





**IDENTIFICATION OF MYANMAR LANDRACE SOYBEAN BY SSR
MARKER AND EVALUATION OF AGRICULTURAL CHARACTERS
IN RECOMBINANT INBRED LINES OF VEGETABLE SOYBEAN**

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Title: Identification of Myanmar Landrace Soybean by SSR
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Recombinant Inbred Line of Vegetable Soybean

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ABSTRACT

Identification of genetic diversity among 14 Myanmar landrace soybean (*Glycine max* L. Merr.) cultivars and comparison with vegetable soybean by using SSR profiling for understanding genetic background, were conducted in this experiment. Simple-sequence repeat (SSR) loci of 14 accessions from different geographic areas of Myanmar and 3 vegetable soybean cultivars was able to detect 174 alleles with 53 markers of 20 molecular linkage groups (MLGs). Genetic similarity was calculated by Dice's coefficient which showed the estimation symmetric similarity value ranging from 0.37 to 0.94 among all accessions.

A dendrogram generated from the SAHN clustering using the unweighted pair group method arithmetic average (UPGMA) by NTSYSpc (version 2.2) was used to separate the three distinct groups of Southern Shan cultivar from Northern Shan, and that of Morbi which was in vegetable soybean group with average tree similarity coefficient estimated at 0.37. All Southern Shan cultivars of SS1, SS2, SS3, SS4 and SS5 were classified into the same cluster. SS1, which stood far from the entire majority of accession. Southern Shan and Northern Shan were separated at an estimated tree similarity coefficient of 0.40.

Identity and purity using the hilum color and that of Northern Shan accessions NS1a and NS1b, showed symmetric similarity coefficient at 0.91 while NS2a and NS2b showed estimate coefficient at 0.93 and Hto Nauk Ahlan HNAA and HNAB at 0.63. Northern Shan 4 and Northern Shan 5 were similar at 0.94 of tree similarity index. Two Northern Shan cultivars of Hto Kai Lyan and Hto Phoshe and accession from lower region of Morbi were clustered in vegetable soybean group.

Among vegetable soybeans, Cha mame was classified into Northern Shan group at symmetric similarity coefficients of 0.37 with AGS292 and No75 while those two cultivars were at 0.43.

The genetic similarity was estimated at a range of 1.00 to 0.00 among all accessions, which meant that some accessions appeared identical while some accessions appeared to have totally different genotypes based on SSRs. The tree plot suggested that all populations formed were from different germplasm pools. The field performances of agricultural characters also significantly showed the distinct characteristics of Myanmar landrace and vegetable soybean.

Evaluation of yield and yield components with the scope of enhancing productivity of the agricultural characters of the recombinant inbred lines derived from a cross of AGS292 versus Nakhon Sawan 1 (NS1) which were carried out in the Ornamental Horticulture Division, Maejo University, Chiang Mai, during 2009, showed an obvious level of limitation for the diverse soybean-growing environments of early rainy and late rainy growing seasons which could quantify and integrate the hypotheses about how crops respond to the environment and specifically to different environmental conditions. The genotypes were planted in a randomized complete block design with three replications in each season and were harvested at the R_6 stage. Maximum and minimum graded fresh pod weights were observed at 9.7 ton/ha (RIL41) and 4.9 ton / ha (RIL3) by combined analysis while fresh green seed yields were at 5.9 ton/ha and 2.7 ton/ha, respectively. Yield of RIL120 with graded fresh pod (11.6 ton/ha) and fresh green seed yields (7.5 ton /ha) showed the highest production potential in late rainy season.

Eating quality was highly significant among all seasons versus varieties or among varieties with highly accepted eating quality of RIL3 for all characters of appearance, taste, texture and flavor. All other RILs also showed higher value than parent lines of AGS292 and NS1. Results observed from this study showed the influence of the environment on some traits of vegetable soybean varieties and also highly significant on season versus varieties which can serve as basic information for future vegetable soybean breeding program in Myanmar.

Keywords: Landraces soybean, vegetable soybean, SSR marker.

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CHAPTER 1

Introduction

Soybean was domesticated in East Asia, where various kinds of landraces have been established as a result of adaptation to different environments and the diversification of food cultures. Asia is thus an important germplasm pool of soybean. The national area production and productivity of soybean in Myanmar are reported to be over 0.116 million hectares. The potential to increase national production of soybeans, both horizontally as well as vertically can be harness through systemic research and development to identify appropriate varieties and production technology for different eco region of the country. There exist bright prospects to repeat the success story of soybeans for improving socioeconomic status of farmers, edible oil economy and national economy of the country. This is feasible, particularly because of the zeal and enthusiasm of all the concerned in the country for the commercial exploitation of this wonder crop.

Most of the area is under local varieties. In the absence of data of the varieties performances in different regions and for the minimum number of three crop seasons, identification of varieties for stable superior performance over the local cultivars would not be reliable at this stage. However, on the basic of the available information and the latitude of the three eco- regions some varieties are likely to have promise in the respective regions of Hilly, Central and Lower Myanmar. The strategy for making available varieties promising soybean varieties for cultivation could be short term and long term. In long term strategy domestic breeding program of soybean varieties should be initiated with collection of germplasm, identifying breeding objectives using existing and induced heritable for selection by different breeding procedures.

There are many landraces from common ancestors that have been segregated by farmers' selections for local environment and farmers' preferences. Even the same cultivars sometimes have different commercial names, perhaps due to sequential local distributions.

In general, the genetic bases among soybean collections are different, and they could be useful sources for increasing the diversity in breeding process (Cui *et al.*, 2000; Zhou *et al.*, 2000; Zhou *et al.*, 2002).

Vegetable soybean has been reported to be better tasting and suitable for human consumption than grain soybean (Weber, 1956). Vegetable soybean is a large-seeded (seed dry wt. >250 mg/seed), green vegetable soybean has a sweet, nutty flavor and can be eaten as snack either boiled in salt water or roasted like peanut seed. Fresh or frozen vegetable soybean can be cooked just like sweet peas (*Pisum sativum* L.) or lima beans (*Phaseolus limensis* L.), either stir fried or added to stews and soups. In Asia, where edamame is an important vegetable, farmers harvest stems with fresh green pods before full maturity when pods are fully filled, nearly 80% matured, and just before turning yellow (Shanmugasundaram *et al.*, 1991). This stage corresponds to the R₆ stage of soybean development (Fehr *et al.*, 1971).

Now a day, vegetable soybean has been introduced to Myanmar and research were done for varietals improvement. Breeding strategy for making available varieties for new introduced vegetable soybean varieties for cultivation should be initiated with collection of germplasm, identifying breeding objectives using existing and induced heritable for selection by different breeding procedures as well as using landraces as genetic resources for specific adaptation to particular environment . Success in plant breeding depends on the selection of parents which are highly diverse for the genes controlling productivity, but which can provide a reasonable high proportion of descendants with adequate product quality.

There is insufficient variation in other morphological traits on which to estimate genetic diversity. Direct estimation of genetic differences the DNA level can be used to identify cultivars and also assists in the choice of parents. The use of neutral DNA markers has been adapted as a method for understanding genetic diversity and for fingerprinting cultivars (Rongwen *et al.*, 1995) especially when details in pedigree information are lacking.

A need exists, therefore, to evaluate and identify the soybean genotypes for potential production as a source of vegetable traits for breeding suitable cultivars.

SSR markers have been used to detect genetic diversity and or important agronomic traits in soybean (Akkaya *et al.*, 1992, 1995; Cregan *et al.*, 1999; Brown Guedira *et al.*, 2000) because of their polymorphic rate and multiple alleles.

In soybeans SSRs detected the higher expected heterozygosity compare to other genetic marker such as RFLP, AFLP, and in soybeans were highly correlated while SSR generated hyper variable polymorphisms (Rongwen *et al.*, 1995). Thus SSR markers were employed as the primary approach to detect differences among accession and elite soybean lines. SSR diversity is used to study the genetic diversity among the elite vegetable soybean.

Broad based rich germplasm collection is the basic requirement of a worthwhile breeding programmed in varietal improvement. Germplasm collection as landraces is a source of genetic variability for soybean breeding program.

Recombinant inbred lines of vegetable soybean quantify and integrate hypotheses about how crops respond to the environment and specifically to different environmental conditions. These simulation models can provide explicit predictions of crop production and impacts on natural resources. They also can guide research on more fundamental questions such as how genetic alterations to partitioning affect crop growth.

The narrow genetic base of modern soybean (*Glycine max*) has been caused by a limited initial based and several decades of intensive breeding and selection. Since genetic variability is necessary for genetic progress, the limitation of genetic diversity may impede further advanced in soybean breeding unless new source of genetic variability are introduced into breeding program or the genetic relationships among the cultivar to assist in choose of parent in future breeding programs. Characterization of diversity by using genetic markers will be useful to breeders for selecting landraces to be used in further research.

Evaluate the agricultural characters of the recombinant inbred lines derived from a cross of AGS292 verses to Nakhon Sawan-1 (NS1) will obviously be a level of limitation for the diverse soybean-growing environments of early rainy and late rainy growing seasons.

The result from this experiment will be useful to breeders for utilizing genetically diverse introductions in soybean improvement or will be efficient breeding strategies for local adaptation by using the selected diverse parents.

This study should be significant for soybean researchers to understand the whole feature of Myanmar- landraces soybean. As well as this study can be used as a model method for selecting a collection for multi-species germplasm collection.

From the agricultural data collection from RILs lines, more adaptable and stable in yield and yield components will indicate the presence of linear and non-linear components of genotype $\times$ environment interaction and we can also observed the favorable environments for recombinant inbred lines for superior, stable and graded green pod yield, or responsive in unfavorable environments.

1.1 Objective

The objectives of this study were;

- (1) To identify the genetic diversity among fourteen Myanmar landraces compare with vegetable soybean varieties of AGS292, No75, and Cha mame by using SSR profiling for understanding genetic background.
- (2) To evaluate the agricultural characters of the recombinant inbred lines derived from a cross of AGS292 verses to Nakhon Sawan-1 (NS1) in two environments.
- (3) To quantify the genetic relationships among the cultivar to assist in choose of parent in future breeding programs.

CHAPTER 2

Literature Review

2.1 Soybean

Soybean (*Glycine max* (L.) Merrill) can be classified into grain soybean and vegetable soybean depending on its form of utilization and nutritional value. The Grain soybean normally has small seed size and consumed as processed products. Soybean is traditionally produced for oil and protein in the seed, which are the economically important seed quality components of the crop. These are “feed grade” or field soybeans that are allowed to dry on the plant and then used as nutritious feed for livestock. The primary constituents of soybean seed, on a dry matter basis, contained about 40% protein, 21% oil, and 11% soluble carbohydrates (Openshaw and Hadley, 1981). Depending on its form of utilization and nutritional value, although both types of soybeans are the same species, the vegetable soybean differs from the field soybean in that the bean is larger in size with a sweeter taste, more tender texture, and better digestibility. There are many soybean cultivars throughout the world but only a few of them are cultivated for consumption as cooked, immature seeds and eat as vegetable.

2.2 Landrace

A landrace is a local variety of a domesticated plant species which has developed largely by natural processes, by adaptation to the natural and cultural environment in which it lives. It differs from a formal breed which has been bred deliberately to conform to a particular standard type. Landraces are usually more genetically and physically diverse than formal breeds. Many formal breeds originated from landraces, and sometimes a particular type has both landrace and formal breed populations. Sometimes a formalized breed retains the “landrace” name, despite no longer being a true landrace.

The term “landrace” has additionally been defined as “an autochthonous landrace is a variety with a high capacity to tolerate biotic and abiotic stress, resulting in high yield stability and an intermediate yield level under a low input agricultural system.”

Landrace populations are often highly variable in appearance, but they are each identifiable morphologically and have a certain genetic integrity. Farmers usually give them local names. A landrace has particular properties or characteristics. Some are considered early maturing and some late. Each has a reputation for adaptation to particular soil types according to the traditional peasant soil classifications, e.g. heavy or light, warm or cold, dry or wet, strong or weak. They also may be classified according to expected usage; among cereals, different landraces are used for flour, for porridge, for bulgur, and for malt to make beer, etc. All components of the population are adapted to local climatic conditions, cultural practices, and disease and pests. But most important, they are genetically diverse. They are balanced populations – variable, in equilibrium with both environment and pathogens and genetically dynamic.

2.3 Vegetable soybean

Vegetable soybean called edamame in Japan, green soybean or eatable soybean in North America and maodou in China. Edamame is a type of soybean selected for fresh or frozen vegetable use at an immature stage which has a similar protein content, mild flavor, nuttier texture and is easier to cook when compared to grain soybean. It is being promoted as a new vegetable for global consumption. Most edamame varieties are bushy plants about two feet tall, similar in appearance to bush beans, with maturity time ranging from about 75 to 125 days. The plants are as easy to grow as bush beans, requiring full sun, fertile, well-drained soil, and warm temperatures. Harvest time is short, as the beans on a plant all mature at about the same time, a month or so after flowering. It can make several small plantings in succession, or plant several varieties of different maturation, to extend the harvest period.

The pods are bright green, about 2.5 inches in length and covered with light hairs, with two to three plump, bright green seeds inside. Refrigerate pods after picking to maintain freshness. It is eaten fresh or salted right from the prepared pod as a snack or appetizer with a sweet, nutty flavor, much as we eat peanuts. It is served as a side dish, just as we would serve garden peas or lima beans, or added to stir-fries, soups, stews, succotash, and salads. It can be eaten hot or cold; and it is also easy to freeze, either shelled or in the pod, for use in winter. Taiwan and China are major suppliers of frozen edamame to the United States.

2.4 Nutritional composition

Fresh vegetable soybean seed has 35% to 38% protein (dry weight basis) and 5% to 7% lipid on fresh weight basis. Brix reading are generally between 8.5% and 12.0%. Monounsaturated fatty acids constitute a greater fraction of lipids in fresh green seeds (Johnson *et al.*, 1999), which makes vegetable soybean a nutritious snack. Soybean is one of the few natural sources of isoflavones (78 to 220 $\mu\text{g/g}$ dried seed depending upon isoflavone type) and tocopherols (vitamin E) which range from 84 to 128 $\mu\text{g/g}$ dried seed (Mohamed *et al.*, 2001). Proximate analysis of seed nutritional composition of edamame in Colorado, US (Johnson *et al.*, 1999), and Japan (Masuda, 1991) indicated that edamame has superior nutritional content than green peas. The calorific value (energy) of vegetable soybean is about 6 times that of green peas. The vegetable soybean contains 60% more Ca, and twice the P and K of green peas. The Na and carotene content of vegetable soybean is about one-third that of green peas and has similar quantities of iron, vitamins B1 and B2. Vegetable soybean is rich in ascorbic acid but low in niacin (Masuda, 1991).

