo A, Ning Y, An G, Meyers BC and Wang GL. 2011. Deep sequencing reveals the complex and coordinated transcriptional regulation of genes related to grain quality in rice cultivars. *BMC Genomics* 12: 190.

Walia H, Wilson C, Condamine P, Liu X, Ismail AM, Zeng L, Wanamaker SI, Mandal J, Xu J, Cui X and Close TJ. 2005. Comparative transcriptional profiling of two contrasting rice genotypes under salinity stress during the vegetative growth stage. *Plant Physiol* 139**:** 822-835.

Walia H, Wilson C, Zeng L, Ismail AM, Condamine P and Close TJ. 2007. Genome-wide transcriptional analysis of salinity stressed *japonica* and *indica* rice genotypes during panicle initiation stage. *Plant Mol Biol* 63**:** 609-623.

Wang D, Pan Y, Zhao X, Zhu L, Fu B and Li Z. 2011a. Genome-wide temporal-spatial gene expression profiling of drought responsiveness in rice. *BMC Genomics* 12**:** 149.

Wang L, Wang A, Huang X, Zhao Q, Dong G, Qian Q, Sang T and Han B. 2011b. Mapping 49 quantitative trait loci at high resolution through sequencing-based genotyping of rice recombinant inbred lines. *Theor Appl Genet* 122**:** 327-340.

Wang L, Xie W, Chen Y, Tang W, Yang J, Ye R, Liu L, Lin Y, Xu C, Xiao J and Zhang Q. 2010. A dynamic gene expression atlas covering the entire life cycle of rice. *Plant J* 61: 752-766.

Wei LQ, Xu WY, Deng ZY, Su Z, Xue Y and Wang T. 2010. Genome-scale analysis and comparison of gene expression profiles in developing and germinated pollen in *Oryza sativa*. *BMC Genomics* 11: 338.

West MA, van Leeuwen H, Kozik A, Kliebenstein DJ, Doerge RW, St Clair DA and Michelmore RW. 2006. High-density haplotyping with microarray-based expression and single feature polymorphism markers in *Arabidopsis*. *Genome Res* 16:787-795.

Wu JL, Wu C, Lei C, Baraoidan M, Bordeos A, Madamba MR, Ramos-Pamplona M, Mauleon R, Portugal A, Ulat VJ, Bruskiewich R, Wang G, Leach J, Khush G and Leung H. 2005. Chemical and irradiation-induced mutants of *indica* rice IR64

for forward and reverse genetics. *Plant Mol Biol* 59**:** 85-97.

Xiang Y, Tang N, Du H, Ye H and Xiong L. 2008. Characterization of OsbZIP23 as a key player of the basic leucine zipper transcription factor family for conferring abscisic acid sensitivity and salinity and drought tolerance in rice. *Plant Physiol* 148**:** 1938-1952.

Xiao B, Huang Y, Tang N and Xiong L. 2007. Over-expression of a LEA gene in rice improves drought resistance under the field conditions. *Theor Appl Genet* 115**:** 35-46.

Xie W, Feng Q, Yu H, Huang X, Zhao Q, Xing Y, Yu S, Han B and Zhang Q. 2010. Parent-independent genotyping for constructing an ultrahigh-density linkage map based on population sequencing. *Proc Natl Acad Sci USA* 107**:** 10578-10583.

Xu GY, Rocha PS, Wang ML, Xu ML, Cui YC, Li LY, Zhu YX and Xia X. 2011. A novel rice calmodulin-like gene, *OsMSR2*, enhances drought and salt tolerance and increases ABA sensitivity in *Arabidopsis*. *Planta* 234**:** 47-59.

Xu H, Gao Y and Wang J. 2012a. Transcriptomic analysis of rice (*Oryza sativa*) developing embryos using the RNA-Seq technique. *PLoS One* 7: e30646.

Xu Y, Lu Y, Xie C, Gao S, Wan J and Prasanna B. 2012b. Whole-genome strategies for marker-assisted plant breeding. *Mol Breed* (first online) DOI: 10.1007/s11032-012-9699-6.

