CHAPTER 2

Characterization of Weedy Rice Populations from Three Rice Production Areas of Thailand

2.1 Introduction

Weedy forms of rice are known as weedy rice (*Oryza sativa* f. *spontanea*). In the rice fields, it is one of the most serious weed, adversely affecting rice areas and yield worldwide, particularly in South and Southeast Asia, South and North America, and southern Europe (Noldin, 2000; Vaughan *et al*., 2001; Gealy *et al*., 2002). Although, weedy rice is taxonomically classified as the same species as cultivated rice (*Oryza sativa* L.), they are still strongly characterized by their seed shattering and dormancy (Cao *et al*., 2006).

Weedy rice in Thailand is originates from interspecific hybridization between common wild rice (*Oryza rufipogon* Griff.) and cultivated rice (Niruntrayakul, 2007) due to Thailand lies partly within the center of genetic diversity of *Oryza*. Weedy rice is frequently found in the sympatric habitats where wild and crop rice are co-exist, and occur in the area where absence of wild populations (Oka, 1988; Song *et al*., 2006). Such hybridization between cultivated and common wild rice is a common occurrence when their flowering period overlaps, through the time of flowering peak of wild rice is either earlier or later than the cultivated rice (Chitrakon, 1995).

In the past decade, weedy rice was firstly observed in rice production areas in Thailand (Maneechote, 2004) and became a serious weed problem where first found in the Central plain. Recently, weedy rices have been increasingly reported widespread in many directed-seed paddy fields in the Lower North and the Northeast. However, weedy rice from each rice production areas originated from independent hybridization between native common wild rice and local popular cultivated rice varieties (Pusadee, 2009). Therefore, high level of genetic differentiation among weedy rice populations indicated that weedy rice populations were differentiated according to their companion cultivated rice of each rice production area.

Generally, weedy rices are diverse and changeable in genetic variation and population structure, which are supposed to be the ongoing evolutionary processes. The significance of population genetic structure and dynamics contributed to increase adaptation to farmers' management and is particularly difficult to control. Therefore, the objectives of the present study are as followed: (1) to evaluate genetic variation of morphological and physiological characters and microsatellite loci of weedy rice populations in Thailand and, (2) to determine population genetic structure and dynamics of weedy rice populations in spatial and temporal terms at the regional scale.

2.2 Materials and Methods

Characterization and collection of weedy rice populations in rice fields

Sixty-three weedy rice populations in farmers' fields were surveyed and studied from three of Thailand's main rice producing regions where heavy infestation of weedy rice has been spreading, namely, the Lower North, the Northeast and the Central Plain (Figure 2.1.1) in four rice growing seasons (wet seasons of 2005, 2008, 2009 and dry season of 2009/10).

In each field, location was geo-referenced, weedy rice type and some morphological and physiological characters of weedy rice; culm length compared with companion cultivated rice, the presence of awn on spikelet, rate of seed shattering, hull and pericarp color, were recorded. Leaves of 10 weedy rice plants of were collected individually, silica-dried following the method described by Chase and Hill (1991) and stored at –20°C until used. Seed of each population were collected and bulked for the pot experiment. In each season, fields with heavy infestation with weedy rice were selected. Leaf samples of 10 plants of crop rice and weedy rice in the same field were collected and DNA analyzed individually. DNA from weedy rice and crop rice samples were compared with seven cultivated rice varieties popular in each region and seven native common wild rice populations collected from the Upper North, the Lower North, the Northeast and the Central Plain of Thailand.

Table 2.1.1 Location and description of weedy rice populations

No.	Weedy rice Accession No.	Region	Province	Year collection*	Rice variety in invaded fields**	UTM
1	WeLN5001	Lower North	Phitsanulok	2005-W	MV	N17.15139 E100.06993
2	WeLN5002		Phichit	2005-W	MV	N16.36638 E100.33548
3	WeLN5003		Kampaeng Phet	2005-W	MV	N16.67962 E099.62512
4	WeNE5001	Northeast	Surin	2005-W	MV/ITV	N14.78988 E103.83912
5	WeNE5002		Buri Rum	2005-W	MV/ITV	N14.62856 E103.24280
6	WeNE5003		Sri Saket	2005-W	MV/ITV	N14.95359 E104.20601
7	WeCP5001	Central Plain	Suphan Buri	2005-W	MV	N14.55998 E100.13917
8	WeCP5002		Sing Buri	2005-W	MV	N14.96038 E100.35778
9	WeLN8001	Lower North	Phitsanulok	2008-W	MV	N17.12766 E100.15588
10	WeLN8002		Phitsanulok	2008-W	MV	N16.79237 E100.38881
11	WeLN8003		Phitsanulok	2008-W	MV	N16.75915 E100.36521
12	WeLN8004		Phichit	2008-W	MV	N16.22924 E100.12810
13	WeLN8005		Phichit	2008-W	MV	N16.51697 E100.14441
14	WeNE8001	Northeast	Maha Sarakham	2008-W	MV/ITV	N15.71499 E103.07142
15	WeNE8002		Surin	2008-W	MV/ITV	N15.37344 E103.26794
16	WeNE8003		Roi Et	2008-W	MV/ITV	N15.80604 E103.93804
17	WeNE8004		Roi Et	2008-W	MV/ITV	N15.66168 E104.13103
18	WeNE8005		Yasothon	2008-W	MV/ITV	N15.62547 E104.29988
19	WeNE8006		Si Saket	2008-W	MV/ITV	N14.88874 E104.52992
20	WeNE8007		Si Saket	2008-W	MV/ITV	N14.90910 E104.41262
21	WeNE8008		Buri Rum	2008-W	MV/ITV	N15.34627 E103.27923
22	WeNE8009		Buri Rum	2008-W	MV/ITV	N15.46947 E103.09297
23	WeNE8010		Ubon Ratchathani	2008-W	MV/ITV	N15.12073 E104.83675
24	WeCP8001	Central Plain	Lop Buri	2008-W	MV	N15.07019 E100.99284
25	WeCP8002		Nakhon Nayok	2008-W	MV	N14.12339 E101.18039
26	WeCP8003		Ayutthaya	2008-W	MV	N14.42070 E100.77249
27	WeCP8004		Saraburi	2008-W	MV	N14.01070 E101.77691
28	WeCP8005		Prachin Buri	2008-W	MV	N14.05872 E101.60651

*year collection; 2005-W = wet 2005, 2008-W = wet 2008, 2009-W = wet 2009 and 2009/10-D = dry 2009/10

**MV=Modern variety, ITV=Improved traditional variety

Table 2.1.1 *(continued)*

No.	Weedy rice Accession No.	Region	Province	Year collection*	Rice variety in invaded fields**	UTM
29	WeLN9101	Lower North	Phitsanulok	2009-W	MV	N16.71645 E100.17924
30	WeLN9102		Phitsanulok	2009-W	MV	N17.14936 E100.08614
31	WeLN9103		Phichit	2009-W	MV	N16.22255 E100.12900
32	WeLN9104		Phichit	2009-W	MV	N16.22255 E100.12900
33	WeLN9105		Uttaradit	2009-W	MV	N17.33998 E100.05503
34	WeLN9106		Nakhon Sawan	2009-W	MV	N15.11861 E100.38908
35	WeNE9101	Northeast	Maha Sarakham	2009-W	MV/ITV	N16.33145 E103.10417
36	WeNE9102		Maha Sarakham	2009-W	MV/ITV	N16.26742 E103.08734
37	WeNE9103		Maha Sarakham	2009-W	MV/ITV	N16.25039 E103.07999
38	WeNE9104		Maha Sarakham	2009-W	MV/ITV	N16.25039 E103.07999
39	WeNE9105		Surin	2009-W	MV/ITV	N15.38288 E103.70244
40	WeNE9106		Surin	2009-W	MV/ITV	N15.24002 E103.66118
41	WeNE9107		Surin	2009-W	MV/ITV	N15.24245 E103.67483
42	WeNE9108		Surin	2009-W	MV/ITV	N14.89289 E103.66179
43	WeNE9109		Buri Rum	2009-W	MV/ITV	N15.16348 E103.19043
44	WeNE9110		Buri Rum	2009-W	MV/ITV	N15.35632 E103.00376
45	WeCP9101	Central Plain	Saraburi	2009-W	MV	N14.01070 E101.77691
46	WeCP9102		Sing Buri	2009-W	MV	N14.96109 E100.36876
47	WeLN9201	Lower North	Phitsanulok	2009/10-D	MV	N17.12307 E100.10931
48	WeLN9202		Phitsanulok	2009/10-D	MV	N17.12261 E100.10836
49	WeLN9203		Uttaradit	2009/10-D	MV	N17.29216 E100.10100
50	WeLN9204		Phichit	2009/10-D	MV	N16.22924 E100.12810
51	WeLN9205		Kampaeng Phet	2009/10-D	MV	N16.20628 E099.75140
52	WeLN9206		Sukhothai	2009/10-D	MV	N17.01357 E099.91210
53	WeLN9207		Nakhon Sawan	2009/10-D	MV	N15.14715 E100.36617
54	WeNE9201	Northeast	Khon Kaen	2009/10-D	MV/ITV	N16.77008 E102.69616
55	WeNE9202		Khon Kaen	2009/10-D	MV/ITV	N16.77008 E102.69616
56	WeNE9203		Kalasin	2009/10-D	MV/ITV	N16.32580 E103.62952
57	WeNE9204		Kalasin	2009/10-D	MV/ITV	N16.32921 E103.62780
58	WeNE9205		Maha Sarakham	2009/10-D	MV/ITV	N16.31350 E103.02975
59	WeNE9206		Ubon Ratchathani	2009/10-D	MV/ITV	N15.29012 E104.59731
60	WeCP9201	Central Plain	Ayutthaya	2009/10-D	MV	N14.42070 E100.77249
61	WeCP9202		Suphan Buri	2009/10-D	MV	N14.77093 E099.89891
62	WeCP9203		Suphan Buri	2009/10-D	MV	N14.77093 E099.89891
63	WeCP9204		Sing Buri	2009/10-D	MV	N14.96038 E100.35778

*year collection; 2005-W = wet 2005, 2008-W = wet 2008, 2009-W = wet 2009

and 2009/10-D = dry 2009/10

**MV=Modern variety, ITV=Improved traditional variety

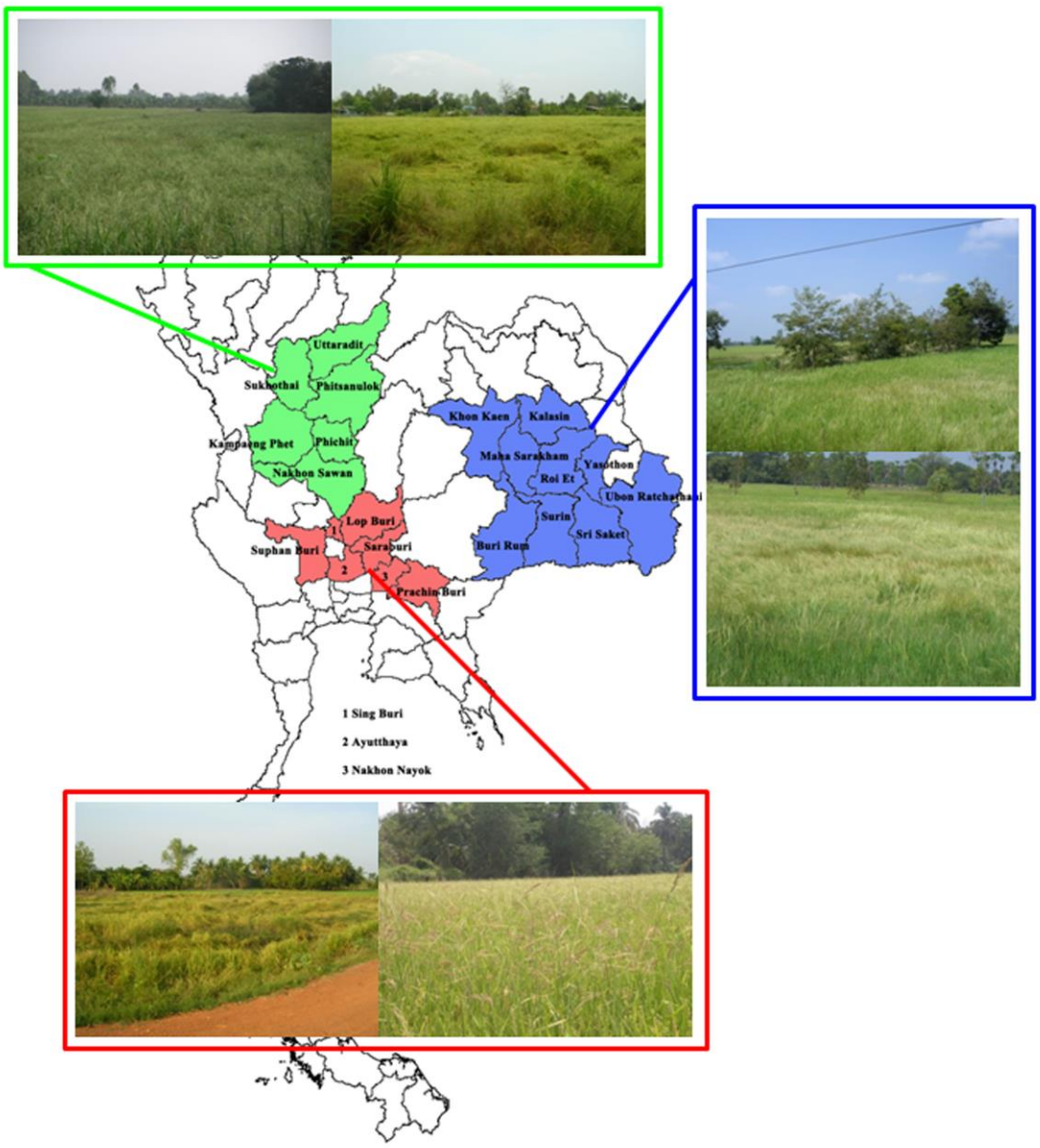


Figure 2.1.1 Location of weedy rice sampling in Thailand of the present study

Table 2.1.2 Common wild rice accession, location and habitat in this study

No.	Accession No.	Region	Province	Habitat condition	Type*	Year collection	UTM
1	WiUN1	Upper North	Chiang Mai	irrigation canal	P	2005	
2	WiUN2		Lamphun	small marsh in rice field	P	2005	N18.57297 E098.98835
3	WiLN1	Lower North	Phitsanulok	marsh	P	2005	N16.48659 E100.21312
4	WiLN2		Phichit	marsh	P	2005	N16.42549 E100.31940
5	WiNE1	Northeast	Sakon Nakon	deep swamp, *in situ* conservation area	P	2005	
6	WiCP1	Central Plain	Ayutthaya	swamp	P	2005	N14.48885 E100.39212
7	WiCP2		Prachin Buri	deep swamp, *in situ* conservation area	P	2005	N14.08506 E101.17453

*Life-history trait type classification; P = perennial type

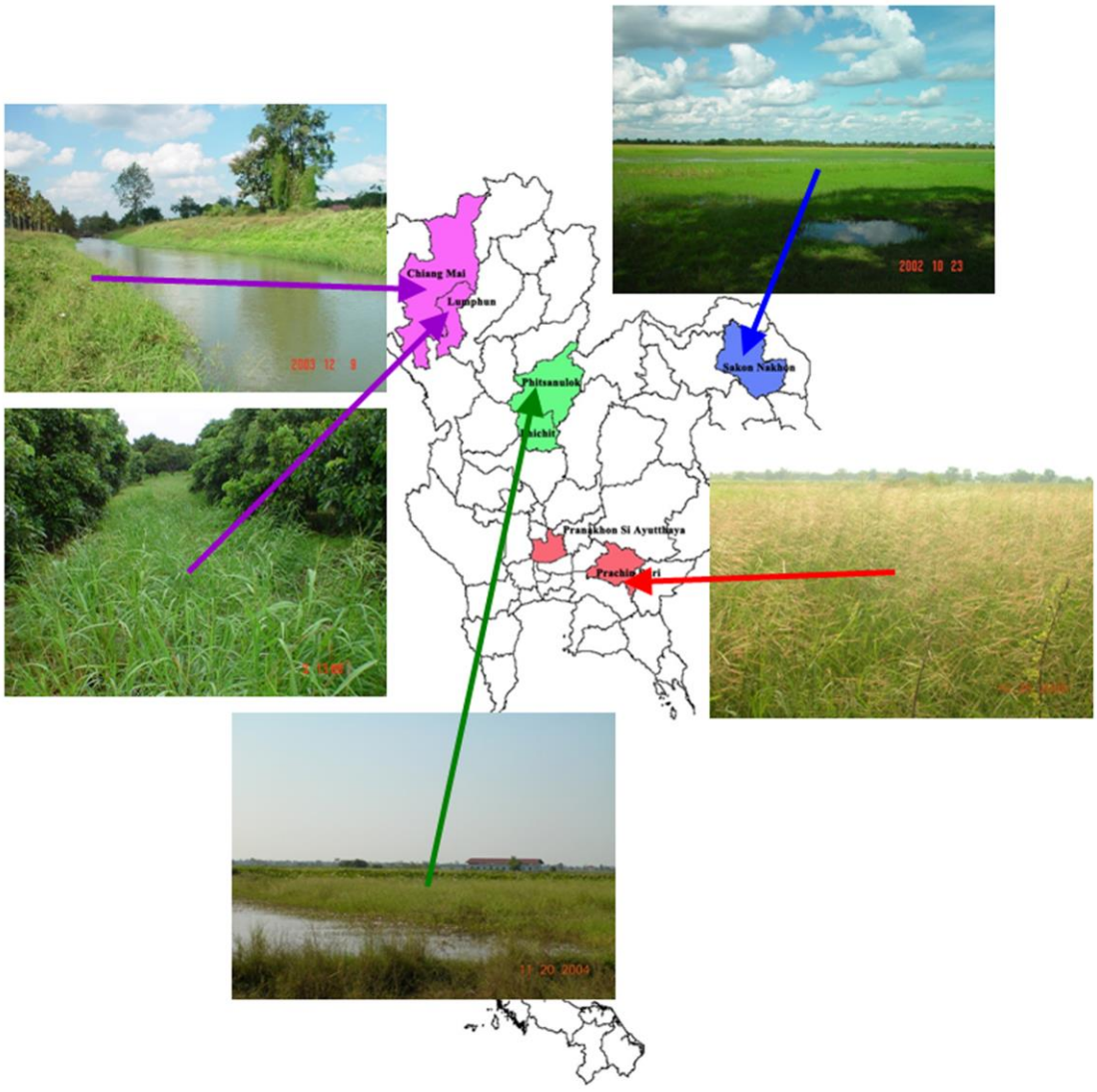


Figure 2.1.2 Location of common wild rice sampling in Thailand of the present study

Table 2.1.3 Name, type, pedigree description and year released of cultivated rice in the present study

No.	Variety	Abbreviation	Thai name	Type*	Type**	Photoperiod sensitivity	Growing area/Source	Description	Year released
1	Chai Nat 1	CNT1	ชัยนาท 1	NG	MV	No	lower north, central and irrigation area of upper north and northeast	IR13146-158-1 / IR15314-43-2-3-3 // BKN6995-16-1-1-2	1993
2	Suphan Buri 1	SPR1	สุพรรณบุรี 1	NG	MV	No	lower north and central	IR25393-57-2-3 / RD23 // IR27316-96-3 -2-2 /// SPRLR77205-3-2-1-1 / SPRLR7 9134-51-2-2	1994
3	Pathum Thani 1	PTT1	ปทุมธานี 1	NG	MV	No	lower north, central and irrigation area of upper north and northeast	BKNA6-18-3-2 / PTT85061-86-3-2-1	2000
4	Phitsanulok 2	PSL2	พิษณุโลก 2	NG	MV	No	lower north and central	CNTLR81122-PSL-37-2-1 / SPRLR81 041-195-2-1 // IR56	2000
5	Khao Dawk Mali 105	KDML105	ขาวดอกมะลิ 105	NG	ITV	Yes	All regions	Pure line selection from traditional variety	1959
6	RD6	RD6	กข 6	G	ITV	Yes	upper north and northeast	Improved by mutation breeding from irradiated KDML105	1977
7	RD15	RD15	กข 15	NG	ITV	Yes	upper north and northeast	Improved by radiation induced mutation breeding from irradiated KDML105	1978

Source: Department of Agriculture

*NG = non-glutinous rice, G = Glutinous rice

**MV = modern variety, ITV = improved traditional variety

Characterization of populations in pots

Morphological and physiological characterization

The experiment was set up in pots. Seeds if each weedy rice population were grown in pot, 10 plants per pot, after year following the sample taking from fields. Ten to 20 plants of each weedy rice populations (Table 2.1.1) were grown and compared with common wild rice (Table 2.1.2) and cultivated rice (Table 2.1.3) populations. Most weedy rice and pure line cultivated rice varieties were planted from seed, but common wild rice populations were planted by vegetative propagation. Morphological and physiological characteristics were recorded individually for each plant using the method of IRRI-IBPGR (1980). At flowering, plants were recorded for color at different plant parts including, leaf blade, leaf sheath, auricle, ligule, internode, apiculus and stigma, plant and panicle type, ligule shape and awn on spikelet and days to flowering. Two panicles per plants were bagged. At maturity, number of tillers and panicles, culm length of each plant was measured. Each plant was harvested separately. Two bagged panicles from each plant were harvested and measured for seed fertility, seed shattering and scored for hull color and pericarp color.

DNA analysis

Dried leaves samples were kept and analyzed separately for each of the 10 plants per population of weedy rice populations collected from the surveys were used to extract DNA. Genomic DNA extraction was modified method from Doyle and Doyle (1987) and the PCR reactions were performed following the description of Panaud *et al*. (1996). Twelve microsatellite loci distributed on 12 chromosomes were

chosen (Pusadee, 2009) and explained in Table 2.1.4. Amplification of DNA was performed in 20 µl reaction consisted of 20-50 ng DNA, 0.25 mM of each dNTP, 2% formamide, 0.2 µM of each primers and 0.5 unit of Taq DNA polymerase in reaction buffer [10 mM of Tris-HCl pH 8.5, 50 mM KCl, 1.5 mM MgCl2, 0.1mM EDTA, 50%(v/v) glycerol]. The amplified polymorphism alleles are distinguishable with the electrophoresis in 10% Polyacrylamide Gel Electrophoresis (PAGE).

Table 2.1.4 Microsatellite markers in the present study

Locus	Chromosome number	Primer sequence (5'to 3')*	Tm (C)	No. of alleles	Expected size (bp.)	Refference
RM1	1	F 5'- GCGAAAACACAATGCAAAAA-3'	55	13	60-154	McCouch *et al.*, 1997
		R 5'- GCGTTGGTTGGACCTGAC-3'				
RM109	2	F 5'- GCCGCCGGAGAGGGAGAGAGAG-3'	55	11	86-104	Temnykh *et al.*, 2000
		R 5'- CCCCGACGGGATCTCCATCGTC-3'				
RM211	2	F 5'- CCGATCTCATCAACCAACTG-3'	55	17	135-163	McCouch *et al.*, 1997
		R 5'- CTTCACGAGGATCTCAAAGG-3'				
RM251	3	F 5'- GAATGGCAATGGCGCTAG-3'	55	14	94-154	McCouch *et al.*, 1997
		R 5'- ATGCGGTTCAAGATTCGATC-3'				
RM280	4	F 5'- ACACGATCCACTTTGCGC-3'	55	11	148-176	Cao *et al.*, 2006
		R 5'- TGTGTCTTGAGCAGCCAGG-3'				
RM133	6	F 5'- TTGGATTGTTTTGCTGGCTCGC-3'	55	12	229-237	Temnykh *et al.*, 2000
		R 5'- GGAACACGGGGTCGGAAGCGAC-3'				
RM234	7	F 5'- ACAGTATCCAAGGCCCTGG-3'	55	7	109-164	McCouch *et al.*, 2000
		R 5'- CACGTGAGACAAAGACGGAG-3'				
RM481	7	F 5'- TAGCTAGCCGATTGAATGGC-3'	55	6	132-170	McCouch *et al.*, 2000
		R 5'- CTCCACCTCCTATGTTGTTG-3'				
RM477	8	F 5'- TCTCGCGGTATAGTTTGTGC-3'	55	20	212-228	McCouch *et al.*, 2000
		R 5'- ACCACTACCAGCAGCCTCTG-3'				
RM316	9	F 5'- CTAGTTGGGCATACGATGGC-3'	55	18	158-250	Temnykh *et al.*, 2000
		R 5'- ACGCTTATATGTTACGTCAAC-3'				
RM206	11	F 5'- CCCATGCGTTTAACTATTCT-3'	55	18	118-190	McCouch *et al.*, 1997
		R 5'- CCCATGCGTTTAACTATTCT-3'				
RM247	12	F 5'- TAGTGCCGATCGATGTAACG-3'	55	18	120-172	McCouch *et al.*, 1997
		R 5'- CATATGGTTTTGACAAAGCG-3'				

* F-forward, R-reverse (www.gramene.org)

Data analysis

Morphological and physiological analysis

For morphological characters, Shannon-Weaver Index *H'* (Shannon and Weaver, 1949 cited by Coffey, 2002) was used to calculate diversity in morphological characters. To illustrating taxonomy relationships among rice groups, neighbor-joining method of phylogenetic tree was constructed using UPGMA dendrogram method on the basis of C.S. Chord (1967) genetic distance based on 14 morphological traits. Physiological characters were calculated for mean and standard deviation (sd). Means were compared the differences between weedy rice populations and check populations were determined by using F-test at a significance level of 95% and 99%.

Population genetic structure and dynamics in spatial and temporal terms of weedy rice populations in Thailand

Weedy rice populations collected from the surveys in four rice cultivation seasons including, wet 2005, 2008, 2009 and dry 2009/10, were used to examine the weedy rice population genetic structure and dynamics. The genetic variation, genetic structure and population structure were used to analyze by partitioned into the following components: (a) for spatial term analysis; within and between regions in each season of weedy rice and (b) for temporal term analysis; within and between regions among seasons of weedy rice.

Genetic diversity analysis

Genetic parameters were calculated base on microsatellite data, number of allele (A), number of allele per population per locus (A_T), observed heterozygosity

(H_O), average gene diversity (H_S), total gene diversity (H_T), degree of gene differentiation among and between populations (F_{ST}) and Wright's inbreeding coefficient (F_{IS}), were calculated using FSTAT version 2.9.3 (Goudet, 2001). In addition, outcrossing rate (*t*) of each population was estimated using the following equation; $t = (1-F_{IS})/(1+F_{IS})$ where *F* is Wright's inbreeding coefficient. Standard measures of genetic diversity were calculated for the estimate of unbiased Nei's (1973) gene diversity (h) using POPGENE version 1.32 (Yeh *et al*., 1999).

Genetic structure analysis

Genetic structure was computed by hierarchical analysis of molecular variance (AMOVA) performed in the software of GeneAlEx version 6.1 (Peakall and Smouse 2006 cited by Pusadee, 2009). In addition, AMOVA provided statistic analogous to Weir and Cockerham (1984) unbiased F_{ST} estimator, to partition genetic variation into components. Total genetic diversity variance was partitioned into the following components: within and between regions of weedy rice populations in each season. The significant of F-statistics was analyzed by permutation, with the probability of non-differentiation for 10,000 randomizations.

Population structure analysis

A Bayesian-clustering program utilizing a Markov Chain Monte Carlo (MCMC) approach, STRUCTURE version 2.2 (Pritchard *et al*., 2000 cited by Pusadee, 2009), was used to elucidated the structuring of genetic variation and identify the number of genetically distinct clusters or gene pool of rice populations. STRUCTURE identifies *K* clusters of individuals based on the multilocus genotypes

of all individuals in the study and differences in allele frequencies at tested loci (Pritchard *et al.,* 2000). STRUCTURE was run four independent times for each value of *K* ranging from 1 to 4 using a burn-in period of 100,000 generations and 100,000 Markov chain Monte Carlo replications and using a model allowing for admixture and correlated allele frequencies. Parameters were set to their default values, following the documentation of STRUCTURE (Pritchard and Wen, 2004). The probability of how the data best fit into each number of assumed clusters (*K*) was estimated by ln probability of the data (ln *L*), so that individuals were assigned to a cluster based on their multilocus genotypic profile. Each test yielded a log-likelihood value of the data (ln *L*), with the highest indicating which test was closest to the actual number of genetically distinct clusters (*K*). Individuals were assigned probabilistically to a population (called inferred populations) or to multiple populations if their genotype profile indicated admixture. In this study, an accession was assigned to a cluster if at least 95% of the given value (*Q*) was estimated to belong to that cluster (*K*). In addition, to trace the origin of widespread weedy rice in various regions proportion of admixture ancestral genotype, different common wild rice genotypes and different cultivated rice genotypes, of each weedy rice population was observed.

Principal coordinate analysis (PCA) was conducted, on the basis of genetic similarity using EIGEN procedure in GeneAlEx 6.1 (Peakall and Smouse, 2006 cited by Pusadee, 2009) to observed the distribution of the rice populations. The PCA is a method to reduce original total variance among the individuals and to clarify the relationship between two or more characters into limited number of uncorrelated new variables (Wiley, 1981). The PCA method contributes to distinguish the differences

among the individuals and identify possible groups or clusters (Mohammadi and Prasanna, 2003).

To illustrating genetic relationships among rice populations, Neighbour-joining (NJ) tree was constructed using MEGA version 4 (Tamura *et al.*, 2007 cited by Pusadee, 2009) based on C.S. chord genetic distance (Cavalli-Sforza and Edwards, 1967) obtained by POWERMARKER version 3.0 (Liu and Muse, 2005) on the basis of 12 microsatellite loci.

