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Genetic Analysis of Pod Dehiscence in Soybean

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ABSTRACT: Pod dehiscence (PD), defined as the opening of pods along both the dorsal and ventral sutures, causes the seed to shatter in the field before harvesting and results in loss of seed yields. However, breeding for resistance to PD is difficult due to the complicated genetic behavior and environmental interaction. The objective of the present research was to analyze the genetic behavior of PD for improving the breeding efficiency of resistance to PD in soybean. PD after oven-drying the sampled pod at 40°C for 24 hours was the most reliable to predict the degree of PD tested in the field. Keunolkong, a dehiscent parent, was crossed with non-dehiscent parents, Sinpaldalkong and Iksan10. Using their F_1 and F_2 seeds, PD was measured after oven drying the pod at 40°C for 24 hours. The gene conferring PD behaved in different manners depending on the genetic populations. In the Keunolkong × Sinpaldalkong population, PD seemed to be governed by single major recessive gene and minor genes, while several genes were probably involved in the resistance to pod dehiscence in the Keunolkong × Iksan10 population. Heritability for PD estimated in F2 population showed over 90% in the two populations. High heritability of PD indicated that selection for resistance to PD should be effective in a breeding program. In addition, genetic mapping of quantitative locus (QTL) for PD in both populations may reveal that genes conferring PD are population-specific.

Keyword: sybean, pod dehiscence, shattering, heritability

The soybean [*Glycire max* (L.) *Merr.*] is one of the major grain crops providing vegetable oil and protein for human and animal consumptions. Soybean seeds in pods tend to dehisce shortly after maturity if harvesting is delayed. Pod dehiscence (PD), defined as the opening of pods along both the dorsal and ventral sutures, causes the seed to shatter in the field before harvesting, and results in the significant loss of seed yields. Related to that, breeding for resistance to PD is one of the most important objectives to reduce the yield losses, particularly in the tropics and subtropics. A high degree of resistance to PD is also needed if soybean is produced commercially using machine harvesting.

In a field experiment, PD was influenced by environmental factors that affected pod moisture content (Tsuchiya & Sunada, 1977). An earlier study revealed that water content in the pod and the accompanying shrinkage were the major factors to PD (Monsi, 1942). PD was caused directly due to differences in tension developed in the inner sclerenchyma layer as a result of loss of moisture. The innermost cell of the sclerenchyma layer, with a parallel orientation of fibrils along the longitudinal axis of the cells, shortens more during the drying process than upper sclerenchyma cells, which have a transverse orientation of fibrils (Carlson & Lersten, 1987).

The degree of PD in the field is considerably genotype-dependent. Piper & Morse (1923) reported that resistance to pod dehiscence appeared to be dominant to susceptibility. Halvankar & Patil (1994) also identified that PD was governed by single gene and late shattering (or non-shattering) habit was dominant over early shattering. They proposed gene symbol *Sh1* for non-shattering and *sh1* for shattering. Contray to this, susceptibility to PD was more likely dominant over resistance (Nagai, 1926; Ting, 1946; Tiwari & Bhatnagar, 1991). Tsuchiya & Sunada (1978) found that susceptibility to PD had incomplete dominance in F_1 and F_2 generations from crosses between Japanese varieties. Caviness (1963) also found that the inheritance of PD was not simple using several populations from the crosses between *Glycine max* and *Glycine soja* as well as from the crosses between *Glycine max*. PD was highly heritable (above 90% of broad-sense heritability) and the minimum number of genes responsible for PD was estimated to be one or two (Caviness, 1969; Tsuchiya, 1987; Bailey *et al.*, 1997).

The field environment is subject to random seasonal weather conditions, and some field environments may not permit the evaluation of PD. Caviness (1969) observed that weather conditions for one year or two years field experiments were not favorable for evaluating phenotypic traits for

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PD. In addition to field evaluation, several methods were developed to screen soybean genotypes reliably for the degree of PD after drying pods (Tiwari & Bhatnagar, 1991), leaving pods in the greenhouse for a limited time (Helms, 1994), and using mechanical methods (Quick, 1974) or anatomical differences (Bhatia & Tiwari, 1994; Tiwari & Bhatia, 1995).

