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Combining ability analysis and phenotypic correlation of nodule parameters and agronomic traits in peanut (*Arachis hypogaea* L.)

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Abstract

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Choice of parental lines is primarily important for the ultimate success of breeding programs, while correlation among traits is also useful information when simultaneous selection for multiple traits is considered. The first objective of this study was to estimate general combining ability (GCA) and specific combining ability (SCA) for fresh and dry weight of nodules, pod yield, seed yield, 100-seed weight, pod number per plant and shelling percentage in the F_3 generation of 25 peanut crosses derived from 5x5 North Carolina Mating Design II to identify the best parents for these traits. The second objective was to determine correlation among these traits. The experiment was conducted under field condition at Khon Kaen University's Agronomy Farm. The 25 crosses and their parents were arranged in a randomized complete block design with 2 replications. GCA mean squares were significant for nodule fresh weight, pod yield, seed yield, 100-seed weight, pod number per plant and shelling percentage, whereas SCA mean squares were not significant for these traits, indicating the importance of additive gene effects governing the inheritance of these traits. PI 269109 had high GCA effects for nodule fresh weight, 100-seed weight, pod number per plant and

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shelling percentage, and PI 243574 had good combining ability for pod yield and seed yield. KK 60-3 was identified as good general combiner for nodule fresh weight, pod yield, seed yield and 100-seed weight. High and positive phenotypic correlation was found between nodule fresh weight and nodule dry weight. Pod yield, seed yield and 100-seed weight were positively correlated with each other.

Key words : general combining ability, specific combining ability, additive gene effect

บทคัดย่อ

สนับน จอกกลอย วิภาวรรณ ตุลา และ ถวัลย์ เกษมมาดา
ความสามารถในการรวมตัวและสหสัมพันธ์ของลักษณะปม
และลักษณะทางการเกษตรในลั่วลิสง
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จากผลการทดลอง พบว่า ความสามารถในการรวมตัวทั่วไปมีนัยสำคัญในลักษณะน้ำหนักปมสต ผลผลิตฝึกผลผลิตเมล็ด น้ำหนัก 100 เมล็ด จำนวนฝึก/ต้น และเบอร์เซ็นต์กะเทาะ ในขณะที่ความสามารถในการรวมตัวเฉพาะไม่มีนัยสำคัญ แสดงให้เห็นว่าการแสดงออกของยีนเป็นแบบบวกในลักษณะเหล่านี้ พันธุ์ PI 269109 มีค่าความสามารถในการรวมตัวทั่วไปสูงในลักษณะน้ำหนักปมสต น้ำหนัก 100 เมล็ด จำนวนฝึก/ต้น และเบอร์เซ็นต์กะเทาะ และพันธุ์ PI 243574 มีความสามารถในการรวมตัวทั่วไปได้ในลักษณะผลผลิตฝึกและผลผลิตเมล็ด พันธุ์ KK 60-3 เป็นพันธุ์ที่มีค่าความสามารถในการรวมตัวทั่วไปสูง ในลักษณะน้ำหนักปมสต ผลผลิตฝึก ผลผลิตเมล็ด และน้ำหนัก 100 เมล็ด พันธุ์ลำปางมีค่าความสามารถในการรวมตัวทั่วไปสูง ในลักษณะน้ำหนักปมสต น้ำหนัก 100 เมล็ด และเบอร์เซ็นต์กะเทาะ ลักษณะน้ำหนักปมสตและน้ำหนักปมแห้งมีสหสัมพันธ์ทางบวกต่อกันสูง ลักษณะผลผลิตฝึกผลผลิตเมล็ด และน้ำหนัก 100 เมล็ดมีสหสัมพันธ์ทางบวกซึ่งกันและกันสูง

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Peanut (*Arachis hypogaea L.*) is an important crop for small-scale farmers in semi-arid tropic regions, which are characterized by low soil fertility, and unpredictable rainfall and rain distribution. As soil nutrients are limited, peanut yield depends mainly on nitrogen derived from the atmosphere by symbiotic nitrogen fixation. Peanut kernel is an important source of inexpensive protein and oil essential for human diet. Peanut

haulm, when incorporated into soils, provides nitrogen for following crops such as rice, cassava, maize and sugarcane. This helps reduce the use of inorganic nitrogen fertilizer and provides sustainable means for agriculture. Improvement of nitrogen fixing ability in peanut can improve yield as well as soil fertility.

