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# Genetic Characterization of Global Rice Germplasm for Sustainable Agriculture

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## 1. Introduction

Crop genebanks or germplasm collections store thousands of crop varieties. Each variety has unique genetic traits to be used in fighting diseases and insects, increasing yield and nutritional value and adjusting to environmental changes such as drought, soil salinity, etc. The Germplasm Resources Information Network (GRIN, 2011) of the United States (US) manages germplasm of plants, animals, microbes and invertebrates. Currently, there are 540,935 accessions of plant germplasm for 95,800 taxonomic names, 13,388 species of 2,208 genera along with 1,866,764 inventory records, 1,628,283 germination records, 7,291,757 characteristic records and 201,156 images in the GRIN (GRIN, 2011).

Rice is one of the most important food crops because it feeds more than half of the world's population (Yang and Hwa, 2008). There are some 4,500,000 accessions in plant germplasm collections worldwide (FAO, 1996), about 9% or 400,000 accessions are rice (Hamilton and Raymond, 2005). The United States Department of Agriculture (USDA) has started collecting rice germplasm from all over the world since the 1800s (Bockelman et al., 2002). At present, the global collection has 18,729 accessions of rice germplasm originated from 116 countries, stored in the National Small Grains Collection (NSGC, 2011) and managed by the GRIN. Great majority of these accessions (18,476 or 98.7%) belong to Asian cultivated species *Oryza sativa* in the US Department of Agriculture (USDA) rice germplasm collection. Africa cultivated species *Oryza glaberrima* has 175 accessions, and other nine species of *Oryza* have very few accessions ranging from 1 for *O. grandiglumis* to 19 for *O. glumipatula*. Some 94% of the accessions in the USDA rice germplasm collection were obtained internationally, and the remainder domestically (Yan et al., 2007). All public cultivars registered in the US can be entered in the collection. Foreign germplasm accessions must be grown for one generation in a plant quarantine greenhouse isolated from commercial rice growing areas to prevent accidental introduction of new disease and insect pests.

Evaluation of germplasm collections is essential for maintenance of the diversity and identification of valuable genes. The USDA-Agricultural Research Service (ARS) coordinates the National Plant Germplasm System (NPGS) and its related germplasm activities in the US, including germplasm acquisition, rejuvenation, storage, distribution, evaluation, and

enhancement (Bretting, 2007). The NPGS is a cooperative effort by public and private organizations to preserve the genetic diversity of plants. Crop Germplasm Committees (CGC), representing the federal, state, and private sectors in various scientific disciplines, determine the set of descriptors to be managed by GRIN for most crops. Rice CGC has requested 42 descriptors plus panicle and kernel images to characterize the collection (Rice Descriptors, 2011). The USDA-ARS Dale Bumpers National Rice Research Center (DBNRRRC) coordinates germplasm activities of rice including evaluation of the collection for the 42 descriptors and constantly updating the GRIN database. Furthermore, the DBNRRRC manages the Genetic Stocks - *Oryza* collection including more than 30,000 accessions of genetic materials donated from national and international research programs (GSOR, 2011).

Comprehensive evaluation of the collection for such a large number of descriptors has been hindered by the sheer number of accessions, particularly those involving grain quality and resistances to biotic and abiotic stresses which require sophisticated instruments and significant resources. It also is difficult to characterize such a large collection using molecular means. For practical evaluation and effective management of large collections in crops, the core collection concept was proposed in the 1980s (Brown, 1989).

## 2. USDA rice core collection

A core collection is a subset of a large germplasm collection that contains chosen accessions capturing most of the genetic variability within the entire gene bank (Brown, 1989). With the strategy of comprehensive evaluation and accurate analysis of the core collection, the genetic diversity of the collection can be assessed, genetic distances among the accessions can be estimated for identification of special divergent subpopulations, genetic gaps of the existing collection can be identified for planning acquisition strategies and joint analysis of phenotype and genotype can be conducted for molecular understanding of the collection (Steiner et al., 2001). These analyses can help users effectively find the traits in which they are interested along with molecular information. The information is useful for determining strategies for transferring desirable traits found in the collection into new commercial cultivars.

### 2.1 Establishment of the core collection

The USDA rice core subset (RCS) or collection was assembled by sampling from over 18,000 accessions in the working collection of the NSGC in 1998 and 2002, respectively (Yan et al., 2007). A method of stratification by country and then random sampling was adapted by: 1) recording the number of accessions from each country or region of origin; 2) calculating the logarithm (log) of the number of accessions from each country or region of origin; 3) randomly choosing the accessions within each country or region based on the relative log numbers, with a minimum of one accession per country or region; and 4) removing obvious duplications by plant introduction (PI) number and cultivar name. In addition to the stratified sampling, additional emphasis was placed on some newly introduced Chinese germplasm (Yan et al., 2002) and newly released accessions from quarantine programs (Yan et al., 2003). The resultant RCS consists of 1,794 entries from 112 countries and represents approximately 10% of the rice whole collection (RWC).

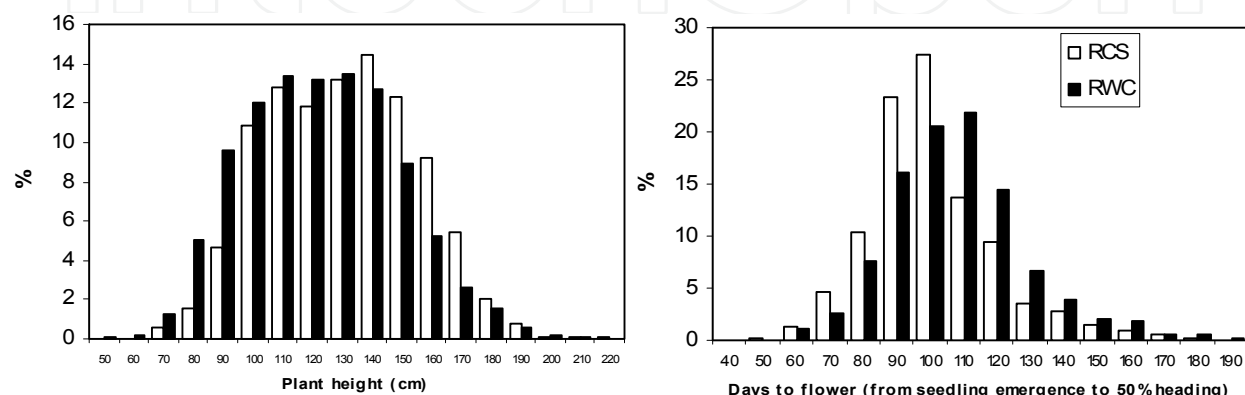
## 2.2 Evaluation of the core collection

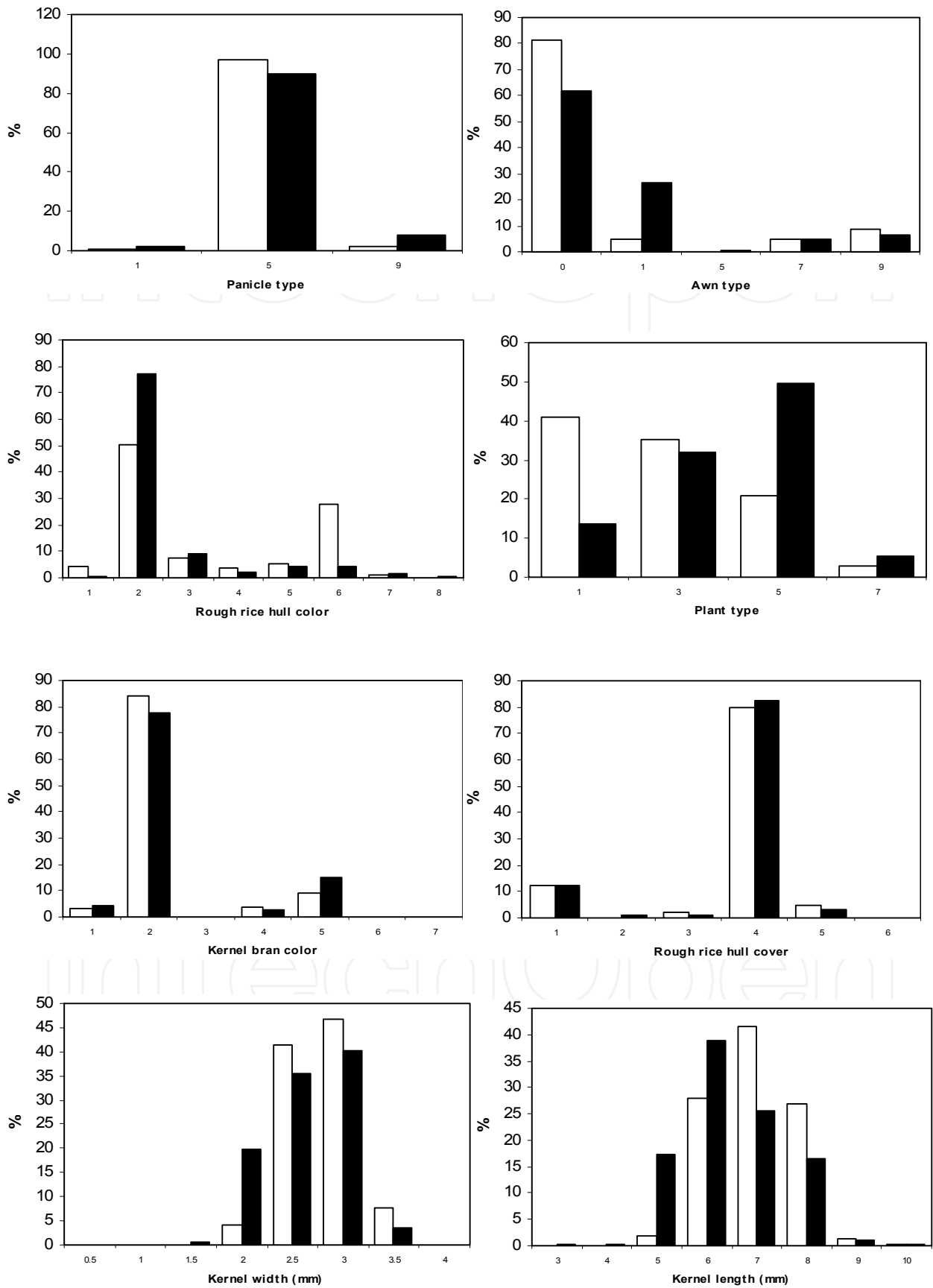
The RCS was evaluated at Stuttgart, Arkansas in 2002. Seeds of each accession were visually purified by seed shape and hull color as described in the GRIN before planting in a plot consisting of two rows, 0.3 m apart and 1.4 m long using a Hege 500 planter. Plots were separated by 0.9 m to avoid biological and mechanical contamination. A permanent flood was established after 67 kg ha<sup>-1</sup> of nitrogen as urea was applied at about 5-leaf stage.

Agronomic descriptors were recorded in the field using standard criteria described in the GRIN. Rough or paddy rice is the mature rice grain as harvested, and becomes brown rice when the hulls are removed. Rough and brown rice samples were analyzed on an automated grain image analyzer (GrainCheck 2312; Foss Tecator AB, Hoganas, Sweden) to determine rice kernel dimensions (length, width and length/width ratio), hull and seed pericarp (bran) colorations, and 1000 grain weight. Samples were milled for determination of apparent amylose content (Pérez and Juliano, 1978; Webb, 1972) and alkali spreading value (ASV) (Little et al., 1958). Fourteen important traits were selected for comparison with the whole collection.

## 2.3 Comparative study of the RCS with RWC

Statistical analysis was conducted using the univariate and correlation procedures of SAS statistical software, Version 9.1.3 (SAS Institute, 2004). Frequency distributions for each of 14 traits were determined using Microsoft Office Excel software. Frequency refers to how often data values occur within a range of values in an Excel bins-array that is an array of data intervals into which the data values are grouped. For example, days to flower had a bins-array of 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180 and 190 (Fig. 1), e.g., all accessions ranging from 36 to 45 days were grouped in bin 40. Frequencies (%) of the respective bins were 0.02, 0.05, 1.15, 2.91, 7.54, 16.01, 20.33, 21.16, 14.91, 6.65, 4.07, 2.29, 1.83, 0.48, 0.52 and 0.10 among 15,097 accessions in RWC, and 0, 0.24, 1.26, 4.56, 10.43, 23.38, 27.40, 13.73, 9.53, 3.54, 2.82, 1.50, 0.96, 0.48, 0.18 and 0 among 1,668 RCS entries that headed in the field (others failed to head). Paired frequencies of the RWC and the RCS on each bin were used for correlation analysis, which measures the correspondence between the two collections. The RCS data of 1,794 accessions were from above field evaluation the RWC data of ~15,000 accessions were extracted from the GRIN.





