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Full Length Research Paper

Genetic diversity analysis and subspecies classification of Thailand rice landraces using DNA markers

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Genetic diversity among 126 rice accessions, including 110 Thai landraces and 16 varieties used as subspecies reference, were evaluated by three types of DNA markers, InDel (Insertion/Deletion), inter-simple sequence repeat (ISSR) and simple sequence repeat (SSR) markers. Twelve InDel primer pairs, based on DNA sequence polymorphism between '93-11' (*indica*) and 'Nipponbare' (*japonica*), were used to identify subspecies of landraces. Most of the local rice samples had either '93-11' alleles or 'Nipponbare' alleles. The scatter plotting of the principal component analysis (PCA) and dendrogram results based on InDel data could clearly classify landraces into two groups, *indica* and *japonica*. InDel and SSR markers showed the average polymorphic information content (PIC) values of 0.3707 and 0.6367, respectively. The dendrogram, based on combining InDel, ISSR and SSR data, could classify rice samples into five clusters at a cut-off genetic similarity value of about 0.70. The genetic similarity within landraces was low, indicating that Thai local rice samples have a great genetic diversity. The results of this experiment provide helpful data for rice germplasm management in breeding program.

Key word: Rice, genetic diversity, DNA markers, simple sequence repeat (SSR), inter-simple sequence repeat (ISSR), insertion/deletion (InDel).

INTRODUCTION

Rice is one of the most important crops. It is grown all over the world and becomes a staple food of more than half of the world population. The cultivated rice species, *Oryza sativa* L., is commonly divided into two subspecies, *indica* and *japonica* (Oka, 1988). In geographical term, *indica* varieties are primarily known as lowland rice generally grown in tropical lowland, whereas *japonica* varieties are typically found in temperate East Asia, upland areas of Southeast Asia and high altitude regions in South Asia (Khush, 1997). *Japonica* is now often subdivided into two subgroups called temperate *japonica* and tropical *japonica* or *javanica* (Oka, 1958). Temperate japonica rice varieties have been found to be distributed mainly in the island of Western Pacific, ranging from Indonesia to Japan, and in certain parts of the continent in China and Korea while tropical *japonica* rice varieties have been distributed on mountainous regions at low-medium elevations in equatorial area (Oka and Chang, 1963).

Thailand has a long history of rice production which has led to diverse native landraces. The local rice germplasm may provide a valuable resource for rice improvement. These landraces are considered to contain high genetic variation which could diverse the gene pool for rice breeding program. Recently, the Indonesian tropical *japonica* landraces are used to develop new plant type (NPT) varieties which have fewer tillers, thick stems and

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Abbreviations: ISSR, Inter-simple sequence repeat; **SSR,** simple sequence repeat; **UPGMA,** unweighted pair group method with arithmetic mean; **PCA,** principal component analysis; **PIC,** polymorphic information content.

large panicles for more efficient grain production (Peng et al., 1999). The vast majority of Thai local rice germplasm remains underutilized. Some of the landraces are difficult to classify into *indica* or *japonica* subspecies when evaluated with morphological characters. The genetic potential of landraces differences from modern cultivars are still not determined. More recently, hybrid rice breeding is one of the promising approaches to develop high-yielding rice. A strong hybrid vigor of *indica-japonica* hybrid has attracted considerable interest of rice geneticists and breeders to developing super hybrid rice (Zhao et al., 1999; Xingxing et al., 2007).

The inter-subspecific crossing between landraces and modern cultivars could be used to broaden the genetic base of parental lines in hybrid rice breeding. Moreover, previous studies revealed that genetic diversity among the parental lines is certainly related to heterosis (Zhang et al., 1997; Liu et al., 2002). Breeding programs in China have integrated *japonica* components into *indica* background and vice versa. These *indica*-inclined and *japonica*-inclined lines have been applied as parental lines to develop super rice varieties (Cheng et al., 2007). Mao (2000) reported that *indica*-inclined or *japonica*-inclined lines are beneficial for a higher F_1 hybrid yield. Thus, subspecies classification and evaluation of genetic diversity of local rice may provide helpful data for germplasm management in rice breeding program.

