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THESIS

GENETIC DIVERSITY OF THE MUNGBEAN (*Vigna radiata*, LEGUMINOSAE) GENEPOOL BASED ON MICROSATELLITE ANALYSIS

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A Thesis Submitted in Partial Fulfillment of
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A large representative collection of mungbean [*Vigna radiata* (L.) Wilczek] consisting of 415 cultivated, 189 wild and 11 intermediate accessions were analyzed using 19 SSR primers. These primers were developed from azuki bean [*V. angularis* (Willd.) Ohwi & Ohashi], and showed polymorphism in wild and cultivated mungbean. One or more SSR locus from each azuki linkage group was used. In total, 309 alleles were detected and of these about twice as many were detected in wild (257 alleles) compared to cultivated accessions (138 alleles). The number of alleles per primer ranged from 2 in CEDG174 to 37 in CEDG304 primers. The average diversity values for each locus was 0.59, ranging from 0.06 in CEDG269 to 0.92 in CEDG304. The results show that cultivated mungbean has its greatest diversity in South Asia, which supports the view that South Asia is the region where this crop was domesticated. SSR marker allelic diversity for cultivated mungbean has a distinct regional distribution with high variation in South and West Asia. Wild Australia and New Guinea represent a distinct center of diversity for wild mungbean. Based on Bayesian algorithm, the entire population was separated into two subgroups with largely belong to two subspecies. Each subspecies was further subdivided into three sub-subgroups. Wild mungbean has a rather clear geographical genetic structure, as compare to the cultivated mungbean. Based on the SSR marker diversity 106 accessions were selected for a useful core collection. This study represents the first comprehensive analysis of cultivated and wild mungbean germplasm diversity. It also highlights specific genetic diversity that might be used to broaden the genetic base of currently grown mungbean cultivars.

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Chontira Sangsiri
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LIST OF ABBREVIATIONS

µl	=	microliter(s)
AFLP	=	amplified fragment length polymorphisms
avg	=	average
bp	=	base pairs
CTAB	=	cetyltrimetyl ammonium bromide
dNTPs	=	deoxynucleotide triphosphate
ISSR	=	inter simple sequence repeats
MCMC	=	Monte Carlo Markov Chain
Ng	=	nanogram(s) (10^{-9}g)
PCR	=	polymerase chain reaction
RAPD	=	random amplified polymorphic DNA
RFLP	=	restriction fragment length polymorphisms
SNP	=	single nucleotide polymorphisms
SSR	=	simple sequence repeats
STS	=	sequence tagged site

GENETIC DIVERSITY OF THE MUNGBEAN (*Vigna radiata*, LEGUMINOSAE) GENEPOOL BASED ON MICROSATELLITE ANALYSIS

INTRODUCTION

Mungbean (*Vigna radiata* (L.) Wilczek) is an important grain legume in Asia. In China alone, it is grown on over 1 million ha and still increasing (Tomooka *et al*., 2005). Most mungbean cultivars are susceptible to diseases such as powdery mildew, mungbean yellow mosaic virus (MYMV), *Cercospora* leaf spot, and susceptible to pests such as bruchids, bean flies, and pod borers. Wild mungbean (*Vigna radiata* var. *sublobata*) is considered a useful source for resistance to mungbean pests and diseases. For example, resistance to bruchids, legume pod borers, and yellow mosaic virus have all been identified in wild mungbean (Singh and Ahuja, 1977; Singh and Emden 1979; Fujii and Miyazaki, 1987; Poonsavasde *et al*., 1996).

Mungbean belongs to the Asian *Vigna*, subgenus *Ceratotropis*. Taxonomically the subgenus *Ceratotropis* was divided into three sections *Ceratotropis*, *Angulares* and *Aconitifoliae*. Section *Ceratotropis* also includes the South Asian cultigen black gram [*V. mungo* (L.) Hepper]. The center of species diversity for section *Ceratotropis* is South Asia (Tomooka *et al*., 2002a).

The presumed progenitor of the cultivated mungbean is the wild form *Vigna radiata* var. *sublobata* (Roxb.) Verdcourt, which is widely distributed across the Old World tropics from West Africa to northern Australia and Papua New Guinea (Tomooka *et al*., 2002a). Archaebotanical finds and literary records suggest that mungbean was domesticated in India where wild mungbean is widely distributed (Smartt, 1990; Tomooka *et al*., 2002a). Archaeobotanical evidence points to both southeastern India between the Godavari and Krishna rivers and western Himalayan foothills as likely places where domestication could have started (Fuller and Harvey, 2006).

The conserved genetic resources of cultivated mungbean number several thousand accessions (Tomooka *et al*., 2002a). The largest collection is that of the World Vegetable Center (formerly the Asian Vegetable Research and Development Center, AVRDC) that has about 6000 accessions. More recent collections particularly by Australian and Japanese scientists have increased the number of wild mungbean accessions have in gene bank (var. *sublobata*) (Lawn and Watkinson, 2002; Tomooka *et al*., 2006a, 2006b; Vaughan *et al*., 2006). To use genetic resources of wild and cultivated germplasm efficiently for research and breeding, a core collection approach has been widely applied (Kojima *et al*., 2005). No molecular studies aiming at developing a mungbean core collection have been reported.

Despite the importance of mungbean, no genome map resolving the 11 linkage groups of this species has been published (Kaga *et al*., 2004). Two papers reported the development of SSR libraries in mungbean, however, these are not associated with linkage groups and the total number of polymorphic markers in mungbean reported was only 14 (Kumar *et al*., 2002a, 2002b; Gwag *et al*., 2006). Among other Asian *Vigna*, SSR marker libraries have been developed for azuki bean [*Vigna angularis* (Willd.) Ohwi & Ohashi] (Wang *et al*., 2004) and used to produce a well-saturated genome map (Han *et al*., 2005). Azuki SSR markers have also been used to produce a genome map of black gram [*V. mungo* (L.) Hepper] that has resolved the 11 linkage groups for this species (Chaitieng *et al*., 2006). Due to their close phylogenetic relationship, azuki bean SSR markers are also likely useful for detecting polymorphism in mungbean. A similar approach was applied in African *Vigna* of the cowpea, *Vigna unguiculata* group (Phansak *et al*., 2005).

In this study, azuki bean SSR primers were screened to determine their usefulness for analysing diversity in the mungbean genepool. Using azuki primers that revealed polymorphism in mungbean, a large collection of wild and cultivated mungbean were analysed. The goal of this study was to provide data useful to mungbean breeders and also to reveal insights into the evolution and dissemination of mungbean.

OBJECTIVES

1. To determine the genetic diversity and relationships within and among cultivated, wild and intermediate mungbean accessions.

2. To develop a core collection from this diverse germplasm as an aid to efficient evaluation of mungbean genetic resources.

LITERATURE REVIEW

1. Population genetics

Population genetics seeks to understand the causes of observable genetic variation in populations and to explain the underlying genetic basis for evolutionary change. It includes an empirical aspect, which measures and quantifies the genetic variation in populations, and a theoretical or statistical side, which attempts to explain the variation in terms of mathematical models of the forces that can change gene frequencies.

The genetic structure of a population is described by the total of all allele frequencies in the gene pool. In the case of diploid, or polyploid sexually interbreeding species, the genetic structure is also characterized by the distribution of alleles into genotypes. The genetic structure of a species can vary both geographically and temporally. The classical and neutral mutation models generate testable hypotheses and are used to explain how much genetic variation should exist within natural populations and what processes could be responsible for the observed variation. Mutation, genetic drift, migration, and natural selection are process that can alter the allelic frequencies of population (Russell, 2003).

2. Genetic diversity

Genetic diversity refers to the variation at the level of heritable characters (polymorphism) and provides a mechanism for populations to adapt to their ever changing environment. The more variation, the higher the chance that at least some of the individuals will have an allelic variant that is suited for the new environment, and will produce offspring with the variant and will in turn reproduce and continue the population into subsequent generations.

When a new allele appears in a population, it has the potential to change the genetic make-up of successive generations. Harmful mutations will likely not persist

because the affected individual will either not survive, or will have limited reproductive success. However, some mutations may be passed on to successive generations because an organism with that allele is better equipped to survive in its environment, that is, it has a selective advantage. Those individuals that produce a greater number of offspring that survive are said to be more fit. Other mutations may have no effect on phenotype, and may persist simply by chance (genetic drift). It is the selective advantage that drives evolution, albeit momentarily, in one direction or another (Russell, 2003).

3. Taxonomic Position and Gene Pools of Mungbean

Mungbean is in a group of agriculturally important, warm season and tropical legumes. It belongs to the genus *Vigna* that is very closely related to *Phaseolus*. The neotropical subgenus of *Vigna*, subgenus *Sigmoidotropis*, is a link between the two genera. The classified differences between *Vigna* and *Phaseolus* have been taxonomic. The close relationship between *Vigna* and *Phaseolus* has enabled comparative genome studies between these genera. Recently, a monograph has been published to discribe the 21 species in the subgenus *Ceratrotopis*, genus *Vigna* (Tomooka *et al*., 2005). Gene pools of mungbean are explained on Figure 1.

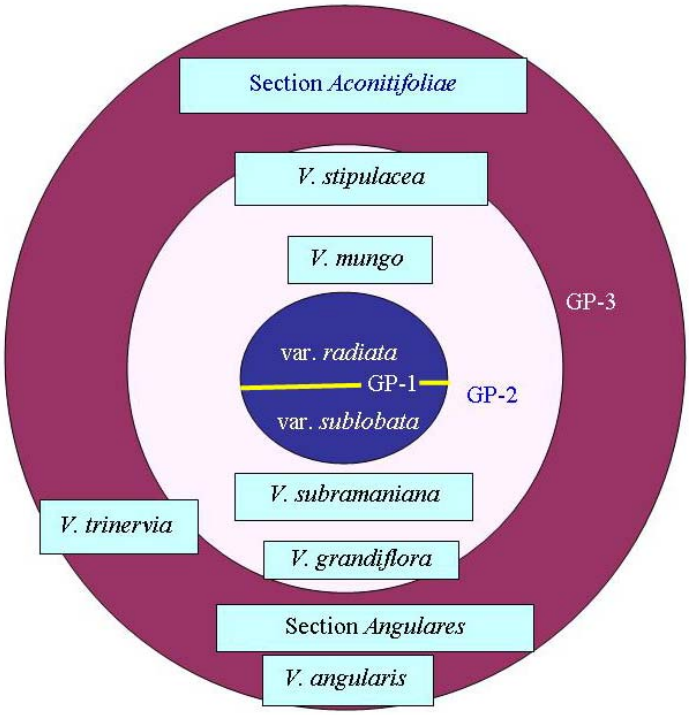

Figure 1 Gene pools of mungbean (*Vigna radiata*): Gene pool 1 (GP1), within which are the cultivated and wild forms of mungbean; Gene pool 2 (GP2), less related species that can be artificially hybridized with mungbean; Gene pool3 (GP3), includes species from which gene transfer to mungbean is impossible that requires *in vitro* embryo rescue, radiation-induced break chromosome and somaclonal fusion.

Source: Tomooka et al. (2005)

The genus *Vigna* is a large tropical genus consisting 82 described species distributed among 6 subgenera. The subgenera are *Ceratotropis*, *Haydonia*, *Lasiospron*, *Plectotropis*, *Sigmoidotropis* and *Vigna*. Within the subgenera in the genus *Vigna* especially subgenus *Ceratotopis* has it center of species diversity in Asia (Tomooka *et al*., 2006a).

The subgenus *Ceratotropis* consists of 21 species of which eight are domesticated including Moth bean [*V. aconitifolia* (Jacq.) Maréchal], Azuki bean [*V. angularis* (Willd.) Ohwi& Ohashi], Black gram [*V. mungo* (L.) Hepper], Mungbean [*V. radiata* (L.) Wilczek], Creole bean [*V. reflexo-pilosa* Hayata var. *glabra*) (Maréchal, Mascherpa & Stainer) N. Tomooka & Maxted], Jungli bean [*V. trilobata* (L.) Verdc], Toapée (Thai) [*V. trinervia* (Heyne ex Wall) Tateishi & Maxted], and Rice bean [*V. umbellata* (Thunb.) Ohwi& Ohashi] (Tomooka *et al*., 2002a). The most important domesticated species are mothbean, azuki bean, black gram and mungbean (Tomooka *et al*., 2006a)

The taxonomy of Asian *Vigna* was described based on several traits, such as seedling characteristics, size of floral parts, and habitat. Three groups within *Ceratotropis* have been recognized as sections, section *Angulares* (Azuki bean group), section *Ceratotropis* (Mungbean group) and section *Aconitifoliae* (Intermediate between azuki and mungbean group).

The characters were taxonomically informative in the subgenus *Ceratotropis* with a high degree of speciation. The species of the Subgenus *Ceratotropis* have flowers colored various shades of yellow, but are never purple, violet, blue or white as is often found in other *Vigna* subgenera (Baudoin and Maréchal, 1988). Tomooka *et al*. (2002b) summarized taxonomic and genetic studies of the subgenus *Ceratotropis* reveal an evolutionary trend from small flowered species with absence of standard appendage and defectively developed keel pocket and style beak in section *Aconitifoliae* to the large flowered species with distinctly developed keel pocket and long style beak in section *Angulares*.

Genetic analyses show that interspecies divergence in section *Aconitifoliae* is greatest. The comparison of genomic DNA sequence data between subgenus *Ceratotropis* and *Vigna* suggests section *Aconitifoliae* is the ancestral section in subgenus *Ceratrotropis*. Species in the section *Angulares* are least diverged and probably derived from species in section *Aconitifoliae* via section *Ceratrotropis*. Section *Ceratrotropis* is intermediate both morphologically and in terms of inter-species diversity and has two distinct phylogenetic lineages each containing one cultigen, *V. radiata* and *V.mungo*, and one wild species.

Molecular analyses and taxonomic studies clearly grouped mungbean with a small group of species, *V. mungo*, *V. subramaniana*, with their diversity centered in South Asia, but also occurring in Southeast Asia. All these species share several morphological traits, such as epigeal germination and first and second leaves being narrowly elliptic to ovate and lacking a petiole. This group of species in the subgenus *Ceratotropis*, was given the section name *Ceratotropis* by Tomooka *et al*. (2002a).

The close relationship between mungbean (*V. radiata*) and black gram (*V. mungo*) has contributed to the confusion surrounding their presumed progenitors, both of which occur in India and which have morphological similarities. Even so, broader stipules, pale yellow flowers, more ovules per pod, a spreading pod with short brown hairs, and a non arillate hilum, as well as chemical and molecular characters, distinguish cultivated and wild mungbean from cultivated and wild black gram.

4. Diversity of Gene Pool and Origin of Mungbean

The wild mungbean (*Vigna radiata* var. *sublobata*) is widely distributed from tropical Africa, through West, South and Southeast Asia to Papua New Guinea and Australia. India is considered the center of diversity of the wild form. Weedy forms of mungbean are also reported from India. In India, wild mungbean is found in hilly tracks, specially in the northern *terai* region and Western Ghats (Tomooka *et al*., 2005). Artificial crossing between *V. radiata* var. *radiata* (cultivate) and var. *sublobata* (wild) is not difficult (James *et al*., 1999), actually natural crossing

where cultivated and wild mungbean are sympatric might be considered. The flowers of cultivated and wild mungbean are similar although cultivated mungbean has larger flowers size ~14 mm than wild mungbean (~12 mm) (Tomooka *et al*., 2006a).

The ancient presence of wild mungbean in Australia is indicated by its ecotype differentiation and use as an aboriginal food. The distribution, ecology and plant types of wild mungbean in Australia and nearby countries have been described in detail. A distinctive form with deeply lobed leaflets is reported from the clay soils of Central Queensland. Besides, a perennial form with thick fleshy roots that has not been seen in other regions was found in northeastern tropical Australia. Wild mungbean in Australia has a twining, more gracile habit compared to taxa from other regions. In addition, Australian accessions of wild mungbean, while varying in seed size and pod color, lack "weedy characteristics" such as green testa color and bushy plant type found in Asia (Lawn and Wakinson, 2002; Lambrides and Goodwin, 2007).

There have been many studies of genetic diversity in Indian wild and landraces of mungbean (Tomooka *et al*., 1992; Lakhanpaul *et al*., 2000; Raje and Rao, 2001; Bisht *et al*., 2005). Tomooka *et al*. (1992) studied landrace collections of germplasm covering much of the distribution of mungbean in Asia using protein banding and plant growth types. The results revealed support for the view of Vavilov (1926) who pointed to Western Asia, viz. Afghanistan, Iran and Iraq, as being an important gene center for the crop, and India a likely place of origin. The report showed primitive forms and diverse protein-type genotypes for mungbean in Afghanistan, Iran and Iraq which may be indicative of a lack of selection there, since that seems to be the case with other crops in the region.

Indian mungbean landraces have the most diverse protein and growth types. India also has many populations of wild and weedy mungbean, and remains of mungbean at Indian archaeological sites have been found (Fuller and Harvey, 2006). The hypothesis has been proposed that mungbean was domesticated in India (Verdcourt, 1970). Tomooka *et al*. (2005) speculated on the route of distribution of mungbean. From India, mungbean might have spread west to the Afghanistan-Iran-

Iraq region and east to Southeast Asia in early times. In the Afghanistan-Iran-Iraq region, mungbean landraces are considered primitive due to the small seeds of various colors, often mixed, plants have many lateral branches, and they retain diverse protein type. Although, Southeast Asian mungbean landraces actually have large shiny green seeds, plants are tall with thick main stems, are late maturing, and protein types are simple. It means in Southeast Asia, mungbean has probably been under high selection pressure from farmers. In addition, in East Asian countries, mungbean landraces include growth type diversity that is intermediate between Afghanistan-Iran-Iraq and Southeast Asian types.

5. Molecular Studies of Genetic Diversity in *Vigna* Species

Genetic marker can be divided into three general groups; morphological, cytological, and molecular (Taji *et al*., 2002). DNA markers can be divided into two general categories, depending on whether they are based on restriction fragment length polymorphisms (RFLP) or on polymerase chain reaction. Molecular markers or DNA markers have becomes important in the genetic characterization and improvement of many crop species. They have been used to identify the genetic region or different alleles of loci on chromosomes. They have contributed to and greatly expanded the assessment of biodiversity and understanding of phylogenetic relations. PCR-based markers can be further sub-divided into two groups, viz. dominant marker and co-dominant markers. Dominant markers include amplified fragment length polymorphisms (AFLP) and random amplified polymorphic DNA (RAPD). Co-dominant markers include simple sequence repeats (SSR), single nucleotide polymorphisms (SNP), and insertion deletions (InDel).

The genetic diversity of several *Vigna* species has been studied using different molecular marker techniques. The specific techniques that have been widely used to characterize and study the genetic relationships within subgenus *Ceratotropis* including AFLP (Yee *et al*., 1999; Yoon *et al*., 2000; Xu *et al*., 2000a; Zong *et al*., 2002; Tomooka *et al*., 2002b; Saravanakumar *et al*., 2004; Seehalak *et al*., 2006), RAPD (Tomooka *et al*., 1995; Kaga *et al*., 1996; Yee *et al*., 1999; Mimura *et al*.,

2000; Xu *et al*., 2000b; Afzal *et al*., 2004; Ba *et al*., 2004; Betal *et al*., 2004), RFLP (Fatokun *et al*., 1993), ribosomal and chloroplast DNA (Doi *et al*., 2002).

5.1 Hybridization-Based Markers

RFLP has been commonly used as informative molecular marker in mapping of crop plant genomes. RFLPs are useful markers because of their co-dominant properties, and thus can distinguish between homozygosity and heterozygosity. RFLP analysis utilizes the difference in nucleotide sequences at specific sites recognized and cut by restriction Enzymes, and then separated according to size under electrophoresis. Individual DNA fragments are identified by labeled probes specific to certain sequences. The presence of particular alleles at these loci is detected by length polymorphisms caused by mutations that have led to loss or gain of a restriction site between genotypes. However, a limitation of RFLP is that it requires a large amount of good quality DNA for analysis. This technique is also time-consuming and expensive, making it less suitable for large-scale screening programs in plant breeding (Taji *et al*., 2002).

5.2 PCR-Based Markers

PCR-based markers use amplification of target DNA sequences by the polymerase chain reaction (PCR) *in vitro*. The basic principles of using PCR in marker systems is similar to RFLP in that it identifies polymorphisms from simple mutations, insertions or deletion in DNA sequence. PCR based markers have advantages over RFLP as the assay requires comparatively little DNA. It can generate a large number of polymorphic markers quickly without the need to develop libraries. Genetic markers based on PCR include, amplified fragment length polymorphisms (AFLP), inter simple sequence repeats (ISSR), random amplified polymorphic DNA (RAPD), sequence tagged sites (STS), simple sequence repeats (SSR), and single nucleotide polymorphisms (SNP). The cost of generating such markers is moderate. However, with the advances in technology and with international collaboration, the

cost has been reduced and greater numbers of such markers have become available to breeders.

Amplified Fragment Length Polymorphisms (AFLP) is a sequence-arbitrary method which amplifies of DNA fragments generated by specific restriction enzymes and oligonucleotide adapters containing few variable nucleotide bases (Vos *et al*., 1995). The method was developed from RFLP combined with RAPD techniques. In this technique, genomic DNA is first digested with one or two restriction endonuclease. Next, an adapter of known sequence is ligated to the ends of the digested genomic DNA. Amplification is carried out using primers with sequence specificity for the adapter. The primer(s) also contains one or more base at their 3' ends which provide amplification selectivity by limiting the number of perfect sequence matches between the primer and pool of available adapter/DNA templates. The resulting amplification products are typically observed by limiting the number of primers concentrations, followed by fragment separation on acrylamide gels. The strengths of this method are the very high multiplex ratio and genotyping throughput while no marker development work is needed. However, AFLP primer screening is often necessary to identify optimal primer specificities and combinations. Then it can be utilized in DNA fingerprint, genetic mapping, and gene tagging (Powell *et al*., 1996; Mohan *et al*., 1997; Cato *et al*., 1999; Tar'an *et al*., 2002; Kelly *et al*., 2003; Peter *et al*., 2004).

Random amplified polymorphic DNA (RAPD) was the first arbitrarily primed PCR markers methodology developed (Welsh and McClelland, 1990) and is still widely used. A number of modifications have been made to the technique, predominantly in primer length (8-12 bp) and detection methodology. RAPD markers are advantageous, since they are easy to generate, rapid, multilocus and do not require radioactivity. This PCR-based technique requires arbitrary short oligonucleotide primers targeting unknown sequences in the genome, usually resulting in presence/absence polymorphism. However, questions have arisen regarding their reliability.

Microsatellites are simple sequence repeats (SSRs) of 2-6 nucleotides. Microsatellites have been detected within the genomes of every eukaryotes so far analyzed, and are often found at frequencies much higher than would be predicted purely on the basis of base composition, although the frequency of microsatellites varies between species. They are abundant, dispersed throughout the genome and show higher level of polymorphism than most other genetic markers. These features, coupled with their ease of detection, have made them useful molecular markers. Their potential for automation and their inheritance in a co-dominant manner are additional advantages when compared with other types of molecular markers (Goldstein and Schlötterer, 2001; Holton, 2001). However, SSR requires sequence information and are relatively expensive to develop.

SSR markers have been developed for plant genomes, including common bean (Blair *et al*., 2003), azuki bean (Wang *et al*., 2004) and mungbean (Kumar *et al*., 2002a, 2002b; Gwag *et al*., 2006). Microsatellite analysis has bean used to study genetic diversity in various legume species including mungbean (Yu *et al*., 1999; Kumar *et al*., 2002a, 2002b; Gwag *et al*., 2006), yardlong bean (Phansak *et al*., 2005), cowpea (Li *et al*., 2001).

Very few SSR markers have been developed for mungbean and thus there is no genome map that has resolved the 11 linkage groups (Kaga *et al*., 2004). SSR marker libraries have been developed for azuki bean (*V. angularis*) (Wang *et al*., 2004) and these have been used to produce a well-saturated genome map for azuki bean (Han *et al*., 2005). Azuki SSR markers have also been used to produce a genome map of black gram (*V. mungo*) that has resolved the 11 linkage groups for this species (Chaitieng *et al*., 2006). Due to their phylogenetic relationship, azuki bean SSR markers are likely to be useful for detecting polymorphism in mungbean.

6. Genetic data analysis

6.1 The parameters used in explaining genetic diversity

Typically, the parameters made to indicate genetic diversity within a population can be estimated based on the change in allele frequencies as followed as:

6.1.1 Number of alleles per locus

Count the number of alleles at each locus in each sample and overall samples. For the average number of alleles per locus, the total numbers of alleles were divided by total numbers of locus.

6.1.2 Determination of allele frequencies

The allele frequencies in each sample and overall average can be estimated from banding patterns created by molecular markers. The overall allele frequencies can be presented either by weighted by sample size or non-weighted frequencies.

6.1.3 Allelic richness (estimate allelic richness per locus and sample (R_s))

The genetic diversity was measured based on allelic richness, which is considered important in the field of conservation genetics, and marker-assisted methods to effectively maximize the number of alleles conserved.

Allelic richness is a measure of the number of alleles independent of sample size, hence allowing comparison of this quantity between different sample sizes. El Mousadik and Petit (1996) proposed to estimate the number of alleles expected in a sample of specified size using rarefaction. The method begins with chosing the sample size of reference to be the number of genes examined in a smaller sample of specified size. The principle is to estimate the expected number of alleles in a sub-

sample of 2n gene, given that 2N genes have been sampled. n is fixed as the smallest number of individuals carrying a locus in a sample. Allelelic richness is then a calculated as:

$$R_s = \sum_{i=1}^{n_{ij}} \left(1 - \frac{\binom{2N - N_i}{2n}}{\binom{2N}{2n}} \right)$$

Where

N_i = the number of alleles of types i among the 2N genes.

n = number of individuals in smallest sampled population

N = the number of samples across all populations

6.1.4 Heterozygosity

Heterozygosity is a measure of heterozygote frequencies per locus. It refers to the fraction of loci within an individual that are heterozygous. It is normally used to refer to the population as a whole. Typically, the observed (H_o) and expected (H_e) heterozygosities are compared, defined as follows for diploid individuals in a population:

Observed heterozygosity

$$H_o = \frac{\sum_{i=1}^{n} (1 \text{ if } a_{i1} \neq a_{i2})}{n}$$

Where

n = the number of individuals in the population

a_{i1} and a_{i2} = the alleles of individual *i* at the target locus

Expected heterozygosity

$$H_e = 1 - \sum_{i=1}^{m} (f_i)^2$$

Where

m = the number of alleles at the target locus

f_i = the allele frequency of the i^{th} allele at the target locus

7. Genetic structure analysis

The measurement of genetic diversity were calculated within and between populations based on different genetic structure analysis. The measure values are follows as:

7.1 Linkage disequilibrium or Gametic phase disequilibrium is the non-random association of alleles at two or more loci, not necessarily on the same chromosome. Linkage disequilibrium explains a situation in which some combinations of alleles occur more or less frequently in a population than would be expected from a random recombination of haplotypes from alleles by their frequencies. Non-random associations between polymorphisms at different loci are measured by the degree of linkage disequilibrium (LD). It is a test of random mating and Mendelian segregation in which independent segregation allele from one genotype to another one (Hedrick, 2005).

7.2 Genetic distance is a measure of the dissimilarity of genetic material between different species or between individuals of the same species. It calculates the allelic substitutions per locus which have occurred during the separate evolution of two populations or species. The calculation of a genetic distance between two populations gives a relative estimation of the time that has passed since the populations have survived as single cohesive units (Nei, 1983).

7.3 F-coefficients are the study of genetic differentiation of populations. Wright (1951) proposed the quantities to measure the degree of relatedness of various pairs of alleles. Wright's F_{IT} is the overall inbreeding coefficient F which correlates of alleles with individual over populations, Wright' F_{IS} (*f*) is the correlation of alleles within individuals within one population and Wright' F_{ST} is the correlation of alleles of different individuals in the same population. F_{ST} is calculated to compare population. It is the correlation between two gametes drawn at random from each subpopulation and measures the degree of genetic differentiation of subpopulations, while F_{IT} and F_{IS} are the correlations between the two uniting gametes or alleles to produce the individuals relative to the total population and relative to the subpopulations, respectively. They are often called fixation index and can be negative, whereas F_{ST} is always positive. The formula used in calculating fixation index is as follow as:

$$1-F_{IT} = (1-F_{IS})\,(1-F_{ST})$$

Where F_{ST} is the measure of different allele frequencies between populations that view for study within population has been separated. For a set of r populations with sample allele frequencies $\tilde{p}_i$ (i = 1, 2, …, r) for some allele A, the statistic could be calculated as:

$$F_{ST} = \frac{\Sigma_i (\tilde{p}_i - \bar{p})^2/(r-1)}{\bar{p}(1-\bar{p})}$$

$$= \frac{s^2}{\bar{p}(1-\bar{p})}$$

Where

$\bar{p} = \Sigma i\ \tilde{p}_{i/r}$ is the average sample frequency of the allele over the sample and s^2 is the sample variance.

Or $$F_{ST} = \frac{\overline{H_T} - \overline{H_S}}{\overline{H_T}}$$

$$F_{IS} = \frac{\overline{H_S} - \overline{H_O}}{\overline{H_S}}$$

$$F_{IT} = \frac{\overline{H_T} - \overline{H_O}}{\overline{H_T}}$$

Where

$\overline{H_S}$ = the average of expected heterozygosity (*He*) of all loci within subpopulation

$\overline{H_T}$ = the average of *He* of all loci within overall population

$\overline{H_O}$ = the average of observed heterozygosity (*Ho*) of all loci within subpopulation

8. The cluster and classification techniques for genetic diversity

Clustering is the classification of objects into different groups, or to reduce the amount of data by categorizing or grouping similar data items together. Data clustering is a common technique for statistical data analysis which is used in many fields, including biological data mining, pattern recognition, image analysis and bioinformatics. Clustering methods can be divided into two types. There are distance-based methods, in which a pair-wise distance matrix is used as an input for analysis by a specific clustering algorithm, leading to a graphical representation, e.g., a dendrogram, in which clusters may be visually identified. Alternatively model-based methods, can be used in which observations from each cluster are assumed to be random draws from some parametric model, and inferences about parameters corresponding to each cluster and cluster membership of each individual are performed jointly using standard statistical methods such as maximum-likelihood or Bayesian methods (Mohammadi and Prasanna, 2003).

Prichard *et al*. (2000) revealed some of the forceful of distance based method and exposed an innovative model-based clustering method on the basis of Bayesian algorithm for inferring population structure using multilocus genotypic data consisting of unliked marker. The strength of which structured associated to approach lies in their effective analysis of population structure, accurate clustering and assignement of individuals into their appropriated populations, normally using a modest number of unlinked markers, and identification of migrants and admixed individuals. Using this approach, one can estimate the proportion of an individual's genome contributed by a specific subpopulation referred to as the genetic background matrix (Q).

9. Multivariate analysis

For increasing in the sample sizes of breeding materials and germplasm accessions used in crop improvement, methods to classify and order genetic variability are assuming considerable significance. The use of established multivariate statistic is an important strategy for a large number of accessions, or analyzing genetic relationships among populations. Multivariate method analyses multiple measurements on each individual under investigation. They can be used in analysis of genetic diversity irrespective of the dataset, for example, morphological, biochemical, or molecular marker data. Among these algorithms, cluster analysis, principal component analysis (PCA), principal coordinate analysis (PCoA), and multidimensional scaling (MDS) are most commonly employed and appear particularly useful (Johns *et al*., 1997; Thompson *et al*., 1998; Brown-Guedira *et al*., 2000).

PCoA is a scaling or ordination method which starts with a matrix of similarities or dissimilarities between a set of individuals and aims to produce a low-dimensional graphical plot of the data in such a way that distances between points in the plot are close to original dissimilarities. Thus the starting point matrix of similarities or dissimilarities for PCoA is different from PCA, which starts with the initial data matrix, for example, presence or absence of alleles in molecular marker data. PCA can be utilized to derive a 2- or 3- dimensional scatter plot of individuals.

The first PC summarizes most of the variability present in the original data relative to all remaining PCs. The second axis explains most of the variability not summarized by the first axis and uncorrelated with first and so on. The axes are of PC plots orthogonal and thus independent of each other. Each axis reveals different properties of the original data and may be interpreted independently. Thus, the total variation in the original data set may be broken down into components which are cumulative. The proportion of variation counted for by each PC is explained as the eigenvalues divided by the sum of eigenvalues. The eigenvector defines the relation of the PC axes to the original data axes (Mohammadi and Prasanna, 2003).

10. Core collection

The concept of a core collection is to obtain a small number of accessions representative a large population of the germplasm so that it can be studied and used more efficiently. It would be less productive to have a core so large that it suffers the same problems as the whole collection. On the other hand, a core that failed to contain a significant fraction of the whole collection's diversity would not serve its purpose either. Its definition extends to a collection that includes a group of related species, or to the one that is the aggregate of several collections of the same taxa held in a network of cooperating genebanks.

The original definition of a core collection is a limited set of accessions representing, with a minimum of repetitiveness, the genetic diversity of crop species and its wild relatives (Frankel and Brown, 1984). In addition, two operational definitions are for an individual genebank and for a whole crop species. The definition for an individual genebank is that a core collection consists of a limited number of the accessions in an existing collection, chosen to represent the genetic spectrum or diversity as much as possible (Brown, 1995). For a whole crop species, a core collection consists of a limited number of entries chosen to represent the genetic diversity of the whole crop species and its wild relatives. It is a synthetic and comprehensive core collection, assembled cooperatively by national and international

genebanks and supplemented with fresh samples of wild or crop populations where needed to fill gaps.

A general procedure for the selection of a core collection can be divided into five steps. These are as follows; (1) identify the material (collection) that will be represented, (2) decide on the size of the core collection, (3) divide the set of material used into distinct groups, (4) decide on the number of entries per group, and (5) choose the entries from each group that will be included in the core. Each of these steps can be more or less complex, depending on the information available and the procedures used, as exemplified in Figure 2 (van Hodgkin *et al*., 1995).

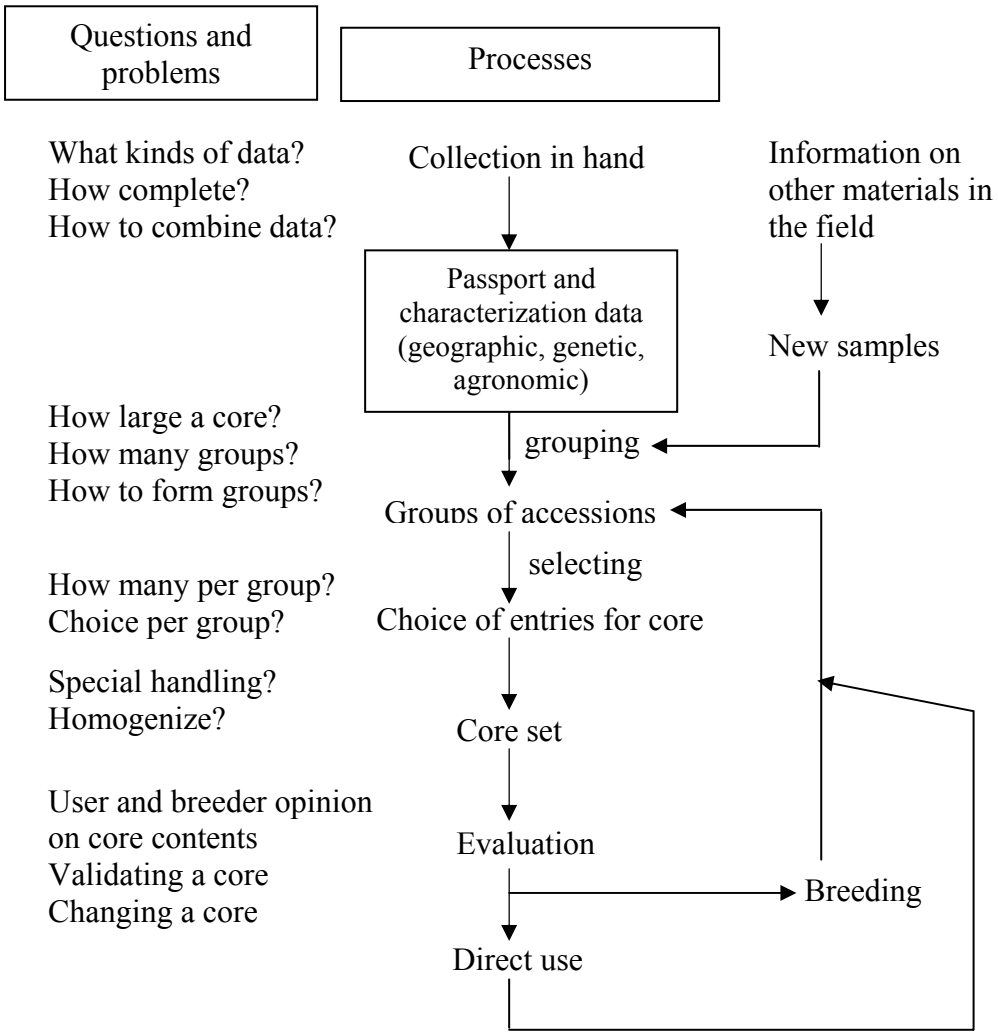

Figure 2 Flow chart illustrating steps in developing a core collection.