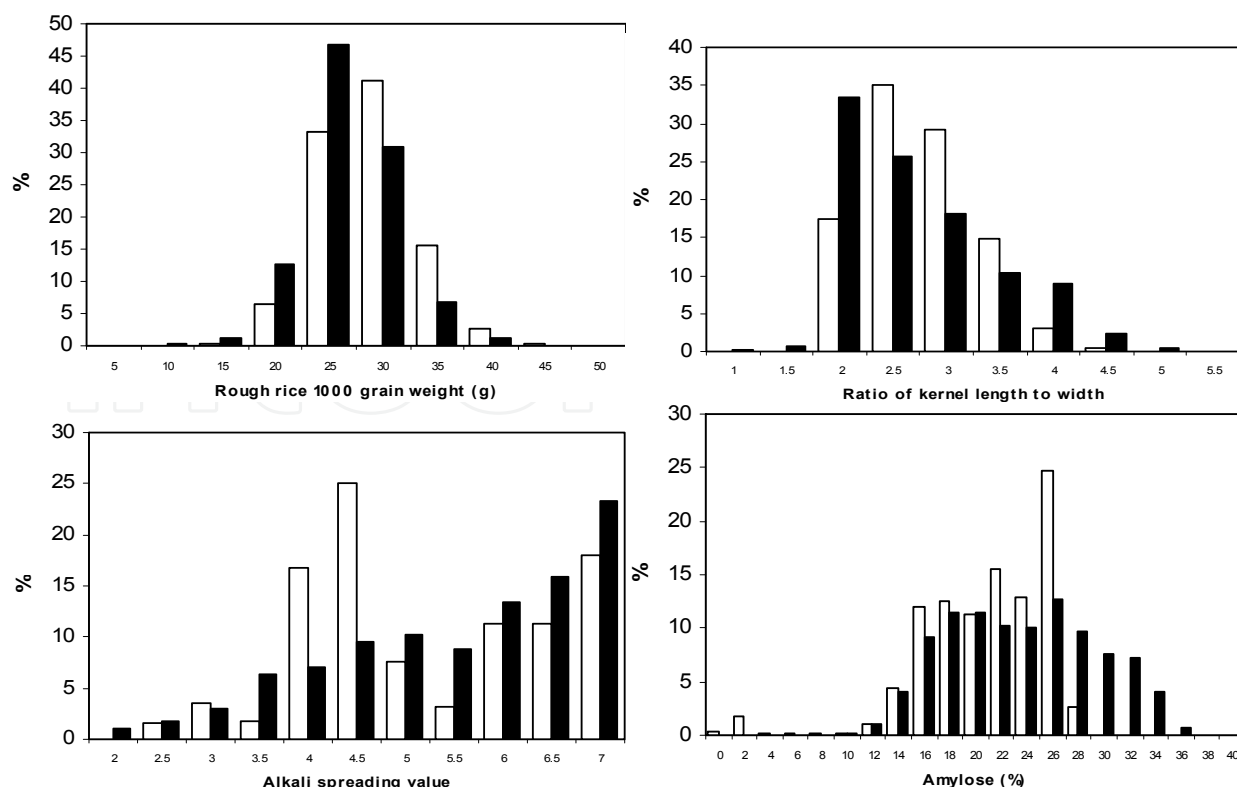


Fig. 1. Comparative distributions of frequency (%) for 14 traits of 1,794 core accessions field-evaluated in 2002 with ~15,000 accessions which data were extracted from the GRIN. Those with no unit are categorical traits, and their category classifications are explained in the GRIN, i.e. Awn type: 0-absent; 1-short and partly awned; 5- short and fully awned; 7-long and partly awned and 9-long and fully awned (Rice Descriptors, 2011).

#### 2.4 Frequency analysis of 14 traits proves that the RCS well represents the RWC

As displayed in Fig. 1, the correlation coefficient ( $r$ ) of the RCS distribution frequency with RWC was 0.90 for Days to flower, 0.93 for Plant height, 0.93 for Awn type, 0.99 for Panicle type, 0.69 for Plant type, 0.88 for Hull color, 0.99 for Hull cover, 0.99 for Bran color, 0.83 for Kernel length, 0.94 for Kernel width, 0.85 for Kernel length/width ratio, 0.91 for Grain weight, 0.82 for Amylose content and 0.65 for Alkali spreading value (Yan et al., 2007). Taken together, the 14 traits had a high correlation of distribution frequency ( $r=0.94$ ,  $P<0.0001$ ) between the RCS and RWC, resulting a determination coefficient ( $r^2$ ) of 0.88. The high correlation of the RCS with the RWC demonstrates that a stratified set of 10% of the accessions can be effectively used to assess the variability in the whole rice collection with 88% certainty. The correlation analysis validates the RCS to be well representative of the RWC for genetic assessment of global rice germplasm.

#### 2.5 The RCS improves genetic characterization of germplasm collection

In an effort to better characterize genetic diversity of the rice collection, the RCS with 10% of over 18,000 accessions in the whole collection is a reasonable size for replicated evaluations. As a result, this core subset has been evaluated for agronomic descriptors (Yan et al., 2005a), kernel dimension traits that impact milling yield and market class (Yan et al., 2005b),

resistance to physiological disease 'straighthead' (Agrama and Yan, 2010) and fungal disease 'sheath blight' (*Rhizoctonia solani*) (Jia et al., 2011) and 'blast' (*Magnaporthe oryzae*) (Agrama et al., 2009), and DNA markers associated with cooking quality and blast resistance (McClung et al., 2004, 2006; Fjellstrom et al., 2006).

### 3. Geographic analysis of global rice germplasm

#### 3.1 Genotyping and statistical analysis

Total genomic DNA was extracted using a rapid alkali extraction procedure (Xin et al., 2003) from a bulk of five plants derived from a single plant selected to represent each accession in the core collection. Seventy-two (71 SSR and an indel) molecular markers, covering the entire rice genome, approximately with an average of one marker per 30 cM, were used to genotype the 1,794 accessions. PCR amplification of the markers followed the procedure that was described by Agrama et al. (2009). DNA samples were separated on an ABI Prism 3730 DNA analyzer according to the manufacturer's instructions (Applied Biosystems, Foster City, CA, USA). Fragments were sized and binned into alleles using GeneMapper v. 3.7 software.

The 112 countries or districts from which the 1,794 accessions originated were classified into 14 geographic regions according to groupings of the United Nations Statistic Division (UNSD, 2009). Each accession was plotted on the global map using its latitude and longitude coordinates according to the GRIN passport database. The map was built using the 'prcomp' procedure in the statistics module (version 2.8.1) of the R statistical package including 'spatial', 'maps' and 'fields' (Venables and Ripley, 1998, Venables et al., 2008).

PowerMarker software (Liu and Muse, 2005) was used to calculate allele frequencies and polymorphism information content (PIC) values (Botstein et al., 1980) for each marker, region and country. Analysis of molecular variance (AMOVA; Excoffier et al., 1992) was conducted for variance components within and among regions and countries of origin, respectively, using ARLEQUIN 3.0 software (Schneider et al., 2000). Significance of variance components was tested using a non-parametric procedure based on 1,000 random permutations of individuals using the software ARLEQUIN 3.0 (Schneider et al., 2000). Genetic diversity was estimated using Nei diversity index for each accession according to Lynch and Milligan (1994). Geographical distribution of diversity index represented by Kriging methods was globally mapped using the R-script (François et al., 2008).

Genetic relationships among accessions represented by regions and countries were determined by the unweighted pair-group method with an arithmetic mean (UPGMA) analysis based on Nei (Nei, 1973) genetic similarity estimated using the 72 markers. The UPGMA trees were constructed from 1,000 bootstrap replicates using the software PowerMarker (Liu and Muse, 2005) and drawn with MEGA v. 3.1 (Kumar et al., 2004). The number of alleles, which are private to a population and do not exist in other populations, is especially informative when populations are studied with highly variable multi-allelic markers, such as SSRs (Szpiech et al., 2008). The average number of private alleles per locus for core accessions originating in each of 14 geographic regions was estimated using ADZE (Allelic Diversity AnalyZER) software (Szpiech et al., 2008) with the 72 molecular markers.

### 3.2 Allelic diversity among 14 geographic regions

A total of 1,005 alleles were revealed by 72 molecular markers, averaging 14 alleles per locus and ranging from 2 to 36. Polymorphic information content (PIC) values averaged  $0.66 \pm 0.02$  ranging from 0.17 to 0.92 with the majority distributed between 0.50 and 0.90. Sixty markers (83%) were highly informative ( $PIC > 0.50$ ), 10 (14%) reasonably informative ( $0.50 > PIC > 0.25$ ) and 2 (3%) slightly informative ( $PIC < 0.25$ ), demonstrating a high discriminatory power of these selected markers (Yan et al., 2010).

The 1,794 core accessions were introduced from 112 countries and distributed to 14 worldwide geographic regions with the most countries in Africa and the least in North America (Table 1). Accession number ranged from 57 in Oceania to 224 in South America. China had the most accessions (135), while 34 countries had less than five accessions each

Geographic region	Countries	Accessions	Alleles/ locus	PIC
Africa	26	198	9.32	0.64
Central America	12	116	8.01	0.59
Central Asia	7	61	6.71	0.59
China	4	212	8.58	0.58
Eastern Europe	7	102	6.96	0.45
Middle East	6	91	7.47	0.62
North America	2	75	6.06	0.46
North Pacific	3	108	7.50	0.52
Oceania	6	57	6.79	0.61
South America	12	224	8.44	0.62
South Pacific	4	120	8.42	0.64
Southeast Asia	6	114	8.86	0.66
Southern Asia	7	215	10.06	0.64
West Europe	10	101	6.00	0.39
Total	112	1794		
Mean			7.80	0.57

Table 1. Allelic analysis of 1,794 accessions in the USDA rice core collection genotyped with 72 DNA markers among 14 geographic regions.

AMOVA showed that the majority (89%) of total genetic variance attributed to differences within regions and the rest (11%) was due to variance among regions (Table 2). Likewise, when countries were taken into account, 82 % of the total variation was due to the differences within countries, and the remaining portion of the variance was equally shared by both among regions and among countries. Genetic variations were significantly differentiated among regions ( $\Phi_{st} = 0.10$ ,  $P < 0.001$ ) and among countries ( $\Phi_{st} = 0.12$ ,  $P < 0.001$ ), and very highly and significantly differentiated within countries ( $\Phi_{st} = 0.85$ ,  $P < 0.001$ ).



Source	df	Sum of squares	Mean squares	$\Phi_{st}$	<i>P-value</i>	Estimated variance	Percentage of total variance
Among regions	13	18956.7	1458.2	0.11	<0.001	10.8	11%
Within regions	1780	164044.8	92.2	0.89	<0.001	92.2	89%
Total	1793	183001.5				103.0	100%
Among region	13	19674.3	1513.4	0.10	<0.001	8.9	8.6%
Among countries	65	17106.2	263.2	0.12	<0.001	9.2	8.9%
Within countries	1672	142286.8	84.8	0.85	<0.001	84.8	82.5%
Total	1750	179067.2				102.9	

Table 2. Analysis of molecular variance (AMOVA) in 14 regions for 1,794 accessions in the USDA rice core collection genotyped with 72 DNA markers.