2.5 Agricultural importance

Development of improved soybean cultivars for soy-foods offers potential for expanding the domestic and international soybean market. The US farmers, particularly organic vegetable producers will have another vegetable crop to choose to extend crop rotations, supplement farm income, and spread risk. As a legume crop, vegetable soybean is a low input, soil enriching crop that could help farmer minimize insect and disease buildup as well.

These cultivars serve dual purposes of pod production and as a green manure crop to replenish soil nutrient levels including nitrogen, soil organic matter and improve soil structure and sustainability (Shanmugasundaram, 2001). Vegetable soybean, due to their short duration (99 to 120 days for MG V-VII), fit well into existing crop rotation patterns, dual purpose varieties serve as main summer crop and enable farmers to turn in green crop residue as green manure before planting crops next spring.

Currently, edamame cultivars that produce high pod yields and also high amount of biomass are being developed through breeding at the Asian Vegetable Research and Development Center, Taiwan.

The initial research on vegetable soybean at AVRDC was a contract research project with Council of Agriculture in Taiwan aimed at developing varieties suitable for export to Japan. Increasing interest and demand for vegetable soybean in other countries encouraged AVRDC to broaden its research scope to cover adaptation of the crop in tropic and subtropics with the objectives of develop varieties suitable for export market, high marketable yield, disease resistance and develop adaption to mechanization.

2.6 Current status of soybean growing in Myanmar

Soybean is traditionally produced for oil and protein in the seed which are economically important seed quality components of the crops. Vegetable soybean is a new introduced variety to Myanmar and for the efficiency of specific research of practical utility in soybeans, it would be of far reaching consequence to systematize the research with multiplication inter disciplinary approach under research project to start with the identification or principal investigation for different soybean landrace cultivars from different regions.

Production of vegetable soybean is similar to grain soybean except that vegetable soybeans are harvested when the pods are still green and full. It is being promoted as a new vegetable for global consumption and will require breeding program for local adoption. A need exist therefore to evaluate and identified the soybean genotypes for potential production as a source of vegetable traits for breeding suitable cultivars. As the demand for vegetable type soybean is increasing in the international market and the need for development of productive cultivars with desirable seed size is increasingly important (Mian *et al.*, 1996), the introduction for growing vegetable soybean will be success in Myanmar.

2.7 Molecular markers

The traditional method of soybean breeding involves artificial hybridization to develop genetic variability followed by self fertilization and phenotypic selection for traits of interest among the offspring (Hoeck *et al.*, 2003). The development of methods to obtain an accurate assessment of genetic variations has been of a great interest to scientists. The rapid classification of different taxonomic groups, breeders are concerned with the determination of valuable variations in breeding programs.

In the context of cultivar registration, plant patents, and breeder's right protection, a quick and reliable method for cultivar identification is particularly appealing. Molecular markers may improve traditional methods of breeding for traits of interest of vegetable soybean by increasing the reliability which desirable progeny are selected. There are three types of genetic markers

1. Morphological markers which themselves are phenotypic traits or characters such as flower color, seed shape, growth habit;
2. Biochemical markers, which include allelic variant of enzymes called isozymes which are limited in number and are often influence by environmental factors or developmental stages of the plant;
3. DNA or molecular markers which reveal site of variation in DNA (Collard *et al.*, 2005).

DNA or molecular markers is the most widely use types of marker predominantly due to their abundance. DNA markers may be broadly divided into three classes based on the method of their detection; 1) hybridization based, 2) polymerase chain reaction (PCR based), and DNA sequence based (Karp and Edwards, 1997). DNA markers are particularly useful if they reveal difference between individuals of the same or different species.

The use of different molecular markers to describe genetic polymorphism in natural populations limits across species and prevent any general conclusions on the structure of genetic diversity. However, trends of variations within species can be different when diversity surveys are conducted with different markers on the same set of populations.

2.8 Simple sequences repeat (SSR) or microsatellite

Simple sequences repeat (SSR) or microsatellites are DNA sequences that consist of two to five nucleotide core units which are tendomly repeated. These small repetitive DNA sequences provide the basic of a polymerase chain reaction (PCR) base, multi-allelic, co-dominant genetic marker system. The region flanking the microsatellite is generally conserved among genotypes of the same species. PCR primers to the flanking regions are used to amplified the SSR- containing DNA fragment length polymorphism is created when PCR product from different individual vary in length as a result of variation in the number of repeat unit in the SSR.

SSR markers are being used for the construction of linkage maps in various plant species *Arabidopsis thaliana* (Bell and Ecker, 1994), soybean (*Glycine max* (L.) Merr; Akkaya *et al.*, 1995) barley (*Hordium vulgare* L.; Liu *et al.*, 1996) and maize (*Zea mays* L.; Senior *et al.*, 1996). SSR provide high level of length polymorphism with as many as 37 alleles at individuals in barley (Saghai- Maroof *et al.*, 1994) and 26 alleles in soybean (Rongwen *et al.*, 1995). In most plant species the level of polymorphism is considerably higher than that found with RFLP markers (Diwan and Cregan, 1997; Plaschke *et al.*, 1995; Saghai- Maroof *et al.*, 1994). A comparative study on the performance of different types of markers in soybean genetic analysis showed that microsatellite markers have a greater degree of polymorphism and, thus, better discrimination between genotypes (Powell *et al.*, 1996).

These DNA markers have been shown to be highly polymorphic in soybean (Akkaya *et al.*, 1992; Diwan and Cregan, 1997). SSRs are composed of a 1- to 6-base pair (bp) DNA sequence that is repeated a variable number of times. SSRs are amplified by PCR with primers that are complementary to the conserved sequences that flank an SSR locus. Polymorphic fragment alleles resulting from variation in SSR repeat length are separated electrophoretically to display genetic profile of individuals. SSR alleles typically show monogenic codominant inheritance that enable classification of homozygote and heterozygote in a segregating population. Given this high level of informative, abundance and apparent random distribution in plant genomes combined with reliable amplification via PCR, SSR markers are likely to become an important and widely used DNA marker system in plant.

More than 600 SSRs have been developed and mapped in 20 molecular linkage groups in soybean (Cregan *et al.*, 1999). These SSRs mostly represent polymorphism at single loci and there is no ambiguity in scoring genotypes, in contrast with analyses with RFLP probes that often hybridize to two or more positions in the soybean genome. High levels of polymorphism at SSRs have been reported for both the numbers of alleles per locus and the gene diversity (Maughan *et al.*, 1995; Powell *et al.*, 1996; Diwan and Cregan, 1997; Song *et al.*, 1999; Narvel *et al.*, 2000). These values are much higher than those reported for RFLP markers (Keim *et al.* 1989; Kisha *et al.*, 1998) and isozymes (Perry *et al.*, 1991; Griffin and Palmer, 1995; Han *et al.*, 1999; Hirata *et al.*, 1999).

2.9 Genetic diversity study by SSR markers

Global production will require breeding programs for local adaptation; however, limited research has been published on genetic diversity of edamame varieties for the assessment of genetic resources. Simple sequence repeats were used to study the genetic diversity among 130 accessions, including edamame cultivars and landraces from Japan, China and the US, and also the new breeding lines in the US. Although it is assumed that elite edamame cultivars would have narrow genetic diversity, seventeen SSRs detected polymorphism to distinguish 99 of the 130 accessions. The cluster analysis generated nine clusters and 18 outliers. Genetic diversity within Japanese edamame was lower than that within Chinese vegetable soybean accessions (maodou), even though only 10 Chinese maodou were analyzed compared to 107 Japanese edamame. Cluster analysis revealed that the patterns of SSR diversity in edamame can generally distinguish maturity classes and testa color and that Japanese edamame have a narrow genetic base different from others and that SSRs can describe the patterns of genetic diversity among the elite vegetable soybean (Makiko *et al.*, 2007).

To obtain a better understanding of the genetic relationships among populations of different geographical regions in Asia, scientists were evaluated the genetic diversity and pattern of genetic variation in Asian cultivated soybeans by examining the length polymorphism of alleles found in 20 SSR loci from different linkage groups. In order to evaluate the genetic structure of the Asian soybean population, allelic profiles at 20 simple-sequence repeat (SSR) loci of 131 accessions introduced from 14 Asian countries were analyzed. The SSR loci produced an average of 11.9 alleles and a mean gene diversity of 0.782 in the accessions tested.

Quantification theory III analysis and cluster analysis with the UPGMA method clearly separated the Japanese from the Chinese accessions, suggesting that the Japanese and Chinese populations formed different germplasm pools. The Korean accessions were involved in both germplasm pools, whereas most of the accessions from Southeast and South/Central Asia were derived from the Chinese pool. Relatively high genetic diversity and the absence of region-specific clusters in the Southeast and South/Central Asian populations suggest that soybean in these areas has been introduced repeatedly and independently from the diverse Chinese germplasm pool. The result indicates that the two germplasm pools can be used as exotic genetic resources to enlarge the genetic bases of the respective Asian soybean populations.

To use, maintain and increase crop germplasm collections efficiently, it is important to assess the diversity of these collections. Chinese spring sowing soybeans (*Glycine max*) were analyzed by using SSR analysis. Random-repeated sampling within landraces of different geographical regions suggested that there was a relationship between genetic distances and geographical distances among North spring soybean and South spring soybean but because of the uneven distribution of SSR variation soybean populations from different regions, indicating a certain degree of geographical differentiation among Chinese soybean germplasm collections.

2.10 Recombinant Inbred Line (RILs)

Populations of recombinant inbred lines (RILs) are obtained by successive self-population from F_2 individuals. At each individual is chosen, which will be the parents of the next generation, single seed descent (SSD). This recurrent process has three important genetic consequences. At each generation of self-pollination each heterozygous locus has one chance in two of being fixed, the mean heterozygosity of the genome decrease rapidly at a rate of $1/2^n$, n being the number of generation after the F_1 . In F_5 93.75% of the genome is fixed, 96.88% in F_6 , 98.44% in F_7 and 99.80% at F_{10} . The individual of a family derived from a given F_2 individual after n generations are all genetically identical, except for the fraction of the genome still heterozygous at the generation at $n-1$. Due to the successive meiosis, recombination has occurred and each family derived from a given F_2 individual has fixed a particular allelic combination at different loci (hence the term recombinant lines).

For a given pair of loci, the proportion R of lines that have recombined can be calculated. But this proportion is not an estimate of r . Indeed, with successive generations, there are several chances for recombination. When self pollination is impossible, recombinant lines can be constructed from full-sib crosses. The rate of fixation is lower.

2.11 Environment effects

Crop performance depends on the genotypes, the environment in which the crops are grown, and the interaction between the genotype and the environment. Genotype and some factors of the environment, such as fertilizer rate, populations and pest control, can be controlled by the researcher.

But the other factors of environment such as sunshine, rainfall, and some soil properties, are generally fixed and difficult to mortify for a given site and planting season. Thus a researcher with a onetime experiment at a single site can vary and evaluate only the controllable factors but not the environmental factors that are beyond the control.

Genotype by environment interaction is the major element in determining many key aspects of a breeding programmed including whether to aim for wide or specific adoptions or choice of locations for selection; whether selection in early generation is conducted in stress or stress free environments; and the trade- off between multi- environment testing of large number of genotypes and subjecting fewer lines to interview trait-based selection. The sampling problem, associated with yearly variation, suggests testing for many crop cycles.

Testing over a wide geographic range can ensure a parallel degree of temporal buffering capacity in their germplasm. Atlin and Frey (1989) concluded that the need for separate breeding programmed for low and high inputs environments depend on the extent to which the yield in the two types of environments are under separate genetic controlled and on the accuracy of selection. Yield in the two environments must be controlled by different alleles to justify different programmed. Probe genotypes, near isogenic pairs with differential responds to known environmental stimuli, provide a more quantitative diagnostic approach to target environments, particularly for factors that are not easily observed. A marked divergence in the performance of a pair in a test environment indicates a high probability of a stress, such as a micronutrient imbalance in the soil or the present of soil borne pathogens.

In general, a plant breeder is concerned with developing a genotype with better mean performance than accepted genotypes and reasonable stability across a wide range of environment and low probability of failing to achieve a maximum acceptable yield. Composition of soybean seed can be affected not only by cultivar but also by environmental factors.

Previous investigations have shown that difference in seed is composition inherent among cultivars (Simpson and Wilcox, 1983; Hartwig and Kilen, 1991; Helms and Orf, 1998). But it has also been frequently observed that the same cultivar, when grown in different year or under different environments in the same year, varies significantly in seed composition. Helms *et al.* (1990) found that protein concentration of soybean increases as planting date is delayed. Shannon *et al.* (1972) and Burton (1985) have reported a negative correlation between soybean seed yield and seed protein concentration but both genetic and environmental factors can strongly affected the seed composition (Krishnan, 2000).

Significant in genetic and environmental impacts on isoflavones concentration in soybean seed has been reported. Wang *et al.* (1994) observed that total isoflavones concentration ranged from 1161 to 2743 $\mu\text{g g}^{-1}$ in 210 soybean cultivars grown in south Dakota. Hoeck *et al.* (2003) showed the genotype, genotype x year, genotype x location, and genotype x year x locations interactions were all significant for both total and individual isoflavones concentrations. Eldridge and Kwolek, (1983) reported that total isoflavones concentration in soybean seed varied from 1160 to 3090 $\mu\text{g g}^{-1}$ among four soybean cultivars grown in the same environment and 460 -1950 $\mu\text{g g}^{-1}$ among four locations with the same cultivar. Wang and Murphy (1994) observed that total isoflavones concentration of 'vinton 81 soybeans' (American soybean and Japanese soybean) ranged from 1176 to 3309 $\mu\text{g g}^{-1}$ among year at the same location and from 1176 to 1749 $\mu\text{g g}^{-1}$ among locations within the same years; thus year seemed to influence isoflavones concentration more than location. Kitamura *et al.* (1991) and Tsukamoto *et al.* (1995) showed that isoflavones concentration was significantly lower in soybean seed that developed in high temperature during seed filling than seed exposed to low temperatures during the filling period.

The development of an improved crop production technology usually involves a series of elimination process starting with a few superior ones that are identified and to be recommended for commercial use. For a given crop at a specific site, planting is usually not staggered uniformly over a 12 month period but is distinctly bunched in some well- defined periods that are consistently repeated over years. The primary objective of a combine analysis over seasons is to examine the interaction between season and treatment and to determine the necessity of a separate technology recommendation for each planting system.