General combining ability (GCA) of the line is the mean performance in all its crosses,

when expressed as deviation from the mean of all crosses. It is the average value of all crosses having this line as one parent, the value being expressed as a deviation from the overall mean of crosses. Any particular cross, then, has an expected value, which is sum of the general combining abilities of its two parental lines. The cross may deviate from this expected value to a greater or lesser extent. This deviation is called specific combining ability (SCA) of the two lines in combination (Falconer and Mackey, 1996). This information provides guidelines for plant breeders to select parent lines to be used in breeding programs and to use promising cross combinations for further selection. Information of relationships among traits is also important when selection for multiple traits is considered.

Early reports on gene action of traits related to nitrogen fixation revealed predominantly non-additive genetic effect. Arrendell *et al.* (1986) reported that specific combining abilities (SCA's) for traits related to nitrogen fixation were significant and accounted for more variability than general combining abilities (GCA's). The parents with high GCA effects for nitrogen fixation were both fastigiate types (Spanish and Valencia), while Virginia-type parents generally had low GCA's. Nigam *et al.* (1985) also found the predominant nature of non-additive genetic variance for nitrogenase activity (measured by acetylene reduction assay) and related traits. For agronomic traits, a number of reports indicated the importance of both additive and non-additive genetic variance. Dwivedi *et al.* (1989) reported that pod and seed traits were controlled largely by additive genetic effect, and pod number/plant and pod weight/plant were controlled by non-additive genetic effect. Both genetic effects were equally important for shelling percentage. Wynne *et al.* (1975) also reported that estimates of both general and specific combining abilities were significant for percent of extra-large kernels, percent of sound mature kernels, kernels/kg, pod length and yield, and estimates of GCA were greater than SCA estimates in magnitude. Jogloy *et al.* (1987) found that general combining ability was highly significant

for pod yield, seed yield, pod length, seed size and shelling percentage, and specific combining ability was significant for pod length and seed size. Swe and Branch (1986) found that estimates of general and specific combining abilities were significant for biomass, harvest index, total pod weight, total pod number, total seed weight, total seed number, seeds per pod, weight per seed, weight per pod and meat content. In general, estimates of specific combining ability are more pronounced in the crosses of more diverse cultivars than in the closely related cultivars (Isleib *et al.*, 1980).

Wynne *et al.* (1980) reported significant and positive correlation among nodulation, plant color and weight, nitrogenase activity and nitrogen content. However, reports on correlation of traits related to nitrogen fixation and yield were inconsistent. Arrendell *et al.* (1985) found that genotypic and phenotypic correlation coefficients of nitrogenase activity with yield were significant with phenotypic correlation coefficients ranged from 0.66 to 0.89 over sampling dates and environments. They also found that correlation between nitrogenase activity and yield was low. Pimratch *et al.* (2004) found that high association between total dry weight and yield was more persistent over a two year experiment than correlation between other traits related to nitrogen fixation and yield, suggesting that environmental influence played an important role for nitrogen fixation in peanut. Evaluation and screening of peanut for high nitrogen fixation should, therefore, be conducted in the target environments.

The objectives of this study were to evaluate general combining ability and specific combining ability for nodule parameters and agronomic traits and to determine phenotypic correlation among these traits. This information can support breeding programs aimed at simultaneously improving high nitrogen fixation and yield in peanut.