### 3.3 Genetic diversity and genetic relationships among geographic regions

Rice accessions collected from Southern Asia had the most number of alleles per locus, followed by Africa, Southeast Asia, China, South America, South Pacific and Central America, while those in Western and Eastern Europe, North America and Central Asia had the least (Table 1). As demonstrated by the PIC value, the accessions derived from Southeast Asia had the greatest diversity, followed by Southern Asia, South Pacific, Africa, Middle East, South America and Oceania, while those in Western and Eastern Europe and North America had the lowest diversity. Visualized by Nei Genetic Diversity index on the world map using the Kriging method, germplasm accessions collected from Southern Asia, Southeast Asia, Central America and Africa were mostly diversified, while those from North Pacific, Oceania, Western and Eastern Europe and North America had the lowest diversity (Fig. 2).

Germplasm accessions that were introduced from Southern Asia had the most private alleles per locus, followed by Africa, Central America, Southeast Asia, South Pacific, China, Oceania and Middle East, while those in Eastern Europe, Central Asia, North and South America and Western Europe had the least private alleles per locus (Fig. 3).

Three main clusters were resulted from the UPGMA analysis based on Nei (Nei, 1973) genetic similarity (Fig. 4). In cluster 1, germplasm accessions from South America were mostly related to Central America, and then to Africa, Oceania and North America. Two sub-groups of the originating region among rice accessions obviously existed in cluster 2, while Eastern Europe and Western Europe were in sub-group 1 and Central Asia, Middle East and North Pacific in sub-group 2. In cluster 3, germplasm accessions originating in Southeast Asia were closest to those in the South Pacific, and then to China and the Southern Asia. Cluster 1 was closer to cluster 2 than to cluster 3.

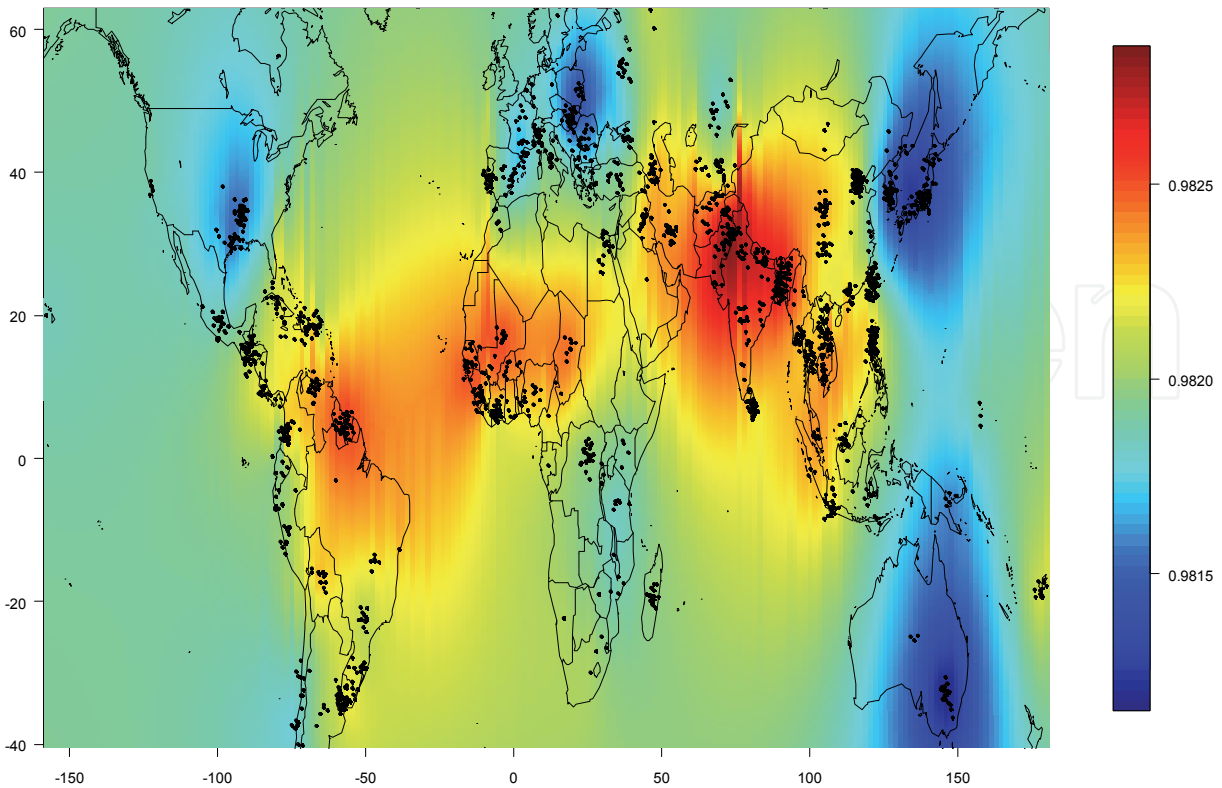


Fig. 2. Geographic diversity of rice germplasm demonstrated by Nei Genetic Diversity Index in the USDA rice core collection genotyped with 72 DNA markers. The deeper the red color is, the greater the genetic diversity is for the area. The deeper the blue color is, the smaller the genetic diversity is for the area. Each dot represents an accession placed on the world map according to its latitude and longitude.

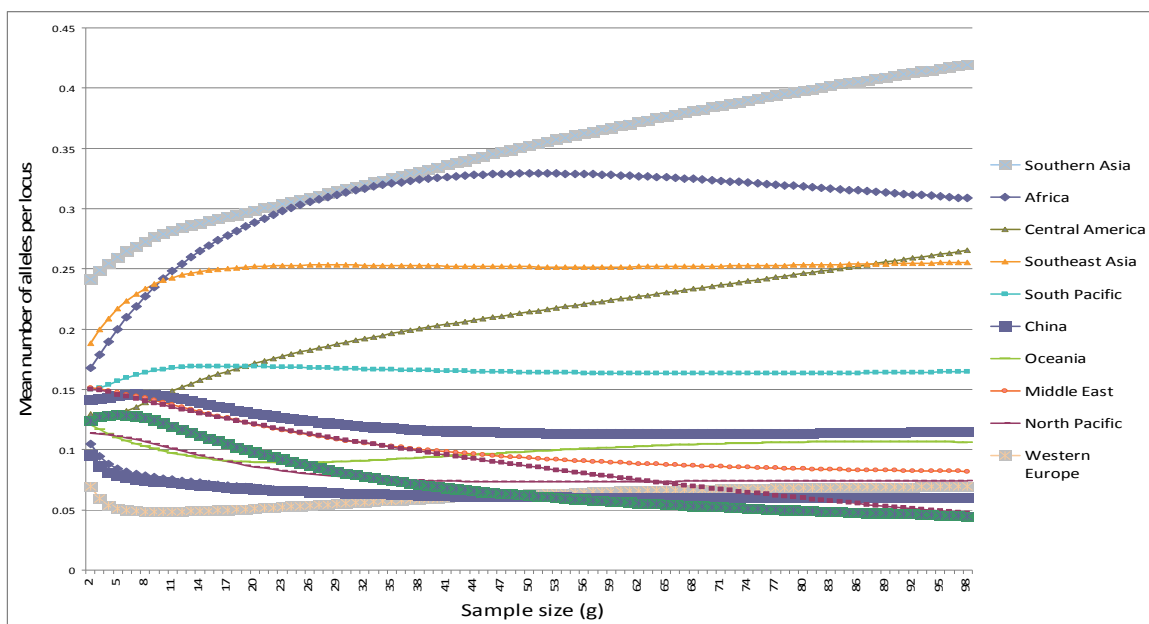


Fig. 3. The mean number of private alleles per locus as a function of standardized sample size (g) for 14 geographic regions arranged from high on the top to low on the bottom for 1,794 accessions in the USDA rice core collection.

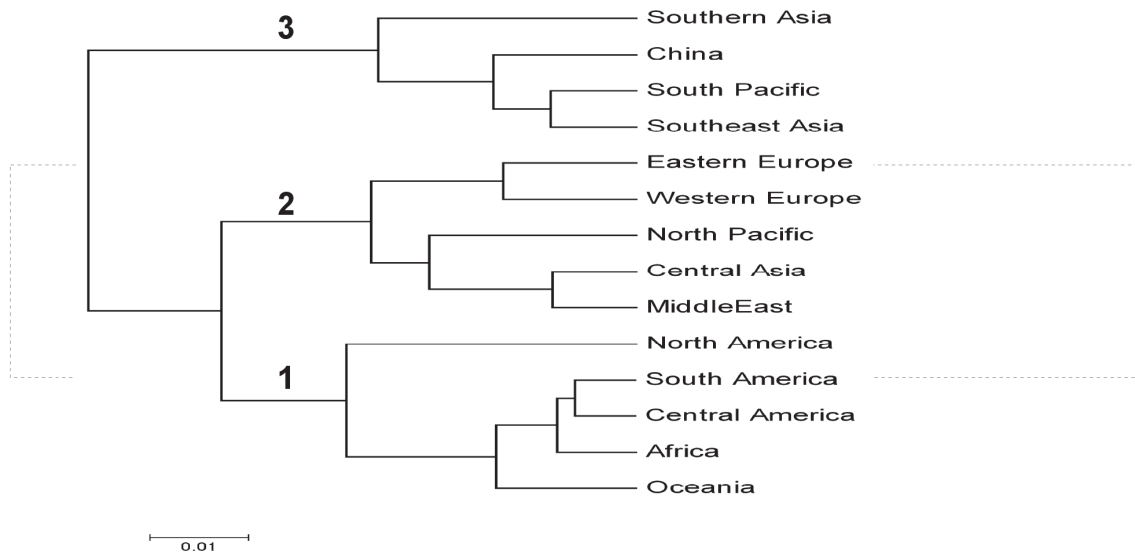


Fig. 4. Cluster analysis of geographic regions using Nei genetic similarity (Nei, 1973) for 1,794 accessions in the USDA rice core collection genotyped with 72 DNA markers

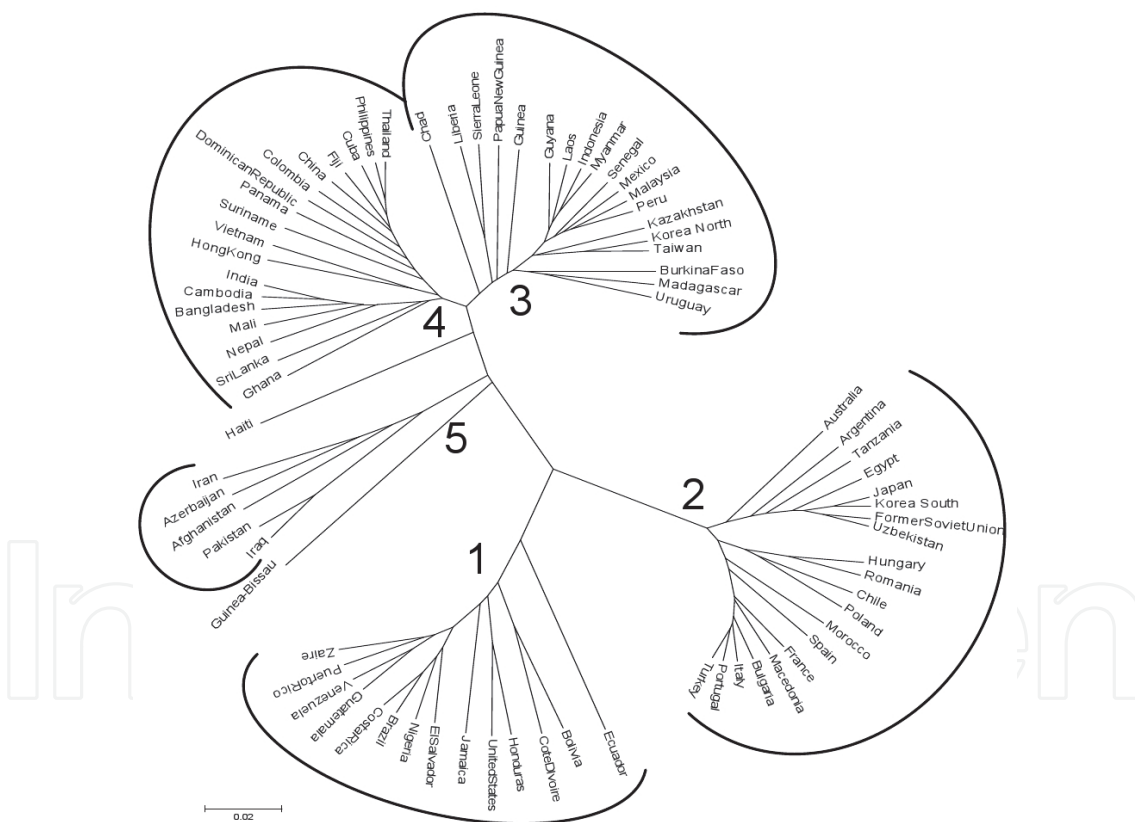


Fig. 5. Cluster analysis of countries having five or more accessions in the USDA rice core collection genotyped with 72 DNA markers

**3.4 Genetic diversity and genetic relationships among countries**

Among the 78 countries from which 5 or more accessions were introduced, Myanmar had the most diversification indicated by the highest PIC (0.65). The PICs measuring genetic

diversities ranged 0.60-0.63 in 13 countries: four in Africa, three in Southeast Asia and two each in South America, South Pacific, and Southern Asia; and 0.50-0.60 in 27 countries: four each in Africa and Central America, three each in South America, and Southern Asia, two each in Central Asia, China, Middle East, and North Pacific, and one each in Eastern Europe, North America, Oceania, Southeast Asia, and South Pacific. There were 22 countries with the PICs ranging 0.40-0.50: five in South America, four each in Africa and Central America, two each in Middle East and Oceania, and one each in Central Asia, China, North Pacific, Southern Asia, and Western Europe. France and Spain in Western Europe and Romania in Eastern Europe had the lowest PIC value.