CHAPTER 3

Materials and methods

3.1 Identification of Myanmar landrace soybean by SSR markers

3.1.1 Plant materials

Fourteen landrace soybean accessions which are commercially produced for local market consumption from different places of Myanmar especially from traditionally soybean growing area of Southern Shan State, Northern Shan State and from the lower Myanmar Morbi and compared sample varieties of introduced cultivars of AGS292, Cha mame, and No75 were used in this experiment. All Myanmar accessions were landraces or pure-line selections (Table 1) and cultivars identity and purity were confirmed using the hilum color, seed coat, corolla color and seed size (Chowdhury *et al.*, 2000).

3.1.2 Laboratory detection

3.1.2.1 DNA extraction

Genomic DNA was extracted from the first trifoliate fresh leaves according to the method of rapid isolation protocol of Doyle and Doyle (1987), a modified CTAB (hexadecyltrimethyl ammonium bromide) extraction. The extracted DNA was air dried and dissolved in Tris-EDTA buffer and prepared the final concentration of sample DNA adjusted to 10 ng / μ l for SSR detection. DNA of all genotypes was amplified with SSR primers of 20 molecular linkage groups. The PCR amplification reactions cycling were consisted of a 1 min denaturation at 94°C, 1 min annealing at 48°C and 2 min extension at 68°C for 32 cycles. The amplification products were analyzed by 3% agarose gel electrophoresis at the electric potential of 80 and 100 volts for 45 minutes. Gel stained with ethidium bromide was exposed to UV light and the images were taken as photographs.

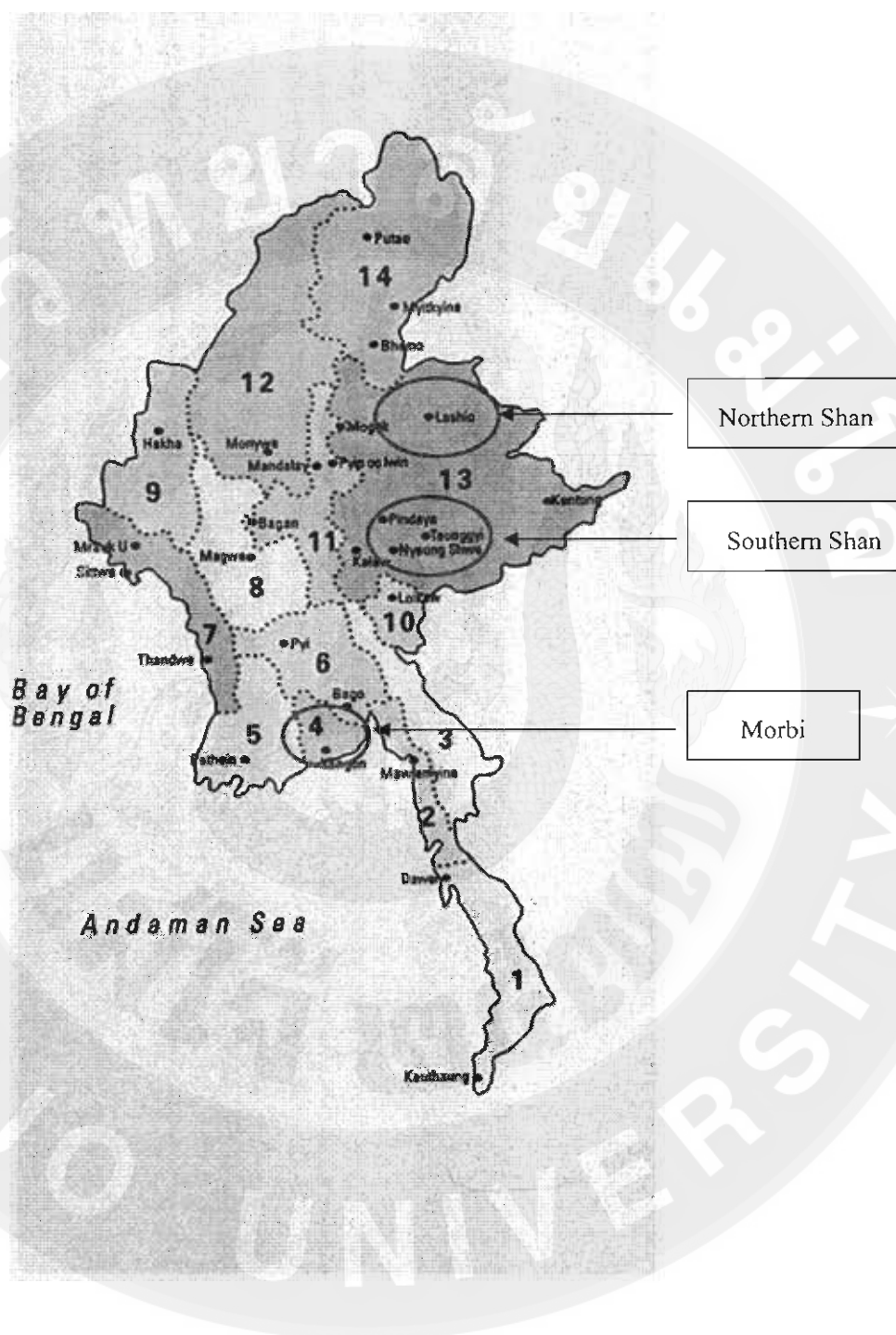


Figure 1 Geographical information of the collection site of Myanmar landrace soybean cultivars

Table 1 The collection site of Myanmar landrace soybean cultivars

Entry	Abbreviation	Part of Myanmar	Original District	Latitude	Altitude (meter)	Temperature Max/Mini	Rain fall 10 years average (inches/days)
Southern Shan 1	SS1	Southern Shan	Taunggyi	20° 39' 0" N, 96° 56' 0" E	1300	31°C/15°C	26.31
Southern Shan 2	SS2						
Southern Shan 3	SS3						
Southern Shan 4	SS4						
Southern Shan 5	SS5						
Northern Shan 1	NS1	Northern Shan	Kyaukse	22° 56' 0" N, 97° 45' 0" E	836	37°C/10°C	45.44
Northern Shan 2	NS2						
Northern Shan 3	NS3						
Northern Shan 4	NS4						
Northern Shan 5	NS5						
Hto Nauk Alan	HNA						
Hto Kai Lyan	HKL						
Hto Pho She	HPS						
Morbi		Lower Region	Morbi	16° 48' 0" N, 96° 9' 0" E	10-100	36°C/21°C	40.00

3.1.2.2 Diversity analysis

Polymorphic DNA segments amplified with SSR primer pair were assigned a letter and each band was scored as present (1) or absent (0). Diversity value for each locus was calculated using Dice's coefficient of similarity " $S = 2p/2p+q+d$ ", where p is the band present in both compared genotypes, q is the total number of polymorphic bands and d is the band absent in both compared genotypes.

A dendrogram was generated with (SAHN) clustering method using the unweighted pair-group method, arithmetic average (UPGMA) (Sneath and Sokal, 1973) by NTSYSpc (Rohlf, 2000, version 2.2) for regional group of accession based on average genetic distances between accessions of different regions.

3.1.3 Field observations

The morphological characters were observed in field which was conducted in dry season of mid December 2008 to March 2009, in the field of Maejo University, Sansai, Chiang Mai, Thailand. The treatments were sown on December 17, 2008 with a single row plot each of 5.0 meter long with 50 cm spacing between rows and 20 cm between plants. Each entry was replicated twice in a randomized complete block design. The seeds were inoculated with Rhizobium (*Bradyrhizobium japonicum*) culture by adopting standard procedure just before drilling. Three seeds were used in each hill and the seedlings were thinned down to two plants per hill at two weeks after germination. Nitrogen at 156.25 kg per hectare was applied to all plots as a basal and 7-10 days after sowing. 14-14-21 with the rate of 312.5 kg per hectare was added 20 days after sowing and 46-0-0 with the rate of 156.25 kg per hectare was applied 45 days after sowing. For insect pests or disease control, Acetamiprid 10g/20 liter and Mancozeb 10g/ 20 liter of water were sprayed 7 days interval. Concerning agricultural characteristics were collected to identify morphologically.

3.1.4 Statistical analysis of data

Data were analyzed using analysis of variance of Statistical Analyses System (SAS). ANOVAs and GLM were performed to test the statistical significance and if the significant difference was detected, further multiple comparisons among samples were conducted.

3.2 Evaluation of agricultural characters in recombinant inbred lines of vegetable soybean

3.2.1 Plant materials

To determine the relationships between yield and its components, and to find the direct and indirect effects of yield-related traits on vegetable soybean yield, the promising line of the commercial source of a population of recombinant inbred lines derived from a cross between AGS292 and NS1 while the former is very popular vegetable soybean cultivars for export and the latter famous for local market of Thailand were used in this study. The RILs selected from the largest seed size were grown for two seasons of early rainy and late rainy to compare the agricultural characters with parents at different environments.

3.2.2 Experimental details

Advanced yield trials of the five RILs compare with parents AGS292 and NS1 were conducted for two environments in the field of Maejo University, Sansai, Chiang Mai, Thailand during early rainy season (June 2009 to August 2009) and late rainy season (late August 2009 to October 2009). The experimental design consisted of a randomized complete block with three replications. The plot size was 4m x 2m with 50 cm spacing between rows and 20 cm between plants sown in four rows per plot. Three seeds were sown in each hill and the seedlings were thinned down to two plants per hill at two weeks after germination. For each replication, ten sample of graded plant were taken at random for data collection on concerning agricultural characteristics such as days to emergence, days to first flowering, plant height at first flowering, days to harvest or plant height at harvesting time, pods per plant, seed per pod, total seed weight, 100 seed weight were collected to identify of morphologically. AVRDC's suggested cultural practices were used in growing the crops, (Shanmugasundaram and Yan, 2001). The whole plot was harvested by cutting the plant at the ground level, and observations were recorded from seedling emergence till harvest.

At R_6 stage, the plant were harvested by cutting or uprooting the whole plant, selected from a random starting point in the center row. The pods from individual plants are than stripped. Stems with leaves were then returned to the field or fed to the cattle.

Initial sorting was done by selecting two- and three-seeded quality pods called graded pods. Single-seeded, malformed or other rejected pods are shelled and beans are marked domestically.

3.2.3 Eating quality test

Because of the complexity of analyzing of overall quality, panel tests are the most frequently used evaluation method (Tsou and Hong, 1991). Major quality requirements of vegetable soybean in terms of palatability are appearance, taste, flavor, and texture. Test on taste were conducted involving representative from students of Maejo University's undergrad students. As soon as after harvest at R_6 stage, (when pods were fully filled, nearly 80% mature, and just before turning yellow (Shanmugasundaram *et al.*, 1991), fresh green pods were put into boiling water and cooked for 10 minutes with or without a pinch of salt, then placed in cool water for a minute or two. Drained and served cold. As a snack, use thumb and forefinger to squeeze beans from pod: eat out of hand (Mentreddy *et al.*, 2002). In order to fetch a good price at the market the appearance of the pods were evaluate and scored for appearance, taste, flavor and texture.

3.2.4 Statistical Analysis

The data were subjected to statistical analysis with SAS software [SAS, 2000]. The important yield contributing components were determined by test for homogeneity of variance in combining data from a series of experiment then combine analysis over season or season x cultivar as random effects, used to test differences between cultivars.

CHAPTER 4

Results

4.1 Identification of Myanmar landraces soybean by SSR markers

4.1.1 Laboratory detection

Out of the 130 SSR primers, 53 SSR primers from 20 molecular linkage groups (MLGs) could be detected total of 174 polymorphic alleles (Table 2). Genetic similarity was calculated by Dice's coefficient which showed the estimation symmetric similarity value ranged from 0.37 to 0.94 among all accessions (Table 3).

A dendrogram generated from the SAHN clustering using the unweighted pair group method arithmetic average (UPGMA) by NTSYSpc (version 2.2) could be separated into three distinct groups of the Southern Shan cultivar from the Northern Shan, and that of Morbi which were in vegetable soybean group with average tree similarity of estimating at 0.37 of coefficient (Figure 1). The tree plot suggested that all the populations formed were from different germplasm pools.

All Southern cultivars SS1, SS2, SS3, SS4 and SS5 were fell into the same cluster. SS1 stood far from all the majority of accessions. Southern Shan and Northern Shan were separated at estimating tree similarity coefficient of 0.40.

The Northern Shan cultivars were scattered in two clusters of Northern Shan and in vegetable soybean group. Among the eight Northern Shan accessions, identity and purity bare confirmed using the hilum color of Northern Shan accessions NS1a and NS1b showed symmetric similarity coefficient at 0.91 while NS2a and NS2b showed 0.93 then HNAA and HNAB showed 0.63. Northern Shan 4 and Northern Shan 5 were similar at coefficient of 0.94 of tree similarity index. Two Northern Shan cultivars of Hto Kai Lyan and Hto Phoshe were clustered in vegetable soybean group.

Among vegetable soybean, Cha mame was fell into Northern Shan group with symmetric similarity coefficients at 0.37 with AGS292 and No75 while those two cultivars were stood at 0.43.

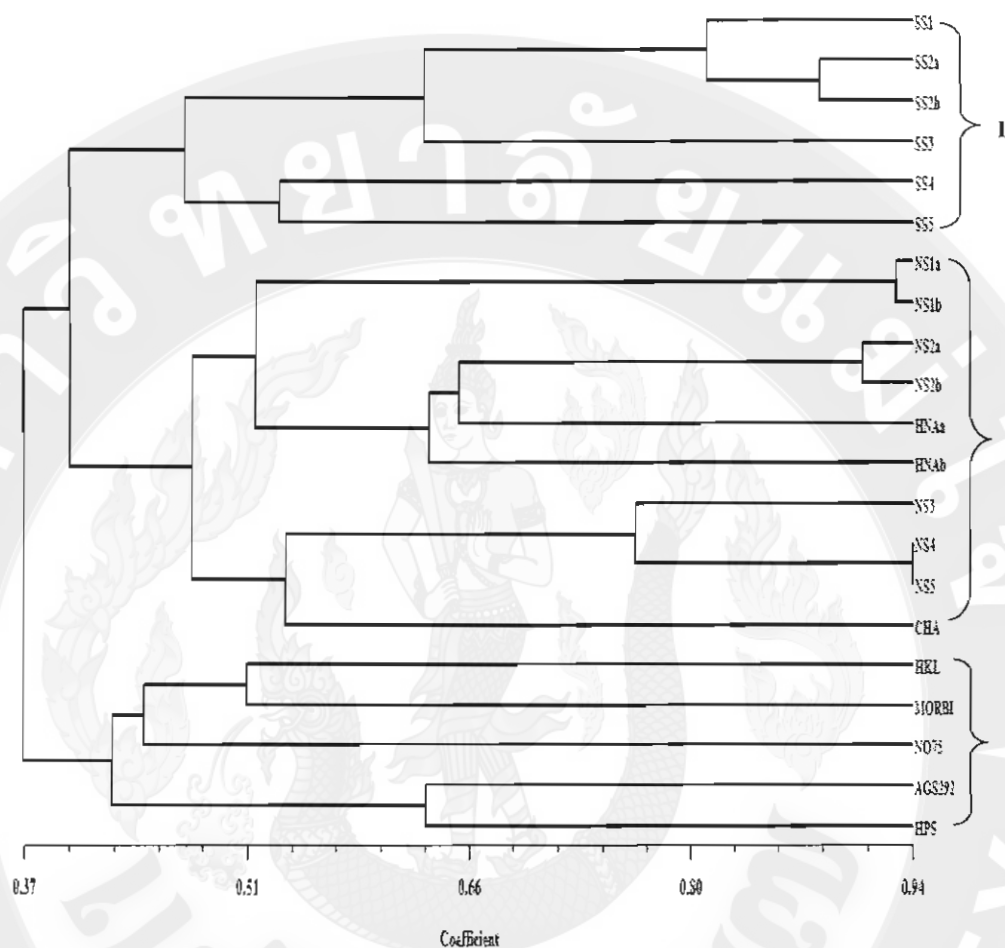


Figure 2 The UPGMA, the dendrogram constructed based on the Dice's similarity coefficient between accessions of different populations by NTSYSpc (version 2.2), representing relationships among three regional populations in Myanmar landrace soybean and three vegetable soybeans.