Recently, molecular markers are powerful tools for evaluation of genetic diversity and easily identification of rice subspecies. Several types of polymerase chain reaction (PCR)-based markers are available today. These include simple-sequence repeats (SSRs) or microsatellites and inter-simple sequence repeats (ISSRs), which are both technical- and cost-efficient. ISSR markers span short chromosomal regions between SSRs (Parsons et al., 1997). The genomic distribution of SSRs in rice seems to be random, with no obvious bias for particular regions (Panaud et al., 1996; Chen et al., 1997). The development of InDel (Insertion/Deletion) markers, possible using tools of comparative genomics DNA sequences between 'Nipponbare' and '93-11', can facilitate the identification of *indica* and *japonica* subspecies (Shen et al., 2004). Some InDel markers can be used to accurately identify the *indica* and *japonica* cultivars (HanWei et al., 2007; Xingxing et al., 2007).

In this experiment, genetic diversity among 126 rice accessions, including 110 Thai landraces, 12 tropical *japonica* varieties from International Rice Research Institute (IRRI), two *indica* and two *japonica* varieties from Thailand Rice Germplasm Bank used as subspecies reference, were determined using three marker systems, InDel, SSR and ISSR markers. The outcome of this study will be useful for research on rice breeding in the future.

MATERIALS AND METHODS

Plant materials and DNA extraction

The 126 rice accessions consisted of four groups: 110 Thai landraces (No.1 to 110, some accessions have the same name), 12 tropical *japonica* accessions (No.111 to 122), two *indica* accession (IR24 and IR36, No. 123 to 124) and two *japonica* accessions ('Nipponbare' and 'Koshihikari', No. 125 to 126) were used in this study. List of rice accessions are shown in Appendix 1. Genomic DNA was extracted from young leaf tissues according to the Cetyl Trimethylammonium Bromide (CTAB) method following the procedure of Webb and Knapp (1990). The genomic DNA was diluted to 20 ng/ul and then used for PCR amplification.

Inter-simple sequence repeats (ISSR) marker genotyping

A total of 15 primers were screened. PCR reactions were carried out in 12.5 µl volumes containing 20 ng of genomic DNA, 20 mM Tris-HCl pH 8.4, 50 mM KCl, 2.0 mM $MgCl_2$, 0.1 mM of each dNTP, 5 pmoles primer and 1 U of *Taq* DNA polymerase (Invitrogen, Brazil). Amplification was performed in a PTC-100 (Perkin Elmer) thermocycler programmed for 5 min at 94°C, followed by 35 cycles of 30 s at 94°C, 1 min at 45°C and 1 h 30 min at 72°C, ending with 10 min at 72°C to complete extension. Amplification products were analyzed by gel electrophoresis on 1.5% agarose in 1× Tris borate EDTA (TBE) buffer and visualized by ethidium bromide staining. Four primers, namely $(CT)_8G$, $(GA)_8T$, $(AG)_8YT$ and $(GATA)_4$, generated clear polymorphic bands were selected to estimate rice genetic diversity.

InDel marker genotyping

Indel primer pairs were selected on the basis of accuracy in identifying *indica* and *japonica* (Shen et al., 2004; HanWei et al., 2007; Xingxing et al., 2007). A total of 12 primer pairs, one primer in each chromosome, were employed to analyze the genetic diversity among 126 rice samples. The PCR reactions were performed in 12.5 µl of a mixture containing 20 ng DNA, 10 mM Tris-HCl pH 8.8, 50 mM KCl, 0.08% Nonidet P40, 0.2 µM of each InDel primer, 2.0 mM $MgCl_2$, 0.1 mM of each dNTP and 1 U of *Taq* DNA polymerase (Fermentas, Canada). The amplification condition was 5 min at 94°C, followed by 35 cycles of 1 min at 94°C, 1 min at 55°C and 30 s at 72°C, and finally for 10 min at 72°C to complete extension. The amplified products were electrophoresed in 1.5% agarose gel, followed by ethidium bromide staining and visualization under ultraviolet (UV) light.

Simple sequence repeats (SSR) marker genotyping

Screening the rice database (www.gramene.org), 18 primers were selected for genotyping. The PCR amplification for SSR analysis in 12.5 µl reaction contained 20 ng DNA, 10 mM Tris-HCl pH 8.8, 50

Table 1. InDel markers used and their chromosome location, total number of alleles, total number of samples containing '93-11' alleles, 'Nipponbare' alleles, other alleles and null alleles, H_o and PIC values generated by each primer.