Cluster analysis of 78 countries from which 5 or more accessions were present in the core collection formed five distinctive groups (Fig. 5). Fourteen countries were placed in Cluster 1, six in Central America, four in South America, three in Africa, and one in North America which is the United States. Cluster 2 contained 20 countries, six in Eastern Europe, four in Western Europe, three in Middle East, two each in North Pacific and South America and one each in Africa, Central Asia and Oceania. Cluster 3 included 19 countries, seven in Africa, three in South America, two each in South Pacific and Southeast Asia, and one each in Central Asia, China, North America, North Pacific and Oceania. Cluster 4 had 18 countries, four in Southern Asia, three each in Central America and Southeast Asia, two each in Africa, China and South America, and one each in Oceania and South Pacific. Cluster 5 was the smallest, including five countries, two each in Middle East and Southern Asia, and one in Central Asia. Two countries each with five accessions were independent of these clusters. Haiti in Central America was between Cluster 4 and 5, while Guinea-Bissau in Africa was between Cluster 1 and 5. The vast diversity found in the USDA global rice collection is an important genetic resource that can effectively support breeding programs in the U.S. and worldwide.

#### 4. Genetic differentiation of global rice germplasm

Cultivated rice (*Oryza sativa* L.) is structured into five genetic groups, *indica* (IND), *aus* (AUS), *tropical japonica* (TRJ), *temperate japonica* (TEJ) and *aromatic* (ARO) (Izawa, 2008; Caicedo et al., 2007; Garriss et al., 2005). Genetic characterization of rice germplasm collections will enhance their utilization by the global research community for improvement of rice.

##### 4.1 Statistical analysis

Genotypic data of 71 SSR plus an indel markers for the core collection plus 23 reference cultivars were used to decide putative number of structures at first. Genetic structure was inferred using the admixture analysis model-based clustering algorithms implemented in TESS v. 2.1 (Chen et al., 2007). TESS implements a Bayesian clustering algorithm for spatial population genetics. Multi-locus genotypes were analyzed with TESS using the Markov Chain Monte Carlo (MCMC) method, with the F-model and a  $\psi$  value of 0.6 which assumes 0.0 as non-informative spatial prior. To estimate the K number of ancestral-genetic populations and the ancestry membership proportions of each individual in the cluster analysis, the algorithm was run 100 times, each run with a total of 70,000 sweeps and 50,000 burn-in sweeps for each K value from 2 to 15. For each run we computed the Deviance Information Criterion (DIC) (Spiegelhalter et al., 2002), a model-complexity penalized measure

to show how well the model fits the data. The putative number of clusters was obtained when the DIC values were the smallest and estimates of data likelihood were the highest in 10% of the runs. Similarity coefficients between runs and the average matrix of ancestry membership were calculated using CLUMPP v. 1.1 (Jakobsson and Rosenberg, 2007).

Each accession in the core collection was grouped to a specific cluster or population by its K value resulted from cluster analysis using TESS. The sub-species ancestry of each K was inferred by the reference cultivars for *indica*, *AUS*, *aromatic*, *temperate japonica*, and *tropical japonica* rices. Analysis of molecular variance (AMOVA; Excoffier et al., 1992) was used to calculate variance components within and among the populations obtained from TESS in the collection. Estimation of variance components was performed using the software ARLEQUIN 3.0 (Schneider et al., 2000). The AMOVA-derived  $\Phi_{ST}$  (Weir and Cockerham, 1984) is analogous to Wright's F statistics differing only in their assumption of heterozygosity (Paun et al., 2006).  $\Phi_{ST}$  provides an effective estimate of the amount of genetic divergence or structuring among populations (Excoffier et al., 1992). Significance of variance components was tested using a non-parametric procedure based on 1,000 random permutations of individuals. The computer package ARLEQUIN was used to estimate pairwise  $F_{ST}$  (Goudet, 1995) for the populations obtained from TESS.

Multivariate analysis such as principle component analysis (PCA) provides techniques for classifying the inter-relationship of measured variables. Multivariate geo-statistical methods combine the advantages of geo-statistical techniques and multivariate analysis while incorporating spatial or temporal correlations and multivariate relationships to detect and map different sources of spatial variation on different scales (Goovaerts, 1992; Wackernagel, 1994). Geographical spatial interpolation of principal coordinates of latitude and longitude and admixture ancestry matrix coefficients (Ks) calculated in TESS for each accession were represented by kriging method (François et al., 2008) as implemented in the R statistical packages 'spatial', 'maps' and 'fields' (Venables and Ripley, 1998; Venables et al., 2008) for visualizing distribution in the world map.

Principal components analysis (PCA) was conducted using GenAlex 6.1 (Peakall and Smouse, 2006) software to structure the core collection genotyped by 72 molecular markers, and generate a PC-matrix. Geo-statistical and geographic analysis was based on CNT coordinates of latitude and longitude where a core accession originated using the R statistical packages. Polymorphism information content (PIC) and number of alleles per locus in each sub-species population were estimated using PowerMarker software (Liu and Muse, 2005). Number of distinct alleles in each population and number of alleles private to each population, that is not found in other populations, were calculated using ADZE program (Allelic Diversity AnalyZER, Szpiech et al., 2008). ADZE uses the rarefaction method to trim unequal accessions to the same standardized sample size, a number equal to the smallest accessions across the populations.

#### 4.2 Number of populations and ancestry determination

Structural analysis resulted in the lowest Deviance Information Criterion (DIC) or highest log likelihood scores when the putative number (K) of populations was set at five, and the ancestry coefficient of each accession in each K was estimated accordingly (Fig. 6) (Agrama et al., 2010). Similarly, principle coordinate (PC) analysis of Nei's genetic distance (Nei, 1973; 1978) classified the core accessions into five clusters by PC1 and PC2 including 71% of total variances (Fig. 7). Both structure and PC analyses indicated that five populations sufficiently

explained the genetic diversity in the core collection. Analysis of molecular variance (AMOVA) showed that 38% of the variance was due to genetic differentiation among the populations (Table 3). The remaining 62% of the variance was due to the differences within the populations. The variances among and within the populations were highly significant ( $P < 0.001$ ).

Source	df	SS	MS	Est. Var.	%	$\Phi_{ST}$	P-value <sup>a</sup>
<b>Among Pops</b>	4	57383	14346	43	38	0.38	<0.001
<b>Within Pops</b>	1781	124086	70	70	62	0.62	<0.001
<b>Total</b>	1785	181470		112	100		

<sup>a</sup> Probability of obtaining a more extreme random value computed from non-parametric procedures (1,000 permutations).

Table 3. Analysis of molecular variance (AMOVA) for the 1,763 core accessions and 23 reference cultivars for five populations (Pops) of ARO, AUS, IND, TEJ and TRJ based on 72 DNA markers.

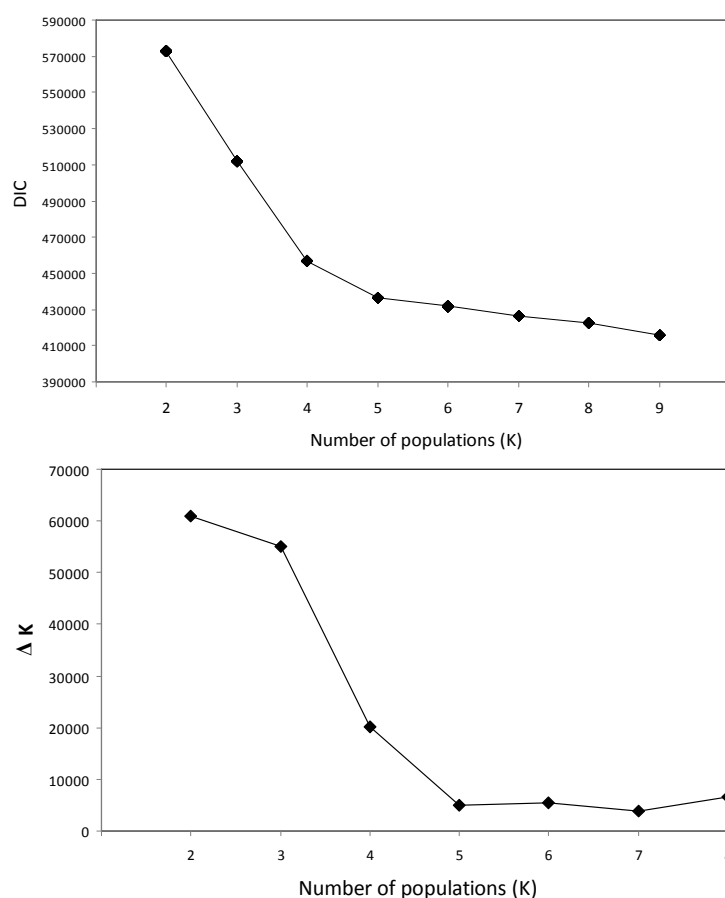


Fig. 6. Five populations should be structured based on both the log-likelihood values (Deviance Information Criterion, DIC) and the change rate of log-likelihood values ( $\Delta K$ ) for estimated number of populations over 50 structure replicated runs using TESS program. Where relatively flat change of both DIC and  $\Delta K$  occurs indicates the most likely number of populations.

Among 40 reference cultivars, 20 that are known *tropical japonica* (TRJ) were classified in K1, four known *temperate japonica* (TEJ) in K2, eight known *indica* (IND) in K3, three known *AUS* (AUS) in K4 and five known *aromatic* (ARO) in K5, indicating the correspondent ancestry of each population. Based on the references, each accession was clearly assigned to a single population when its inferred ancestry estimate was 0.6 or larger and admixture between populations when its estimate was less than 0.6. Admixture was based on proportion of the estimate, i.e. GSOR 310002 was assigned TEJ-TRJ because of its estimate 0.5227 in K2 and 0.4770 in K1.

K1 or TRJ population included 353 (19.8%) absolute accessions, 41 (2.3%) admixtures with K2 or TEJ population, 26 (1.5%) admixtures with K3 or IND and one admixture with K4 or AUS. In K2, 420 (23.5%) accessions had absolute ancestry, 52 (2.9%) admixed with K1 and seven admixed with other populations. K3 or IND population had 625 (35.0%) accessions among which 595 were clearly assigned, twelve admixed with K4 or AUS, and 18 admixed with other populations. One hundred sixty-five (9.8%) accessions were clearly grouped in K4, 13 were admixed with K3 and two admixed with K5 or ARO population. Seventy-two (4.0%) accessions were clearly structured in K5, five were admixed with K2 and three admixed with other population.