2.3 Results

Characterization of weedy rice populations in rice fields

Weedy rice forms

Three types of weedy rice, Khao Harng (weedy rice with awn), Khao Deed (weedy rice without awn) and Khao Lai (rice with dark-brown hull and red pericarb) as described by Maneechote *et al.* (2004) were found in the survey (Table 2.2.1 and Figure 2.2.1). Khao Harng and Khao Deed were found in all regions in four growing seasons in 2005, 2008 and 2009 while Khao Lai found only in the Northeast (NE) in dry 2009/10. In the Lower North (LN) and the Central Plain (CP), proportion of Khao Deed increased with years of survey. In 2009, about 50-100% of weedy rice samples were Khao deed. In contrast, in 2005 both Khao Harng and Khao Deed were found at similar proportion (Table 2.2.1 and Figure 2.2.1).

Plant height

Weedy rice populations were classified for plant height into two types, the same as and taller than crop rice in the field. Only one population from the NE in wet 2009 was shorter than crop rice (Table 2.2.1). In 2005, most weedy rice were taller than crop but at 2008 and 2009 survey, most weedy rice populations from in the LN and the CP (50-86%) were at the same height as crop rice. However, in wet 2009 most (67%) weedy rice samples from the NE were taller than crop (Table 2.2.1).

Awn characteristics

Weedy rice populations were classified for awn characteristics into two characters, awn length and awn color. In 2005, most weedy rice had seeds with long

awn (>5 cm) (50-67%) but at 2008 and 2009 survey, most weedy rice populations from all regions (40-100%) were awnless. However, in dry 2009/10 some (14-33%) weedy rice sample from the LN and the NE were seeds with short awn (<5 cm) (Table 2.2.1).

Weedy rice populations with awn on seeds were presented two colors, red and white. In 2005, most weedy rice populations in all regions had red awn but in 2008 and 2009 survey, most weedy rice populations were mixture between red and white awns (Table 2.2.1).

Hull and pericarp color

Three hull colors, dark brown, straw and black were found in weed rice populations. Straw hull weedy rice seeds were found in all regions in four growing seasons in 2005, 2008 and 2009 while black hull weedy rice seeds were found only in wet 2005 and 2008 in all regions. However, weedy rice seeds with dark brown hull were found in some populations (17-33%) only in the NE (Table 2.2.1).

Most weedy rice populations had red pericarp in all regions in four growing seasons in 2005, 2008 and 2009 while in dry 2009/10 white pericarp were found in some (29-50%) populations from the LN and the CP (Table 2.2.1).

Seed shattering

Heavy seed shattering at maturity were found in most populations from all regions in four growing seasons in 2005, 2008 and 2009. The rest of populations were moderate shattering of weedy rice seeds (Table 2.2.1).

Table 2.2.1 Frequency (%) of type, plant height and seed characters of weedy rice populations from surveyed data from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d).

Character	Lower North				Northeast				Central Plain			
	05-w	08-w	09-w	09/10-d	05-w	08-w	09-w	09/10-d	05-w	08-w	09-w	09/10-d
n	3	5	6	7	3	10	10	6	2	5	2	4
Type[1]												
Khao-lai	0	0	0	0	0	0	0	16.7	0	0	0	0
Khao-harng	66.7	40.0	16.7	14.3	66.7	40.0	30.0	16.7	50.0	20.0	50.0	0
Khao-deed	33.3	60.0	83.3	85.7	33.3	60.0	70.0	66.7	50.0	80.0	50.0	100
Plant height												
Shorter than crop rice	0	0	0	0	0	0	10.0	0	0	0	0	0
As tall as crop rice	33.3	60.0	83.3	85.7	33.3	60.0	30.0	66.7	50.0	80.0	50.0	75.0
Taller than crop rice	66.7	40.0	16.7	14.3	66.7	40.0	60.0	33.3	50.0	20.0	50.0	25.0
Awn length												
None	33.3	40.0	83.3	85.7	33.3	40.0	70.0	66.7	50.0	80.0	50.0	100
<5 cm	0	40.0	16.7	14.3	0	40.0	30.0	33.3	0	20.0	50.0	0
>5 cm	66.7	20.0	0	0	66.7	20.0	0	0	50.0	0	0	0
Awn color												
Red	66.7	33.3	0	0	100	50.0	33.3	33.3	50.0	20.0	0	-
White and red	33.3	66.7	100	100	0	50.0	66.7	66.7	50.0	30.0	100	-
Seed shattering[2]												
Heavy	66.7	80.0	83.3	100	66.7	80.0	88.9	83.3	50.0	100	100	100
Moderate	33.3	20.0	16.7	0	33.3	20.0	11.1	16.7	50.0	0	0	0
Hull color												
Dark brown	0	0	0	0	33.3	0	0	16.7	0	0	0	0
Straw	66.7	80.0	100	100	33.3	80.0	100	83.3	50.0	60.0	100	100
Black	33.3	20.0	0	0	33.3	20.0	0	0	50.0	40.0	0	0
Pericarp color												
Red	100	100	100	71.4	100	100	100	100	100	100	100	50.0
White	0	0	0	28.6	0	0	0	0	0	0	0	50.0

[1]"Khao-lai" means one type of weedy rice with dark brown hull and length awn at the tip of the spikelet,

"Khao-harng" means one type of weedy rice with long awn at the tip of the spikelet,

"Khao-deed" means one type of weedy rice with jumping of seeds when their mature.

[2]Heavy seed shatter – most of seeds on spikelet were shattered when they were swing

Moderate seed shatter – around 50% of all seeds on spikelet were shattered when they were swing

(a) “Khao-harng” means one type of weedy rice with long awn .

(b) “Khao-deed” means one type of weedy rice with jumping of seeds when their mature.

(c) “Khao-lai” means one type of weedy rice with dark brown hull and length awn at the tip of the spikelet.

Figure 2.2.1 Three types of weedy rice, Khao-harng (a), Khao-deed (b) and Khao-lai (c).

Morphological and physiological characterization of weed rice in pot

Morphological characterization

Characteristics of stem, leaf, panicle, spikelets and seeds of weedy rice populations were recorded and compared with cultivated rice and common wild rice (Table 2.2.2, Figure 2.2.2 and Figure 2.2.3). For cultivated rice checks, no variation was found for all characters while large variation in leaf blade, leaf sheath, auricle, ligule and stigma color, ligule shape and anther size were found in common wild rice. For weedy rice, no variation was found in five morphological characters of weedy rice populations in all regions in four growing seasons. All plant had green leaf blade, 2 cleft of ligule, green inter-node, open panicle and anther size of about 1/2 of seed as observed as the same cultivated rice. Nine characters were varied in most populations collected in 2005 and 2008 including, erect and spreading plant type, green and green with purple at margin leaf sheath, colorless and light purple auricle and ligule, colorless and red apiculus and straw and dark gray hull. Exception in awnless to long awn, colorless and purple stigma and red and white pericarp were varied in populations collected in all four growing seasons in 2005, 2008 and 2009 (Table 2.2.2 and Figure 2.2.2).

All morphological characteristics were subjected to cluster analysis using UPGMA dendrogram method (Figure 2.2.3). In all seasons, common wild rice populations were classified as a distinct group from cultivated rice and weedy rice at the distance parameter around 0.35. Morphological relationship between weedy rice and crop rice were increased with year of infestation, with the distance of 0.2 in 2005 and decreased to 0.18 and 0.15 in 2008 and 2009, respectively. Within the weedy rice populations, in 2005 populations were separated into three regions. In 2008

populations from the LN and the CP were classified in the same group. In 2009, there was no relationship between locations of collection and morphological distance (Figure 2.2.3)

Table 2.2.2 Morphological characteristics and Shannon-Weaver Index (H') of weedy rice (We) populations from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were characterized in pot compared with 7 pure line rice varieties and 7 common wild rice (Wi) populations.

No.	Type	Abbreviation/ Accession No.	N	n	Stem and leaf		
					Plant type	Leaf blade color	Leaf sheath color
1	Cultivated rice	CNT1	1	10	erect	green	green
2	Cultivated rice	SPR1	1	10	erect	green	green
3	Cultivated rice	PTT1	1	10	erect	green	green
4	Cultivated rice	PSL2	1	10	erect	green	green
5	Cultivated rice	KDML105	1	10	erect	green	green
6	Cultivated rice	RD6	1	10	erect	green	green
7	Cultivated rice	RD15	1	10	erect	green	green
8	Weedy rice	WeLN05-w	3	30	erect, spreading (H'=0.4792)	green	green
9	Weedy rice	WeNE05-w	3	30	erect, spreading (H'=0.1271)	green	green, green with purple at margin (H'=0.3250)
10	Weedy rice	WeCP05-w	2	20	erect	green	green
11	Weedy rice	WeLN08-w	5	50	erect	green	green
12	Weedy rice	WeNE08-w	10	100	erect	green	green
13	Weedy rice	WeCP08-w	5	50	erect	green	green
14	Weedy rice	WeLN09-w	6	60	erect	green	green
15	Weedy rice	WeNE09-w	10	100	erect	green	green
16	Weedy rice	WeCP09-w	2	20	erect	green	green
17	Weedy rice	WeLN09/10-d	7	70	erect	green	green
18	Weedy rice	WeNE09/10-d	6	60	erect	green	green
19	Weedy rice	WeCP09/10-d	4	40	erect	green	green
20	Common wild rice	WiUN	2	20	prostrate	green, green with purple at margin (H'=0.4505)	green, green with purple at margin (H'=0.4505)
21	Common wild rice	WiLN	2	20	prostrate	green with purple at margin	green
22	Common wild rice	WiNE	1	10	prostrate	green with purple at margin	green with purple at margin
23	Common wild rice	WiCP	2	20	prostrate	green with purple at margin	green with purple at margin

Table 2.2.2 *(continued)*

No.	Abbreviation/ Accession No.	Stem and leaf			
		Auricle color	Ligule shape	Ligule color	Internode color
1	CNT1	colorless	2 cleft	colorless	green
2	SPR1	colorless	2 cleft	colorless	green
3	PTT1	colorless	2 cleft	colorless	green
4	PSL2	colorless	2 cleft	colorless	green
5	KDML105	colorless	2 cleft	colorless	green
6	RD6	colorless	2 cleft	colorless	green
7	RD15	colorless	2 cleft	colorless	green
8	WeLN05-w	colorless	2 cleft	colorless	green
9	WeNE05-w	colorless, light purple (H'=0.325)	2 cleft	colorless, light purple (H'=0.3250)	green
10	WeCP05-w	colorless	2 cleft	colorless	green
11	WeLN08-w	colorless	2 cleft	colorless	green
12	WeNE08-w	colorless	2 cleft	colorless, light purple (H'=0.2453)	green
13	WeCP08-w	colorless	2 cleft	colorless	green
14	WeLN09-w	colorless	2 cleft	colorless	green
15	WeNE09-w	colorless	2 cleft	colorless	green
16	WeCP09-w	colorless	2 cleft	colorless	green
17	WeLN09/10-d	colorless	2 cleft	colorless	green
18	WeNE09/10-d	colorless	2 cleft	colorless	green
19	WeCP09/10-d	colorless	2 cleft	colorless	green
20	WiUN	colorless, light purple (H'=0.8486)	2, 3 cleft (H'=0.3767)	colorless, light purple (H'=0.3767)	green
21	WiLN	colorless	2 cleft	colorless	green
22	WiNE	colorless, light purple (H'=0.8486)	2, 3 cleft (H'=0.6365)	colorless, light purple (H'=0.3250)	green
23	WiCP	colorless	2 cleft	colorless	green

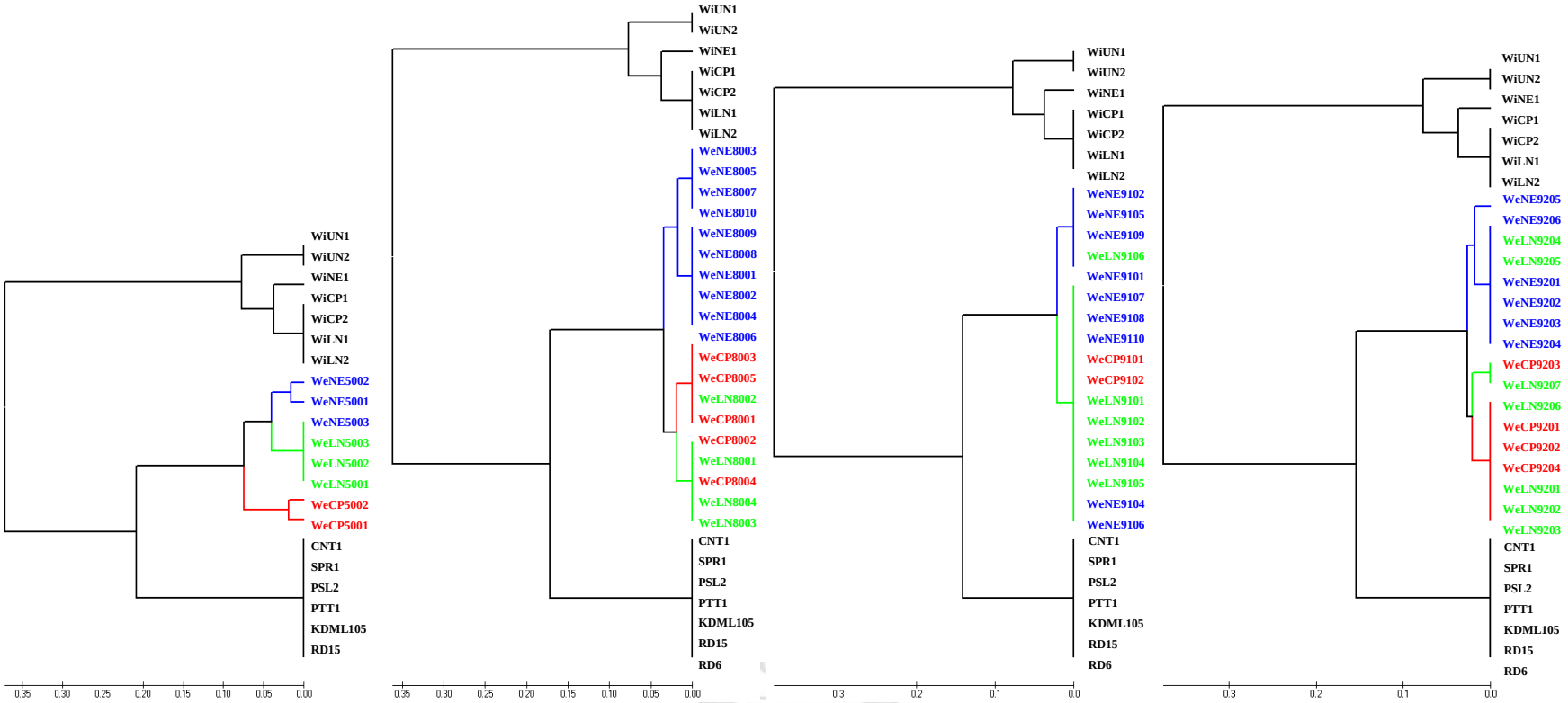
Table 2.2.2 *(continued)*

No.	Abbreviation/ Accession No.	Panicle and spikelet			
		Panicle type	Apiculus color	Spikelet awning	Stigma color
1	CNT1	compact	colorless	awnless	colorless
2	SPR1	compact	colorless	awnless	colorless
3	PTT1	compact	colorless	awnless	colorless
4	PSL2	compact	colorless	awnless	colorless
5	KDML105	compact	colorless	awnless	colorless
6	RD6	compact	colorless	awnless	colorless
7	RD15	compact	colorless	awnless	colorless
8	WeLN05-w	open	colorless, red (H'=0.6931)	long awn, awnless (H'=0.6734)	colorless, purple (H'=0.6931)
9	WeNE05-w	open	colorless, red (H'=0.6931)	long awn, awnless (H'=0.6734)	colorless, purple (H'=0.6931)
10	WeCP05-w	open	colorless, red (H'=0.6931)	long awn, awnless (H'=0.5072)	colorless, purple (H'=0.6931)
11	WeLN08-w	open	colorless, red (H'=0.6931)	long awn, awnless (H'=0.4327)	colorless, purple (H'=0.6931)
12	WeNE08-w	open	colorless, red (H'=0.5953)	long awn, awnless (H'=0.5953)	colorless, purple (H'=0.6931)
13	WeCP08-w	open	colorless	long awn, awnless (H'=0.3892)	colorless, red (H'=0.6931)
14	WeLN09-w	open	colorless	long awn, awnless (H'=0.1667)	colorless, purple (H'=0.3931)
15	WeNE09-w	open	colorless	long awn, awnless (H'=0.3029)	colorless, purple (H'=0.3931)
16	WeCP09-w	open	colorless	long awn, awnless (H'=0.1667)	colorless
17	WeLN09/10-d	open	colorless	long awn, awnless (H'=0.1428)	colorless
18	WeNE09/10-d	open	colorless	long awn, awnless (H'=0.1428)	colorless
19	WeCP09/10-d	open	colorless	awnless	colorless
20	WiUN	open	red	long awn	purple
21	WiLN	open	red	long awn	colorless, purple (H'=0.6931)
22	WiNE	open	red	long awn	purple
23	WiCP	open	red	long awn	purple

Table 2.2.2 *(continued)*

No.	Abbreviation/ Accession No.	Panicle and spikelet	Seed	
		Anther size	Hull color	Pericarp color
1	CNT1	1/2 seed	straw	white
2	SPR1	1/2 seed	straw	white
3	PTT1	1/2 seed	straw	white
4	PSL2	1/2 seed	straw	white
5	KDML105	1/2 seed	straw	white
6	RD6	1/2 seed	straw	white
7	RD15	1/2 seed	straw	white
8	WeLN05-w	1/2 seed	straw, dark gray (H'=0.6931)	red
9	WeNE05-w	1/2 seed	straw dark gray (H'=0.6931)	red
10	WeCP05-w	1/2 seed	straw dark gray (H'=0.3767)	red
11	WeLN08-w	1/2 seed	straw	red
12	WeNE08-w	1/2 seed	straw dark gray (H'=0.3767)	red
13	WeCP08-w	1/2 seed	straw	white, red (H'=0.2475)
14	WeLN09-w	1/2 seed	straw	white, red (H'=0.2475)
15	WeNE09-w	1/2 seed	straw	red
16	WeCP09-w	1/2 seed	straw	white, red (H'=0.3767)
17	WeLN09/10-d	1/2 seed	straw	white, red (H'=0.3767)
18	WeNE09/10-d	1/2 seed	straw	red
19	WeCP09/10-d	1/2 seed	straw	white, red (H'=0.5953)
20	WiUN	1/2, 2/3 seed (H'=0.3251)	gray, dark gray (H'=0.6365)	red
21	WiLN	1/2 seed	gray, dark gray (H'=0.6931)	red
22	WiNE	1/2, 2/3 seed (H'=0.3251)	dark gray	red
23	WiCP	1/2, 2/3 seed (H'=0.3768)	gray, dark gray (H'=0.6931)	red

Figure 2.2.2 Morphological characteristics of weedy rice populations. Weedy rice exhibited the intermediate between wild and crop traits.



***(a)* Wet 2005** ***(b)* Wet 2008** ***(c)* Wet 2009** ***(d)* Dry 2009/10**

Figure 2.2.3 Cluster analysis using UPGMA dendrogram method on the basis of C.S. Chord (1967) genetic distance among pure line rice varieties, weedy rice and common wild rice populations in Thailand among four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10 constructed based on 14 morphological traits.

Physiological characterization

Days to flowering

Overall mean of days to flowering for weedy rice populations from three rice production areas in four growing seasons was 69 days after sowing that earlier than both seven pure line rice varieties and common wild rice with 82 and 108 days, respectively (Table 2.2.3 and Figure 2.2.4). For among seasons, days to flowering were decreased with year of infestation from 71-73 days in 2005 to 66-67 days in dry 2009/10 but there were no difference between locations of collection and this character (Table 2.2.3). However, day to transplant in pot of wet 2005 populations was July 2008, wet 2008 populations was August 2009, wet 2009 and dry 2009/10 were August 2010.

Number of tillers plant^{-1}

Number of tillers plant^{-1} for weedy rice populations from three rice production areas in four growing seasons was 7 tillers that was the same as seven pure line rice varieties (6 tillers) but lower than common wild rice (14 tillers) (Table 2.2.3 and Figure 2.2.5). For among weedy rice populations, the number of tillers plant^{-1} were decreased with year of infestation from 7 in 2005 to 6 in 2009, except in dry 2009/10 collected from Northeast and Central Plain were 8 (Table 2.2.3).

Number of panicles plant^{-1}

Number of panicles plant^{-1} for weedy rice populations (10 panicles) from three rice production areas in four growing seasons was distributed in the range between seven pure line rice varieties (7 panicles) and common wild rice (14 panicles) (Table

2.2.3 and Figure 2.2.6). For among weedy rice populations, the number of panicles plant^{-1} was no difference between locations and year of collected in this character (Table 2.2.3).

Culm length (cm)

Overall mean of culm length of seven pure line rice varieties were 88 cm while common wild rice populations 138 cm (Table 2.2.3 and Figure 2.2.7). For weedy rice populations, culm length was distributed in the range between seven pure line rice varieties and common wild rice but there was decreased with year of infestation from 138-144 cm in 2005 to 96-104 cm in dry 2009/10 but there were no difference between locations of collection and this character (Table 2.2.3).

Panicle length (cm)

Panicle length for weedy rice populations from three rice production areas in four growing seasons was 23 cm that was the same as common wild rice (22 cm) but lower than seven pure line rice varieties (26 cm) (Table 2.2.3 and Figure 2.2.8). For weedy rice populations, panicle length was no difference between locations and year of collected in this character (Table 2.2.3).

Number of primary branches panicle^{-1}, spikelets panicle^{-1} and seeds panicle^{-1}

Number of primary branches panicle^{-1} for weedy rice populations from three rice production areas in four growing seasons was 8 branches that was the same as seven pure line rice varieties and common wild rice with 9 and 7 branches, respectively (Table 2.2.3 and Figure 2.2.9).

Number of spikelets panicle^{-1} for weedy rice populations from three rice production areas in four growing seasons was 125 spikelets that was the same as seven pure line rice varieties (133 spikelets) but higher than common wild rice (108 spikelets) (Table 2.2.3 and Figure 2.2.10).

As the same number of spikelets panicle^{-1}, number of seeds panicle^{-1} for weedy rice populations from three rice production areas in four growing seasons was 117 seeds that was the same as seven pure line rice varieties (120 seeds) but higher than common wild rice (93 seeds) (Table 2.2.3 and Figure 2.2.11). In weedy rice populations, number of primary branches panicle^{-1}, number of spikelets panicle^{-1} and number of seeds panicle^{-1} was no difference between locations and year of collected in this character (Table 2.2.3).

Seed fertility (%)

Overall mean of seed fertility rate of common wild rice populations were 64% while seven pure line rice varieties were 90% (Table 2.2.3 and Figure 2.2.12). For weedy rice populations, rate of seed fertility was distributed in the range between seven pure line rice varieties and common wild rice but there was increased with year of infestation from 69-72% in 2005 to 74-78% in dry 2009/10 but there were no difference between locations of collection and this character (Table 2.2.3).

Seed shattering (%)

Very low rate of seed shattering was recorded in seven pure line rice varieties (3%) while all seeds of common wild rice were shattered at maturity (100%). Overall mean of the rate of seed shattering for weedy rice populations from three rice

production areas in four growing seasons was 92% that was the same as common wild rice (Table 2.2.3 and Figure 2.2.13). However, the rate of seed among weedy rice populations was no difference between locations and year of collected in this character (Table 2.2.3).

Table 2.2.3 Physiological characteristics of weedy rice populations from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pot compared with 7 pure line rice varieties and 7 common wild rice populations (mean±sd).

Population	N	n	Days to Flowering[1]	Tillers plant-1	Panicle plant-1	Culm length (cm)	Panicle length (cm)
Pure line rice varieties	**7**	**70**	**82±10**	**6±2**	**7±3**	**88±17**	**26±3**
Common wild rice	**4**	**40**	**108±14**	**14±8**	**14±3**	**138±12**	**22±2**
Weedy rice	**63**	**630**	**69±7**	**7±2**	**10±3**	**111±16**	**23±3**
Lower North							
We2005-w	3	30	73±7A	7±3AB	10±4	138±14A	24±3
We2008-w	5	50	70±6AB	7±1AB	10±2	108±5C	23±3
We2009-w	6	60	67±2AB	6±1B	10±2	104±10CD	23±3
We2009/10-d	7	70	66±6B	6±2B	9±2	102±11CD	23±3
Northeast							
We2005-w	3	30	72±6A	7±2AB	10±4	144±8A	24±3
We2008-w	10	100	69±9AB	7±1AB	11±3	108±6C	22±2
We2009-w	10	100	67±7B	6±2B	9±2	107±5C	22±3
We2009/10-d	6	60	67±7B	8±1A	10±2	104±11	23±2
Central Plain							
We2005-w	2	20	71±6AB	7±3AB	10±4	143±9A	24±2
We2008-w	5	50	69±9AB	8±2A	11±3	107±22B	22±3
We2009-w	2	20	69±8AB	7±1AB	11±2	108±8C	22±4
We2009/10-d	4	40	67±8B	6±2B	11±4	96±11D	23±3
F-test			*	*	ns	***	ns
LSD (0.05)			**6.68**	**1.57**		**9.46**	

[1]Day to transplant in pot; Wet 2005 – July 2008, Wet 2008 – August 2009, Wet and dry 2009/10 - August 2010

Table 2.2.3 *(continued)*

Population	N	n	Number of primary branches panicle^{-1}	Number of spikelets panicle^{-1}	Number of seeds panicle^{-1}	Seed fertility (%)	Seed shattering (%)
Pure line rice varieties	**7**	**70**	**9±1**	**133±14**	**120±13**	**90±5**	**3±2**
Common wild rice	**4**	**40**	**7±4**	**108±44**	**93±42**	**64±19**	**100**
Weedy rice	**63**	**630**	**8±2**	**125±31**	**117±33**	**77±13**	**92±9**
Lower North							
We2005-w	3	30	10±2	125±14	116±13	72±3B	85±11
We2008-w	5	50	9±2	124±14	114±17	77±19A	95±7
We2009-w	6	60	9±2	122±14	106±27	78±7A	90±11
We2009/10-d	7	70	8±3	118±11	103±11	77±8A	91±9
Northeast							
We2005-w	3	30	10±1	125±14	123±31	72±6B	94±11
We2008-w	10	100	8±3	124±14	115±14	70±11C	92±9
We2009-w	10	100	8±3	121±13	105±13	77±7A	92±9
We2009/10-d	6	60	9±2	124±27	109±30	74±18B	96±6
Central Plain							
We2005-w	2	20	9±1	118±18	103±18	69±5C	93±11
We2008-w	5	50	8±2	113±24	100±29	73±12B	88±11
We2009-w	2	20	9±3	120±12	110±22	72±15B	91±7
We2009/10-d	4	40	9±2	119±23	105±12	78±13A	93±10
F-test			**ns**	**ns**	**ns**	*******	**ns**
LSD (0.05)						**6.76**	

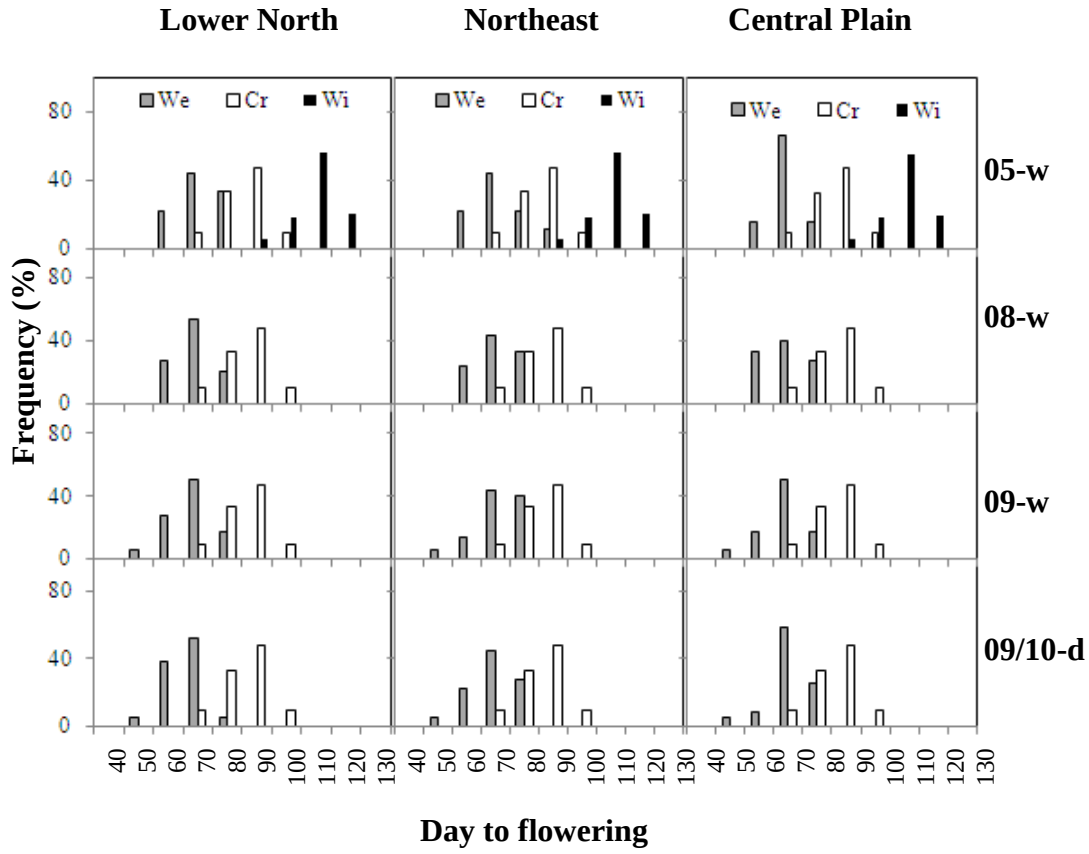


Figure 2.2.4 Distribution of days to flowering of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pot compared with seven pure line rice varieties and seven common wild rice populations.