*InDel primer	Chromosome location	Number of alleles	Number of samples containing '93-11' alleles	Number of samples containing 'Nipponbare' alleles	Number of samples containing other alleles	Number of samples containing null alleles	H_o	PICs or H_e
R1M7	1	3	87	41	2	-	0.0072	0.2922
R2M10	2	2	87	41	-	-	0.0048	0.2888
R3M10	3	2	94	34	-	-	0.0048	0.2464
R4M13	4	4	68	51	9	-	0.0482	0.5663
R5M20	5	3	65	62	2	-	0.0096	0.4733
R6M14	6	3	90	38	2	-	0.0120	0.3651
R7M7	7	2	43	83	-	1	0.0120	0.5050
R8M33	8	3	65	64	3	-	0.0193	0.4196
R9M42	9	2	87	41	-	-	0.0048	0.2925
R10M17	10	3	108	20	3	-	0.0193	0.2110
R11M40	11	3	62	65	2	1	0.0144	0.3977
R12M27	12	3	80	47	1	-	0.0120	0.3901
Average		2.75	78	49	2	0.17	0.0179	0.3707

*InDel primer designed by Shen et al. (2004).

mM KCl, 0.08% Nonidet P40, 0.1 µM of each SSR primer, 1.5 mM $MgCl_2$, 0.1 mM of each dNTP and 1 U of *Taq* DNA polymerase (Fermentas, Canada). The amplification using a denaturation period of 5 min at 95°C followed by 35 cycles of 1 min at 95°C, 45 s at 55°C and 45 s at 72°C, and then 5 min at 72°C for final extension. PCR products were resolved on 6% denaturing polyacrylamide gel. After electrophoresis, bands were revealed using the silver-staining procedure described by Caetano-Anolles (1997).

Data analysis

The amplified DNA fragments from SSR and ISSR markers were scored as present (1) or absent (0) for analysis with NTSYS-pc version 2.2k (Rohlf, 2005). Genetic similarity were calculated using dice coefficient (Sneath and Sokal, 1973) by SIMQUAL subprogram and SAHN subprogram was used for cluster analysis by unweighted pair group method with arithmetic mean (UPGMA) method. Relationships between the rice accessions were portrayed in the form of dendrogram.

For InDel markers, banding patterns of each sample were scored based on the banding patterns of '93-11' (typical reference for *indica*) and 'Nipponbare' (typical reference for *japonica*) which were reported earlier by Shen et al. (2004). The allele patterns that were different from '93-11' and 'Nipponbare' were scored according to molecular weight. Other known *indica* and *japonica* references, for example IR20, IR22, IR48, IR50, IR64, PSBRc1, PSBRc2, Matatag1, Matatag2, Akitakomachi, were also examined and were found to contain at least eight alleles of '93-11' (*indica*) or eight alleles of 'Nipponbare' (*japonica*) (data not shown). Therefore, rice samples that have at least eight alleles of '93-11' or eight alleles of 'Nipponbare' were identified as *indica* and *japonica* types, respectively. The rice accessions which cannot be identified into *indica* or *japonica* were classified into intermediate type. All InDel bands were transferred into 0, 1 data matrix, and then were subjected to principal component analysis (PCA) using the implemented program in NTSYS-pc. The dendrogram was also performed as described above. Polymorphic information contents (PICs) or expected heterozygosity (H_e) were estimated for each marker by $1 - \Sigma^{k}_{i=1} p_i^2$ where p_i is the frequency of i^{th} allele and k is the number of alleles (Ott, 1991). PIC values range from 0 to 1. Observed heterozygosity (H_o) is a proportion of the observed heterozygotes.

RESULTS AND DISCUSSION

Subspecies classification of landraces by InDel markers

A total of 12 InDel markers, designed by comparative genomic study on DNA sequences between '93-11' and 'Nipponbare', one marker in each chromosome, were used to classify subspecies of 110 Thai landrace rice samples. The number of alleles per marker varied between two to four, with an average of 2.75. The PIC values ranged from 0.2110 to 0.5663, with an average of 0.3707. Observed heterozygosity (H_o) ranged from 0.0048 to 0.0482, with an average score of 0.0179 (Table 1). PCR analysis indicated that most of the landraces had the same alleles as referenced *indica* ('93-11') and *japonica* ('Nipponbare') cultivars. The other alleles and null alleles appeared in some samples (Table 1), suggesting that the mutation occurred in the DNA regions of InDel marker. Although the polymorphic regions might not directly affect the phenotyphic variations between *indica* and *japonica* rice, the polymorphic markers were proved to be effective in identifying rice subspecies. The *indica* cultivars, IR24 and IR36, possessed 11 and 9 '93-11' alleles, respectively; whereas *japonica* cultivar, 'Koshihikari', carried 'Nipponbare' alleles in all 12 loci.

Table 2. Total number of amplified bands and polymorphism revealed by ISSR analyses.