Source: van Hodgkin et al. (1995)

MATERIALS AND METHODS

1. Plant materials

In this study, 615 accessions were used from representative population. Among them, 415 accessions were cultivated mungbean, 189 accessions were wild mungbean and 11 accessions were classified as intermediate between wild and cultivated mungbean (Table 1, Appendix Figure 1, 2 and 3). The plants were sampled from 29 countries in three continents, viz. Africa, Asia and Australia as shown in Figure 3.

2. DNA Extraction

Genomic DNA was extracted from 200-300mg of young leaves on a single plant per accession with the EZ-1 kit (QIAGEN, Valencia, CA) or modified CTAB method (Puchooa, 2004). DNA concentration was adjusted to 10 ng μL^{-1} as determined on 1% agarose gel by comparing bands with 10 ng μL^{-1} of λDNA.

3. PCR Amplification

78 SSR primer pairs developed from azuki bean were tested for PCR amplification and their usefulness to detect polymorphism by screening them on one cultivated (JP1648) and three wild mungbean (JP107876, JP217427, JP81648) accessions. From the results 19 primers (Table 1) located on all 11 linkage groups of azuki bean were selected for use with all the mungbean accessions (Table 2).

The 5' end of the reverse primers in each set were labeled with one of the four fluorescent dyes, 5-FAM, VIC, NED or PET (Applied Biosystems). Six multiplex sets of PCR reactions per sample were set up for the amplification.

DNA amplification was carried out on the GeneAmp PCR System 9700 (Applied Biosystems). PCR reaction were performed in a 10 μL volume containing

10 ng of genomic DNA, 1x KOD-plus PCR buffer, 1 mM $MgSO_4$, 0.2 mM dNTPs, 1U KOD-plus (*Thermococcus kodakaraensis*, KOD strain) DNA polymerase (TOYOBO, Japan) and 5 *p*mol of forward and fluorescent labeled reverse primers loaded in a standard 96-well plate format. The cycles were programmed as follows: 2 min at 94^{o}C followed by 35 cycles of denaturing for 15s at 94^{o}C, annealing for 15s at 55^{o}C and extension for 15s at 68^{o}C and finally maintained products at 4^{o}C. PCR product was diluted by MQ water 10 times as necessary to prevent off-scale fluorescent signals.

4. Genotyping

A volume of 1μL of PCR product or 1/10 diluted PCR product was mixed with 9 μL of Hi-Di formamide containing 0.1 μL of GeneScan 500 LIZ size standards (Applied Biosystems). The mixture was denatured at 95^{o}C for 5 min then placed on ice immediately. The denatured products were run on an automated capillary DNA sequencer (ABI Prism3100 Genetic analyser). Size of SSR fragment was determined with Gene Mapper ver. 3.0 (Applied Biosystems).

5. Data analysis and core collection selection

5.1 Intra population variation. Genetic variability of each population was measured as the number of alleles per locus (*A*), gene diversity, expected and observed heterozygosity (H_E and H_O), fixation index (*Fis*) and allelic richness. All parameters were calculated by FSTAT ver. 2.9.3.2 (Goudet, 2002). H_E is equal to H_O in random mating populations. Fixation index (Wright, 1965) shows deviation from Hardy-Weinberg expectation. The testing to evaluate either excess or deficit in heterozygotes was computed using FSTAT software.

5.2 Outcrossing rate (*t*) was calculated from the fixation index using the equation $t = (1-Fis)/(1+Fis)$ (Weir, 1996).

5.3 Diversity values for each locus (Heterozygosity) were calculated using Nei's genetic diversity index according to the formula H =1-$\sum P^2_{ij}$ where P_{ij} is the frequency of the *j* th alleles for *i* th loci.

5.4 The D_A genetic distance (Nei *et al*., 1983) for all possible pairs of wild, intermediate and cultivated mungbean samples were calculated using POPULATIONS ver. 1.2.28 (Langella *et al*., 2001). The D_A distance-based method was used to calculate likelihood values for each individual belonging to the sample population. This distance method does not require Hardy-Weinberg equilibrium or absence of linkage disequilibrium among loci and the D_A distance showed higher percentage of individuals assigned to the correct population than other distance methods such as standard genetic distance (D_S) and minimum genetic distance (D_m) (Cornuet *et al*., 1999).The equation of D_A is revealed as followed.

$$D_A = \frac{1}{r} \sum_{j=1}^{r} \left(1 - \sum_{j=1}^{mj} \sqrt{X_{ij} Y_{ij}}\right)$$

Where

Consider two populations, X and Y, and let X_{ij} and Y_{ij} be the frequencies of the i-th allele at the j-th locus in population X and Y, respectively.

m_j is the number of alleles at the j-th locus in population and r is the number of loci studied.

5.5 The Principal coordinate analysis (PCoA**)** was used to display genetic divergence among samples in a multidimensional space. The D_A distances computed among all samples were coordinated in two dimensions using NTSYSpc. 2.1.0 (Rohlf, 2001) based on D_A distance.

5.6 Core collection selection

A core collection was developed using the simulated annealing algorithm developed by Liu (2002) based on the maximum richness of SSR allelic data. Given

the complete set of lines (*L*), the algorithm works by first randomly selecting a subset of lines (*l*). Each line has a calculating based on the number of private alleles in the line. Adding new lines from the number of alleles is then evaluated and swap is accepted if increase number of lines and some probability same or less. The probability of acceptance is dependent on the level of decrease in allelic richness and on the iteration number such that probability is larger in earlier iteration. Swapping is continued for a predefined number of iterations. Probability gradually decreases with iterations (time). Under this approach, lines with more private mean of data set alleles have a larger probability to be included in the core set. The algorithm can also incorporate a weights suches the agronomic quality of the lines and can allow some lines to be designated as "conserved" such that they are automatically included in the core set (Liu *et al*., 2003).

5.7 Analysis of genetic structure

The accessions were subdivided into genetic clusters using a model-based approach with the software package STRUCTURE (Pritchard and Wen, 2007). The Bayesian clustering algorithm was implemented to identify clusters of genetically similar individuals and to test the proportion of genetic admixture among the clusters at the individual level. The algorithm identifies subpopulations and allocates individuals into group based on estimation of allele frequencies.

In this experiment, the data set was separated into 3 subgroups according to types (wild, intermediate and cultivate). One to six *K* was applied to infer the number of clusters for population. In each run, a Markov chain Monte Carlo (MCMC) method was used to estimate allele frequencies in each of *K* populations and the degree of admixture in each individual under the condition of 100,000 burn-in (process required to prepare for running MCMC) period and 500,000 MCMC steps and a model allowing for no admixture and correlated allele frequencies. Twenty replicated analysis were done. Lines with membership probabilities ≥ 0.8 were assigned to clusters (Liu *et al*., 2003). The number of clusters was identified from clear cluster group. Upon separationnof the entire population, two subspecies groups were clearly

seen (Figure 8 and Figure 9). Then the data were separated into each subspecies (wild and cultivated population) and calculated accoding Evano *et al*. (2005). This equation finally detected the number of true clusters in the sample of individuals when patterns of dispersal within populations are not homogeneous has not been tested. The correct estimation of the number of clusters using an ad hoc statistic based on the rate of change in the log of probability of data between successive cluster values.

Despite in this experiment, the reports have been revealed in the part of the number of the clusters at the highest of log likelihood (Prichard *et al*., 2000; Falush *et al*., 2003) and the highest of the second rate of change in the probability of data which modal value of the distribution of ΔK was used as an indicator of the strength of the signal detected by STRUCTURE as described by Evanno *et al*. (2005).

The details of equations were calculated by Bayesian algorithm. The first step was calculated by mean of likelihood of each K, after that we calculated first and second rates of change of the likelihood function with respect to K and the final step to analyze absolute ΔK (Evanno *et al*., 2005). In the last step, the wild and cultivated data set was run at K = 1 to 5 under the condition with 100,000 burn-in period and 500,000 MCMC replications for five times (Figure11 and Figure 17).

5.8 Geographical analysis of the population clusters

Each accession was located on map individual origins based on latitude and longitude by Google earth 4.3.7191 software (Google, 2009). However, for some accessions the original collection sites were unknown. Thus we used only known locations accessions, viz. 176 accesions of wild (93.12%) and 316 accessions of cultivated (76.14%) for locating on the world map. The maps for the wild accessions are in Figure 14-16 and for the cultivated are in Figure 20.

RESULTS AND DISCUSSION

Results

1. SSR primers

Of the 78 azuki SSR primers screened, 27 were found to be useful for detecting polymorphism in the test accessions JP107876, JP217427, JP218942 and JP81648. From these, 19 primers located on each of the 11 linkage groups of azuki bean were used to analyse the entire set of mungbean germplasm (Table 2).

The number of alleles detected among the 19 SSR primers ranged from two (CEDG174) to 37 (CEDG304) and the average genetic diversity per locus for all accessions was 0.59 in a range of 0.06 to 0.92. A sample SSR peak is shown in Figure 4. In total, 309 alleles were detected, with about twice as many detected in wild accessions (257 alleles) as in cultivated accessions (138 alleles), eventhough twice as many cultivated accessions were analyzed.

In all, 136 alleles are detected exclusively in wild germplasm and 33 only in cultivated germplasm. From the 19 primers used, 10 primers revealed more than 10 alleles in the wild germplasm analysed, and four of these revealed more than 20 alleles (Table 2). In addition, the allele frequency distribution of these 10 primers did not show a clear peak with respect to the number of accessions that had the same allele. In contrast to wild mungbean, the cultigen had fewer alleles per primer, with the exception of primer CEDG304 that produced 30 alleles in cultivated mungbean, but only 22 in wild mungbean.

In cultivated mungbean, 10 primers showed bimodal allele distribution and many accessions had the same allele, e.g. primers G108, G150 and G247 (Figure 4 and Appendix Figure 4).

2. Principal coordinate analysis (PCoA)

Results of the PCoA are shown in Figure 5 and Figure 6a–h. The first axis of the PCoA separates cultivated (Figure 6a–f) from wild (Figure 6g–h) accessions and explains 43.9% of the variation. The second axis primarily distinguishes among cultivated mungbean germplasm and accounts for 11.3% of the variation. Figure 6a–f show cultivated mungbean accessions from different geographic areas. This reveals that Central and East Asian accessions harbour less diversity. Other regions had broader diversity, particularly accessions from South Asia (Figure 6c and 6d). Five accessions from Africa are in the upper left of the PCoA plot and two (from Madagascar) are in the lower-left side (Figure 6f).

Intermediate accessions (Figure 6g) consisted of some accessions on the PCoA plot in a position similar to cultivated accessions. Other intermediate accessions are associated with wild accessions. Wild accessions (Figure 6h) showed no overlap with cultivated accessions, although wild accessions from India and Africa are closest to cultivated mungbean on the PCoA plot (Figure 5).

3. Gene diversity and genetic distance

The gene diversity of the cultivated mungbean is higher in South and West Asia than in other regions, being 0.44 and 0.45, respectively (Table 1). Central and East Asia have the lowest gene diversity of 0.22 and 0.27, respectively. Among the wild germplasm analyzed, the highest gene diversity is found in South Asia (0.68), despite consisting of far fewer accessions than germplasm from Australia and neighbouring countries. The genetic distance (D_A) of the cultivated mungbean among different countries of Southeast and West Asia was low ($D_A \leq 0.11$) (Table 3). However, D_A between cultivated mungbean from Nepal and Sri Lanka and other parts of South Asia is relatively high ($D_A \geq 0.18$). In East Asia, D_A between Japanese-cultivated mungbean and Chinese or Korean germplasm is higher than D_A between Chinese and Korean germplasm. The D_A between Central and West Asian cultivated mungbean is lower than that among the other regions. African-cultivated germplasm

have low D_A from South, Southeast and West Asian germplasm (0.13–0.14). Wild mungbean germplasm from Pakistan, India and Bangladesh is most similar to cultivated germplasm of the same region. Wild germplasm from Africa, like cultivated germplasm from Africa, show low D_A from mungbean germplasm from South and Southeast Asia. Within wild germplasm, the D_A between wild germplasm from Australia and that from Indonesia, Timor Leste and Papua New Guinea was the lowest. The D_A between Japanese and Chinese intermediate populations and cultivated mungbean is less than that between them and wild mungbean. This is also the case with Indian and Nepalese intermediate populations, but to a lesser extent. On the other hand, the D_A between intermediate populations from Indonesia and wild populations is less than that between other intermediate populations and wild populations.

4. Core collection

Accessions selected for the mungbean core collection are shown in Table 4. This collection consists of almost equal numbers of wild (49) and cultivated (52) accessions plus five intermediate accessions. The core collection consists of ~17% of the entire collection analysed and includes 90.6% of the SSR alleles. The PCA plot of core collection as revealed by individual coordinated position was similar to entire population (Table 4 and Figure 7). The genetic diversity, heterozygosity and diversity values for each locus of the core collection were higher than the original population (0.69, 0.02 and 0.66 vs 0.62, 0.01 and 0.59, respectively).

5. Outcrossing rate

Average estimated outcrossing rate in cultivated mungbean is 1.68%, with variation ranging among regions from 0.96% in East Asia to 5.21% in central Asia (Table 1). The average outcrossing rate for smaller-flowered wild mungbean is lower at 0.81%, ranging from 0.40 to 2.77%. Intermediate populations showed the highest average level of outcrossing at 2.62%.

6. Genetic structure and geographical analysis of the population clusters

Bayesian analysis of population structure revealed two clearly separated groups largely corresponding to cultivated wild accessions. The intermediate accessions were grouped on either wild or cultivated without any accessions of mixed ancestry.

Analysis of the population structure within each of the two subgroups separately showed a separation into there clusters within each of them (Figure 11-13 and 17-19 and Table 6).

The wild population has a rather clear geographical genetic structure. One group was widely distribution origin except for the Australian ones which not also found in this group (Figure 15). There were from Africa to Southeast Asia. Wild accessions of this group might be dispersed from their origin to other countries in the early time. All accessions revealed minor adaptation from the primitive alleles of wild mungbean.

Groups two and three were distinct groups of the accesions from Australia, Papua New Guinea and Indonesia. The number of group two comprised clear geographical North-East Australia (Queensland) to Papua New Guinea (Figure 16). The climate of this part is subtropical to tropical (Wikipedia, 2008 and Appendix Figure 5) which may be affected adaptation and selection of specific alleles in wild mungbean of this part.

Group three occupied in North-West Australia to East Timor in Indonesia (Figure 16). The climate in this part is tropical (Wikipedia, 2008 and Appendix Figure 5). It affects adaptation of certain alleles and geographical genetic structure of wild mungbean in this part.

The cultivated mungbean was classified into three subgroups (Figure 17-19). There were no clear geographical genetic structures (Figure 20). However, cluster two and three were rather distributed in South and West Asia.

The distribution of group one (G1) had the largest number of accessions and ranged from Turkey (West Asia) to South East Asia (Figure 20). Accessions of this group have more green-and yellow-seeded varieties than the other groups. This group comprised 182 green (84.65%), 27 yellow (12.56%) and 6 black mottling (2.79%) seed colors (Table 9-11).

Group two (G2) was from distributing in West and South Asia (Figure 20). Accessions of this group have highest frequencies of black mottling seed. There were 55 green (57.89%), 6 yellow (6.32%) and 34 black mottling (35.79%) seed colors (Table 9-11).

Group three (G3) was mostly distributed in South and West Asia. This group comprised some accessions from Central Asia (Figure 20). Accessions of this group have more black mottling seed color accessions than group one. There were 73 green (70%), 8 yellow (7.62%) and 24 black mottling (22.86) seed colors (Table 9-11).

The percentage of green seed color in G1, G2 and G3 were 58.71%, 17.74% and 23.55%, respectively. The percentage of yellow seeded accessions of G1, G2 and G3 were 65.85%, 14.63% and 19.51%, respectively. While the percentage of black mottling seed color of G1, G2 and G3 were 9.38%, 53.13% and 37.50%, respectively (Table 11).

Group Two and three showed no clear geographical distribution. However, some accessions of group two were located at Northern India and India-Pakistan region, while some accessions of group three were located at Central India. This is supported the hypothesis of Fuller and Harvey (2006) that mungbean might be domestricated twice in India. Both groups revealed more black mottling seed color accessions which is considered a primitive mungbean seed color.

Table 1 List of SSR primers used for the entire population.

	Forward Primer	Reverse Primer	Repeat	Annealing temperature	Linkage Group
CEDG013	CGTTCGAGTTTCTTCGATCG	ACCATCCATCCATTCGCATC	(AG)22	55	1
CEDG087	CCTCTTGAAATTCTCCTTGA	CCTCTTGTGAACCTCAATAA	(AG)10	55	1
CEDG149	GGCTGAAGGTGATGACAGAAG	GGCACTGGTTTTCTAAGGTTGTTG	(AT)12(AG)16	60	1
CEDC050	TCCCACTTCTCCATTACCTCCAC	GAGATTATCTTCTGGGCAGCAAGG	(AC)8	65	2
CEDG108	TCCCAGCTACCCACCTCT	CTTCTACCCAGCCAAACC	(AG)14	60	2
CEDG305	GCAGCTTCACATGCATAGTAC	GAACTTAACTTGGGTTGTCTGC	(AG)22	60	3
CEDG088	TCTTGTCATTTAGCACTTAGCACG	TTGTTGTTTACTAAGAGCCCGTGT	(AG)7	65	4
CEDG139	CAAACTTCCGATCGAAAGCGCTTG	GTTTCTCCTCAATCTCAAGCTCCG	(AG)19	60	4
CEDG264	GATTCCCTTCCTAGCTATGG	CTGCTGGACATGAAGATTCAG	(AG)10 AT(AG)16	60	5
CEDG015	CCCGATGAACGCTAATGCTG	CGCCAAAGGAAACGCAGAAC	(AG)27	60	6
CEDG191	CAATAAGCAATCTGTGGAGAG	CTGCAGGAAACTTGGAATTGC	(AG)21	60	6
CEDG174	GAGGGATCTCCAAAGTTCAACGG	GAAGGCTCCGAAGTTGAAGGTTG	(AG)22	60	7
CEDG247	GTAGACACTGATCATCACC	GACCATCATCGATACGATTC	(AG)16	60	8
CEDG269	CTGTTACGGCACCTGGAAAG	GCAGAGACACACCTTAACCTTG	(AG)14	60	8
CEDG056	TTCCATCTATAGGGGAAGGGAG	GCTATGATGGAAGAGGGCATGG	(AG)14	60	9
CEDG304	ACCACTTCATAATCCCTGAG	GTTGCATGCTATATTTTGGTTCAC	(AG)9	55	9
CEDG075	Note	Note	Note	Note	10
CEDG150	GAAGGGAATGAAAATGAAACCC	GTTCAATCCATTCAGTCTCC	(AG)14	50	10
CEDG100	CCCATCAAGTAACTACATAACA	ATGTGGGACTGGACAAATAAAA	(AG)4A(AG)2A(AG)3	55	11

Note: No available from National Institute of Agrobiological Science

Table 2 Origin and number of cultivated, wild and intermediated mungbean accessions from different countries used in this study, together with their gene diversity, observed heterozygosity and estimated outcrossing rate.

Populations (Code)	No. of accessions	Loci typed	No. of alleles	Gene diversity	Observed heterozygosity	Outcrossing rate(t%)
Cultivated	415	19	138	0.41	0.01	1.68
East Asia	50	19	61	0.27	0.01	0.96
Japan (CJPN)	10	19	34	0.19	0.01	1.37
South Korea (CPRK)	10	19	39	0.28	0.01	0.91
China (CCHN)	20	19	44	0.26	0.01	1.01
Taiwan ($CCHN_t$)	10	19	35	0.20	0.01	1.27
Southeast Asia	131	19	97	0.32	0.01	1.99
Philippines (CPHL)	21	19	47	0.27	0.01	0.91
Indonesia (CIDN)	35					
Timor-Leste (CTLS)	2					
(CIDN+CTLS)	37	19	81	0.37	0.02	2.35
Vietnam (CVNM)	20					
Laos (CLAO)	5					
(CVNM+CLAO)	25	19	49	0.26	0.01	2.93
Thailand (CTHA)	28	19	47	0.25	0.01	2.67
Myanmar (CMMR)	20	19	45	0.26	0.01	1.47
South Asia	101	19	105	0.44	0.01	1.63
Nepal (CNPL)	9	19	35	0.21	0.01	1.37
Bangladesh (CBGD)	2					
India (CIND)	80					
(CBGD+CIND)	82	19	101	0.45	0.02	1.78
Sri Lanka (CLKA)	10	19	37	0.23	0.01	1.06
West Asia	112	19	92	0.45	0.02	2.04
Pakistan (CPAK)	50	19	75	0.46	0.01	0.81
Afghanistan (CAFG)	30	19	59	0.37	0.04	4.88
Iran (CIRN)	20					
Iraq (CIRQ)	2					
(CIRN+CIRQ)	22	19	62	0.35	0.02	3.15
Turkey (CTUR)	10	19	43	0.36	0.01	1.42
Central Asia	14	19	38	0.22	0.02	5.21
Uzbekistan (CUZB)	9					
Tadhikistan (CTJK)	2					
Krygystan (CKGZ)	3					
Africa	7	19	44	0.40	0	0
Mauritius (CMUS)	1					
Madagascar (CMDG)	4					
Nigeria (CNGA)	2					
Wild	189	19	257	0.63	0.01	0.81
Australia (WAUS)	126	19	189	0.59	0.01	0.40
Indonesia (WIDN)	8					
Timor-Leste (WTLS)	6					

Table 2 (Continued)

Populations (Code)	No. of accessions	Loci typed	No. of alleles	Gene diversity	Observed heterozygosity	Outcrossing rate(t%)
Myanmar (WMMR)	7					
India (WIND)	2					
Sri Lanka (WLKA)	2					
(WMMR+WIND+ WLKA)	11	19	103	0.68	0.04	2.77
Cameroon (WCMR)	7					
Madagascar (WMDG)	2					
(WCMR+WMDG)	9	19	64	0.48	0.01	1.16
Intermediate	11	19	84	0.63	0.03	2.62
Japan (MJPN)	3	19	23	0.11	0	0
China (MCHA)	2	19	24	0.16	0.05	14.29
Indonesia (MIDN)	3	19	41	0.49	0.09	7.93
Nepal (MNPL)	1	19	19	-	-	-
India (MIND)	2	19	34	0.53	0	0
Total/Mean	**615**	**19**	**309**	**0.62**	**0.01**	**1.06**

Table 3 SSR primers used linkage group (LG), number of alleles per locus, alleles size range, Diversity values for each locus (Heterozygosity) and allelic richness for cultivated, wild and intermediate mungbean accessions.

No.	Primer	LG	No. of alleles				Alleles size range (bp)			
			All	Cultivated	Wild	Intermediate	All	Cultivated	Wild	Intermediate
1	CEDG013	1	24	14	19	7	78-122 (46)	80-122 (42)	78-116 (44)	80-122 (42)
2	CEDG087	1	6	1	6	2	116-128 (12)	120	116-128 (12)	120-122 (2)
3	CEDG149	1	19	7	16	4	156-211 (55)	156-185 (29)	156-211 (55)	156-180 (24)
4	CEDC050	2	8	4	7	4	108-128 (20)	122-128 (6)	108-126 (18)	116-126 (10)
5	CEDG108	2	5	5	3	4	125-130 (5)	125-130 (5)	127-129 (2)	127-130 (3)
6	CEDG305	3	10	7	8	4	102-124 (22)	102-124 (22)	102-120 (18)	102-124 (22)
7	CEDG088	4	19	6	17	5	104-141 (37)	105-119 (14)	104-141 (37)	111-129 (18)
8	CEDG139	4	21	6	19	6	192-234 (42)	192-230 (38)	196-234 (38)	192-228 (36)
9	CEDG264	5	18	2	17	5	156-198 (42)	160-162 (2)	156-198 (42)	160-180 (20)
10	CEDG015	6	27	1	27	4	162-218 (62)	173	162-218 (62)	167-189 (22)
11	CEDG191	6	22	14	21	6	152-188 (36)	156-186 (30)	152-188 (36)	165-181 (16)
12	CEDG174	7	2	2	2	2	183-187 (4)	183-187 (4)	183-187 (4)	183-187 (4)

Table 3 (Continued)

No.	Primer	LG	No. of alleles				Alleles size range (bp)			
			All	Cultivated	Wild	Intermediate	All	Cultivated	Wild	Intermediate
13	CEDG247	8	8	6	6	3	152-164 (12)	152-164 (12)	152-162 (10)	152-154 (2)
14	CEDG269	8	10	1	9	2	130-143 (13)	130	130-143 (13)	130-139 (9)
15	CEDG056	9	35	15	23	7	161-255 (94)	201-255 (54)	161-233 (72)	193-245 (52)
16	CEDG304	9	37	30	22	6	63-156 (93)	63-156 (93)	63-130 (67)	80-151 (70)
17	CEDG075	10	18	9	18	6	240-274 (34)	244-268 (24)	240-274 (34)	248-262 (14)
18	CEDG150	10	11	5	9	4	148-175 (27)	148-162 (14)	154-175 (22)	154-164 (10)
19	CEDG100	11	9	3	8	3	180-202 (22)	185-202 (17)	180-189 (9)	185-189 (4)
	Total		309	138	257	84				
	Average		16.26	7.26	13.53	4.42	35.68	21.53	31.31	20

Table 3 (Continued)

No.	Primer	Diversity values for each locus				Allelic richness		
		All	Cultivated	Wild	Intermediate	Cultivated	Wild	Intermediate
1	CEDG013	0.78	0.63	0.9	0.84	5.46	10.31	7
2	CEDG087	0.38	0	0.35	0.46	1	3.78	2
3	CEDG149	0.65	0.39	0.81	0.55	3.79	7.34	4
4	CEDC050	0.57	0.26	0.58	0.63	2.14	4.39	4
5	CEDG108	0.46	0.44	0.11	0.64	3.03	1.88	4
6	CEDG305	0.71	0.68	0.27	0.67	4.88	3.22	4
7	CEDG088	0.71	0.45	0.8	0.73	3.06	7.27	5
8	CEDG139	0.61	0.22	0.91	0.66	2.4	10.3	6
9	CEDG264	0.52	0.05	0.87	0.45	1.45	9.12	5
10	CEDG015	0.5	0	0.94	0.45	1	13.22	4
11	CEDG191	0.76	0.54	0.92	0.78	5.66	11.25	6
12	CEDG174	0.38	0.33	0.04	0.5	2	1.38	2
13	CEDG247	0.59	0.33	0.52	0.56	2.56	3.79	3
14	CEDG269	0.06	0	0.18	0.17	1	2.71	2
15	CEDG056	0.82	0.64	0.82	0.83	5.24	8.25	7
16	CEDG304	0.92	0.87	0.9	0.73	9.96	10.79	6
17	CEDG075	0.83	0.75	0.86	0.81	5.52	8.75	6
18	CEDG150	0.36	0.3	0.43	0.45	2.18	3.85	4
19	CEDG100	0.55	0.3	0.67	0.58	2.05	4.77	3
	Total							
	Average	0.59	0.38	0.62	0.6	3.39	6.65	4.42

Table 4 Genetic distance, { D_A from Nei *et al*., (1983)} among mungbean populations.

	CJPN	**CPRK**	**CCHN**	**CCHN$_t$**	**CPHL**	**CIDN+ CTLS**	**CVNM+ CLAO**	**CTHA**	**CMMR**	**CNPL**	**CBGD+ CIND**	**CLKA**
East Asian cultivated population												
CJPN	0	0.14	0.15	0.18	0.16	0.14	0.19	0.13	0.13	0.13	0.24	0.17
CPRK	0.14	0	0.07	0.08	0.07	0.12	0.13	0.09	0.11	0.15	0.19	0.09
CCHN	0.15	0.07	0	0.05	0.06	0.09	0.09	0.06	0.1	0.14	0.17	0.05
CCHN$_t$	0.18	0.08	0.05	0	0.06	0.13	0.11	0.08	0.12	0.17	0.22	0.08
Southeast Asian cultivated population												
CPHL	0.16	0.07	0.06	0.06	0	0.08	0.1	0.08	0.1	0.18	0.15	0.08
CIDN+ CTLS	0.14	0.12	0.09	0.13	0.08	0	0.09	0.08	0.07	0.13	0.07	0.11
CVN+ CLAO	0.19	0.13	0.09	0.11	0.1	0.09	0	0.05	0.06	0.1	0.17	0.11
CTHA	0.13	0.09	0.06	0.08	0.08	0.08	0.05	0	0.05	0.09	0.18	0.09
CMMR	0.13	0.11	0.1	0.12	0.1	0.07	0.06	0.05	0	0.06	0.14	0.09
South Asian cultivated population												
CNPL	0.13	0.15	0.14	0.17	0.18	0.13	0.1	0.09	0.06	0	0.2	0.14
CBG+ CIND	0.24	0.19	0.17	0.22	0.15	0.07	0.17	0.18	0.14	0.2	0	0.18
CLKA	0.17	0.09	0.05	0.08	0.08	0.11	0.11	0.09	0.09	0.14	0.18	0
West Asian cultivated population												
CPAK	0.25	0.22	0.21	0.26	0.19	0.12	0.21	0.23	0.19	0.22	0.05	0.21
CAFG	0.22	0.17	0.15	0.19	0.13	0.1	0.13	0.16	0.15	0.2	0.08	0.17

Table 4 (Continued)

	CJPN	CPRK	CCHN	$CCHN_t$	CPHL	CIDN+ CTLS	CVNM+ CLAO	CTHA	CMMR	CNPL	CBGD+ CIND	CLKA
CIRN+ CIRQ	0.22	0.17	0.15	0.17	0.14	0.13	0.14	0.15	0.15	0.18	0.13	0.16
CTUR	0.22	0.18	0.15	0.18	0.14	0.1	0.14	0.14	0.13	0.17	0.13	0.16
Cetral Asian Cultivated population												
CUZB+ CTJK+ CKGK	0.24	0.2	0.19	0.19	0.16	0.15	0.16	0.18	0.2	0.23	0.18	0.18
African cultivated population												
CMU+ CMDG	0.21	0.17	0.19	0.2	0.15	0.13	0.21	0.19	0.18	0.23	0.13	0.2
Wild population												
WAUS	0.68	0.69	0.71	0.7	0.65	0.63	0.71	0.68	0.69	0.74	0.62	0.73
WIDN+ WTLS+ WPNG	0.68	0.69	0.72	0.7	0.66	0.65	0.71	0.68	0.69	0.74	0.66	0.73
WMMR+ WIND+ WLKA	0.65	0.65	0.65	0.68	0.64	0.59	0.66	0.65	0.63	0.69	0.57	0.67
WCMR+ WMDG	0.63	0.63	0.65	0.64	0.61	0.56	0.66	0.63	0.62	0.69	0.55	0.68
Intermediate population												
MJPN	0.33	0.29	0.22	0.22	0.22	0.24	0.24	0.22	0.26	0.26	0.33	0.26
MCHA	0.28	0.32	0.27	0.3	0.25	0.23	0.19	0.25	0.24	0.26	0.27	0.27
MIDN	0.52	0.53	0.58	0.57	0.55	0.53	0.59	0.56	0.54	0.58	0.55	0.6
MNPL	0.62	0.63	0.59	0.62	0.58	0.5	0.55	0.6	0.58	0.61	0.43	0.59
MIND	0.59	0.59	0.59	0.6	0.55	0.51	0.58	0.6	0.57	0.6	0.45	0.62

Table 4 (Continued)

	CPAK	CAFG	CIRN+CIRQ	CTUR	CUZB+CTJK+CKGK	CMUS+CMDG
East Asian cultivated population						
CJPN	0.25	0.22	0.22	0.22	0.24	0.21
CPRK	0.22	0.17	0.17	0.18	0.2	0.17
CCHN	0.21	0.15	0.15	0.15	0.19	0.19
$CCHN_t$	0.26	0.19	0.17	0.18	0.19	0.2
Southeast Asian cultivated population						
CPHL	0.19	0.13	0.14	0.14	0.16	0.15
CIDN+ CTLS	0.12	0.1	0.13	0.1	0.15	0.13
CVNM+ CLAO	0.21	0.13	0.14	0.14	0.16	0.21
CTHA	0.23	0.16	0.15	0.14	0.18	0.19
CMMR	0.19	0.15	0.15	0.13	0.2	0.18
South Asian cultivated population						
CNPL	0.22	0.2	0.18	0.17	0.23	0.23
CBGD+ CIND	0.05	0.08	0.13	0.13	0.18	0.13
CLKA	0.21	0.17	0.16	0.16	0.18	0.2
West Asian cultivated population						
CPAK	0	0.08	0.14	0.15	0.23	0.14
CAFG	0.08	0	0.08	0.11	0.15	0.12
CIRN+ CIRQ	0.14	0.08	0	0.1	0.14	0.17
CTUR	0.15	0.11	0.1	0	0.18	0.18

Table 4 (Continued)

	CPAK	CAFG	CIRN+CIRQ	CTUR	CUZB+CTJK+CKGK	CMUS+CMDG
Cetral Asian Cultivated population						
CUZB+ CTJK+ CKGK	0.23	0.15	0.14	0.18	0	0.14
African cultivated population						
CMUS+ CMDG	0.14	0.12	0.17	0.18	0.14	0
Wild population						
WAUS	0.63	0.65	0.69	0.68	0.68	0.61
WIDN+ WTLS+ WPNG	0.66	0.65	0.69	0.69	0.69	0.64
WMMR+ WIND+ WLKA	0.57	0.62	0.66	0.63	0.7	0.61
WCMR+ WMDG	0.56	0.62	0.66	0.59	0.65	0.55
Intermediate population						
MJPN	0.34	0.27	0.24	0.23	0.31	0.33
MCHA	0.29	0.21	0.22	0.2	0.25	0.32
MIDN	0.54	0.54	0.61	0.6	0.6	0.52
MNPL	0.43	0.43	0.5	0.51	0.56	0.51
MIND	0.46	0.53	0.57	0.54	0.62	0.53

Table 4 (Continued)

	WAUS	WIDN+WTLS+WPNG	WMMR+WIND+WLKA	WCMR+WMDG	MJPN	MCHA	MIDN	MNPL	MIND
East Asian cultivated population									
CJPN	0.68	0.68	0.65	0.63	0.33	0.28	0.52	0.62	0.59
CPRK	0.69	0.69	0.65	0.63	0.29	0.32	0.53	0.63	0.59
CCHN	0.71	0.72	0.65	0.65	0.22	0.27	0.58	0.59	0.59
$CCHN_t$	0.7	0.7	0.68	0.64	0.22	0.3	0.57	0.62	0.6
Southeast Asian cultivated population									
CPHL	0.65	0.66	0.64	0.61	0.22	0.25	0.55	0.58	0.55
CIDN+CTLS	0.63	0.65	0.59	0.56	0.24	0.23	0.53	0.5	0.51
CVNM+CLAO	0.71	0.71	0.66	0.66	0.24	0.19	0.59	0.55	0.58
CTHA	0.68	0.68	0.65	0.63	0.22	0.25	0.56	0.6	0.6
CMMR	0.69	0.69	0.63	0.62	0.26	0.24	0.54	0.58	0.57
South Asian cultivated population									
CNPL	0.74	0.74	0.69	0.69	0.26	0.26	0.58	0.61	0.6
CBGD+CIND	0.62	0.66	0.57	0.55	0.33	0.27	0.55	0.43	0.45
CLKA	0.73	0.73	0.67	0.68	0.26	0.27	0.6	0.59	0.62
West Asian cultivated population									
CPAK	0.63	0.66	0.57	0.56	0.34	0.29	0.54	0.43	0.46
CAFG	0.65	0.65	0.62	0.62	0.27	0.21	0.54	0.43	0.53
CIRN+CIRQ	0.69	0.69	0.66	0.66	0.24	0.22	0.61	0.5	0.57

Table 4 (Continued)

	WAUS	WIDN+WTLS+WPNG	WMMR+WIND+WLKA	WCMR+WMDG	MJPN	MCHA	MIDN	MNPL	MIND
CTUR	0.68	0.69	0.63	0.59	0.23	0.2	0.6	0.51	0.54
Cetral Asian Cultivated population									
CUZB+ CTJK+ CKGK	0.68	0.69	0.7	0.65	0.31	0.25	0.6	0.56	0.62
African cultivated population									
CMUS+ CMDG	0.61	0.64	0.61	0.55	0.33	0.32	0.52	0.51	0.53
Wild population									
WAUS	0	0.13	0.43	0.56	0.73	0.75	0.41	0.81	0.65
WIDN+ WTLS+ WPNG	0.13	0	0.42	0.62	0.7	0.74	0.4	0.82	0.68
WMMR+ WIND+ WLKA	0.43	0.42	0	0.61	0.68	0.73	0.57	0.65	0.66
WCMR+ WMDG	0.56	0.62	0.61	0	0.73	0.74	0.63	0.77	0.63
Intermediate population									
MJPN	0.73	0.7	0.68	0.73	0	0.32	0.65	0.65	0.69
MCHA	0.75	0.74	0.73	0.74	0.32	0	0.68	0.61	0.59
MIDN	0.41	0.4	0.57	0.63	0.65	0.68	0	0.78	0.69
MNPL	0.81	0.82	0.65	0.77	0.65	0.61	0.78	0	0.75
MIND	0.65	0.68	0.66	0.63	0.69	0.59	0.69	0.75	0

See Table 1 for abbreviations of population names

Table 5 List of mungbean accessions in the core collection.