The genetic similarity estimated was in a range of 1.00 to 0.00 among all accessions, which means that some accessions appear identical, while some accessions appear to be totally different genotypes based on the SSRs. The tree plot suggested that all the populations formed were from different germplasm pools. The field performances of agricultural characters were also significantly showed the distinct of Myanmar landrace and vegetable soybean.

Table 2 SSR markers and number of alleles at 20 MLGs

Locus	MLGs ^s	No. of alleles	Locus	MLGs	No. of alleles
Satt545	A1	6	Satt569	F	2
Satt211	A1	2	Sat_262	F	4
Satt480	A2	2	Satt663	F	4
Sat_232	A2	2	Satt313	F	3
Satt332	B1	2	Satt656	F	2
Sct_064	B2	2	Satt199	G	5
Satt399	C1	2	Sat_210	G	4
Satt682	C1	2	Sat_038	G	6
Satt460	C2	2	Sat_185	G	3
Satt681	C2	3	Sat_541	G	3
Satt357	C2	3	Satt700	I	4
Sat346	D1a+Q	2	Satt562	I	2
Sat_160	D1a+Q	4	Satt529	J	3
Satt701	D1b+W	2	Sat_396	J	6
Satt604	D1b+W	2	Satt555	K	2
Satt372	D2	2	Satt326	K	7
Satt514	D2	5	Satt499	K	3
Satt082	D2	3	Sat_293	K	3
Satt268	E	3	Sat_408	K	4
Satt720	E	3	Satt652	K	3
Satt403	E	3	Sat_286	L	5
Satt117	E	2	Sat_244	M	9
Sat_381	E	4	Satt323	M	4
Satt425	F	5	Sat_422	M	2
Satt039	F	2	Satt255	N	5
Satt595	F	2	Sat038	O	2
Sat_120	F	2			
Total					174
Average					3.28

MLG = Molecular Linkage Group

Primers showed high polymorphic band

Satt357 (MLG C2)



Sat_396 (MLG J)



Sat_244 (MLG M)



Primers showed monomorphic band

Satt364 (MLG D1a+Q)



Satt033 (MLG F)

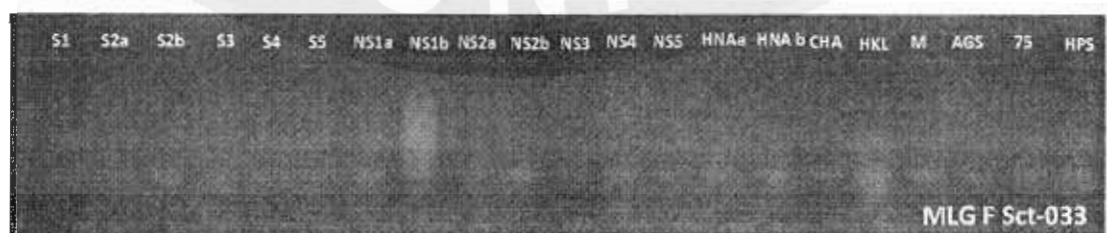


Figure 3 Amplification products showed polymorphic or monomorphic band by SSRs

Table 3 Symmetric similarity between accessions of different populations analyzed with Dice's coefficient of similarity by NTSYSpC (version 2.2), representing relationships among three regional populations in Myanmar landraces and three vegetable soybeans cultivars.

	SS1	SS2a	SS2b	SS3	SS4	SS5	NS1a	NS1b	NS2a	NS2b	NS3	NS4	NS5	HNAa	HNAb	CHA	HKL	MORBI	AGS292	NO75	HPS
SS1	1.00																				
SS2a	0.81	1.00																			
SS2b	0.81	0.88	1.00																		
SS3	0.63	0.63	0.63	1.00																	
SS4	0.47	0.47	0.47	0.47	1.00																
SS5	0.47	0.47	0.47	0.47	0.53	1.00															
NS1a	0.40	0.40	0.40	0.40	0.40	0.40	1.00														
NS1b	0.40	0.40	0.40	0.40	0.40	0.40	0.93	1.00													
NS2a	0.40	0.40	0.40	0.40	0.40	0.40	0.52	0.52	1.00												
NS2b	0.40	0.40	0.40	0.40	0.40	0.40	0.52	0.52	0.91	1.00											
NS3	0.40	0.40	0.40	0.40	0.40	0.40	0.48	0.48	0.48	0.48	1.00										
NS4	0.40	0.40	0.40	0.40	0.40	0.40	0.48	0.48	0.48	0.48	0.76	1.00									
NS5	0.40	0.40	0.40	0.40	0.40	0.40	0.48	0.48	0.48	0.48	0.76	0.94	1.00								
HNAa	0.40	0.40	0.40	0.40	0.40	0.40	0.52	0.52	0.65	0.65	0.48	0.48	0.48	1.00							
HNAb	0.40	0.40	0.40	0.40	0.40	0.40	0.52	0.52	0.63	0.63	0.48	0.48	0.48	0.63	1.00						
CHA	0.40	0.40	0.40	0.40	0.40	0.40	0.48	0.48	0.48	0.48	0.54	0.54	0.54	0.48	0.48	1.00					
HKL	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	1.00				
MORBI	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.51	1.00			
AGS292	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.43	0.43	1.00		
NO75	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.45	0.45	0.43	1.00	
HPS	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.43	0.43	0.63	0.43	1.00

4.1.2 Agricultural characters

For agricultural characters yield component analysis, 10 plants were harvested at maturity at R_8 stage from each accession. Yield of soybean plants and yield components obtained from the experiment conducted on dry season of 2008 have been recorded and the result showed were also highly significant between that of Northern Shan, Southern Shan, Morbi and vegetable soybean (Table 4-6).

Days to emergence were 6.0 and 5.1 days that of landraces and vegetable soybean. Days to first flower was 47.8 and 42.1 days then days to harvest at R_8 stage were highly significant at 110.5 days and 86.8 days of landraces and that of vegetable soybean by mean value (Table 7).

Plant height at first flower was significant at 24.2 cm and 23.2 cm between landraces and vegetable soybean while plant height at harvesting time was 51.4 cm and 25.8 cm (Table 7). Among landraces, Morbi showed the highest plant height at first flower of 25.7 cm (Table 8) and significant at 23.3 cm of Northern Shan and 24.6 cm of Southern Shan as shown in (Table 9).

For each group of plants, data were recorded according to node position on the main stem. Node one was the unifoliate node, being the first node above the cotyledons. Number of nodes per plant showed average of 14.2 among landraces and 8.2 in vegetable soybean.

Pod per plant were highly significant between landraces and vegetable soybean of 53.3 and 15.7 gram per plant (Table 7). Among landraces Morbi produced 54.0 gram per plant as shown in table 8, and Northern Shan produced 46.2 gram per plant, Southern Shan produced 57.7 gram per plant respectively (Table 9). Total dry pod yield varied 2.0 ton/ha of landraces and 0.8 ton/ha that of vegetable soybean (Table 7) and 2.5 ton/ha of Morbi (Table 8), 2.1 ton/ha of Northern Shan and Southern Shan (Table 9). Average pod weight was 45.0 Morbi in gram per plant (Table 8), 34.3 g of Northern Shan, 39.9 g Southern Shan as shown in table 9.

100 dry seed weight were 27.0 g of vegetable soybean (Table 7), 19.9 g of Morbi (Table 8), 18.6 g of Northern Shan, and 16.7 g of Southern Shan (Table 9). Total dry seed yield of 3 days after under natural sun dried were 1.9 and 0.8 ton / ha while the biomass yield were 7.38 ton / ha of landraces and 4.0 ton/ha of vegetable soybean (Table 7).

Total biomass yield were 4.0 ton/ha of vegetable soybean (Table 7), 6.5 ton/ha of Morbi (Table 8), 7.0 ton / ha of Northern Shan, and 7.7 ton/ha of Southern Shan (Table 9).

The mean value of biomass yield at R_8 stage varied between 4.0 ton/ha of vegetable soybean and 7.4 ton/ha of landraces on dry weight basis at R_8 maturity stage. Among landraces, Morbi produced 6.5 ton/ha (Table 8) and Northern Shan produced total biomass yield of 7.0 ton/ha and Southern Shan produced 7.7 ton/ha (Table 9).

The harvest index was significantly different in all cultivars of vegetable soybean and landraces (Table 7).

Information on yield by correlations of coefficient on days to emergence, days to first flower, days to harvest, plant height at first flower, plant height at harvest, number of nodes per plant, pods per plant, average pod weight per plant (g), total pod yield ton per ha, 100 dry seed weight (g), total seed yield (ton/ha), biomass yield (ton/ha), and harvesting index respectively were showed positive and significantly association among all characters except that of 100 dry seed weight which was negatively correlated with all of that characters (Table 10).

Total seed yield were positively and significantly correlated with number of nodes per plant (0.335**), pods per plant (0.438**), total pod yield ton / ha (0.570**), biomass yield ton/ha (0.508**) and harvesting index (HI) (0.662**) showed highly significant among all characters. Total pod yield (ton/ha) were positively and high significantly correlated with days to first flower (0.34**), days to harvest, plant height at harvest (R_8), number of nodes per plant, pod per plant or average pod weight per plant. According to this result, seed yield could be improved by selecting for all these characters (Table 10).

Table 4 Agricultural characters of Myanmar landrace soybean and vegetable soybean grown in dry season of December 2008-March 2009

Cultivars	Days to emergence	Days to first flower	Days to harvest	Plant height at first flower (cm)	Plant height at harvest (cm)	Node per plant
Southern Shan 1	5.0d-g	44.3bcd	101.0a-d	15.5h	22.5g	10.4efg
Southern Shan 2	6.3abc	46.7bcd	107.0abc	18.9fg	29.4fg	11.4def
Southern Shan 3	4.3fg	48.7abc	104.7a-d	28.2a	49.0cde	12.7cde
Southern Shan 4	6.0bcd	49.0abc	118.0a	26.9abc	66.1abc	14.7abc
Southern Shan 5	5.3c-f	45.7bcd	114.3a	26.7abc	67.6ab	16.2a
Northern Shan 1	5.7cde	49.7ab	105.3a-d	27.5ab	60.8a-d	14.7abc
Northern Shan 2	4.7efg	47.0bed	108.3ab	25.1c	46.0def	14.2a-d
Northern Shan 3	4.3fg	46.7bcd	114.7a	25.3bc	46.8def	14.6abc
Northern Shan 4	6.3ab	50.3ab	118.3a	23.0d	44.5def	15.0ab
Northern Shan 5	7.3a	48.0abc	117.7a	25.7bc	55.2bcd	16.1a
HtoNaukAJan	5.0d-g	46.3bcd	110.7a	25.9be	72.9a	16.8a
HtoPhoShc	7.0ab	53.7a	116.0a	27.3abc	71.0ab	15.7ab
HtoKaiLyan	5.3e-f	47.7abc	111.0a	17.3gh	33.5efg	11.6c-f
Morbi	4.3fg	46.0bcd	100.3a-d	25.7bc	54.6bcd	15.1ab
Cha mame	4.0g	41.0d	85.0d	22.1de	24.5g	8.6fg
No.75	5.3c-f	42.7ed	88.7bcd	27.0abc	30.6fg	8.3g
AGS292	6.0bed	42.7cd	86.7dc	20.5ef	22.3g	7.5g
F-Test	**	*	**	**	**	**
CV (%)	12.4	7.2	10.6	4.9	20.4	12.9

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Table 5 Pods per plant, average pod weight per plant (g), and 100 dry seed weight of Myanmar landrace soybean and vegetable soybean grown in dry season of December 2008-March 2009

Cultivars	Pod per plant	Average pod weight per plant (g)	100 dry seed weight (g)
Southern Shan 1	35.8cde	38.9a-d	22.4bcd
Southern Shan 2	35.0cde	30.5b-e	22.2bcd
Southern Shan 3	37.8b-e	34.2a-e	23.1bc
Southern Shan 4	62.2ab	28.7b-e	12.4g
Southern Shan 5	60.3abc	39.2a-d	12.7fg
Northern Shan 1	60.6abc	44.2abc	15.8efg
Northern Shan 2	56.9a-d	40.4a-d	15.5efg
Northern Shan 3	48.1bcd	37.4a-e	20.1cde
Northern Shan 4	62.4ab	41.0a-d	14.0fg
Northern Shan 5	58.9abc	35.0a-e	14.0fg
HtoNaukAlan	80.3a	54.5a	15.5efg
HtoPhoShe	62.9ab	44.5ab	17.8def
HtoKaiLyan	31.3de	22.0c-f	21.0bcd
Morbi	54.0bcd	45.0ab	19.9cde
Cha mame	17.5e	11.1f	24.7bc
No.75	16.0e	18.7def	30.7a
AGS292	13.7e	15.5ef	25.6b
F-Test	**	**	**
CV (%)	29.1	32.7	14.2

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Table 6 Total pod yield, total seed yield, biomass yield and harvesting index of Myanmar landrace soybean and vegetable soybean grown in dry season of December 2008-March 2009