ISSR primer	Total number of bands	Number of polymorphic bands	% polymorphic band	PIC
$(GA)_8T$	7	6	85.71	0-0.3367
$(CT)_8G$	5	2	40.00	0-0.4938
$(AG)_8Y^*T$	10	6	60.00	0-0.4980
$(GATA)_4$	11	8	72.73	0-0.4390
Average	8.25	5.5	66.67	0.1798

Y* = C or T.

This result was consistent with the previous studies (HanWei et al., 2007; Xingxing et al., 2007), demonstrated that some *japonica* alleles might still exist in the *indica* rice. All 12 tropical *japonica* accessions had at least 7 'Nipponbare' alleles, demonstrating that tropical *japonica* has close relationships to *japonica* at the molecular level.

The genetic diversity among the rice samples was shown by PCA in the scatter plotting (Figure 1). The first, second and third principal components were accounted for 42.17, 10.57 and 7.79% of the total variation, respectively. The *indica* and *japonica* samples were clearly separated into two major groups. The tropical *japonica* samples dispersed near *japonica* accessions. There were two landraces (No. 76 and 109) scattered between two major groups because many InDel loci of both landraces were heterozygous. A dendrogram was also constructed with InDel genotypic data (Figure 2). Two major groups among *indica* and *japonica* were observed, similar to the scatter plotting. A low genetic similarity coefficient (GS) of about 0.27 was detected between the two groups compared to the high similarity within the *indica* (GS = 0.72 to 1.00) and the *japonica* (GS = 0.67 to 1.00). The *japonica* clade could be divided into three subgroups, tropical *japonica*, *japonica* and the two heterozygous landraces. Interestingly, twenty-four Thai landraces were classified into tropical *japonica* subgroup. These tropical *japonica* landraces could be used in rice breeding program to develop a new *indica-japonica* hybrid with a strong hybrid vigor. Moreover, the rice sample accession no. 24 was placed in the same cluster as the typical japonica, 'Nipponbare' and 'Koshihikari'.

Most of tropical *japonica* landraces were grown under upland (unflooded) conditions and some of them were from plain region in northern Thailand. It has long been reported that some cultivated rice varieties grown by the native people in northern Thailand exhibited *japonica*-like features. Oka and Chang (1963) investigated the rice varieties obtained from northern Thailand. Many characteristics and F_1 sterility relationship were evaluated. The results reveal that some rice varieties from the mountainous region appeared to be intermediate between *indica* and *japonica* subspecies. The intermediate types were difficult to identify using phenotypical characteristics. In this study, the InDel markers were proved to be very useful as a tool for classifying rice subspecies in breeding program.

Genetic diversity revealed by combination of inter-simple sequence repeats (ISSR), simple sequence repeat (SSR) and InDel markers

Two marker systems, SSR and ISSR, were also employed to analyze the genetic diversity of 126 rice accessions. Genetic diversity was assessed by four ISSR primers, and a total of 33 DNA bands were generated with an average of 8.25 bands per primer (Table 2). The ISSR pattern generated by $(GA)_8T$ primer was highly polymorphic, containing 85.71% polymorphic bands. Totally, 22 out of 33 (66.67%) DNA bands were found to be polymorphic. PICs ranged from 0 to 0.4980, with an average of 0.1798. Eighteen SSR markers were also performed, and 92 alleles were produced, with an average of 5.11 alleles per locus (Table 3). All the SSR markers used, RM241 produced 10 alleles which were the maximum number of alleles obtained from SSR primers on this set of rice samples; while the minimum of alleles (two alleles) generated from RM390 were observed. PIC scores ranged from 0.1189 to 0.8513, with an average of 0.6367. SSR markers provided high PIC values compare to ISSR because of their multiallelism. Most of the SSR markers (14 out of 18 markers) were highly informative with PIC values higher than 0.5. The loss of allelic diversity for RM390 may be described by its location in the transcribed region of the cytochrome *b5* gene rather than in non-transcribed DNA (Blair and McCouch, 1997). Observed heterozygosity (H_o) ranged from 0 to 0.0317, with an average of 0.0119.

As shown in Figure 3, genetic relationships among rice samples were evaluated with combined data from ISSR, SSR and InDel markers. A combination of markers will provide whole genome coverage and reduce the errors in genetic similarity evaluation based on any one marker system alone (Davierwala et al., 2000). The dendrogram revealed five clusters at a cut-off similarity coefficient level of about 0.70. The *indica* landraces were divided into cluster 1 and 2, while the two typical *indica* cultivars were in cluster 3; suggesting *indica* landraces have some

Table 3. SSR markers used and their chromosome location, H_o and PIC values generated by each primer.