No.	Label	Status	Accessions no.	Country of Origin	State/Province	Collection site	Note
1	W001	Wild	-	Australia	Queensland	Georgetown	-
2	W010	Wild	-	Australia	Queensland	Queensland	powdery mildew and halo bligth resistance
3	W026	Wild	-	Australia	Northern Territory	Northern Territory	-
4	W039	Wild	-	Australia	Queensland	Queensland	-
5	W046	Wild	-	Australia	Queensland	Queensland	-
6	W053	Wild	-	Australia	Queensland	Queensland	-
7	W055	Wild	-	Australia	Queensland	Queensland	-
8	W058	Wild	-	Australia	Queensland	Northern Territory	-
9	W063	Wild	-	Australia	Northern Territory	Northern Territory	-
10	W065	Wild	-	Australia	Northern Territory	Northern Territory	-
11	W073	Wild	-	Australia	Queensland	Queensland	halo bligth resistance
12	W077	Wild	-	Australia	Queensland	Queensland	short pod (4.1 cm, 0.88 g)
13	W080	Wild	-	Australia	Northern Territory	Northern Territory	-
14	W083	Wild	-	Australia	Northern Territory	Northern Territory	-
15	W101	Wild	-	Australia	Queensland	Queensland	-
16	W104	Wild	-	Australia	Northern Territory	Northern Territory	-
17	W105	Wild	-	Australia	Queensland	Queensland	-
18	W109	Wild	-	Australia	Queensland	Queensland	-

Table 5 (Continued)

No.	Label	Status	Accessions no.	Country of Origin	State/Province	Collection site	Note
19	W111	Wild	-	Australia	Queensland	Queensland	-
20	W115	Wild	-	Australia	Queensland	Queensland	-
21	W116	Wild	-	Australia	Western Australia	Western Australia	-
22	W128	Wild	JP218943	Cameroon	Lara	Lara	-
23	W130	Wild	JP218945	Cameroon	Ngutchmi	Ngutchmi	-
24	W131	Wild	JP227260	Carmeroon	Dembo	Dembo	short pod (3.8 cm, 1.51 g)
25	W133	Wild	JP227262	Carmeroon	Nigba	Nigba	-
26	W134	Wild	JP227263	Carmeroon	Pate maga	Pate maga	-
27	W138	Wild	JP 226584	Timor-Leste	Manatuto	Manatuto	-
28	W141	Wild	JP 226609	Timor-Leste	Ainaro	Ainaro	short pod (3.9 cm, 0.64 g)
29	W147	Wild	JP107875	India	-	-	-
30	W148	Wild	JP110831	India	Rishikesh	Rishikesh	-
31	W158	Wild	JP 202270	Indonesia	-	-	-
32	W159	Wild	JP 202271	Indonesia	-	-	-
33	W162	Wild	JP107877	Madagascar	-	-	TC 1966 : bruchid resistane
34	W164	Wild	JP 210796	Myanmar	Mandalay	Kyauk Tham Pat	-
35	W165	Wild	JP 210804	Myanmar	Shan	Kalaw	-
36	W168	Wild	JP 217436	Myanmar	Shan	Kalaw	-
37	W169	Wild	JP 217437	Myanmar	Shan	Khar Lein	-
38	W172	Wild	-	Papua New Guinea	Bogola	Bogola	-
39	W174	Wild	JP202294	Papua New Guinea	-	-	-
40	W175	Wild	JP218942	Papua New Guinea	Port Moreby	Port Moreby	-
41	W176	Wild	JP222454	Papua New Guinea	Port Moreby	Waigani	-
42	W177	Wild	JP222455	Papua New Guinea	Port Moreby	Napa Napa	-

Table 5 (Continued)

No.	Label	Status	Accessions no.	Country of Origin	State/Province	Collection site	Note
43	W179	Wild	JP222457	Papua New Guinea	Port Moreby	Napa Napa	-
44	W182	Wild	JP222460	Papua New Guinea	Central	Kubuna Mission	-
45	W190	Wild	JP226873	Papua New Guinea	East Sepik	Timbun	collected from low land near river; Sepik
46	W191	Wild	JP226874	Papua New Guinea	East Sepik	Ari John	collected from low land near river; Sepik
47	W192	Wild	JP226875	Papua New Guinea	East Sepik	Savanaut	collected from low land near river; Sepik
48	W203	Wild	JP 210617	Sri Lanka	Mahiyangana	Welpallewela	-
49	W204	Wild	JP 217528	Sri Lanka	-	-	-
50	H011	Cultivated	JP229109	Korea, South	ChungChonh NamDo	ChungChonh NamDo	-
51	H022	Cultivated	JP229144	China	Shaanxi	Shaanxi	-
52	H023	Cultivated	JP229145	China	Henan	Henan	-
53	H027	Cultivated	JP229215	China	Shanxi	Xian	-
54	H030	Cultivated	JP229216	China	Shanxi	Xian	long pod (14.9 cm, 6.1 g)
55	H050	Cultivated	JP99049	Taiwan	-	-	-
56	H071	Cultivated	-	Philippines	-	-	V 2802: bruchid resistance
57	H082	Cultivated	JP229133	Indonesia	Sabu	Sabu	glabrous plant
58	H090	Cultivated	JP229222	Indonesia	Makassar	Makassar	-
59	H095	Cultivated	JP229227	Indonesia	Makassar	Makassar	-
60	H102	Cultivated	JP229233	Indonesia	Makassar	Makassar	-
61	H110	Cultivated	JP78939	Vietnam	Bac Thai	Phu Yen	-
62	H149	Cultivated	JP229096	Thailand	Sukhothai	Sukhothai	long pod (12.2 cm, 7.8 g)

Table 5 (Continued)

No.	Label	Status	Accessions no.	Country of Origin	State/Province	Collection site	Note
63	H150	Cultivated	JP229097	Thailand	Kamphaeng Phet	Kamphaeng Phet	long pod (12.8 cm, 8.4 g)
64	H151	Cultivated	JP229098	Thailand	Nakhon Phatom	Kapaeng saen	long pod (14.7 cm, 8.0 g)
65	H152	Cultivated	JP229099	Thailand	Uthai Thani	Lan sak	long pod (14.4 cm, 7.4 g)
66	H157	Cultivated	-	Thailand	Chinat	Chinat	resistance to iron-deficient soil
67	H162	Cultivated	-	Thailand	-	-	VC 1210A: powdery mildew resistance
68	H192	Cultivated	JP229130	Bangladesh	-	-	-
69	H205	Cultivated	JP229213	India	United	Tehri	-
70	H209	Cultivated	JP229170	India	Kathiawar	Veraval	-
71	H211	Cultivated	JP229163	India	Cutch	Bhuj	-
72	H215	Cultivated	JP229180	India	Mysore	Mysore	-
73	H219	Cultivated	JP229200	India	Jodhpur	Jodhpur	-
74	H229	Cultivated	JP229181	India	Gwalior	Gwalior	-
75	H230	Cultivated	JP229181	India	Rajasthan	Pali	-
76	H234	Cultivated	JP229185	India	Sihora	Sihora	-
77	H241	Cultivated	JP229177	India	Srinagar	Srinagar	-
78	H242	Cultivated	JP229175	India	Dhariwal	Dhariwal	-
79	H243	Cultivated	JP229193	India	Punjab	Amraili	-
80	H248	Cultivated	JP229211	India	Uttar Pradesh	Jamnagar	-
81	H250	Cultivated	JP2292160	India	Baroda	Baroda	-
82	H262	Cultivated	JP229190	India	Bhawanipatna	Bhawanipatna	-
83	H273	Cultivated	JP99039	India	-	-	-
84	H274	Cultivated	JP99046	India	-	-	V 2709: bruchid resistance

Table 5 (Continued)

No.	Label	Status	Accessions no.	Country of Origin	State/Province	Collection site	Note
85	H279	Cultivated	JP187898	Sri Lanka	Mihintale	Anuradhapura	-
86	H280	Cultivated	JP81649	Sri Lanka	Mihintale	Anuradhapura	-
87	H285	Cultivated	JP81653	Pakistan	North-West Frontier	Chakiatan	-
88	H287	Cultivated	JP103115	Pakistan	North-West Frontier	Maindam	-
89	H296	Cultivated	JP103128	Pakistan	Punjab	Mair	-
90	H306	Cultivated	JP103136	Pakistan	Balochistan	Dandar	-
91	H307	Cultivated	JP103138	Pakistan	Balochistan	Dandar	-
92	H334	Cultivated	JP73290	Pakistan	-	-	-
93	H337	Cultivated	JP229240	Afghanistan	Kandahar	Kandahar	-
94	H351	Cultivated	JP74721	Afghanistan	-	-	-
95	H357	Cultivated	JP99009	Afghanistan	-	-	-
96	H366	Cultivated	JP229253	Iran	Masanderan	Masanderan	-
97	H370	Cultivated	JP229256	Iran	Kerman	Kerman	-
98	H377	Cultivated	JP229263	Iran	Emamshahr (Shahrud)	Emamshahr (Shahrud)	-
99	H384	Cultivated	JP31331	Iran	-	-	control line
100	H412	Cultivated	JP98808	Madagascar	-	-	-
101	H417	Cultivated	JP212360	Nigeria	-	-	V 2817: bruchid resistance
102	M135	Intermediate	-	China	-	-	high out crossing rate
103	M136	Intermediate	-	China	-	-	high out crossing rate
104	M146	Intermediate	-	India	Tamilnadu	Tamil nadu	-
105	M151	Intermediate	-	Indonesia	Lesser Sunda	Lesser Sunda	-
106	M153	Intermediate	-	Indonesia	Lesser Sunda	Lesser Sunda	-

Table 6 List of accession name were assigned by Bayesian method at K2 under no admixture model.

Group	Accession name									
Group1	W001	W002	W003	W004	W005	W006	W007	W009	W010	W011
	W012	W013	W014	W015	W016	W017	W018	W019	W020	W021
	W022	W023	W024	W025	W026	W027	W028	W029	W030	W031
	W032	W033	W034	W035	W036	W037	W038	W039	W040	W041
	W042	W043	W044	W045	W046	W047	W048	W049	W050	W051
	W052	W053	W054	W055	W056	W057	W058	W059	W060	W061
	W062	W063	W064	W065	W066	W067	W068	W069	W070	W071
	W072	W073	W074	W075	W076	W077	W078	W079	W080	W081
	W082	W083	W084	W085	W086	W087	W088	W089	W090	W091
	W092	W093	W094	W095	W096	W097	W098	W099	W100	W101
	W102	W103	W104	W105	W106	W107	W108	W109	W110	W111
	W112	W113	W114	W115	W116	W117	W118	W119	W120	W121
	W122	W123	W124	W125	W126	W127	W128	W129	W130	W131
	W132	W133	W134	W137	W138	W139	W140	W141	W142	W147
	W148	W150	W154	W155	W156	W157	W158	W159	W160	W162
	W163	W164	W165	W166	W167	W168	W169	W170	W171	W172
	W173	W174	W175	W176	W177	W178	W179	W180	W181	W182
	W183	W184	W185	W186	W187	W188	W189	W190	W191	W192
	W193	W194	W195	W196	W197	W198	W199	W203	W204	
	M146	M151	M152	M153						
Group2	H001	H002	H003	H004	H005	H006	H007	H008	H009	H010
	H011	H012	H013	H014	H015	H016	H017	H018	H019	H020
	H021	H022	H023	H024	H025	H026	H027	H028	H029	H030
	H031	H032	H033	H034	H035	H036	H037	H038	H039	H040
	H041	H042	H043	H044	H045	H046	H047	H048	H049	H050
	H051	H052	H053	H054	H055	H056	H057	H058	H059	H060

Table 6 (Continued)

Group	Accession name									
Group2	H061	H062	H063	H064	H065	H066	H067	H068	H069	H070
	H071	H072	H073	H074	H075	H076	H077	H078	H079	H080
	H081	H082	H083	H084	H085	H086	H087	H088	H089	H090
	H091	H092	H093	H094	H095	H096	H097	H098	H099	H100
	H101	H102	H103	H104	H105	H106	H108	H109	H110	H111
	H112	H113	H114	H115	H116	H117	H118	H119	H120	H121
	H122	H123	H124	H125	H126	H127	H128	H129	H130	H131
	H132	H133	H134	H135	H136	H137	H138	H139	H140	H141
	H142	H143	H144	H145	H146	H147	H148	H149	H150	H151
	H152	H153	H154	H155	H156	H157	H158	H159	H160	H161
	H162	H163	H164	H165	H166	H167	H168	H169	H170	H171
	H172	H173	H174	H175	H176	H177	H178	H179	H180	H181
	H182	H183	H184	H185	H186	H187	H188	H189	H190	H191
	H192	H193	H194	H195	H196	H197	H198	H199	H200	H201
	H202	H203	H204	H205	H206	H207	H208	H209	H210	H211
	H212	H213	H214	H215	H216	H217	H218	H219	H220	H221
	H222	H223	H224	H225	H226	H227	H228	H229	H230	H231
	H232	H233	H234	H235	H236	H237	H238	H239	H240	H241
	H242	H243	H244	H245	H246	H247	H248	H249	H250	H251
	H252	H253	H254	H255	H256	H257	H258	H259	H260	H261
	H262	H263	H264	H265	H266	H267	H268	H269	H270	H271
	H273	H274	H275	H276	H277	H278	H279	H280	H281	H282
	H283	H284	H285	H286	H287	H288	H289	H290	H291	H292
	H293	H294	H295	H296	H297	H298	H299	H300	H301	H302
	H303	H304	H305	H306	H307	H308	H309	H310	H311	H312
	H313	H314	H315	H316	H317	H318	H319	H320	H321	H322
	H323	H324	H325	H326	H327	H328	H329	H330	H331	H332
	H333	H334	H335	H336	H337	H338	H339	H340	H341	H342
	H343	H344	H345	H346	H347	H348	H349	H350	H351	H352

Table 6 (Continued)

Group	Accession name									
Group2	H353	H354	H355	H356	H357	H358	H359	H360	H361	H362
	H363	H364	H365	H366	H367	H368	H369	H370	H371	H372
	H373	H374	H375	H376	H377	H378	H379	H380	H381	H382
	H383	H384	H385	H386	H387	H388	H389	H390	H391	H392
	H393	H394	H395	H396	H397	H398	H399	H400	H401	H402
	H403	H404	H405	H406	H407	H408	H409	H410	H411	H412
	H413	H414	H415	H416	H417					

Table 7 List of wild accessions name were assigned by Bayesian method at K3 under no admixture model.

Group	Accession name								
Group 1	W128	W129	W130	W131	W132	W133	W134	W147	W148
	W159	W162	W163	W164	W165	W166	W167	W168	W169
	W172	W203	W204						
Group 2	W001	W002	W003	W004	W006	W007	W009	W010	W011
	W013	W014	W015	W016	W017	W018	W019	W020	W021
	W025	W030	W031	W032	W033	W034	W035	W038	W040
	W042	W043	W044	W045	W052	W053	W055	W056	W059
	W067	W068	W069	W071	W072	W073	W074	W075	W076
	W079	W082	W098	W099	W100	W101	W102	W106	W108
	W110	W111	W112	W114	W115	W117	W118	W122	W123
	W125	W126	W127	W138	W171	W173	W174	W175	W176
	W178	W179	W180	W181	W182	W183	W184	W185	W186
	W188	W189	W190	W194	W195	W196	W197	W198	W199
Group 3	W005	W023	W024	W026	W027	W028	W029	W036	W037
	W046	W047	W048	W049	W050	W051	W054	W057	W058
	W062	W063	W064	W065	W066	W070	W078	W080	W081
	W084	W085	W086	W087	W088	W089	W090	W091	W092
	W094	W095	W096	W097	W103	W104	W105	W107	W113
	W119	W120	W121	W137	W139	W140	W141	W142	W150
	W155	W156	W157	W160	W191	W192	W193		

Table 8 List of cultivated accessions name were assigned by Bayesian method at K3 under no admixture model

Group	Accession name									
Group1	H001	H002	H003	H004	H005	H006	H007	H008	H009	H010
	H011	H012	H013	H014	H015	H016	H017	H018	H019	H020
	H021	H022	H023	H024	H026	H027	H028	H029	H031	H032
	H033	H034	H035	H036	H037	H038	H039	H040	H041	H042
	H043	H044	H045	H046	H047	H048	H049	H050	H051	H052
	H053	H054	H055	H057	H058	H059	H060	H061	H062	H063
	H064	H065	H066	H067	H069	H070	H071	H072	H073	H074
	H075	H076	H077	H078	H079	H080	H081	H082	H084	H087
	H094	H097	H099	H100	H101	H102	H103	H104	H105	H106
	H108	H109	H110	H111	H112	H113	H114	H115	H116	H117
	H118	H120	H121	H122	H123	H124	H125	H126	H127	H128
	H129	H130	H131	H132	H133	H134	H135	H136	H137	H138
	H139	H140	H141	H142	H143	H144	H145	H146	H147	H148
	H149	H150	H151	H152	H153	H154	H155	H156	H157	H158
	H159	H160	H161	H162	H163	H164	H165	H166	H167	H168
	H169	H170	H171	H172	H173	H174	H175	H176	H177	H179
	H181	H182	H183	H184	H185	H186	H187	H188	H189	H190
	H191	H193	H195	H196	H197	H198	H199	H200	H201	H202
	H207	H211	H212	H214	H217	H267	H273	H275	H276	H278
	H279	H280	H281	H282	H283	H284	H289	H290	H331	H332
	H333	H351	H355	H381	H383	H384	H385	H389	H391	H392
	H393	H394	H403	H412	H417					
Group2	H025	H068	H086	H088	H092	H095	H098	H178	H180	H192
	H203	H204	H205	H206	H209	H210	H213	H219	H221	H222
	H223	H224	H225	H226	H228	H232	H235	H236	H239	H240
	H242	H243	H244	H245	H246	H247	H248	H249	H250	H252
	H253	H254	H255	H256	H257	H258	H259	H260	H261	H262
	H263	H265	H266	H270	H271	H274	H277	H286	H287	H291
	H292	H293	H294	H295	H296	H297	H298	H299	H300	H301
	H305	H306	H307	H308	H309	H310	H311	H314	H317	H318
	H319	H320	H321	H322	H330	H334	H343	H344	H352	H356

Table 8 (Continued)

Group	Accession name									
Group2	H363	H380	H395	H396	H416					
Group3	H030	H056	H083	H085	H089	H090	H091	H093	H096	H119
	H194	H208	H215	H216	H218	H220	H227	H229	H230	H231
	H233	H234	H237	H238	H241	H251	H264	H268	H269	H285
	H288	H302	H303	H304	H312	H313	H315	H316	H323	H324
	H325	H326	H327	H328	H329	H335	H336	H337	H338	H339
	H340	H341	H342	H345	H346	H347	H348	H349	H350	H353
	H354	H357	H358	H359	H360	H361	H362	H364	H365	H366
	H367	H368	H369	H370	H371	H372	H373	H374	H375	H376
	H377	H378	H379	H382	H386	H387	H388	H390	H397	H398
	H399	H400	H401	H402	H404	H405	H406	H407	H408	H409
	H410	H411	H413	H414	H415					

Table 9 The numbers of accessions of cultivated were separated by seed color and each group was assigned from Bayesian method at K3 under no admixture model.

Group	Green	Yellow	Black mottling (BLM)	Total
G1	182	27	6	215
G2	55	6	34	95
G3	73	8	24	105
Total	310	41	64	415

Table 10 Percentage of each seed color accessions of cultivated within each group. All groups were assigned by Bayesian method at K3 under no admixture model.

Group	%Green	%Yellow	%BLM	%Total
G1	84.65	12.56	2.79	100
G2	57.89	6.32	35.79	100
G3	69.52	7.62	22.86	100
Total	74.70	9.88	15.42	100

Table 11 Percentage of each seed color accessions of cultivated within population. All groups were assigned by Bayesian method at K3 under no admixture model.

Group	%Green	%Yellow	%BLM
G1	58.71	65.85	9.38
G2	17.74	14.63	53.13
G3	23.55	19.51	37.50
Total	100	100	100

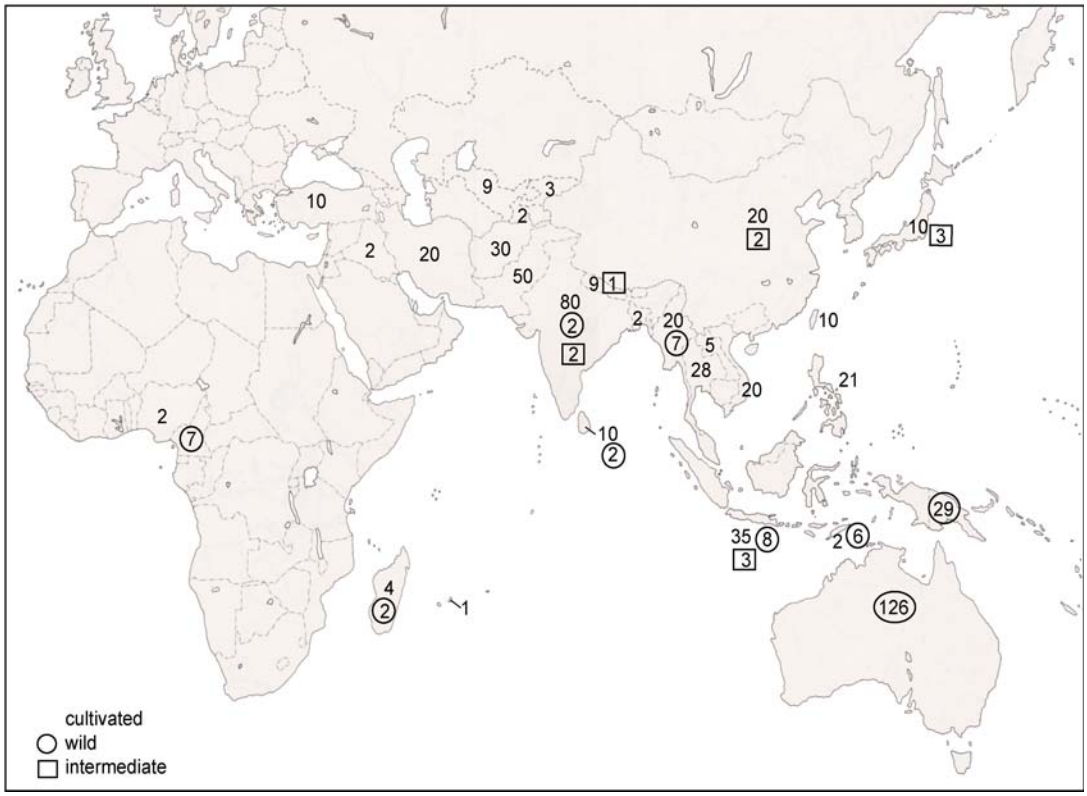

Figure 3 Map showing distribution of the mungbean samples.

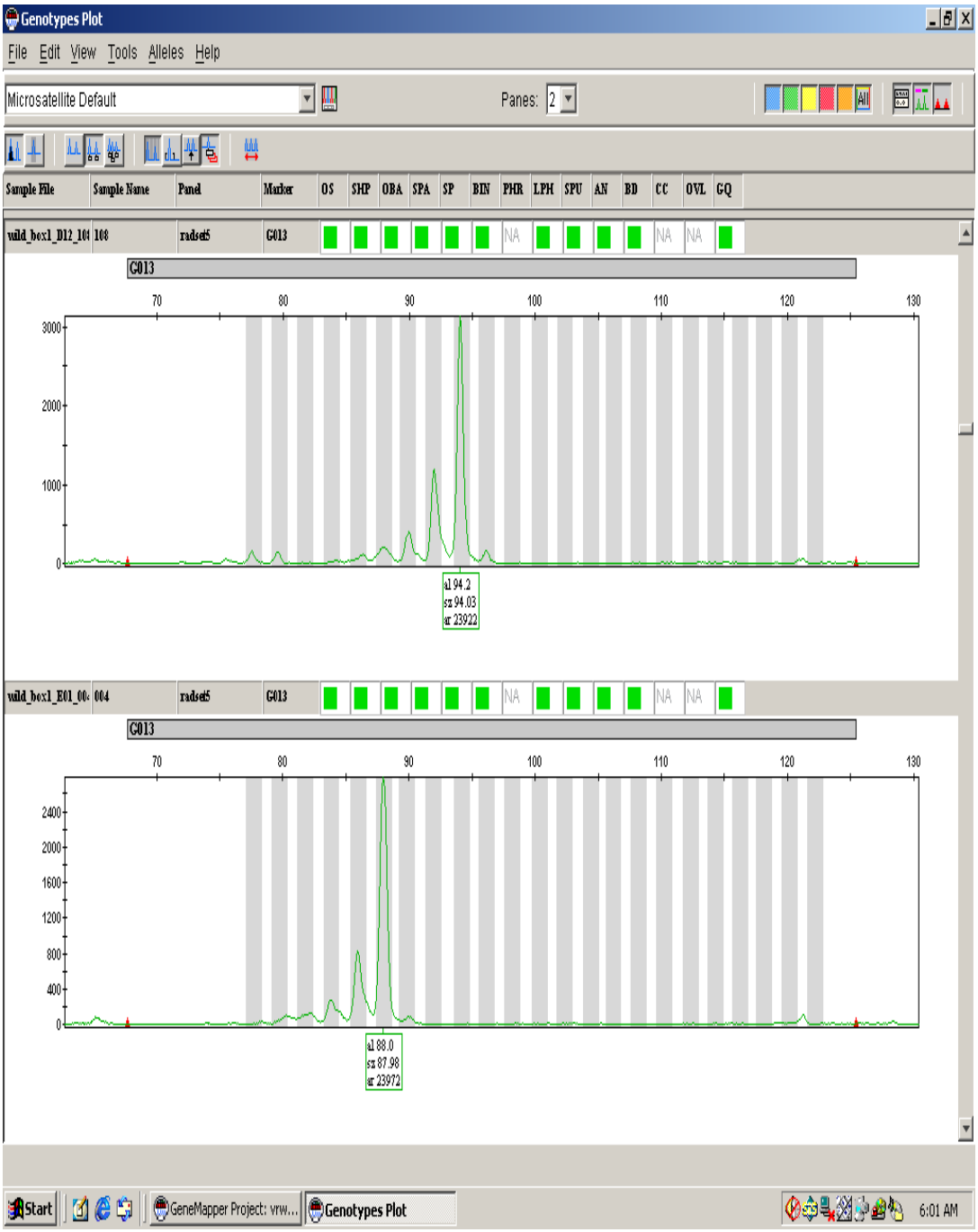

Figure 4 The SSR fragments of CEDG013 primer showing allele sizes of 94 and 88 bp.

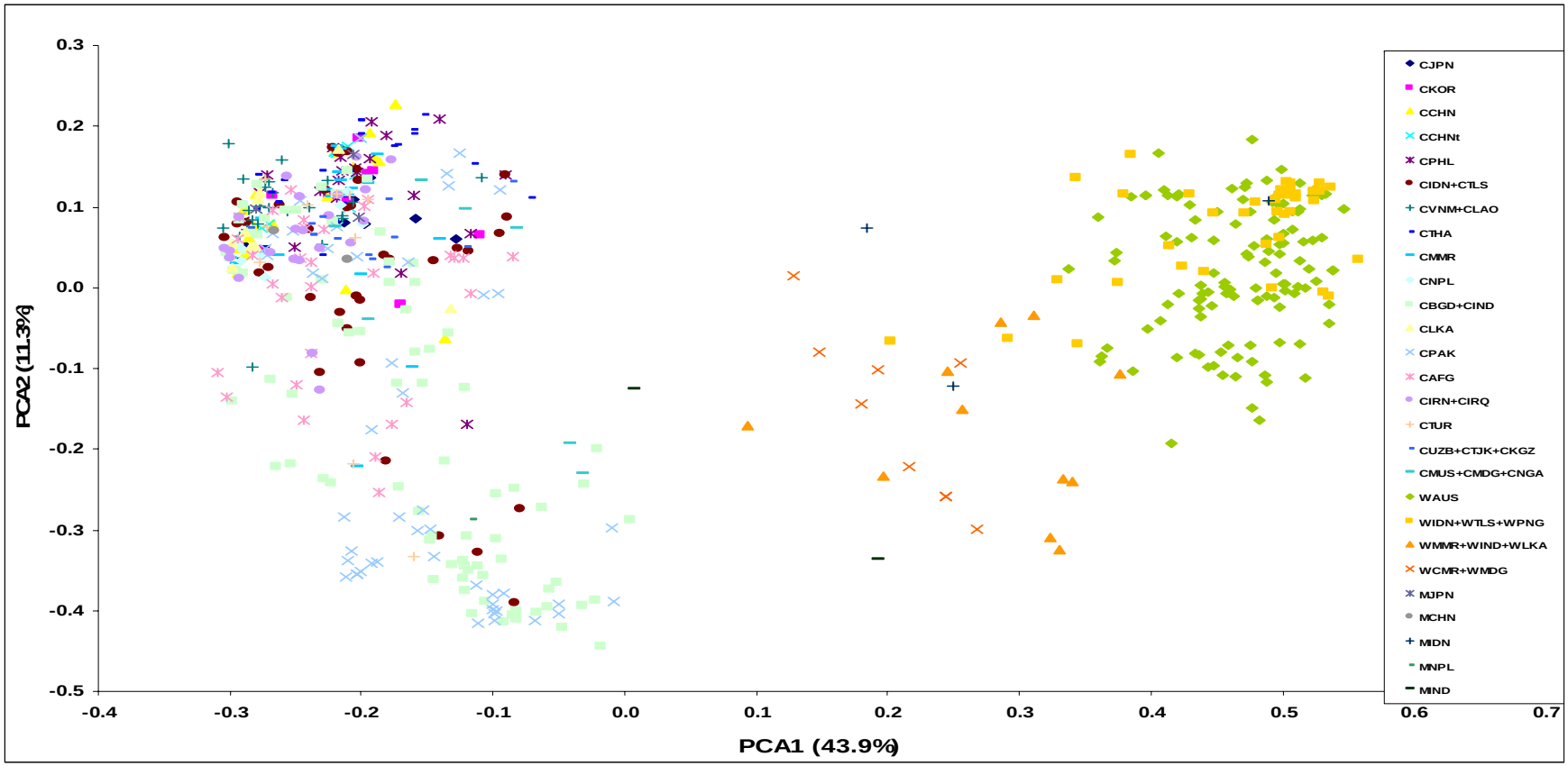

Figure 5 Principal component analysis showing overall distribution of 615 mungbean accessions. See Table 1 for abbreviations of population name

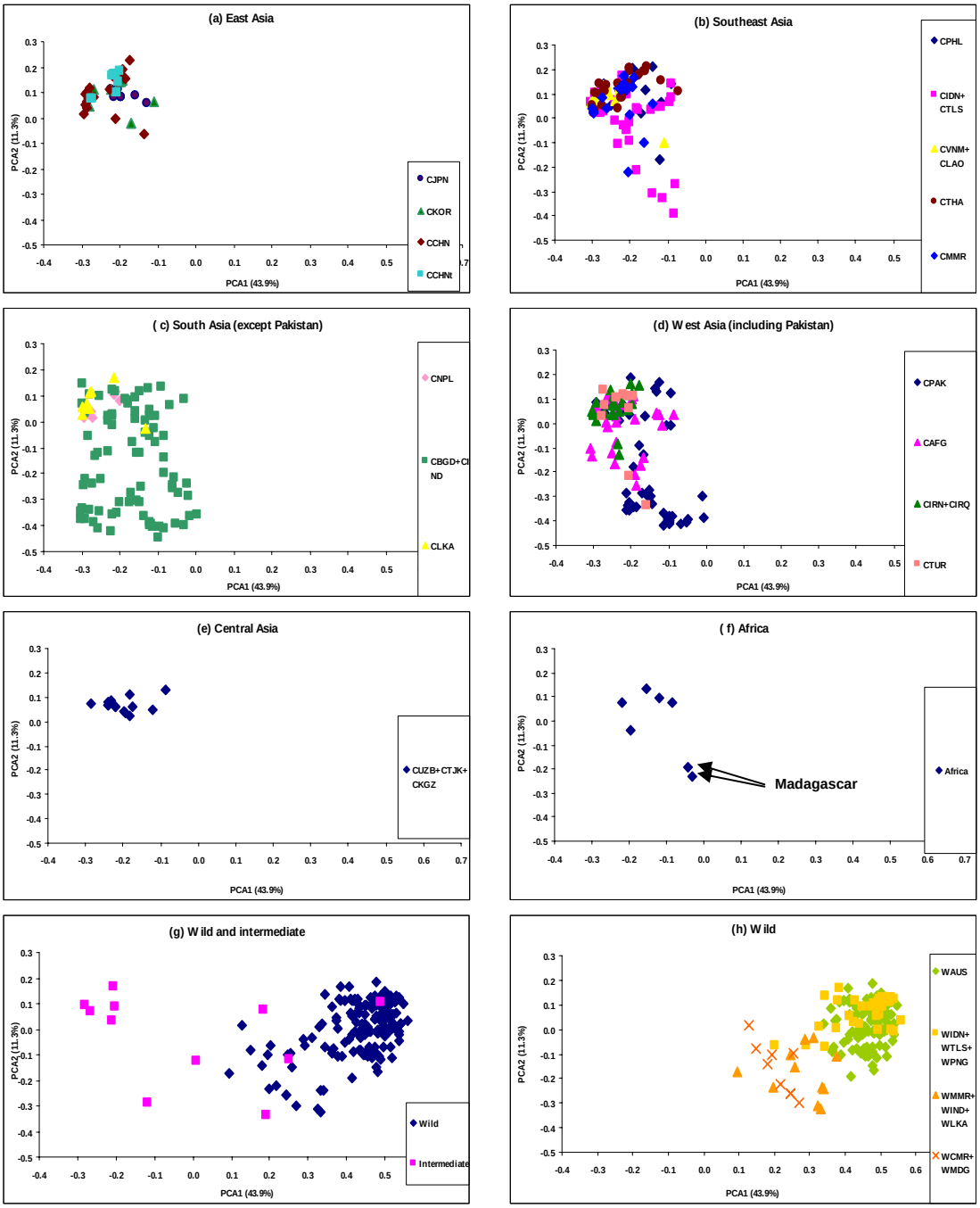

Figure 6 Principal component analysis showing distribution of mungbean germplasm from different origins and types. (a) East Asia, (b) Southeast Asia, (c) South Asia (except Pakistan), (d) West Asia (including Pakistan), (e) Central Asia, (f) Africa, (g) wild and intermediate germplasm, (h) wild germplasm. See Table 1 for abbreviations of population name

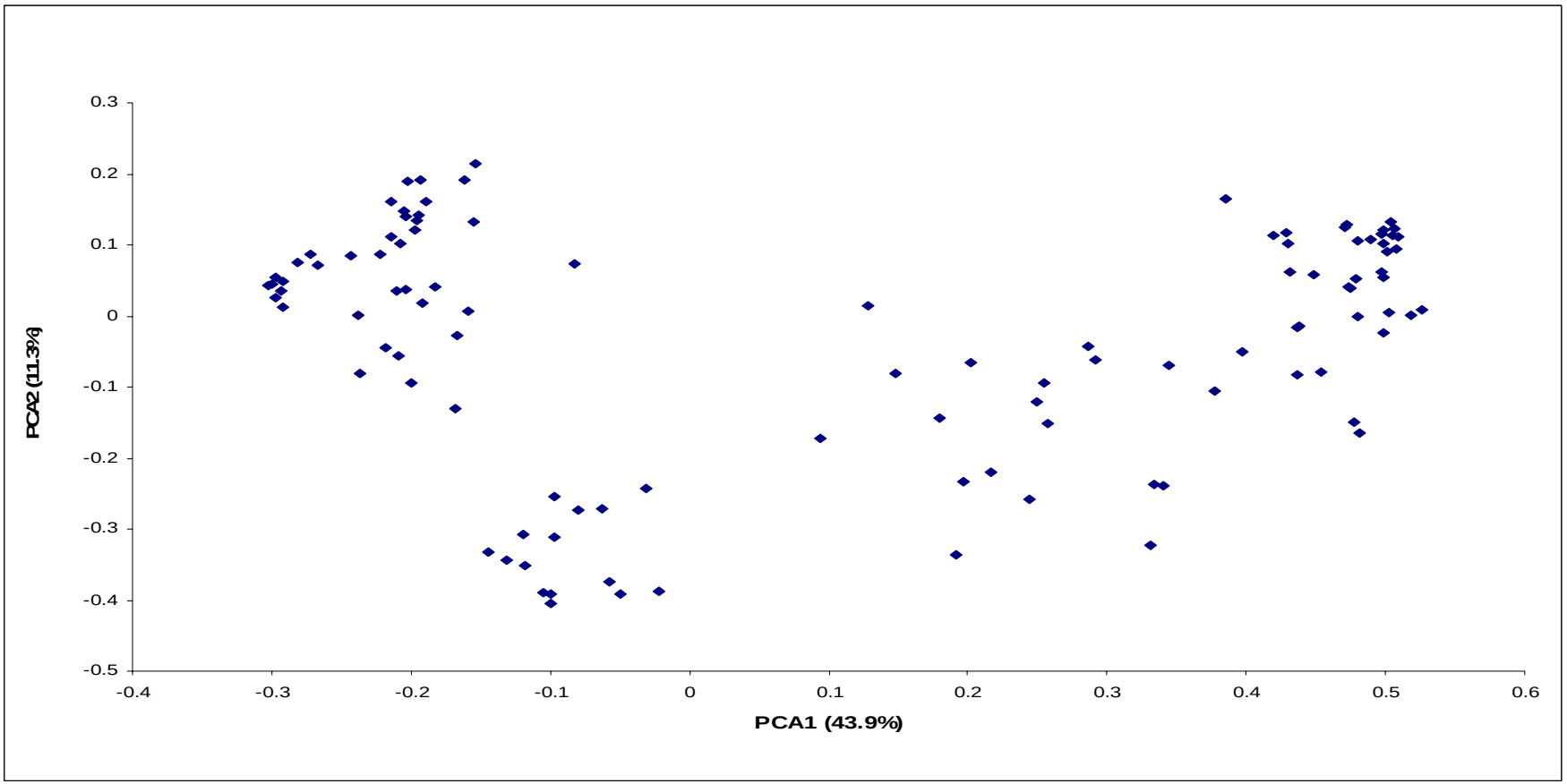

Figure 7 Principal component analysis showing distribution of mungbean accessions in the core collection.