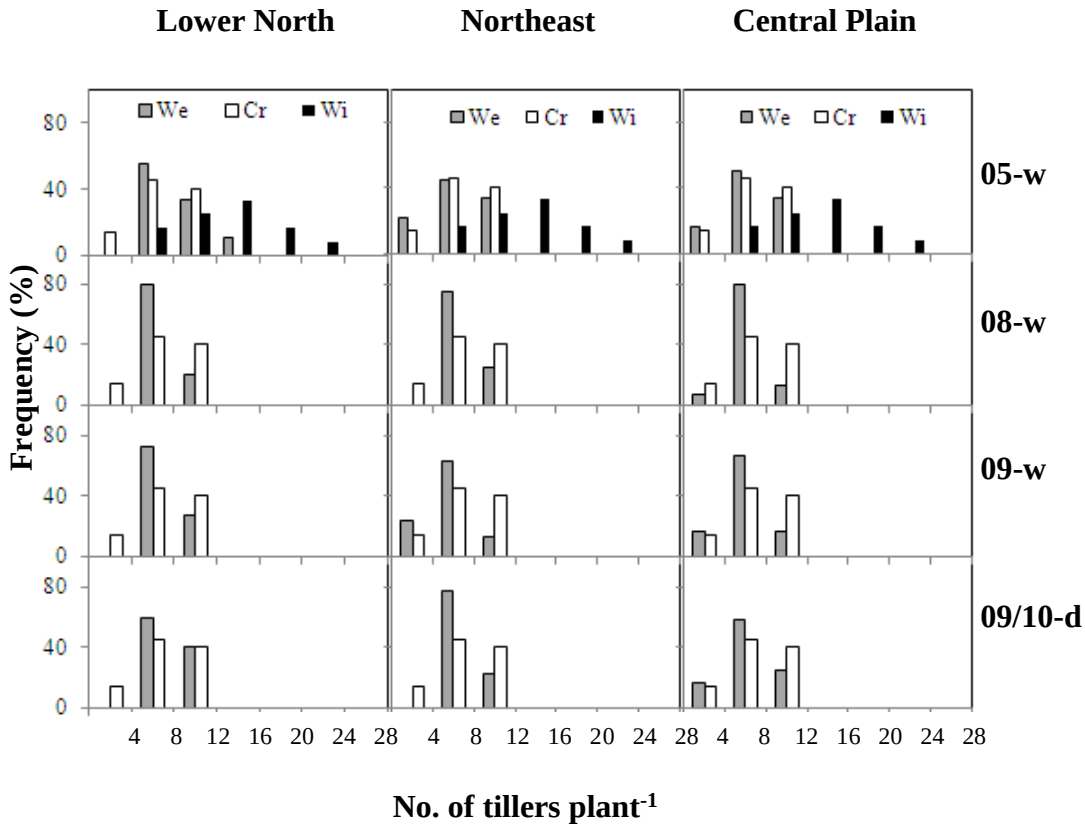


Figure 2.2.5 Distribution of tillers plant^{-1} of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pot compared with seven pure line rice varieties and seven common wild rice populations.

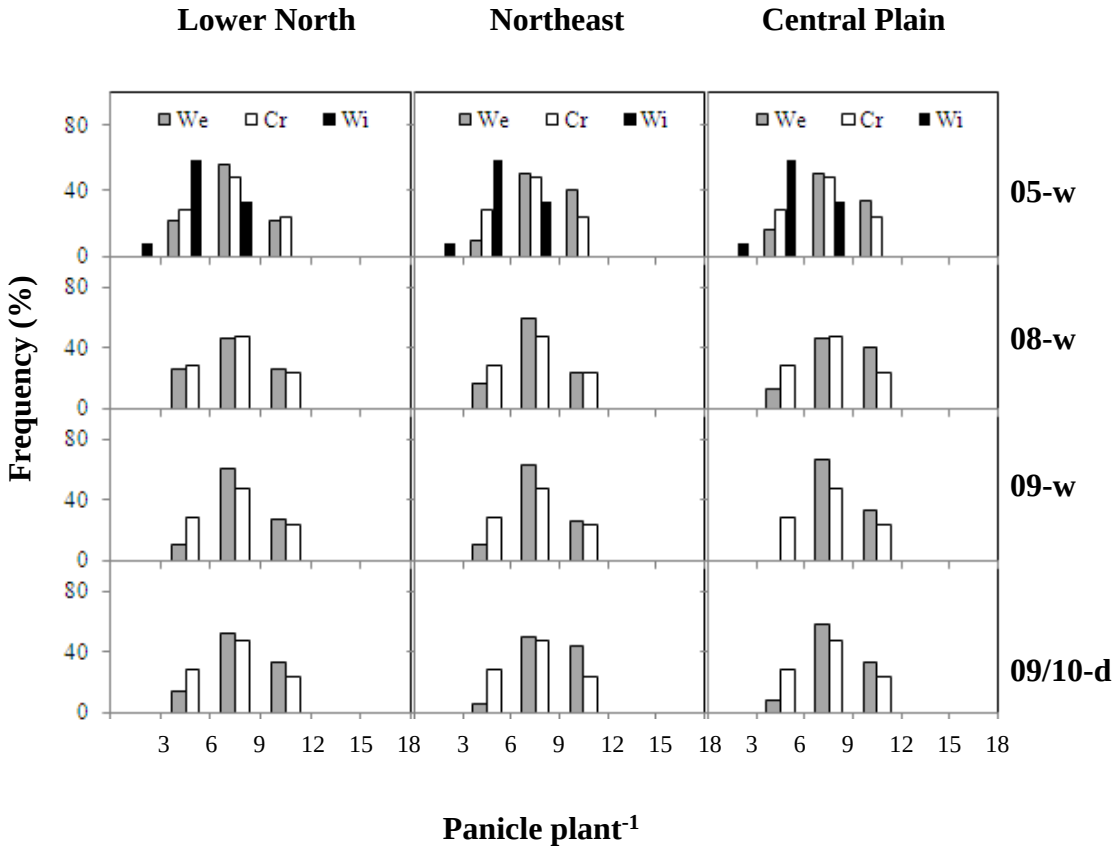


Figure 2.2.6 Distribution of panicle plant^{-1} of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

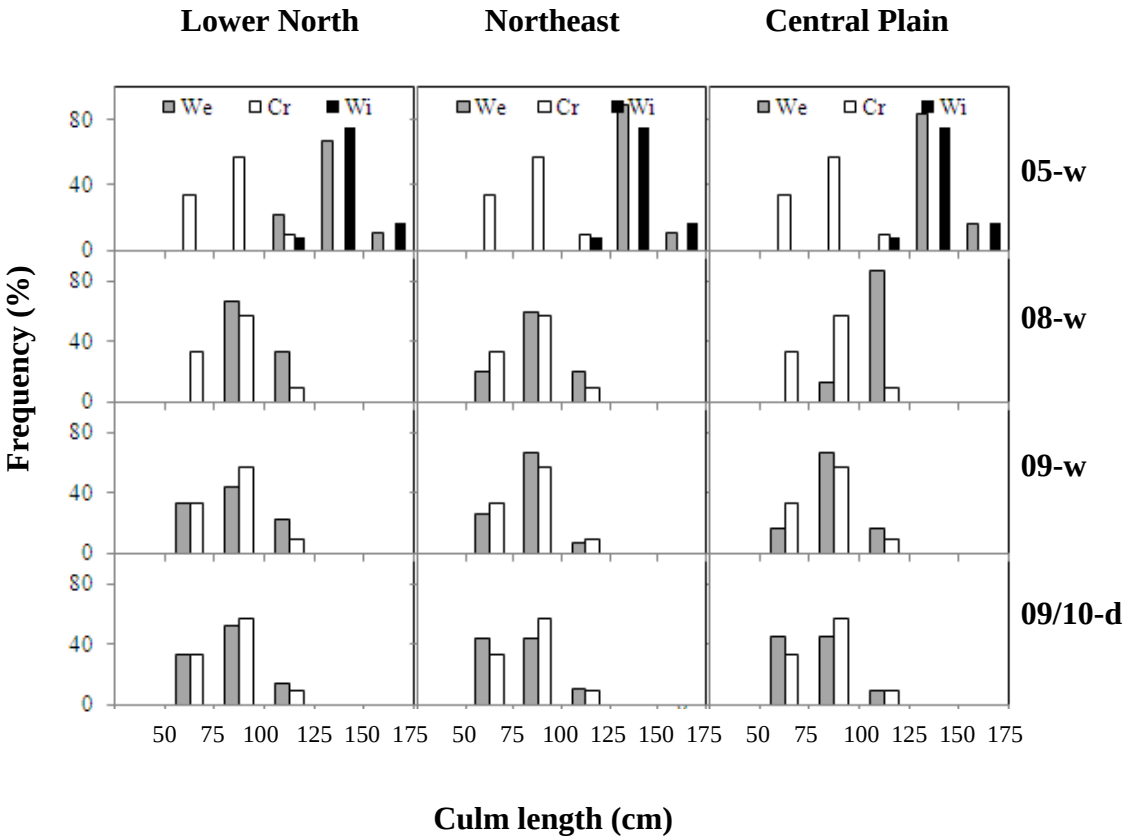


Figure 2.2.7 Distribution of culm length (cm) of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

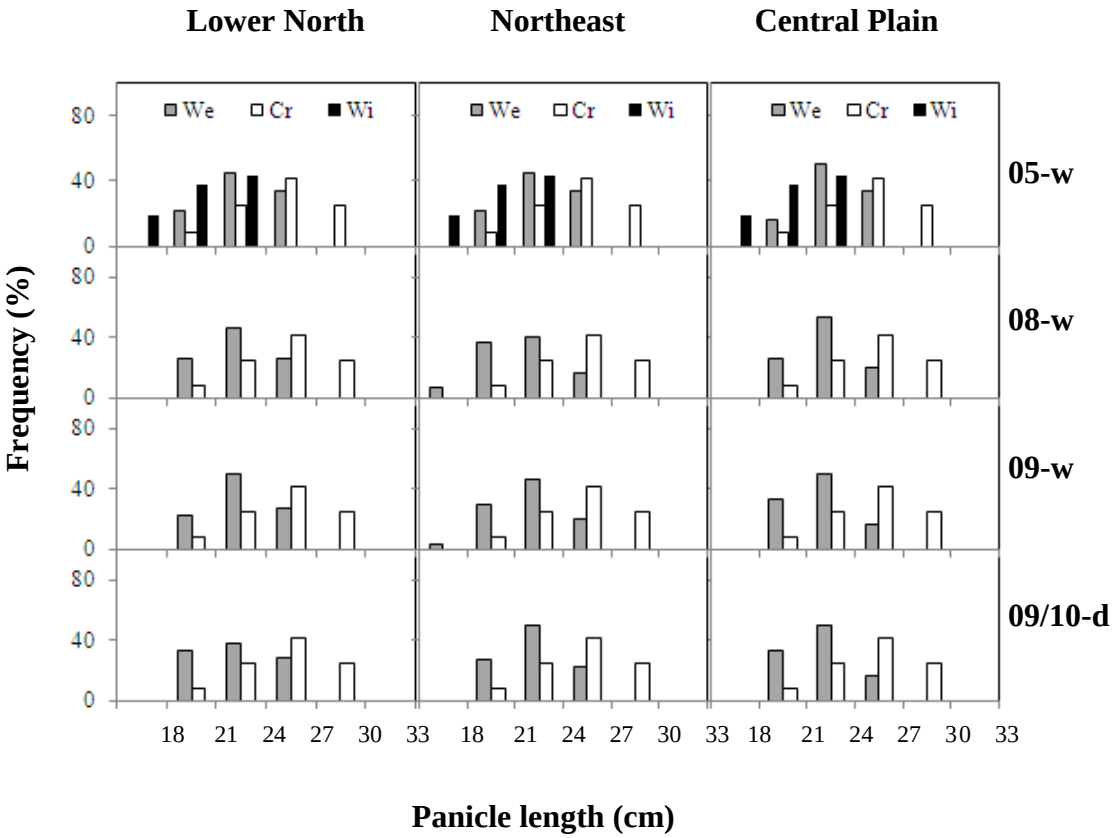


Figure 2.2.8 Distribution of panicle length (cm) of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

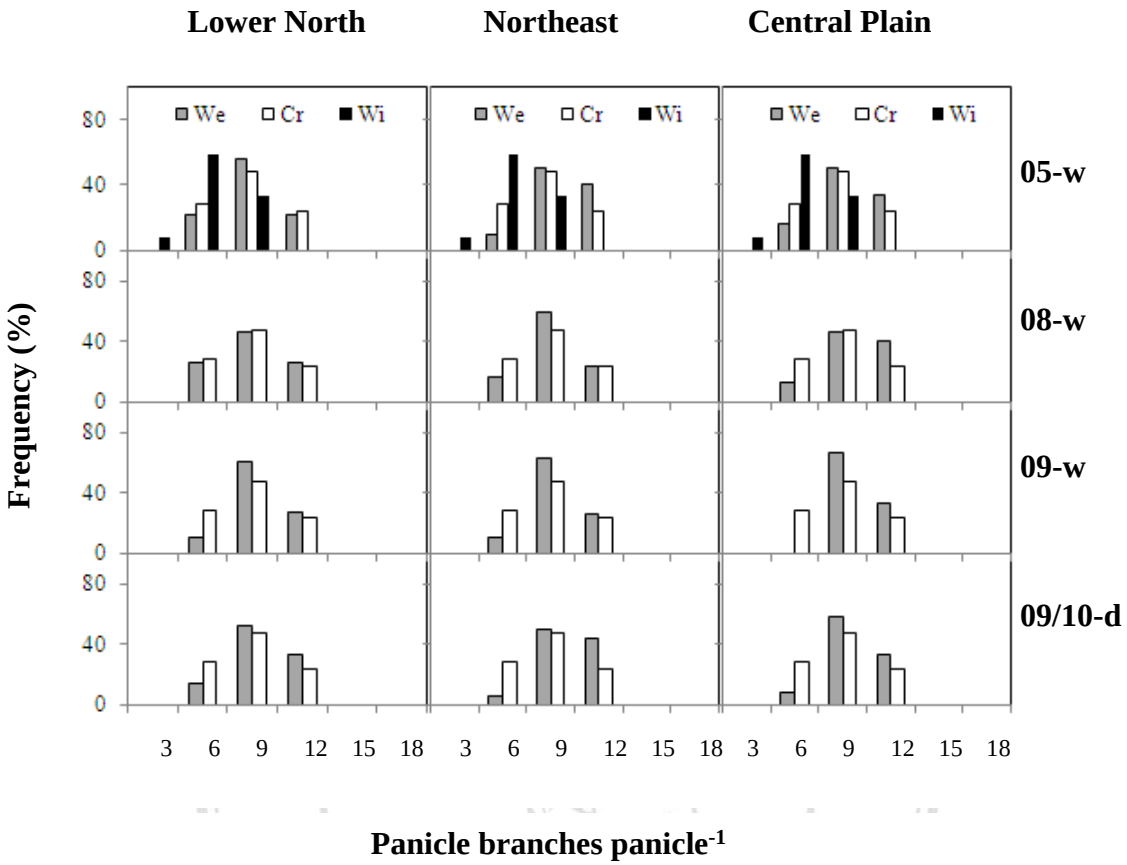


Figure 2.2.9 Distribution of number of primary branches panicle^{-1} of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

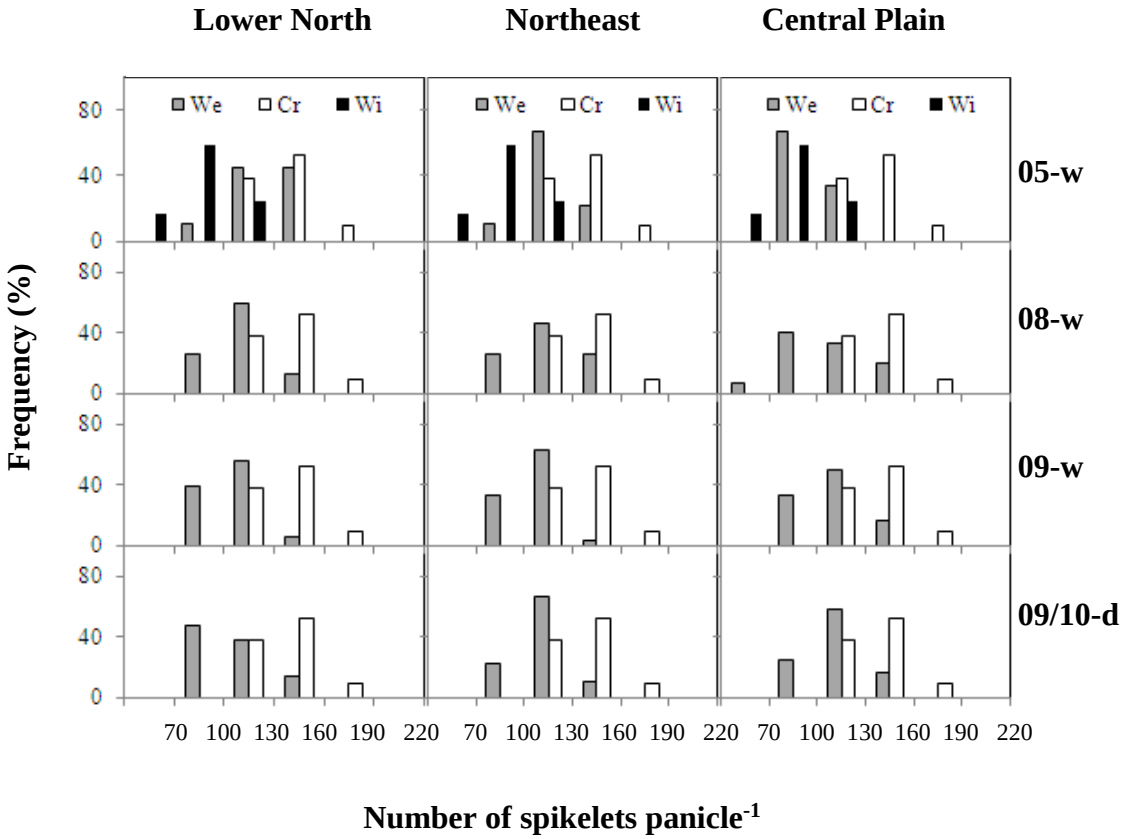


Figure 2.2.10 Distribution of number of spikelets panicle^{-1} of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

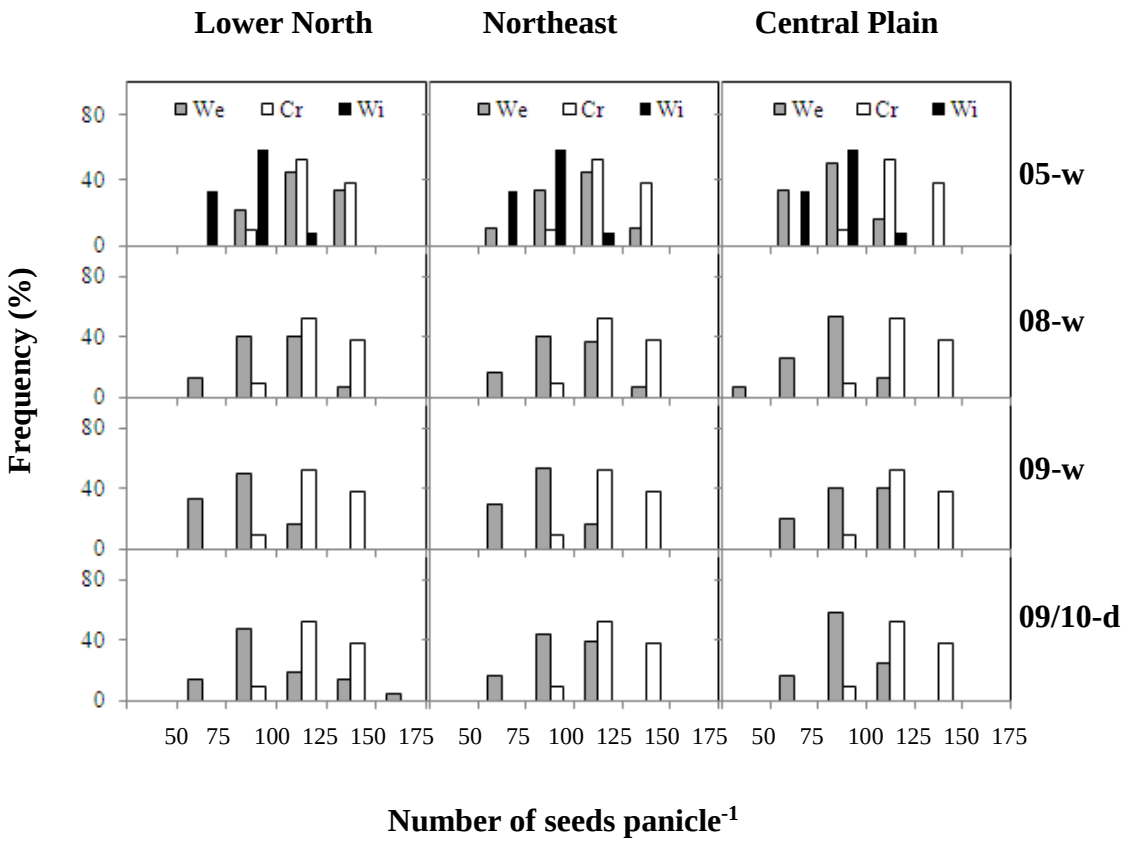


Figure 2.2.11 Distribution of number of seeds panicle^{-1}of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

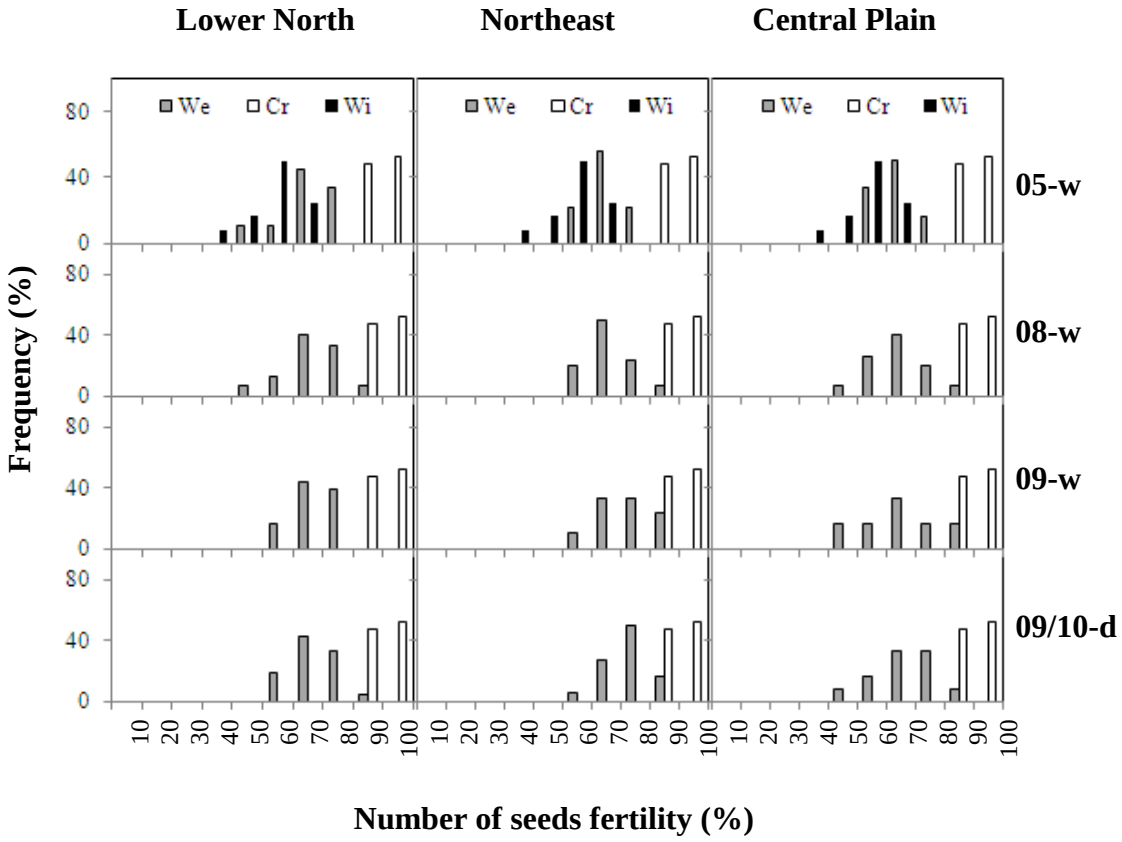


Figure 2.2.12 Distribution of seed fertility (%) of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

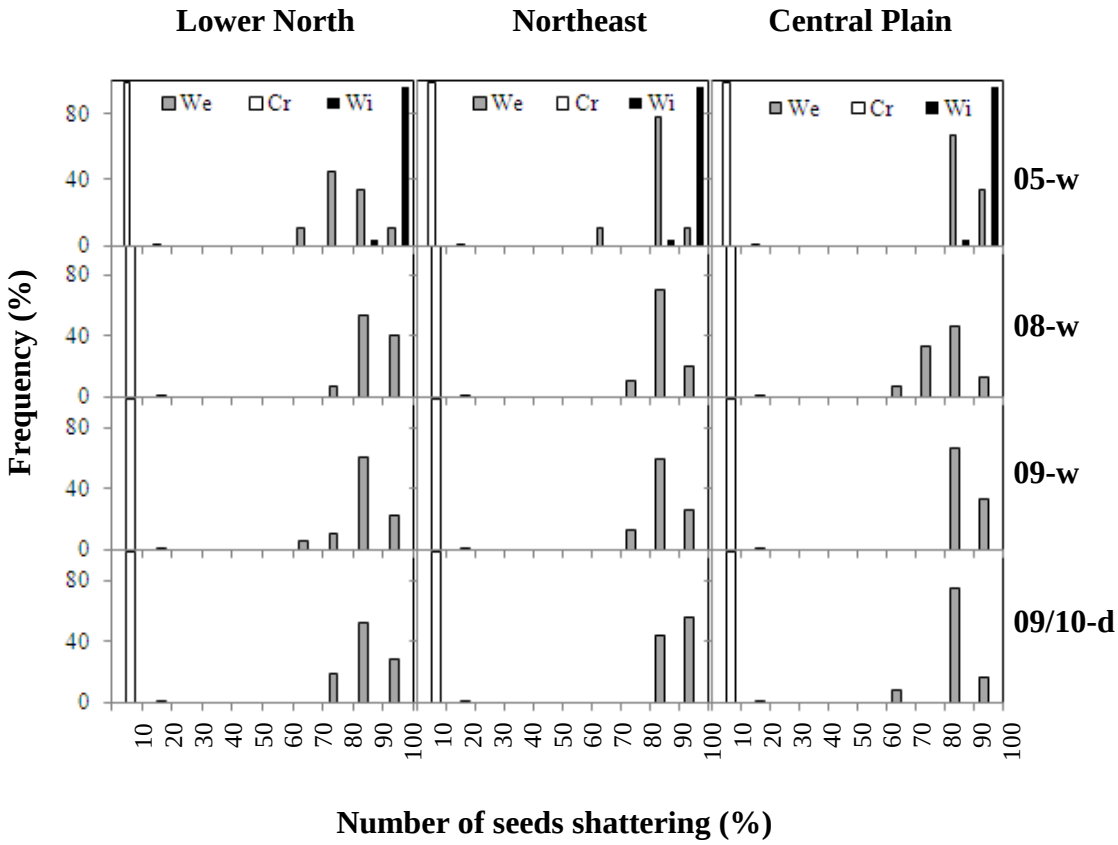


Figure 2.2.13 Distribution of seed shattering (%) of weedy rice populations from three rice production areas; Lower North, Northeast and Central Plain in Thailand among four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) were measured in pots compared with seven pure line rice varieties and seven common wild rice populations.

Population genetic structure and spatial dynamics of weedy rice populations in Thailand

Weedy rice populations from wet 2005

Allele diversity

Various levels of allele diversities of 8 weedy rice populations from wet season of 2005 were presented in Table 2.3.1. Total number of 100 alleles of weedy rice was observed with 12 SSR loci with the average of 8.3 alleles. Number of alleles per locus ranged from 7 alleles in RM234 and RM481 to 10 alleles in RM1 and RM211. For 12 loci, average number of allele per population was 3.6 alleles.

At individual population level, total number of alleles were ranging from 36 alleles in WeLN5001 (Phitsanulok) to 53 alleles in WeNE5001 (Surin) with 43 alleles in average (Table 2.3.1). Number of alleles per locus were ranging from 2 to 7 alleles per population with an average ranging from 3.0 allele per locus in WeLN5001 (Phitsanulok) to 4.4 alleles per locus in WeNE5001 (Surin).

Among the regions, the highest number of alleles was observed in the Northeast totally 66 alleles, followed with 59 alleles in the Central Plain while the Lower North was the lowest with 53 alleles. Average number of alleles per region was 49.7 alleles in the Northeast, 41.0 in the Central Plain and the lowest in the Lower North, 38.0 alleles. Average number of alleles per region per locus was the highest in the Northeast (5.5 alleles), followed in the Central Plain were 4.9 and the Lower North were 4.4 alleles, respectively (Table 2.3.1).