*Locus name	Chromosome	Total number of alleles	H_o	PICs or H_e
RM11	7	6	0.0238	0.7370
RM12	12	4	0.0159	0.6136
RM13	6	6	0.0079	0.7446
RM19	12	9	0.0238	0.7919
RM72	8	3	0	0.6541
RM126	8	3	0.0079	0.4841
RM190	6	4	0.0079	0.5880
RM224	11	7	0.0159	0.6807
RM241	4	10	0.0238	0.8513
RM250	2	5	0.0079	0.4965
RM264	8	5	0.0159	0.7687
RM276	6	6	0.0079	0.7352
RM287	11	4	0.0159	0.4934
RM307	4	4	0	0.5378
RM333	10	3	0	0.6209
RM334	5	5	0.0079	0.7726
RM335	4	6	0.0317	0.7702
RM390	5	2	0	0.1189
Average		5.11	0.0119	0.6367

*From rice database (www.gramene.org).

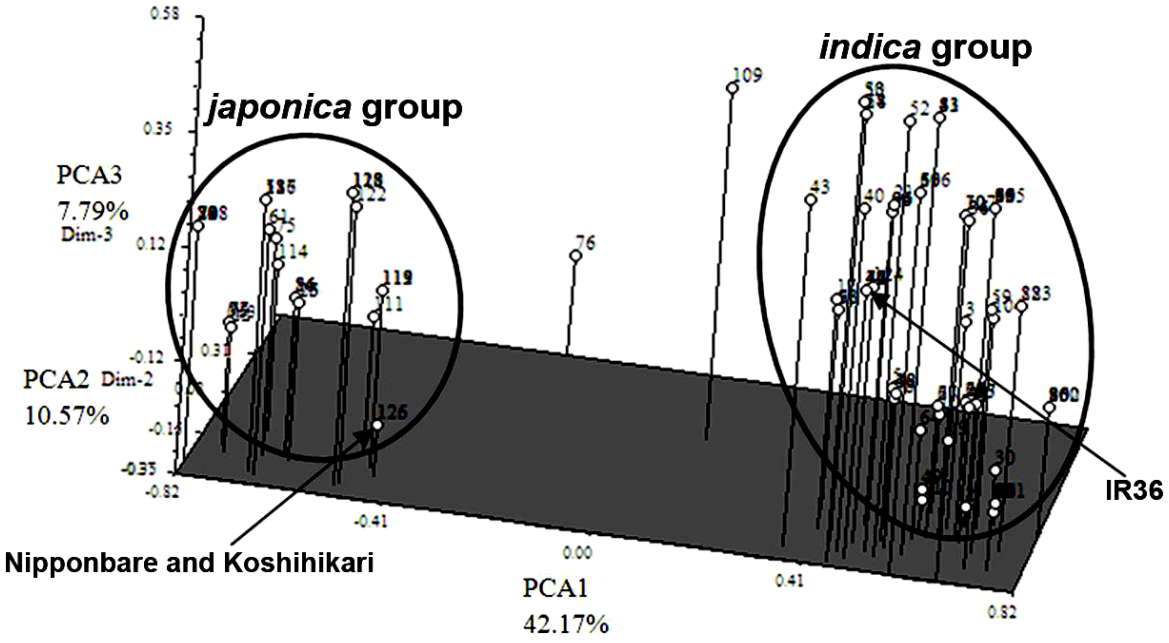

Figure 1. The PCA scatter plotting showing the relationships among the *indica* and *japonica* rice accessions based on 12 InDel markers. Reference varieties are labeled.

degree of genetic differences from the typical *indica* cultivars. The seeds of *indica* landraces came from cultivated rice varieties grown by the native people from different parts of the country, whereas the seeds of the two typical *indica* cultivars came from Pathum Thani Rice Research Center, Thailand. All samples are collected and