The objectives of the present research was 1) to develop the most reliable screening method by ove-drying the harvested pods for evaluation of PD in soybean and 2) to analyze the genetic behavior of pod dehiscence in two different soybean populations.

MATERIALS AND METHODS

Plant materials

To analyze genetic behavior of PD, two soybean populations were made from the crosses of Keunolkong × Sinpaldakong and of Keunolkong × Iksan10, respectively. Keunolkong was selected from local cultivars and was chosen as a parent for its susceptibility to PD, which was characterized with early maturity, and large seed size. Sinpaldalkong was selected from a cross between Will × (Elf × SS74185), showing resistance to PD. Iksan10 was derived from a cross of KW552 × Bangsakong, showing resistance to PD with small seed size. For developing the most reliable screening method for PD in soybean, shattering degree in both field and oven dry method were compared for forty seven and one hundred thirty five soybean cultivar and breeding lines respectively in 1997 at Yeongnam Agricultural Research Institute.

Analysis of inheritance

Three parents, F_1 and F_2 populations were grown in a green house sheltered from rainfall at the Asian Vegetable Research and Development Center (AVRDC) from 28 February to late June 2000. Six inch pots filled with mixture of soil and medium texture sand (2 : 3) were autoclaved for sterilization. Two or three seeds were planted per pot, and thinned to two plants per pot after emergence. Plants were watered twice a day. Data were recorded for days to flowering, days to maturity (R8 stage) days from flowering to maturity, pod number per plant, degree of PD after oven-drying (40°C, 24 hours), flower color, pubescence color, and seed size of each plant in F_1, F_2, and each parental line. PD was evaluated on the basis of single plant.

Statistical analyses were made on single plant basis for parents, F_1, and F_2 generations. Broad sense heritability in the F_2 generation was estimated using the following formula, heritability of $X=[(V_{F2}-V_E)/V_{F2}] \times 100$, where V_{F2} is the total phenotypic variance for a specific character in the F_2 generation, and V_E is the environmental variance that was estimated by the arithmetic mean of the variance of the two parents and F_1 populations.

Measurement of pod dehiscence

Field test

To evaluate the degree of pod dehiscence in the field at maturity (R8 stage), ten representative plants were carefully taken from each of three replications and naturally dried for 7 days in the field and the dehiscent pods were detached every 7 days for 4 weeks and the number of non-dehiscent pods was counted.

Oven-dry test

Pods from each plant were carefully sampled at maturity. The sampled pods were kept and allowed to equilibrate for a week at room temperature. The sampled pods were subjected to oven drying at 40°C, 50°C and 60°C for 24 hours. Percentage of pod dehiscence was recorded.

RESULTS AND DISCUSSION

PD is one of major factors to soybean yield. However, due to the absence of efficient method for evaluating PD, soybean breeding program for reducing the PD has been limited. Oven-dry method is known to be simple and rapid for estimating the degree of PD. When the degree of PD is evaluated using oven-dry method, temperature is considered as a major factor. The degree of PD measured by oven dry method using three different temperatures was compared with that tested in the field (Fig. 1). As the temperature increased, PD at field was less correlated with that evaluated at drying oven. The results indicated that the oven-dry method at 40°C for 24 hours was the most reliable to predict the degree of PD tested in the field. This was in a good agreement with the previous result by Tiwari & Bhatnagar (1991), in which two different methods were tested for evaluating PD. They evaluated PD for field-grown plants by drying the harvested pods at maturity at room temperature for a week or by oven-drying at 40°C for 24 hours. It was reported that oven-drying method was highly time- and cost-effective in estimating the field PD.