Table 2.3.1 Allele diversity of 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005 using 12 SSR markers

Population	Number of allele (A)												Total No. of allele	Average No. of allele
	RM1	RM109	RM211	RM251	RM280	RM133	RM234	RM481	RM477	RM316	RM206	RM247		
WeLN5001	2	4	2	2	3	3	2	2	5	2	5	4	36	3.0
WeLN5002	4	4	3	4	3	3	2	3	4	3	3	3	39	3.3
WeLN5003	5	4	4	4	2	3	3	2	4	2	3	3	39	3.3
Total LN	**5**	**4**	**5**	**6**	**5**	**4**	**3**	**3**	**5**	**4**	**5**	**4**	**53¶ 38.0‡**	**4.4§**
WeNE5001	5	4	5	5	3	4	3	3	6	5	5	5	53	4.4
WeNE5002	5	2	6	3	4	3	4	3	2	6	3	4	45	3.8
WeNE5003	7	5	7	2	4	5	3	4	2	4	5	3	51	4.3
Total NE	**7**	**5**	**8**	**5**	**4**	**5**	**4**	**5**	**6**	**6**	**6**	**5**	**66¶ 49.7‡**	**5.5§**
WeCP5001	2	4	4	5	4	4	2	2	5	3	4	3	42	3.5
WeCP5002	2	3	4	5	5	4	2	3	4	2	4	2	40	3.3
Total CP	**4**	**4**	**3**	**8**	**7**	**7**	**3**	**3**	**7**	**3**	**6**	**4**	**59¶ 41.0‡**	**4.9§**
Average	**4.0**	**3.8**	**4.4**	**3.8**	**3.5**	**3.6**	**2.6**	**2.8**	**4.0**	**3.4**	**4.0**	**3.4**	**43.1**	**3.6*§**
Range	**2-7**	**2-5**	**2-7**	**2-5**	**2-5**	**3-5**	**2-4**	**2-4**	**2-6**	**2-6**	**3-5**	**2-5**	**36-53**	
Total	**10**	**8**	**10**	**8**	**8**	**8**	**7**	**7**	**9**	**9**	**8**	**8**	**100**	**8.3§§**

¶ Number of alleles per region ‡Average number of alleles per region § Number of alleles per region §§Average number of alleles per locus *§Average number of alleles per population

Genetic diversity

Eight weedy rice populations exhibited high level of genetic diversity demonstrated by average gene diversity (H_S = 0.555) and total gene diversity (H_T = 0.827). The results of genetic differentiation (F_{ST}) illustrated that almost half (43%) of genetic variation of those found in weedy rice was the differences between 8 weedy rice populations while the rest 57% was distributed between 151 individuals within 8 populations. In mating system, moderate level of total inbreeding coefficient (F_{IS} = 0.572) was detected, lead to the moderate level of total outcrossing rate (t = 0.272) (Table 2.3.2).

For each population, eight weedy rice populations exhibited broad level of genetic diversity (Table 2.3.2). Population of WeNE5001 (Surin) showed the highest of the observed heterozygosity (H_O = 0.558) while WeLN5001 (Phitsanulok) had the lowest (0.183) with 0.292 in average. Similarly, the highest Nei's (1973) gene diversity (h) was also found in WeNE5001 (0.673) and the lowest was in WeLN5001 (0.475) with 0.575 in average. Weedy rice displayed various levels of inbreeding coefficient (F_{IS}). The highest level was found in WeLN5001 (Phitsanulok, 0.722) while the lowest level was WeNE5001 (Surin, 0.172). On the other hand, population of WeNE5001 (Surin) showed the highest level of outcrossing rate (71%) while WeLN5001 (Phitsanulok) showed the lowest (16%).

For each region (Table 2.3.3), weedy rice collected from three regions showed lower level of observed heterozygosity (H_O) than Nei's (1973) gene diversity (h). Weedy rice from the Northeast exhibited the highest genetic variation including observed heterozygosity (0.399), Nei's gene diversity (0.689) and average gene diversity (0.629) while the Lower North was the lowest (H_O = 0.227, h = 0.512 and

H_S = 0.499, respectively). Weedy rice from the Northeast exhibited highest total gene diversity (H_T = 0.766), following with the Central Plain (0.717) while the Lower North was lowest (0.697). Considering mating system, the highest level of inbreeding coefficient (F_{IS}) was found in the Lower North (0.597) following with the Central Plain (0.556) and the lowest was the Northeast (0.421). Conversely, the Northeast weedy rice exhibited the highest level of outcrossing rate (41%) following with the Central Plain (29%) and the lowest in the Lower North (24%).

Genetic structure

(a) Analysis of Molecular Variance (AMOVA)

Analysis of molecular variance (AMOVA) was used to examine the partitioning of total variation in weedy rice (Table 2.3.4). About 43% of total variance from total genetic diversity (0.827, Table 2.3.2) was distributed among 8 populations of weedy rice while the rest 57% was distributed between 151 individuals. Partitioning among regions showed that 20% of the total genetic variation was distributed among regions. For within each region, about 34%, 50% and 42% were the distribution of genetic variation among populations within the Lower North, the Northeast and the Central Plain, respectively (Table 2.3.4).

(b) Genetic differentiation (F_{ST})

In term of genetic differentiation (F_{ST}), weedy rice exhibited higher level of F_{ST} within region than between regions. Weedy rice populations of the Northeast (0.503) illustrated the highest level of genetic differentiation within populations while the Lower North (0.335) was the lowest. Considering pairwise genetic differentiation between regions, weedy rice of the Lower North to the Central Plain (0.102) were

more similar than those weed rice of the Lower North to the Northeast (0.183) and the Northeast to the Central Plain (0.205) (Table 2.3.5).

Table 2.3.2 Genetic parameters of 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005 using 12 SSR markers

Population	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
WeLN5001	18	0.475				0.183	0.722	0.162
WeLN5002	20	0.521				0.269	0.634	0.224
WeLN5003	20	0.501				0.229	0.518	0.318
WeNE5001	23	0.673				0.558	0.172	0.707
WeNE5002	18	0.580				0.218	0.625	0.231
WeNE5003	18	0.633				0.420	0.335	0.498
WeCP5001	15	0.504				0.240	0.484	0.348
WeCP5002	19	0.552				0.217	0.691	0.183
Total weedy rice in wet season of 2005 (8 populations)	**151**	**0.575**	**0.555**	**0.827**	**0.428**	**0.292**	**0.572**	**0.272**

Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Table 2.3.3 Summary of genetic parameters of weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005 based on 12 SSR loci

Accession	Region	N	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
LN	Lower North	3	58	0.512	0.499	0.697	0.335	0.227	0.597	0.235
NE	Northeast	3	59	0.689	0.629	0.766	0.503	0.399	0.421	0.407
CP	Central Plain	2	34	0.567	0.528	0.717	0.415	0.229	0.556	0.285
Among regions	**Among region**	**8**	**151**	**0.575**	**0.555**	**0.827**	**0.198**	**0.292**	**0.572**	**0.272**

Number of populations (N), Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Average gene diversity (H_S), Total gene diversity (H_T), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Table 2.3.4 Analysis of molecular variance (AMOVA) among populations and three rice production areas for 151 individuals of 8 weedy rice populations in wet 2005 based on 12 SSR loci

Source	df	SS	Variance component	% of the total variance
Among Populations	7	877.432	7.489	43%
Among Regions	2	307.871	1.094	20%
Populations/region	5	569.561	3.646	23%
Populations/Lower North	2	191.688	6.686	34%
Populations/Northeast	2	206.178	8.071	50%
Populations/Central Plain	1	160.434	5.305	42%
Individuals/Population	143	661.922	5.218	57%

Table 2.3.5 Pairwise of genetic differentiation (F_{ST}) within and between three rice production areas of 8 weedy rice populations in wet 2005 based on 12 SSR loci

Pairwise F_{ST}		Region	
		Within	**Between**
Lower North		0.335	
	Northeast		0.183
	Central Plain		0.102
Northeast		0.503	
	Central Plain		0.205
Central Plain		0.415	

Population structure

Eight weedy rice populations were structured into 2 inferred populations (K=2) on the basis of 12 microsatellite loci using STRUCTURE program (Figure 2.3.1). The admixtures of first inferred population consisted of weedy rice populations from the Lower North and the Central Plain which were collected from the fields with modern varieties particularly CNT1, SPR1, PTT1 and PSL2 (Figure 2.3.1 and Figure 2.3.2). Whereas the second inferred population consisted of weedy rice populations from the Northeast which were collected from the fields with improved traditional varieties particularly KDML105, RD15 and RD6 (Figure 2.3.1 and Figure 2.3.2).

Principal coordinate analysis (PCA) clustered the 8 weedy rice populations into 2 groups. The EIGEN analysis of the pairwise Chord genetic distance measures for among 8 weedy rice populations explained 58.01% of the variation presented in the two weedy rice groups within the first and second axes. The first group consisted of weedy rice populations collected from the Lower North and the Central Plain with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2 while weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6 were clustered in second group (Figure 2.3.3).

Similarly, NJ clustering method base on C.S. Chord genetic distance also divided 8 weedy rice populations into 2 major clusters (Figure 2.3.4). The first cluster consisted of weedy rice populations collected from the Lower North and the Central Plain with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2. The second cluster consisted of weedy rice populations collected from the

Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6.

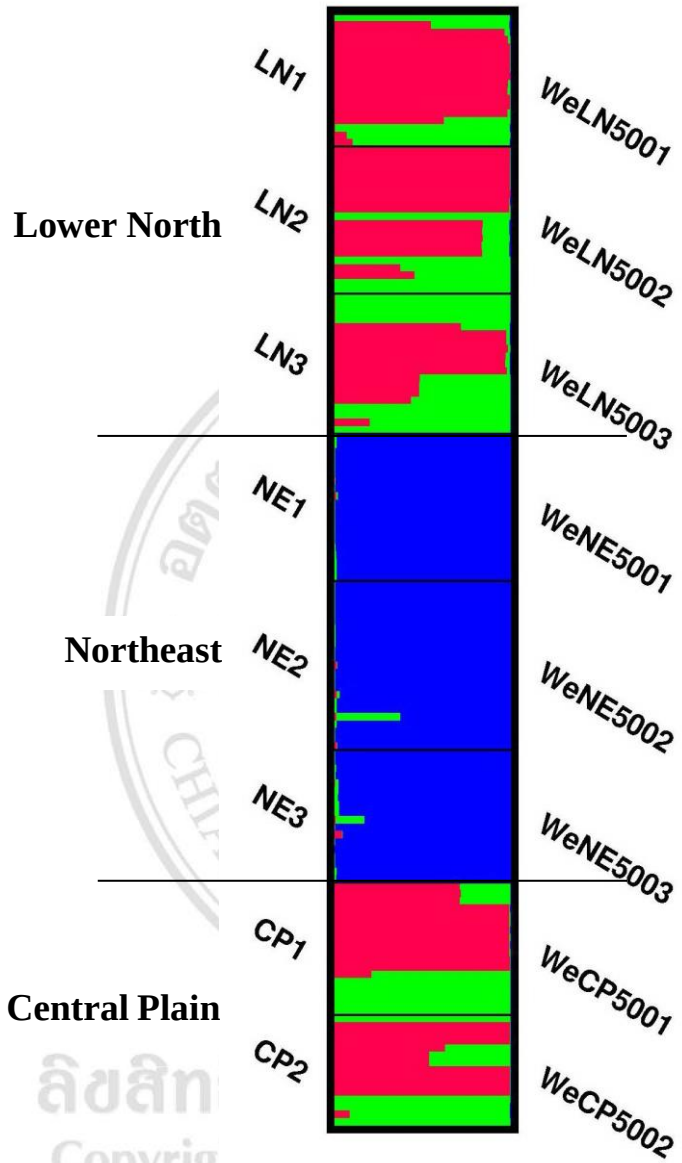


Figure 2.3.1 Population assignment of 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005 reveal K=2. Each bar represent each population consisted between 15 to 23 individuals. Different colors represent different inferred populations, referred to different K.

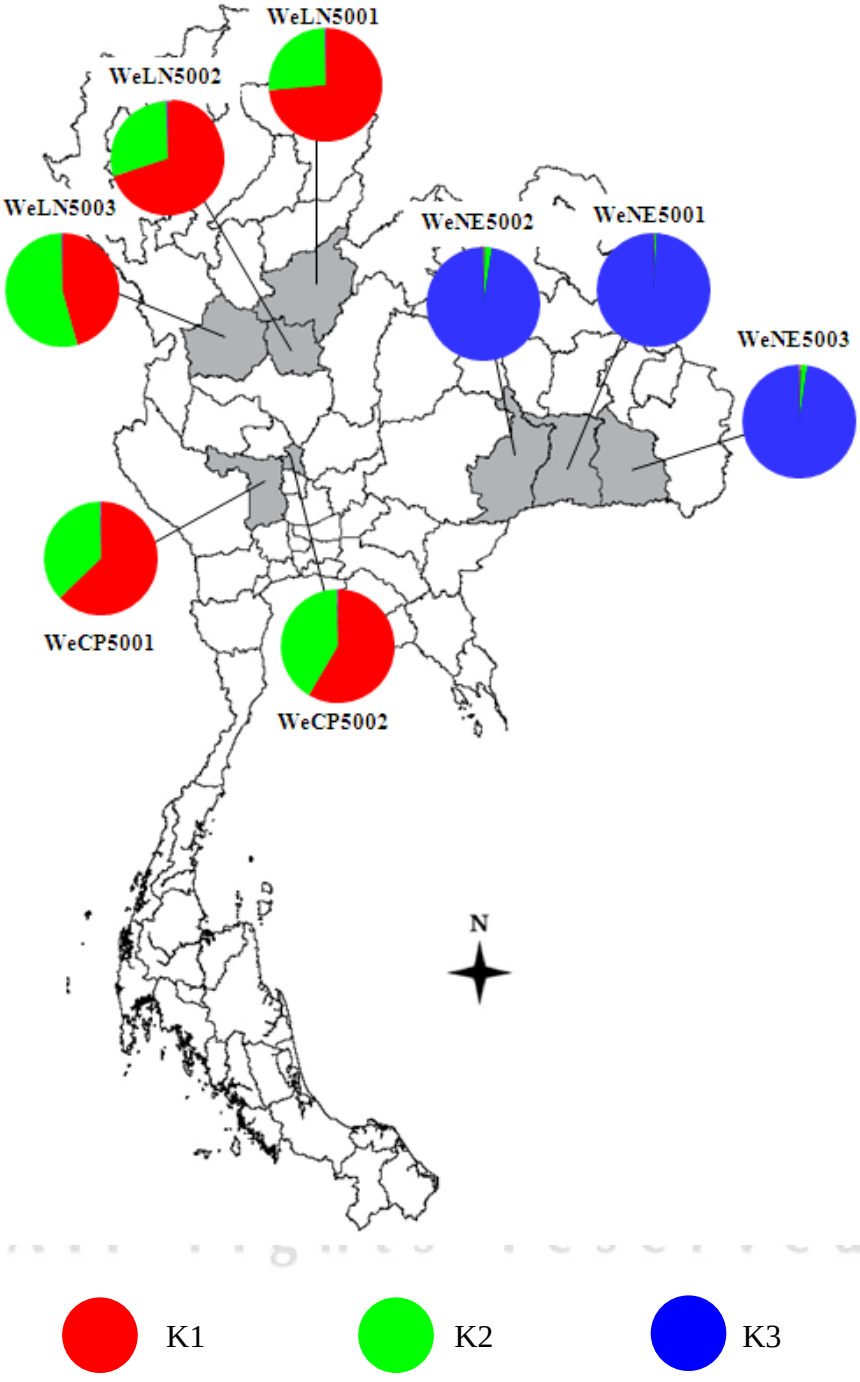


Figure 2.3.2 Genetic proportion of 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005 (K=2).

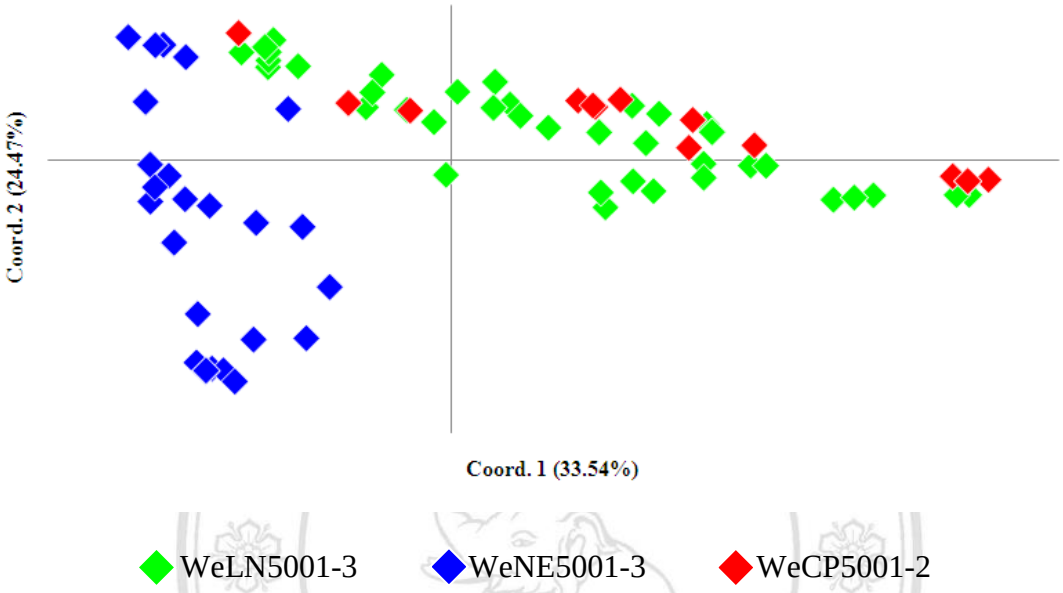


Figure 2.3.3 Distribution of 151 individuals of 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005. Different colors represent 2 clusters referred to the assignment obtained from STRUCTURE: Green-weedy rice collected from the Lower North fields, Blue-weedy rice populations collected from the Northeast fields and Red- weedy rice populations collected from the Central Plain fields, formed by the principal coordinate analysis (PCA) on the basis of 12 microsatellite markers.

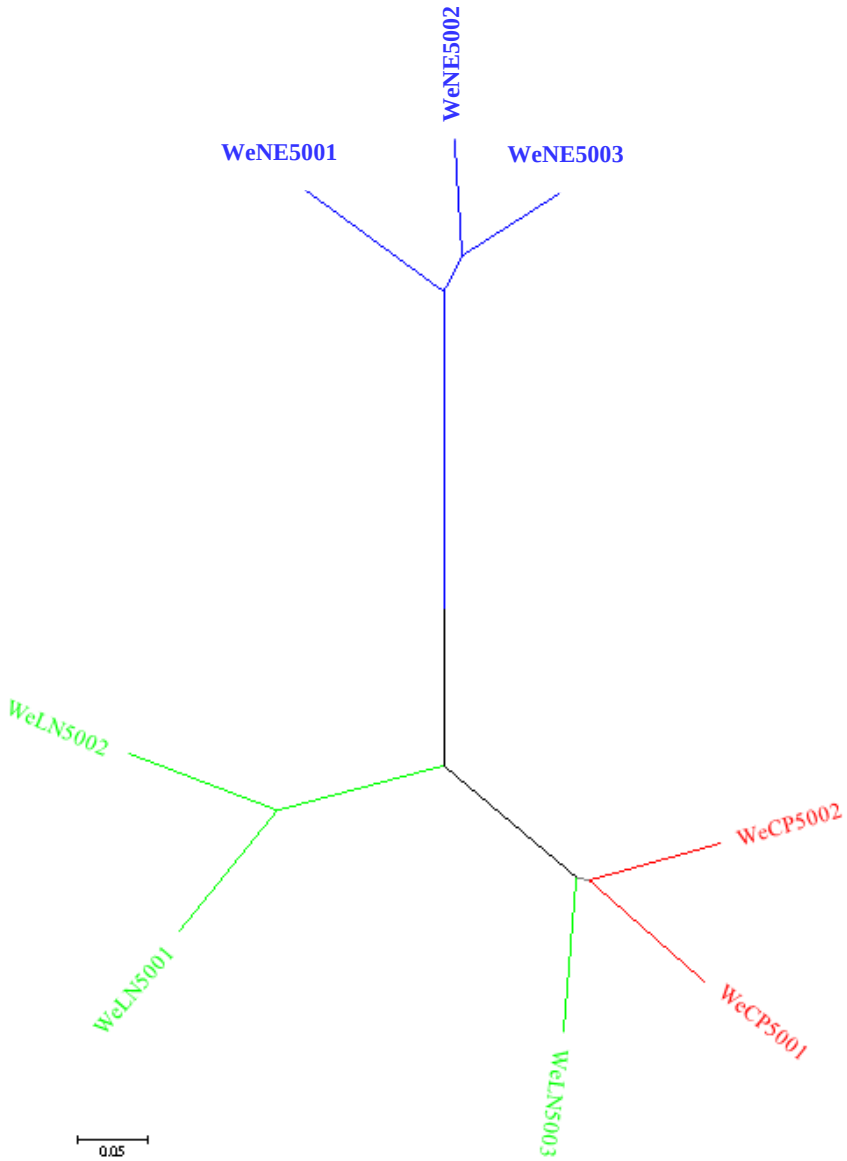


Figure 2.3.4 Cluster analysis using NJ method based on C.S. Chord (1967) genetic distance among 8 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2005

Weedy rice populations from wet 2008

Allele diversity

Various levels of allele diversities of 20 weedy rice populations from wet season of 2008 were presented in Table 2.3.6. Total number of 89 alleles of weedy rice was observed with 12 SSR loci with the average of 7.4 alleles. Number of alleles per locus ranged from 6 alleles in RM481 to 9 alleles in RM206 and RM247. For 12 loci, average number of allele per population was 3.1 alleles.

At individual population level, total number of alleles were ranging from 29 alleles in WeLN8005 (Phichit) to 49 alleles in WeNE8003 (Roi Et) with 37.5 alleles in average (Table 2.3.6). Number of alleles per locus were ranging from 1 to 6 alleles per population with an average ranging from 2.2 allele per locus in WeLN8005 (Phichit) to 4.1 alleles per locus in WeNE8003 (Roi Et).

Among the regions, the highest number of alleles was observed in the Northeast totally 64 alleles, followed with 51 alleles in the Central Plain while the Lower North was the lowest with 47 alleles. Average number of alleles per region was 41.0 alleles in the Northeast, 36.4 in the Central Plain and the lowest in the Lower North, 34.6 alleles. Average number of alleles per region per locus was the highest in the Northeast (5.3 alleles), followed in the Central Plain and the Lower North were 4.3 and 4.1 alleles, respectively.

Table 2.3.6 Allele diversity of 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008 using 12 SSR markers

Population	Number of allele (A)												Total No. of allele	Average No. of allele
	RM1	RM109	RM211	RM251	RM280	RM133	RM234	RM481	RM477	RM316	RM206	RM247		
WeLN8001	3	3	3	4	1	5	2	3	4	1	5	2	36	3.0
WeLN8002	3	4	5	3	3	3	3	3	4	3	4	3	41	3.4
WeLN8003	2	3	4	4	4	3	2	2	2	3	3	1	33	2.8
WeLN8004	2	2	2	1	2	3	4	2	4	5	2	1	31	2.5
WeLN8005	3	2	3	2	3	2	3	1	3	4	3	2	29	2.2
Total LN	**3**	**4**	**5**	**4**	**4**	**5**	**4**	**3**	**4**	**5**	**5**	**3**	**47**[¶] **34.6**[‡]	**4.1**[§]
WeNE8001	5	3	4	5	4	3	2	3	5	3	4	5	46	3.8
WeNE8002	2	4	5	2	3	3	2	3	5	5	3	3	40	3.3
WeNE8003	3	4	5	3	5	4	4	3	4	5	5	4	49	4.1
WeNE8004	5	4	3	3	3	4	1	2	5	4	3	4	41	3.4
WeNE8005	4	3	5	3	2	3	2	1	3	2	3	3	34	2.8
WeNE8006	3	3	4	3	3	2	3	2	5	3	3	4	38	3.2
WeNE8007	4	4	2	3	3	3	3	2	3	3	3	3	36	3.0
WeNE8008	4	3	5	4	2	5	3	2	3	3	5	5	44	3.7
WeNE8009	3	2	3	4	3	2	3	2	5	4	5	5	41	3.4
WeNE8010	3	4	4	2	3	3	3	2	5	4	4	4	41	3.4
Total NE	**6**	**5**	**6**	**5**	**5**	**6**	**4**	**4**	**6**	**5**	**6**	**6**	**64**[¶] **41.0**[‡]	**5.3**[§]
WeCP8001	3	2	3	4	2	4	2	2	4	2	4	2	34	2.8
WeCP8002	2	2	2	2	2	2	3	2	2	3	3	2	30	2.3
WeCP8003	4	3	3	4	2	3	3	2	4	2	3	2	35	2.9
WeCP8004	3	2	2	2	3	4	2	2	3	3	3	3	32	2.7
WeCP8005	3	5	3	2	2	2	2	2	2	2	2	3	30	2.5
Total CP	**5**	**5**	**4**	**5**	**3**	**5**	**4**	**3**	**5**	**4**	**4**	**4**	**51**[¶] **36.4**[‡]	**4.3**[§]
Average	**3.2**	**3.1**	**3.5**	**3.0**	**2.8**	**3.3**	**2.6**	**2.2**	**3.8**	**3.2**	**3.7**	**3.3**	**37.5**	**3.1**[*§]
Range	**2-5**	**2-5**	**2-5**	**1-5**	**1-5**	**2-5**	**1-4**	**1-3**	**2-5**	**1-5**	**2-6**	**1-6**	**27-49**	
Total	**7**	**7**	**7**	**7**	**7**	**7**	**7**	**6**	**8**	**8**	**9**	**9**	**89**	**7.4**[§§]

[¶] Number of alleles per region [‡]Average number of alleles per region [§] Number of alleles per region [§§]Average number of alleles per locus [*§]Average number of alleles per population

Genetic diversity

Twenty weedy rice populations exhibited high level of genetic diversity demonstrated by average gene diversity (0.473) and total gene diversity (0.734) (Table 2.3.7). The results of F_{ST} illustrated that half (50%) of genetic variation of those found in weedy rice was the differences between 20 weedy rice populations while the rest 50% was distributed between 386 individuals within 20 populations (Table 2.3.7). In mating system, moderate to high level of total inbreeding coefficient (0.682) was detected, lead to the moderate level of total outcrossing rate (0.189).

For each population, twenty weedy rice populations exhibited broad level of genetic diversity (Table 2.3.7). Population of WeNE8003 (Roi Et) showed the highest observed heterozygosity (0.408) while WeLN8005 (Phichit) had the lowest (0.108) with 0.172 in average. Similarly, the highest Nei's (1973) gene diversity (h) was also found in WeNE8003 (0.601) while the lowest was in WeLN8005 (0.288) with 0.494 in average. Weedy rice displayed various levels of inbreeding coefficient (F_{IS}). The highest level was found in WeLN8005 (Phichit) while the lowest level was WeNE8003 (Roi Et). On the other hand, population of WeNE8003 (Roi Et) showed the highest level of outcrossing rate (52%) while WeLN8005 (Phichit) showed the lowest (14%).

For each region (Table 2.3.8), weedy rice collected from three regions showed lower level of observed heterozygosity (H_O) than Nei's (1973) gene diversity (h). Weedy rice from the Northeast exhibited the highest genetic variation including observed heterozygosity (0.203), Nei's gene diversity (0.545) and average gene diversity (0.507) while the Lower North was the lowest (H_O = 0.126, h = 0.460 and H_S = 0.423, respectively). Weedy rice from the Northeast exhibited highest total

gene diversity (H_T = 0.664), following with the Central Plain (0.623) while the Lower North was lowest (0.602). Considering mating system, the highest level of inbreeding coefficient (F_{IS}) was found in the Lower North (0.727) following with the Central Plain (0.692) and the lowest was the Northeast (0.627). Conversely, the Northeast weedy rice exhibited the highest level of outcrossing rate (23%) following with the Central Plain (18%) and the lowest in the Lower North (16%).

Genetic structure

(a) Analysis of Molecular Variance (AMOVA)

Analysis of molecular variance (AMOVA) was used to examine the partitioning of total variation in weedy rice (Table 2.3.9). About 50% of total variance (0.734, Table 2.3.7) was distributed among 20 populations of weedy rice while the rest 50% was distributed between 386 individuals. Partitioning among regions showed that 26% of the total genetic variation was distributed among regions. For within each region, about 42%, 56% and 48% were the distribution of genetic variation among populations within the Lower North, the Northeast and the Central Plain, respectively (Table 2.3.9).

(b) Genetic differentiation (F_{ST})

In term of genetic differentiation (F_{ST}), weedy rice exhibited higher level of F_{ST} within region than between regions. Weedy rice populations of the Northeast (0.564) illustrated the highest level of genetic differentiation within populations while the Lower North (0.421) was the lowest. Considering pairwise genetic differentiation between regions, weedy rice of the Lower North to the Central Plain (0.121) were

more similar than those weed rice of the Lower North to the Northeast (0.202) and the Northeast to the Central Plain (0.249) (Table 2.3.10).