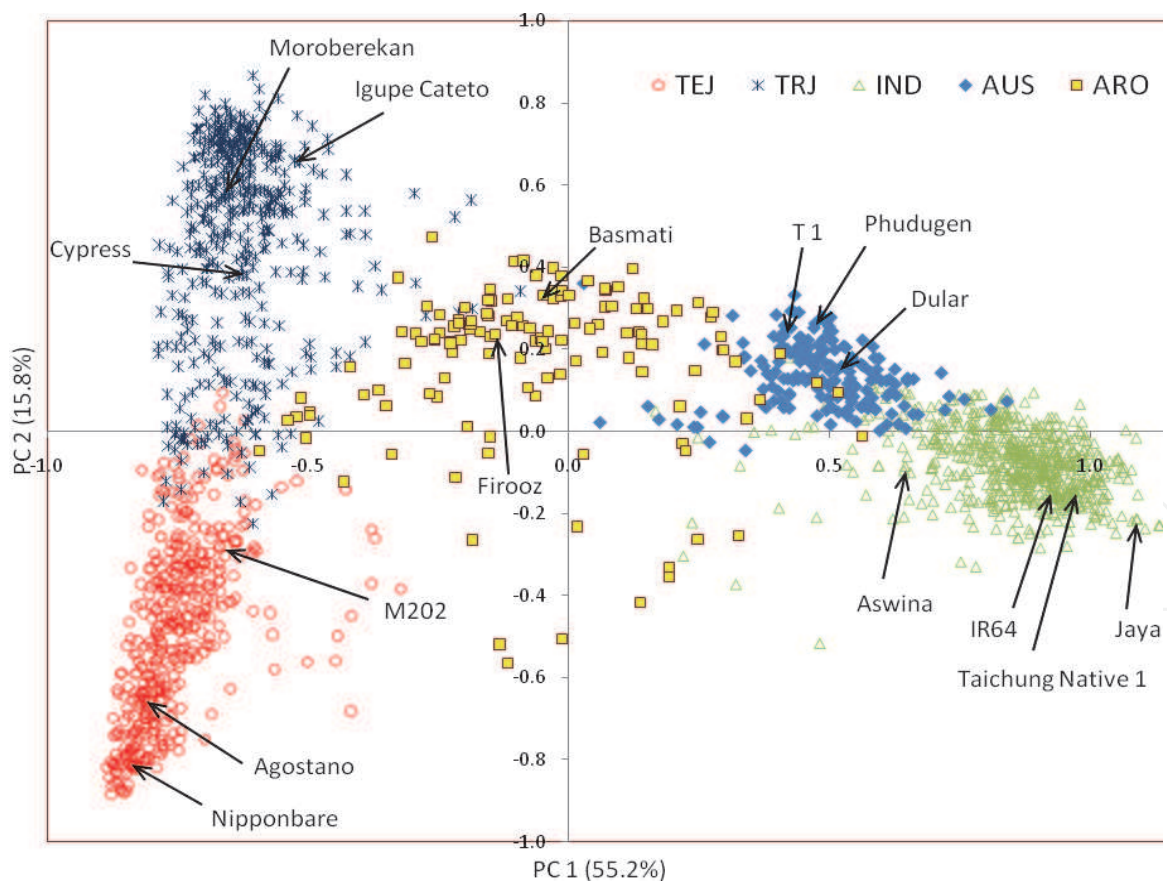


Fig. 7. Principle coordinates analysis of five populations inferred by highlighted reference cultivars (*temperate japonica* - TEJ, *tropical japonica* - TRJ, *indica* - IND, *aus* - AUS and *aromatic* - ARO) for the core accessions genotyped with 72 DNA markers.

### 4.3 Genetic relationship and global distribution of ancestry populations

All pair-wise estimates of  $F_{ST}$  using AMOVA for the populations were highly significant ranging from 0.240 to 0.517 (Table 4). IND was equally distant from ARO and AUS, but more distant from TEJ and TRJ. AUS and IND were mostly differentiated from TEJ. However, TEJ, TRJ and ARO were close to each other in comparison with others. These relationships were consistent with structure analysis revealed by the PCA (Fig. 7).

	ARO	AUS	IND	TEJ	TRJ
ARO		0.001	0.001	0.001	0.001
AUS	0.253		0.001	0.001	0.001
IND	0.284	0.308		0.001	0.001
TEJ	0.317	0.517	0.500		0.001
TRJ	0.240	0.475	0.462	0.273	

Table 4. Pairwise estimates of  $F_{ST}$  (lower diagonal) and their corresponding probability values (upper diagonal) for five rice populations, K5 - *aromatic* (ARO), K4 - *aus* (AUS), K3 - *indica* (IND), K2 - *temperate japonica* (TEJ) and K1 - *tropical japonica* (TRJ) for 1,763 core accessions genotyped with 72 DNA markers based on 999 permutations.

Among 421 accessions of TRJ rice in the core collection, the majority is collected from Africa (23%) and South America (21%), followed by Central America (15%), North America (13%), South Pacific (6%), Southeast Asia and Oceania (5% each) (Fig. 8A). North America had 75 accessions in total and 55 were grouped in TRJ, which was the highest percentage (73%) among 14 regions, followed by Central America (56%), Africa (49%) and South America (41%). Among 112 countries, the U.S. in North America had the highest percentage (92%) of accessions, followed by Cote d'Ivoire and Zaire (91%) in Africa and Puerto Rico (72%) in Central America.

Most TEJ rice is collected from Western and Eastern Europe (20% each), followed by North Pacific (14%), South America (10%), Central Asia (7%) and North China (7%) (Fig. 8B). Similarly, Western and Eastern Europe had the highest percentage (85% each) of TEJ, followed by North Pacific (55%) and South America (20%). Hungary accessions had the highest percentage (97%), followed by Italy (89%), Russian Federation and Portugal (83% each).

Based on United Nations' classification, region China includes Mongolia, Hong Kong, Taiwan and China itself. Most IND rice (25%) is collected from region China, followed by the South Asia (14%), South America (13%), Southeast Asia and Africa (10% each) (Fig. 8C). Region China had the highest percentage (72%) of IND, followed by South Pacific (57%), Southeast Asia (53%), Southern Asia (38%) and Africa (29%). Also, country China had the highest percentage (84%) of IND, followed by Columbia (81%), Sri Lanka (80%) and Philippines (68%).

About half of the AUS rice in the collection was sampled from the South Asia (48%), followed by Africa (16%), Middle East (11%), South America and Southeast Asia (7% each) (Fig. 8D). South Asia had the highest percentage (40%) of AUS, followed by

Middle East (21%), Africa (14%) and Southeast Asia (10%). Bangladesh had the highest percentage (63%) of AUS, followed by Iraq (64%), Pakistan (49%) and India (40%).



Aromatic rice in the collection originated mainly from Pakistan (20%) and Afghanistan (13%) in the South Asia and Azerbaijan (15%) in Central Asia, representing 37%, 44% and 57% of total core accessions from these countries, respectively (Fig. 8E).

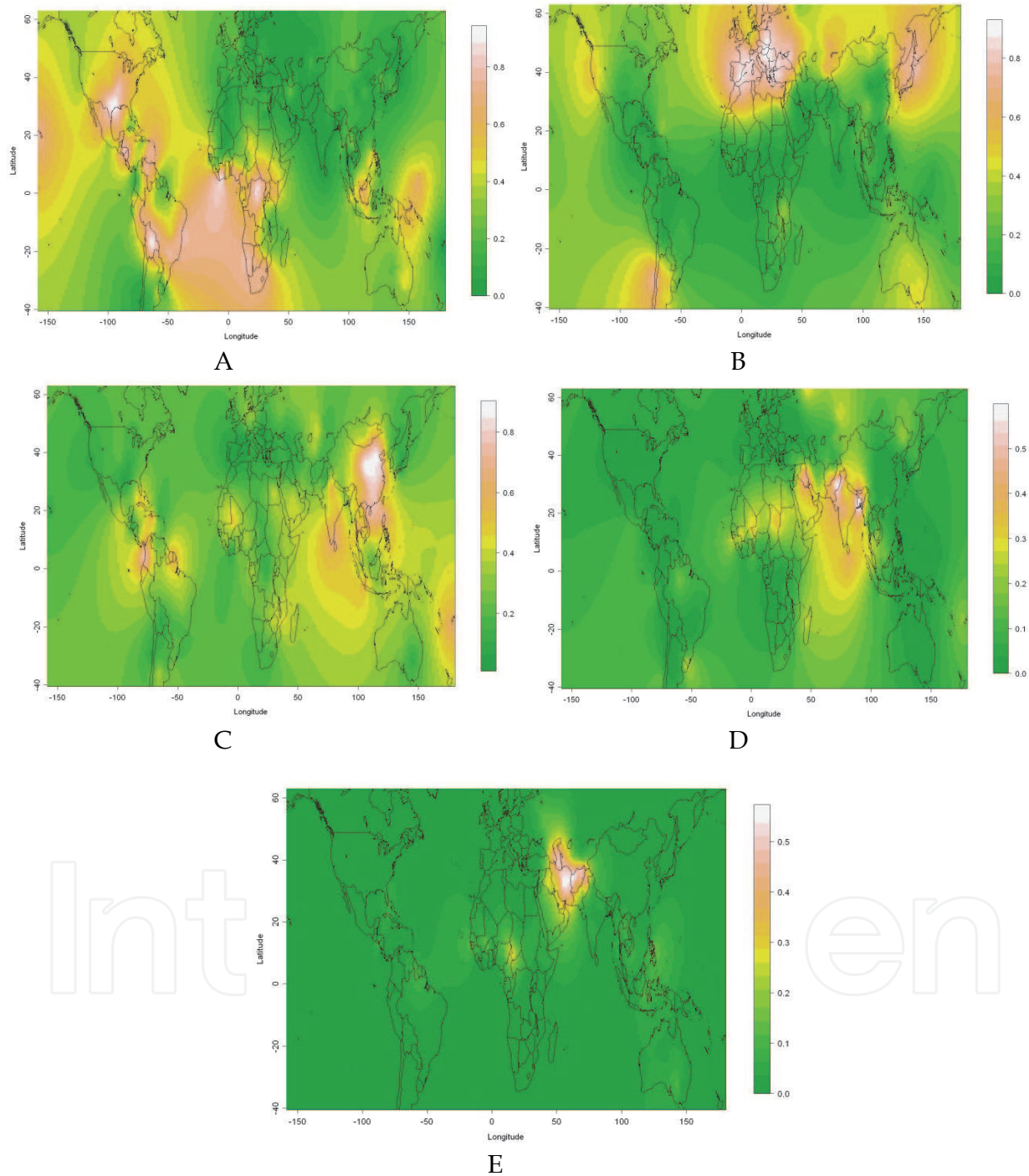


Fig. 8. Global distribution of core accessions in each population resulted from cluster analysis and inferred by reference cultivars based on geographical coordinates of latitude and longitude in K1 (*tropical japonica* – TRJ), A; K2 (*temperate japonica* – TEJ), B; K3 (*indica* – IND), C; K4 (*aus* – AUS), D and K5 (*aromatic* – ARO), E.

#### 4.4 Genetic diversity of the populations

Average alleles per locus were the highest in IND, followed by AUS, ARO, TRJ and TEJ (Fig. 9). IND had 45% more alleles per locus than TEJ. ARO had the highest polymorphic information content (PIC), followed by AUS, IND, TRJ and TEJ. The PIC value of TEJ was 72% less than that of ARO. AUS had the most alleles per locus corrected for difference in sample size distinctly (Fig. 10A) and privately (Fig. 10B) from others. Although IND and ARO had same distinct alleles per locus, which was next to AUS, there were much more private alleles per locus in IND than in ARO. TEJ had either the lowest distinct alleles or private alleles per locus among the populations.

Genetic characterization of the USDA rice world collection for genetic structure, diversity, and differentiation will help design cross strategy to avoid sterility for gene transfer and exchange in breeding program and genetic studies, thus better serve the global rice community for improvement of cultivars and hybrids because this collection is internationally available, free of charge and without restrictions for research purposes. Seed may be requested from GRIN (GRIN, 2011) for the whole collection, and from GSOR (GSOR, 2011) for the core collection.