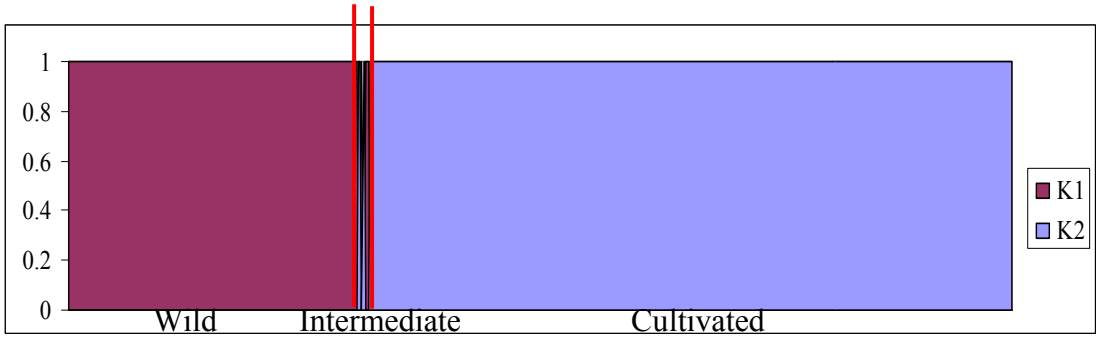

Figure 8 The structure of population was divided by Bayesian method at K2 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications.

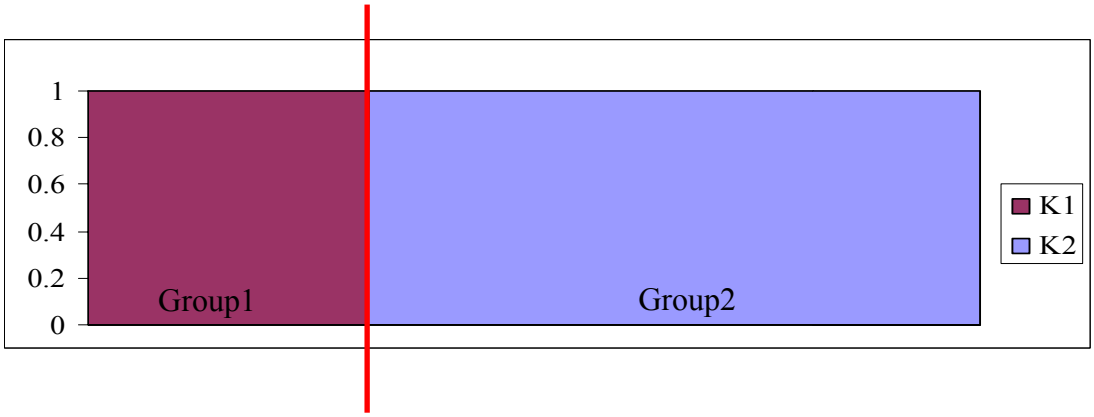

Figure 9 The structure of population was divided by Bayesian method at K2 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications. The data were sorted by Q-value.

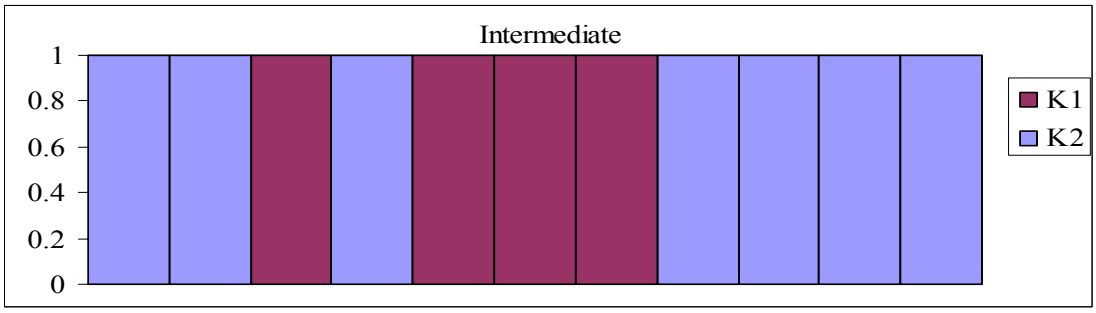

Figure 10 The structure of the intermediate population was divided by Bayesian method at K2 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications.

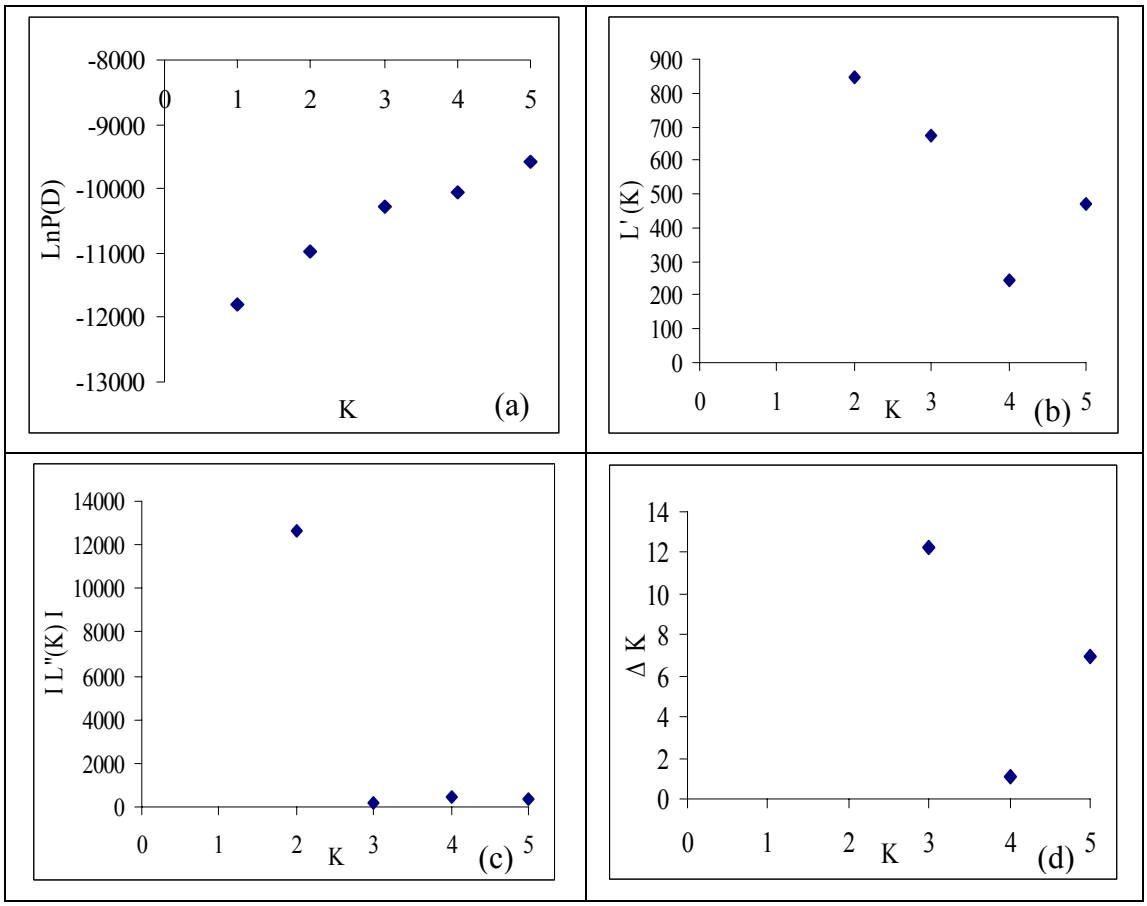

Figure 11 True group of wild mungbean was evaluated by equation step (Evano *et al*., 2005). (a) mean likelihood distribution of L(K)1 to 5, (b) mean difference between successive likelihood values of K, $L'(K) = L(K) - L(K - 1)$ distribution, (c) absolute value of the difference between successive values of $L'(K)$, $|L''(K)| = |L'(K + 1) - L'(K)|$ distribution, (d) mean of the absolute values of $L''(K)$ averaged over 20 runs divided by the standard deviation of $L(K)$, $\Delta K = m(|L''(K)|)/s[L(K)]$.

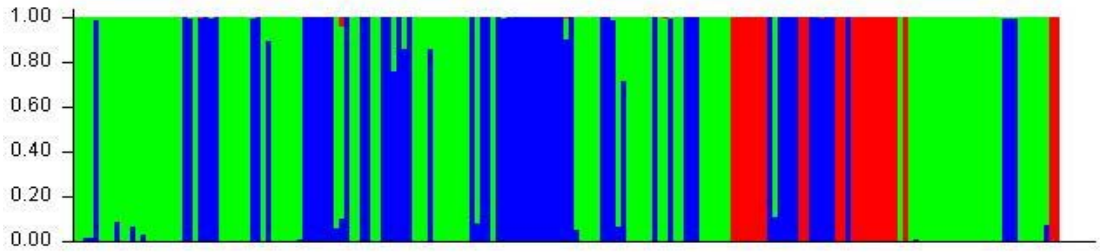

Figure 12 The structure of wild population was divided by Bayesian method at K3 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications.

Figure 13 The structure of wild population was divided by Bayesian method at K3 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications. The data were sorted by Q-value.

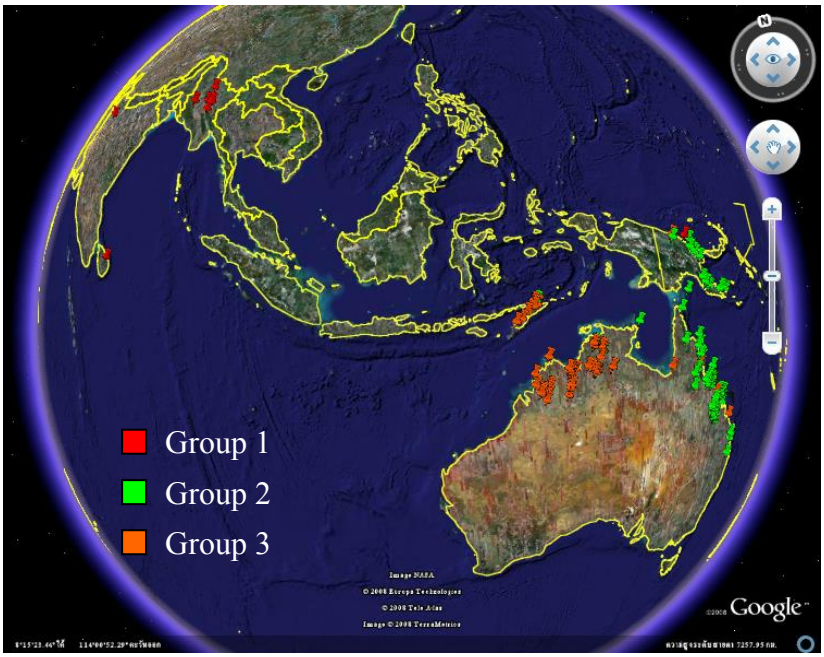

Figure 14 Locations of wild mungbean in each group were assigned by Bayesian method (all group).

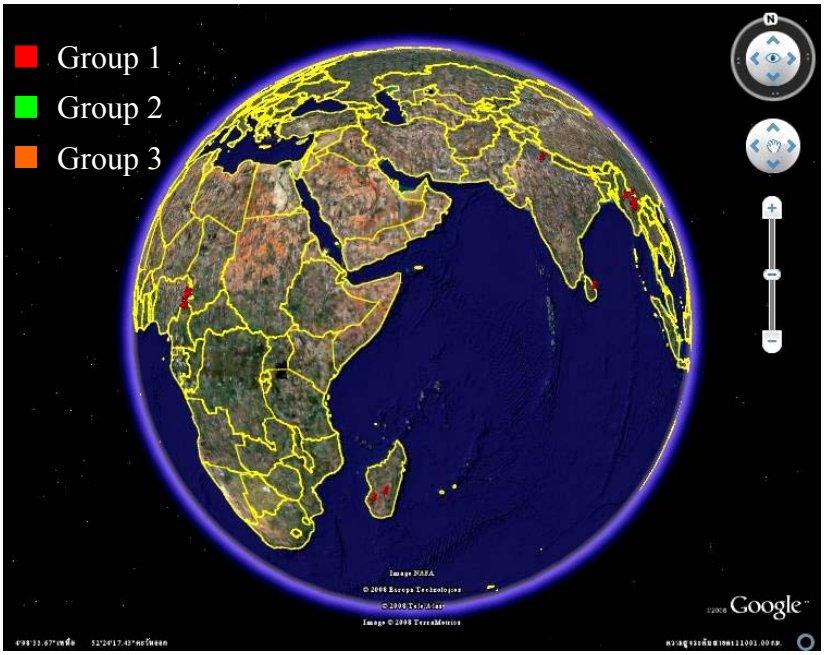

Figure 15 Locations of wild mungbean in each group were assigned by Bayesian method (group 1).

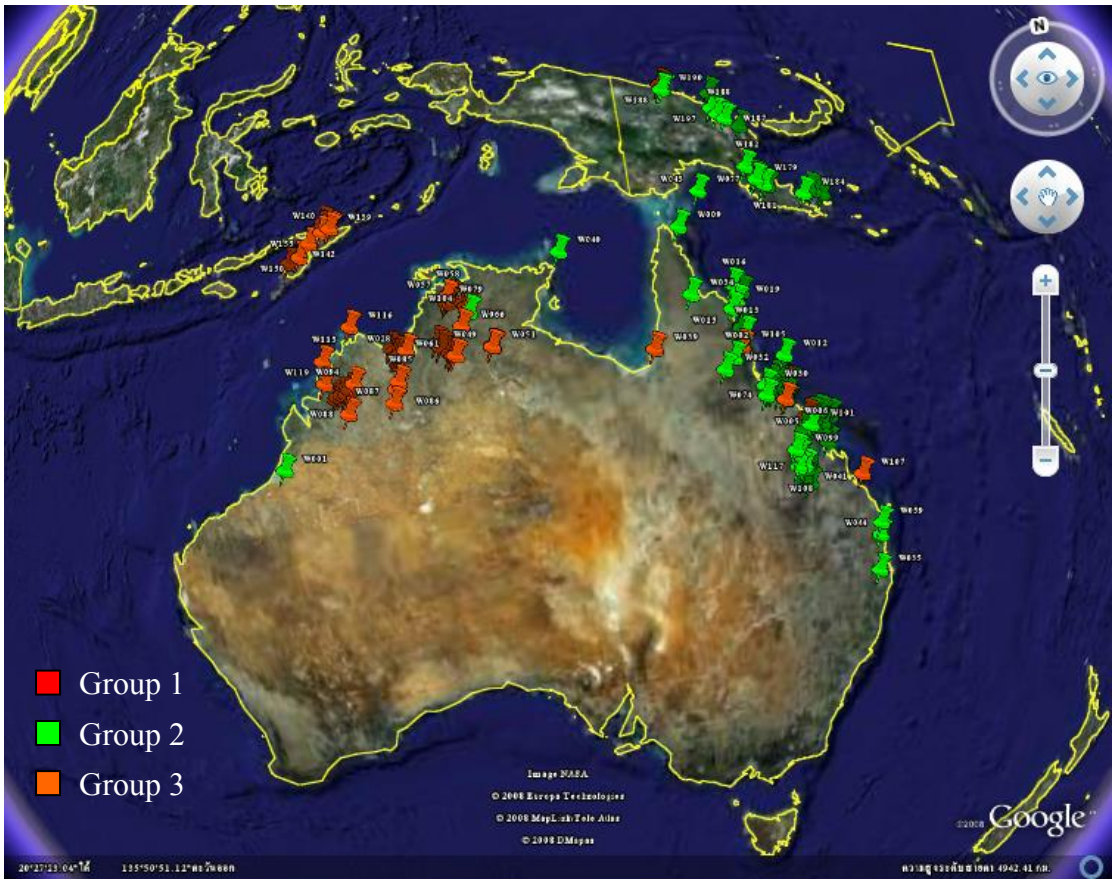

Figure 16 Locations of wild mungbean in each group were assigned by Bayesian method (group 2 and 3).

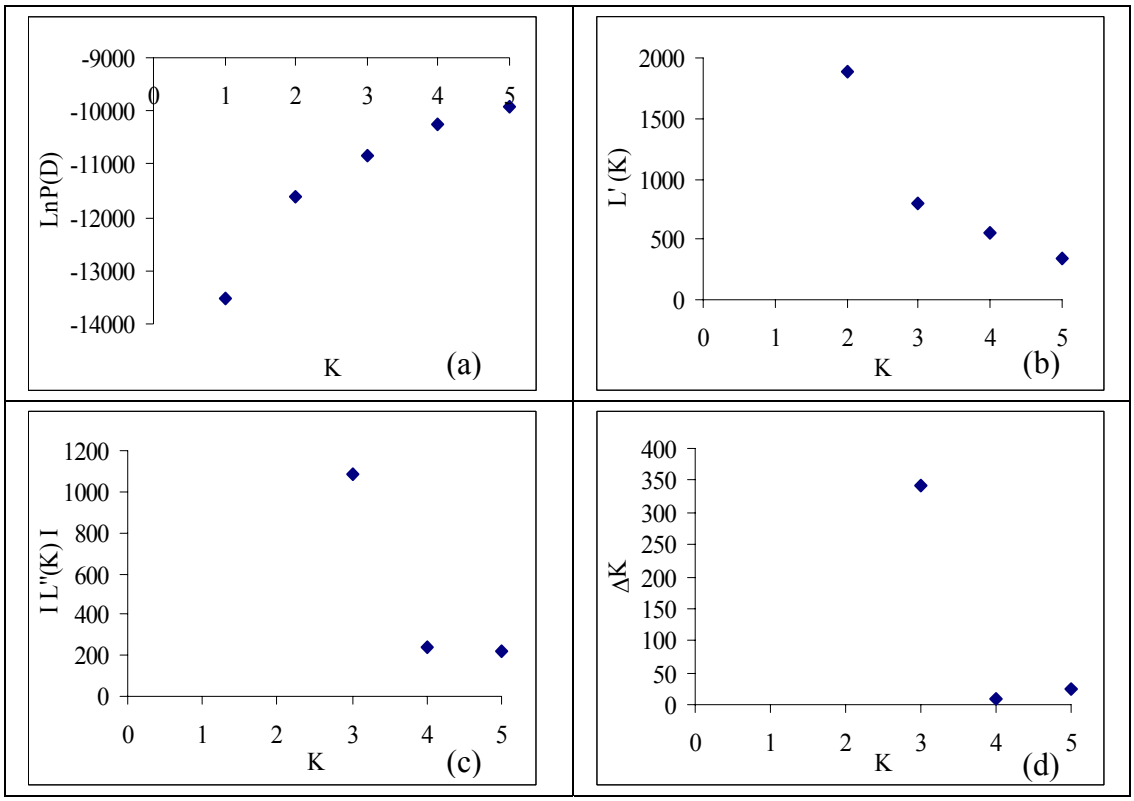

Figure 17 True group of cultivated mungbean was evaluated following equation step by Evano *et al*. (2005). (a) mean likelihood distribution of L(K)1 to 5, (b) mean difference between successive likelihood values of K, $L'(K) = L(K) - L(K - 1)$ distribution, (c) absolute value of the difference between successive values of $L'(K)$, $|L''(K)| = |L'(K + 1) - L'(K)|$ distribution (d) mean of the absolute values of $L''(K)$ averaged over 20 runs divided by the standard deviation of $L(K)$, $\Delta K = m(|L''(K)|)/s[L(K)]$.

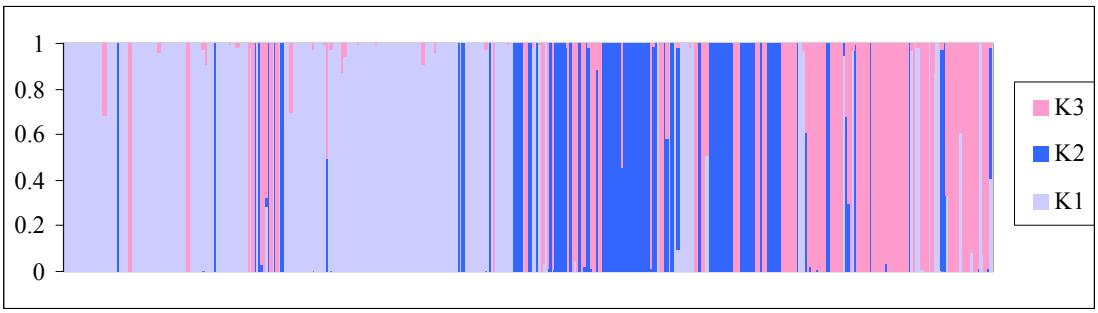

Figure 18 The structure of cultivated mungbean was divided by Bayesian method at K3 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications.

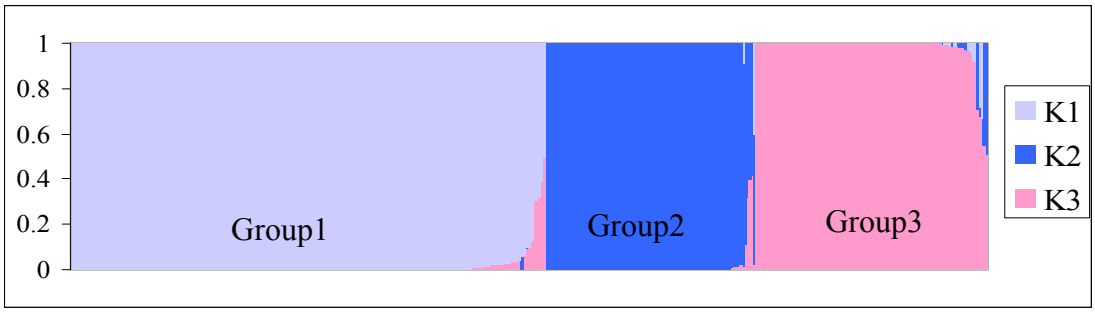

Figure 19 The structure of cultivated mungbean was divided by Bayesian method at K3 under no admixture model with the condition of 100,000 burn-in periods and 500,000 MCMC replications. The data were sorted by Q-value.

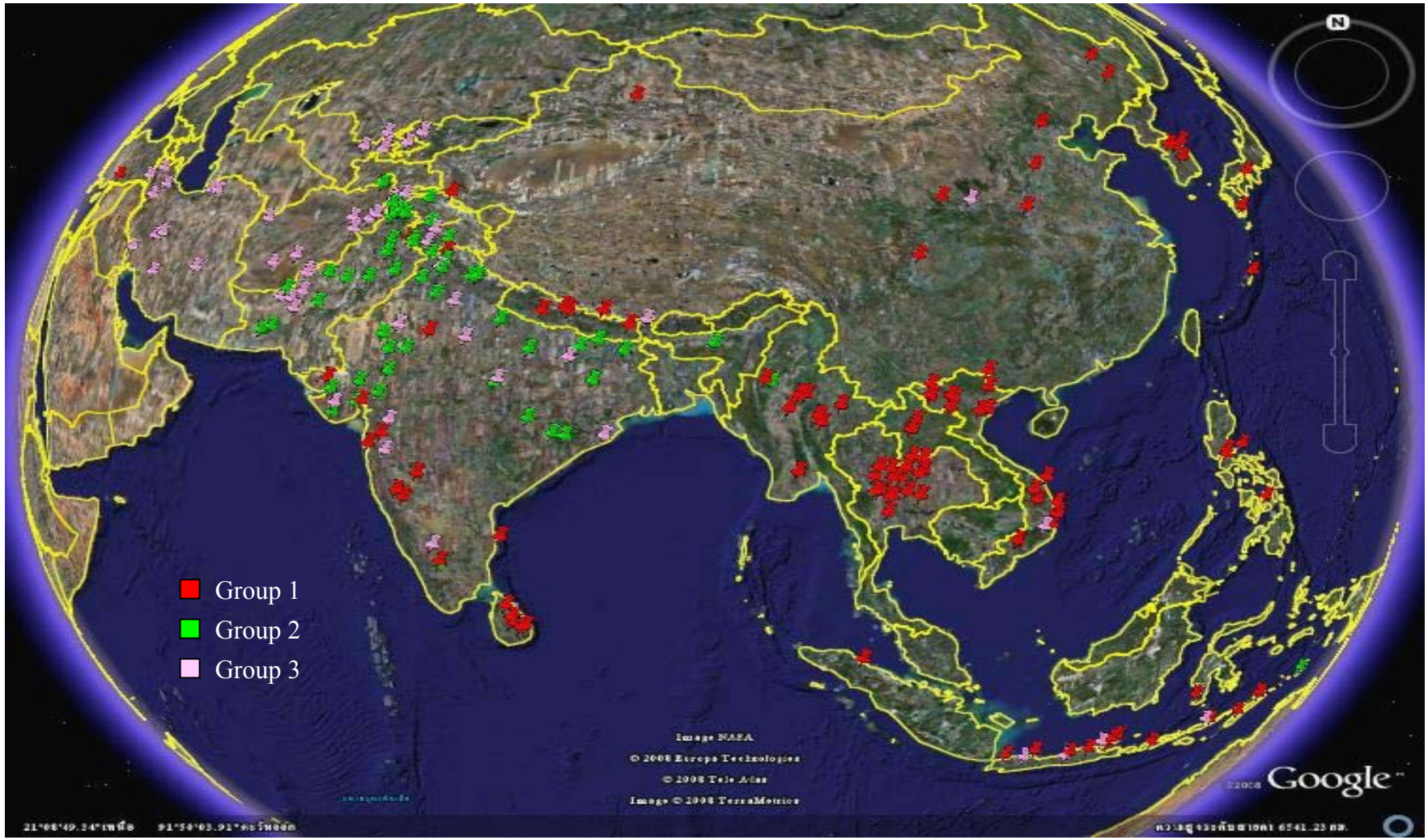

Figure 20 Locations of cultivated mungbean were assigned by Bayesian method at K3.

Discussion

Previous studies using large sets of mungbean genetic resources have focussed on the analysis for morpho-agronomic characteristics (Lawn and Rebetzke, 2006) or protein banding (Tomooka *et al.*, 1992). Small sets (11-12 accessions) of wild mungbean germplasm have been analysed in relation to phenology (Rebetzke and Lawn, 2006a), growth, biomass and seed yield (Rebetzke and Lawn, 2006a), root system (Rebetzke and Lawn, 2006b), and AFLP banding (Saravanakumar *et al*., 2004). In this study, a large collection was analysed with 19 microsatellite markers. The result reveals successful for using cross-species amplification of mungbean. However, the complexity and the possibility of size homoplasy affect from interspecies (Peakall *et al*., 1998). Moreover, this study has few Indian and no West and Central Asia wild include the experiment. The experiment may be no clear for where is mungbean domesticate, however, this study can be apply for mungbean improvement to select the core collection and parents for breeding program in the future. This study represents the first large scale analysis of cultivated and wild mungbean germplasm by molecular markers.

1. Genetic relationship between wild and cultivated mungbean

This study has shown that the cultivated and wild genepools of mungbean are well differentiated (Figure 8). Based on genetic distance (D_A), wild mungbeans from South Asia (India, Myanmar and Sri Lanka) and Africa (Cameroon and Madagascar) or most similar to the cultivated genepool (Table 3). Similarly, cultivated mungbean from South Asian (Bangladesh, India and Pakistan) germplasm is most similar to wild mungbean from mainland Asia and Africa (Table 3). Therefore, results from analysis of SSR allelic diversity support the view that mungbean was domesticated in mainland South Asia. The results also suggest that from there, both wild and cultivated mungbean were introduced into Africa, perhaps recently.

Wild mungbean in India is widely distributed in the Western Ghats and sporadically distributed in Rajastan, Madhya Pradesh and the north-western Himalayas (Arora and Nayar, 1984; Bisht *et al*., 2005). Archaeobotanical remains of *V. radiata* have been found in Neolithic sites in southern India where wild mungbean occurs. In addition, early finding of *V. radiata* occur in Eastern Harappan sites not far from the western Himalayas (Fuller and Harvey, 2006). Since only a few South Asian accessions of wild mungbean were analysed here it is not possible to comment on potential areas in South Asia where domestication occurred. To seek further information related to mungbean domestication it would be helpful to analyse the diversity of the cytoplasmic genomes in wild and cultivated mungbean germplasm.

Domestication results in a genetic bottleneck due to the restricted number of plants involved in domestication (Gepts, 2004). Based on the mungbean germplasm analysed ~50% of the genetic variation present in wild mungbean is found in the cultigen suggesting the genetic bottleneck resulting from domestication is weaker than reported for cereals. Cereals have been characterized by genetic bottlenecks of about 30% or less when considering nucleotide diversity (Buckler *et al*., 2001; Zhu *et al.*, 2007). This may reflect differences in the domestication process of legumes compared to cereals, for example, in relation to wild population size and the domestication related genes that have different developmental origin in legumes compared to cereals such as those associated with shattering and seed dispersal.

2. Genetic diversity of the cultivated mungbean

Gene diversity in cultivated mungbean of Asia is highest in South Asia (India, Bangladesh 0.45, Pakistan 0.46) followed by West Asia (Afghanistan 0.37, Turkey 0.36, Iran and Iraq 0.35) and insular Southeast Asia (Indonesia and East Timor 0.37) (Table 1). High gene diversity was also recorded for Africa (0.40) because two accessions from Madagascar differ greatly from the other African accessions (Figure 6f). The results suggest that cultivated germplasm spread from South Asia to both West Asia and Southeast Asia at possibly a similar time and that it was introduced into Africa more than once.

By using seed protein-banding variation Tomooka *et al*. (1992) studied a large set of 590 cultivated mungbean accessionsand found most genetic diversity in West Asia (Afghanistan, Iran and Iraq). In this current study, most allelic diversity was found in cultivated germplasm from South Asia (India, Bangladesh and Pakistan) followed by West Asia. The difference between results here and those using protein banding may reflect the type of marker used but do point to the high level of mungbean diversity that went to West Asia and has been retained there.

This study supports the hypothesis of Tomooka *et al*. (1992) regarding spread of cultivated mungbean from its presumed areas of domestication in South Asia. Diverse cultivars from South Asia spread to both West Asia and Southeast Asia. While mungbean would have been introduced by land routes to West Asia the high level of diversity in Indonesia suggests early dispersal by sea routes. Restricted germplasm spread to Central Asia and East Asia was perhaps due to selection for mungbean germplasm adapted to its ecological limits at the northern edge of its distribution. The close relationship, based on genetic distance of South Asian and Chinese cultivated mungbean to Indonesian cultivated mungbean suggests introduction of mungbean to Indonesia from both places has occurred. Genetic distance results also suggest that China may have been a source of cultivated mungbean germplasm to other parts of Southeast Asia as a result of migration from China.

Cultivated mungbean in Africa appears to have spread by two routes. A northern route perhaps via West Asia, and a southern route perhaps with the Indonesian expansion to Madagascar that consists of highly diverse germplasm.

3. Wild mungbean in Australasia

This study included a large number of wild mungbean accessions from Australia and neighboring countries (East Timor, Indonesia and Papua New Guinea) where it is indigenous (Lawn and Cottrell, 1988). This germplasm is genetically closely related (D_A = 0.13) but with high allelic diversity. Previous studies have also

shown both the close genetic relationship of germplasm from the extreme east of the wild mungbean distribution range, and also its distinctiveness from germplasm of other regions (Saravanakumar *et al*., 2004). Results here support the view that *V. radiata* var. *sublobata* has a long history in Australasia (Lawn and Cottrell, 1988; Saravanakumar *et al*., 2004).