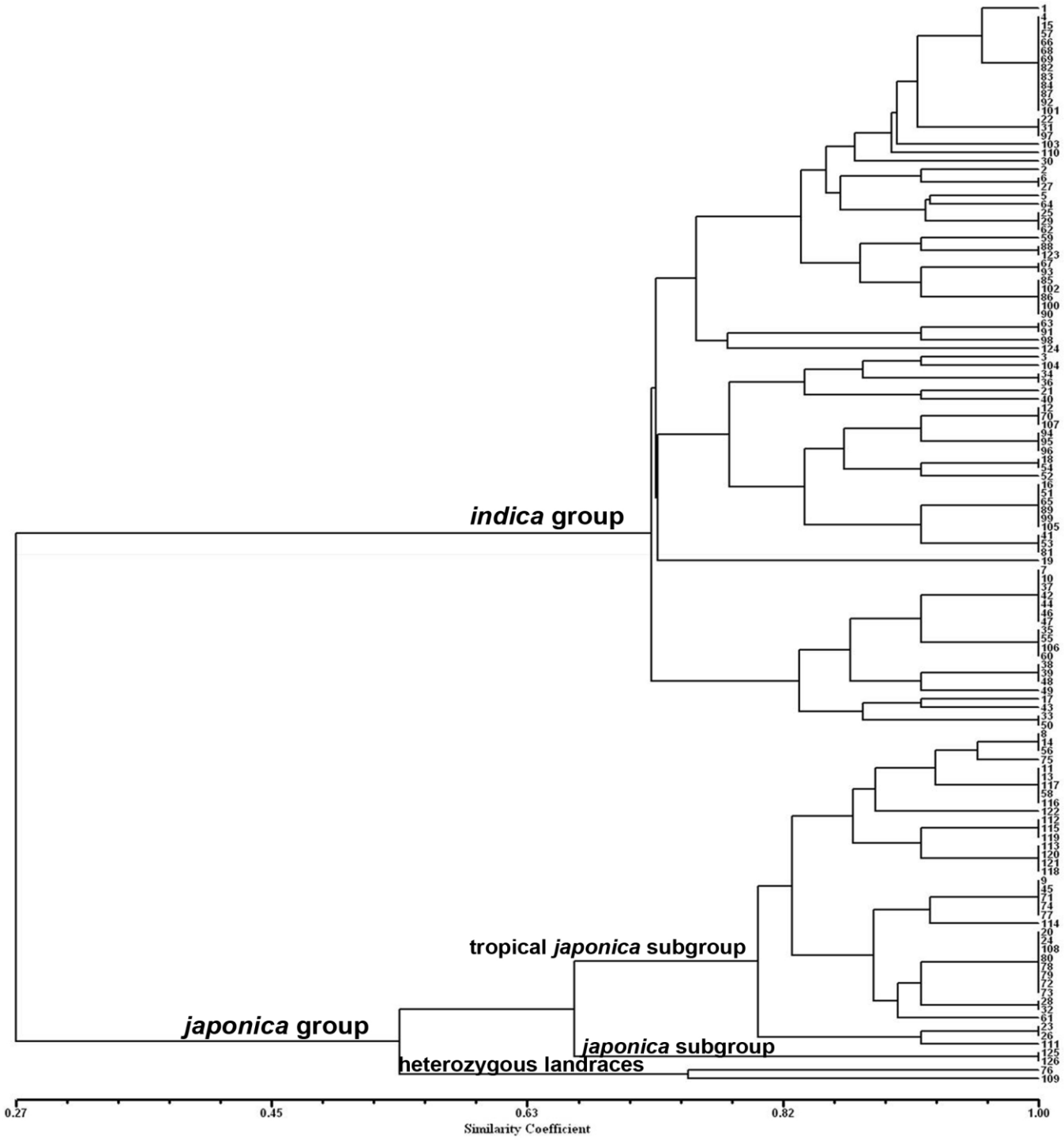

Figure 2. The UPGMA dendrogram showing the genetic relationships among 126 rice accessions based on 12 InDel markers. The major groups and subgroups are labeled.

maintained lines at Thailand Rice Germplasm Bank. Although some local rice samples collected from different parts of the country had the same name, these landraces were not genetically identical. The tropical *japonica* and *japonica* rice samples were in cluster 4 and the two heterozygous landraces were in cluster 5.

When the genetic similarity was analyzed within *indica* and *japonica* groups, the *japonica* group had slightly higher genetic similarity at 0.71 compared to 0.67 for the *indica* group. The genetic similarity coefficient within only