Cultivars	Total pot yield (ton/ha)	Total dry seed yield(ton/ha)	Biomass Yield (ton/ha)	Harvesting Index (%)
Southern Shan 1	2.0a-d	1.8b-f	6.6b-f	26.8abc
Southern Shan 2	1.5b-e	2.1a-d	6.9b-f	33.5abc
Southern Shan 3	1.9a-d	2.9a	7.6b-e	40.1a
Southern Shan 4	1.4b-e	1.3c-g	5.2def	24.7abc
Southern Shan 5	2.1a-d	2.6ab	8.8a-d	26.2abc
Northern Shan 1	2.4ab	1.9a-e	6.8b-f	32.1abc
Northern Shan 2	2.2abc	1.9a-e	6.3b-f	32.0abc
Northern Shan 3	1.8a-e	1.2d-g	7.1b-e	18.5bc
Northern Shan 4	2.4ab	1.7b-g	11.4a	13.8c
Northern Shan 5	1.9a-e	2.2abc	9.7ab	23.6abc
HtoNaukAlan	2.9a	1.8b-e	9.5abc	20.1bc
HtoPhoShe	2.4ab	1.6a-e	5.9c-f	34.7ab
HtoKaiLyan	1.1cde	1.1d-g	4.9ef	22.8abc
Morbi	2.5ab	1.8b-e	6.4b-f	28.2abc
Cha mame	0.6e	0.7fg	4.4ef	17.3bc
No.75	0.9de	0.9efg	4.3ef	21.2abc
AGS292	0.9de	0.6g	3.2f	18.3bc
F-Test	**	**	**	*
CV (%)	35.1	32.8	28.3	39.5

ns= non significant, ** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Table 7 Contrast of agricultural characters of Myanmar landrace soybean with vegetable soybeans

Characters	Landrace varieties	Vegetable soybean	F-Test	CV (%)
Days to emergence	6.0	5.1	ns	12.4
Days to 1 st flower	47.8	42.1	**	7.2
Days to harvest	110.5	86.8	**	10.6
Plant height at 1 st flower	24.2	23.2	*	4.9
Plant height at harvest (cm)	51.4	25.8	**	20.4
Node per plant	14.2	8.2	**	12.9
Pod per plant	53.3	15.7	**	29.1
Average pod weight (g/plant)	38.3	13.2	**	32.7
Total pod yield (ton/ha)	2.0	0.8	**	35.1
Total dry seed yield (ton/ha)	1.9	0.7	**	32.8
100 dry seed weight (g)	17.6	27.0	**	14.2
Biomass yield (ton/ha)	7.4	4.0	**	28.3
Harvesting index (%)	27.0	18.9	*	39.5

ns = non significant;

*, ** = Significant at 5% and 1% level of probability, respectively.

Table 8 Contrast of agricultural characters of Myanmar landrace soybean of Northern Shan and Southern Shan cultivars with Morbi

Characters	Landrace varieties		F-Test	CV (%)
	Northern Shan and Southern Shan	Morbi		
Days to emergence	5.6	4.3	**	12.4
Days to 1 st flower	48.0	46.0	ns	7.2
Days to harvest	111.3	100.3	*	4.9
Plant height at 1 st flower	24.1	25.7	ns	10.6
Plant height at harvest (cm)	51.2	54.6	ns	20.4
Node per plant	14.2	15.1	ns	12.9
Pod per plant	53.3	54.0	ns	29.1
Average pod weight (g/plant)	37.7	45.0	ns	32.7
Total pod yield (ton/ha)	2.0	2.5	ns	35.1
Total dry seed yield (ton/ha)	1.9	1.8	ns	32.8
100 dry seed weight (g)	17.4	19.9	ns	14.2
Biomass yield (ton/ha)	7.4	6.5	ns	28.3
Harvesting Index (%)	26.9	28.2	*	39.5

ns = non significant;

*,** = Significant at 5% and 1% level of probability, respectively.

Table 9 Contrast of agricultural characters of Myanmar landrace soybean of Northern Shan cultivar with Southern Shan

Characters	Landrace varieties		F-Test	CV (%)
	Northern Shan	Southern Shan		
Days to emergence	5.4	5.7	ns	12.4
Days to 1 st flower	46.9	48.7	ns	7.2
Days to harvest	109.0	112.8	**	4.9
Plant height at 1 st flower	23.2	24.6	ns	10.6
Plant height at harvest (cm)	46.9	53.9	*	20.4
Node per plant	13.1	14.8	**	12.9
Pod per plant	46.2	57.7	*	29.1
Average pod weight (g/plant)	34.3	39.9	ns	32.7
Total pod yield (ton/ha)	2.1	2.1	ns	35.1
100 dry seed weight (g)	18.6	16.7	*	14.2
Total dry seed yield (ton/ha)	1.8	1.7	ns	32.8
Biomass yield (ton/ha)	7.0	7.7	ns	28.3
Harvesting Index (%)	30.3	24.7	*	39.5

ns = non significant; *,** = Significant at 5% and 1% level of probability, respectively.

Table 10 Correlation between agricultural characters of soybean grown in dry season of 2008 December – March 2009

	L1	L2	L3	L4	L5	L6	L7	L8	L9	L10	L11	L12	L13
L1	1.000												
L2	0.434**	1.000											
L3	0.407**	0.683**	1.000										
L4	-0.018	0.300	0.190	1.000									
L5	0.253	0.595**	0.599**	0.663**	1.000								
L6	0.288	0.627**	0.726**	0.455**	0.893	1.000							
L7	-0.275	0.279*	0.517**	0.643**	0.356**	0.787**	1.000						
L8	0.094	0.313*	0.306*	0.195	0.519**	0.581**	0.769**	1.000					
L9	0.098	0.341*	0.299*	0.218	0.524**	0.588**	0.776**	0.980**	1.000				
L10	-0.321*	-0.329	-0.659	-0.248*	-0.677**	-0.807**	-0.683	-0.322*	-0.329*	1.000			
L11	0.038	0.240	0.151	0.245	0.287	0.335*	0.438**	0.595	0.570**	-0.203	1.000		
L12	0.233	0.309	0.443	0.197	0.409	0.564**	0.616**	0.532**	0.547**	-0.422**	0.508**	1.000	
L13	-0.131	0.128	-0.106	0.143	0.023	-0.017	0.053	0.233	0.205	0.045	0.662**	-0.248	1.000

** Significant at 1% level of probability.

L1 = Days to emergence, L2 = Days to first flower, L3 = Days to harvest, L4 = Plant height at first flower, L5 = plant height at harvest, L6 = nodes per plant, L7 = pods per plant, L8 = average pod weight per plant (g), L9= Total pod yield ton per ha, L10 = 100 dry seed (g) , L11 = Total seed yield (ton/ha) , L12 = Biomass yield (ton/ha), L13= Harvesting index

4.2 Evaluation of agricultural characters in recombinant inbred lines of vegetable soybean

Days to first flower and days to R_6 stage

Days to first flower were ranged from 27 to 36 days in early rainy season and 25 to 30 days in rainy season respectively. The RILs were falling between the two parents of AGS292 of 27 days in early rainy and 25 days in late rainy while 36 days and 30 days for NS1 in two seasons (Table 11). The parent showed significantly different in this trait but no homogeneity between early and late rainy season.

Planting early and late maturing genotypes in a sequence will enable the farmer to market fresh vegetable soybean over a long duration. There were significant genotypic differences for days to achieve R_6 stage, when the green pods could be harvested (Table 11). The average number of days from planting to R_6 stage ranged from 63-76 days. The two early flowering cultivars, RIL3 and RIL120, also achieved R_6 stage significantly earlier than all other genotypes as AGS292, while RIL1, RIL41 and RIL87 were similar to NS1 in early rainy season, however, 65 – 68 days in late rainy season.

Plant height

Plant height at first flowering and at R_6 stage varied significantly due to variety (Table 12). The tallest plant (63.0 cm) was found in RIL41 which was resembled to parent NS1 (66.1 cm) in early rainy season and the shortest plant was found in RIL3 (39.2 cm) similar to AGS292 of 35.3 cm. In late rainy season, the maximum plant height 44.0 cm was recorded in RIL41 and the minimum plant height 25.2 cm in AGS292. All RILs stand between the parent cultivars. The plant height at R_6 stage was also highly significant on both early and late rainy season and also highly significant on combine analysis which showed homogeneity of variance on both season. The highest plant height among RILs was observed at RIL41 (97.1 cm) and the shortest RIL3 (50.6 cm) in early rainy season while the parents line AGS292 showed the plant height 42.4 cm and NS1 was 114.9 cm. In late rainy season, the RIL41 got the highest (69.7 cm) and the shortest RIL120 was 52.2 cm, and AGS292 had 36.9 cm and NS1 was 73.8 cm, respectively. From the result of combine analysis, the plant height ranged from 50.0 cm- 83.5 cm among RILs while the parent cultivars were 39.7 cm of AGS292 and 94.3 cm of NS1 (Table 12).

Table 11 Days to first flower (R_f) and days to harvest (R_h) of vegetable soybean RILs growing in early and late rainy season of 2009.

Variety	Days to first flower (R_f)		Days to harvest (R_h)	
	Early rainy	Late rainy	Early rainy	Late rainy
AGS292	27d	25d	63f	65c
NS1	36a	30a	76a	68a
RIL1	34b	28b	75b	68a
RIL3	29c	25d	65e	65c
RIL41	33b	30a	68d	66b
RIL87	34b	30a	74c	68a
RIL120	28c	27c	65e	65c
F-Test	**	**	**	**
CV (%)	1.8	-	-	-

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Table 12 Plant height at first flower and at R_6 stage of vegetable soybean RILs growing in early and late rainy season of 2009.

Variety	Plant height at first flower (cm)		Plant height at R_6 (cm)		
	Early rainy	Late rainy	Early rainy	Late rainy	Combined
AGS292	35.3c	25.2d	42.4f	36.9d	39.7e
NS1	66.1a	39.9b	114.9a	73.8a	94.3a
RIL1	47.3b	33.9c	82.2c	58.5bc	70.4c
RIL3	39.2c	34.5c	50.6f	49.3cd	50.0d
RIL41	63.0a	44.0a	97.1b	69.7ab	83.5b
RIL87	50.4b	34.2c	77.3cd	61.7abc	69.5c
RIL120	49.4b	38.3b	64.4de	52.2c	58.3d
F-Test	**	**	**	**	**
CV (%)	6.4	4.8	10.4	12.6	11.4

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Number of nodes per plant

Positive correlation between characters showed highly significant of node per plant with plant height which varies from 12.0-17.7 in early rainy then 11.0-15.1 in late rainy season and 11.1-16.4 on combined (Table 13).

Table 13 Number of node per plant of vegetable soybean RILs growing in early and late rainy season of 2009

Variety	Node per plant		
	Early rainy	Late rainy	Combined
AGS292	12.0de	11.0c	11.5d
NS1	17.7a	15.1a	16.4a
RIL1	14.0c	13.4b	13.7c
RIL3	11.3e	10.9c	11.1d
RIL41	13.3cd	13.7ab	13.4c
RIL87	16.0b	14.2ab	15.1b
RIL120	12.0de	11.0c	11.5d
F-Test	**	**	**
CV (%)	5.3	6.5	6.0

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Total biomass yield and total pod yield

The total biomass yield varied from a minimum of 10.1 ton / ha to a maximum of 20.4 ton / ha of mean value in early rainy season among RILs while AGS292 produced 10.4 ton/ha and NS1 produced 13.8 ton/ha. In late rainy, RIL1 produced minimum 13.7 ton / ha and maximum 19.5 ton/ ha among RILs compare to the parent line of AGS292 15.3 ton/ha and 'NS1' 13.5 ton/ha as shown in Table 14.

The total pod yield could be as high as 11.4 ton /ha (RIL87) in early rainy and 11.6 ton /ha of RIL41 in late rainy (Table 15). From the result, graded pod yield (up to 2 seeded pods) was the lowest 5.3 ton/ ha of RIL3 in early rainy and highest 9.4 ton/ha of RIL87 and 5.2 ton/ha of AGS292 and 5.4 ton/ ha of NS1. In late rainy season 4.4 ton/ ha of RIL3 showed the lowest yield and 11.6 ton/ ha of RIL41 showed the highest graded pods while AGS292 was 6.8 ton/ha and NS1 was 7.1 ton / ha could be commercially produced for export quality, respectively. The remaining pods in early and late rainy could be sold as shelled bean in domestic market.

Table 14 Total biomass yield, total pod weight, and standard (graded) pod yield of RILs compare with parent lines.

Characters		Total biomass yield		Total pod yield		Graded pod yield	
		(ton/ha)		(ton/ha)		(ton/ha)	
		Early	Late	Early	Late	Early	Late
		rainy	rainy	rainy	rainy	rainy	rainy
Parents	AGS292	10.4	15.3	7.5	8.3	5.2	6.8
	NS1	13.8	13.5	5.9	8.8	5.4	7.1
RILs	Maximum	20.4	19.5	11.4	11.6	9.4	11.6
	Minimum	10.1	13.5	5.4	4.6	5.3	5.0
CV (%)		9.5	7.1	18.3	12.6	13.1	14.0

Graded pod yield

From the result, RIL41 produced maximum graded pod yield in both season then in combined not very much different from RIL120 which has the highest 58.5% of total fresh green seed yield (Table 15). RIL3 produced fewer pods and but similar number of seeds per pod as AGS292 had higher graded pod yield but the lowest total fresh green seeds yield. RIL1 produced total fresh green seed yield 2.9 ton/ha similar to AGS292, even graded pod yield (7.4 ton/ha) higher than AGS292 (6.0 ton/ha).

Fresh green seed yield

The total fresh green seed yield percent on graded pod yield was the highest 58.6% for RIL120 compare to 58.2% in RIL41 which has higher graded pod yield value than parent cultivar. Except RIL1 which produced the lowest ratio of 39.0% of total fresh green seed yield over graded pod yield, even it has higher graded pod yield than AGS29 and NS1 (Table 15).

Table 15 Total pod yield, graded pod yield and total fresh green seed yield of vegetable soybean grown in early and late rainy season of 2009.