Wild germplasm of *V. radiata* var. *sublobata* in Australia and neighbouring countries comes from a wide range of ecological habitats including riverbanks, savannah grassland and lightly wooded areas (Lawn and Watkinson, 2002; Tomooka *et al*., 2006b; Vaughan *et al*., 2006). Some morphological characters of accessions reflect the ecologically diverse habitats this germplasm comes from. For example wild mungbean that comes from clay soils of central Queensland, Australia, where an extended dry season occurs, have well-developed taproots (Lawn and Watkinson, 2002; Rebetzke and Lawn, 2006b). However, wild mungbean from other habitats, such as the permanently wet alluvial banks of the Sepik river of Papua New Guinea, have extensive fibrous roots.

4. Genetic structure and location of genetic structure

Genetic structure of mungbean gene pool was divided into two groups as have been reported. Wild mungbean gene pool was separated into three groups supporting the view that wild mungbean in Australia is distinct from germplasm of other regions (Saravanakumar *et al*., 2004). *V. radiata* var. *sublobata* has a long history in Australasia (Lawn and Cottrell, 1988; Saravanakumar *et al*., 2004). However, this experiment has a rather small population from South and Southeast wild mungbean germplasm.

Wild mungbean in Australia and neighbouring countries comes from a wide range of ecological habitats and geography (Lawn and Watkinson, 2002; Tomooka *et al*., 2006b; Vaughan *et al*., 2006). These results showed different genetic structure between wild accessions come from Indean and Pacific coastline. Wild mungbean accessions from West Australia and East Timor of Indian coastline was classified into

one group and accessions from Queensland and Papua New Guinea of Pacific coastline was identified into another one. Genetic structure reflects the ecologically diverse habitats this germplasm comes from.

Cultivated mungbean gene pool was divided into three groups. However, there is no clear geopgrapical genetic structure, as it might be transported by human (Vaughan *et al*., 2006). The genetic structure might be related to seed color. One group of cultivated mungbean comes from wide range of countries with ~97% of seed colors are green and yellow seed color. Two and Three group come from India-Pakistan and neighbouring countries with ~40 and 50 % had primitive wild seed colors, other cultivated seed colors are only 50-60%. This result supported that mungbean might be twice domestricated (Fuller and Harvey, 2006) as the in India-Pakistan varieties show two genetic structures together with primive seed color in this area.

5. Core collection development

The core collection developed here represents 17% (106 accessions) of the original collection (615 accessions) (Table 5). This is a higher proportion than the 10% originally proposed by Frankel and Brown (1984). However, this set has ~50 accessions each of wild and cultivated mungbean and that should enable either the wild and/or cultivated germplasm to be easily evaluated for traits of importance. It would be expected that the wild germplasm may be a useful source of pest, disease and abiotic stress resistance while the cultivated germplasm may be most useful for traits related to improved plant type or life cycle.

This study represents the first large-scale genome level analysis of the mungbean crop complex. While the collection analyzed is poorly represented in germplasm from some areas, particularly wild mungbean from South Asia, the broad relationships among components of the mungbean complex have been revealed. This study should assist mungbean breeders in selecting germplasm for evaluation and use

in breeding programs. Further efforts are needed to develop mungbean molecular linkage map that resolves the 11 linkage groups.

CONCLUSION AND RECOMMENDATION

Conclusion

A large representative collection of mungbean [*Vigna radiata* (L.) Wilczek] consisting of 415 cultivated, 189 wild and 11 intermediate accessions were analyzed using 19 SSR primers. These SSR primers were developed from azuki bean [*V. angularis* (Willd.) Ohwi & Ohwi], and showed polymorphism in wild and cultivated mungbean. One or more SSR locus from each azuki linkage group was analyzed. In total, 309 alleles were detected and of these about twice as many were detected in wild (257 alleles) compared to cultivated accessions (138 alleles). The results show that cultivated mungbean has its greatest diversity in South Asia, which supports the view that South Asia is where this crop was domesticated. SSR marker allelic diversity for cultivated mungbean has a distinct regional distribution with high variation in South and West Asia. Australia and New Guinea represent a distinct center of diversity for wild mungbean. Based on Bayesian algorithm, the entire population was separated into two subgroups with largely belong to two subspecies. Each subspecies was further subdivided into three sub-subgroups. Wild mungbean has a rather clear geographical genetic structure, although the cultivated mungbean has not revealed geographical genetic structure. This study represents the first comprehensive diversity analysis of cultivated and wild mungbean germplasm. Based on the SSR marker diversity 106 accessions were selected for a useful core collection. This study highlights specific genetic diversity that might be used to broaden the genetic base of currently grown mungbean cultivars. Although *Vigna radiata* is morphologically diverse and widely distributed, a relatively few SSR markers created polymorphic alleles in the genepools as compare to the other *Vigna* spp.

Recommendation

The results from this study provided the basic information on DNA fragment information found in both cultivated and wild mungbeans. It can also be used as information for selecting parents in mungbean breeding project that will eventually accomplish the goal of increasing genetic variation in mungbean in the future.

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APPENDIX

Appendix Table 1 Accession number and origin of mungbean samples analyzed

No.	Label	Status	Accessions no.	Country	Province	Collection site
1	H001	Cultivated	JP212394	Japan	Okinawa	Okinawa
2	H002	Cultivated	JP31224	Japan	Kagawa	Kagawa
3	H003	Cultivated	JP31225	Japan	Kagawa	Kagawa
4	H004	Cultivated	JP53827	Japan	Unknown	Unknown
5	H005	Cultivated	JP74739	Japan	Okinawa	Okinawa
6	H006	Cultivated	JP74740	Japan	Okinawa	Okinawa
7	H007	Cultivated	JP78922	Japan	Okinawa	Okinawa
8	H008	Cultivated	JP78923	Japan	Kagoshima	Kagoshima
9	H009	Cultivated	JP78924	Japan	Okinawa	Okinawa
10	H010	Cultivated	JP78971	Japan	Kagoshima	Kagoshima
11	M200	Intermediate	JP 225160	Japan	Saga	Hottate
12	M201	Intermediate	JP 226798	Japan	Saga	Hyogomachi-Kawara
13	M202	Intermediate	JP 226803	Japan	Saga	Hyogomachi-Kawara
14	H011	Cultivated	JP229109	Korea, South	ChungChonh NamDo	ChungChonh NamDo
15	H012	Cultivated	JP229109	Korea, South	ChungChonh NamDo	ChungChonh NamDo
16	H013	Cultivated	JP229110	Korea, South	Kangwondo	Kangwondo
17	H014	Cultivated	JP229112	Korea, South	Kyonggido	Kyonggido
18	H015	Cultivated	JP229115	Korea, South	Kyonggido	Kyonggido
19	H016	Cultivated	JP229117	Korea, South	Kyonggido	Kyonggido
20	H017	Cultivated	JP229121	Korea, South	Chuongchong Bukto	Chuongchong Bukto
21	H018	Cultivated	JP229122	Korea, South	Chuongchong Bukto	Chuongchong Bukto
22	H019	Cultivated	JP229127	Korea, South	Chuongchong Bukto	Chuongchong Bukto
23	H020	Cultivated	JP229128	Korea, South	Chuongchong Bukto	Chuongchong Bukto

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
24	H021	Cultivated	JP229137	China	Jilin	Jilin
25	H022	Cultivated	JP229144	China	Shaanxi	Shaanxi
26	H023	Cultivated	JP229145	China	Henan	Henan
27	H024	Cultivated	JP229146	China	Hebei	Hebei
28	H025	Cultivated	JP229104	China	Beijing	Beijing
29	H026	Cultivated	JP229105	China	Harbin	Harbin
30	H027	Cultivated	JP229215	China	Shanxi	Xian
31	H028	Cultivated	JP229215	China	Shanxi	Xian
32	H029	Cultivated	JP229215	China	Shanxi	Xian
33	H030	Cultivated	JP229216	China	Shanxi	Xian
34	H031	Cultivated	JP229219	China	Beijing	Beijing
35	H032	Cultivated	JP229217	China	Heilongjiang	Harbin
36	H033	Cultivated	JP229218	China	Chengdu, Sichuan	Chengdu
37	H034	Cultivated	-	China	Urumqi	Turpan
38	H035	Cultivated	JP31240	China	-	-
39	H036	Cultivated	JP31241	China	-	-
40	H037	Cultivated	JP31243	China	-	-
41	H038	Cultivated	JP31242	China	-	-
42	H039	Cultivated	JP98915	China	-	-
43	H040	Cultivated	JP73326	China	-	-
44	M135	Intermediate	-	China	-	-
45	M136	Intermediate	-	China	-	-
46	H041	Cultivated	JP98831	Taiwan	-	-

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
47	H042	Cultivated	JP98832	Taiwan	-	-
48	H043	Cultivated	JP98834	Taiwan	-	-
49	H044	Cultivated	JP98835	Taiwan	-	-
50	H045	Cultivated	JP98836	Taiwan	-	-
51	H046	Cultivated	JP98840	Taiwan	-	-
52	H047	Cultivated	JP31246	Taiwan	-	-
53	H048	Cultivated	JP31247	Taiwan	-	-
54	H049	Cultivated	JP31248	Taiwan	-	-
55	H050	Cultivated	JP99049	Taiwan	-	-
56	H051	Cultivated	-	Philippines	Mandaluyoung	Mandaluyoung
57	H052	Cultivated	-	Philippines	Quezon	Cubao
58	H053	Cultivated	-	Philippines	Quezon	Cubao
59	H054	Cultivated	-	Philippines	Cebu	Cebu
60	H055	Cultivated	JP98754	Philippines	-	-
61	H056	Cultivated	JP31254	Philippines	-	-
62	H057	Cultivated	JP98759	Philippines	-	-
63	H058	Cultivated	JP98851	Philippines	-	-
64	H059	Cultivated	JP98862	Philippines	-	-
65	H060	Cultivated	JP98868	Philippines	-	-
66	H061	Cultivated	JP98956	Philippines	-	-
67	H062	Cultivated	JP98957	Philippines	-	-
68	H063	Cultivated	JP73329	Philippines	-	-
69	H064	Cultivated	JP73331	Philippines	-	-

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
70	H065	Cultivated	JP73193	Philippines	-	UPCA
71	H066	Cultivated	JP73194	Philippines	-	UPCA
72	H067	Cultivated	JP98990	Philippines	-	-
73	H068	Cultivated	JP98998	Philippines	-	-
74	H069	Cultivated	JP99003	Philippines	-	-
75	H070	Cultivated	JP99095	Philippines	-	-
76	H071	Cultivated	-	Philippines	-	-
77	H072	Cultivated	-	Indonesia	Wonogiri	Wonogiri
78	H073	Cultivated	-	Indonesia	Makassar	Makassar
79	H074	Cultivated	-	Indonesia	Jatibarang	Jatibarang
80	H075	Cultivated	-	Indonesia	Jatibarang	Jatibarang
81	H076	Cultivated	-	Indonesia	Makassar	Makassar
82	H077	Cultivated	-	Indonesia	Makassar	Makassar
83	H078	Cultivated	-	Indonesia	Pamekasan	Pamekasan
84	H079	Cultivated	-	Indonesia	Pamekasan	Pamekasan
85	H080	Cultivated	JP229132	Indonesia	West Timor	NTT Livestock project
86	H081	Cultivated	JP229133	Indonesia	Sabu	Sabu
87	H082	Cultivated	JP229133	Indonesia	Sabu	Sabu
88	H083	Cultivated	JP229134	Indonesia	Madura island	Madura island
89	H084	Cultivated	JP229135	Indonesia	Madura island	Madura island
90	H085	Cultivated	JP229136	Indonesia	Madura island	Madura island
91	H086	Cultivated	JP229138	Indonesia	Sumbawa Besar NTB	Sumbawa Besar NTB
92	H087	Cultivated	JP229138	Indonesia	Sumbawa Besar NTB	Sumbawa Besar NTB

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
93	H088	Cultivated	JP229220	Indonesia	Soemenep	Soemenep
94	H089	Cultivated	JP229221	Indonesia	Oost Flore	Oost Flore
95	H090	Cultivated	JP229222	Indonesia	Makassar	Makassar
96	H091	Cultivated	JP229223	Indonesia	Ketjamatan	Ketjamatan
97	H092	Cultivated	JP229224	Indonesia	Timur	Timur
98	H093	Cultivated	JP29225	Indonesia	Timur	Timur
99	H094	Cultivated	JP229226	Indonesia	Tegal	Tegal
100	H095	Cultivated	JP229227	Indonesia	Makassar	Makassar
101	H096	Cultivated	JP229228	Indonesia	Djawa	Djawa
102	H097	Cultivated	JP229229	Indonesia	Ngandguk	Ngandguk
103	H098	Cultivated	JP229230	Indonesia	Probolingo	Probolingo
104	H099	Cultivated	JP229230	Indonesia	Probolingo	Probolingo
105	H100	Cultivated	JP229231	Indonesia	Bandjar	Bandjar
106	H101	Cultivated	JP229232	Indonesia	Soemenep	Soemenep
107	H102	Cultivated	JP229233	Indonesia	Makassar	Makassar
108	H103	Cultivated	JP229234	Indonesia	Makassar	Makassar
109	H104	Cultivated	JP229235	Indonesia	Sulawesi	Sulawesi
110	H105	Cultivated	JP229236	Indonesia	Sulawesi	Sulawesi
111	H106	Cultivated	JP229237	Indonesia	Ketjamatan	Ketjamatan
112	W150	Wild	-	Indonesia	Lesser Sunda	Lesser Sunda
113	W154	Wild	-	Indonesia	Lesser Sunda	Lesser Sunda
114	W155	Wild	-	Indonesia	Lesser Sunda	Lesser Sunda

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
115	W156	Wild	-	Indonesia	Lesser Sunda	Lesser Sunda
116	W157	Wild	JP 202267	Indonesia	-	-
117	W158	Wild	JP 202270	Indonesia	-	-
118	W159	Wild	JP 202271	Indonesia	-	-
119	W160	Wild	JP 227257	Indonesia	E. Nusatenggara	E. Nusatenggara
120	M151	Intermediate	-	Indonesia	Lesser Sunda	Lesser Sunda
121	M152	Intermediate	-	Indonesia	Lesser Sunda	Lesser Sunda
122	M153	Intermediate	-	Indonesia	Lesser Sunda	Lesser Sunda
123	H108	Cultivated	JP226587	Timor-Leste	Baucau	Baucau
124	H109	Cultivated	JP226600	Timor-Leste	Viqueque	Viqueque
125	W137	Wild	JP 226583	Timor-Leste	Dili	Dili
126	W138	Wild	JP 226584	Timor-Leste	Manatuto	Manatuto
127	W139	Wild	JP 226601	Timor-Leste	Manatuto	Manatuto
128	W140	Wild	JP 226602	Timor-Leste	Manatuto	Manatuto
129	W141	Wild	JP 226609	Timor-Leste	Ainaro	Ainaro
130	W142	Wild	JP 226612	Timor-Leste	Ainaro	Ainaro
131	H110	Cultivated	JP78939	Vietnam	Bac Thai Prov	Phu Yen
132	H111	Cultivated	JP78941	Vietnam	Cao Bang	Quang Hoa
133	H112	Cultivated	JP78944	Vietnam	Lang Son Prov	Huu Lung Dis
134	H113	Cultivated	JP78945	Vietnam	Da Nang	Da Nang
135	H114	Cultivated	JP78947	Vietnam	Gia Lai	Chu Se
136	H115	Cultivated	JP78948	Vietnam	Kon Tum	Kon Tum
137	H116	Cultivated	JP78950	Vietnam	Dac Lac	Cum Gar

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
138	H117	Cultivated	JP78952	Vietnam	Song Be	Tan Uyen
139	H118	Cultivated	JP78954	Vietnam	Dong Nai	Bien Hoa
140	H119	Cultivated	JP78956	Vietnam	Lam Dong	Duc Trang
141	H120	Cultivated	JP78958	Vietnam	Ninh Thuan	Ninh Son
142	H121	Cultivated	JP78963	Vietnam	Khanh Hoa	Ninh Hoa
143	H122	Cultivated	JP78964	Vietnam	Khanh Hoa	Van Ninh
144	H123	Cultivated	JP78966	Vietnam	Phu Yen	Tuy An
145	H124	Cultivated	JP78967	Vietnam	Binh Dinh	An Nhan
146	H125	Cultivated	JP78969	Vietnam	Quang Ngai	Quang Ngai
147	H126	Cultivated	JP207879	Vietnam	Son La	Yen Chau
148	H127	Cultivated	JP207883	Vietnam	Son La	Mai Son
149	H128	Cultivated	JP207905	Vietnam	Lai Chau	Dien Bien
150	H129	Cultivated	JP207919	Vietnam	Lai Chau	Lai Chau
151	H130	Cultivated	JP226668	Laos	Houa Phan	Xam Neua
152	H131	Cultivated	JP226686	Laos	Sayabouli	Sayabouli
153	H132	Cultivated	JP226692	Laos	Sayabouli	Sayabouli
154	H133	Cultivated	JP226695	Laos	Luang Prabang	Muang Thadua
155	H134	Cultivated	JP226698	Laos	Luang Prabang	Luang Prabang
156	H135	Cultivated	-	Thailand	Pichit	Taphan Hin
157	H136	Cultivated	-	Thailand	Nakhon Sawan	Lat Yao
158	H137	Cultivated	-	Thailand	Lopburi	Chai Badan
159	H138	Cultivated	-	Thailand	Singburi	Ranam
160	H139	Cultivated	-	Thailand	Chai Nat	Homkrajui
161	H140	Cultivated	-	Thailand	Nongbualamphu	Non Mueang

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
162	H141	Cultivated	-	Thailand	Phitsanulok	Wang Thong
163	H142	Cultivated	-	Thailand	Phetchabun	Chondaen
164	H143	Cultivated	-	Thailand	Khonkean	Chum Phae
165	H144	Cultivated	-	Thailand	Loei	Loei
166	H145	Cultivated	-	Thailand	Phetchabun	Lom Sak
167	H146	Cultivated	-	Thailand	-	-
168	H147	Cultivated	JP229067	Thailand	Phetchabun	Ban Rai
169	H148	Cultivated	-	Thailand	-	-
170	H149	Cultivated	JP229096	Thailand	Sukhothai	Sukhothai
171	H150	Cultivated	JP229097	Thailand	Kamphaeng Phet	Kamphaeng Phet
172	H151	Cultivated	JP229098	Thailand	Nakhon Phathom	Kamphaeng saen
173	H152	Cultivated	JP229099	Thailand	Uthai Thani	Lan Sak
174	H153	Cultivated	JP229077	Thailand	Chaiyaphum	Khon San
175	H154	Cultivated	JP229082	Thailand	Nakhon Ratchasima	Sung Noen
176	H155	Cultivated	JP81647	Thailand	-	-
177	H156	Cultivated	JP110830	Thailand	-	-
178	H157	Cultivated	-	Thailand	Chinat	Chinat
179	H158	Cultivated	JP81648	Thailand	-	-
180	H159	Cultivated	-	Thailand	-	-
181	H160	Cultivated	-	Thailand	-	-
182	H161	Cultivated	-	Thailand	-	-
183	H162	Cultivated	-	Thailand	-	-
184	H163	Cultivated	JP211977	Myanmar	Shan	Min Thaut

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
185	H164	Cultivated	JP211977	Myanmar	Shan	Min Thaut
186	H165	Cultivated	JP211982	Myanmar	Shan	Methway Phote
187	H166	Cultivated	JP211982	Myanmar	Shan	Methway Phote
188	H167	Cultivated	JP212131	Myanmar	Mandalay	Mandalay
189	H168	Cultivated	JP212132	Myanmar	Mandalay	Mandalay
190	H169	Cultivated	JP212149	Myanmar	Mandalay	Pyin Oo Lwin
191	H170	Cultivated	JP217422	Myanmar	Ayeyarwaddy	Nyaungdon township
192	H171	Cultivated	JP217422	Myanmar	Ayeyarwaddy	Nyaungdon township
193	H172	Cultivated	JP217423	Myanmar	Ayeyarwaddy	Nyaungdon township
194	H173	Cultivated	JP217423	Myanmar	Ayeyarwaddy	Nyaungdon township
195	H174	Cultivated	JP217430	Myanmar	Shan	Taunggyi
196	H175	Cultivated	JP217430	Myanmar	Shan	Taunggyi
197	H176	Cultivated	JP217432	Myanmar	Shan	Taunggyi
198	H177	Cultivated	JP217432	Myanmar	Shan	Taunggyi
199	H178	Cultivated	JP217473	Myanmar	Magway	Gangaw
200	H179	Cultivated	JP217501	Myanmar	Sagaing	Kalemyo
201	H180	Cultivated	JP217510	Myanmar	Magway	Pakoku
202	H181	Cultivated	JP217511	Myanmar	Magway	Pakoku
203	H182	Cultivated	-	Myanmar	Mandalay	Pyin Oo Lwin
204	W164	Wild	JP 210796	Myanmar	Mandalay	Kyauk Tham Pat
205	W165	Wild	JP 210804	Myanmar	Shan	Kalaw
206	W166	Wild	JP 211874	Myanmar	Mandalay	Pyin-Oo-Lwin
207	W167	Wild	JP 217427	Myanmar	Shan	Kyawt Nge
208	W168	Wild	JP 217436	Myanmar	Shan	Kalaw
209	W169	Wild	JP 217437	Myanmar	Shan	Khar Lein
210	W170	Wild	JP 217469	Myanmar	Sagain	Ggangaw

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
211	H183	Cultivated	JP229139	Nepal	-	-
212	H184	Cultivated	JP229140	Nepal	-	-
213	H185	Cultivated	JP229141	Nepal	-	-
214	H186	Cultivated	JP229143	Nepal	-	-
215	H187	Cultivated	JP85556	Nepal	Ghorahi	Ghorahi
216	H188	Cultivated	JP85557	Nepal	Dhankuta	Dhankuta
217	H189	Cultivated	JP108249	Nepal	Patichaur	Patichaur
218	H190	Cultivated	JP108250	Nepal	Tatopani	Tatopani
219	H191	Cultivated	JP108251	Nepal	Tatopani	Tatopani
220	M207	Intermediate	-	Nepal	Jhapa	Chanawa
221	H192	Cultivated	JP229130	Bangladesh	-	-
222	H193	Cultivated	JP229131	Bangladesh	-	-
223	H194	Cultivated	-	India	Pune	Pune
224	H195	Cultivated	JP105628	India	Solapur	Solapur
225	H196	Cultivated	JP229149	India	Punjab	Katla
226	H197	Cultivated	JP229149	India	Punjab	Katla
227	H198	Cultivated	JP229206	India	Coimbatore	Coimbatore
228	H199	Cultivated	JP229196	India	Chatsu	Chatsu
229	H200	Cultivated	JP229179	India	Bombay	Hubli
230	H201	Cultivated	JP229178	India	Bombay	Belgaum
231	H202	Cultivated	JP229178	India	Bombay	Belgaum
232	H203	Cultivated	JP229209	India	United	Dehra Dun
233	H204	Cultivated	JP229212	India	United	Mussoorie

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
234	H205	Cultivated	JP229213	India	United	Tehri
235	H206	Cultivated	JP229195	India	Jodhpur	Araba
236	H207	Cultivated	JP229161	India	Kathiawar	Bhavnagar
237	H208	Cultivated	JP229165	India	Kathiawar	Junagadh
238	H209	Cultivated	JP229170	India	Kathiawar	Veraval
239	H210	Cultivated	JP229184	India	Central	Nawapara
240	H211	Cultivated	JP229163	India	Cutch	Bhuj
241	H212	Cultivated	JP229163	India	Cutch	Bhuj
242	H213	Cultivated	JP229189	India	Assam	Shillong
243	H214	Cultivated	JP229187	India	Bombay	Mahabaleshwar
244	H215	Cultivated	JP229180	India	Mysore	Mysore
245	H216	Cultivated	JP229158	India	New Delhi	New Delhi
246	H217	Cultivated	JP229186	India	Igatpuri bazaar	Igatpuri bazaar
247	H218	Cultivated	JP229172	India	Mandi	Mandi
248	H219	Cultivated	JP229200	India	Jodhpur	Jodhpur
249	H220	Cultivated	JP229204	India	Assam	Sikkim
250	H221	Cultivated	JP229204	India	Assam	Sikkim
251	H222	Cultivated	JP229202	India	Gwalior	Gwalior
252	H223	Cultivated	JP229197	India	Rajasthan	Desu
253	H224	Cultivated	JP229164	India	Gujrat	Goeing
254	H225	Cultivated	JP229154	India	Ranchi	Ranchi
255	H226	Cultivated	JP229183	India	Jabalpur	Jabalpur
256	H227	Cultivated	JP229188	India	Gwalior	Gwalior

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
257	H228	Cultivated	JP229152	India	Pusa	Pusa
258	H229	Cultivated	JP229181	India	Gwalior	Gwalior
259	H230	Cultivated	JP229181	India	Rajasthan	Pali
260	H231	Cultivated	JP229156	India	Bihar	Sahasram
261	H232	Cultivated	JP229151	India	Arrah	Arrah
262	H233	Cultivated	JP229182	India	Satna	Satna
263	H234	Cultivated	JP229185	India	Sihora	Sihora
264	H235	Cultivated	JP229167	India	Sihora	Sihora
265	H236	Cultivated	JP229157	India	Bihar	Sahasram
266	H237	Cultivated	JP229174	India	Boggar	Boggar
267	H238	Cultivated	JP229173	India	Achabal	Achabal
268	H239	Cultivated	JP229176	India	Ispur	Ispur
269	H240	Cultivated	JP229177	India	Srinagar	Srinagar
270	H241	Cultivated	JP229177	India	Srinagar	Srinagar
271	H242	Cultivated	JP229175	India	Dhariwal	Dhariwal
272	H243	Cultivated	JP229193	India	Punjab	Amraili
273	H244	Cultivated	JP229194	India	Bhatinda	Bhatinda
274	H245	Cultivated	JP229171	India	Hansi	Hansi
275	H246	Cultivated	JP229205	India	Coimbatore	Coimbatore
276	H247	Cultivated	JP229150	India	Punjab	Samirala
277	H248	Cultivated	JP229211	India	Uttar Pradesh	Jamnagar
278	H249	Cultivated	JP229211	India	Uttar Pradesh	Jamnagar
279	H250	Cultivated	JP2292160	India	Baroda	Baroda

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
280	H251	Cultivated	JP229153	India	Gwalior	Gwalior
281	H252	Cultivated	JP229153	India	Gwalior	Gwalior
282	H253	Cultivated	JP229198	India	Gurlan	Gurlan
283	H254	Cultivated	JP229203	India	Patanpura	Patanpura
284	H255	Cultivated	JP229201	India	Moodi	Moodi
285	H256	Cultivated	JP229166	India	Gujrat	Laiza
286	H257	Cultivated	JP229166	India	Gujrat	Laiza
287	H258	Cultivated	JP229159	India	Gujrat	Barai
288	H259	Cultivated	JP229159	India	Gujrat	Barai
289	H260	Cultivated	JP229168	India	Toda	Toda
290	H261	Cultivated	JP229155	India	Sabaur	Sabaur
291	H262	Cultivated	JP229190	India	Bhawanipatna	Bhawanipatna
292	H263	Cultivated	JP229191	India	Puri	Puri
293	H264	Cultivated	JP229192	India	Puri	Puri
294	H265	Cultivated	JP229208	India	Allahabad	Allahabad
295	H266	Cultivated	JP229210	India	Hardoi	Hardoi
296	H267	Cultivated	JP229207	India	Madras	Madras
297	H268	Cultivated	JP98922	India	-	-
298	H269	Cultivated	JP73313	India	-	-
299	H270	Cultivated	JP99005	India	-	-
300	H271	Cultivated	JP99013	India	-	-
301	H273	Cultivated	JP99039	India	-	-
302	H274	Cultivated	JP99046	India	-	-

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
303	W147	Wild	JP107875	India	-	-
304	W148	Wild	JP110831	India	Rishikesh	Rishikesh
305	M146	Intermediate	-	India	Tamilnadu	Tamilnadu
306	M149	Intermediate	JP 218941	India	Mysore	Mysore
307	H275	Cultivated	-	Sri Lanka	Elukwella	Matale
308	H276	Cultivated	JP187894	Sri Lanka	Elukwella	Matale
309	H277	Cultivated	JP187894	Sri Lanka	Galakulugolla	Moneragala
310	H278	Cultivated	JP187898	Sri Lanka	Galakulugolla	Moneragala
311	H279	Cultivated	JP187898	Sri Lanka	Mihintale	Anuradhapura
312	H280	Cultivated	JP81649	Sri Lanka	Mihintale	Anuradhapura
313	H281	Cultivated	JP81650	Sri Lanka	Kahatagahamadiththa	Monaragala
314	H282	Cultivated	JP81651	Sri Lanka	Monaragala	Siyabalagune Weeawanya
315	H283	Cultivated	JP81652	Sri Lanka	Monaragala	Siyabalagune Weeawanya
316	H284	Cultivated	JP81652	Sri Lanka	Monaragala	Wellawaya
317	W203	Wild	JP 210617	Sri Lanka	Mahiyangana	Welpallewela
318	W204	Wild	JP 217528	Sri Lanka	-	-
319	H285	Cultivated	JP81653	Pakistan	North-West Frontier	Chakiatan
320	H286	Cultivated	JP103111	Pakistan	North-West Frontier	Drosh
321	H287	Cultivated	JP103115	Pakistan	North-West Frontier	Maindam
322	H288	Cultivated	JP103118	Pakistan	North-West Frontier	Dir
323	H289	Cultivated	JP103119	Pakistan	North-West Frontier	Chikar
324	H290	Cultivated	JP81407	Pakistan	Punjab	Dina

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
325	H291	Cultivated	JP103121	Pakistan	Punjab	Main Channu
326	H292	Cultivated	JP73258	Pakistan	Punjab	Mauza Qadimi
327	H293	Cultivated	JP103127	Pakistan	Punjab	Khanwana
328	H294	Cultivated	JP73259	Pakistan	Punjab	Chinji
329	H295	Cultivated	JP73260	Pakistan	Punjab	Chinji
330	H296	Cultivated	JP103128	Pakistan	Punjab	Mair
331	H297	Cultivated	JP73261	Pakistan	Punjab	Mair
332	H298	Cultivated	JP73261	Pakistan	Punjab	Dhok Tayab
333	H299	Cultivated	JP73262	Pakistan	Punjab	Dhok Tayab
334	H300	Cultivated	JP73262	Pakistan	Balochistan	Rarkan
335	H301	Cultivated	JP103131	Pakistan	Balochistan	MikhtR Baz Barad
336	H302	Cultivated	JP73264	Pakistan	Balochistan	Kuchlak
337	H303	Cultivated	JP103132	Pakistan	Balochistan	Surab
338	H304	Cultivated	JP103134	Pakistan	Balochistan	Khuzdar
339	H305	Cultivated	JP103135	Pakistan	Balochistan	Paro Besimia
340	H306	Cultivated	JP103136	Pakistan	Balochistan	Dandar
341	H307	Cultivated	JP103138	Pakistan	Balochistan	Dandar
342	H308	Cultivated	JP103138	Pakistan	Balochistan	Mushkai
343	H309	Cultivated	JP103140	Pakistan	Punjab	Khushab
344	H310	Cultivated	JP73267	Pakistan	Punjab	Shab Din Wal
345	H311	Cultivated	JP73268	Pakistan	Balochistan	Dera Murad Jamali
346	H312	Cultivated	JP103354	Pakistan	Balochistan	Pishin
347	H313	Cultivated	JP103355	Pakistan	Balochistan	Pishin

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
348	H314	Cultivated	JP103356	Pakistan	Balochistan	Noshki
349	H315	Cultivated	JP103357	Pakistan	Balochistan	Kharan
350	H316	Cultivated	JP73270	Pakistan	Balochistan	Mangochar
351	H317	Cultivated	JP103360	Pakistan	Punjab	Multan
352	H318	Cultivated	JP74762	Pakistan	Punjab	Multan
353	H319	Cultivated	JP74763	Pakistan	Punjab	Waind
354	H320	Cultivated	JP74777	Pakistan	North-West Frontier	Surji Gali
355	H321	Cultivated	JP104256	Pakistan	North-West Frontier	Thakot
356	H322	Cultivated	JP74787	Pakistan	North-West Frontier	Sarai Paen
357	H323	Cultivated	JP74797	Pakistan	North-West Frontier	Baroz
358	H324	Cultivated	JP104262	Pakistan	North-West Frontier	Randor-Bakhtol
359	H325	Cultivated	JP104266	Pakistan	North-West Frontier	Shoghor
360	H326	Cultivated	JP104268	Pakistan	North-West Frontier	Mogh
361	H327	Cultivated	JP74804	Pakistan	North-West Frontier	Kari
362	H328	Cultivated	JP104270	Pakistan	North-West Frontier	Kughzi
363	H329	Cultivated	JP74805	Pakistan	North-West Frontier	Parwak
364	H330	Cultivated	JP104274	Pakistan	North-West Frontier	Gulapur
365	H331	Cultivated	JP74809	Pakistan	-	-
366	H332	Cultivated	JP31310	Pakistan	-	-
367	H333	Cultivated	JP98891	Pakistan	-	-
368	H334	Cultivated	JP73290	Pakistan	-	-
369	H335	Cultivated	JP99006	Afghanistan	Kabul	Kabul
370	H336	Cultivated	JP229139	Afghanistan	Kandahar	Kandahar

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
371	H337	Cultivated	JP229240	Afghanistan	Kandahar	Kandahar
372	H338	Cultivated	JP229241	Afghanistan	Helmand	Helmand
373	H339	Cultivated	JP229242	Afghanistan	Jalalabad	Momokhil
374	H340	Cultivated	JP229243	Afghanistan	Heart	Heart
375	H341	Cultivated	JP229244	Afghanistan	Paktia	Paktia
376	H342	Cultivated	JP229245	Afghanistan	Kabul	Kabul
377	H343	Cultivated	JP229246	Afghanistan	Kabul	Kabul
378	H344	Cultivated	JP229246	Afghanistan	Faizabad	Faizabad
379	H345	Cultivated	JP229247	Afghanistan	Nangarhar	Nangarhar
380	H346	Cultivated	JP229248	Afghanistan	-	-
381	H347	Cultivated	JP229250	Afghanistan	-	-
382	H348	Cultivated	JP229251	Afghanistan	-	-
383	H349	Cultivated	JP229252	Afghanistan	-	-
384	H350	Cultivated	JP31320	Afghanistan	-	-
385	H351	Cultivated	JP74721	Afghanistan	-	-
386	H352	Cultivated	JP31322	Afghanistan	-	-
387	H353	Cultivated	JP74724	Afghanistan	-	-
388	H354	Cultivated	JP74726	Afghanistan	-	-
389	H355	Cultivated	JP98828	Afghanistan	-	-
390	H356	Cultivated	JP73291	Afghanistan	-	-
391	H357	Cultivated	JP99009	Afghanistan	-	-
392	H358	Cultivated	JP31324	Afghanistan	-	-