Table 2.3.7 Genetic parameters of 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008 using 12 SSR markers

Population	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
WeLN8001	18	0.393				0.125	0.682	0.189
WeLN8002	20	0.550				0.125	0.591	0.157
WeLN8003	18	0.354				0.125	0.647	0.214
WeLN8004	15	0.489				0.183	0.625	0.231
WeLN8005	19	0.288				0.108	0.810	0.142
WeNE8001	20	0.596				0.183	0.693	0.182
WeNE8002	23	0.496				0.167	0.664	0.202
WeNE8003	18	0.601				0.408	0.320	0.515
WeNE8004	18	0.525				0.225	0.571	0.273
WeNE8005	17	0.385				0.136	0.646	0.215
WeNE8006	19	0.441				0.175	0.603	0.248
WeNE8007	20	0.391				0.133	0.659	0.206
WeNE8008	20	0.561				0.217	0.614	0.239
WeNE8009	22	0.538				0.183	0.659	0.206
WeNE8010	20	0.535				0.207	0.613	0.240
WeCP8001	20	0.369				0.140	0.620	0.235
WeCP8002	23	0.588				0.208	0.616	0.201
WeCP8003	18	0.388				0.148	0.618	0.236
WeCP8004	20	0.433				0.123	0.717	0.165
WeCP8005	18	0.509				0.128	0.787	0.119
Total weedy rice in wet season of 2008 (20 populations)	**386**	**0.494**	**0.473**	**0.734**	**0.499**	**0.172**	**0.682**	**0.189**

Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Table 2.3.8 Summary of genetic parameters of weedy rice collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008 based on 12 SSR loci

Region	N	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
Lower North	5	90	0.460	0.423	0.602	0.421	0.126	0.727	0.158
Northeast	10	197	0.545	0.507	0.664	0.564	0.203	0.627	0.230
Central Plain	5	99	0.511	0.457	0.623	0.479	0.157	0.692	0.182
Among region	**20**	**386**	**0.494**	**0.473**	**0.734**	**0.263**	**0.172**	**0.682**	**0.189**

Number of populations (N), Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Average gene diversity (H_S), Total gene diversity (H_T), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Genetic structure

Table 2.3.9 Analysis of molecular variance (AMOVA) among populations and three rice production areas for 386 individuals of 20 weedy rice populations in wet 2008 based on 12 SSR loci

Source	df	SS	Variance component	% of the total variance
Among Populations	19	737.991	6.476	50%
Among Regions	2	496.313	3.785	26%
Populations/region	17	241.679	1.928	24%
Populations/Lower North	4	83.784	6.264	42%
Populations/Northeast	9	101.506	7.842	56%
Populations/Central Plain	4	56.388	4.859	48%
Individuals/Population	366	886.032	7.538	50%

Table 2.3.10 Pairwise of genetic differentiation (F_{ST}) within and between three rice production areas of weedy rice populations in wet 2008 based on 12 SSR loci

Pairwise F_{ST}		Region	
		Within	**Between**
Lower North		0.421	
	Northeast		0.202
	Central Plain		0.121
Northeast		0.564	
	Central Plain		0.249
Central Plain		0.479	

Population structure

Twenty weedy rice populations were structured into 2 inferred populations (K=2) on the basis of 12 microsatellite loci using STRUCTURE program (Figure 2.3.5). The first inferred population consisted of weedy rice populations were found in WeLN8001, WeLN8002, WECP8001 and WeCP8003. The second inferred population consisted of weedy rice populations were found in WeLN8003, WeLN8004, WeLN8005 and WeCP8005. In term of genome proportions (*Q*), the admixtures of the first inferred populations, were found in 3 weedy populations (WeCP8001, WeCP8002 and WeCP8004). Weedy rice populations consisted in the first inferred populations were collected from the fields with modern varieties particularly CNT1, SPR1, PTT1 and PSL2 (Figure 2.3.5 and Figure 2.3.6). Whereas the second inferred population consisted of weedy rice populations of WeNE8001 to WeNE8010 which were collected from the fields with improved traditional varieties particularly KDML105, RD15 and RD6 (Figure 2.3.5 and Figure 2.3.6).

Principal coordinate analysis (PCA) clustered the 20 weedy rice populations into 2 groups. The EIGEN analysis of the pairwise Chord genetic distance measures for among 20 weedy rice populations explained 45.25% of the variation presented in the two weedy rice groups within the first and second axes. The first group consisted of weedy rice populations collected from the Lower North, the Central Plain and some populations from the Northeast with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2 while weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6 were clustered in second group (Figure 2.3.7).

Similarly, NJ clustering method base on C.S. Chord genetic distance also divided 20 weedy rice populations into 2 major clusters (Figure 2.3.8). The first cluster consisted of weedy rice populations collected from the Lower North, the Central Plain and WeNE8005, with collected from the Northeast, with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2. The second cluster consisted of weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6.

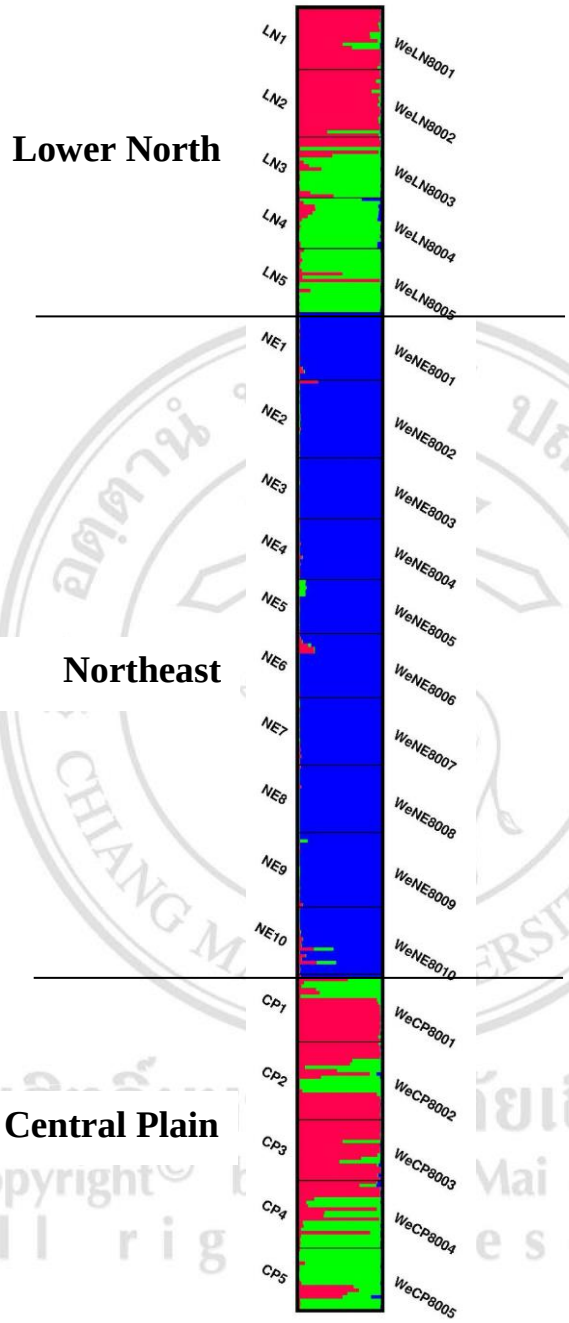


Figure 2.3.5 Population assignment of 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008 reveal K=2. Different colors represent different inferred populations, referred to different K.

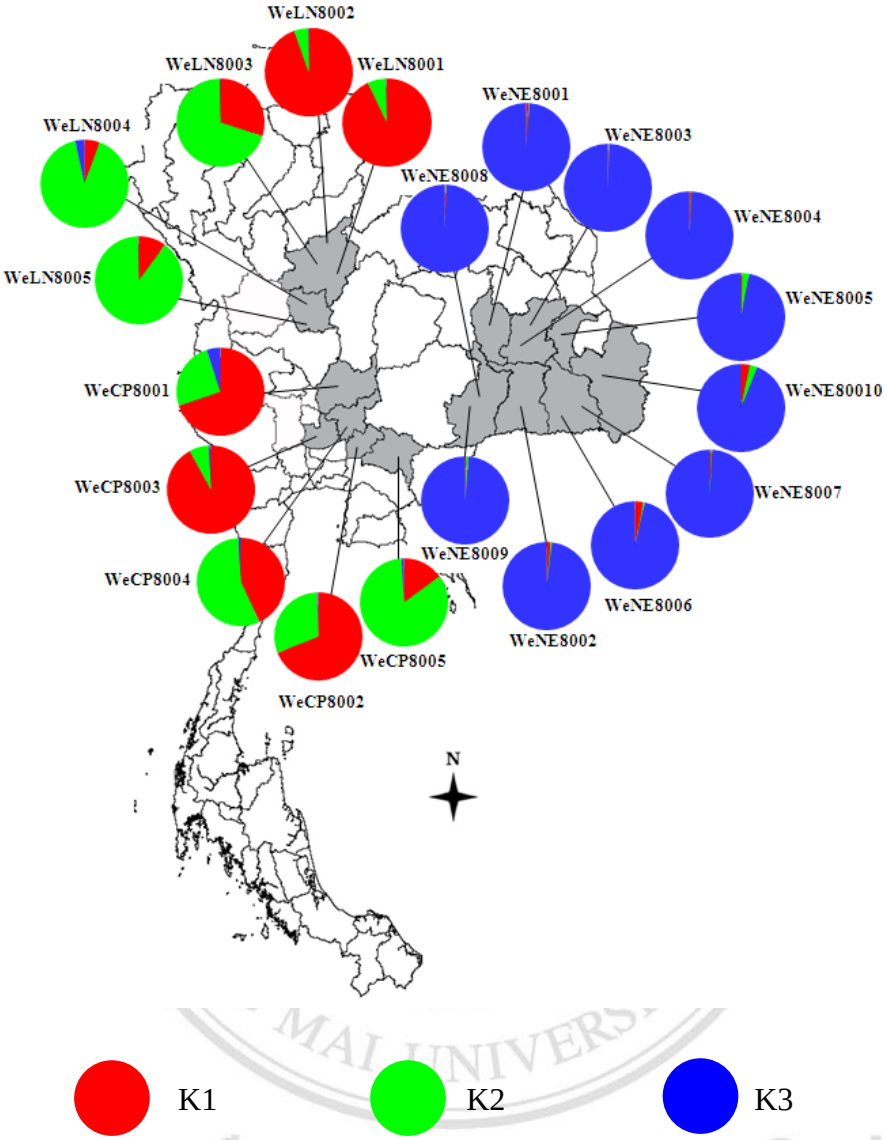


Figure 2.3.6 Genetic proportion of 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008 (K=2)

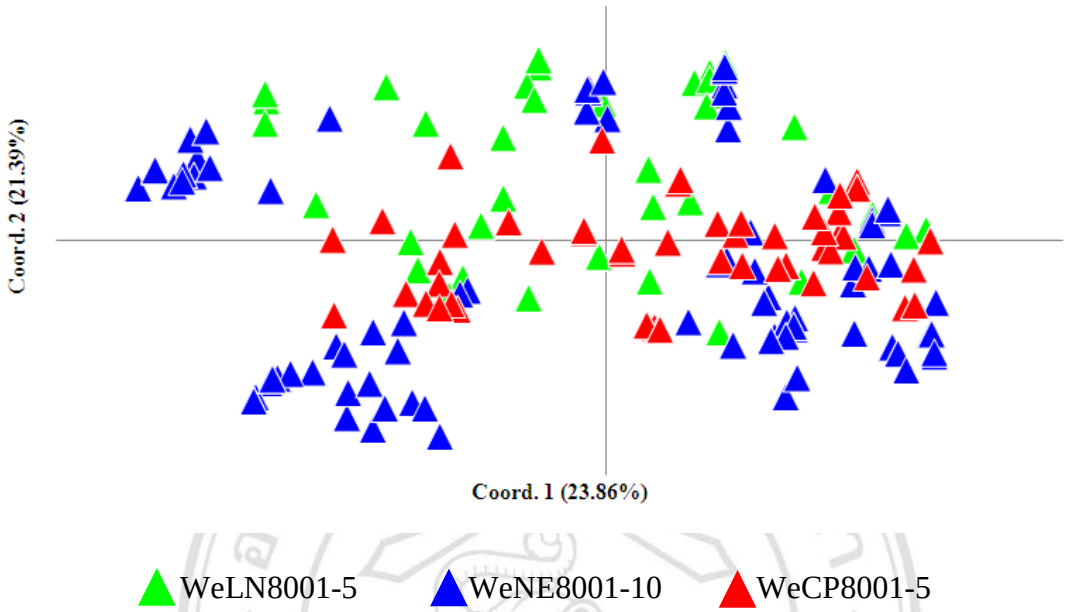


Figure 2.3.7 Distribution of 386 individuals of 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008. Different colors represent 2 clusters referred to the assignment obtained from STRUCTURE: Green-weedy rice collected from the Lower North fields, Blue-weedy rice populations collected from the Northeast fields and Red- weedy rice populations collected from the Central Plain fields, formed by the principal coordinate analysis (PCA) on the basis of 12 microsatellite markers.

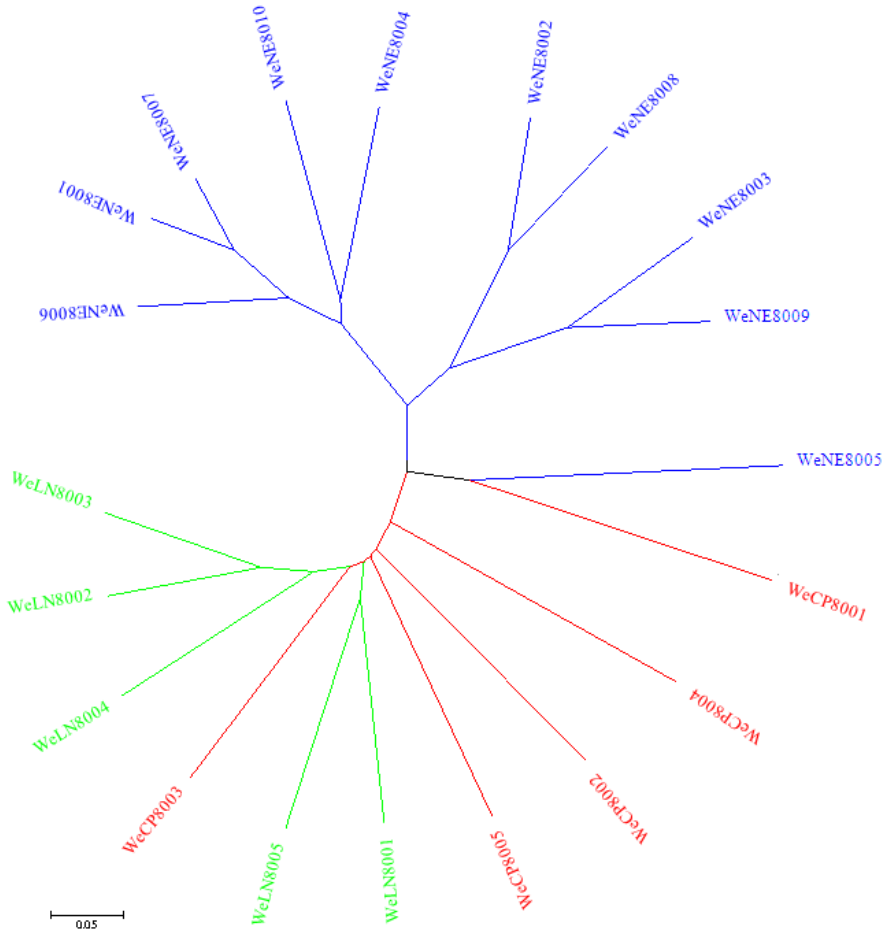


Figure 2.3.8 Cluster analysis using NJ method based on C.S. Chord (1967) genetic distance among 20 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2008

Weedy rice populations from wet 2009

Allele diversity

Various levels of allele diversities of 18 weedy rice populations from wet season of 2009 were presented in Table 2.3.11. Total number of 85 alleles of weedy rice was observed with 12 SSR loci with the average of 7.1 alleles. Number of alleles per locus ranged from 6 alleles in RM211, RM280, RM133, RM234 and RM481 to 9 alleles in RM247. For 12 loci, average number of allele per population was 3.0 alleles.

At individual population level, total number of alleles were ranging from 28 alleles in WeLN9106 (Nakhon Sawan) to 43 alleles in WeNE9103 (Maha Sarakham) with 36.5 alleles in average (Table 2.3.11). Number of alleles per locus were ranging from 1 to 6 alleles per population with an average ranging from 2.6 allele per locus in WeLN9106 (Nakhon Sawan) to 3.6 alleles per locus in WeNE9103 (Maha Sarakham).

Among the regions, the highest number of alleles was observed in the Northeast totally 61 alleles, followed with 53 alleles in the Central Plain while the Lower North was the lowest with 47 alleles. Average number of alleles per region was 38.2 alleles in Northeast, 37.5 in the Central Plain and the lowest in the Lower North, 33.7 alleles. Average number of alleles per region per locus was the highest in the Northeast (5.1 alleles), followed in the Central Plain and the Lower North were 4.4 and 4.0 alleles, respectively.

Table 2.3.11 Allele diversity of 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009 using 12 SSR markers

Population	Number of allele (A)												Total No. of allele	Average No. of allele
	RM1	RM109	RM211	RM251	RM280	RM133	RM234	RM481	RM477	RM316	RM206	RM247		
WeLN9101	3	4	4	4	3	3	2	2	4	3	3	3	38	3.2
WeLN9102	1	4	4	3	2	4	1	3	1	4	4	4	35	2.9
WeLN9103	4	4	3	3	3	2	1	2	4	3	5	3	37	3.1
WeLN9104	3	4	4	2	2	3	2	2	3	3	3	2	33	2.8
WeLN9105	5	3	4	3	2	3	2	4	4	3	2	3	38	3.2
WeLN9106	3	2	4	2	3	3	3	2	3	2	4	2	28	2.6
Total LN	**4**	**5**	**4**	**4**	**3**	**4**	**3**	**4**	**3**	**4**	**5**	**4**	**47¶ 33.7‡**	**4.0§**
WeNE9101	2	3	3	3	3	3	2	2	3	4	4	4	36	3.0
WeNE9102	3	2	4	2	3	3	4	2	2	4	2	3	34	2.8
WeNE9103	2	4	3	5	3	4	3	3	2	5	3	6	43	3.6
WeNE9104	2	3	3	2	4	2	3	2	3	5	4	4	37	3.1
WeNE9105	4	4	3	5	3	4	2	3	5	3	4	2	42	3.5
WeNE9106	4	2	4	4	4	4	1	3	3	3	4	5	41	3.4
WeNE9107	4	2	4	2	3	3	3	2	4	3	4	5	39	3.3
WeNE9108	5	4	3	3	3	3	3	2	3	3	3	3	38	3.2
WeNE9109	3	4	3	3	2	2	2	1	5	2	3	3	33	2.8
WeNE9110	3	4	4	3	3	3	3	2	3	4	3	4	39	3.3
Total NE	**6**	**5**	**5**	**6**	**4**	**4**	**4**	**4**	**6**	**6**	**5**	**6**	**61¶ 38.2‡**	**5.1§**
WeCP9101	2	4	4	4	4	3	5	3	4	2	5	4	40	3.4
WeCP9102	4	4	4	3	5	3	4	2	4	3	5	3	34	3.1
Total CT	**5**	**5**	**5**	**4**	**3**	**4**	**3**	**4**	**4**	**4**	**4**	**3**	**53¶ 37.5‡**	**4.4§**
Average	**2.8**	**3.1**	**3.4**	**3.2**	**3.0**	**3.2**	**2.2**	**2.3**	**3.1**	**3.4**	**3.6**	**3.7**	**36.5**	**3.0*§**
Range	**1-5**	**2-4**	**3-4**	**2-5**	**2-4**	**2-4**	**1-4**	**1-4**	**1-5**	**2-5**	**2-5**	**2-6**	**28-43**	
Total	**8**	**7**	**6**	**7**	**6**	**6**	**6**	**6**	**8**	**8**	**8**	**9**	**85**	**7.1§§**

¶ Number of alleles per region ‡Average number of alleles per region § Number of alleles per region §§Average number of alleles per locus *§Average number of alleles per population

Genetic diversity

Eighteen weedy rice exhibited high level of genetic diversity demonstrated by average gene diversity (0.434) and total gene diversity (0.720) (Table 2.3.12). The results of F_{ST} illustrated that more than half (52%) of genetic variation of those found in weedy rice was the differences between 18 weedy rice populations while the rest 48% was distributed between 180 individauls within 18 populations (Table 2.3.12). In mating system, moderate to high level of total inbreeding coefficient (0.695) was detected, lead to the moderate level of total outcrossing rate (0.180).

For each population, eighteen weedy rice populations exhibited broad level of genetic diversity (Table 2.3.12). Population of WeNE9103 (Maha Sarakham) showed the highest observed heterozygosity (0.226) while WeLN9106 (Nakhon Sawan) had the lowest (0.118) with 0.161 in average. The highest Nei's (1973) gene diversity (h) was also found in WeNE9103 (0.558) while the lowest was in WeLN9106 (0.317) with 0.437 in average. Weedy rice displayed various levels of inbreeding coefficient (F_{IS}). The highest level was found in WeLN9106 (Nakhon Sawan) while the lowest level was WeNE9103 (Maha Sarakham). On the other hand, population of WeNE9103 (Maha Sarakham) showed the highest level of outcrossing rate (30%) while WeLN9106 (Nakhon Sawan) showed the lowest (9%).

For each region (Table 2.3.13), weedy rice collected from three regions showed lower level of observed heterozygosity (H_O) than Nei's (1973) gene diversity (h). Weedy rice from the Northeast exhibited the highest genetic variation including observed heterozygosity (0.176), Nei's gene diversity (0.513) and average gene diversity (0.456) while the Lower North was the lowest (H_O = 0.083, h = 0.407 and H_S = 0.393, respectively). All weedy rice from three regions exhibited similar total

gene diversity (H_T), 0.637 in the Northeast, 0.630 in the Central Plain and 0.603 in the Lower North. Considering mating system,the highest level of inbreeding coefficient (F_{IS}) was found in the Lower North (0.794) following with the Central Plain (0.692) and the lowest was the Northeast (0.658). Conversely, the Northeast weedy rice exhibited the highest level of outcrossing rate (22%) following with the Central Plain (20%) and the lowest in the Lower North (12%).

Genetic structure

(a) Analysis of Molecular Variance (AMOVA)

Analysis of molecular variance (AMOVA) was used to examine the partitioning of total variation in weedy rice (Table 2.3.14). About 52% of total variance (0.720, Table 2.3.12) was distributed among 18 populations of weedy rice while the rest 48% was distributed between 180 individuals. Partitioning among regions showed that 29% of the total genetic variation was distributed among regions. For within each region, about 44%, 57% and 50% were the distribution of genetic variation among populations within the Lower North, the Northeast and the Central Plain, respectively (Table 2.3.14).

(b) Genetic differentiation (F_{ST})

In term of genetic differentiation (F_{ST}), weedy rice exhibited higher level of F_{ST} within region than between regions. Weedy rice populations of the Northeast (0.571) illustrated the highest level of genetic differentiation within populations while the Lower North (0.445) was the lowest. Considering pairwise genetic differentiation between regions, weedy rice of the Lower North to the Central Plain (0.125) were

more similar than those weed rice of the Lower North to the Northeast (0.211) and the Northeast to the Central Plain (0.282) (Table 2.3.15).

Table 2.3.12 Genetic parameters of 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009 using 12 SSR markers

Population	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
WeLN9101	10	0.413				0.175	0.577	0.269
WeLN9102	10	0.387				0.156	0.598	0.252
WeLN9103	10	0.412				0.167	0.636	0.123
WeLN9104	10	0.329				0.142	0.569	0.274
WeLN9105	10	0.461				0.183	0.602	0.148
WeLN9106	10	0.317				0.118	0.675	0.085
WeNE9101	10	0.410				0.167	0.594	0.255
WeNE9102	10	0.386				0.153	0.604	0.247
WeNE9103	10	0.558				0.226	0.513	0.304
WeNE9104	10	0.413				0.158	0.616	0.237
WeNE9105	10	0.549				0.200	0.595	0.254
WeNE9106	10	0.508				0.222	0.563	0.280
WeNE9107	10	0.488				0.175	0.642	0.218
WeNE9108	10	0.429				0.165	0.616	0.238
WeNE9109	10	0.348				0.147	0.665	0.201
WeNE9110	10	0.472				0.183	0.612	0.241
WeCP9101	10	0.442				0.147	0.636	0.152
WeCP9102	10	0.453				0.152	0.586	0.160
Total weedy rice in wet season of 2009 (18 populations)	**180**	**0.437**	**0.434**	**0.720**	**0.515**	**0.161**	**0.695**	**0.180**

Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Table 2.3.13 Summary of genetic parameters of weedy rice collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009 based on 12 SSR loci

Region	N	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
Lower North	6	60	0.407	0.393	0.603	0.445	0.083	0.794	0.124
Northeast	10	100	0.513	0.456	0.637	0.571	0.176	0.658	0.216
Central Plain	2	20	0.444	0.448	0.630	0.502	0.164	0.692	0.196
Among region	**18**	**180**	**0.437**	**0.434**	**0.720**	**0.288**	**0.161**	**0.695**	**0.180**

Number of populations (N), Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Average gene diversity (H_S), Total gene diversity (H_T), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Genetic structure

Table 2.3.14 Analysis of molecular variance (AMOVA) among populations and 3 three rice production areas for 180 individuals of 18 weedy rice populations in wet 2009 based on 12 SSR loci

Source	df	SS	Variance component	% of the total variance
Among Populations	17	687.484	6.488	52%
Among Regions	2	474.940	4.321	29%
Populations/region	15	212.544	1.980	23%
Populations/Lower North	5	77.124	6.318	44%
Populations/Northeast	9	102.194	7.153	57%
Populations/Central Plain	1	33.226	5.012	50%
Individuals/Population	162	840.357	7.561	48%

Table 2.3.15 Pairwise of genetic differentiation (F_{ST}) within and between three rice production areas of weedy rice populations in wet 2009 based on 12 SSR loci

Pairwise F_{ST}		Region	
		Within	**Between**
Lower North		0.445	
	Northeast		0.211
	Central Plain		0.125
Northeast		0.571	
	Central Plain		0.282
Central Plain		0.502	

Population structure

Eighteen weedy rice populations were structured into 2 inferred populations (K=2) on the basis of 12 microsatellite loci using STRUCTURE program (Figure 2.3.9). In term of genome proportions (*Q*), the admixtures of the first and second inferred populations, were found in 4 weedy populations (WeLN9101, WeLN9104 WeCP9101 and WeCP9102). Weedy rice populations consisted in the first inferred populations were collected from the fields with modern varieties particularly CNT1, SPR1, PTT1 and PSL2 (Figure 2.3.9 and Figure 2.3.10). Whereas the second inferred population consisted of weedy rice populations of WeNE9101 to WeNE9110 which were collected from the fields with improved traditional varieties particularly KDML105, RD15 and RD6 (Figure 2.3.9 and Figure 2.3.10).

Principal coordinate analysis (PCA) clustered the 18 weedy rice populations into 2 groups. The EIGEN analysis of the pairwise Chord genetic distance measures for among 18 weedy rice populations explained 50.50% of the variation presented in the two weedy rice groups within the first and second axes. The first group consisted of weedy rice populations collected from the Lower North and the Central Plain with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2 while weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6 were clustered in second group (Figure 2.3.11).

Similarly, NJ clustering method base on C.S. Chord genetic distance also divided 18 weedy rice populations into 2 major clusters (Figure 2.3.12). The first cluster consisted of weedy rice populations collected from the Lower North and the Central Plain with collected from Northeast, with modern varieties popular cultivation

particularly CNT1, SPR1, PTT1 and PSL2. The second cluster consisted of weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6.

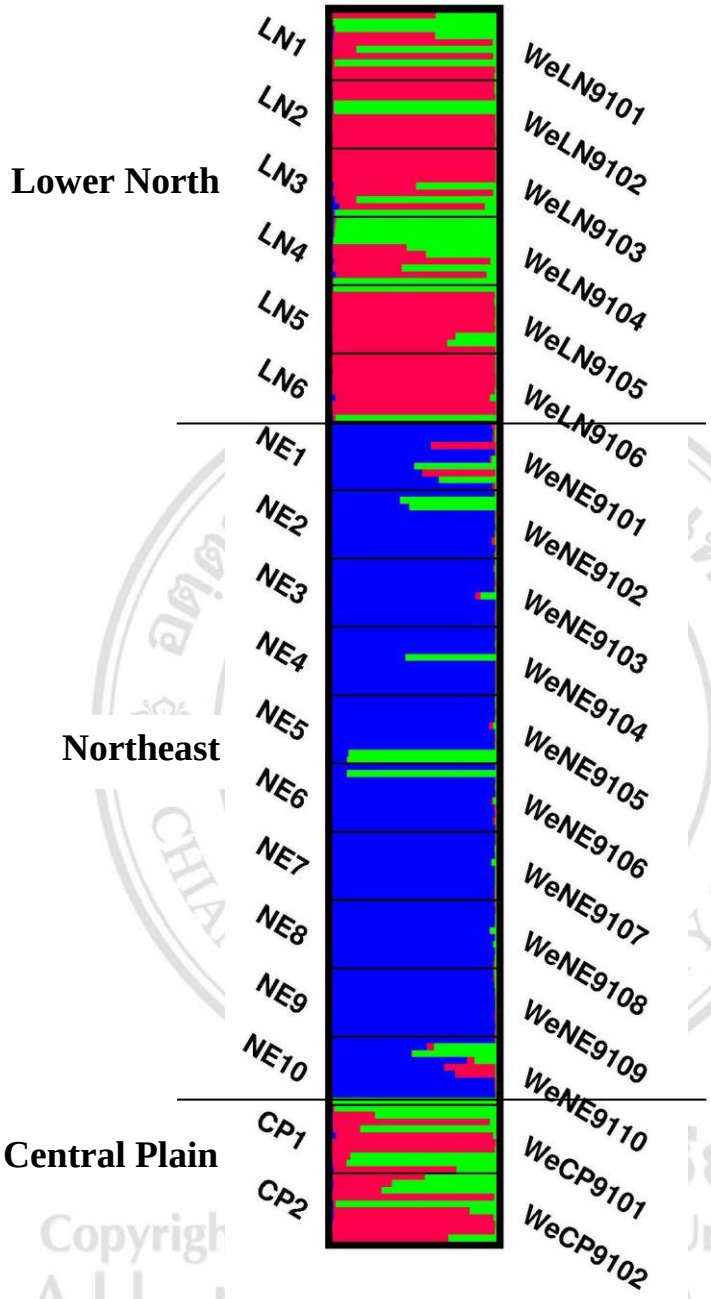


Figure 2.3.9 Population assignment of 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009 reveal K=2. Each bar represent each population consisted 10 individuals. Different colors represent different inferred populations, referred to different K.