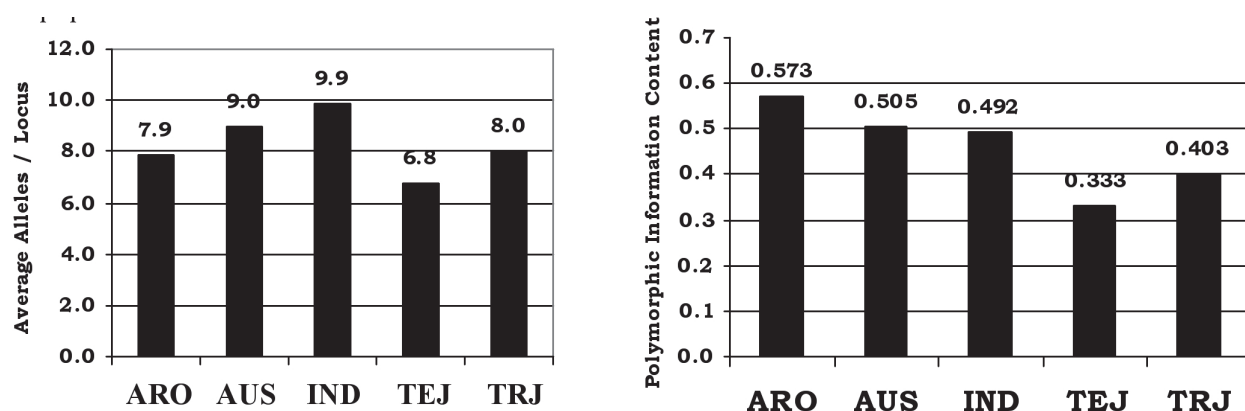


Fig. 9. Average alleles per locus and polymorphic information content for five populations resulted from cluster analysis and inferred by reference cultivars K1 (*tropical japonica* - TRJ), K2 (*temperate japonica* - TEJ), K3 (*indica* - IND), K4 (*aus* - AUS) and K5 (*aromatic* - ARO).

#### 5. USDA rice mini-core collection

Development of core collections is an effective tool to extensively characterize large germplasm collections, and the utilization of a mini-core sub-sampling strategy further increases the effectiveness of genetic diversity analysis at detailed phenotype and molecular levels (Agrama et al., 2009). Using the advanced M strategy, Kim et al. (2007) presented PowerCore software that possesses the power to represent all the alleles identified by molecular markers and classes of the phenotypic observations in the development of core collections.

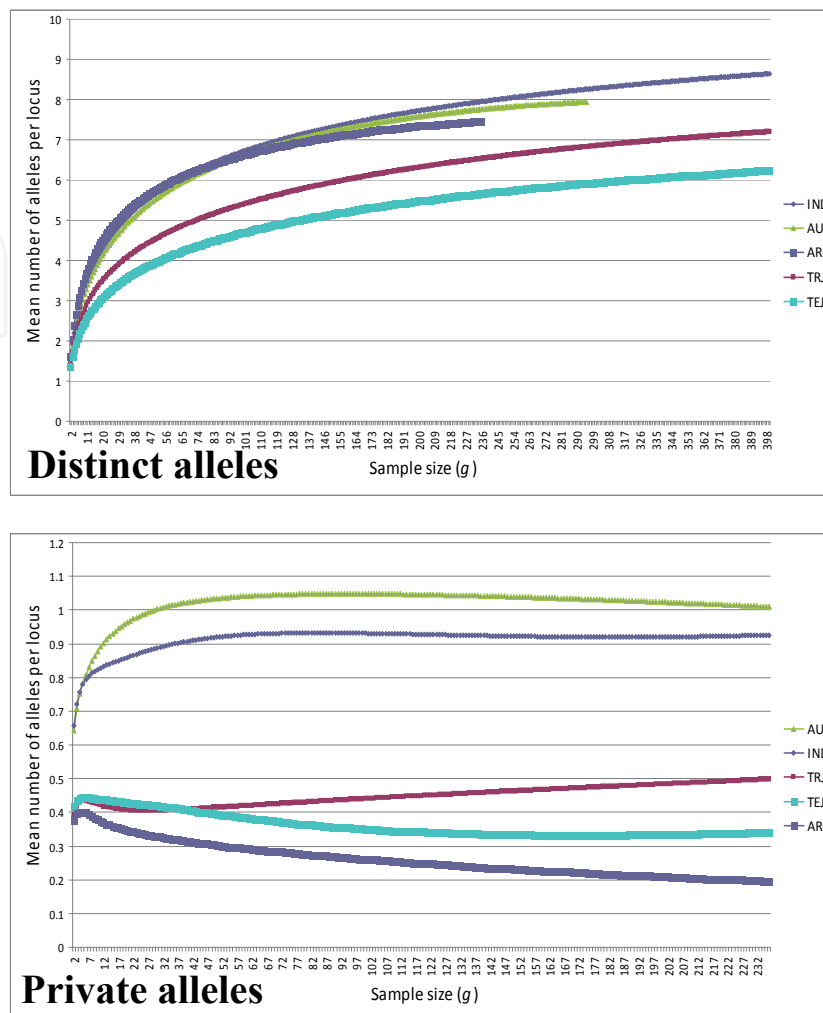


Fig. 10. The mean number of (A) distinct alleles per locus and (B) private alleles per locus to each of five populations, K1 (*tropical japonica* – TRJ), K2 (*temperate japonica* – TEJ), K3 (*indica* – IND), K4 (*aus* – AUS) and K5 (*aromatic* – ARO), as functions of standardized sample size  $g$ .

### 5.1 Phenotypic and genotypic data used to develop the USDA rice mini-core collection

Data of 26 phenotypic traits, 69 SSRs and one indel marker generated from 1,794 accessions in the USDA rice core collection at Stuttgart, Arkansas, USA were used to develop the mini-core. The phenotypic traits included 13 for morphology, two for cooking quality, 10 for rice blast disease resistance ratings from individual races of *Magnaporthe oryzae* Cav., and one for physiological disease, straighthead. Field evaluations of blast were conducted at the University of Arkansas Experiment Station, Pine Tree, AR following inoculation using a mixture of the most prevalent races (IB-1, IB-49, IC-17, IE-1, IE-1K, IG-1 and IH-1) found in the southern US rice production region using the method described by Lee et al. (2003). In greenhouse, seven blast races, IB-1, IB-33, IB-49, IC-17, IE-1K, IG-1, and IH-1 were individually inoculated and rated in a scale from 0 (no lesions) to 9 (dead).

## 5.2 Sampling strategy and representation analysis

Sampling the core collection was performed by the PowerCore software with an effort to maximize both the number of observed alleles at SSR loci and the number of phenotypic trait classes using the advanced M (maximization) strategy implemented through a modified heuristic algorithm (Agrama et al., 2009). The phenotypic traits were automatically classified into different categories or classes by the PowerCore program based on Sturges' rule =  $1 + \text{Log}_2(n)$ , where  $n$  is the number of observed accessions (Kim et al., 2007).

The resulting mini-core was compared with the original core collection to assess its homogeneity. Nei genetic diversity index (Nei, 1973) was estimated for each molecular marker in both the core and mini-core collections. Chi-squared ( $\chi^2$ ) tests were used to test the similarity for number of marker alleles and frequency distribution of accessions. Homogeneity was further evaluated for the 26 phenotypic traits using the Newman-Keuls test for means, the Levene test (Levene, 1960) for variances, and the mean difference (MD%), variance difference (VD%), coincidence rate of range (CR%) and variable rate of coefficient of variance (VR%) according to Hu et al. (2000). Coverage of all the phenotypic traits in the original core collection was estimated in the mini-core as proposed by Kim et al. (2007):

$$\text{Coverage (\%)} = \frac{1}{m} \sum_{j=1}^m \frac{D_c}{D_e} \times 100$$

Where  $D_c$  is the number of classes occupied in the mini-core and  $D_e$  is the number of classes occupied in the original core collection for each trait and  $m$  is the number of traits which is 26 in this case.

## 5.3 Distribution frequency of accessions in the core and mini-core collections

The heuristic search based on the 26 phenotypic traits and the 70 markers sampled 217 accessions (12.1%) out of 1,794 accessions in the core collection. The 217 mini-core entries originated from 76 countries covering all the 15 geographic regions (Table 5). Five regions, Subcontinent, South Pacific, Southeast Asia, Africa and China accounted for the majority, 63.6% of the mini-core entries, while the fewest entries came from three regions, Australia, Mideast and North America, accounting for 5.5%. Two accessions in the mini-core are of unknown origin.

The similarity of distribution frequencies between the core and mini-core collections for each of the 15 regions was tested using  $\chi^2$  with one degree of freedom (Table 5). All 15 regions had non-significant  $\chi^2$  values ranging from 0.095 to 0.996 with probability ( $P$ ) from 0.303 to 0.758, which proved a homogeneous distribution between the two collections.

Among the 217 mini-core *Oryza* entries, eight belong to *O. glaberrima*; two each of *O. nivara* and *rufipogon*; one each of *O. glumaepatula*, *latifolia*, and the remaining 203 entries belong to *O. sativa*.

Region	USDA Rice Core collection		Mini-core		$\chi^2$	<i>P</i>
	Number	%	Number	%		
Africa	198	11.0	24	11.1	0.996	0.318
Australia	24	1.3	1	0.5	0.513	0.474
Balkans	61	3.4	9	4.2	0.786	0.375
Central America	116	6.5	12	5.5	0.787	0.375
China	208	11.6	20	9.2	0.602	0.438
Eastern Europe	102	5.7	9	4.2	0.624	0.430
Mideast	91	5.1	5	2.3	0.308	0.579
North America	71	4.0	6	2.8	0.646	0.422
North Pacific	108	6.0	11	5.1	0.775	0.379
South America	224	12.5	15	6.9	0.206	0.650
South Pacific	152	8.5	24	11.1	0.558	0.455
Southeast Asia	114	6.4	23	10.6	0.303	0.303
Subcontinent	215	12.0	47	21.7	0.095	0.758
Western Europe	101	5.6	9	4.2	0.635	0.425
Unknown	9	0.5	2	0.9	0.725	0.725
Total	1794	100	217	100		

\* $\chi^2$  values with one degree of freedom and the corresponding probability (*P*).

Table 5. Distribution frequency comparison of origin of accessions between the USDA rice core and mini-core collections among 15 geographical regions.

#### 5.4 Phenotypic diversity in the core and mini-core collections

Comparative analysis of the ranges, means and variances for 26 phenotypic traits demonstrated that the mini-core covered full range of variation for each trait. The Newman-Keuls test results indicate the presence of homogeneity of means between the core collection and mini-core for 22 traits (85%). Sixteen (62%) of the traits had homogeneous variances revealed by the Levene's test. Among the 10 traits having heterogeneous variances, five morphological traits and amylose content had greater variances in the mini-core than in the core collection. However, hull cover and color, and two disease traits had smaller variances.

The mean difference percentage (MD%), the variance difference percentage (VD%), the coincidence rate (CR%) and the variable rate (VR%) are designed to comparably evaluate

the property of core collection with its initial collection. Over the entire 26 phenotypic traits, the MD% was 6.3%, far less than the significance level of 20%. The VD% was 16.5%, less than the significance level of 20%, and six traits had much greater variances in the mini-core than in the core collection (Table 6). The VR% compares the coefficient of variation values and determines how well the variance is being represented in the mini-core. More than 100% of VR is required for a core collection to be representative of its original collection (Hu et al., 2000). The mini-core had 102.7% VR over its originating core, indicating good representation.