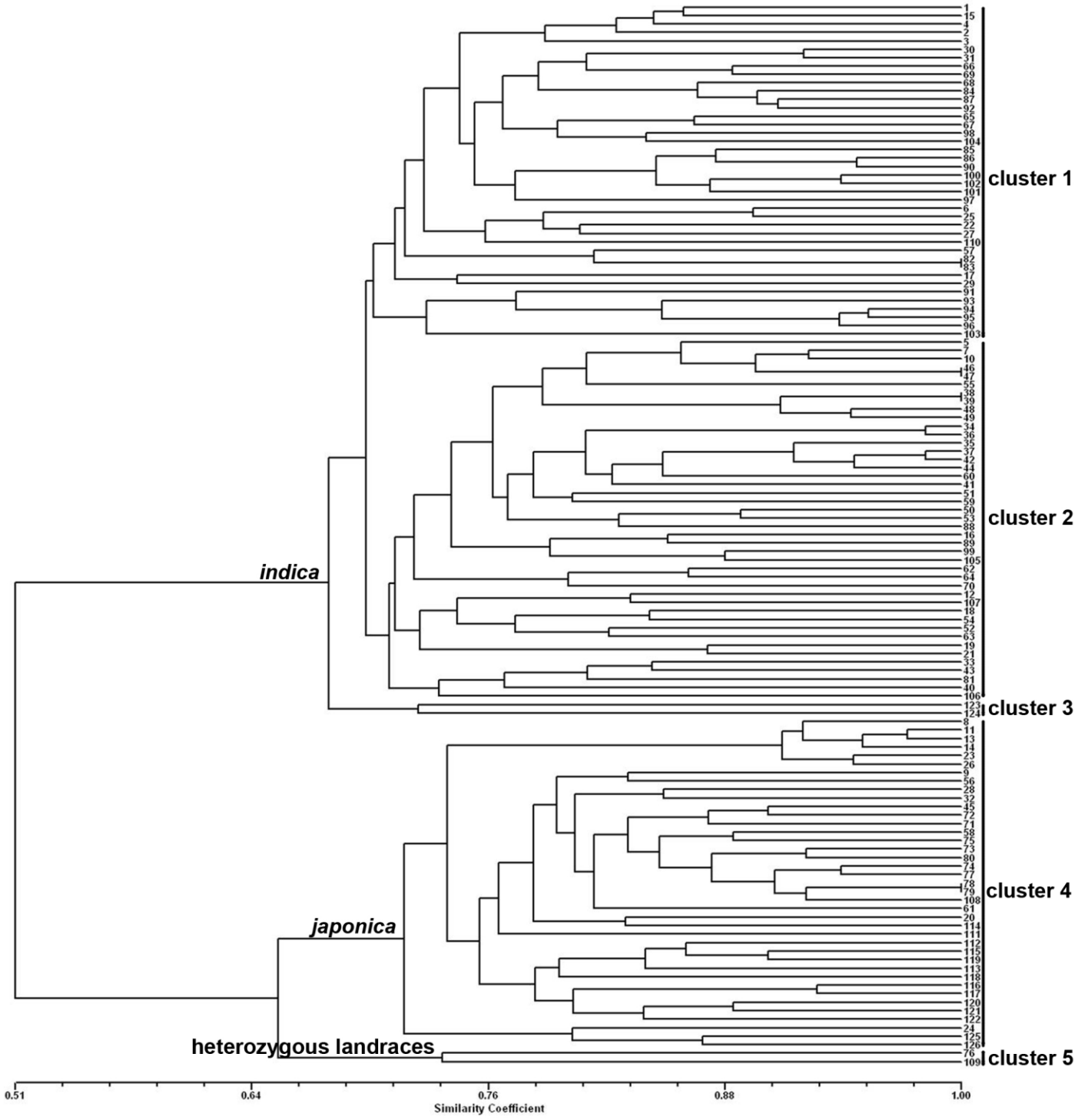

Figure 3. The UPGMA dendrogram showing the genetic relationships among 126 rice accessions based on combined data from ISSR, SSR and InDel markers.

landrace samples was 0.52, indicating that landrace germplasms had a great genetic diversity. Based on three marker systems in this study, 25 landraces were classified as *japonica* or tropical *japonica*. These *japonica* or tropical *japonica* local rice samples could be used to broaden the genetic diversity of Thai modern cultivars. The IRRI's research has demonstrated the potential of tropical *japonica* to improve rice varieties. The new plant

type varieties have been developed by IRRI, using Indonesian tropical *japonica* germplasm (Virk et al., 2004). Notably, the heterozygous landraces were found when investigated by InDel and SSR markers. Thus, the pure line selection is required before using these landraces in breeding program.

Conclusion

Genetic diversity among 126 rice accessions, including 110 Thai landraces and 16 varieties used as subspecies reference, were determined using three marker systems. The results reveal that Thai landraces have a great genetic diversity. The subspecies classification using InDel markers and genetic diversity evaluated by InDel, SSR and ISSR markers provided very useful data to exploit landraces for future research on rice breeding.

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Appendix 1. List of rice samples used in this experiment.