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
393	H359	Cultivated	JP74728	Afghanistan	-	-
394	H360	Cultivated	JP74729	Afghanistan	-	-
395	H361	Cultivated	JP99078	Afghanistan	-	-
396	H362	Cultivated	JP74732	Afghanistan	-	-
397	H363	Cultivated	JP99079	Afghanistan	-	-
398	H364	Cultivated	JP99092	Afghanistan	-	-
399	H365	Cultivated	JP99195	Iran	Khorasan	Khorasan
400	H366	Cultivated	JP229253	Iran	Masanderan	Masanderan
401	H367	Cultivated	JP229254	Iran	Azerbaijan-e Gharbi	Rezaiyeh
402	H368	Cultivated	JP229255	Iran	Azerbaijan-e Sharqi	Tabriz
403	H369	Cultivated	JP229256	Iran	Azerbaijan-e Sharqi	Tabriz
404	H370	Cultivated	JP229256	Iran	Kerman	Kerman
405	H371	Cultivated	JP229257	Iran	Shiraz	Shiraz
406	H372	Cultivated	JP229258	Iran	Ahwaz	Ahwaz
407	H373	Cultivated	JP229260	Iran	Esfahan	Esfahan
408	H374	Cultivated	JP229261	Iran	Chahar Mahal-e Bakhtiari	Saman
409	H375	Cultivated	JP229261	Iran	Chahar Mahal-e Bakhtiari	Saman
410	H376	Cultivated	JP229262	Iran	Isfahan	Isfahan (Esfahan) bazaar
411	H377	Cultivated	JP229263	Iran	Emamshahr (Shahrud)	Emamshahr (Shahrud)
412	H378	Cultivated	JP229264	Iran	Zanjan	Zanjan
413	H379	Cultivated	JP31326	Iran	-	-
414	H380	Cultivated	JP31328	Iran	-	-
415	H381	Cultivated	JP31329	Iran	-	-
416	H382	Cultivated	JP73288	Iran	-	-

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
417	H383	Cultivated	JP31330	Iran	-	-
418	H384	Cultivated	JP31331	Iran	-	-
419	H385	Cultivated	JP98811	Iraq	-	-
420	H386	Cultivated	JP31333	Iraq	-	-
421	H387	Cultivated	JP229103	Turkey	-	-
422	H388	Cultivated	JP229106	Turkey	-	-
423	H389	Cultivated	JP229107	Turkey	Gaziantep	Gaziantep
424	H390	Cultivated	JP229108	Turkey	Harari	Sapatan
425	H391	Cultivated	JP98772	Turkey	-	-
426	H392	Cultivated	JP98774	Turkey	-	-
427	H393	Cultivated	JP73287	Turkey	-	-
428	H394	Cultivated	JP31334	Turkey	-	-
429	H395	Cultivated	JP31335	Turkey	-	-
430	H396	Cultivated	JP98983	Turkey	-	-
431	H397	Cultivated	JP78925	Uzbekistan	Tashkent	Tashkent
432	H398	Cultivated	JP78926	Uzbekistan	Talake	Zizak
433	H399	Cultivated	JP78927	Uzbekistan	Zamin	Lailakuas
434	H400	Cultivated	JP78928	Uzbekistan	Zaharabad	Zaharabad
435	H401	Cultivated	JP78932	Uzbekistan	Saultepa	Saultepa
436	H402	Cultivated	JP78933	Uzbekistan	Fergana	Yagiabad

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
437	H403	Cultivated	JP78934	Uzbekistan	Fergana	Fergana
438	H404	Cultivated	JP78935	Uzbekistan	Fergana	Fergana
439	H405	Cultivated	JP78936	Uzbekistan	Andidjan	Kaleninjan
440	H406	Cultivated	JP78930	Tadzikistan	Leninabad	Cistakos
441	H407	Cultivated	JP78931	Tadzikistan	Leninabad	Karachum
442	H408	Cultivated	JP78929	Kirgizstan	Isfana	Isfana
443	H409	Cultivated	JP78937	Kirgizstan	Kensai	Kensai
444	H410	Cultivated	JP78938	Kirgizstan	Kodksai	Kodksai
445	H411	Cultivated	JP229102	Mauritius	-	-
446	H412	Cultivated	JP98808	Madagascar	-	-
447	H413	Cultivated	JP98934	Madagascar	-	-
448	H414	Cultivated	JP98935	Madagascar	-	-
449	H415	Cultivated	JP98935	Madagascar	-	-
450	W162	Wild	JP107877	Madagascar	-	-
451	W163	Wild	JP 227259	Madagascar	Mahaboboka	Mahaboboka
452	H416	Cultivated	JP212359	Nigeria	-	-
453	H417	Cultivated	JP212360	Nigeria	-	-
454	W001	Wild	-	Australia	Queensland	Georgetown
455	W002	Wild	-	Australia	Queensland	Queensland
456	W003	Wild	-	Australia	Queensland	Queensland
457	W004	Wild	-	Australia	Queensland	Queensland
458	W005	Wild	-	Australia	Queensland	Queensland
459	W006	Wild	-	Australia	Queensland	Queensland

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
460	W007	Wild	-	Australia	Queensland	Queensland
461	W009	Wild	-	Australia	Queensland	Queensland
462	W010	Wild	-	Australia	Queensland	Queensland
463	W011	Wild	-	Australia	Queensland	Queensland
464	W012	Wild	-	Australia	Queensland	Queensland
465	W013	Wild	-	Australia	Queensland	Queensland
466	W014	Wild	-	Australia	Queensland	Queensland
467	W015	Wild	-	Australia	Queensland	Queensland
468	W016	Wild	-	Australia	Queensland	Queensland
469	W017	Wild	-	Australia	Queensland	Queensland
470	W018	Wild	-	Australia	Queensland	Queensland
471	W019	Wild	-	Australia	Queensland	Queensland
472	W020	Wild	-	Australia	Queensland	Queensland
473	W021	Wild	-	Australia	Queensland	Queensland
474	W022	Wild	-	Australia	Queensland	Queensland
475	W023	Wild	-	Australia	Western Australia	Western Australia
476	W024	Wild	-	Australia	Northern Territory	Northern Territory
477	W025	Wild	-	Australia	Northern Territory	Northern Territory
478	W026	Wild	-	Australia	Northern Territory	Northern Territory
479	W027	Wild	-	Australia	Northern Territory	Northern Territory
480	W028	Wild	-	Australia	Northern Territory	Northern Territory
481	W029	Wild	-	Australia	Northern Territory	Northern Territory
482	W030	Wild	-	Australia	Queensland	Queensland
483	W031	Wild	-	Australia	Queensland	Queensland

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
484	W032	Wild	-	Australia	Queensland	Queensland
485	W033	Wild	-	Australia	Queensland	Queensland
486	W034	Wild	-	Australia	Queensland	Cape York
487	W035	Wild	-	Australia	Queensland	Queensland
488	W036	Wild	-	Australia	Northern Territory	Northern Territory
489	W037	Wild	-	Australia	Northern Territory	Northern Territory
490	W038	Wild	-	Australia	Queensland	Queensland
491	W039	Wild	-	Australia	Queensland	Queensland
492	W040	Wild	-	Australia	Northern Territory	Northern Territory
493	W041	Wild	-	Australia	Queensland	Queensland
494	W042	Wild	-	Australia	Queensland	Queensland
495	W043	Wild	-	Australia	Queensland	Queensland
496	W044	Wild	-	Australia	Queensland	Queensland
497	W045	Wild	-	Australia	Queensland	Queensland
498	W046	Wild	-	Australia	Queensland	Queensland
499	W047	Wild	-	Australia	Northern Territory	Northern Territory
500	W048	Wild	-	Australia	Northern Territory	Northern Territory
501	W049	Wild	-	Australia	Northern Territory	Northern Territory
502	W050	Wild	-	Australia	Northern Territory	Northern Territory
503	W051	Wild	-	Australia	Northern Territory	Northern Territory
504	W052	Wild	-	Australia	Queensland	Queensland
505	W053	Wild	-	Australia	Queensland	Queensland
506	W054	Wild	-	Australia	Queensland	Queensland

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
507	W055	Wild	-	Australia	Queensland	Queensland
508	W056	Wild	-	Australia	Queensland	Queensland
509	W057	Wild	-	Australia	Queensland	Northern Territory
510	W058	Wild	-	Australia	Queensland	Northern Territory
511	W059	Wild	-	Australia	Queensland	Queensland
512	W060	Wild	-	Australia	Queensland	Queensland
513	W061	Wild	-	Australia	Northern Territory	Northern Territory
514	W062	Wild	-	Australia	Northern Territory	Northern Territory
515	W063	Wild	-	Australia	Northern Territory	Northern Territory
516	W064	Wild	-	Australia	Northern Territory	Northern Territory
517	W065	Wild	-	Australia	Northern Territory	Northern Territory
518	W066	Wild	-	Australia	Northern Territory	Northern Territory
519	W067	Wild	-	Australia	Queensland	Queensland
520	W068	Wild	-	Australia	Queensland	Queensland
521	W069	Wild	-	Australia	Queensland	Queensland
522	W070	Wild	-	Australia	Queensland	Queensland
523	W071	Wild	-	Australia	Queensland	Queensland
524	W072	Wild	-	Australia	Queensland	Queensland
525	W073	Wild	-	Australia	Queensland	Queensland
526	W074	Wild	-	Australia	Queensland	Queensland
527	W075	Wild	-	Australia	Queensland	Queensland
528	W076	Wild	-	Australia	Queensland	Queensland
529	W077	Wild	-	Australia	Queensland	Queensland
530	W078	Wild	-	Australia	Northern Territory	Northern Territory

Appendix Table 1 (Continued)

No.	Label	Status	Accessions no.	Country	Province	Collection site
531	W079	Wild	-	Australia	Northern Territory	Northern Territory
532	W080	Wild	-	Australia	Northern Territory	Northern Territory
533	W081	Wild	-	Australia	Northern Territory	Northern Territory
534	W082	Wild	-	Australia	Northern Territory	Northern Territory
535	W083	Wild	-	Australia	Northern Territory	Northern Territory
536	W084	Wild	-	Australia	Western Australia	Western Australia
537	W085	Wild	-	Australia	Northern Territory	Northern Territory
538	W086	Wild	-	Australia	Western Australia	Western Australia
539	W087	Wild	-	Australia	Western Australia	Western Australia
540	W088	Wild	-	Australia	Western Australia	Western Australia
541	W089	Wild	-	Australia	Western Australia	Western Australia
542	W090	Wild	-	Australia	Western Australia	Western Australia
543	W091	Wild	-	Australia	Western Australia	Western Australia
544	W092	Wild	-	Australia	Western Australia	. Western Australia
545	W093	Wild	-	Australia	Western Australia	Western Australia
546	W094	Wild	-	Australia	Western Australia	Western Australia
547	W095	Wild	-	Australia	Western Australia	Western Australia
548	W096	Wild	-	Australia	Western Australia	Western Australia
549	W097	Wild	-	Australia	Western Australia	Western Australia
550	W098	Wild	-	Australia	Queensland	Queensland
551	W099	Wild	-	Australia	Queensland	Queensland
552	W100	Wild	-	Australia	Queensland	Queensland
553	W101	Wild	-	Australia	Queensland	Queensland
554	W102	Wild	-	Australia	Queensland	Queensland

Appendix Table 1 (Continued)

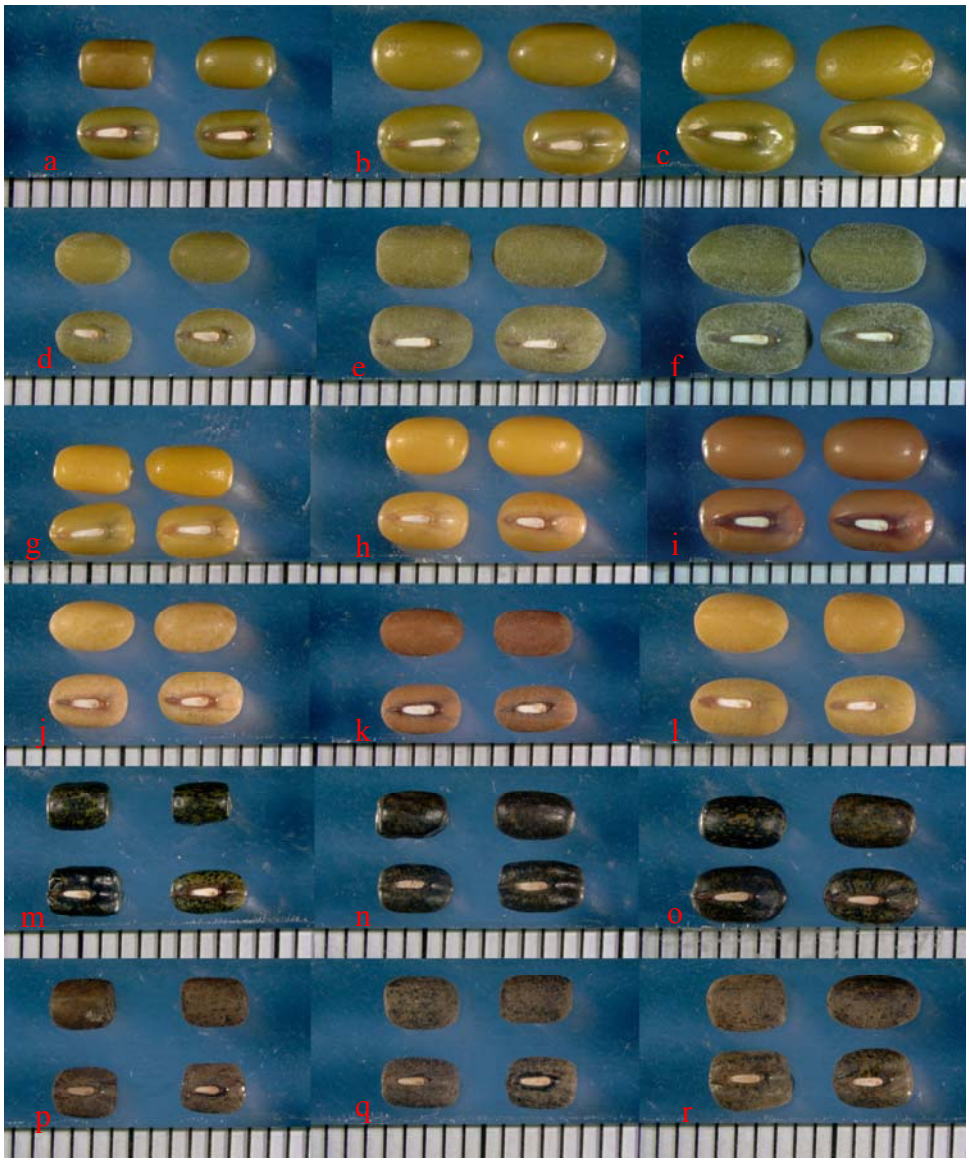
No.	Label	Status	Accessions no.	Country	Province	Collection site
555	W103	Wild	-	Australia	Northern Territory	Northern Territory
556	W104	Wild	-	Australia	Northern Territory	Northern Territory
557	W105	Wild	-	Australia	Queensland	Queensland
558	W106	Wild	-	Australia	Queensland	Queensland
559	W107	Wild	-	Australia	Queensland	Queensland
560	W108	Wild	-	Australia	Queensland	Queensland
561	W109	Wild	-	Australia	Queensland	Queensland
562	W110	Wild	-	Australia	Queensland	Queensland
563	W111	Wild	-	Australia	Queensland	Queensland
564	W112	Wild	-	Australia	Queensland	Queensland
565	W113	Wild	-	Australia	Queensland	Queensland
566	W114	Wild	-	Australia	Queensland	Queensland
567	W115	Wild	-	Australia	Queensland	Queensland
568	W116	Wild	-	Australia	Western Australia	Western Australia
569	W117	Wild	-	Australia	Queensland	Queensland
570	W118	Wild	-	Australia	Queensland	Queensland
571	W119	Wild	-	Australia	Western Australia	Western Australia
572	W120	Wild	-	Australia	Northern Territory	Northern Territory
573	W121	Wild	-	Australia	Northern Territory	Northern Territory
574	W122	Wild	-	Australia	Queensland	Queensland
575	W123	Wild	JP107876	Australia	-	-
576	W124	Wild	JP110843	Australia	Queensland	Cape York
577	W125	Wild	JP202280	Australia	-	-

Appendix Table 1 (Continued)

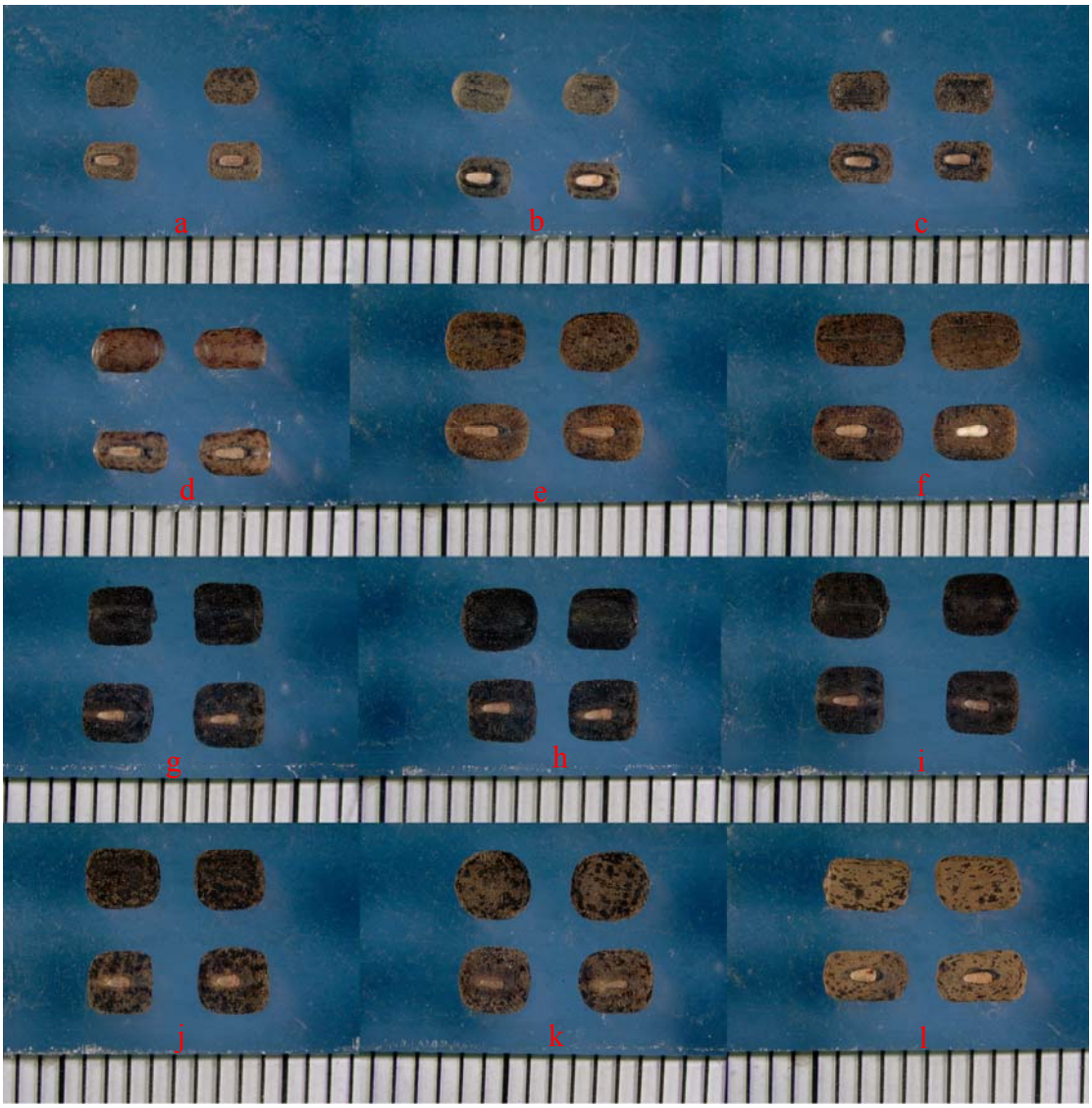
No.	Label	Status	Accessions no.	Country	Province	Collection site
578	W126	Wild	JP202281	Australia	-	-
579	W127	Wild	JP227254	Australia	Queensland	Queensland
580	W171	Wild	-	Papua New Guinea	Nadzab	Nadzab
581	W172	Wild	-	Papua New Guinea	Bogola	Bogola
582	W173	Wild	JP202293	Papua New Guinea	-	-
583	W174	Wild	JP202294	Papua New Guinea	-	-
584	W175	Wild	JP218942	Papua New Guinea	Port Moresby	Port Moresby
585	W176	Wild	JP222454	Papua New Guinea	Port Moresby	Waigani
586	W177	Wild	JP222455	Papua New Guinea	Port Moresby	Napa Napa
587	W178	Wild	JP222456	Papua New Guinea	Port Moresby	Napa Napa
588	W179	Wild	JP222457	Papua New Guinea	Port Moresby	Napa Napa
589	W180	Wild	JP222458	Papua New Guinea	Central	Saroa
590	W181	Wild	JP222459	Papua New Guinea	Central	Manu Goro
591	W182	Wild	JP222460	Papua New Guinea	Central	Kubuna Mission
592	W183	Wild	JP222461	Papua New Guinea	Milne Bay	Awayama
593	W184	Wild	JP222462	Papua New Guinea	Milne Bay	Garuahi
594	W185	Wild	JP222464	Papua New Guinea	Milne Bay	Raba Raba
595	W186	Wild	JP222468	Papua New Guinea	Milne Bay	Dogura
596	W187	Wild	JP222485	Papua New Guinea	Morobe	Marajabong
597	W188	Wild	JP222486	Papua New Guinea	Madang	Dunpu
598	W189	Wild	JP226872	Papua New Guinea	East Sepik	Aibom
599	W190	Wild	JP226873	Papua New Guinea	East Sepik	Timbun
600	W191	Wild	JP226874	Papua New Guinea	East Sepik	Ari John

Appendix Table 1 (Continued)

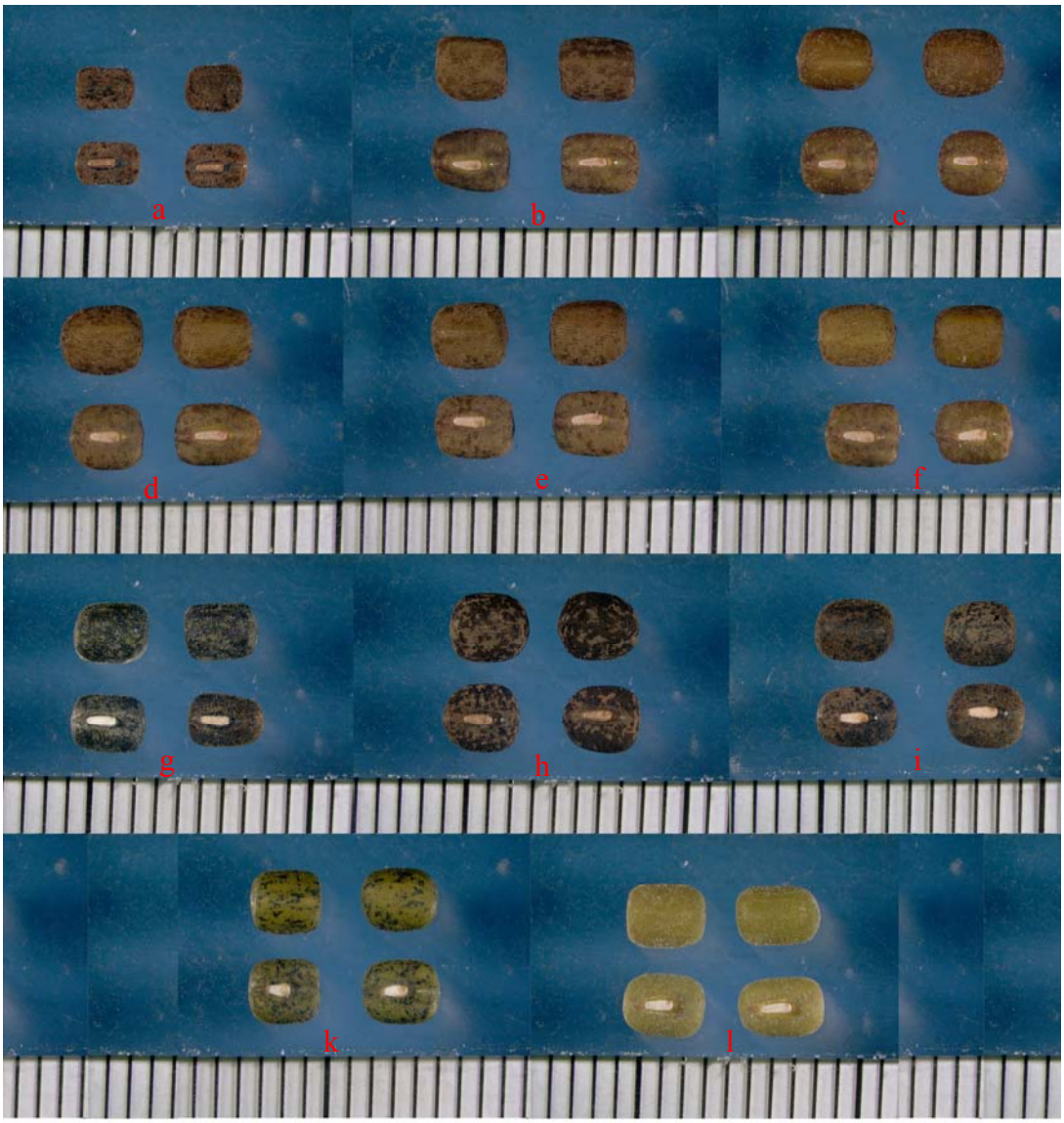
No.	Label	Status	Accessions no.	Country	Province	Collection site
601	W192	Wild	JP226875	Papua New Guinea	East Sepik	Savanaut
602	W193	Wild	JP226876	Papua New Guinea	East Sepik	Pagwi
603	W194	Wild	JP226878	Papua New Guinea	Madang	Madang
604	W195	Wild	JP226879	Papua New Guinea	Madang	Madang
605	W196	Wild	JP226880	Papua New Guinea	Madang	Yagumbu
606	W197	Wild	JP226881	Papua New Guinea	Madang	Madang
607	W198	Wild	JP226883	Papua New Guinea	Madang	Tapopo
608	W199	Wild	JP227258	Papua New Guinea	Port Moresby	Port Moresby
609	W128	Wild	JP218943	Cameroon	Lara	Lara
610	W129	Wild	JP218944	Cameroon	Ngutchmi	Ngutchmi
611	W130	Wild	JP218945	Cameroon	Ngutchmi	Ngutchmi
612	W131	Wild	JP227260	Cameroon	Dembo	Dembo
613	W132	Wild	JP227261	Cameroon	Nigba	Nigba
614	W133	Wild	JP227262	Cameroon	Nigba	Nigba
615	W134	Wild	JP227263	Cameroon	Pate maga	Pate maga



Appendix Figure 1 Pictures of seeds some mungbean landraces used in this experiment. a-c pictures show shiny-green seeds arraying from small to large seeds. g-i pictures shiny-yellow seeds arraying from small to large seeds size. j-l pictures show dull-yellow seeds arraying from small to large seeds size. m-o pictures show shiny-black mottling seeds arraying from small to large seeds size. p-r pictures show dull-brown mottling seeds arraying from small to large seeds size.

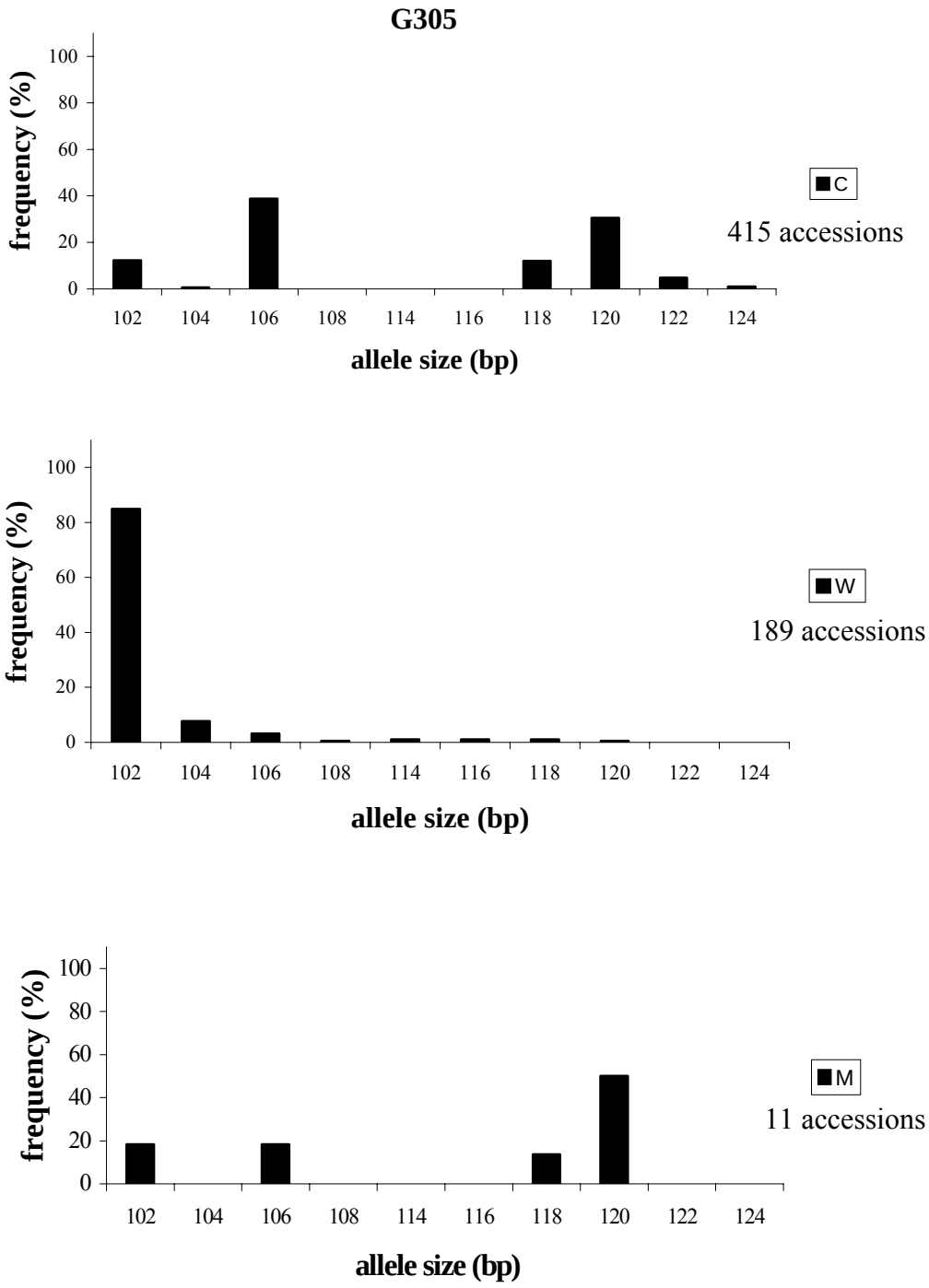


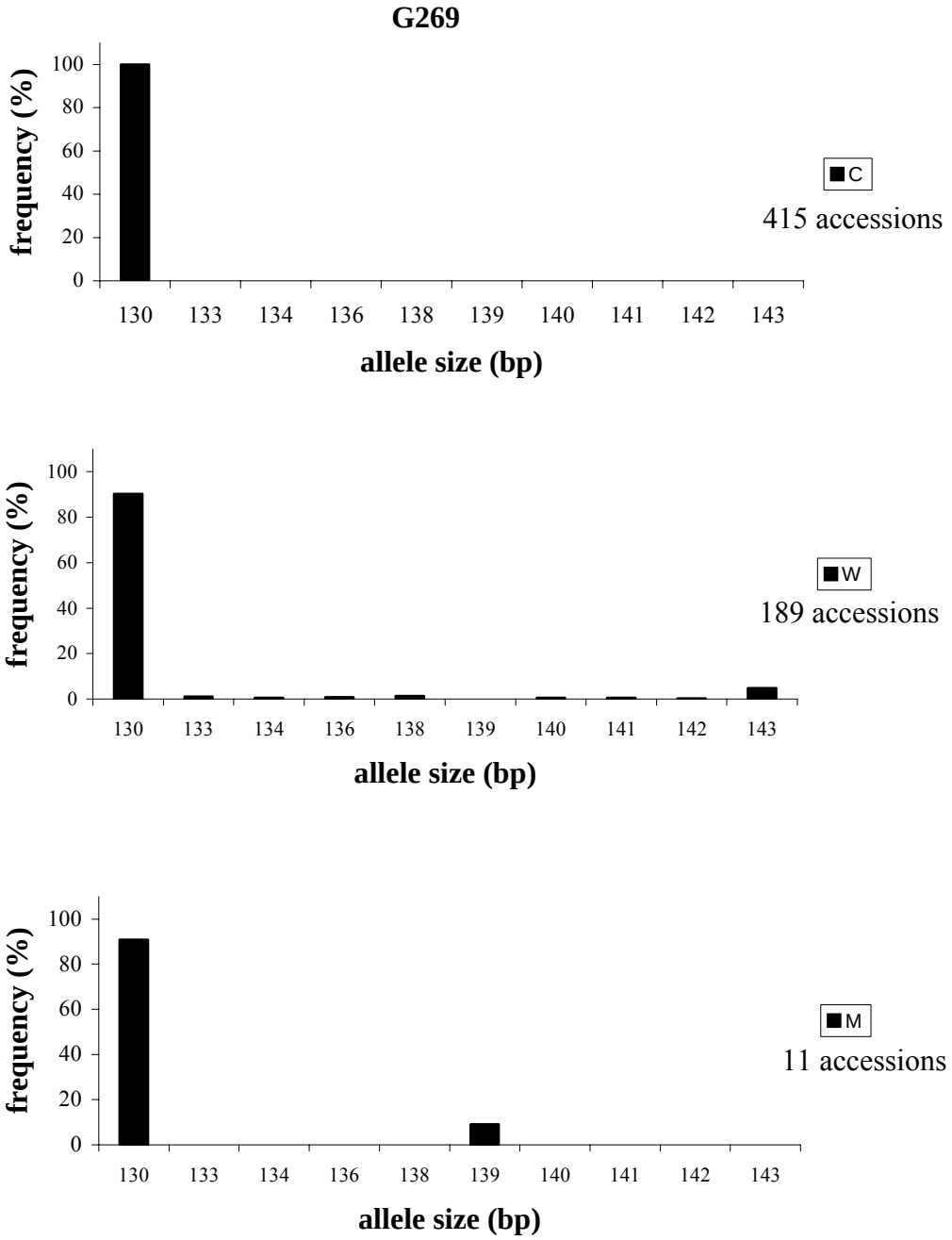
Appendix Figure 2 Pictures of seeds some wild mungbean accessions used in this experiment. a-l pictures show dull-black mottling seeds arraying from small to large seeds size.