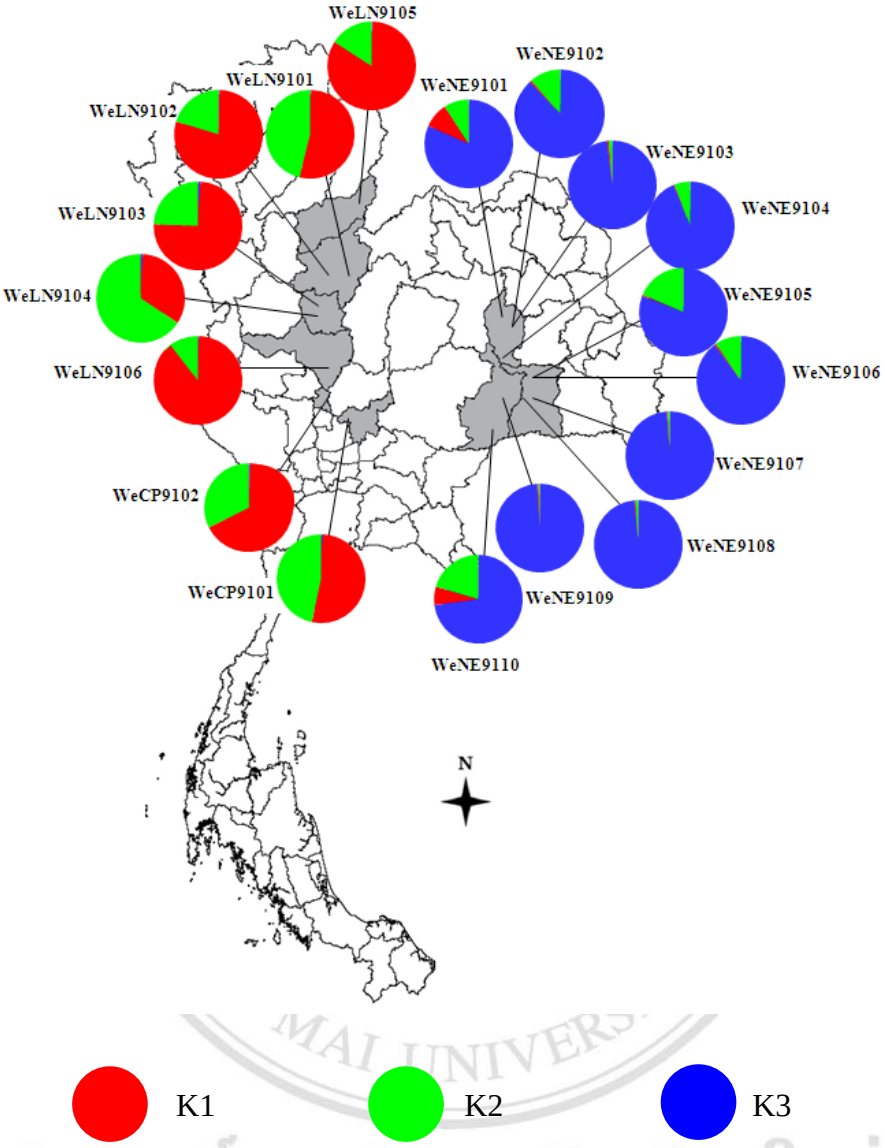


Figure 2.3.10 Genetic proportion of 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009 (K=2)

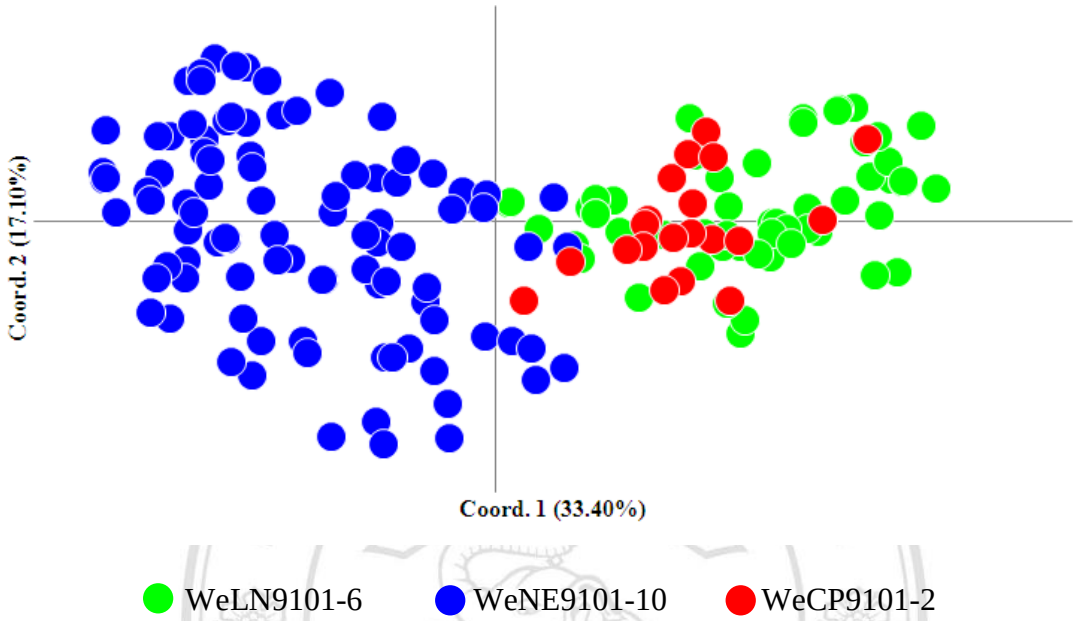


Figure 2.3.11 Distribution of 180 individuals of 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009. Different colors represent 2 clusters referred to the assignment obtained from STRUCTURE: Green-weedy rice collected from the Lower North fields, Blue-weedy rice populations collected from the Northeast fields and Red- weedy rice populations collected from the Central Plain fields, formed by the principal coordinate analysis (PCA) on the basis of 12 microsatellite markers.

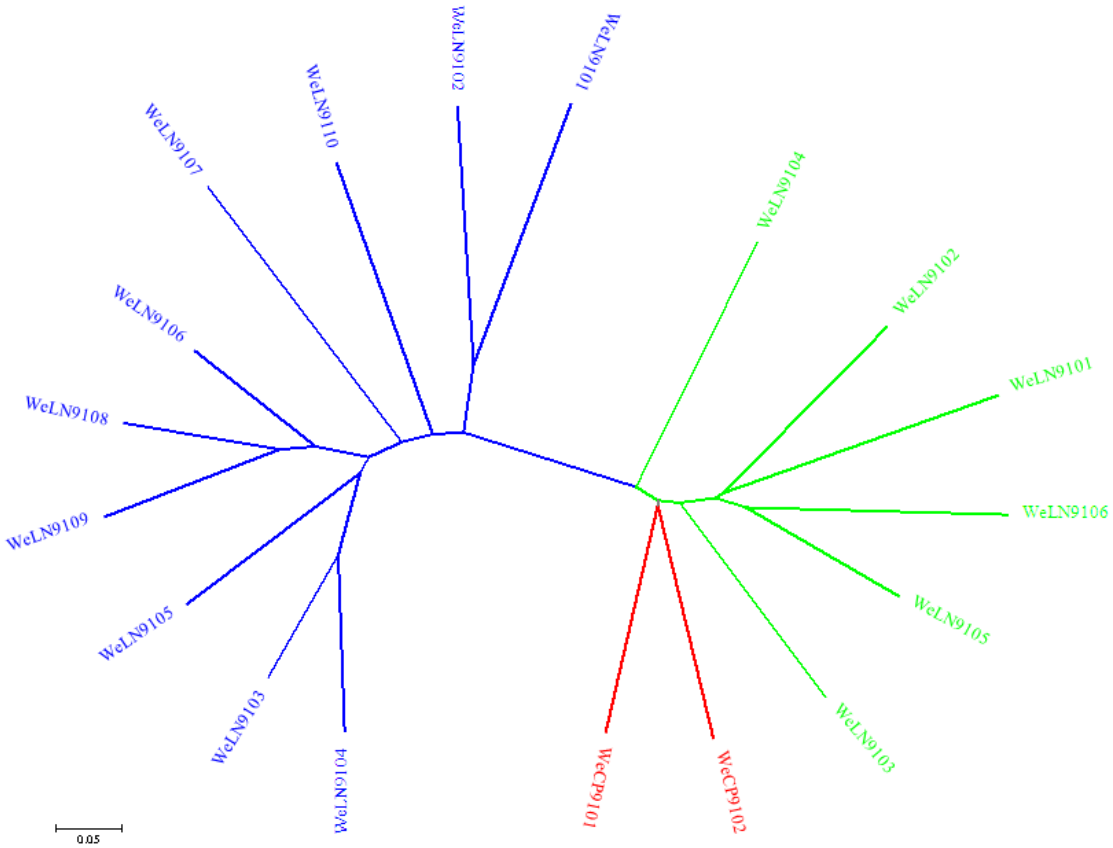


Figure 2.3.12 Cluster analysis using NJ method based on C.S. Chord (1967) genetic distance among 18 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in wet 2009

Weedy rice populations from dry 2009/10

Allele diversity

Various levels of allele diversities of 17 weedy rice populations from dry season of 2009 were presented in Table 2.3.16. Total number of 83 alleles of weedy rice was observed with 12 SSR loci with the average of 6.9 alleles. Number of alleles per locus ranged from 5 alleles in RM234 and RM481 to 9 alleles in RM206. For 12 loci, average number of allele per population was 2.9 alleles.

At individual population level, total number of alleles were ranging from 24 alleles in WeLN9204 (Phichit) to 42 alleles in WeNE9202 (Khon Kaen) with 34.8 alleles in average (Table 2.3.16). Number of alleles per locus were ranging from 1 to 6 alleles per population with an average ranging from 2.1 allele per locus in WeLN9204 (Phichit) to 3.5 alleles per locus in WeNE9202 (Khon Kaen).

Among the regions, the highest number of alleles was observed in the Northeast totally 57 alleles, followed with 47 alleles in the Central Plain while the Lower North was the lowest with 45 alleles. Average number of alleles per region was 38.7 alleles in Northeast, 31.7 in the Lower North and the lowest in the Central Plain, 29 alleles. Average number of alleles per region per locus was the highest in the Northeast (4.8 alleles), followed in the Lower North and the Central Plain were 3.8 and 3.7 alleles, respectively.

Table 2.3.16 Allele diversity of 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10 using 12 SSR markers

Population	Number of allele (A)												Total No. of allele	Average No. of allele
	RM1	RM109	RM211	RM251	RM280	RM133	RM234	RM481	RM477	RM316	RM206	RM247		
WeLN9201	2	3	2	2	3	3	2	2	3	4	4	3	35	2.9
WeLN9202	3	3	3	1	3	4	3	2	3	4	4	4	37	3.1
WeLN9203	3	3	3	3	3	4	2	2	3	3	3	3	39	3.3
WeLN9204	2	2	3	2	2	2	2	2	2	2	3	2	24	2.1
WeLN9205	2	3	3	3	3	3	2	2	3	2	2	2	30	2.5
WeLN9206	2	3	4	3	3	2	2	1	3	4	5	3	38	3.2
WeLN9207	3	3	4	3	3	3	2	3	3	3	3	1	38	3.2
Total LN	**4**	**3**	**4**	**4**	**3**	**4**	**3**	**3**	**4**	**4**	**5**	**4**	**45**[¶] **31.7**[‡]	**3.8**[§]
WeNE9201	4	3	3	3	3	2	2	2	3	3	2	3	33	2.8
WeNE9202	3	4	4	4	3	3	3	3	3	4	4	4	42	3.5
WeNE9203	4	4	3	3	2	2	3	2	3	3	6	5	40	3.3
WeNE9204	1	2	3	4	4	5	2	2	5	4	4	3	39	3.3
WeNE9205	2	4	4	3	2	4	2	3	3	2	4	3	36	3.0
WeNE9206	4	4	4	4	3	4	1	2	4	3	4	5	41	3.4
Total NE	**5**	**5**	**5**	**4**	**4**	**5**	**3**	**4**	**6**	**5**	**6**	**5**	**57**[¶] **38.7**[‡]	**4.8**[§]
WeCP9201	4	4	2	2	5	6	2	3	3	2	3	3	34	2.8
WeCP9202	3	2	3	2	2	2	3	3	3	4	3	2	30	2.5
WeCP9203	2	3	2	2	2	2	4	3	4	4	4	2	27	2.3
WeCP9204	3	2	4	2	2	2	2	2	2	2	2	2	25	2.1
Total CP	**4**	**4**	**4**	**3**	**5**	**6**	**4**	**3**	**4**	**4**	**4**	**3**	**47**[¶] **29.0**[‡]	**3.7**[§]
Average	**3.1**	**3.0**	**2.9**	**2.5**	**3.0**	**3.3**	**2.5**	**2.4**	**3.5**	**3.1**	**3.6**	**3.4**	**34.8**	**2.9**[*§]
Range	**1-5**	**2-4**	**2-4**	**1-5**	**2-5**	**2-6**	**1-3**	**1-3**	**2-5**	**2-5**	**2-6**	**1-5**	**25-42**	
Total	**8**	**6**	**6**	**7**	**7**	**8**	**5**	**5**	**7**	**7**	**9**	**8**	**83**	**6.9**[§§]

[¶] Number of alleles per region [‡]Average number of alleles per region [§] Number of alleles per region [§§]Average number of alleles per locus [*§]Average number of alleles per population

Genetic diversity

Seventeen weedy rice populations exhibited high level of genetic diversity demonstrated by average gene diversity (0.426) and total gene diversity (0.714) (Table 2.3.17). The results of F_{ST} illustrated that more half (53%) of genetic variation of those found in weedy rice was the differences between 17 weedy rice populations while the rest 47% was distributed between 170 individuals within 17 populations (Table 2.3.17). In mating system, high level of total inbreeding coefficient (0.723) was detected, lead to the low level of total outcrossing rate (0.160).

For each population, seventeen weedy rice populations exhibited broad level of genetic diversity (Table 2.3.17). Population of WeNE9202 (Khon Kaen) showed the highest observed heterozygosity (0.183) while WeLN9204 (Phichit) had the lowest (0.102) with 0.134 in average. Similarly, the highest Nei's (1973) gene diversity (h) was also found in WeNE9202 (0.552) while the lowest was in WeLN9204 (0.262) with 0.426 in average. Weedy rice displayed various levels of inbreeding coefficient (F_{IS}). The highest level was found in WeLN9204 (Phichit) while the lowest level was WeNE9202 (Khon Kaen). On the other hand, population of WeNE9202 (Khon Kaen) showed the highest level of outcrossing rate (40%) while WeLN9204 (Phichit) showed the lowest (8%).

For each region (Table 2.3.18), weedy rice collected from three regions showed lower level of observed heterozygosity (H_O) than Nei's (1973) gene diversity (h). Weedy rice from the Northeast exhibited the highest genetic variation including observed heterozygosity (0.157), Nei's gene diversity (0.519) and average gene diversity (0.468) while the Lower North was the lowest (H_O = 0.143, h = 0.392 and H_S = 0.392, respectively). All weedy rice from three regions exhibited similar total

gene diversity (H_T), 0.619 in the Northeast, 0.617 in the Central Plain and 0.601 in the Lower North. Considering mating system, the highest level of inbreeding coefficient (F_{IS}) was found in the Lower North (0.815) following with the Central Plain (0.698) and the lowest was the Northeast (0.658). Conversely, the Northeast weedy rice exhibited the highest level of outcrossing rate (21%) following with the Central Plain (18%) and the lowest in the Lower North (10%).

Genetic structure

(a) Analysis of Molecular Variance (AMOVA)

Analysis of molecular variance (AMOVA) was used to examine the partitioning of total variation in weedy rice (Table 2.3.19). About 53% of total variance (0.714, Table 2.3.17) was distributed among 17 populations of weedy rice while the rest 47% was distributed between 170 individuals. Partitioning among regions showed that 30% of the total genetic variation was distributed among regions. For within each region, about 48%, 61% and 51% were the distribution of genetic variation among populations within the Lower North, the Northeast and the Central Plain, respectively (Table 2.3.19).

(b) Genetic differentiation (F_{ST})

In term of genetic differentiation (F_{ST}), weedy rice exhibited higher level of F_{ST} within region than between regions. Weedy rice populations of Northeast (0.608) illustrated the highest level of genetic differentiation within populations while the Lower North (0.477) was the lowest. Considering pairwise genetic differentiation between regions, weedy rice of the Lower North to the Central Plain (0.124) were

more similar than those weed rice of the Lower North to the Northeast (0.219) and the Northeast to the Central Plain (0.284) (Table 2.3.20).

Table 2.3.17 Genetic parameters of 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10 using 12 SSR markers

Population	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
WeLN9201	10	0.390				0.125	0.679	0.191
WeLN9202	10	0.412				0.156	0.622	0.233
WeLN9203	10	0.468				0.161	0.656	0.208
WeLN9204	10	0.262				0.102	0.759	0.081
WeLN9205	10	0.315				0.175	0.444	0.385
WeLN9206	10	0.451				0.133	0.705	0.173
WeLN9207	10	0.447				0.163	0.636	0.222
WeNE9201	10	0.326				0.146	0.551	0.290
WeNE9202	10	0.552				0.183	0.668	0.399
WeNE9203	10	0.504				0.137	0.728	0.157
WeNE9204	10	0.483				0.175	0.638	0.221
WeNE9205	10	0.400				0.158	0.604	0.247
WeNE9206	10	0.543				0.142	0.739	0.150
WeCP9201	10	0.385				0.158	0.649	0.182
WeCP9202	10	0.416				0.108	0.740	0.150
WeCP9203	10	0.489				0.181	0.634	0.191
WeCP9204	10	0.398				0.143	0.693	0.156
Total weedy rice in dry season of 2009 (17 populations)	**170**	**0.419**	**0.426**	**0.714**	**0.528**	**0.134**	**0.723**	**0.160**

Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Table 2.3.18 Summary of genetic parameters of weedy rice collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10 based on 12 SSR loci

Region	N	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
Lower North	7	70	0.392	0.392	0.601	0.477	0.143	0.805	0.102
Northeast	6	60	0.519	0.468	0.619	0.608	0.157	0.658	0.206
Central Plain	4	40	0.440	0.422	0.617	0.515	0.151	0.698	0.178
Among region	**17**	**170**	**0.419**	**0.426**	**0.714**	**0.302**	**0.134**	**0.723**	**0.160**

Number of populations (N), Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Average gene diversity (H_S), Total gene diversity (H_T), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

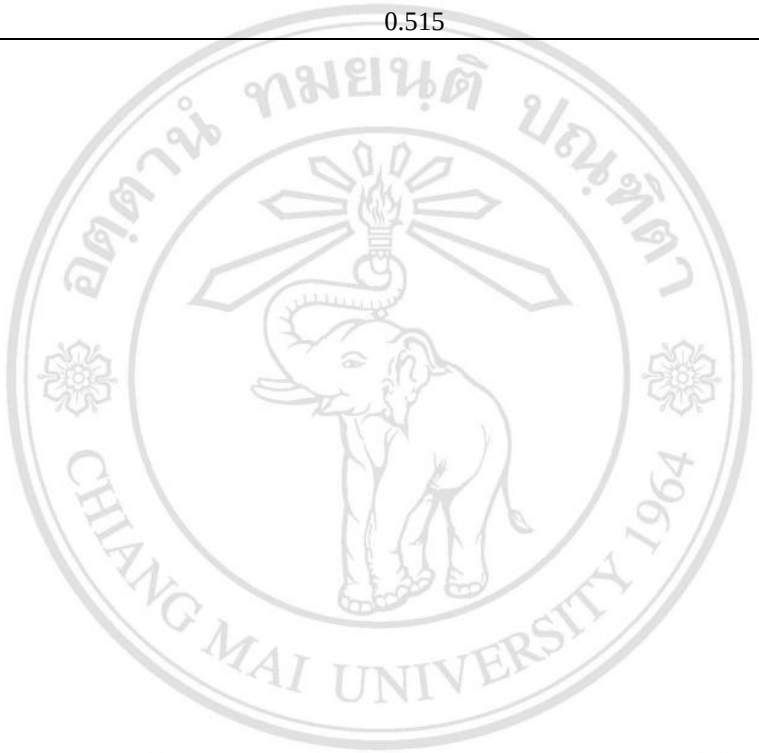
Genetic structure

Table 2.3.19 Analysis of molecular variance (AMOVA) among populations and three rice production areas for 180 individuals of 17 weedy rice populations in dry 2009/10 based on 12 SSR loci

Source	df	SS	Variance component	% of the total variance
Among Populations	16	674.573	6.170	53%
Among Regions	2	502.008	4.591	30%
Populations/region	14	172.565	1.578	23%
Populations/Lower North	6	58.745	6.088	48%
Populations/Northeast	5	63.546	6.569	61%
Populations/Central Plain	3	50.275	5.786	51%
Individuals/Population	153	753.466	7.128	47%

Table 2.3.20 Pairwise of genetic differentiation (F_{ST}) within and between three rice production areas of weedy rice populations in dry 2009/10 based on 12 SSR loci

Pairwise F_{ST}		Region	
		Within	**Between**
Lower North		0.477	
	Northeast		0.219
	Central Plain		0.124
Northeast		0.608	
	Central Plain		0.284
Central Plain		0.515	



Population structure

Seventeen weedy rice populations were structured into 2 inferred populations (K=2) on the basis of 12 microsatellite loci using STRUCTURE program (Figure 2.3.13). In term of genome proportions (*Q*), the admixtures of the first inferred populations, were found in 4 weedy populations (WeLN9202, WeLN9203 WeLN9204 and WeCP9203). Weedy rice populations consisted in the first inferred populations were collected from the fields with modern varieties particularly CNT1, SPR1, PTT1 and PSL2 (Figure 2.3.13 and Figure 2.3.14). Whereas the second inferred population consisted of weedy rice populations of WeNE9201 to WeNE9206 which were collected from the fields with improved traditional varieties particularly KDML105, RD15 and RD6 (Figure 2.3.13 and Figure 2.3.14).

Principal coordinate analysis (PCA) clustered the 17 weedy rice populations into 2 groups. The EIGEN analysis of the pairwise Chord genetic distance measures for among 18 weedy rice populations explained 48.93% of the variation presented in the two weedy rice groups within the first and second axes. The first group consisted of weedy rice populations collected from the Lower North and the Central Plain with modern varieties popular cultivation particularly CNT1, SPR1, PTT1 and PSL2 while weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6 were clustered in second group (Figure 2.3.15).

Similarly, NJ clustering method base on C.S. Chord genetic distance also divided 17 weedy rice populations into 2 major clusters (Figure 2.3.16). The first cluster consisted of weedy rice populations collected from the Lower North and the Central Plain with collected from the Northeast, with modern varieties popular

cultivation particularly CNT1, SPR1, PTT1 and PSL2. The second cluster consisted of weedy rice populations collected from the Northeast with improved traditional varieties cultivation fields particularly KDML105, RD15 and RD6.

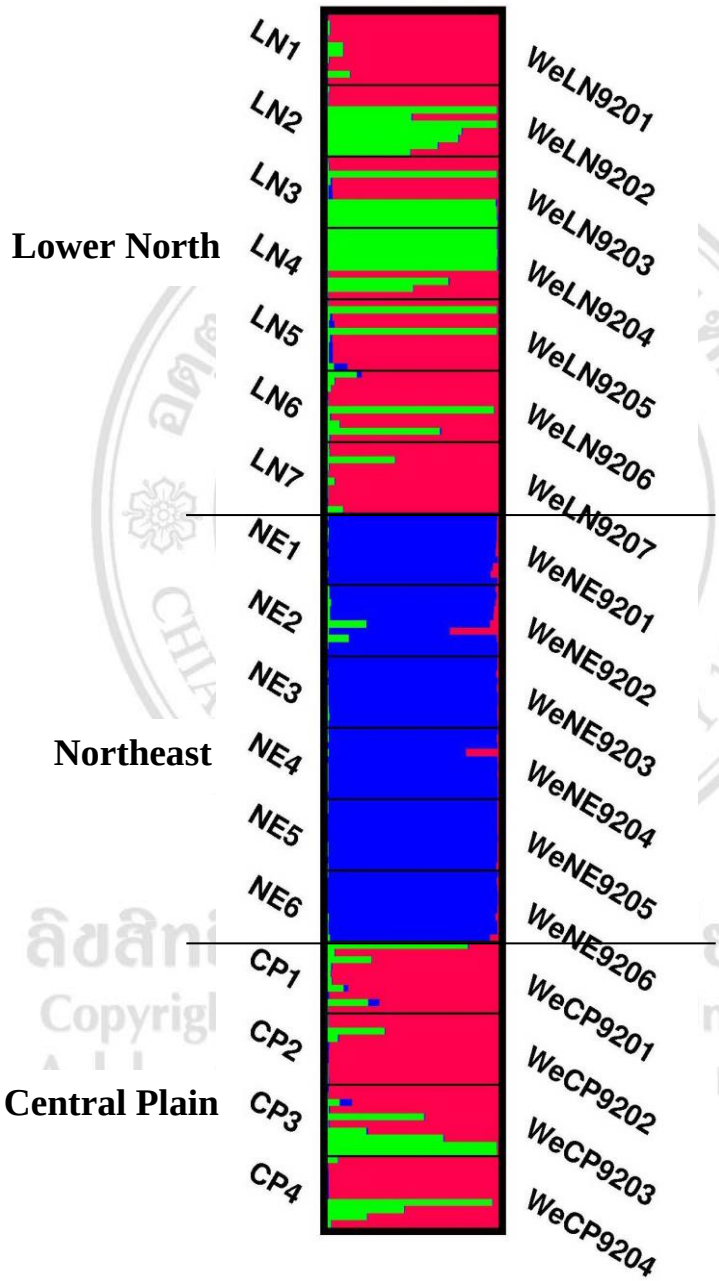


Figure 2.3.13 Population assignment of 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP)

of Thailand in dry 2009/10 reveal K=2. Each bar represent each population consisted 10 individuals. Different colors represent different inferred populations, referred to different K.

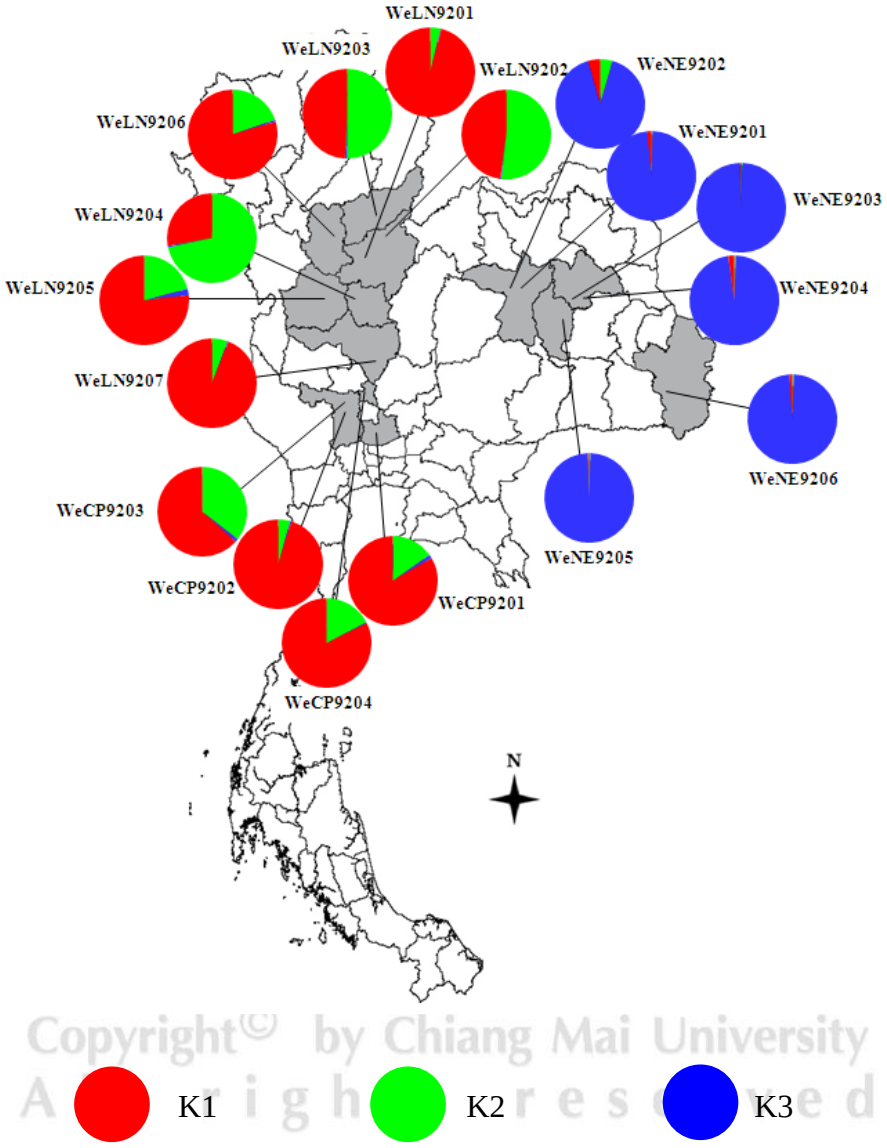


Figure 2.3.14 Genetic proportion of 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10 (K=2)

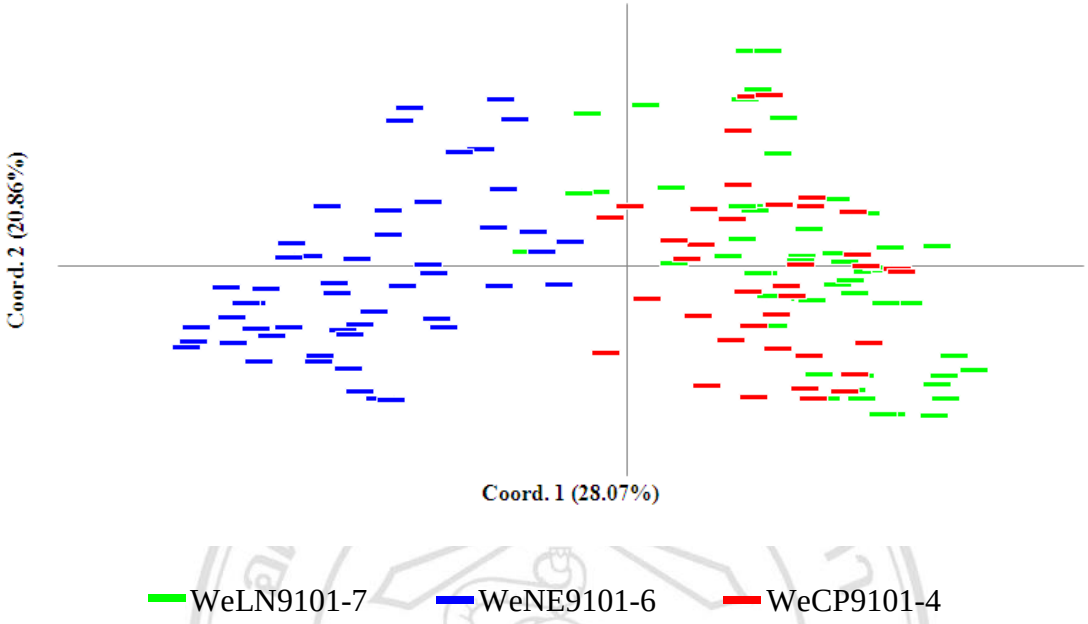


Figure 2.3.15 Distribution of 170 individuals of 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10. Different colors represent 2 clusters referred to the assignment obtained from STRUCTURE: Green-weedy rice collected from the Lower North fields, Blue-weedy rice populations collected from the Northeast fields and Red- weedy rice populations collected from the Central Plain fields, formed by the principal coordinate analysis (PCA) on the basis of 12 microsatellite markers.