Variety	Total pod yield (ton/ha).			Graded pod yield up to 2 seeded (ton/ha)			Total fresh green seed yield (ton/ha)			Graded pod yield (%)
	Early rainy	Late rainy	Combined	Early rainy	Late rainy	Combined	Early rainy	Late rainy	Combined	
AGS292	7.5b	6.8bc	7.7b	5.2d	6.8d	6.0cd	2.1e	3.6c	2.9b	47.3
NS1	5.9b	7.1b	7.4b	5.4cd	7.1c	6.3bc	2.3c	3.6c	3.0b	47.5
RIL1	7.1b	8.0b	8.3ab	6.9bc	7.9b	7.4b	3.1cd	2.6c	2.9b	40.0
RIL3	5.4b	4.4d	5.0c	5.3cd	4.4d	4.9d	2.5de	2.8c	2.7b	55.5
RIL41	8.1b	11.6a	10.1a	7.7b	11.6a	9.7a	3.8b	7.9a	5.9a	58.2
RIL87	11.4a	5.0cd	8.9ab	9.4a	5.0d	7.2a	3.6bc	2.9c	3.3b	48.4
RIL120	7.6b	10.6a	9.2ab	7.5b	10.6b	9.0a	5.0a	5.2b	5.1a	58.6
F-test	*	**	**	**	**	**	**	**	**	**
CV (%)	22.3	14.1	18.6	13.1	14.0	13.6	10.9	21.0	17.9	9.6

** Significant at 1% and 5 % level of probability, respectively.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Number of fresh green pods per 500 gram

The means number of green pods across seasons ranged from the highest pods per 500g of NS1 was 253.3 and the lowest AGS292 of 145.3 in early rainy and 292.0 for NS1 and 168.3 pods per 500 g of RIL120 in late rainy season. Other RILs of RIL1, RIL3, RIL41 and RIL87 fall between two parents. RIL3 and RIL87 produced significantly greater number of pods than all other genotypes in both seasons. RIL1 and RIL120 produced fewer pods per 500g than most other genotypes (Table 16).

Table 16 Number of pod per 500 g of vegetable soybean RILs grown in early and late rainy season of 2009

Variety	Number of pod per 500 g		
	Early rainy	Late rainy	Combined
AGS292	145.3c	176.7b	161.0d
NS1	253.3a	292.0a	272.7a
RIL1	195.3b	175.0b	185.2c
RIL3	215.0b	280.3a	247.2b
RIL41	207.3b	258.7a	233.3b
RIL87	218.3b	274.0a	246.2b
RIL120	198.3b	168.3b	183.3c
F-Test	**	**	**
CV (%)	7.2	8.1	7.7

** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Pod size

Pod length exerted non significant influence on variety in early rainy but was highly significant in late rainy season range from 5.4 cm-5.7 cm among RILs (Table 17). Numerically highest pod length (6.5 cm) was found in AGS292 and 5.5cm in NS1. For 3 seeded graded pods, the size was significant in length but not in width among RILs and parents on both seasons and highly significant on combined over season (Table18).

Table 17 Graded pod size 2 seed pod of RILs growing in early and late rainy season of 2009

Variety	Graded pod size 2 seeded pod (cm)			
	Early rainy		Late rainy	
	Length	Width	Length	Width
AGS292	5.3	1.7	6.5a	1.6
NS1	5.5	1.4	5.5b	1.4
RIL1	5.7	1.5	5.7b	1.5
RIL3	5.4	1.5	5.4b	1.5
RIL41	6.0	1.5	5.7b	1.5
RIL87	5.5	1.4	5.4b	1.5
RIL120	5.8	1.4	5.6b	1.5
F-Test	*	ns	**	ns
CV (%)	5.8	8.4	5.9	5.1

ns = non significant, ** Significant at 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Table 18 Graded pod size 3 seeded pod of vegetable soybean RILs by combined analysis on both season of early, late rainy season of 2009.

Variety	Graded pod size (3 seeded)					
	Early rainy		Late rainy		Combined	
	Length (cm)	Width (cm)	Length (cm)	Width (cm)	Length (cm)	Width (cm)
AGS292	6.6ab	1.6	6.4ab	1.6	6.4bc	1.6a
NS1	6.5a	1.5	6.0b	1.5	6.3c	1.5b
RIL1	6.6ab	1.5	6.7a	1.5	6.6ab	1.5b
RIL3	6.2ab	1.5	6.6ab	1.5	6.3c	1.5b
RIL41	6.7a	1.5	6.7a	1.5	6.7a	1.5b
RIL87	5.8b	1.5	5.9b	1.5	5.9d	1.5b
RIL120	6.3b	1.5	6.3ab	1.5	6.3c	1.5b
F-Test	*	ns	*	ns	**	**
CV (%)	4.7	4.5	4.6	4.1	4.6	4.3

ns = non significant, *, ** Significant at 5 % and 1% level of probability.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Number of fresh green seed per pod

The number of seeds per pod is one of the important quality characteristics that determine the marketability and profitability of vegetable soybean. In early rainy season, RIL3 and RIL41 had the lowest number of green seeds per pod, whereas RIL120 had significantly more seeds per pod than all other genotypes (Table 19). 'NS1' and RIL87 retained significantly fewer seeds per pod than did AGS292 and RIL1. The RIL3 had significantly greater number of seeds per pod than RIL120 but RIL87 and a similar number of seeds per pod were reported for AGS292, NS1, RIL1, and RIL41 in late rainy season.

100 Fresh-seed weight

The results showed highly significant on 100 fresh seed weight among parents verses RILs in both seasons (Table 19). RIL120 produced the highest 100 seed weight of 66.7 g in early rainy season and 62.9 g in late rainy while RIL1 was the lowest with 47.9 and 40.5 g but all RILs' fresh green seed weight were still higher than parent cultivars of AGS292 of 60.0g in early rainy and 65.5 and NS1 was 37.5 g and 38.2 g.

Table 19 The number of fresh green seeds per pod and 100 Fresh seed weight of vegetable soybean grown in early and late rainy season 2009

Variety	Fresh green seed per pod		100 Fresh weight (g)	
	Early rainy	Late rainy	Early rainy	Late rainy
	season	season	season	season
AGS292	2.4abc	2.3abc	60.0ab	65.5a
NS1	2.3bc	2.3abc	37.5e	38.2d
RIL1	2.4ab	2.3abc	47.9cd	40.5d
RIL3	2.2c	2.4a	51.3bc	47.8c
RIL41	2.2c	2.3bc	54.3bc	56.3b
RIL87	2.3bc	2.2c	38.6de	48.2c
RIL120	2.6a	2.4ab	66.7a	62.9a
F-test	**	*	**	**
CV (%)	5.4	2.7	10.4	3.4

*, ** Significant at 5 % and 1% level of probability, respectively

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

Eating quality test

Tests on taste were conducted involving representatives from 14 persons of the test panel members. Results showed that all lines are well accepted compare with the export variety AGS292. The appearance and the texture were highly significant among variety but no significant for taste and flavor, while the appearance and texture were not significant between different environments, however that of taste was highly significant and as for flavor was significant between early rainy and late rainy season. Among RILs, RIL3 was accepted the highest rank with all characters compare with that of export variety of AGS292.

Combine analysis after verifying homogeneity of variance showed that the environment effect on taste and flavor except the appearance and texture of the cultivars. There are non-significant on appearance and texture over season or season verses variety. Within RILs, there are no significant in taste and flavor but significant and highly significant for appearance and texture (Table 20).

Table 20 Eating quality of vegetable soybean RILs grown in early and late rainy season of 2009

Characters	Appearance ¹⁻⁵	Taste ¹⁻⁵	Flavor ¹⁻⁵	Texture ¹⁻⁵
Varieties				
AGS292	1.4d	2.4	2.3	1.6c
NS-1	2.4bc	2.2	2.3	2.3b
RIL-1	2.3c	2.5	2.3	2.3b
RIL-3	3.1a	2.6	2.6	3.1a
RIL-41	2.5bc	2.2	2.2	2.3b
RIL-87	2.8ab	2.6	2.4	2.4b
RIL-120	2.5bc	2.6	2.4	2.3b
F-test				
Season	ns	**	**	ns
Variety	**	ns	ns	**
Season*Variety	ns	**	*	ns
CV (%)	28.5	32.1	30.7	28.1

¹⁻⁵, 1=Excellent, 2= Good, 3= Moderate, 4= Low, and 5= Bad

ns = non significant

*,** Significant at 5 % and 1% level of probability, respectively.

Within column, means followed by the same letter are not significantly different at the 0.05 probability level based on the DMRT procedure.

The environmental effects

The combined analysis of variance over early rainy season and late rainy season of homogeneity characters indicated that the main effects of season (S), and the G x S interaction were highly significant for the traits of plant height at R₆ stage, total pod yield, graded pod yield, total fresh green seed yield except node per plant, 3 seed graded pod size (Table 21). The result indicated that the effect of season was influence on these traits and the interaction between season and varieties as well. The difference between parents and recombinant inbred lines were also highly significant. The mean value of the plant height at R₆ stage was 75.5 cm in early rainy season and 57.5 cm in late rainy season. Positively correlated between plant height and node per plant was found early rainy season produced more nodes than late rainy in average as shown in (Table 21). The homogeneity on both seasons produced 3 seed graded pod size was not significant. The difference of graded pod yield and total fresh green seed yield were found 7.6, 6.8 and 3.2 ton per ha in early then 8.6, 7.7 and 4.1 ton per ha in rainy season. The interaction between season and genotype (E x G) affected all the characters, except pod size width which had shown non-significant.

Table 21 The environment effects on morphological characters of vegetable soybean RILs which showed homogeneity on both early rainy and late rainy season of 2009

Characters	Environment		F-test
	Early rainy	Late rainy	
Plant height at R ₆ stage(cm)	75.5	57.5	**
Node per plant	13.8	12.8	ns
3 seeded pod size Length (cm)	6.4	6.4	ns
3 seeded pod size Width (cm)	1.5	1.5	ns
Total pod yield (ton/ha)	7.6	8.6	**
Graded pod yield (ton/ha)	6.8	7.7	**
Total Fresh green seed yield (ton/ha)	3.2	4.1	**

ns = non- significant

*, **= Significant at 5% and 1% level respectively.

CHAPTER 5

Discussion

5.1 Identification of Myanmar Landrace soybean by SSR marker

Knowing the genealogy of cultivars was essential for interpretation of the dendrogram, a fact in line with other studies (Abdelnoor *et al.*, 1995; Diwan and Cregan, 1997; Priolli *et al.*, 2002). Abe, *et al.* (2003) observed the genetic diversity among the Asian accession with the results suggest that the Japanese and Chinese soybean populations have different germplasm pools, then all of the accession from Southeast Asian and South / Central Asia countries were included in the Chinese germplasm pool. A cluster analysis with the UPGMA clearly separated the Japanese and Chinese accessions. The cluster analysis assigned the 131 accessions into two groups, A and B. The Chinese accessions were included scattered into two clusters. The accession of Myanmar MYA-1 was clustered in Chinese group A6; the others accessions originated from Southern Shan (Pindaya) MYA-2 and Eastern Shan MYA-4 were clustered into Chinese Group B10, while MYA-3 (Taunggyi) was clustered to Chinese group B11. The results from this experiment confirmed that most of the Myanmar landraces soybean cultivars were descendent from Chinese group.

Li *et al.* (2008) report that from a representative collection of 1,863 landraces, a total of 1,160 SSR alleles at 59 SSR loci were detected including 97 unique and 485 low-frequency alleles, which indicated great richness and uniqueness of genetic variation in this core collection. Seven clusters were inferred by STRUCTURE analysis, which is in good agreement with a neighbor-joining tree. A high proportion (95.1%) of pairs of alleles from different loci was in LD in the complete dataset. The low value of LD within the clusters can be seen as evidence that much of the recombination events in the past have been maintained in soybean, fixed in homozygous self-fertilizing landraces.

High genetic similarity among Brazilian soybean cultivars was also detected by Abdelnoor *et al.* (1995), who used RAPD analysis and obtained a mean GS coefficient of 0.82 with a range of 0.69 to 1.00. The most divergent cultivars were Tropical and UFV-6, whereas the most similar cultivars were Ocepar-9 and Paranagoiana.

Narvel *et al.* (2000) studied the use of molecular markers to facilitate the introgression of plant introducing (PI) germplasm into elite soybean [*Glycine max.* (L.)] cultivars depend on the amount of polymorphism that exists between elite genotypes and PIs. A total of 397 alleles were detected among the 79 genotypes at 74 SSR marker loci. Average marker diversity among the PIs was 0.56 and ranged from 0.0 to 0.84. Average marker diversity among the Elites was 0.50 and ranged from 0.0 to 0.79. Genetic similarity estimate based on simple matching coefficients revealed more genetic diversity among the PIs than Elites.

The present analysis detected a high level of length polymorphism at 20 SSR loci in the Myanmar landrace soybean accessions tested. Analyses of the 20 SSR loci clearly allocated the observed diversity into three major groups. According to Abe, *et al.* (2003) Myanmar landraces soybean cultivar of Northern Shan and Southern Shan are descent from the Chinese group. The relationship of the three regional populations is reflected in a UPGMA dendrogram based on the coefficient of similarity between accessions of the different populations. The genetic similarity estimated was in a range of 0.37 to 0.94 among all accessions, which means that some accessions appear identical, while some accessions appear to be totally different genotypes based on the SSRs (Table 3).

The observed diversity among the accessions is thus sufficient to evaluate their genetic relatedness, although the variation among individuals within accessions was not evaluated in this study. Relatively high genetic distance from Southern Shan with that of vegetable soybean suggest that soybean in these areas could be introduced as genetic resources to enlarge the genetic bases.

For yield component analysis, 10 plants were harvested at maturity R_8 stage from each accession. Yield and yield components obtained from the experiment conducted on dry season of 2008 have been summarized in (Table 4-6). Among the data recorded of landraces from field experiment were also highly significant among Northern Shan, Southern Shan and Morbi.

The result analyzed by contrast showed days to emergence between landraces and vegetable soybean were ranged from 6.0 and 5.1 days. Days to first flower were 47.8 days of landraces and 42.1 days of vegetable soybean of mean value. Days to harvest at R_8 stage were highly significant at 110.5 days and 86.8 days of vegetable soybean (Table 7).

Plant height at first flower was significant at 24.2 cm and 23.2 cm between landraces and vegetable soybean, while plant height at harvest was 51.4 cm and 25.8 cm, respectively (Table 7). Among landraces, plant height at first flower was significant at 23.3 cm of Northern Shan, 24.6 cm of Southern Shan and 25.7 cm of Morbi then plant height at harvest at R_8 stage was highly significant with 54.6 cm, 46.9 cm and 53.9 cm respectively (Table 8 and 9).

For each group of plants, data were recorded according to node position on the main stem. Node 1 was the unifoliate node, being the first node above the cotyledons. (Table 7) showed number of nodes per plant varied from 10.4-16.8 among landraces and 7.5-8.6 in vegetable soybean (Table 4).