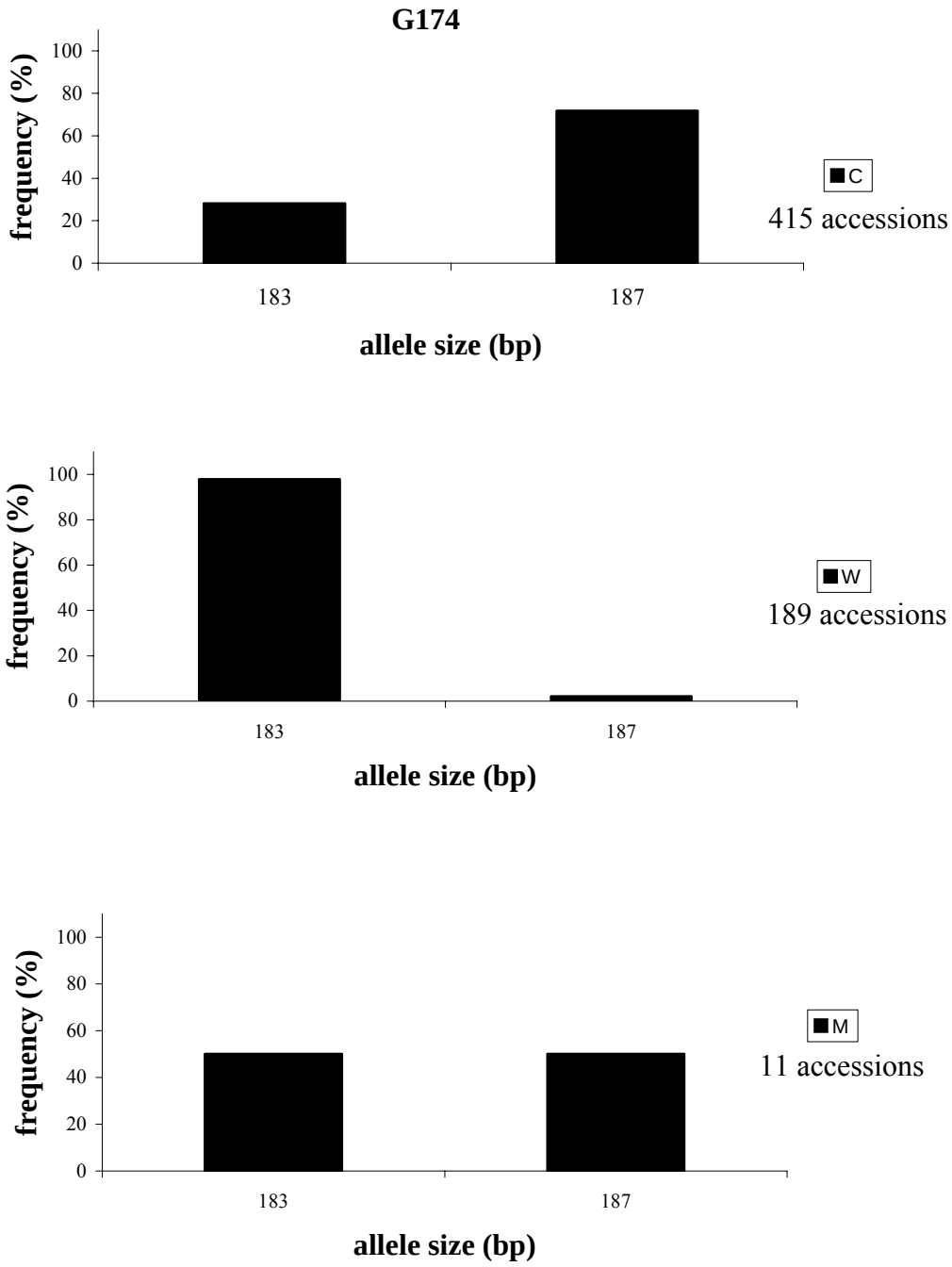


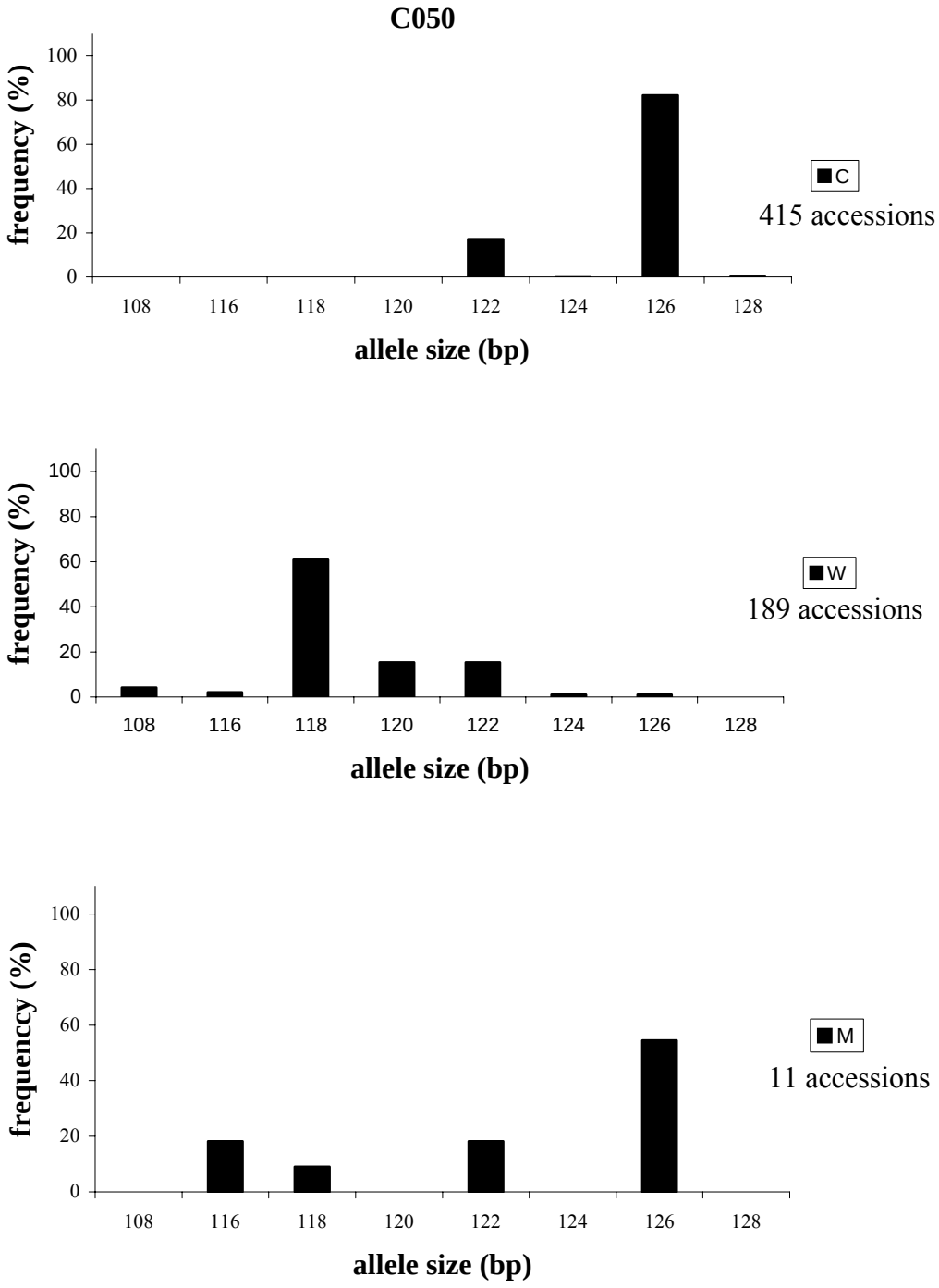
Appendix Figure 3 Pictures of seeds intermediate mungbean accessions used in this experiment. a-l pictures show intermediate mungbeans. viz, M153, M136, M135, M202, M200, M201, M151, M146, M207, M149 and M152.

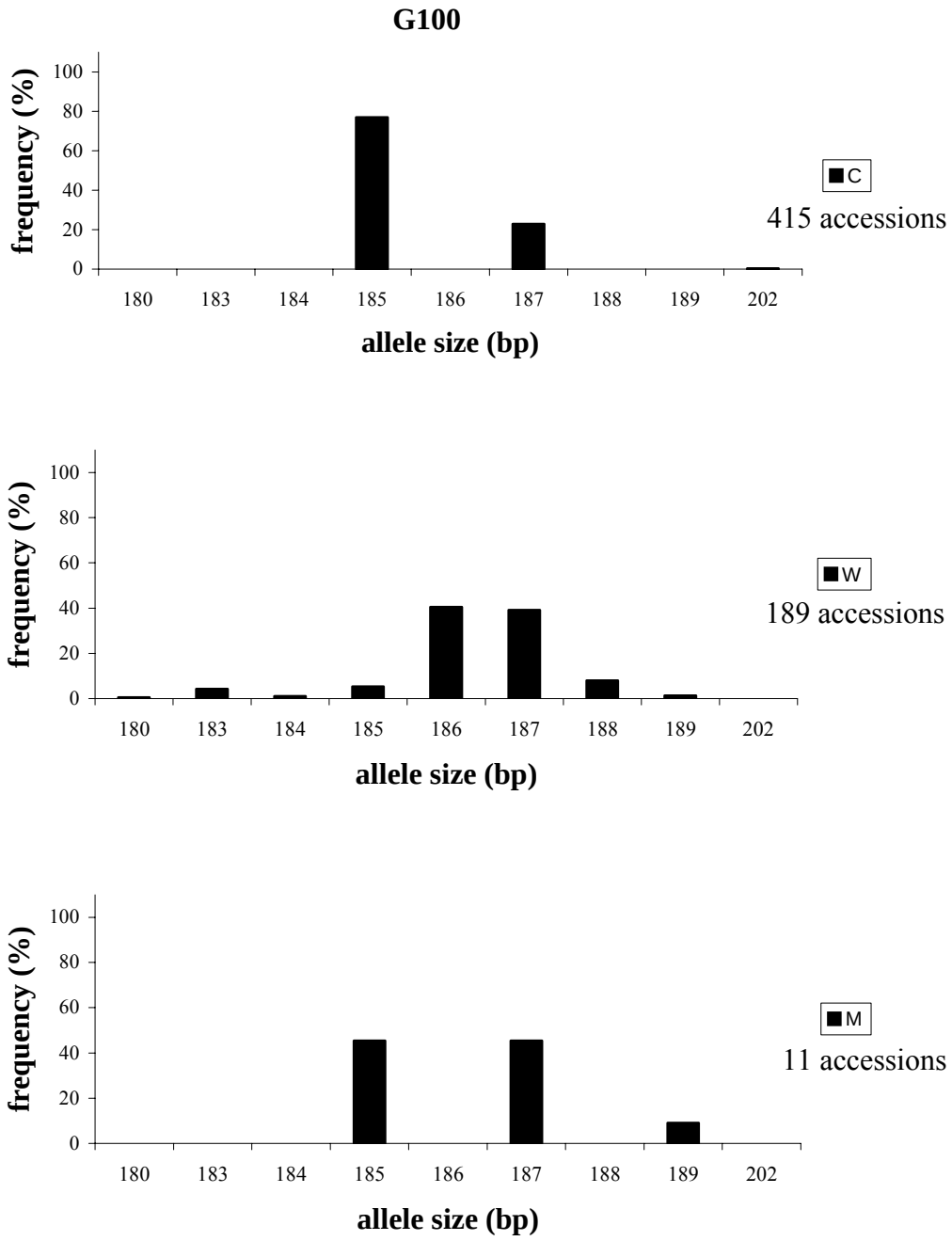
Appendix Figure 4 Histograms of allele frequency of loci per primer in cultivated wild, and intermediate mungbean samples.



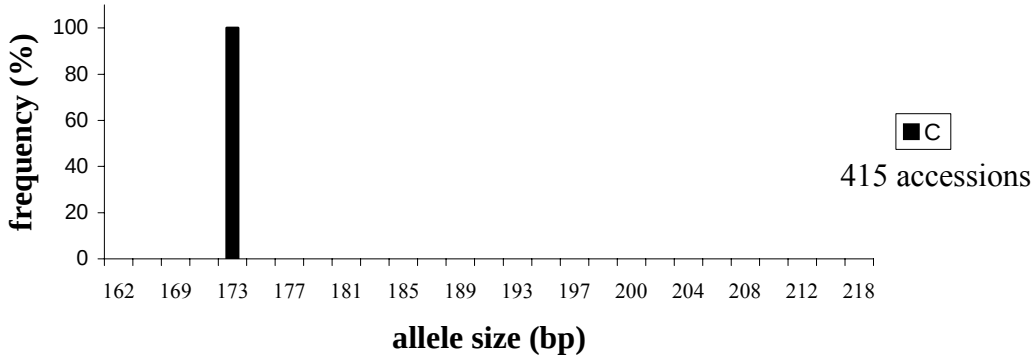

G269
frequency (%)
100
80
60
40
20
0
130
133
134
136
138
139
140
141
142
143
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
130
133
134
136
138
139
140
141
142
143
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
130
133
134
136
138
139
140
141
142
143
allele size (bp)
M
11 accessions

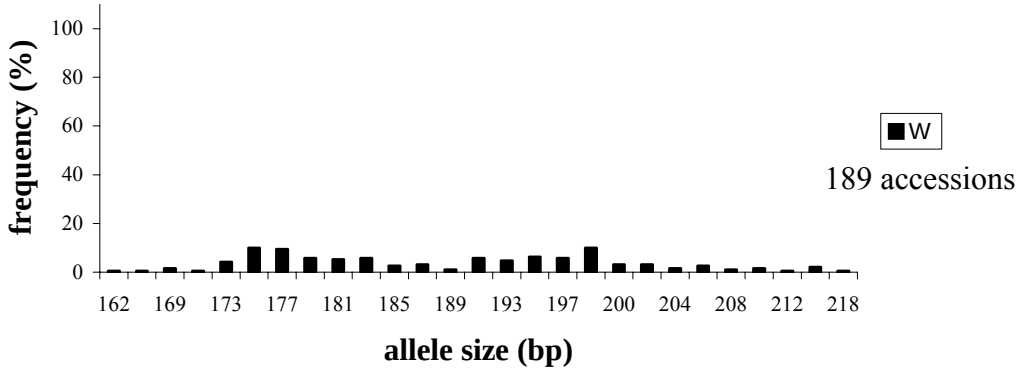

G174
frequency (%)
100
80
60
40
20
0
183
187
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
183
187
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
183
187
allele size (bp)
M
11 accessions

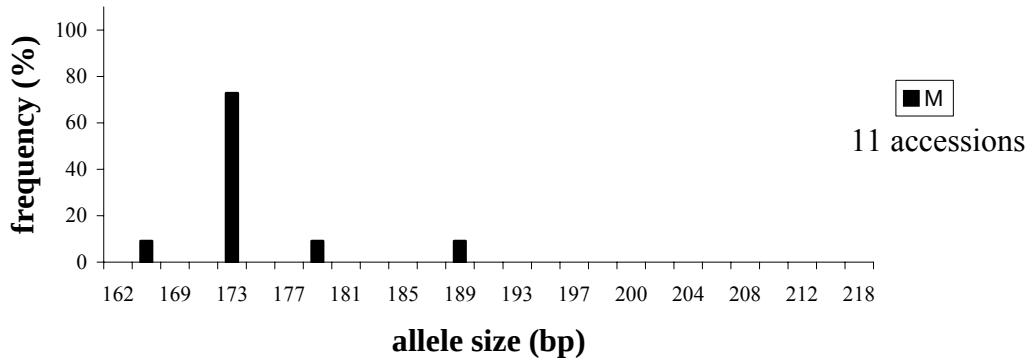

C050
frequency (%)
100
80
60
40
20
0
108
116
118
120
122
124
126
128
allele size (bp)
C
415 accessions
frequency (%)
W
189 accessions
allele size (bp)
frequenccy (%)
M
11 accessions
allele size (bp)

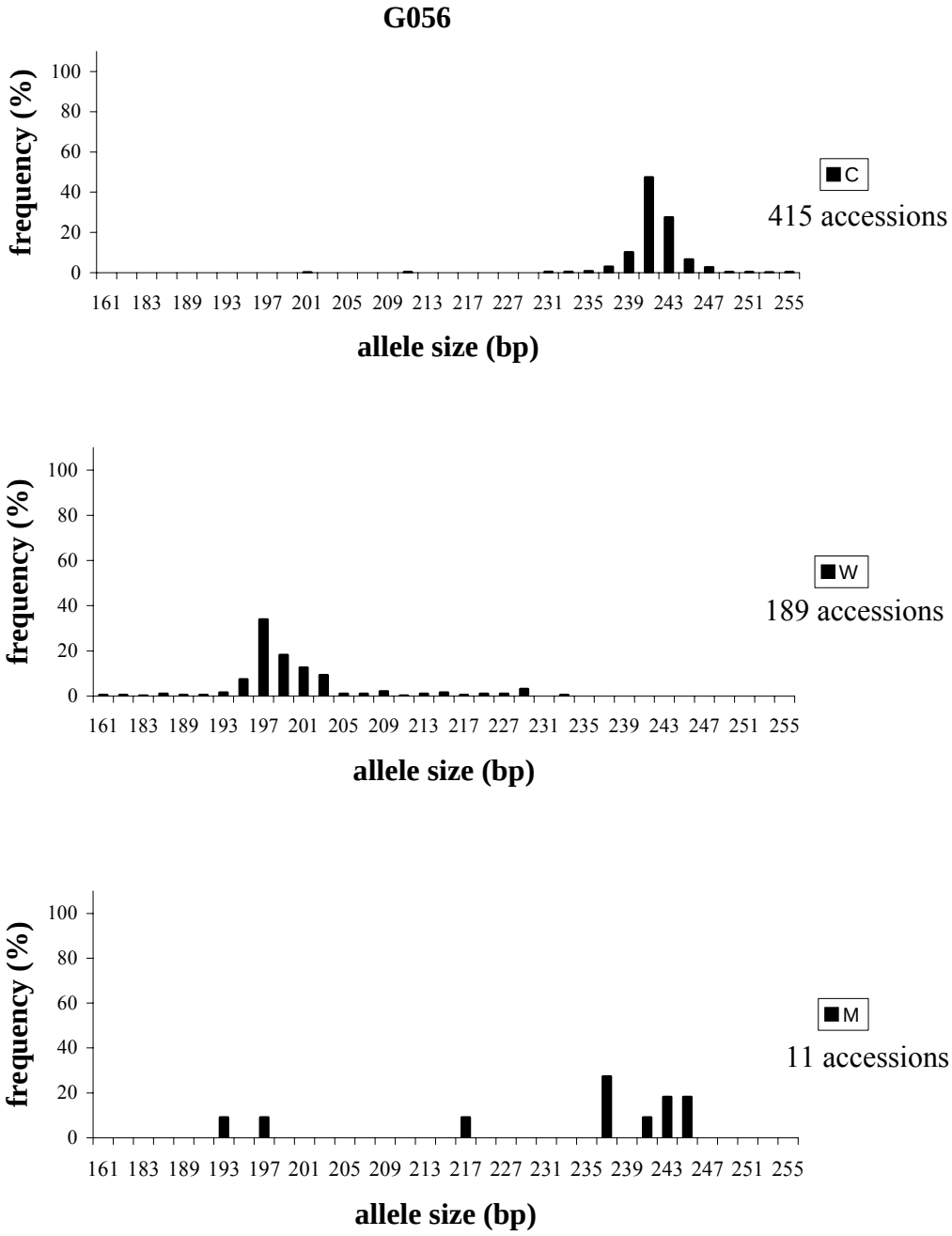

G100
frequency (%)
100
80
60
40
20
0
180
183
184
185
186
187
188
189
202
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
180
183
184
185
186
187
188
189
202
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
180
183
184
185
186
187
188
189
202
allele size (bp)
M
11 accessions

G015

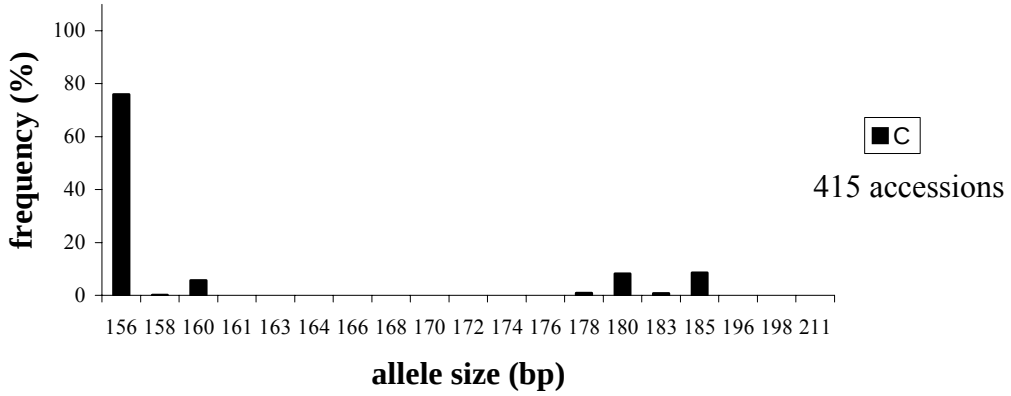

frequency (%)
100
80
60
40
20
0
162 169 173 177 181 185 189 193 197 200 204 208 212 218
allele size (bp)
C
415 accessions

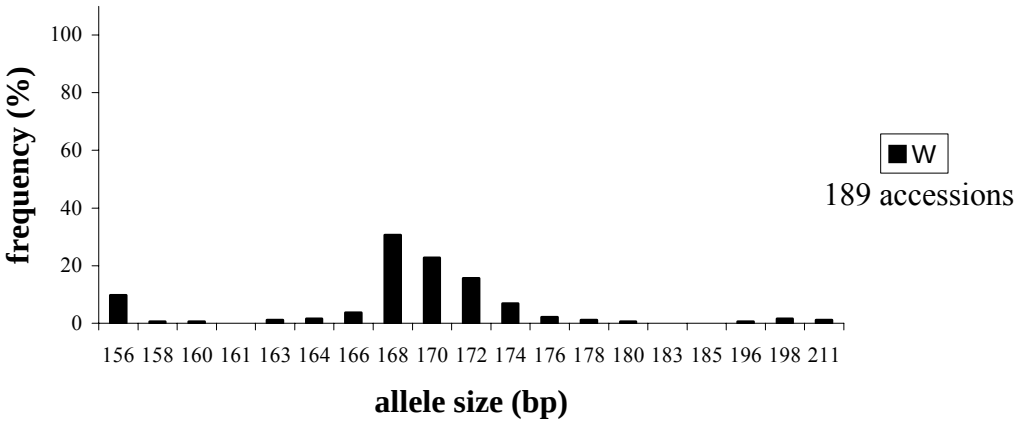

frequency (%)
100
80
60
40
20
0
162 169 173 177 181 185 189 193 197 200 204 208 212 218
allele size (bp)
W
189 accessions

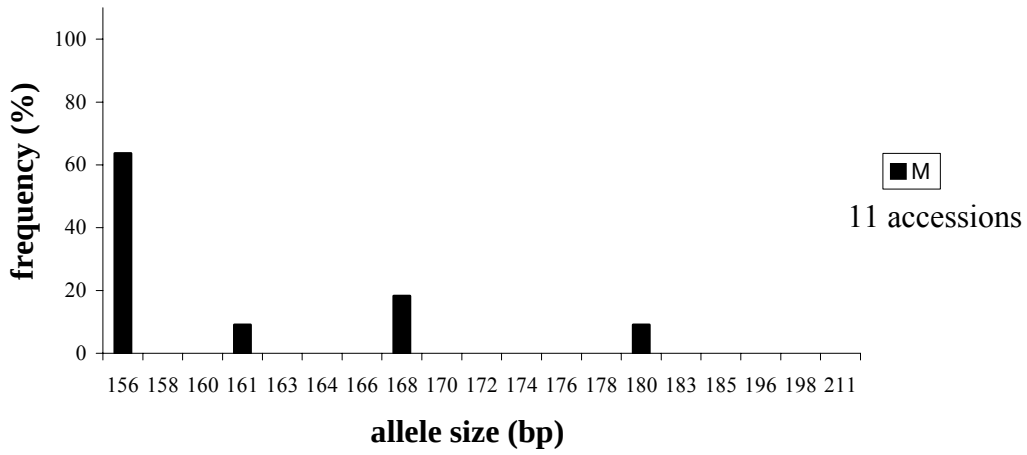

frequency (%)
100
80
60
40
20
0
162 169 173 177 181 185 189 193 197 200 204 208 212 218
allele size (bp)
M
11 accessions

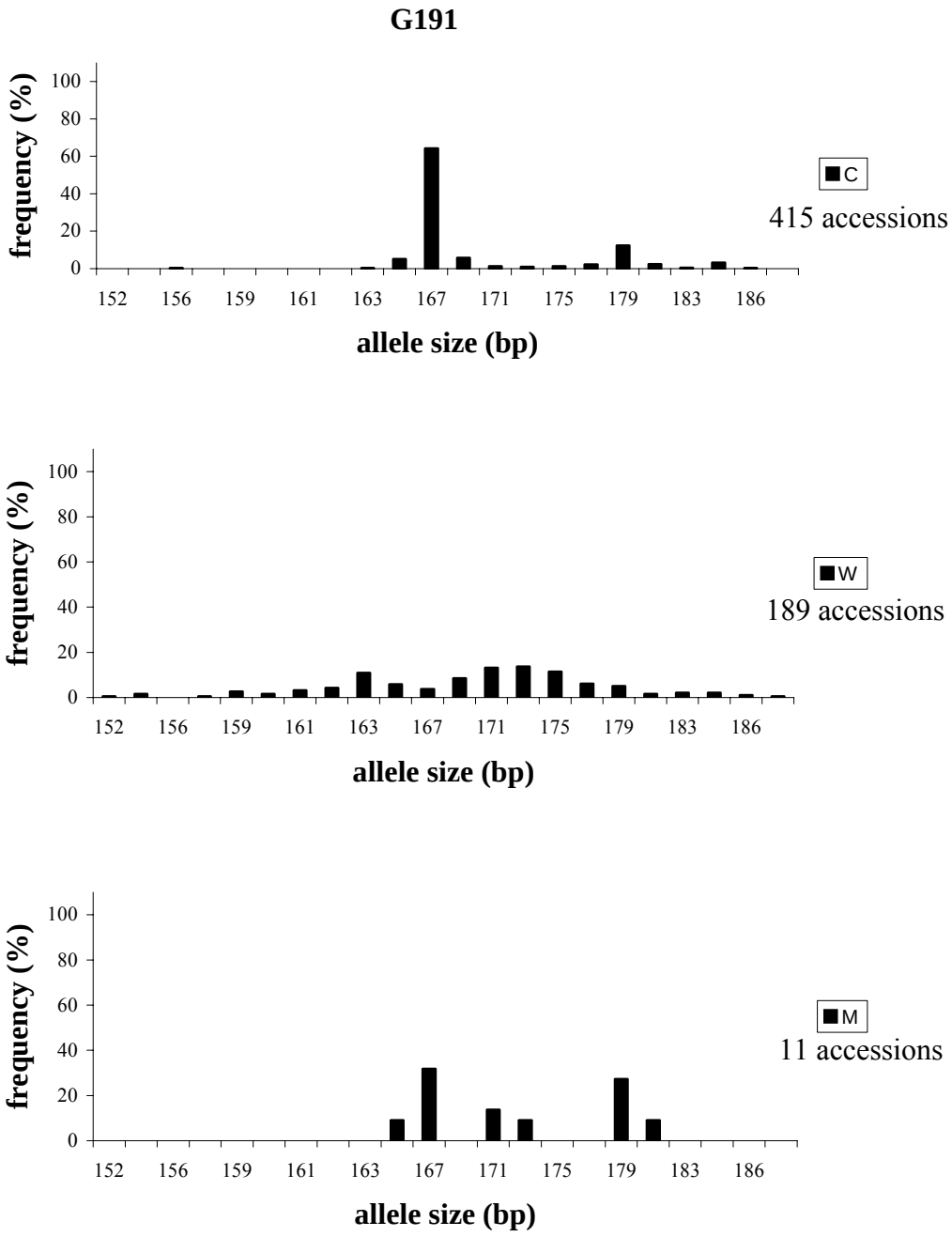

G056
frequency (%)
100
80
60
40
20
0
161 183 189 193 197 201 205 209 213 217 227 231 235 239 243 247 251 255
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
161 183 189 193 197 201 205 209 213 217 227 231 235 239 243 247 251 255
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
161 183 189 193 197 201 205 209 213 217 227 231 235 239 243 247 251 255
allele size (bp)
M
11 accessions

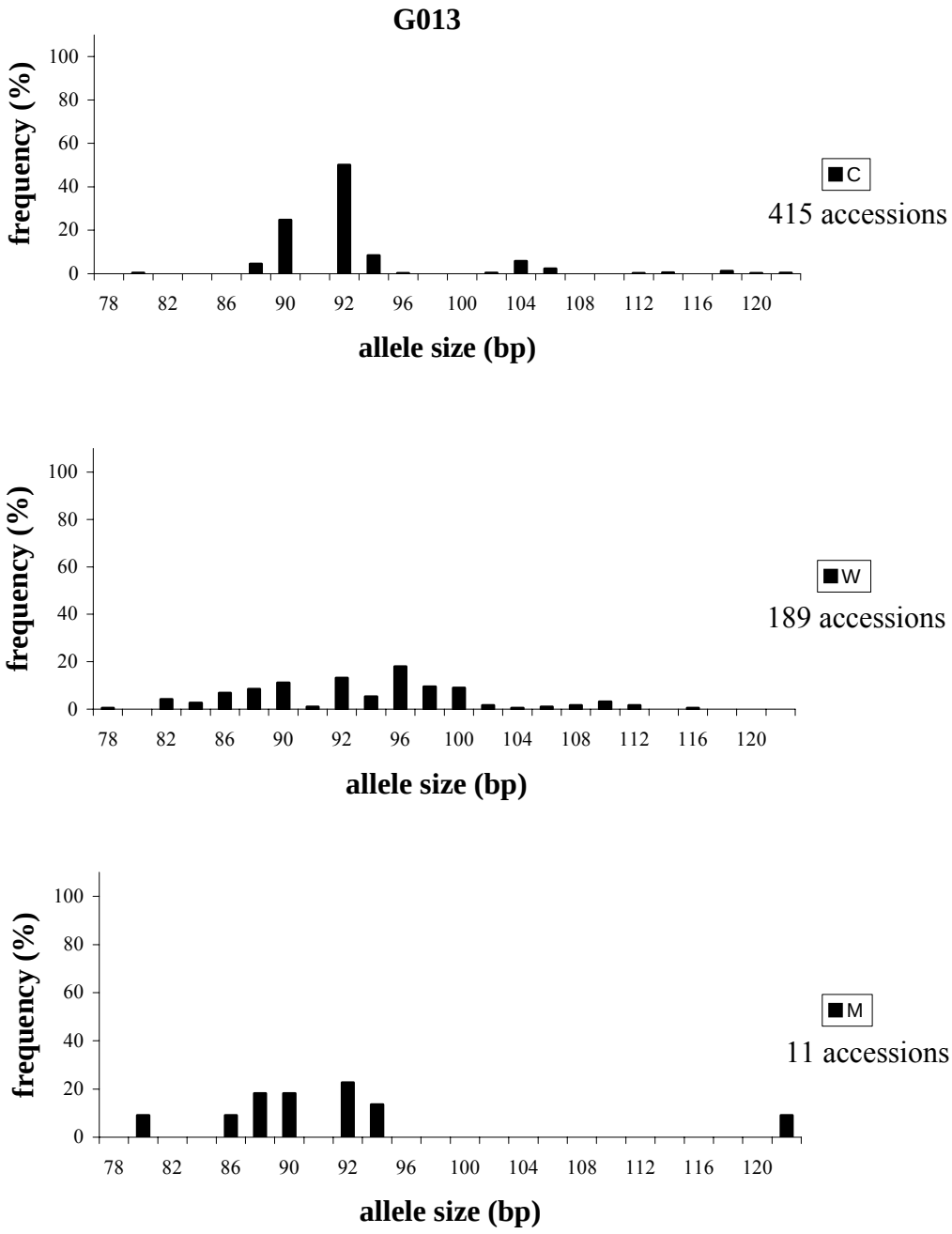
G149

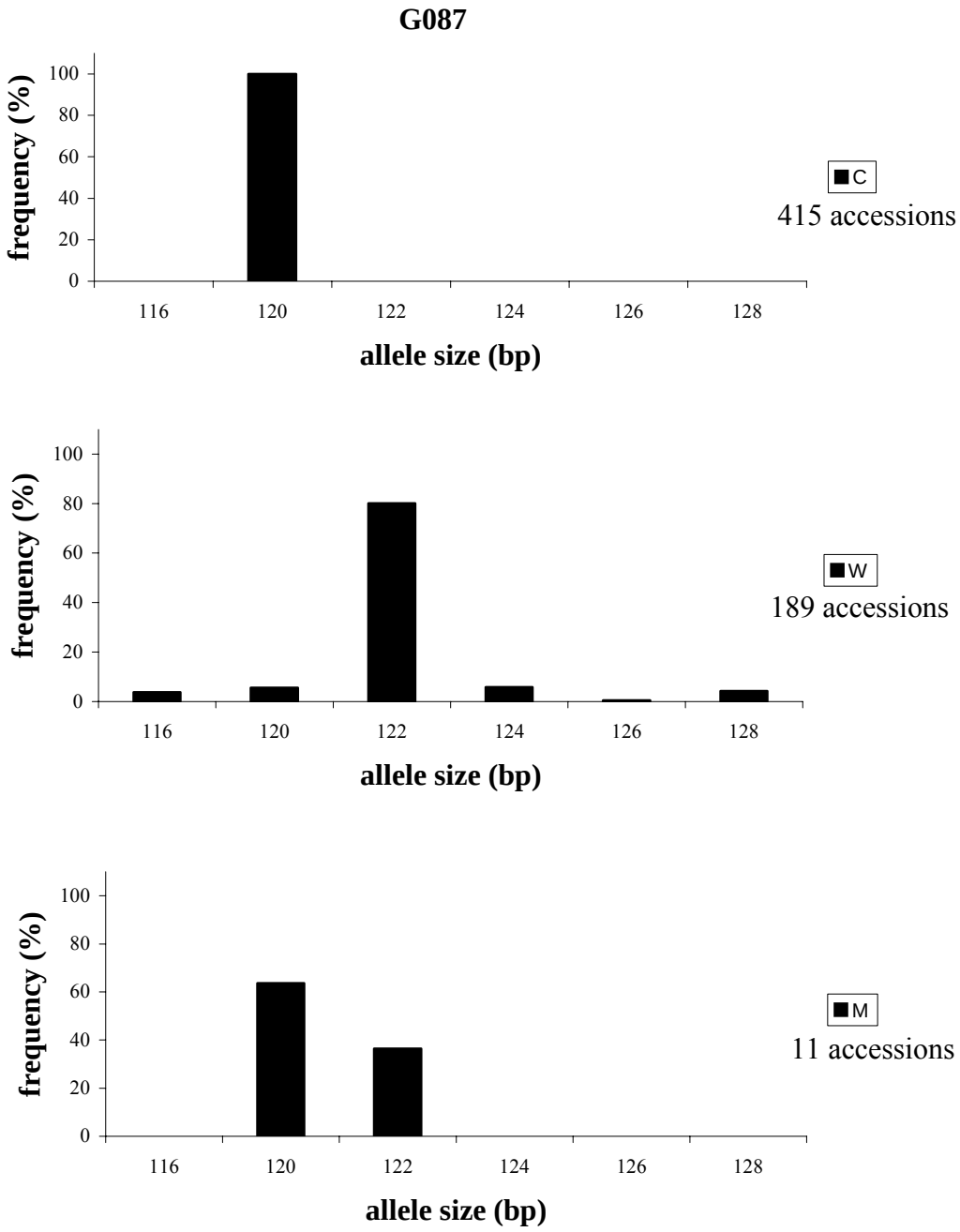

frequency (%)
100
80
60
40
20
0
156 158 160 161 163 164 166 168 170 172 174 176 178 180 183 185 196 198 211
allele size (bp)
C
415 accessions