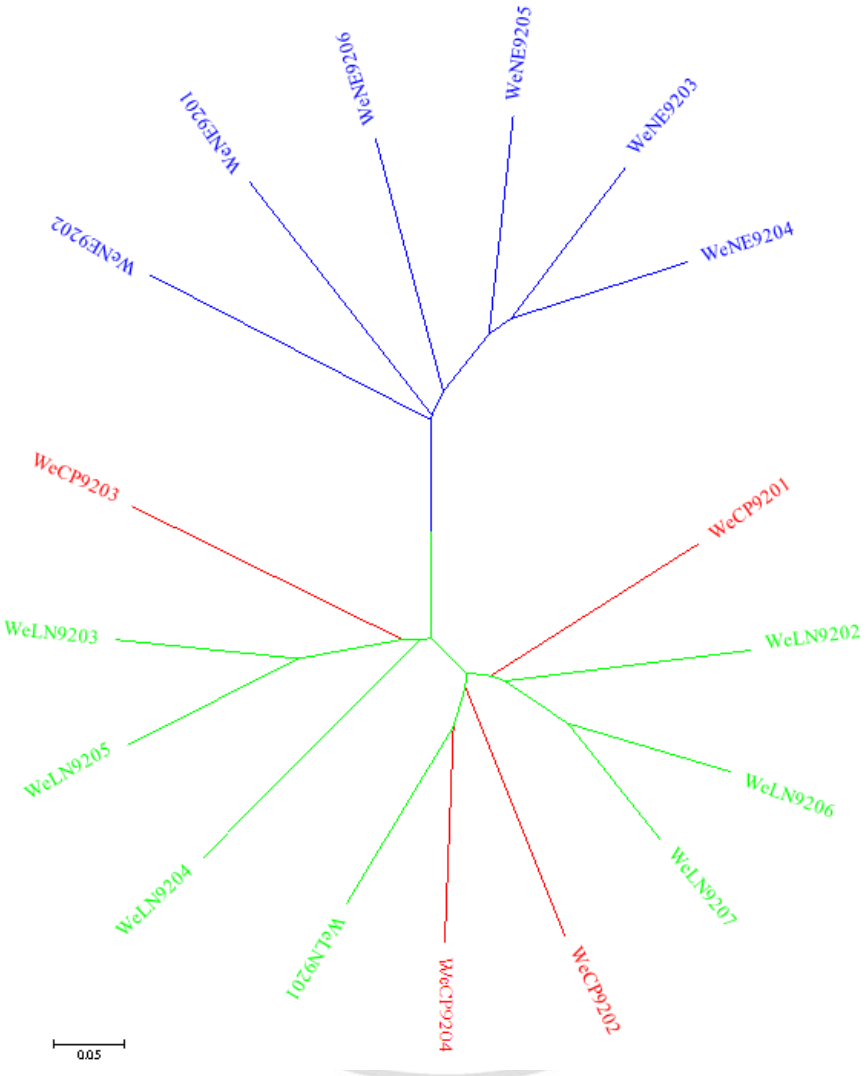


Figure 2.3.16 Cluster analysis using NJ method based on C.S. Chord (1967) genetic distance among 17 weedy rice populations collected from three rice production areas; Lower North (LN), Northeast (NE) and Central Plain (CP) of Thailand in dry 2009/10

Population genetic structure and temporal dynamics of weedy rice populations in Thailand and compared with pure line rice varieties and common wild rice

Allele diversity

Level of allele diversities of 63 weedy rice populations in four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009 were investigated and compared with cultivated rice and common wild rice (Table 2.3.21). For cultivated rice checks, no allele diversity was detected within seven pure line cultivated rice. Total number of polymorphic alleles observed among 37 cultivated rice varieties in 12 SSR loci was 42 alleles with average of 3.5 alleles per locus. Number of alleles for each locus ranged from 2 alleles in RM109 to 5 alleles in RM211. Each variety was fixed at one allele per locus per variety (Table 2.3.21). For common wild rice checks, high level of genetic variation was found in seven common wild rice populations (Table 2.3.21). Total number of polymorphic alleles detected with 12 SSR loci was 169 with an average of 14.1 alleles per locus. For each locus, number of alleles ranged from 4 alleles in RM133 to 21 alleles in RM1 and RM477 with an average of 14.1 alleles per locus. For weedy rice, various levels of allele diversities of 63 weedy rice populations from 4 seasons were presented in Table 2.3.21. Total number of 160 alleles of weedy rice was observed with 12 SSR loci with the average of 13.3 alleles. Number of alleles per locus ranged from 12 alleles in RM234 and RM481 to 15 alleles in RM1 and RM211. For 12 loci, average number of allele per population was 3.1 alleles.

For among four seasons, the polymorphism of allele was decreased with year of infestation. The highest number of alleles was observed in wet 2005 totally 100 alleles, followed with 89 alleles in wet 2008, 85 alleles in wet 2009 while dry 2009/10

was the lowest with 83 alleles. Average number of alleles per season was 43.1 alleles in wet 2005, 37.5 in wet 2008, 36.5 in wet 2009 and the lowest in dry 2009/10, 34.8 alleles. Average number of alleles per season per locus was the highest in wet 2005 (3.6 alleles), followed in wet 2008, wet 2009 and dry 2009/10 were 3.1, 3.0 and 2.9 alleles, respectively (Table 2.3.21).

Table 2.3.21 Allele diversity of 63 weedy rice populations collected from three rice production areas; Lower North, Northeast and Central Plain of Thailand in four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10 using 12 SSR markers.

Population	Number of allele (A)												Total No. of allele	Average No. of allele
	RM1	RM109	RM211	RM251	RM280	RM133	RM234	RM481	RM477	RM316	RM206	RM247		
Cultivated rice														
Average	1	1	1	1	1	1	1	1	1	1	1	1	12	1.0*§
Range	1	1	1	1	1	1	1	1	1	1	1	1	1	
Total	4	2	5	4	3	3	4	3	2	4	4	4	42	3.5§§
Common wild rice														
Average	6.1	4.3	6.3	4.9	3.7	2.0	5.0	4.6	5.0	4.1	5.1	3.9	55.0	4.6*§
Range	4-9	3-6	3-11	2-7	2-6	1-3	3-8	3-7	3-7	2-7	3-8	2-6	37-71	
Total	21	15	18	10	8	4	16	15	21	8	17	16	169	14.1§§
Weedy rice in wet 2005														
Average	4.0	3.8	4.4	3.8	3.5	3.6	2.6	2.8	4.0	3.4	4.0	3.4	43.1	3.6*§
Range	2-7	2-5	2-7	2-5	2-5	3-5	2-4	2-4	2-6	2-6	3-5	2-5	36-53	
Total	10	8	10	8	8	8	7	7	9	9	8	8	100	8.3§§
Weedy rice in wet 2008														
Average	3.2	3.1	3.5	3.0	2.8	3.3	2.6	2.2	3.8	3.2	3.7	3.3	37.5	3.1*§
Range	2-5	2-5	2-5	1-5	1-5	2-5	1-4	1-3	2-5	1-5	2-6	1-6	27-49	
Total	7	7	7	7	7	7	7	6	8	8	9	9	89	7.4§§
Weedy rice in wet 2009														
Average	2.8	3.1	3.4	3.2	3.0	3.2	2.2	2.3	3.1	3.4	3.6	3.7	36.5	3.0*§
Range	1-5	2-4	3-4	2-5	2-4	2-4	1-4	1-4	1-5	2-5	2-5	2-6	28-43	
Total	8	7	6	7	6	6	6	6	8	8	8	9	85	7.1§§
Weedy rice in dry 2009/10														
Average	3.1	3.0	2.9	2.5	3.0	3.3	2.5	2.4	3.5	3.1	3.6	3.4	34.8	2.9*§
Range	1-5	2-4	2-4	1-5	2-5	2-6	1-3	1-3	2-5	2-5	2-6	1-5	25-42	
Total	8	6	6	7	7	8	5	5	7	7	9	8	83	6.9§§
Total of weedy rice														
Average	3.2	3.2	3.5	3.0	2.9	3.2	2.4	2.3	3.5	3.2	3.6	3.3	37.2	3.1*§
Range	1-7	2-5	2-7	1-5	1-5	2-6	1-4	1-4	1-5	1-6	2-6	1-6	25-53	
Total	15	13	15	13	13	13	12	12	14	14	13	13	160	13.3§§

§§Average number of alleles per locus *§Average number of alleles per population

Genetic diversity

Genetic diversity from all weedy rice populations in four growing seasons were analyzed together with cultivated rice and common wild rice (Figure 2.3.17 and Table 2.3.22). No variation was detected within variety of cultivated rice but high level of total genetic diversity was found (H_S = 0 and H_T = 0.625, respectively). In contrast, common wild rice showed the highest level of average and total genetic diversity (H_S = 0.611 and H_T = 0.842, respectively). Sixty three weedy rice populations showed intermediate value of genetic diversity (H_S = 0.436 and H_T = 0.825, respectively) between seven common wild rice populations and seven cultivated rice varieties but had the same high value of total genetic diversity as common wild rice (Table 2.3.22).

For compared among four seasons, genetic diversity of the 63 weedy rice populations was decreased with year of infestation (Figure 2.3.17 and Table 2.3.22). Weedy rice in wet 2005 exhibited the highest genetic variation including observed heterozygosity (H_O = 0.274), Nei's gene diversity (h = 0.575), average gene diversity (H_S = 0.555) and total gene diversity (H_T = 0.827), following in populations of wet 2008 (H_O = 0.172, h = 0.494, H_S = 0.473 and H_T = 0.734) and wet 2009 (H_O = 0.161, h = 0.437, H_S = 0.434 and H_T = 0.720) while the populations of dry 2009/10 was the lowest (H_O = 0.134, h = 0.419, H_S = 0.426 and H_T = 0.714). On the other hand, the highest level of genetic differentiation (F_{ST}) was found in dry 2009/10 (0.528) following with wet 2009 (0.515), in wet 2008 (0.499) and the lowest in wet 2005 (0.428). Considering mating system, inbreeding coefficient (F_{IS}) was the highest in weedy rice in dry 2009/10 (0.723) following with wet 2009 (0.695), in wet 2008 (0.682) and the lowest in wet 2005 (0.572), respectively. Conversely, outcrossing rate

(*t*) was the highest in weedy rice in wet 2005 (27%) while the lowest in dry 2009/10 was16% (Figure 2.3.17 and Table 2.3.22).

Genetic structure

(a) Analysis of Molecular Variance (AMOVA)

Analysis of molecular variance (AMOVA) was conducted to investigate the overall distribution of genetic diversity among rice groups (Table 2.3.23). About 9% of total variance was partitioned among rice groups. For weedy rice, partitioning among seasons showed that 33% of the total genetic variation was distributed among four seasons. For within each season, about 43%, 50%, 52% and 53% were the distribution of genetic variation among populations within wet 2005, wet 2008, wet 2009 and dry 2009/10, respectively, with increased with year of infestation (Table 2.3.23).

(b) Genetic differentiation (F_{ST})

In term of genetic differentiation (F_{ST}), weedy rice exhibited higher level of F_{ST} within season than between seasons. Degrees of genetic differentiation (F_{ST}) from the highest to the lowest were weedy rice in dry 2009/10, wet 2009, wet 2008 and wet 2005, respectively. Pairwised F_{ST} was highest between weedy rice in wet 2009 vs dry 2009/10 (0.025) while the lowest was found between weedy rice in wet 2005 vs dry 2009/10 (0.190) (Table 2.3.24).

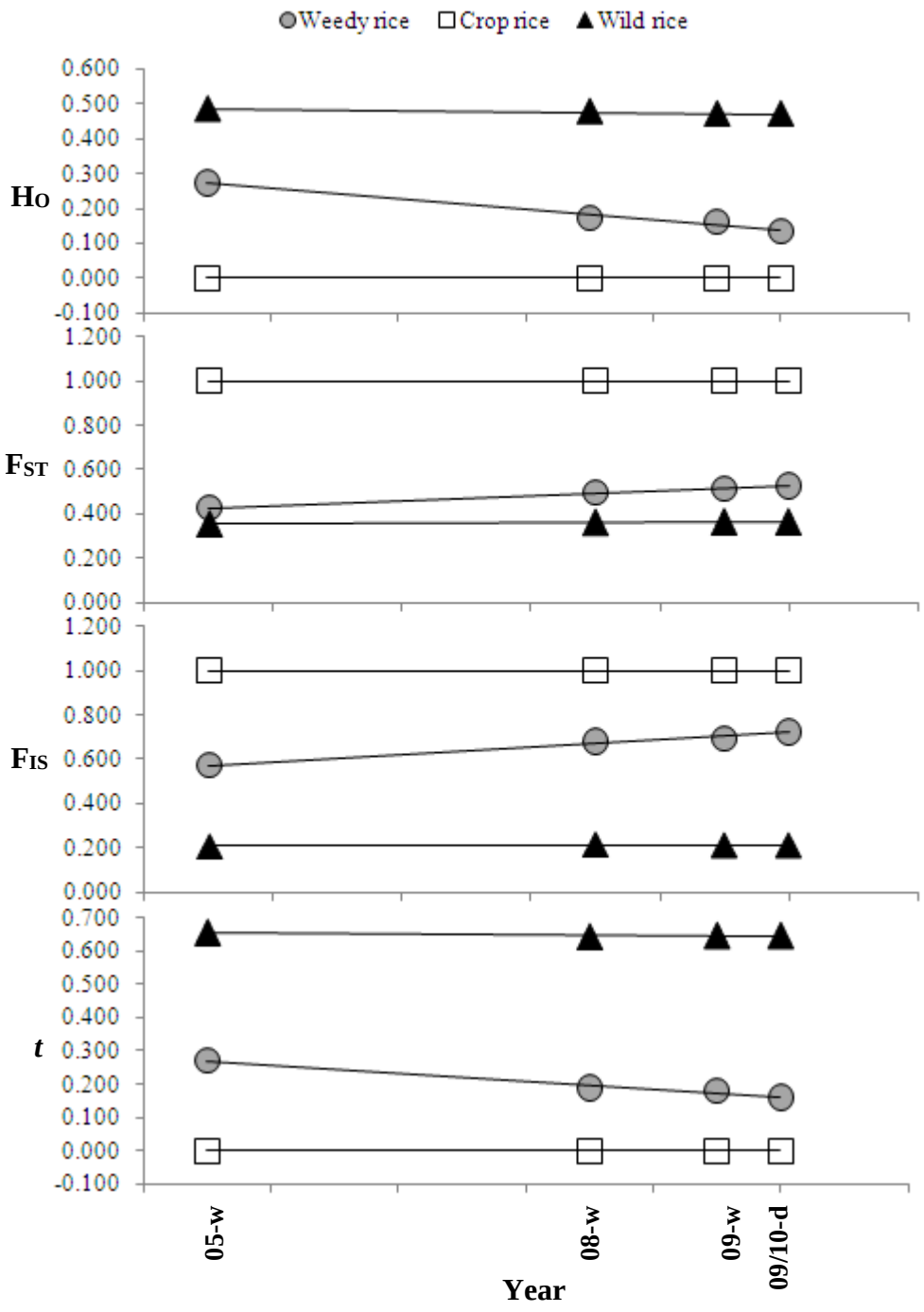


Figure 2.3.17 Observed heterozygosity (H_O), Genetic differentiation (F_{ST}), Inbreeding coefficient (F_{IS}) and Outcrossing rate (*t*) of 63 weedy rice populations collected in four growing seasons in wet 2005 (05-w), wet 2008 (08-w), wet 2009 (09-w) and dry 2009/10 (09/10-d) using 12 SSR markers.

Table 2.3.22 Genetic parameters of 63 weedy rice populations collected from three rice production areas; Lower North, Northeast and Central Plain of Thailand in four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10 using 12 SSR markers.

Population	N	n	*h*	H_S	H_T	F_{ST}	H_O	F_{IS}	*t*
Weedy rice	**63**	**887**	**0.536**	**0.436**	**0.825**	**0.327**	**0.172**	**0.614**	**0.239**
wet 2005	8	151	0.575	0.555	0.827	0.428	0.274	0.572	0.272
wet 2008	20	386	0.494	0.473	0.734	0.499	0.172	0.682	0.189
wet 2009	18	180	0.437	0.434	0.720	0.515	0.161	0.695	0.180
dry 2009/10	17	170	0.419	0.426	0.714	0.528	0.134	0.723	0.160
Cultivated rice	**7**	**70**	**0**	**0**	**0.625**	**1.000**	**0**	**1.000**	**0**
Common wild rice	**7**	**70**	**0.645**	**0.611**	**0.842**	**0.419**	**0.504**	**0.176**	**0.701**

Number of populations (N), Number of individuals (n), Observed heterozygosity (H_O), Nei's (1973) gene diversity (*h*), Average gene diversity (H_S), Total gene diversity (H_T), Inbreeding coefficient (F_{IS}), and Outcrossing rate (*t*)

Genetic structure

Table 2.3.23 Analysis of molecular variance (AMOVA) among populations of each for 887 individuals of 63 weedy rice populations collected from three rice production areas; Lower North, Northeast and Central Plain of Thailand in four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10 using 12 SSR markers.

Source	df	SS	Variance component	% of the total variance
Among rice groups	2	1626.186	2.201	9%
Populations/Group	74	6129.469	12.080	66%
Populations/Weedy rice	62	1155.843	7.088	33%
wet 2005	7	339.603	14.052	43%
wet 2008	19	285.982	11.833	50%
wet 2009	17	274.066	11.340	52%
dry 2009/10	16	256.192	10.601	53%
Populations/Cultivated rice	6	3502.554	76.569	100%
Populations/Common wild rice	6	1471.073	9.128	42%
Individuals/Population	950	4753.466	7.786	25%

Table 2.3.24 Pairwise of genetic differentiation (F_{ST}) within and between four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10 of 63 weedy rice populations collected from three rice production areas; Lower North, Northeast and Central Plain of Thailand using 12 SSR markers.

Pairwise F_{ST}		**Season**	
		Within	**Between**
wet 2005		0.428	
	wet 2008		0.135
	wet 2009		0.165
	dry 2009/10		0.190
wet 2008		0.499	
	wet 2009		0.030
	dry 2009/10		0.055
wet 2009		0.515	
	dry 2009/10		0.025
dry 2009/10		0.528	

Population structure

Seven pure line rice varieties (Cr) were structured into 3 inferred populations (K = 3) (Figure 2.3.18) on the basis of 12 microsatellite loci using STRUCTURE program, the first inferred population was SPR1 or CNT1, the second inferred population was PTT1 or PSL2 and the third inferred population was KDML105 or RD15 or RD6, each interred population represent by the presence of red, green and blue colors, respectively. Seven native common wild rice (Wi) from three regions were structured into 1 inferred populations (K = 1), represent by the presence of gray color (Figure 2.3.18).

The sixty-three weedy rice populations in the present study resulted from hybridization between common wild rice and crop rice in each region. Evidence of their genetic admixtures in STRUCTURE model was composed genetic proportion between common wild rice and crop rice but the higher proportions of cultivar genotypes were found than wild genotype and tended to increase with years of infestation (Figure 2.3.18 and Figure 2.3.19). Consequently, the population structure of weedy rice populations can be structured into 2 clusters according to its companion crop rice varieties of each region (*K* = 2) (Figure 2.3.18). The admixtures of the first inferred populations consisted of weedy rice populations from the Lower North and the Central Plain. These admixtures contained cultivar genotypes, CNT1 or SPR1 or PTT1 or PSL2, and wild genotype. These modern cultivated rice varieties are commonly found cultivation in paddy fields of these regions (Figure 2.3.18 (b) and (d) and Figure 2.3.19 (a) and (c)). The rest of second inferred population consisted of weedy rice populations from the Northeast. The admixtures contained cultivar genotypes, KDML105 or RD6 or RD15, and wild genotype. These improved

traditional rice varieties are commonly found cultivation in paddy fields of this region (Figure 2.3.18 (c) and Figure 2.3.19 (b)). When considering among seasons, the proportions of cultivar genotypes in weedy rice from all regions were found more than wild genotype and trend to increase with years of infestation. This increasing of the proportion of cultivar genotypes may caused by the subsequently backcross of weedy rice to cultivated rice in rice fields resulting the rapidly loss of wild traits in segregating populations.

The result of genetic admixtures of weedy rice populations by STRUCTURE is consistent with the distribution of weedy rice by principal coordinate analysis (PCA) analysis (Figure 2.3.20 (a-d)). The relationship among wild, weedy and crop rice was displayed that crop rice and common wild rice were widely distributed across the graph while weedy rice groups were distributed between the groups of crop rice and common wild rice but tended to closer toward the group of cultivar with years of infestation. When all weedy rice populations from four seasons were analyzed together and compared with seven pure line rice varieties and common wild rice checks displayed that the pattern of weedy rice groups were unified group in seasons later. Neighbor-join tree also revealed relationship among rice groups illustrated that weedy rice populations each region were also divided among companion crop rice varieties and common wild rice populations group but tended to closer toward the group of cultivar with years of infestation (Figure 2.3.21 (a-d)).

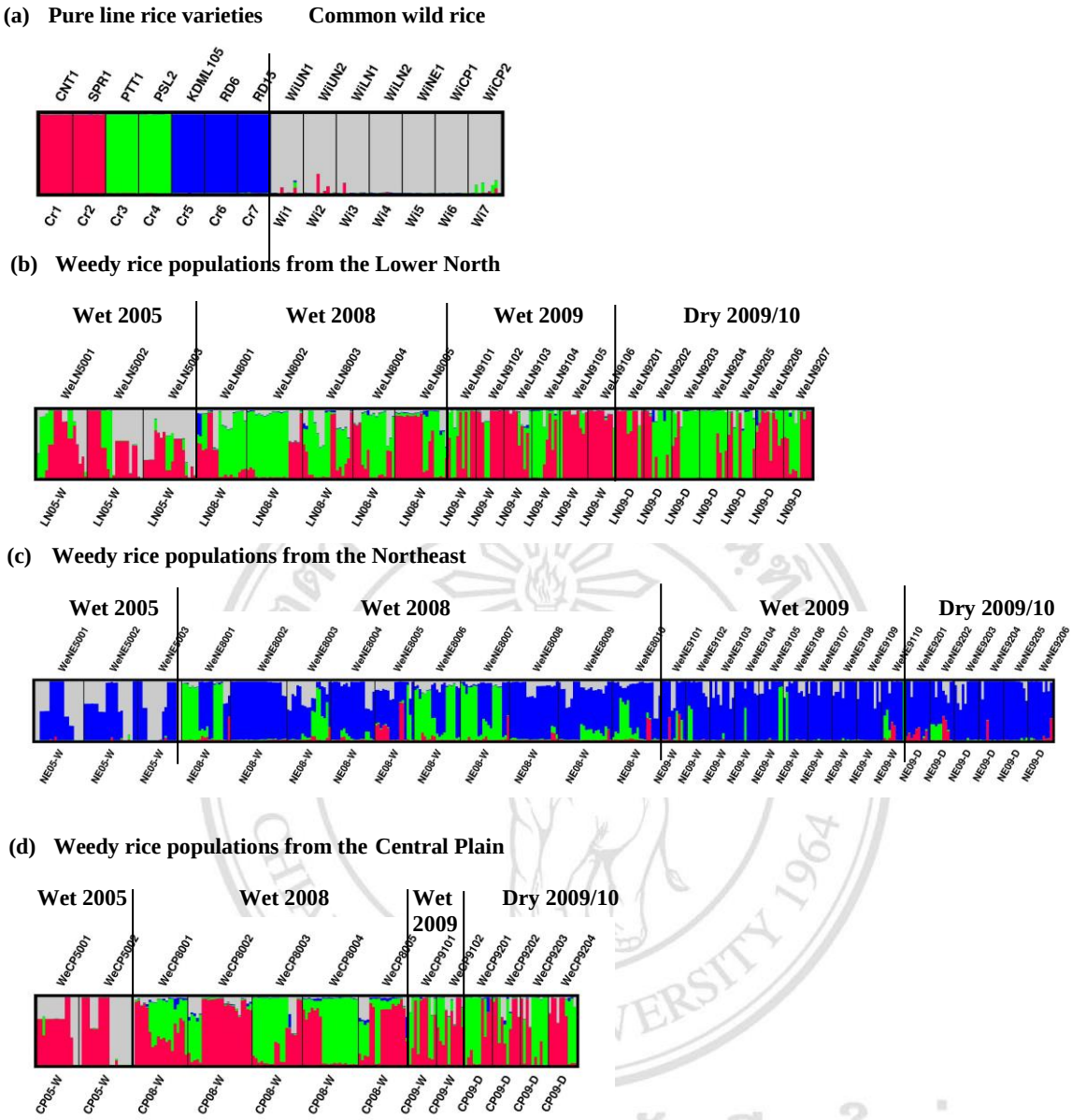


Figure 2.3.18 Population assignment using STRUCTURE analysis of 63 weedy rice populations collected from three rice production areas; Lower North (b), Northeast (c) and Central Plain (d) in four growing seasons in wet 2005, wet 2008, wet 2009 and dry 2009/10, compared with 7 pure line cultivated rice varieties; SPR1 (Cr1), CNT1 (Cr2), PTT1 (Cr3), PSL2 (Cr4), KDML105 (Cr5), RD15 (Cr6) and RD6 (Cr7), and 7 common wild rice populations (Wi1-7) (a). Each bar represented each population consist 10-23 individuals. Different colors represent different inferred populations (*K* = 4).

(a) Lower North

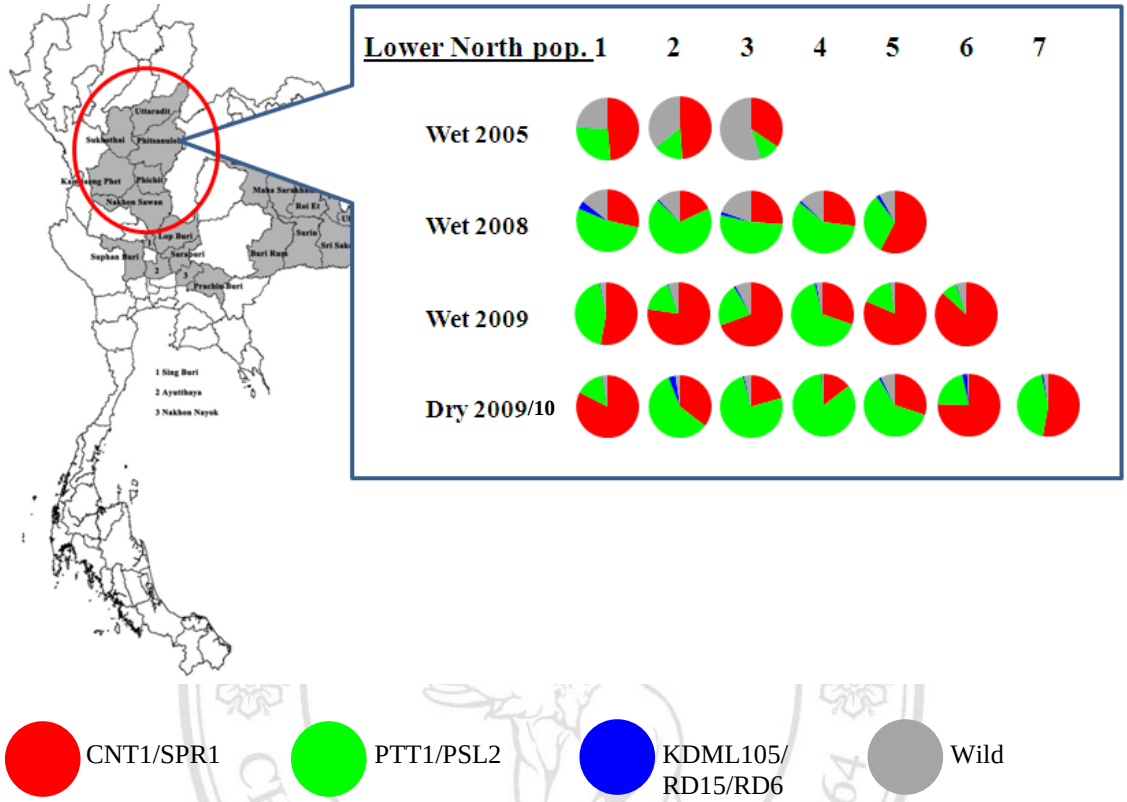
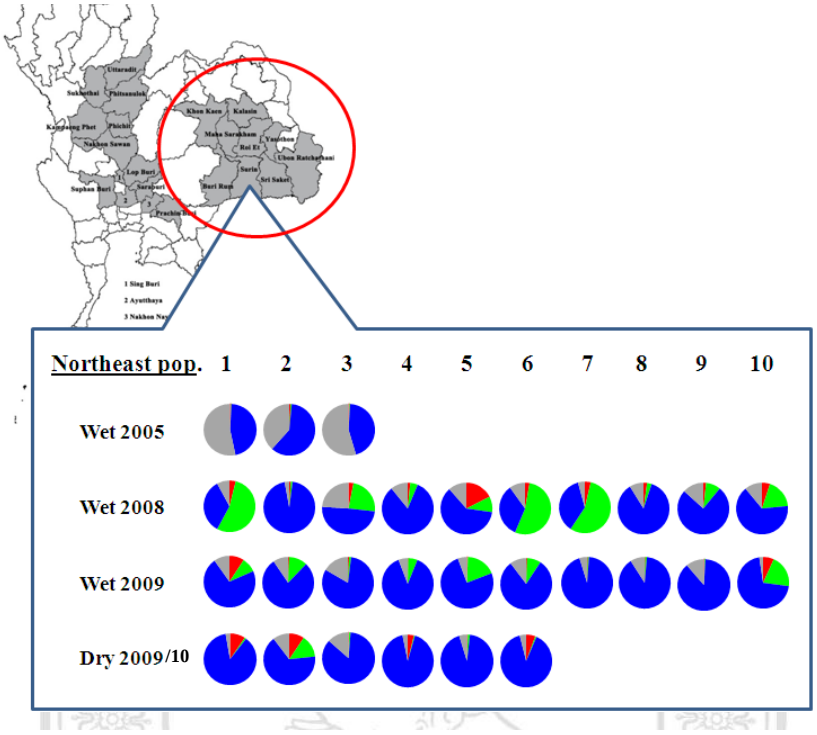


Figure 2.3.19 Genetic proportion of 63 weedy rice populations collected from three rice production areas; Lower North (a), Northeast (b) and Central Plain (c) in different four growing seasons in wet 2005, 2008, 2009, dry 2009/10 compared with 7 pure line cultivated rice varieties and 7 common wild rice populations. Each color represent on STRUCTURE.