Xue LJ, Zhang JJ and Xue HW. 2012. Genome-wide analysis of the complex transcriptional networks of rice developing seeds. *PLoS One* 7: e31081.

Yamakawa H and Hakata M. 2010. Atlas of rice grain filling-related metabolism under high temperature: joint analysis of metabolome and transcriptome demonstrated inhibition of starch accumulation and induction of amino acid accumulation. *Plant Cell Physiol* 51**:** 795-809.

Yamamoto MP, Onodera Y, Touno SM and Takaiwa F. 2006. Synergism between RPBF Dof and RISBZ1 bZIP activators in the regulation of rice seed expression genes. *Plant Physiol* 141: 1694-1707.

Yang A, Dai X and Zhang WH. 2012. A R2R3-type MYB gene, *OsMYB2*, is involved in salt, cold, and dehydration tolerance in rice. *J Exp Bot* 63**:** 2541-2556.

Yang HJ, Shen H, Chen L, Xing YY, Wang ZY, Zhang JL and Hong MM. 2002. The *OsEBP-89* gene of rice encodes a putative EREBP transcription factor and is temporally expressed in developing endosperm and intercalary meristem. *Plant Mol Biol* 50: 379-391.

Yano K, Takashi T, Nagamatsu S, Kojima M, Sakakibara H, Kitano H, Matsuoka M and Aya K. 2012. Efficacy of microarray profiling data combined with QTL mapping for the identification of a QTL gene controlling the initial growth rate in rice. *Plant Cell Physiol* 53**:** 729-739.

Yin LL and Xue HW. 2012. The MADS29 transcription factor regulates the degradation of the nucellus and the nucellar projection during rice seed development. *Plant Cell* 24: 1049-1065.

Yu H, Xie W, Wang J, Xing Y, Xu C, Li X, Xiao J and Zhang Q. 2011. Gains in QTL detection using an ultra-high density SNP map based on population sequencing relative to traditional RFLP/SSR markers. *PLoS One* 6: e17595.

Yu S, Liao F, Wang F, Wen W, Li J, Mei H and Luo L. 2012. Identification of rice transcription factors associated with drought tolerance using the ECOTILLING method. *PLoS ONE* 7**:** e30765.

Zhang H, Zhang D, Wang M, Sun J, Qi Y, Li J, Wei X, Han L, Qiu Z, Tang S and Li Z. 2011. A core collection and mini core collection of *Oryza sativa* L in China. *Theor Appl Genet* 122**:** 49-61.

Zhao K, Tung CW, Eizenga GC, Wright MH, Ali ML, Price AH, Norton GJ, Islam MR, Reynolds A, Mezey J, McClung AM, Bustamante CD and McCouch SR. 2011. Genome-wide association mapping reveals a rich genetic architecture of complex traits in *Oryza sativa*. *Nat Commun* 2**:** 467.

Zheng X, Chen B, Lu G and Han B. 2009. Overexpression of a NAC transcription factor enhances rice drought and salt tolerance. *Biochem Biophys Res Commun* 379**:** 985-989.

Zhou J, Wang X, Jiao Y, Qin Y, Liu X, He K, Chen C, Ma L, Wang J, Xiong L, Zhang Q, Fan L and Deng XW. 2007. Global genome expression analysis of rice in response to drought and high-salinity stresses in shoot, flag leaf, and panicle. *Plant Mol Biol* 63**:** 591-608.

Citation: Agarwal P, Parida S, Kothari KS, Sharma G, Baranwal V, Kapoor S and Tyagi A. 2013. Transcriptome resources for function analysis and genetic enhancement of rice. In: Muralidharan K and Siddiq EA, eds. 2013. *International Dialogue on Perception and Prospects of Designer Rice*. Society for Advancement of Rice Research, Directorate of Rice Research, Hyderabad 500030, India, pp 9-24.