	USDA Rice Core Collection			Mini-core			Test <sup>1</sup>	
	Range	Mean	Variance	Range	Mean	Variance	N-K	Lev
<b>Morphology</b>								
Days to flower	42 - 174	95.8	355.5	46 - 166	96.2	469.6	n.s.	*
Plant height cm	60 - 212	125.8	627.3	70 - 202	135.7	646.6	**	n.s.
Plant type <sup>2</sup>	1 - 9	2.7	2.82	1 - 9	2.7	3.01	n.s.	n.s.
Lodging <sup>2</sup>	0 - 9	2.3	4.98	0 - 9	3.1	7.71	**	**
Panicle type <sup>2</sup>	1 - 9	4.9	1.20	1 - 9	4.8	2.31	n.s.	*
Awn type <sup>2</sup>	0 - 9	1.2	8.27	0 - 9	2.0	12.56	**	**
Hull cover <sup>2</sup>	1 - 6	3.6	1.20	1 - 6	3.7	0.78	n.s.	*
Hull color <sup>2</sup>	1 - 8	3.5	3.55	1 - 8	3.7	1.94	n.s.	*
Bran color <sup>2</sup>	1 - 7	2.3	1.09	1 - 7	2.5	1.75	n.s.	*
Kernel length mm	4.2 - 10.0	6.5	0.63	4.2 - 10.1	6.5	0.95	n.s.	n.s.
Kernel width mm	1.5 - 3.5	2.6	0.11	1.5 - 3.5	2.6	0.10	n.s.	n.s.
Kernel Length/Width	2.0 - 5.0	2.6	0.35	2.0 - 5.0	2.6	0.45	n.s.	n.s.
1000 kernel weight g	6.72 - 37.4	21.2	14.76	10.0 - 37.4	21.0	18.45	n.s.	n.s.
<b>Quality</b>								
Amylose %	0 - 26.9	19.9	25.57	0.10 - 26.5	10.5	38.46	n.s.	*
ASV <sup>2</sup>	2.1 - 7	5.1	1.59	2.3 - 7.0	4.9	1.47	n.s.	n.s.
<b>Disease</b>								
Leaf blast	0 - 9	4.5	7.50	0.3 - 9	4.9	7.88	n.s.	n.s.
Early panicle blast	0 - 9	4.1	8.63	0 - 9	4.1	8.26	n.s.	n.s.
Final panicle	0 - 9	5.0	8.00	0 - 9	4.9	8.40	n.s.	n.s.

	USDA Rice Core Collection			Mini-core			Test <sup>1</sup>	
	Range	Mean	Variance	Range	Mean	Variance	Levene's	N-K
blast								
Blast IB-1	0 - 8	4.0	9.24	0 - 8	3.9	8.60	n.s.	n.s.
Blast IB-33	0 - 8	6.1	1.7	0 - 8	6.1	1.74	n.s.	n.s.
Blast IB-49	0 - 8	5.0	9.27	0 - 8	5.0	8.60	n.s.	n.s.
Blast IC-17	0 - 8	4.0	10.58	0 - 8	3.4	9.93	*	n.s.
Blast IG-1	0 - 8	4.0	10.68	0 - 8	4.0	9.73	n.s.	*
Blast IE-1K	0 - 8	4.3	8.74	0 - 8	4.6	7.75	n.s.	*
Blast IH-1	0 - 8	1.8	5.78	0 - 8	2.0	5.45	n.s.	n.s.
Straighthead <sup>2</sup>	1 - 9	7.3	1.90	1.3 - 9	7.5	1.83	n.s.	n.s.

<sup>1</sup> Means were tested using Newman-Keuls test (N-K) and variances were tested by Levene's test (Lev) for homogeneity between the USDA rice core collection and mini-core, \* and \*\* significant at 0.05 and 0.01 probability, respectively.

<sup>2</sup>Categorical data as described in the GRIN (GRIN, 2011).

Table 6. Comparison of range, mean and variance between the USDA rice core collection and the mini-core for 26 phenotypic traits.

The coincidence rate (CR%) indicates whether the distribution ranges of each trait in the mini-core are well represented when compared to the core collection. The resulting CR over the 26 traits was 97.5%, indicating homogeneous distribution ranges of the phenotypic traits because it was larger than the recommended 80% (Kim et al., 2007). The calculated Coverage value for the resulting mini-core was 100%, suggesting there is full coverage of all the diversity present in each class of phenotypic traits in the USDA rice core collection.

### 5.5 Molecular diversity in the core and mini-core collections

Both the USDA rice core collection and mini-core contained the same total number of polymorphic alleles (= 962 alleles) produced by the 70 markers, with an average of 14 alleles per locus, ranging from two for RM338 to 37 for RM11229 (Fig. 7A). Total alleles per locus ranged from 2 to 9 for 24 markers, from 10 to 19 for 32 markers and from 20 to 37 for 14 markers. The Nei genetic diversity index values reveal the allelic richness and evenness in the population. Distributions of the Nei indices among the 70 markers were very similar between the core and mini-core collections (Fig. 7B). The core collection had an average Nei diversity index of 0.72 with a minimum of 0.24 for AP5625-1 and maximum of 0.94 for RM11229 and RM302, while the average was 0.76 with a minimum of 0.37 for RM338 and AP5625-1 and maximum of 0.95 for RM11229 and RM302 in the mini-core. The minor difference of the molecular diversity was not statistically significant. Similarly, none of the 70 markers had significantly different Nei diversity index between the core and mini-core collections, indicated by the  $\chi^2$  test with values ranging from 0.000 to 0.022 and probabilities ranging from 0.882 to 0.999. More than 60% of the markers have a diversity index higher than 0.60 indicating high diversity across the markers (Fig. 7).

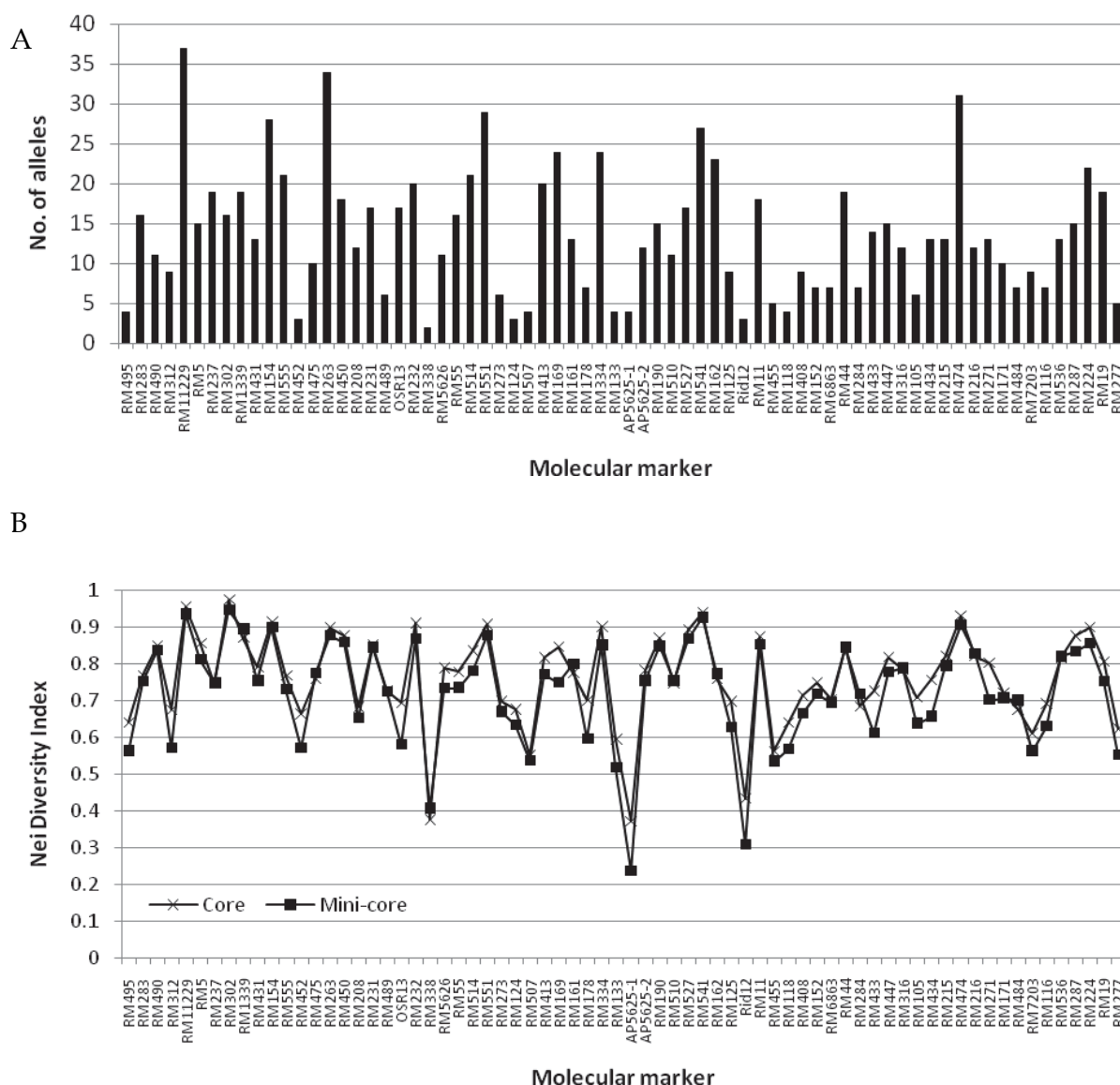


Fig. 7. Distribution of number of alleles per locus and Nei diversity index among the 70 DNA markers in the USDA rice core collection (Core) and mini-core (Mini-core). The markers were placed according to their position within the rice genome.

## 6. Use the USDA rice mini-core collection for mining valuable genes

Demonstrated both phenotypically and genotypically, the USDA rice mini-core collection of 217 entries is a good representative of the core of 1,794 entries as well as the entire rice global genebank of more than 18,000 accessions in the US (Yan et al., 2007; Agrama et al., 2009). The vast genetic diversity means the richness of valuable genes that could be extracted for cultivar improvement (Li et al., 2010). The reasonable number of entries in the mini-core allows extensively phenotyping and genotyping for mining valuable genes. The phenotyping could be performed in replicated tests and in multi-locations for the traits that are largely affected by environments such as yield (Li et al., 2011) and that require large amount of resources such as biotic and abiotic stresses. The genotyping could be done



genome-wide with high density of molecular markers such as simple sequence repeat (SSR) or single nucleotide polymorphism (SNP), or with sequencing the entire genome. The reliably phenotyping and densely genotyping genome-wide will improve the efficiency and accuracy of mining valuable genes for a globally sustainable agriculture. The core and mini-core collections are managed by the Genetic Stock *Oryza* Collection (GSOR, 2011) at the USDA-ARS Dale Bumpers National Rice Research Center and are available to the global research community.

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## 8. References

- Agrama H.A., W.G. Yan, F.N. Lee, R. Fjellstrom, M-H. Chen, M. Jia and A. McClung. 2009. Genetic assessment of a mini-core developed from the USDA rice genebank. *Crop Sci.* 49:1336-1346.
- Agrama H.A., W.G. Yan, M. Jia, R. Fjellstrom and A.M. McClung. 2010. Genetic structure associated with diversity and geographic distribution in the USDA rice world collection. *Natural Science* 2:247-291.
- Agrama, H.A. and W.G. Yan. 2010. Genetic diversity and relatedness of rice cultivars resistant to straighthead disorder. *Plant Breeding* 129:304-312.
- Bockelman, H.E., R.H. Dilday, W. Yan, and D.M. Wesenberg. 2002. Germplasm collection, preservation, and utilization. C.W. Smith and R. H. Dilday (eds.) *Rice: Origin, History, Technology, and Production. Crop Production*, pp. 597-625. Ser. #6149. John Wiley & Sons, Inc. New York, NY.
- Botstein, D., R.L. White, M. Skolnick, and M.R. Davis. 1980. Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *American J. of Human Genet.* 32:314-331.
- Bretting, P.K. 2007. The U.S. national plant germplasm system in an era of shifting international norms for germplasm exchange. *Proc. XXVII IHC-S1 Plant Gen. Reso. Acta Hort.* 760:55-60.
- Brown, A.H.D. 1989. Core collections: a practical approach to genetic resources management. *Genome*, 31: 818-824.
- Caicedo, A.L., S.H. Williamson, R.D. Hernandez, A. Boyko, et al. 2007. Genome-wide patterns of nucleotide polymorphism in domesticated rice. *PloS Genetics* 3:1745-1756.
- Chen, C., E. Durand, F. Forbes, and O. François. 2007. Bayesian clustering algorithms ascertaining spatial population structure: A new computer program and a comparison study. *Molecular Ecology Notes* 7:747-756.
- Excoffier, L., P.E. Smouse, and J.M. Quattro. 1992. Analysis of molecular variance inferred from metric distances among DNA haplotypes: Application to human mitochondrial DNA restriction sites. *Genetics* 131:479-491.