(b) Northeast



(c) Central Plain

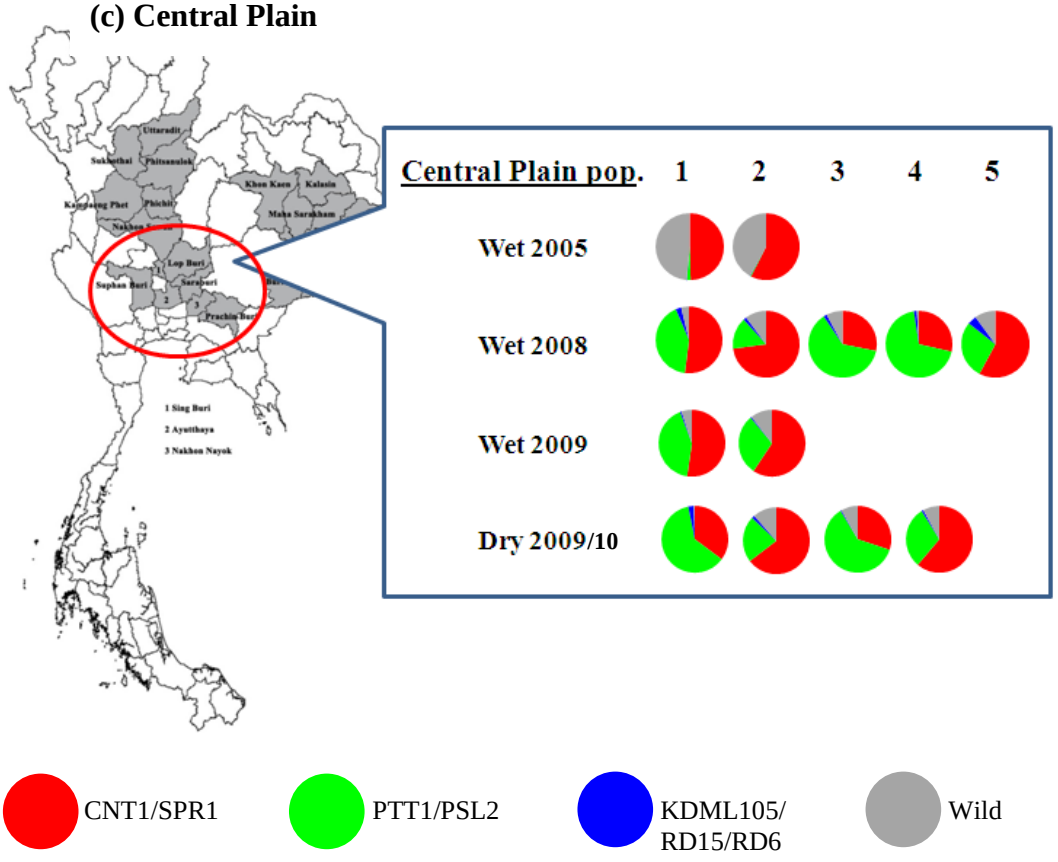
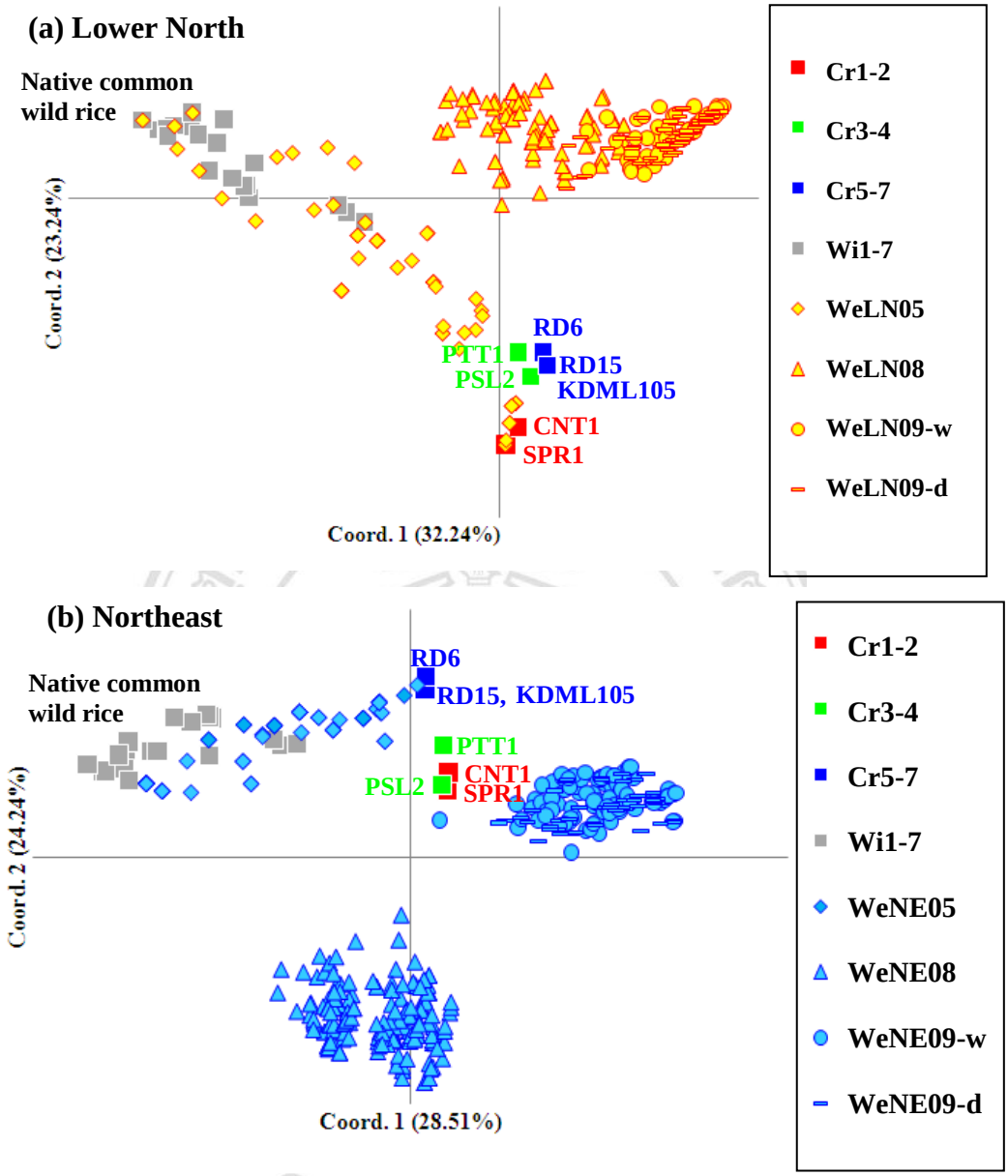
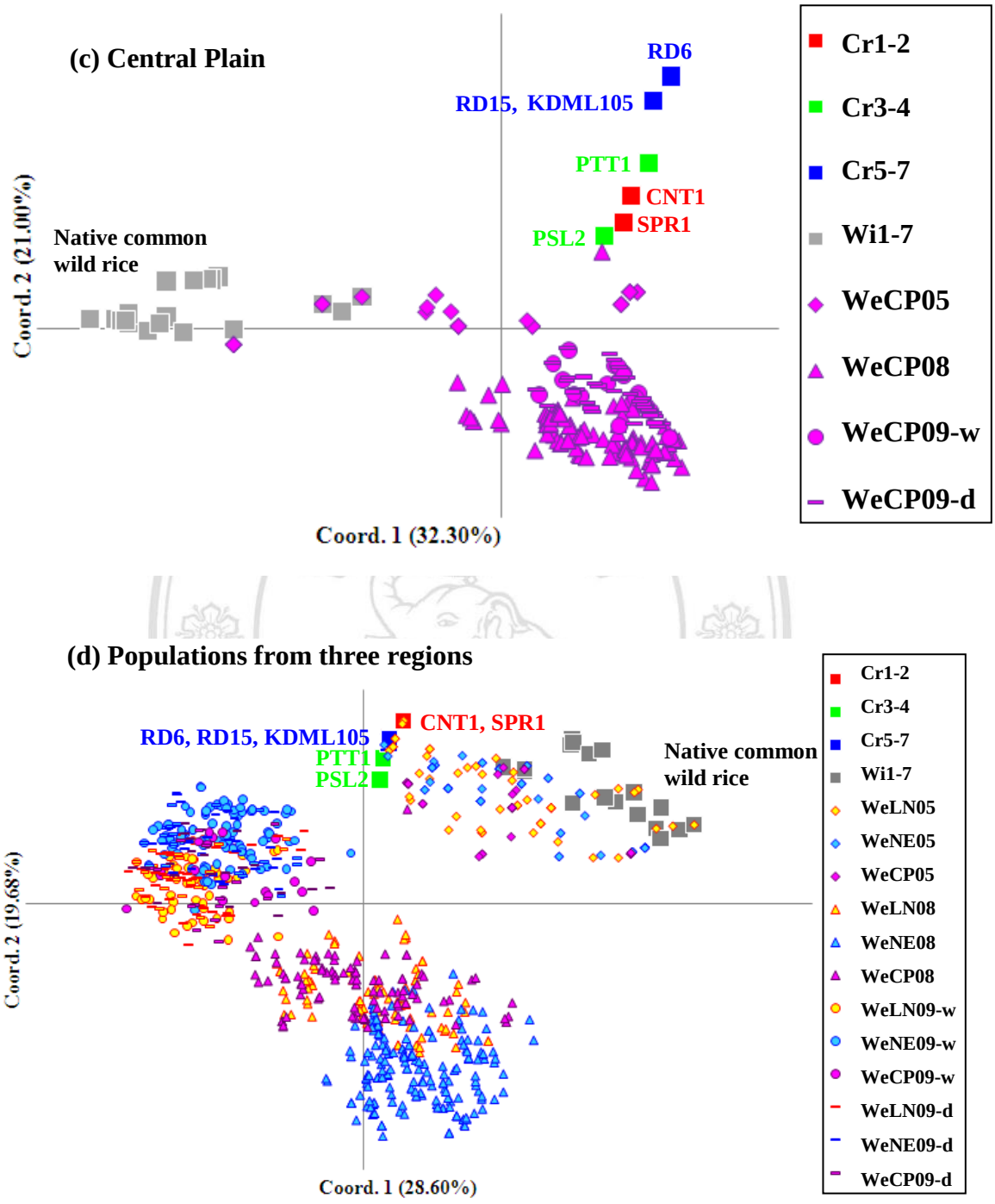


Figure 2.3.19 *(continued)*



Cr1-2= SPR1, CNT1 **Cr3-4 = PTT1, PSL2** **Cr5-7 = KDML105, RD15, RD6**

Figure 2.3.20 Principle component analysis (PCA) of 63 weedy rice populations collected from three rice production areas; Lower North (a), Northeast (b) and Central Plain (c) in different four growing seasons in wet 2005, 2008, 2009, dry 2009/10 and all four seasons (d) compared with 7 pure line cultivated rice varieties and 7 common wild rice populations. Different colors of weedy rice samples represent different regions.



Cr1-2= SPR1, CNT1 **Cr3-4 = PTT1, PSL2** **Cr5-7 = KDML105, RD15, RD6**

Figure 2.3.20 *(continued)*

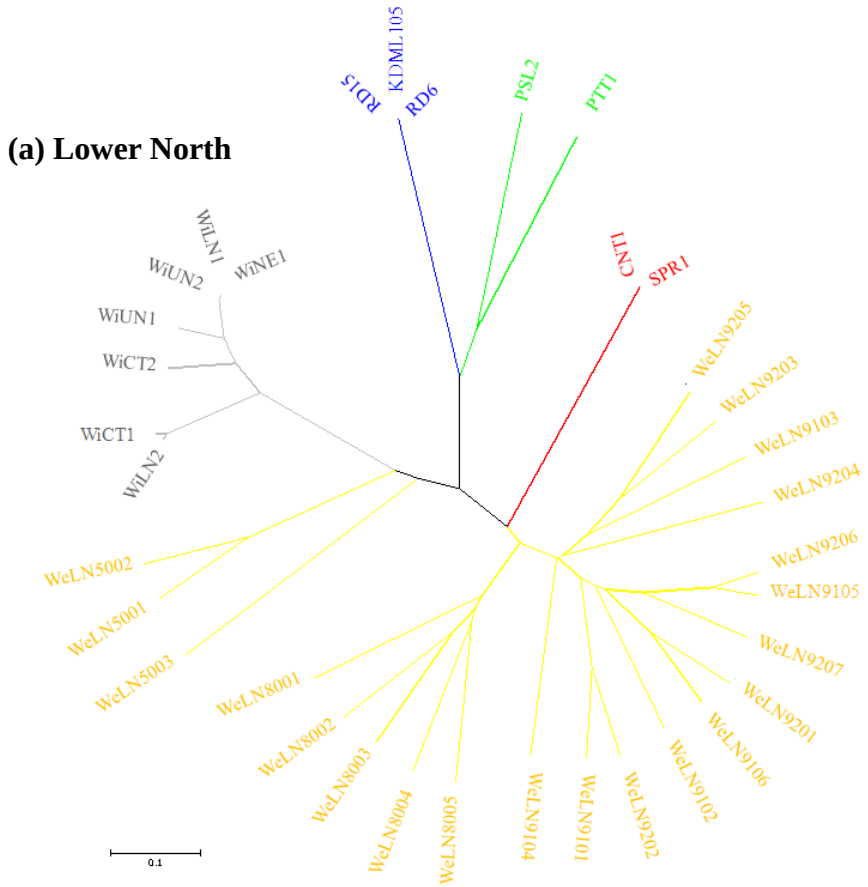
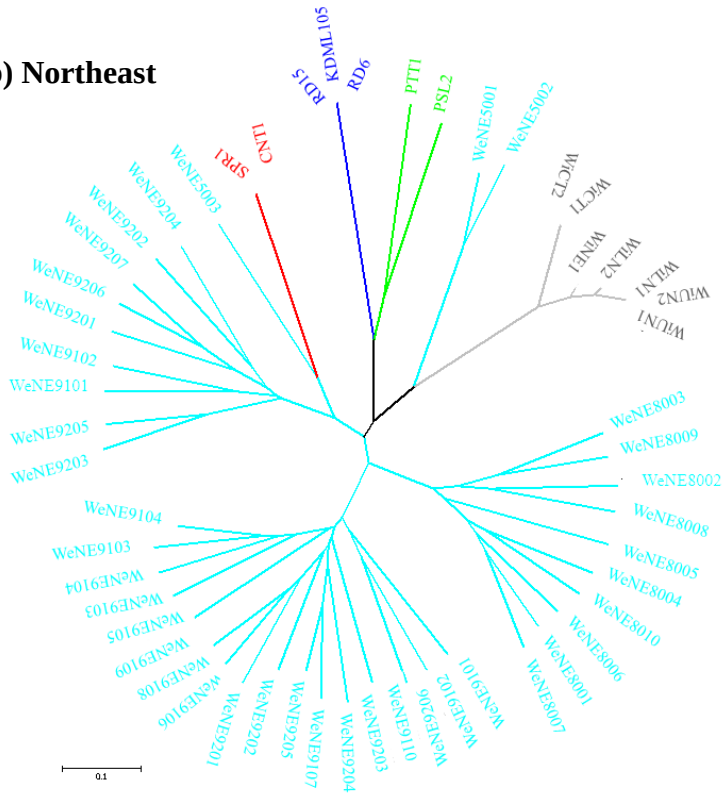


Figure 2.3.21 Cluster analysis using NJ method based on C.S. Chord (1967) genetic distance among 63 weedy rice populations collected from three rice production areas; Lower North (a), Northeast (b) and Central Plain (c) in different four growing seasons in wet 2005, 2008, 2009, dry 2009/10 and all four seasons (d) compared with 7 pure line cultivated rice varieties and 7 common wild rice populations. Different colors of weedy rice samples represent different regions; WeLN: yellow, WeNE: light blue, WeCP: pink, Cr1-2: red, Cr3-4: green, Cr5-7: blue and Wi: gray.

(b) Northeast



(c) Central Plain

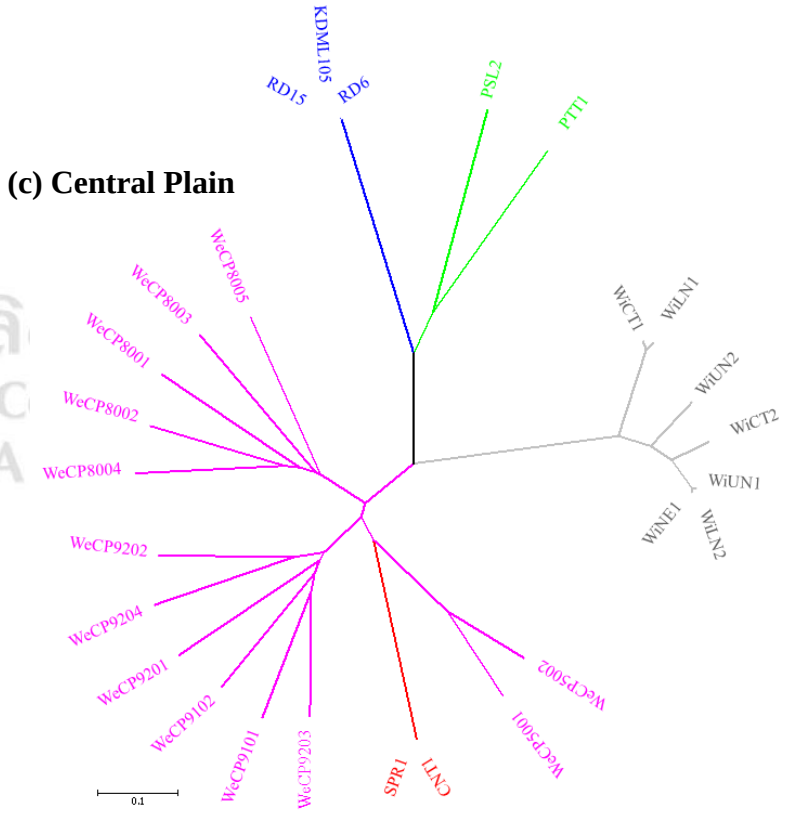


Figure 2.3.21 *(continued)*

(d) Populations three regions

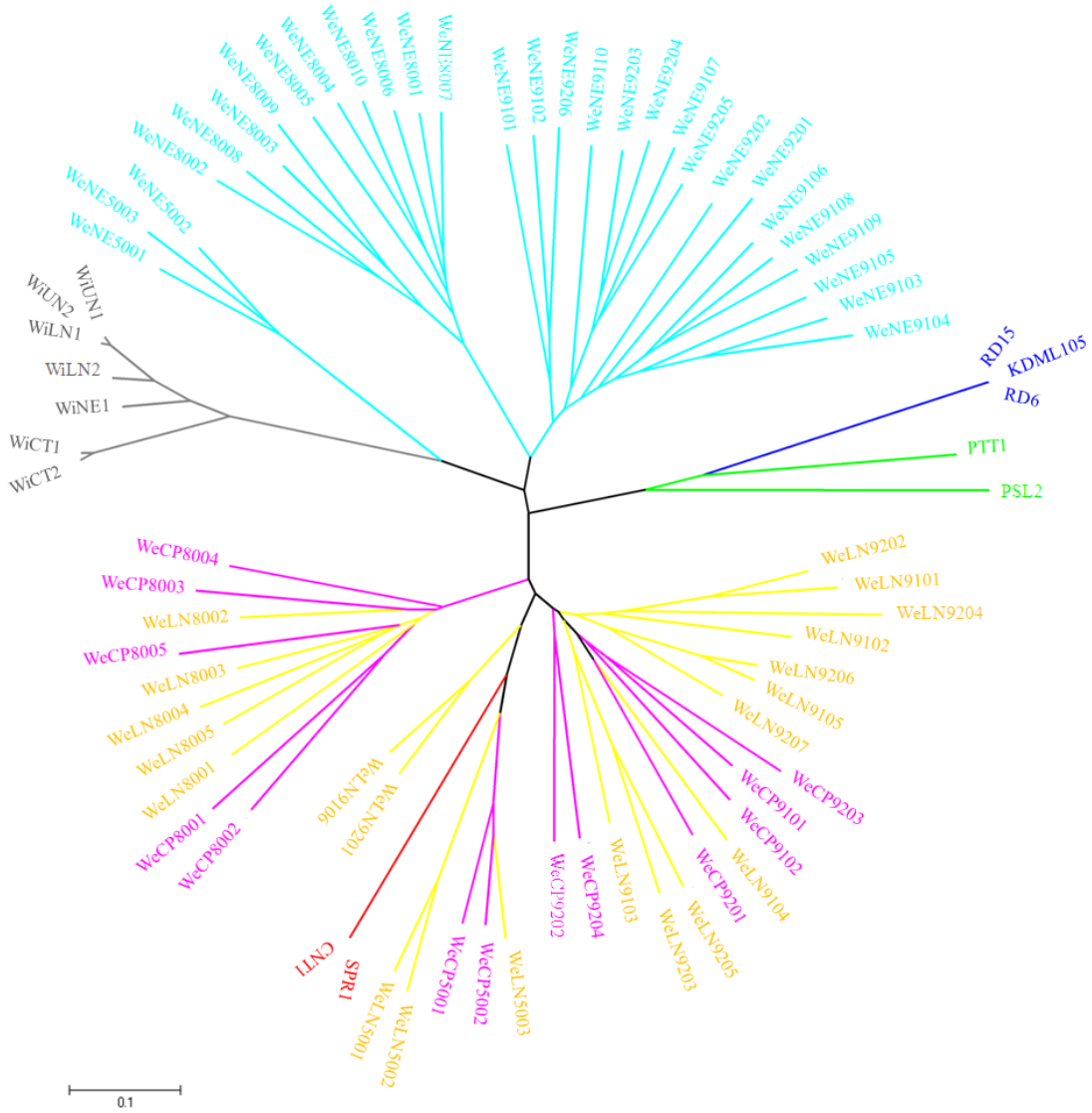


Figure 2.3.21 *(continued)*

2.4 Discussion

Genetic variation in morphological, physiological characters and microsatellite loci of weedy rice populations in Thailand

The present results based on genetic variation of morphological, physiological characters and microsatellite loci demonstrated that Thailand weedy rice populations possessed relatively high level of genetic variation, both within and between populations. Morphologically and physiologically, highly variation of intermediate characteristics between common wild rice and pure line rice checks were observed in weedy rice populations including, spreading to erect plant types, long awn to awnless on spikelet, dark gray to straw hull and present anthocyanin to none on the parts of leaf, stem, panicle and seed. This result is similar from that of previous studies by Niruntrayakul (2008) that wild and crop traits were mixed on many parts of weedy rice plants from Kanchanaburi province.

Likewise, Very high level of genetic diversity based on microsatellite loci was maintained, both within and between Thailand weedy rice populations. These were demonstrated by high levels of average gene diversity (h), total genetic diversity (H_T) and genetic differentiation among weedy rice populations (F_{ST}). Similar relatively high levels of genetic diversity for SSR markers were also detected in other studies of weedy rice in Thailand by Niruntrayakul (2007) and Pusadee (2009). While over all genetic diversity of weedy rice populations from both north-eastern Chaina (Cao *et al.,* 2006) and US (Londo and Schaal, 2007), where absent wild progenitor, exhibited very much lower levels comparing with weedy rice in the present study. The similarity in genetic variation between this study and previous reports are probably due to weedy rice in Thailand originates from interspecific hybridization between

wild progenitor and crop rice. This high level of the genetic variation contributed to their success in agroecosystems and infests a wide range of diverse habitats.

The results also showed that weedy rice in present study more produced yield components, especially in tillers plant^{-1}, panicle plant^{-1} and plant height, which contributed to high competitiveness of weedy rice in rice fields. Interestingly, most weedy rice plants in this study were earlier flowering than both common wild rice populations and seven pure line rice varieties and they were maintained high level of seed shattering when their mature as the same common wild rice traits. Vaughan *et al.*, (2005) confirmed that weedy rice showed high adaptation to the agronomic practices by earlier flowering and seed mature than cultivated rice and they shatter their seeds during the cultivated rice harvest. General, most weedy rice seeds with non-shattering are harvested together with the cultivated rice led to contaminate in seed stock. The remainder seeds are shattered and fall on the ground for escape the harvest and investing crop rice by weedy rice plants in the next crop (Maneechote *et al*., 2004).

Population genetic structure and dynamics of weedy rice populations in Thailand

The results showed that population of weedy rice in Thailand were highly structured by its companion crop rice varieties popular in each region. This is clearly reflected by the results from the genetic assignments from the STRUCTURE model, the distribution of weedy rice plants on PCA graph and the grouped weedy rice from the cluster analysis. The results from STRUCTURE model, PCA graph and the cluster analysis showed a relatively close genetic relationship of weedy rice populations from the Lower North and Central Plain of Thailand with modern rice

varieties while weedy rice populations from the Northeast showed a relatively close genetic relationship with improved traditional rice varieties. Consequently, significant differentiation among weedy rice populations from different regions confirmed the origin of weedy rice in Thailand. This result suggested that weedy rice in the present study resulted from independent hybridization between native common wild rice and cultivated rice varieties popular each region (Niruntrayakul, 2007; Pusadee, 2009). Pusadee (2009) also found similarly result that the population structure of weedy rice in Thailand were differentiated into 2 groups according to its companion cultivated rice varieties each region, the weedy rice of modern rice varieties fields was found in the Lower North and the Central Plain while the weedy rice of improved traditional varieties fields was found in the Upper North and the Northeast.

In addition, the results of genetic differentiation (F_{ST}) and analysis of molecular variance (AMOVA) demonstrated that major genetic variation existed among weedy rice populations from different regions. The observed differentiation of weedy rice populations from different regions is probably caused by genetic introgression from different cultivated rice varieties in different regions into weedy rice populations through time may increase variation among weedy rice populations from different regions. Weedy rice plants can easily hybridize with crop rice because those plants are always surrounded by rice cultivars in fields. Ellstrand *et al.* (1999) and Song *et al.* (2006) also reported the importance of introgression from crop species into weedy rice, which may have a substantial impact on differentiation and evolutionary processes in weedy populations.

In general, most of the weedy rice types are essentially adapt and mimic to cultivated rice for their highly successful as weeds in terms survival and distribution (Cao *et al*., 2006). In this study, the results based on morphological relationship, genetic diversity and population structure demonstrated that weedy rice populations temporally changed trend toward crop rice over the years. This may caused by accumulating continuous backcrossing of weedy rice to companion crop rice in the sympatric rice fields (Ellstrand *et al*., 1999). Such increasing morphological relationship and population structure with crop rice and decreasing genetic diversity of weedy rice indicated that weedy rice in Thailand showed high adaptation by mimicry to crop rice. They are difficult to recognize during the periodic weeding of the cultivars lead to increase their weediness (Ellstrand *et al*., 1999).

In conclusion, weedy rice populations in present study were diverse and changeable over time in morphological, physiological characters, genetic diversity and population structure. As considering among regions, weedy rice populations from different regions have differently structured base on cultivated rice varieties with popular in each region with high genetic differentiation. When considering among seasons, weedy rice populations were temporally changed. These dynamics of weedy rice has tended to parallel with cultivated rice for high adaptation to the agronomic practices by mimicry to cultivated rice led to the convergence of homogeneity of weedy population toward the crop rice. However, they were strongly some characters, produced more yield components and high seed shattering, for increasing their fitness and distribution. Therefore, the level of these adaptations by mimic to crop rice had tended to increase over years in each region for successful as weeds in terms survival and distribution when considering at the regional scale. Genetic

introgression and gene flow could play a major process in the long-evolution of weedy rice population in Thailand. The pattern of population structure and dynamics of wild-weedy-crop rice in sympatric habitats under gene flow process were evaluated at the field scale for their evolution in the next chapter.