Therefore, PD was evaluated using oven dry method for further genetic analysis in two segregating RIL populations. Fig. 2 represented the frequency distribution of PD in a cross between Keunolkong (dehiscent) × Sinpaldalkong (non-dehiscent). The PD of F_1 was similar to non-dehiscent parent, and that of F_2 progeny was skewed towards non-dehiscent parent,

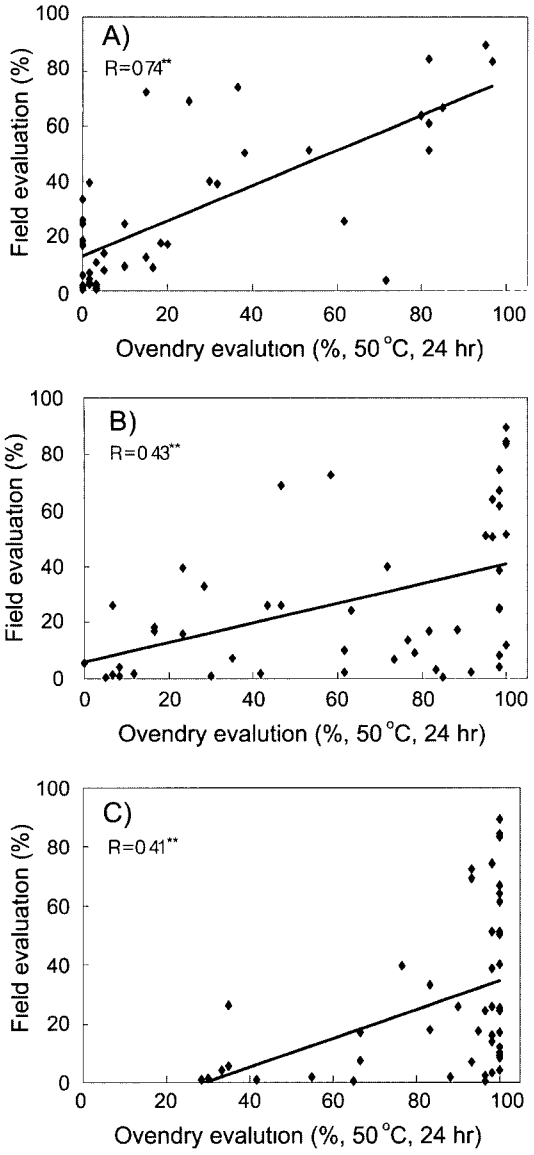

Fig. 1. Relationship of the degree of pod dehiscence between field evaluation and oven-dry evaluation as affected by three different temperatures.

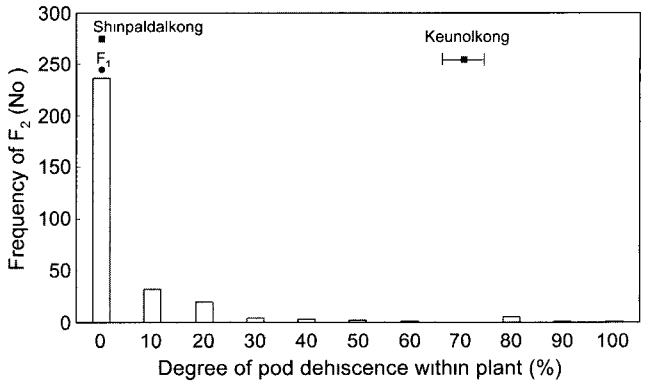

Fig. 2. Frequency distribution of degree of pod dehiscence in P_1, P_2, F_1, and F_2 of a cross between Keunolkong and Sinpaldalkong.