- FAO. 1996. Report on the state of the world's plant genetic resources for food and agriculture, prepared for the FAO International Technical Conference on Plant Genetic Resources. FAO Leipzig, Germany, pp 17–23.
- Fjellstrom, R.G., W. Yan, M.H. Chen, R.J. Bryant, H. Bockelman, and A.M. McClung. 2006. Genotypic and phenotypic assessment of the NSGC rice core collection for amylose content and alkali spreading value. Proc. 31st Rice Technical Working Group Conference. Feb. 26-Mar. 1, 2006, Houston, TX.
- François, O., M.G.B. Blum, M. Jakobsson, and N.A. Rosenberg. 2008. Demographic history of European populations of *Arabidopsis thaliana*. PLoS Genetics 4(5): e1000075.
- Garris, A.J., T.H. Tai, J. Coburn, S. Kresovich, and S. McCouch. 2005. Genetic structure and diversity in *Oryza sativa* L. Genetics 169:1631–1638
- Goovaerts, P. 1992. Factorial kriging analysis: a useful tool for exploring the structure of multivariate spatial soil information. J Soil Sci 43:597–619.
- Goudet, J. 1995. Fstat version 1.2: a computer program to calculate F statistics. J of Heredity 86: 485–486.
- GRIN, 2011. Germplasm Resources Information Network.  
<http://www.ars-grin.gov/> (Verified on September 12, 2011).
- GSOR. 2011. Genetic Stock Oryza Clection.  
<http://www.ars.usda.gov/Main/docs.htm?docid=8318> (Verified on September 12, 2011).
- Hamilton, R.S., and R. Raymond. 2005. Toward a global strategy for the conservation of rice genetic resources. In: Toriyama K, Heong KL, Hardy B (ed) Rice is life: scientific perspectives for the 21<sup>st</sup> century. Proceedings of the World Rice Research Conference held in Tsukuba, Japan, CD-ROM, pp 47–49.
- Hu, J., J. Zhu, and H.M. Xu. 2000. Methods of constructing core collections by stepwise clustering with three sampling strategies based on the genotypic values of crops. Theor. Appl. Genet. 101:264–268.
- Izawa, T. 2008. The process of rice domestication: a new model based on recent data. Rice 1:127–134.
- Jakobsson, M., and N. A. Rosenberg. 2007. CLUMPP: a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. Bioinformatics 23:1801–1806.
- Jia, L.M., W.G. Yan, H.A. Agrama, K. Yeater, X.B. Li, B.L. Hu, K. Moldenhauer, A. McClung and D.X. Wu. 2011. Searching for germplasm resistant to sheath blight from the USDA rice core collection. Crop Sci. 51:1507–1517.
- Kim, K.W., H.K. Chung, G.T. Cho, K.H. Ma, D. Chandrabalan, J.G. Gwag, T.S. Kim, E.G. Cho, and Y.J. Park. 2007. PowerCore: a program applying the advanced M strategy with a heuristic search for establishing core sets. Bioinformatics 23:2155–2162.
- Kumar, S., K. Tamura, and M. Nei. 2004. MEGA3: Integrated software for Molecular Evolutionary Genetics Analysis and sequence alignment. Brief Bioinform. 5: 150–163.
- Lee, F.N., W.G. Yan, J.W. Gibbons, M.J. Emerson, and S.D. Clark. 2003. Rice blast and sheath blight evaluation results for newly introduced rice germplasm. In R.J. Norman, J-F.

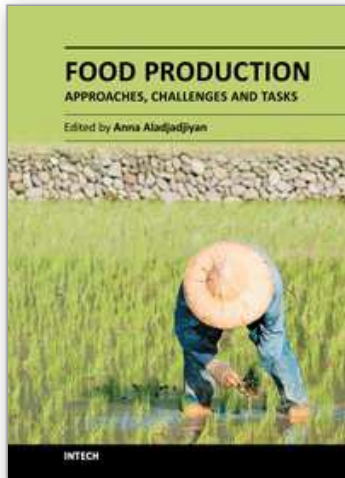
- Meullenet (eds.) BR Wells Rice Research Studies 2002. Univ. of Arkansas, Agric. Exp. Stn., Res. Ser. 504:85-92.
- Levene, H. 1960. Robust test for equality of variances. *In* I. Olkin (ed). Contributions to probability and statistics: essays in honor of Harold Hotelling. Stanford University Press, Stanford, pp 278-292.
- Li, X.B., W.G. Yan, H. Agrama, B.L. Hu, L.M. Jia, M. Jia, A. Jackson, K. Moldenhauer, A. McClung and D.X. Wu. 2010. Genotypic and phenotypic characterization of genetic differentiation and diversity in the USDA rice mini-core collection. *Genetica* 138:1221-1230.
- Li, X.B., W.G. Yan, L.M. Jia, X. Sheng, A. Jackson, K. Moldenhauer, K. Yeater, A. McClung and D.X. Wu. 2011. Mapping QTLs for improving grain yield using the USDA rice mini-core collection. *Planta*, DOI 10.1007/s00425-011-1405-0.
- Little, R. R., G. B. Hilder and E. H. Dawson. 1958. Differential effect of dilute alkali on 25 varieties of milled white rice. *Cereal Chem.* 35: 111-126.
- Liu, K., and S.V. Muse. 2005. PowerMarker: integrated analysis environment for genetic marker data. *Bioinformatics* 21:2128-2129.
- Lynch, M., and B.G. Milligan. 1994. Analysis of population genetic structure with RAPD markers. *Mol. Ecol.* 3: 91-99.
- McClung, A., M. Chen, H.E. Bockelman, R.J. Bryant, W. Yan, and R. Fjellstrom. 2004. Characterization of a core collection of rice germplasm and elite breeding lines in the US with genetic markers associated with cooking quality. *Proc. 2nd Int'l Sym. Rice Functional Genomics*, Poster 127, Tucson, AZ, Nov. 15-17, 2004.
- McClung, A.M., W. Yan, Y. Jia, F.N. Lee, M.A. Marchetti, and R.G. Fjellstrom. 2006. Genotypic and phenotypic assessment of the NSGC core collection of rice for resistance to *Pyricularia grisea*. *Proc. 31st Rice Technical Working Group Conference*. Feb. 26-Mar. 1, 2006, Houston, TX.
- Nei, M. 1973. The theory and estimation of genetic distance. *In*: Morton NE (ed) *Genetic structure of populations*, Uni. Press Hawaii, Honolulu, HI, USA: 45-54.
- Nei, M. 1978. Estimation of average heterozygosity and genetic distance from a small number of individuals. *Genetics* 89:583-590.
- NSGC. 2011. National Small Grains Collection. [http://www.ars-grin.gov/cgi-bin/npgs/html/site\\_holding.pl?NSGC](http://www.ars-grin.gov/cgi-bin/npgs/html/site_holding.pl?NSGC) (verified on August 29, 2011)
- Paun, O., J. Greilhuber, E.M. Temsch, and E. Hörandl. 2006. Patterns, sources and ecological implications of clonal diversity in apomictic *Ranunculus carpaticola* (*Ranunculus auricomus* complex, Ranunculaceae). *Mol Ecol* 15:897-910.
- Peakall, R., and P.E. Smouse. 2006. GenAIEx 6: Genetic analysis in Excel. Population genetic software for teaching and research. *Mol Ecol Notes* 6:288-295.
- Pérez, C. M., and B.O., Juliano. 1978. Modification of the simplified amylose test for milled rice. *Starch/Stärke*, 30: 424-426.
- Rice Descriptors. 2011. Multiple Descriptors Query for Rice. [http://www.ars-grin.gov/cgi-bin/npgs/html/desc\\_form.pl?75](http://www.ars-grin.gov/cgi-bin/npgs/html/desc_form.pl?75) (Verified on September 12, 2011).
- SAS Institute. 2004. SAS/STAT user's guide. Version 9.1.3 ed. SAS Inst., Cary, NC.
- Schneider, S., D. Roessli., and L. Excoffier. 2000. Arlequin: a software for population genetic data. *Genetics and Biometry Laboratory, University of Geneva, Switzerland.*

- Spiegelhalter, D.J., N.G. Best, B.P. Carlin, and van der Linde A. 2002. Bayesian measures of model complexity and fit (with discussion). *J R Stat Soc B* 64:583–639.
- Steiner, J.J., P.R. Beuselinck, S.L. Greene, J.A. Kamn, J.H. Kirkbride, and C.A. Roberts. 2001. A description and interpretation of the NPGS birdsfoot trefoil core subset collection. *Crop Sci.* 41: 1968-1980.
- Szpiech, Z.A., M. Jakobsson, and N.A. Rosenberg. 2008. ADZE: a rarefaction approach for counting alleles private to combinations of populations. *Bioinform.* 24:2498-2504.
- UNSD. 2009. United Nations Statistics Division, Geographic regions and composition. <http://unstats.un.org/unsd/methods/m49/m49regin.htm>
- Venables, W.N. and B.D. Ripley. 1998. *Modern Applied Statistics with S+*, 2<sup>nd</sup> Edition. New York: Springer.
- Venables, W.N., D.M. Smith and Development Core Team. 2008. *Notes on R: A Programming Environment for Data Analysis and Graphics Version 2.8.1*. Vienna: R Foundation for Statistical Computing.
- Wackernagel, H. 1994. Cokriging versus kriging in regionalized multivariate data analysis. *Geoderma* 62:83–92.
- Webb, B.D. 1972. An automated system of amylose analysis in whole-kernel rice. *Cereal Sci. Today*, 30: 284.
- Weir, B., and C.C. Cockerham. 1984. Estimating *F* statistics for the analysis of population structure. *Evolution* 38:1358–1370.
- Xin, Z., J.P. Velten, M.J. Oliver, and J.J. Burke. 2003. High-throughput DNA extraction method suitable for PCR. *Bio. Techniques* 34:820-826.
- Yan, W., F.N. Lee, J.Neil Rutger, K.A.K. Moldenhauer, and J.W. Gibbons. 2002. Chinese germplasm evaluation for yield and disease resistance. R.J. Norman and J.-F. Meullenet, (eds.). *B.R. Wells Rice Research Studies 2001*. pp. 349-358, Univ. of AR, Agri. Exp. Sta., Res. Ser. 495.
- Yan, W., J.Neil Rutger, R.J. Bryant, F.N. Lee, and J.W. Gibbons. 2003. Characteristics of newly introduced accessions in the USDA-ARS rice quarantine program. R.J. Norman and J.-F. Meullenet, (eds.). *B.R. Wells Rice Research Studies 2002*. pp. 112-124, Univ. of AR, Agri. Exp. Sta., Res. Ser. 504.
- Yan, W., J. Neil Rutger, H.E. Bockelman, and T.H. Tai. 2005a. Agronomic evaluation and seed stock establishment of the USDA rice core collection. R.J. Norman, J.-F. Meullenet and K.A.K. Moldenhauer, (eds.), *B.R. Wells Rice Research Studies 2004*. pp. 63-68, Univ. of AR, Agri. Exp. Sta., Res. Ser. 529.
- Yan, W., J. Neil Rutger, H.E. Bockelman, and T.H. Tai. 2005b. Evaluation of kernel characteristics of the USDA rice core collection. R.J. Norman, J.-F. Meullenet and K.A.K. Moldenhauer, (eds.), *B.R. Wells Rice Research Studies 2004*. pp. 69-74, Univ. of AR, Agri. Exp. Sta., Res. Ser. 529.
- Yan, W., J. N. Rutger, R.J. Bryant, H.E. Bockelman, R.G. Fjellstrom, M.H. Chen, T.H.Tai, and A.M. McClung. 2007. Development and evaluation of a core subset of the USDA rice germplasm collection. *Crop Sci.* 47:869-878.
- Yan, W.G., H. Agrama, M. Jia, R. Fjellstrom and A. M. McClung. 2010. Geographic description of genetic diversity and relationships in the USDA rice world collection. *Crop Science* 50:2406-2417.

Yang, X.C., and C.N. Hwa. 2008. Genetic modification of plant architecture and variety improvement in rice. *Heredity*, 101:396-404.

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## **Food Production - Approaches, Challenges and Tasks**

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