Materials and Methods

Plant materials and experimental procedures

Ten peanut genotypes were used as parental lines in a 5x5 North Carolina Mating Design II to

generate F_1 hybrids. These lines were hybridized at Khon Kaen University's Agronomy Farm. The male parents were Virginia Bunch (PI 152133), 42-G-105 (PI 269109), AG-1-1 (Bc 164) (PI 268770), Koyoma Runner (PI 244602) and Local (PI 243574). For convenience, the male parents are hereafter designated as PI 152133, PI 269109, PI 268770, PI 244602 and PI 243574, respectively. The female parents were Tainan 9, Lampang, KK 60-1, KK 60-2 and KK 60-3. They were the local cultivars and the newly released cultivars in Thailand. The 25 F_1 hybrids were self-pollinated to produce F_2 seeds. The F_2 seeds were multiplied for seed increase.

The 25 crosses in the F_3 generation and their parents were evaluated at Khon Kaen University's Agronomy Farm during wet season of 2003. The treatments of 35 entries were arranged in a randomized complete block design with 2 replications under natural infection of *Rhizobium* population. The test site was frequently grown to peanut in previous years, and peanut varieties did not respond to *Rhizobium* inoculation (Toomsan *et al.*, 1988). Fungicide (captan) was used for seed treatment to control crown rot disease caused by *Aspergillus niger*. Plot size was a 2-row plot with 5 m long and spacing of 10 cm between plants within row and 50 cm between rows. Plants were kept at final density of 1 plant per hill. Inorganic fertilizer with 12-24-12 of N-P₂O₅-K₂O at the rate of 25 kg rai⁻¹ (1 rai = 0.16 ha) was applied at 15 days after planting (DAP). Gypsum (CaSO₄) at the rate of 50 kg rai⁻¹ and carbofuran 3% G at the rate of 5 kg rai⁻¹ were applied at 35 DAP. Mechanical weeding was practised at 15 and 35 DAP. Insecticides and fungicides were used as necessary to obtain optimum growth and yield of peanut crop.

Data collection

Nodule parameters and agronomic data were recorded at harvest. Plants at the two ends of each plot were discarded and the remainders were used as sample for data collection. Plants were dug carefully to recover nodules as much as possible and cut at hypocotyls. Pods and roots were separated and each placed in a paper bag. Nodules

were removed from roots and weighed to obtain nodule fresh weight. Then nodules were oven-dried at 70°C for 48 h and weighed to obtain nodule dry weight. Pods were air-dried to obtain approximately 8% moisture and then pod yield, seed yield, 100-seed weight, pod number per plant and shelling percentage were determined. Pod yield and seed yield were converted into yield per rai scale, but the remaining characters were reported on per plant basis.

Data analysis

Analyses of variance were done separately for parental lines and the F_3 crosses according to a randomized complete block design. Mean comparison was done for parent lines using Duncan's multiple range test (Gomez and Gomez, 1984). Mean squares caused by difference among crosses were partitioned into difference due to male parents and female parents, which was attributed to general combining ability (GCA), and difference due to male x female interaction, which was attributed to specific combining ability (SCA). In case of the presence of significant GCA and SCA mean squares, GCA and SCA effects were calculated using the method described by Simmonds (1979). The genetic model is as follows:

$$X = \mu + GCA_p + GCA_Q + SCA_{PQ},$$

Where; X is the true mean of a cross between lines P and Q; μ is the mean of all crosses and GCA and SCA are general and specific combining abilities, respectively. The error term was assumed to be zero. If it is present, it should be associated with GCA and SCA effects. Mean separation of GCA effects was performed using LSD. Simple phenotypic correlation was calculated as described by Gomez and Gomez (1984).