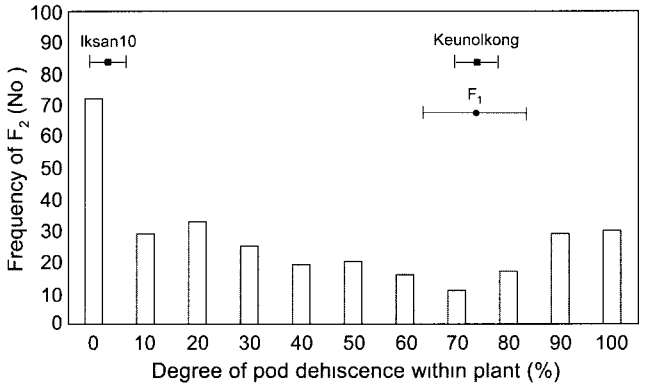

Fig. 3. Frequency distribution of degree of pod dehiscence in P_1, P_2, F_1, and F_2 of a cross between Keunolkong and Iksan10.

Sinpaldalkong. Chi-square test for segregation ratio of F_2 population revealed a goodness of fit to the expected ratio of 1 (dehiscent) : 3 (non-dehiscent), indicating the possible existence of a major recessive gene in the Keunolkong×Sinpaldalkong population (Table 4). Two independent research groups, Halvankar & Patil (1994) and Piper & Morse (1923), reported that a single gene governed PD and dehiscence was recessive over non-dehiscence More recently, Bailey *et al* (1997) identified five putatively independent QTLs conditioning PD in a cross of Young and PI416937. Of five QTLs, a single QTL, linked to RFLP B122_1 on linkage group (LG J) based on the genetic map constructed by the USDA/Iowa State University, accounted for 44% of the phenotypic variation for PD. This indicated that a major QTL on LG J for PD was also observed in the Young and PI 416937 population. However, it can not be concluded that a major gene conferring PD in the Keunolkong × Sinpaldalkong population is the same that was observed by two independent studies (Halvankar & Patil, 1994; Piper & Morse, 1923). Further study using the molecular marker technology may be needed to clarify the gene for PD is the same across populations.

The frequency distribution of PD in a cross between Keunolkong × Iksan10 was clearly different from that between Keunolkong and Sinpaldalkong. PD of F_1 was similar to the dehiscent parent, Keunolkong whereas the F_2 progeny showed a more complex distribution (Fig. 3), implying that PD seems to be incompletely dominant in F_1 but its inheritance might be complex in this cross. Previous studies revealed that susceptibility to PD was dominant to resistant reactions (Nagai, 1926; Ting, 1946; Tiwari & Bhatnagar, 1991). Caviness (1963) found that the inheritance of PD was complex in two different crosses between wild and cultivated species and between varieties of cultivated species. In this

Table 1. Segregation for pod dehiscence in F_2 of soybean population tested against expected ratios (pooled reciprocal).

	Dehiscence	Non-dehiscence	Total	χ^2 (1 : 3)	P
	----------no----------				
Keunolkong×Sinpaldalkong	69	236	305	0.92	0.5~0.1

Table 2. Estimates of broad-sense heritabilities for five agronomic characters in F_2 generation of two soybean crosses.

	Pod dehiscence	Days to flowering	Days to maturity	Days from flowering to maturity	Seed size
	---------- % ----------				
Keunolkong × Sinpaldalkong	94.28	81.08	89.73	86.86	77.16
Keunolkong × Iksan 10	94.99	89.96	88.43	82 90	86.38

$h^2=(V_{F2}-V_E)/V_{F2}$, $Ve=(V_{P1}+V_{P2}+V_{F1})/3$

study using both populations the genes conferring PD might be population-specific. In the Keunolkong × Sinpaldalkong population, a major gene and several minor genes might be responsible for PD whereas several minor genes might be involved in the resistance to PD in the Keunolkong × Iksan10 population. Also, degree of dominance was clearly different across two populations. While F_1 plants showed resistance to PD in the Keunolkong x Sinpaldalkong population, F_1 plants showed susceptibility to PD in the Keunolkong x Iksan10 population. It could be surmised that the genes conferring PD in both populations were controlled in a different manner depending on the genetic populations.