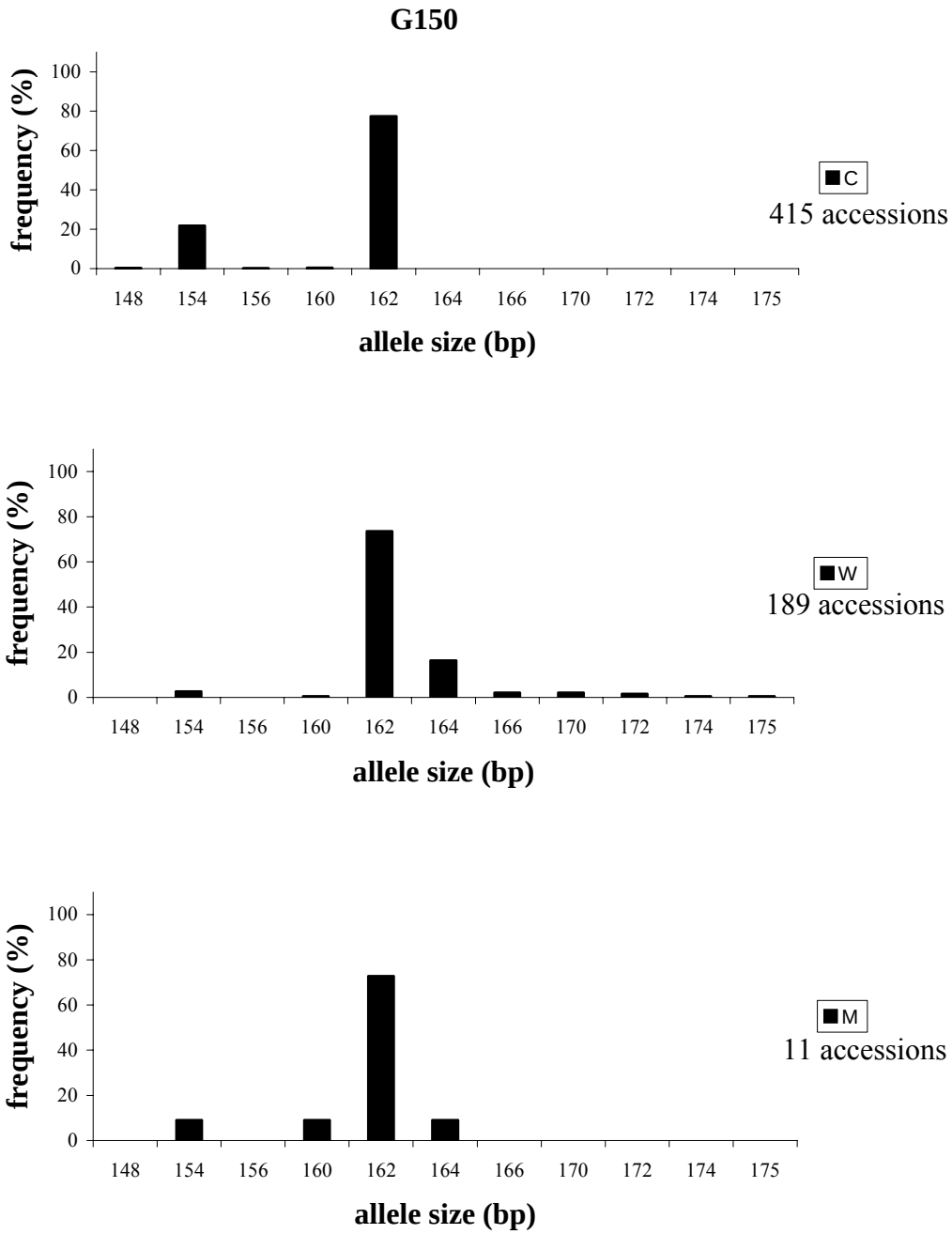

frequency (%)
100
80
60
40
20
0
156 158 160 161 163 164 166 168 170 172 174 176 178 180 183 185 196 198 211
allele size (bp)
W
189 accessions

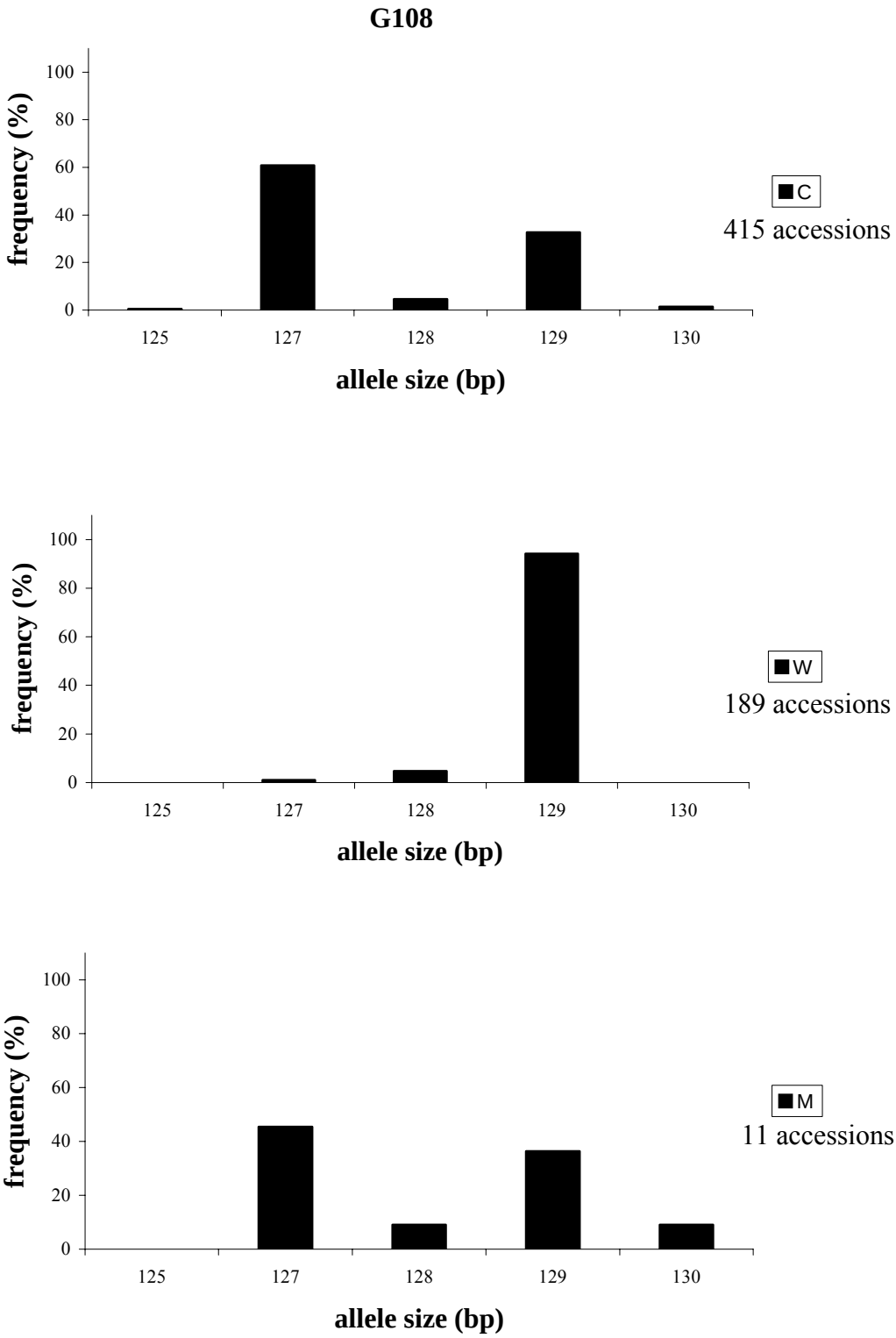

frequency (%)
100
80
60
40
20
0
156 158 160 161 163 164 166 168 170 172 174 176 178 180 183 185 196 198 211
allele size (bp)
M
11 accessions

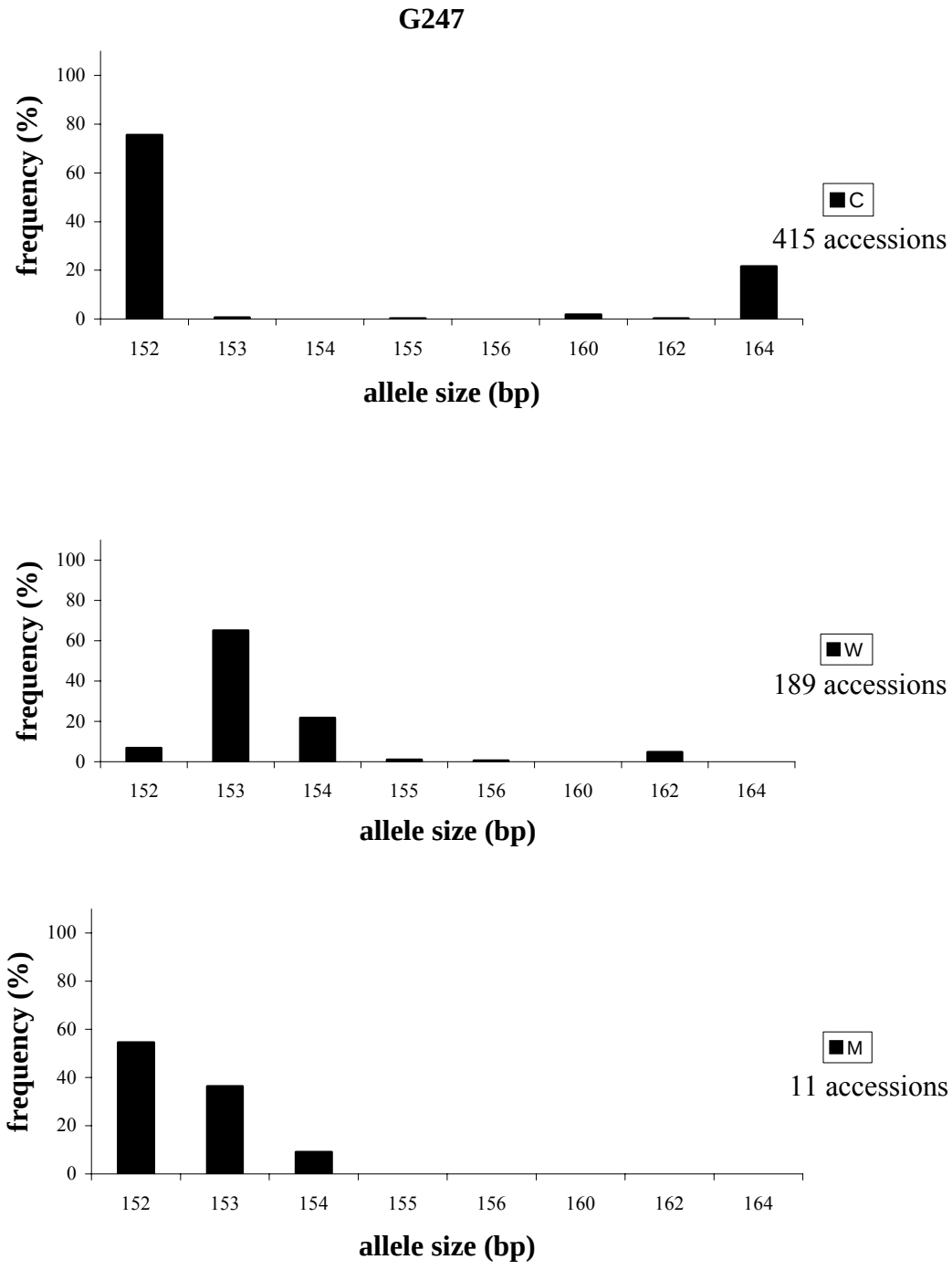

G191
frequency (%)
100
80
60
40
20
0
152
156
159
161
163
167
171
175
179
183
186
allele size (bp)
C
415 accessions
frequency (%)
allele size (bp)
W
189 accessions
frequency (%)
allele size (bp)
M
11 accessions

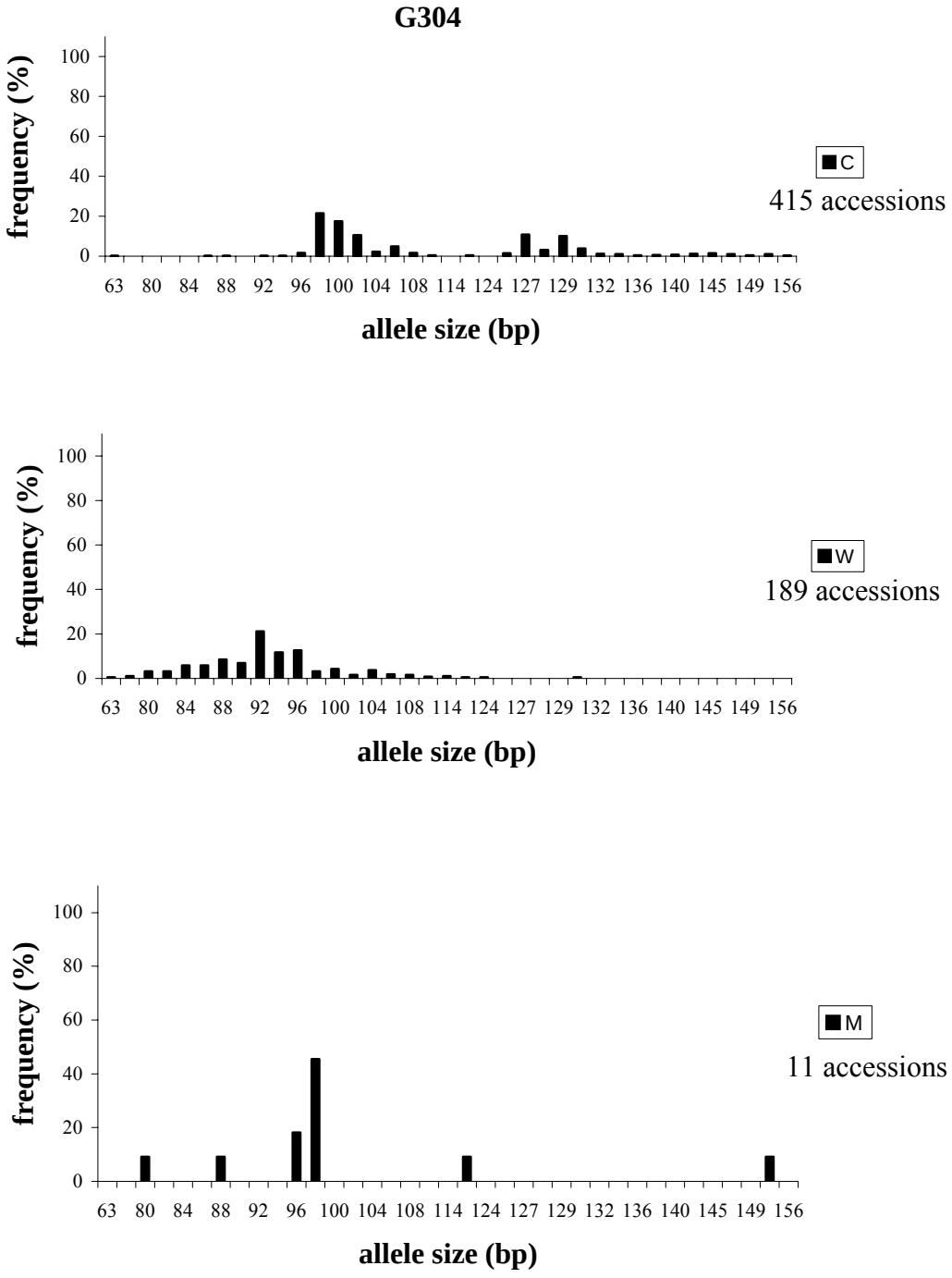

G013
frequency (%)
100
80
60
40
20
0
78
82
86
90
92
96
100
104
108
112
116
120
allele size (bp)
C
415 accessions
frequency (%)
allele size (bp)
W
189 accessions
frequency (%)
allele size (bp)
M
11 accessions

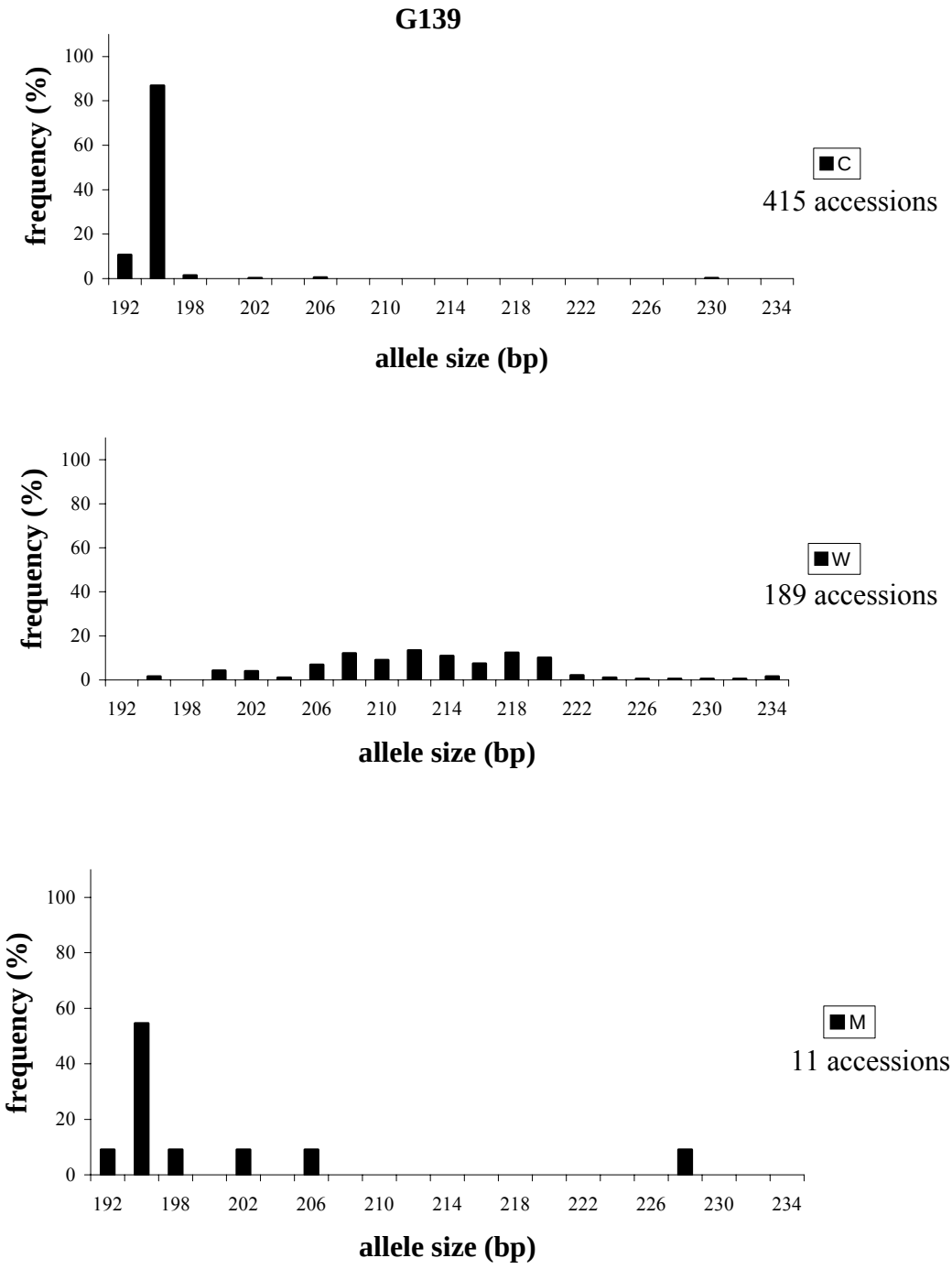

G087
frequency (%)
100
80
60
40
20
0
116
120
122
124
126
128
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
116
120
122
124
126
128
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
116
120
122
124
126
128
allele size (bp)
M
11 accessions

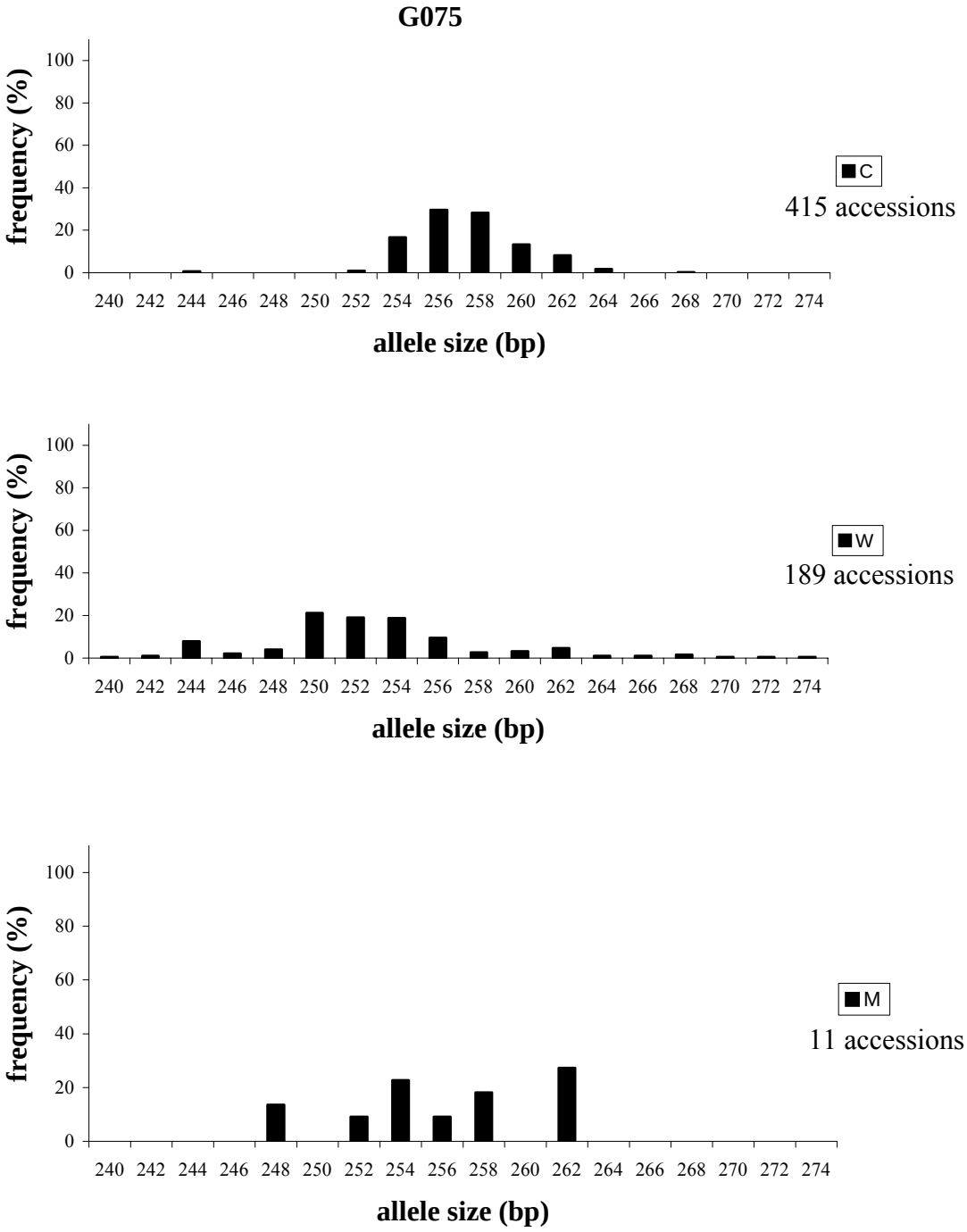

G150
frequency (%)
100
80
60
40
20
0
148
154
156
160
162
164
166
170
172
174
175
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
148
154
156
160
162
164
166
170
172
174
175
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
148
154
156
160
162
164
166
170
172
174
175
allele size (bp)
M
11 accessions

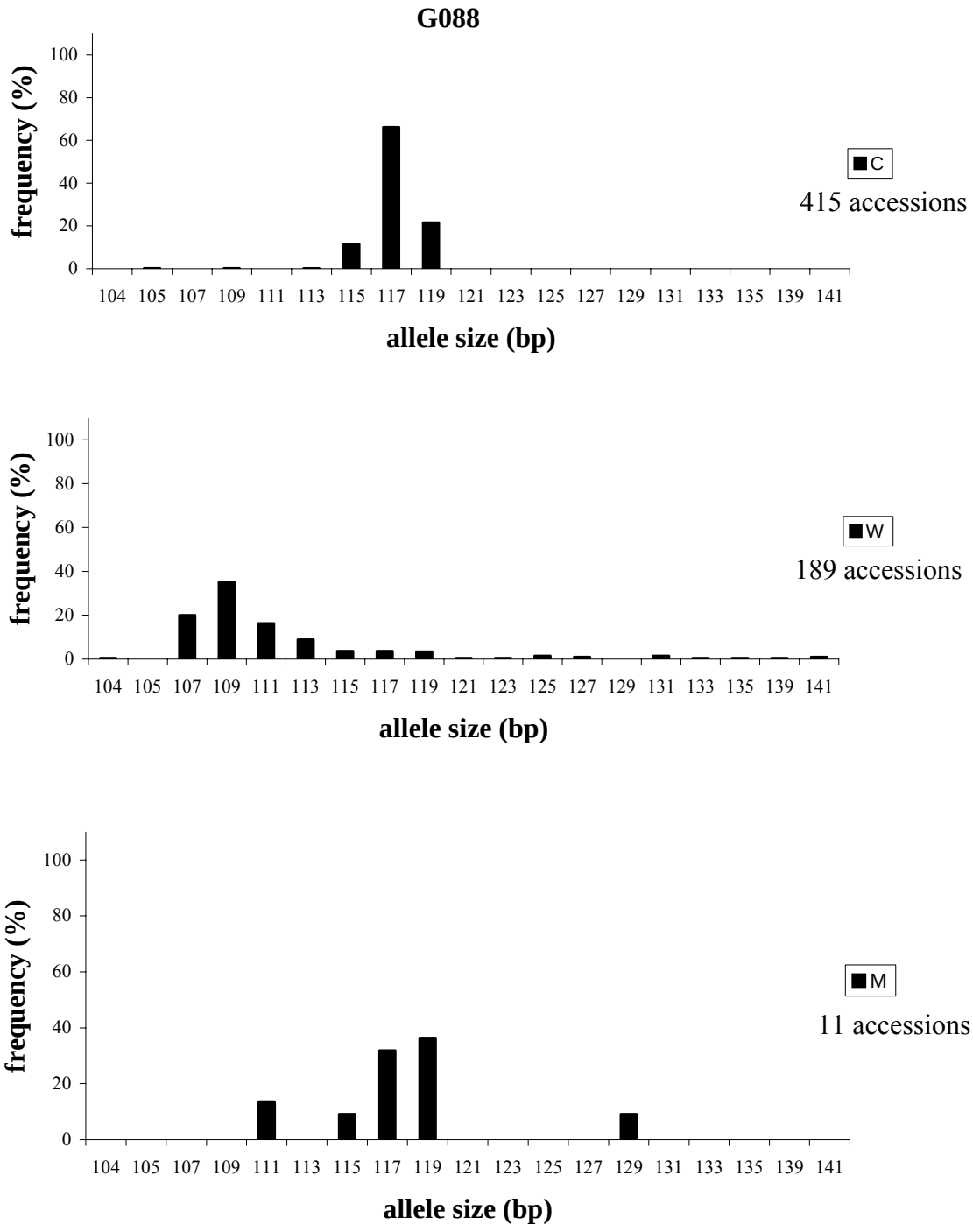

G108
frequency (%)
100
80
60
40
20
0
125
127
128
129
130
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
125
127
128
129
130
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
125
127
128
129
130
allele size (bp)
M
11 accessions


G247
frequency (%)
100
80
60
40
20
0
152
153
154
155
156
160
162
164
allele size (bp)
C
415 accessions
frequency (%)
allele size (bp)
W
189 accessions
frequency (%)
allele size (bp)
M
11 accessions

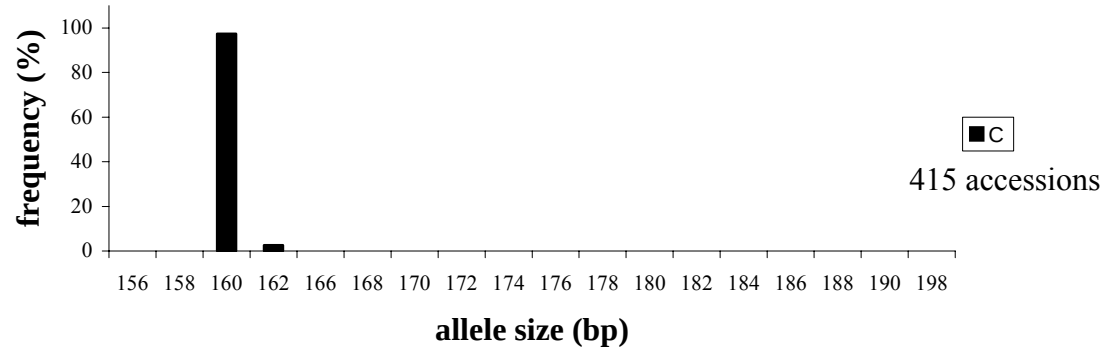

G304
frequency (%)
100
80
60
40
20
0
63 80 84 88 92 96 100 104 108 114 124 127 129 132 136 140 145 149 156
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
63 80 84 88 92 96 100 104 108 114 124 127 129 132 136 140 145 149 156
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
63 80 84 88 92 96 100 104 108 114 124 127 129 132 136 140 145 149 156
allele size (bp)
M
11 accessions

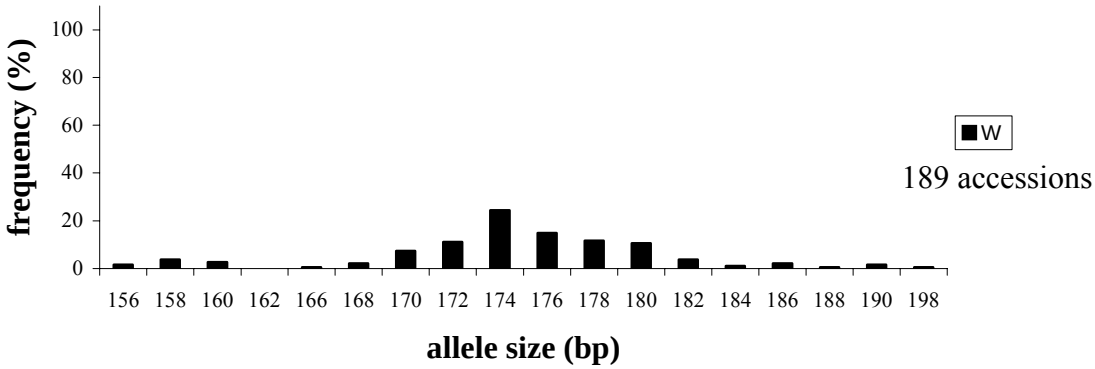

G139
frequency (%)
100
80
60
40
20
0
192
198
202
206
210
214
218
222
226
230
234
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
192
198
202
206
210
214
218
222
226
230
234
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
192
198
202
206
210
214
218
222
226
230
234
allele size (bp)
M
11 accessions

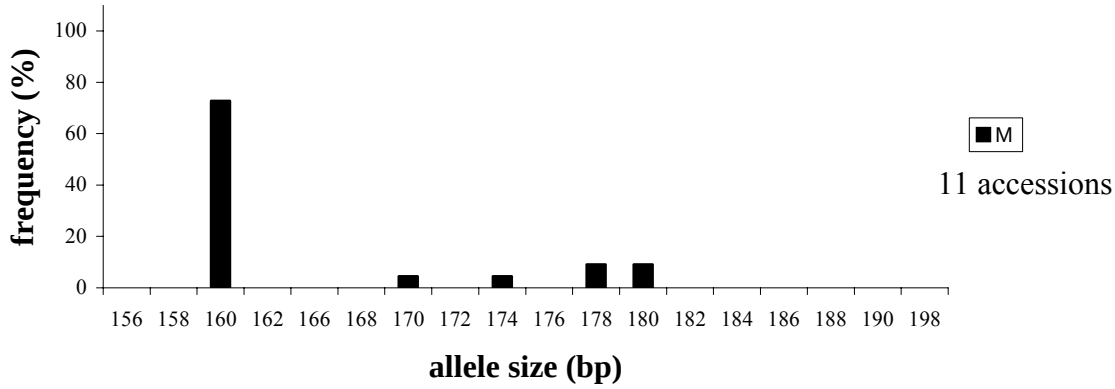

G075
frequency (%)
100
80
60
40
20
0
240 242 244 246 248 250 252 254 256 258 260 262 264 266 268 270 272 274
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
240 242 244 246 248 250 252 254 256 258 260 262 264 266 268 270 272 274
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
240 242 244 246 248 250 252 254 256 258 260 262 264 266 268 270 272 274
allele size (bp)
M
11 accessions

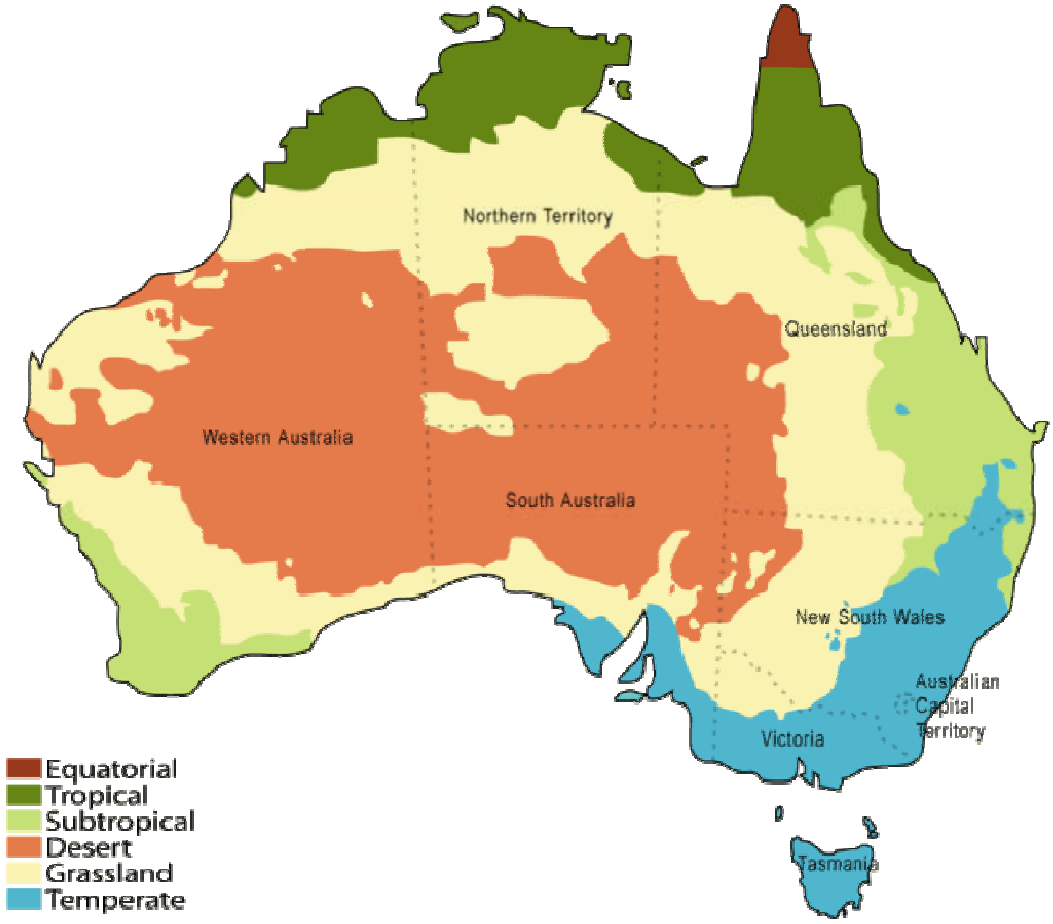

G088
frequency (%)
100
80
60
40
20
0
104 105 107 109 111 113 115 117 119 121 123 125 127 129 131 133 135 139 141
allele size (bp)
C
415 accessions
frequency (%)
100
80
60
40
20
0
104 105 107 109 111 113 115 117 119 121 123 125 127 129 131 133 135 139 141
allele size (bp)
W
189 accessions
frequency (%)
100
80
60
40
20
0
104 105 107 109 111 113 115 117 119 121 123 125 127 129 131 133 135 139 141
allele size (bp)
M
11 accessions

G264


frequency (%)
100
80
60
40
20
0
156 158 160 162 166 168 170 172 174 176 178 180 182 184 186 188 190 198
allele size (bp)
C
415 accessions


frequency (%)
100
80
60
40
20
0
156 158 160 162 166 168 170 172 174 176 178 180 182 184 186 188 190 198
allele size (bp)
W
189 accessions


frequency (%)
100
80
60
40
20
0
156 158 160 162 166 168 170 172 174 176 178 180 182 184 186 188 190 198
allele size (bp)
M
11 accessions



Appendix Figure 5 Climatic zones in Australia, on the basis of Köppen classification.

Source: Wikipedia (2008)

CIRRICULUM VITAE

NAME : Ms. Chontira Sangsiri

BIRTH DATE : May 25, 1981

BIRTH PLACE : Phitsanulok, Thailand

EDUCATION :	**YEAR**	**INSTITUTE**	**DEGREE/DIPLOMA**
	2002	Kasetsart Univ.	B.S. (Agriculture)

POSITION/TITLE : -

WORK PLACE : -

SCHOLARSHIP/AWARD : 2002-2004: Assistant researcher scholarship (Center for Agricultural Biotechnology)
2004-2008: Thailand Research Fund; Thai Royal Golden Jubilee Scholarship