The result of pod per plant was ranged from 53.3 and 15.7 of landraces and vegetable soybean. Average pod weight was 54.0 gram per plant of Morbi and 34.3 gram per plant of Northern Shan, 39.9 gram per plant of Southern Shan (Table 8 and 9). Maximum number of pods per plant of (80.3) was recorded in Hto Naut Anlan which differed significantly from the rest of all other accessions. Minimum number of pods was observed in Hto Kai Lyan (31.3) among landraces and 15.7 pods per plant when contrast with vegetable soybean means value (Table 5).

100 dry seed weight of 17.6 g and 27.0 g among Landraces and vegetable soybean results showed highly significant on (Table 7). The highest 100 dry seed weight (30.7 g) was obtained in AGS292 and the lowest Southern Shan 5 (12.7 g). There are also highly significant within landraces of Northern Shan 18.6 g, Southern Shan 16.7 g and 19.9 g of Morbi. The variation in seed yield among the varieties may be attributed to the genetic makeup of the varieties. 100 dry seed weight were 18.6 g Northern Shan, 16.7 g Southern Shan, 19.9 g Morbi and 27.0 g of vegetable soybean. Total biomass yield were 7.0 ton per hectare of Northern Shan, 7.7 ton per hectare of Southern Shan, 6.5 ton/ha of Morbi and 4.0 ton per hectare of vegetable soybean.

Total dry seed yield of 3 days after under natural sun dried were observed 1.9 ton/ha of landraces and 0.8 ton per hectare of vegetable soybean while the biomass yield were 7.4 ton/h and 4.0 ton/ha. The mean biomass at R_8 stage varied between landraces for Northern Shan 7.0 ton/ha and Southern Shan 7.7 ton/ha and Morbi for 6.5 ton/ha on dry weight basis (Table 8).

The N075 which had the lowest biomass yield 3.2 ton/ha and Northern Shan 4 had the highest amount of biomass 11.4 ton/ha.

Harvesting index, which is the ratio of economical yield per total biomass yield in percentage. The data indicate that maximum harvesting index (HI) 40.1 was recorded in Southern Shan and minimum HI value of 13.8 in Northern Shan which was significantly different among Myanmar landraces (Table 6) and 18.9 in average value of vegetable soybean cultivars (Table 7).

Information on yield correlations of coefficient on days to emergence, days to first flower, days to harvest, plant height at first flower, plant height at harvest, number of nodes per plant, pods per plant, average pod weight per plant (g), total pod yield ton per ha, 100 dry seed yield (g), total seed yield (ton/ha), biomass yield (ton/ha), and harvesting index respectively were showed positive and significantly association among all characters except that of 100 dry seed weight which was negatively correlated with all of that characters (Table 10).

5.2 Evaluation of agricultural characters in recombinant inbred lines of vegetable soybean

In the early years, vegetable soybean types from Japan were crossed with grain soybean types adapted to tropics and subtropics. Back crossing was used to recover seed size. Disruptive seasonal selection was used to select for broader adaptation (Shanmugasundaram *et al.*, 1991). According to the research from varietal improvement of vegetable soybean in Taiwan by Shanmugasundaram *et al.*, (2000), AGS292 was recognized as the best selection based on the selection based on the results of the regional yield traits by the Kaohsiung DAIS. This selection was found to be less sensitive to photoperiod and temperature. Therefore, as the first strategy, AGS292 was used as one of the adapted parents to cross with other vegetable type or large vegetable soybeans.

From this experiment, five recombinant inbred lines derived from a cross between commercial vegetable soybean cultivar AGS292 crossed with NS1 were used to determine the relationships between yield and its components.

All results from the concerning morphological characters of the RILs evaluated in this experiment were a close to ideal plant type of vegetable soybean characters reported by (Shanmugasundaram *et al.*, 1991) from the qualification based on the experience gained from the

research on vegetable soybean in tropic (including Taiwan) as; non-lodging with strong stem with good rooting system, delayed flowering >40 days, ten to fourteen nodes, fewer branches, longer R_6 to R_7 period, large narrow leaflet preferred, about 15-20 pods per plant, less sensitive to photoperiod and temperature. pod width ≥ 1.4 cm and pod length 5.0 cm, > 75% 2 to 3 seeded pod, bright green pod and seed coat color, gray pubescence, gray or light brown color, easy to strip pods, pods clearance from soil at least 10cm, resistance to bacteria pustule and downy mildew, tolerant to soybean rust, dry weight of 100 seed more than 30 gram, free from undesirable pigmentation and spots on stem or pods, and preferably lipoxygenase null.

The data collected on days to first flower were ranged from 27 days to 36 days in early rainy season and 25 to 30 days in rainy season. The RILs were falling between the two parents of AGS292 of 27 days in early rainy and 25 days in late rainy while 36 days and 30 days for NS1 in two different seasons (Table 11).

The time of harvesting is a critical factor in determining consumer acceptability and marketability of fresh vegetable soybean (Mbuvi and Litchfield, 1999). The optimum time for harvesting fresh vegetable soybean to combine the best product quality with maximum yield is a function of a dynamic relationship between maturity, yield, and quality parameters. Quality properties such as color, texture, and seed size of vegetable soybeans are a function of development time (Mbuvi and Litchfield, 1995). Since these quality parameters do not peak at the same time, it is necessary to compromise time of harvest of green beans. Shanmugasundaram *et al.* (1991) reported that the optimum time for harvesting green beans was when the pods are still green, immature, and tight with fully developed immature green seeds. This stage coincides with the R_6 stage of soybean development as staged by Fehr *et al.* (1971). Thus, harvest at R_6 stage is very critical for ensuring bean yield and quality.

Planting early and late maturing genotypes in a sequence will enable the farmer to market fresh vegetable soybean over a long duration. There were significant genotypic differences for days to achieve R_6 stage, when the green pods could be harvested (Table 11). The average number of days from planting to R_6 stage ranged from 63 DAP of AGS292 and 76 days of NS1. The two early flowering cultivars, RIL3 and RIL120, also achieved R_6 stage significantly earlier than all other genotypes as AGS292, while RIL1 and RIL87 were similar to NS1, 74 and 75 DAP in early rainy season but 65 – 68 DAP in late rainy season.

Plant height at first flowering and at R_6 stage were one of the important character needed non-lodging with strong stem with good rooting system for an ideal plant type of vegetable soybean.

The result from this experiment varied significantly due to variety (Table 12). The tallest plant (63.0cm) was found in RIL41 which was resembled to NS1 (66.1 cm) in early rainy season and the shortest plant was found in RIL3 (39.2 cm) similar with AGS292 (35.3 cm). In late rainy season, the plant height was highly significant with the maximum (43.97 cm) recorded in RIL41 and the minimum with 33.9 cm of RIL1.

The plant height at R_6 stage was also highly significant on both early and late rainy season and also highly significant on combine analysis which showed homogeneity of variance on both season. The maximum plant height among RILs was observed at RIL41 (97.1 cm) and the minimum RIL3 (50.6 cm) in early rainy season while the parents line AGS292 showed the plant height 42.4 cm and NS1 was 114.9 cm. In late rainy season, the RIL41 got the highest (69.7 cm) and the shortest RIL120 (52.2 cm), and AGS292 (36.9 cm) and NS1 (94.3 m), respectively. From the result of combine analysis, the plant height ranged from (50.0 cm- 83.5 cm) among RILs (Table 12).

According to ideal plant type of vegetable soybean characters reported by (Shanmugasundaram *et al.*, 1991) from the qualification based on the experience gained from the research on vegetable soybean in tropic (including Taiwan), node per plant should be 10 - 14. Positive correlation between characters showed highly significant of node per plant with plant height which varies from 12.0-17.7 in early rainy then 11.0-15.1 in late rainy season and 11.1-15.1 in combine value for two seasons (Table 13).

Yield and Yield Components at the R_6 Stage

Suggested cultural practices for vegetable soybean from AVRDC revealed that, the good marketable yields are 7–10 ton /ha of pods, or 4–7 ton /ha of green beans, or 18–25 ton /ha of whole plants. (Lal, G., S.H. Lai, and S. Shanmugasundaram, online pdf.). From this experiment, the total biomass yield varied from a minimum of 18.1 ton / ha of RIL3 to a maximum of 36.3 ton / ha in early rainy season.

In late rainy, 13.7 ton / ha of RIL1 was the lowest and maximum 19.5 ton/ ha of RIL120 among RILs compare to that of AGS292 15.3 ton/ha and NS1 was 13.5 ton/ha (Table 14).

The total pod yield was 11.4 ton /ha in early rainy and 11.6 ton /ha in late rainy (Table 15). The graded pod yield up to two seeded was the lowest 5.3 ton/ ha of RIL3 in early rainy and maximum 9.4 ton/ha of RIL87. AGS292 produced 5.2 ton/ha and for NS1 5.4 ton/ha. In late rainy season 4.4 ton/ ha of RIL3 has the lowest yield and 10.6 ton/ ha of RIL87 had the highest while AGS292 was 11.6 ton/ha and NS1 was 6.8 ton / ha of pods could be commercially produced for export quality in early rainy and late rainy, respectively were followed the varietal improvement conducted for vegetable soybean at AVRDC science 1991 with the objective of to improve total pod yield ≥ 10 ton/ha, and graded pod yield (> 7 t/ha); 2) to identify and select varieties adapted to tropical and subtropical latitudes; and to improve consumer quality attributes including pod and seed color, appearance, flavor, texture, taste, size of the pod and the number of seeds per pod were presented by Shanmugasundaram (2001).

Graded pod yield (up to 2 seeded pods) determined the export quality of vegetable soybean. According to the fifth strategy to select for larger proportion of two and three seeded pods in varietal improvement of vegetable soybean at AVRDC, described in the previous report by Shanmugasundaram *et al.*, (1991). From the result, RIL41 produced maximum graded pod in both season then in combined not very much different from RIL120 which has the highest 58.5% of total fresh green seed yield. RIL3 produced fewer graded pods and but similar number of seeds per pod as AGS292 which showed higher graded pod yield but the lowest total fresh green seeds yield. RIL1 produced total fresh green seed yield 2.9 ton/ha similar to AGS292, even graded pod yield 7.4 ton/ha higher than AGS292 of 6.0 ton/ha as shown in (Table 15).

Number of fresh green pods per 500 gram

The mean number of green pods across seasons ranged from the highest pods per 500 g of NS1 was 253.3 and the lowest of AGS292 was 145.3 in early rainy and 292.0 for NS1 and 168.3 of RIL120 in late rainy season. Other RILs of RIL1, RIL3, RIL41 and RIL87 fall between two parents. RIL3 and RIL87 produced significantly greater number of pods than all other genotypes in both seasons. RIL1 and RIL120 produced fewer pods than most other genotype (Table 16).

Pod size

Among physical characteristics appearance and size of fresh pod and seeds are important. Pod bright green in color with gray pubescence and approximately > 5.0 cm in length and 1.4 cm in width with two or more bright green seed having light buff or gray hilum are considered important for fetching high prices (Shanmugasundaram *et al.*, 1991).

According to the fourth strategy of varietal improvement on vegetable soybean cultivar (Bravo *et al.* 1980; Frank and Fehr, 1981 and Shanmugasundaram *et al.*, 1991) was used the pod length and pod width of two seeded pods to select for large seed size since there was a good correlation between them. In this experiment, pod length exerted non significant influence on variety in early rainy but was highly significant in late rainy season range from 5.4 cm-5.7 cm among RILs (Table 17).

Numerically highest pod length of 6.5 cm was found in AGS292 and 5.5cm in NS1. For 3 seeded graded pods, the size was significant in length but not in width among RILs and parents and highly significant on combined over season (Table 18).

The number of fresh green seeds per pod is one of the important quality characteristics that determine the marketability and profitability of vegetable soybean. In early rainy season, RIL3 and RIL41 had the lowest number of green seeds per pod, whereas RIL120 had significantly more seeds per pod than all other genotypes. NS1 and RIL87 retained significantly fewer seeds per pod than did 'AGS292' and RIL1. The RIL3 had significantly greater number of seeds per pod than RIL120 but RIL87 and a similar number of seeds per pod were reported for AGS292 NS1, RIL1, and RIL41 in late rainy season (Table 19).

100- Fresh seed weight

The results showed highly significant on 100 fresh seed weight among parent verses RILs in both seasons (Table 19). The maximum 100 seed weight (66.7 g) was obtained in RIL120 and the minimum NS1 (37.5 g) in early rainy while the highest of AGS292 (65.5 g) which was statistically different with NS1 (38.1 g) in late rainy season. The 100 seed weight obtained in RILs was fall between their parents AGS292 and NS1 in both seasons. The variation in seed yield among the varieties may be attributed to the genetic makeup of the varieties.

The result agreed with pods more than two seeds are generally preferred and fetch premium prices in the Asian markets (Shanmugasundaram *et al.*, 1991).

The experiment on cultivation and nutritional constituents of Virginia grown Edamame by Carson (2010), the range of seed weight in 2008 was from 50.1 g to 71.0 g per 100 seeds and 47.5 g to 68.0 g in 2009. The green seed yield at R_6 stage was significantly correlated with number of green pods and seeds. Board *et al.* (1996) reported that seed yield at maturity of late planted soybean was more strongly correlated with number of pods than with seeds per pod or seed size. The fresh green seed yield showed a greater correlation with pod yield than with number of pods and seeds, perhaps, because pod yield is the product of number of pods and seeds per pod.

Although, AGS292 produced fewer pods and fewer seeds per pod, it had higher pod yield because of heavier seeds. On the other hand, RIL41 had a greater number of pods and seeds per pod, but its pod yield was not as high as many other genotypes because of smaller seeds.

Fresh seed yield is an important yield determinant and quality parameter that determines consumer acceptability (Shanmugasundaram *et al.*, 1991; Mbuvi and Litchfield, 1995). Generally, seed quality characteristics achieve their peak levels when the seed size is also at its maximum. The number of seeds per pod is one of the yield determinants of soybean. Shanmugasundaram *et al.* (1991) reported higher seed fresh weights for some vegetable soybean breeding lines in Taiwan. A similar number of seeds per pod were reported for several vegetable soybean genotypes grown in Virginia (Mebratu, *et al.*, 1997) and (Konovsky *et al.*, 1994). The number of seeds per pod and seed weight was generally negatively related as they compete for the same resources. A compensatory mechanism between number of seeds per pod and seed weight may have been operative with smaller, and lighter seeds could retain more seeds per pod than vegetable soybean genotypes which produced heavier seeds.

The total fresh green seed yield percent on graded pod weight was the highest 58.6% for RIL120 compare to 58.2% in RIL41 which has higher graded pod yield value. Except RIL1 which produced the lowest 40.0% of total fresh green seed weight even it has higher graded pod yield than AGS292 and NS1 (Table 19).