Heritability was calculated to estimate the portion of variability present in F_2. Table 2 represented the heritabilities for PD, days to flowering, days to maturity, days from flowering to maturity and seed size. Heritability of PD was more than 90 % in both crosses. It was greater than those for days to flowering, days to maturity, days from flowering to maturity, and seed size. Heritability estimates for days to flowering, days to maturity, days from flowering to maturity and seed size were more than 80% in both populations except that of seed size in Keunolkong × Sinpaldalkong. Previous studies also indicated that heritability for PD was more than the 90% and the minimum number of genes responsible for PD was estimated to be one or two (Caviness, 1969; Tsuchiya, 1987; Bailey *et al.*, 1997), indicating that PD is highly heritable and selection for resistance to PD should be effective in a breeding program.

Common parent, Keunolkong, was shared to construct both populations in this study. Genetic mapping using molecular marker technologies for both populations will dissect QTLs for PD and will identify the population-specific QTL. In addition, detection of a major QTL as well as development of a molecular marker tightly linked to the QTL may acclerate the marker-assisted selection for enhancing PD resistance in a soybean breeding program.

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REFERENCES

Bailey, M. A., M. A. R. Mian, T. E. Carter Jr., D A. Ashley, and H. R. Boerma. 1997. Pod dehiscence of soybean Identification of quantitative trait loci. J. Heredity 88 : 152-154.

Bhatia, V. S. and S. P Tiwari. 1994. Pod-wall thickness as a selection criterion for shattering resistance in soybean. Soybean Genet. Newsletter 21 : 98-100

Carlson, J. B. and N. R. Lersten. 1987. Reproductive Morphology. p 95-134 In J. R. Wilcox (ed.) Soybeans: Improvement, production, and uses. 2nd ed. Agron. Monogr 16. ASA, CSSA, SSSA, Madison, WI.

Caviness, C. E. 1963. A Physiological and genetic study of shattering in the soybean. Ph. D Dissertion. University of Missouri, Columbia.

Caviness, C. E. 1969. Heritability of pod shattering and its association with some agronomic characters in soybeans. Crop Sci 9 : 207-209.

Halvankar, G. B. and V. P. Patil. 1994. Inheritance and linkage studies in soybean. Indian J. of Genet. & Plant Breed. 54(3) : 216-224.

Helms, T. C. 1994. Greenhouse and field evaluation of pod dehiscence in soybean. Canadian J of Plant Sci 74(4) · 699-701.

Monsi, M. 1942 Untersuchungen uber den Mechanismus der schleuder- bewegung der Sojabohnen-Hulse Jap. J Bot. 12 437-474.

Nagai, I. 1926. On the inheritance of the Soybean. Separate Mag. Noggo and Engei. I : nos. 1-2.

Piper, C. V and W J. Morse. 1923. The soybean. Mcgraw Hill. New York.

Quick, G. R. 1974. A quantitative shatter index for soybeans.

Experimental- Agriculture 10(2) . 149-158.

Ting, C. L. 1946. Genetical studies on the wild and cultivated soybean. J Amer Soc. Agron. 38 : 381-393.

Tiwari, S. P. and P. S Bhatnagar. 1991. Genetics of pod shattering in soybean. Soybean Genetics Newsletter 18 . 150-153.

Tiwari, S. P. and V. S. Bhatia. 1995. Characters of pod anatomy associated with resistance to pod-shattering in soybean. Annals of Botany 76(5) · 483-485

Tsuchiya, T and K. Sunada. 1977. Shattering of pods in soya bean breeding 1. Relations between degree of shattering and moisture content in pods Bull. Of Hokkaido Prefectural Agri. Experi. Stations. No 37 : 17-24.

Tsuchiya, T. and K. Sunada 1978. Breeding studies on pod shattering in soya bean. II. Methods of testing for shattering and varietal differences. Bull. of Hokkaido Prefectural Agri. Experi. Stations Hokkaidoritsu Nogyo Shikenjo Shuho, No. 39 : 19-26.

Tsuchiya, T. 1987. Physiological and genetic analysis of pod shattering in soybeans. JARQ Japan Agricultural Research Quarterly. No 21(3) : 166-175.