Sample #	Name (*)	Sample #	Name (*)
1	Daeng (A001)	64	Kee Tom Leuang (D015)
2	Lueang Bunma (D064)	65	Tub Mai (D016)
3	Dor (D065)	66	Ta Mai (D017)
4	Khao Ai Choet (A022)	67	Gor Diaw (D018)
5	Hom Look Rung (A023)	68	Ngun Khao (D019)
6	Tum (A026)	69	Mah Mum (D020)
7	Hom Peun Murng (A027)	70	Sarn Tua (D021)
8	Dok Phayom (A030)	71	Dawk Gai (D022)
9	Jao (A035)	72	Dawk Tong (D023)
10	(A036)	73	Ma Kaw (D024)
11	Med Pum Lai (A037)	74	Nuan Yai (D025)
12	Khai Riang (A038)	75	Nuan Noi (D026)
13	Khai Riang (A039)	76	Peek (D027)
14	Khai Riang (A040)	77	Yuak (D028)
15	Chao Daeng (A041)	78	Hom Mah Teun (D029)
16	Chao Daeng (A042)	79	Sarn Tua (D030)
17	Nok (A043)	80	Tab Daeng (D031)
18	Leuang Tong Lao Tak (D066)	81	Pla sew Dawk Kuu (D033)
19	Ngow Ratin (A045)	82	Pla sew Dawk Kuu (D034)
20	Ngow Boo Meu Sa Ngur-ah (A046)	83	Gor Diaw (D035)
21	Ngow Peun (A047)	84	Nuan Dor (D036)
22	Khao Yai (A048)	85	Pla sew (D037)
23	Khai Riang (A052)	86	Dor, Khao Mae Hahng (D038)
24	Ja Gor Law (A053)	87	Hom Tung (D039)
25	E-To (A057)	88	Dor Ubon (D040)
26	Kai Reeang (A058)	89	Dor Pla sew (D041)
27	Gum (C001)	90	E-Nang Toon (D042)
28	Bao Hua Bon (C004)	91	Khao Dor (D043)
29	Mali Daeng (C015)	92	Bug Kham Dor (D044)
30	Gum (C017)	93	Dor Nang Bunma (D045)
31	Gum Lau Suu (C021)	94	Dor Nang Bunma (D047)
32	Niao Daeng (C022)	95	Leuang Bunma (D048)
33	Peun Murng (C023)	96	Dor Nang Wee (D049)
34	Peun Murng (C024)	97	E-Ped Noi (D050)
35	Peun Murng (C025)	98	Hom Tom (D051)
36	Gum (C026)	99	Pong Aow (D052)
37	Daeng (C027)	100	Niaw Pla sew (D053)
38	Gum (C028)	101	Niaw Pong Aow (D054)
39	How (C029)	102	E-Gerd (D055)
40	Niao Dam (C031)	103	Mun Ped (D056)
41	Gum, Niaw (C032-1)	104	E-Tom (D057)
42	E-Non Daeng (C033)	105	Niaw phrae Rai (D058)
43	Dam, Gum (C034)	106	Dor Mer-ei (D059)
44	Ah Bai (C035)	107	Leb Chang (D060)
45	Man Pu (C037)	108	Pleek (D061)
46	Jib, Daeng (C038)	109	Pong Aow (D062)
47	Niaw Dam (C040)	110	Leuang Bunma (D063)
48	Niaw Daeng Num Mahk (C041)	111	Ase Bolong
49	Gum (C042)	112	Djawa Pelet
50	Khao Yai (D001)	113	Gundil Kuning IRGC 16428
51	Kee Tom Yai (D002)	114	Gundil Kuning IRGC 1983DS

Appendix 1. Contd.

52	Lao Tak (D003)	115	Gundil Kuning IRGC 27129
53	Mae Peung (D004)	116	Kemandi Pance
54	Hom Puu Parn (D005)	117	Ketan Lumbu IRGC 16461
55	E-Glieng (D006)	118	Loas Gedjeh IRGC9243
56	Sew Mae Jan (D007)	119	Ribon
57	Khao Pla Sew (D008)	120	Sarimahi IRGC34632
58	Gan Lau Hug (D009)	121	Sengkeu
59	Gum Num Poon (D010)	122	Sopongono
60	Praya Leum Gang (D011)	123	IR24
61	Kiow (D012)	124	IR36
62	E-Po (D013)	125	Nipponbare
63	Som Samai (D014)	126	Koshihikari

*****Thailand Rice Germplasm Bank accession number and names are based on Thai Royal Institute (http://www.royin.go.th). #1 to 110, Thailand rice landraces; #111 to 122, tropical *japonica* rice (from IRRI); #123 to 124, *indica* rice; #125 to 126, *japonica* rice.