The eating quality

Tsou and Hong (1991) reported that the parameter used to evaluate the quality of vegetable soybean include appearance, eating quality and nutrient content. A grading system for appearance has been developed based on pod size, number of seed per pods, pod color and the degree of pest damage. Hence appearance becomes criterion in quality evaluation for vegetable soybean processing. However there are no specifications established for the eating quality and nutrient content in Taiwan. These two qualities often vary with the preference of consumers, and thus the evaluation of these two properties can only provide descriptive information rather than grading parameters. Panel tests have showed that the quality of vegetable soybean consists of its sweetness, flavor, texture and taste. The contributions of each component to overall eating quality are almost the same. Thus evaluation methods for all four parameters are required in order to provide a more complete description on quality of vegetable soybean.

Young (2000) determined the thirty-one green soybean genotypes from maturity groups III to VI harvested between R_6 and R_7 , were frozen, boiled, shelled, and evaluated for color, texture, sweetness, nuttiness, beaniness, oiliness, after taste and overall eating quality by three sensory panels. Results showed highly significant ($p < 0.01$) variability among the genotypes in all sensory parameters and highly significant correlations among several parameters. The green soybeans, when boiled, ranged from lightly green to green, were slightly resistant to chewing, slightly nutty, slightly beany, and not oily, imparted a pleasant aftertaste and had a fairly good overall eating quality. The green soybeans would be potentially acceptable as a vegetable in the frozen state and for use in recipes. When selecting genotypes for production, consideration should be given to the sensory attributes of the genotypes, because there was significant variability among the characteristics of the green soybeans, and several characteristics together enhanced the overall acceptability of the vegetable soybeans.

From this experiment, the appearance and texture were highly significant among varieties but not significantly different on taste and flavor. The effect of the environment was influenced on taste and texture but not on the appearance and texture. For the appearance, RIL1 was highly accepted than both of the parents as well as the taste flavor and texture. All RILs used in this experiment were equal or higher eating quality compared to parent lines AGS292 and NS1 (Table 20).

To further increase the acceptability of green soybeans as a vegetable, geneticists need to consider breeding a vegetable soybean that has as many desirable sensory attributes as possible.

The environment effects

The stability of yield is an important characteristic to be considered when judging the value of a cropping system relative to others. In the context of agricultural research, the analysis of yield stability has been largely confined to multi-environment trials of crop cultivars (Piepho, 1998). The trials on stability and adaptability of cultivars or lines have been more emphasized and investigated by commonly used method for modeling statistical interaction is a simple regression of the cultivar performance on the site index (Yates and Cochran, 1938; Finlay and Wilkinson, 1963; Eberhart and Russel, 1966).

When cultivars are tested in terms of seed yield at the multi-environmental trials, cultivars of a crop are grown under a wide range of conditions and great differences are commonly observed in yield performance over environments. This differential yield response of cultivars from one environment to another is called Genotype x Environment (G x E) interaction (Allard, 1960; Vargas *et al.*, 1998). Karasu *et al.*, (2009) reported that effective identification of superior genotypes is generally complicated by the presence of G x E interactions, whereby cultivar relative yields vary across different environments and their performance relative to each other may not be the same. One cultivar may have the highest yield in some environment and a second cultivar may excel in others. Changes in the relative performance of genotype across different environments determine the importance of an interaction.

The combined analysis of variance indicated that the main effects of season (S), and genotype (G) and the S x G interaction were significant for the traits of plant height at mature, node per plant 3 seeded pod size, total pod weight, graded pod weight, total fresh green seed weight, harvest index and daily production weight.

At AVRDC, three crops can be grown in a year. February, June and September were the three planting seasons. The sixth strategy was to use disruptive seasonal selection for adaptation. Specific season adapted types and two and three season adapted types were selected.

The results from four-year study suggested that the graded pod yield of February planting was the highest followed by September planting (AVRDC 1995, 1996, 1997 and 1998). The July planting was quite variable due to typhoons and heavy rains. One hundred bean weights were similar in February and September planting and smaller in July planting. Sugar content was high in September planting and similar in February and July planting. The best planting date of vegetable soybean is dependent upon temperature and day-length. The optimum temperature range of soybean cultivation is 20–30 °C with short day-length 14 hours or less. However, planting should be avoided at cooler temperatures during winter. AVRDC training suggested that the cultural management practices can make modifications to suit local conditions which are adopted for high yields of good quality vegetable soybean.

A plant's yield is genetically determined but changes in environment and cultural practices can affect the maximum. In soybeans, plant population and planting date can influence the number of beans per pod, the average seed size, and yield (Ray *et al.*, 2008; Penderson and Lauer, 2004).

From this experiment, the planting duration was early rainy season from June 1, 2009- August 15, 2009 and for late rainy season from August 27, 2009 to November 3, 2009. The plant height at maturity stage was higher in early rainy than in late rainy with 75.5 cm and 57.5 cm mean value. Positively correlated with plant height, node per plant was also more in early rainy season (Table 13). Most of the pods left on the soybean plants after harvest were at the bottom of the plant and missed by the reel on the harvester. Smith *et al.* (1961) reported that losses could be expected if conventional soybeans were set lower than 13 to 15cm above the soil.

In this study, cultivar and season were significantly interact with respect to percent marketable pods but did so with average seed size. It is difficult to determine if there was a difference between the numbers of pods with two or three beans because it would not have affected the percent marketable pods. The difference in seed size is most likely a function of relative maturity at harvest.

Total pod weight was highly significant and could be produced more weight in late rainy season 8.6 ton/ha and 7.6 ton/ ha in early rainy by combine analysis. The graded pod weight, was 6.8 ton /ha and 7.7 ton/ha; the fresh green seed weight was 3.2 ton/ha and 4.1 ton/ha (Table 15).

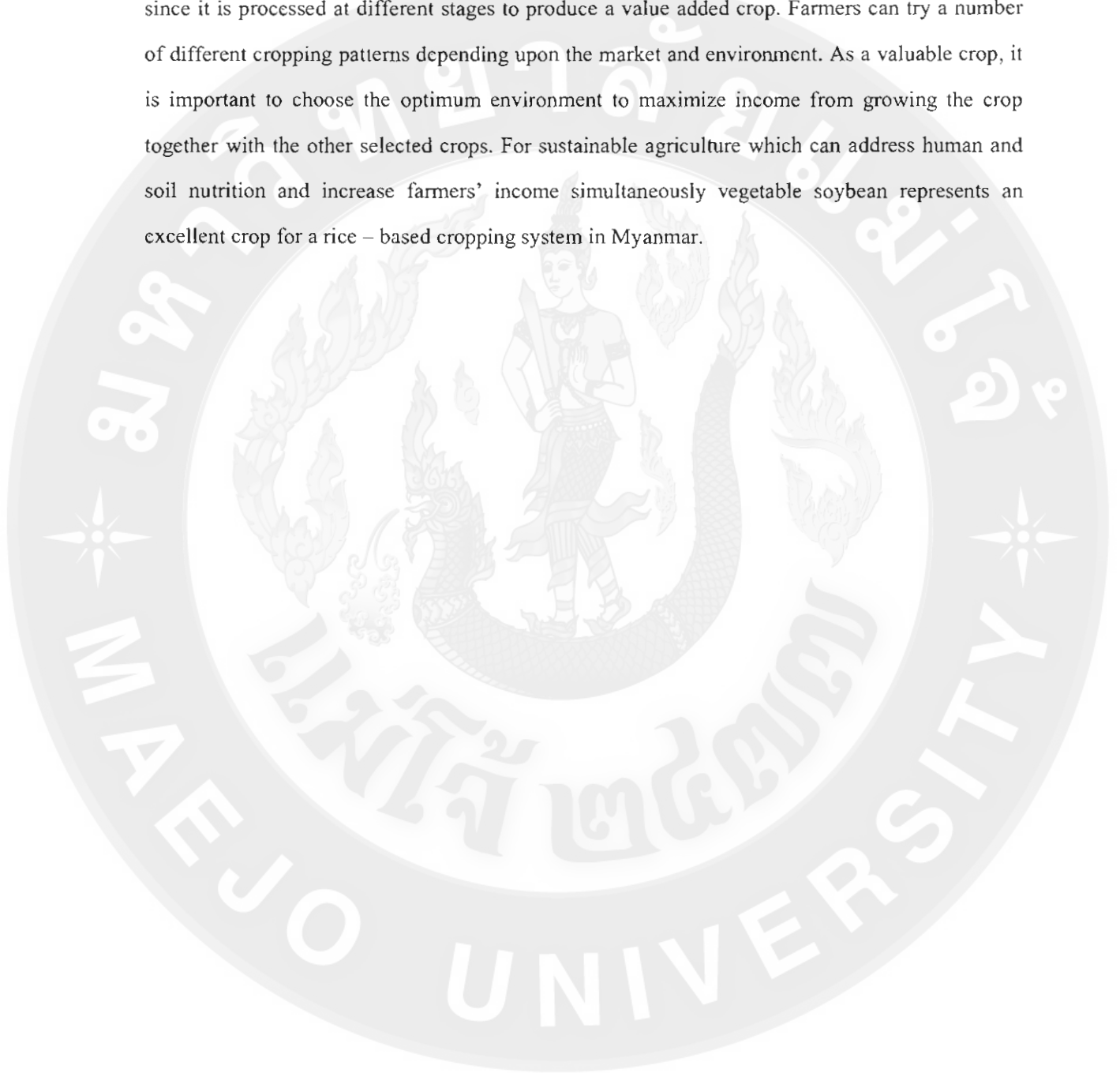
This study has not only helped identify several potential high yielding vegetable soybean cultivars for production to cater to the needs of export or local market, it also provides valuable information that could be used for further improvement of soybean for food uses through classical breeding combined with modern molecular biological approaches as suggested for the improvement of desirable characters of soybean (Boerma and Mian, 1998). In this study, it was possible to investigate and select more adapted and stable in relation to these traits in the absence of genotype and environment interaction.

Future research work is required on vegetable soybean to find answers to several existing issues such as: incorporation of vegetable soybean into the existing cropping systems; identification of suitable production areas, seasons and cropping system for vegetable soybean in different locations in Myanmar; achieving high yield and good quality; improving the total productivity of a given piece of land and the total profit to the farmer diversification of uses and improving quality standards for vegetable soybean products; seed production and storage studies; and crop, disease, insect and weed management.

Landraces are a valuable source of genetic diversity for improvement of a cultivated species. They are rapidly being lost as farmers are provided with improved cultivars developed by plant breeders. It is important that landraces be collected and preserved throughout the world, particularly in areas where modern cultivars are replacing germplasm that has used for crop production for a long time. When a crop is heterogeneous and contains many different genotypes, natural and artificial selection result in the development of thousands of cultivars, commonly referred to as landraces and the plant breeder collect diverse genotypes of cultivated, wild and weedy species that may have potential as breeding material in the future.

Consequently various kinds of landraces have been established as a result of adaptation to different environments and the diversification of food cultures. Vegetable soybean is a new crop for Myanmar and there is still a need to explore acceptance of vegetable soybean, especially in areas where nutritional deficiency is prevalent. Although new crop can surely go a long way towards generating farm family income, improve nutrition and sustain the productivity of farmland as well as offer farmers a wider choice in a cropping system with other crops to enhance productivity and to increase their income. The income from growing vegetable soybean is considerably higher than grain soybean and hence it is an attractive crop to farmers.

Growing vegetable soybean provides employment opportunities to farm families since it is processed at different stages to produce a value added crop. Farmers can try a number of different cropping patterns depending upon the market and environment. As a valuable crop, it is important to choose the optimum environment to maximize income from growing the crop together with the other selected crops. For sustainable agriculture which can address human and soil nutrition and increase farmers' income simultaneously vegetable soybean represents an excellent crop for a rice – based cropping system in Myanmar.



CHAPTER 6

Conclusion

Fourteen Myanmar landrace were identified by using SSR markers could detect 174 alleles with 53 markers of 20 molecular linkage groups (MLGs). Genetic similarity was calculated by Dice's coefficient which showed the estimation symmetric similarity value ranged from 0.37 to 0.94 among all accessions. The dendrogram created by SAHN clustering, the result with the UPGMA distinctly separated into three major groups of Northern Shan, Southern Shan, and vegetable soybean group. All Southern Shan cultivars were cluster in the same group while Northern Shan cultivars of Hto Kyai Lyan and Hto Phoshe and Morbi were clustered into vegetable soybean group. Vegetable soybean cultivar Cha mame has genetic similarity with Northern Shan variety. The relationship of the three regional populations is reflected in a UPGMA dendrogram based on the coefficient of similarity between accessions of the different population. The agricultural characters from the field experiment were significantly different from landraces with vegetable soybean cultivars.

Recombinant inbred lines of vegetable soybean, evaluated in two environments of early and late rainy seasons showed highly significant among varieties verses seasons. From the agricultural data collection form RILs lines, more adaptable and stable in yield and yield components were indicated the presence of linear and non-linear components of genotype $\times$ environment interaction and we can also observed the favorable environments for recombinant inbred lines for superior, stable and graded green pod yield, or responsive in unfavorable environments.

Compare with the parental line of AGS292 and NS1, RILs gave pod and seed yields similar to or better than the adapted cultivar, their possible adaptation to early rainy season verses late rainy season. RIL87 and RIL120 could produce higher graded pod weight and fresh green seed weight than that of export cultivar AGS292. RIL1 had the largest seed on fresh weight basis and this component was probably the key to high pod and seed yields of this cultivar. Genotypes that produced more pods and seeds also produced high pod and seed yields.

Taste panel tests involving fourteen people have proved that the RILs vegetable soybean is acceptable for both cultivation and utilization. Among the cultivars used in this study, RIL3, it showed high acceptable characters compare with parent line and was found to be stable over the environments and therefore; could be used in the breeding programmed for the development of high yielding stable genotypes over environments for future use.

The result from this experiment showed the presence of and the type of GE interactions among the five RILs soybean genotypes and their yield components. High-yielding genotypes with broad adaptation and some genotypes with specific adaptation were identified. Further investigations on GE interactions at important crop growth stages for yield components and biochemical profiles would help to develop strategies that integrate traditional plant breeding with modern molecular marker-based selection for tailoring soybean cultivars for high yield and target environments.

This study has not only helped identify several potential high yielding vegetable soybean cultivars, it also provides valuable information that could be used for further improvement of soybean for food uses through classical breeding combined with modern molecular biological approaches and which also high- lighted to breeders for utilizing genetically diverse introductions in vegetable soybean improvement, and efficient breeding strategies for local adaptation by using the selected diverse parents.

In Summary, this study was significant for soybean researchers to understand the whole feature of Myanmar- landraces soybean as well as can be used as a model method for selecting a collection for multi-species germplasm collection.

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