Results and Discussion

Combining ability

The results indicated that differences among parental lines were significant ($P \leq 0.05$) for most traits except for nodule dry weight. Nodule dry

weight was significant at 0.10 probability level. KK 60-3 was the best parent for pod yield, seed yield and 100-seed weight. Tainan 9 was the best parent for shelling percentage and pod number per plant. Lampang was the best parent for shelling percentage and PI 243574 was the best genotype for nodule fresh weight and nodule dry weight (Table 1). Differences among parental lines indicated sufficient diversity for most traits under study, and thus genetic variability would be expected in the progenies in segregating generation from crossing. Although parental lines were statistically different, their progenies were not significantly different for nodule fresh weight and shelling percentage possibly due to high errors as indicated by high C.V. values, when compared with those in their parents.

Differences among crosses were also significant ($P \leq 0.05$) for pod yield, seed yield and 100-seed weight, but not significant for nodule fresh weight, nodule dry weight, pod number per plant and shelling percentage, while that for pod number per plant was significant at 0.10 probability level (Table 2).

Further partitioning of variance of crosses into that due to male and female parents attributed to GCA, and that due to male x female interaction attributed to SCA showed that GCA mean squares of male and female parents were significant ($P \leq 0.05$) for pod yield, seed yield and 100-seed weight. For nodule fresh weight, pod number per plant and shelling percentage, GCA mean squares of either male or female parents were significant at 0.10 probability level. Male x female variances, which were attributed to SCA, were not significant for all traits. GCA/SCA ratios were high for pod yield (10.2:1) and 100-seed weight (61.1:1) (Table 2).

The significance of GCA mean squares indicated the importance of additive gene effect governing the inheritance of nodule fresh weight, pod yield, seed yield and 100-seed weight. Selection of superior genotypes in segregating generation of crosses should be possible for these traits. Lack of significance of SCA mean squares revealed that non-additive gene effect was not important for these traits, and thus selection in early segregating generations should be possible. However, the

Table 1. Mean comparison of 10 peanut genotypes for nodule parameters and agronomic traits

Cultivar	Nodule parameter		Agronomic trait				
	Fresh weight(g)	Dry weight(g)	Pod yield (kg rai ⁻¹)	Seed yield (kg rai ⁻¹)	100-seed weight (g)	Pod number/plant	Shelling percentage
Tainan 9	1.00bcd	0.55bcd	239.3abcd	153.8abc	42.5b	19.0a	64.1ab
Lampang	0.45d	0.28cd	307.8ab	212.5a	43.5b	12.5bc	69.0a
KK 60-1	0.63cd	0.31cd	304.6ab	189.3ab	44.0b	15.0ab	62.2abc
KK 60-2	1.02bcd+	0.61abc	220.2bcd	129.3bcd	40.0b	14.5abc	58.4abc
KK 60-3	0.99bcd	0.53bcd	325.0a	201.8a	76.0a	14.0abc	62.0abc
PI 152133	0.37d	0.22d	162.1d	105.6cd	40.5b	15.5ab	65.1ab
PI 268770	1.75ab	0.76ab	280.3abc	152.2abc	56.0b	14.0abc	55.9bcd
PI 269109	1.64b	0.64abc	311.5ab	177.1ab	56.0b	11.0bc	56.6bcd
PI 244602	1.59bc	0.65abc	192.5cd	101.9cd	37.0b	11.0bc	52.8cd
PI 243574	2.61a	0.93a	157.1d	74.1d	44.0b	9.5c	46.9d
F-ratio	9.0**	3.1 [†]	6.0**	5.2*	3.8*	3.2*	4.3*
CV (%)	27.0	34.8	14.8	19.2	17.7	15.9	7.6

Figures in the same column followed by a common letter are not statistically significant at 0.05 probability level by DMRT

[†], *, ** Significant at 0.10, 0.05 and 0.01 probability levels, respectively

Table 2. Analysis of variance in the F_3 generation from 25 peanut crosses for nodule parameters and agronomic traits.

Sources of variation	df	Nodule parameter		Agronomic trait				
		Fresh weight	Dry weight	Pod yield	Seed yield	100-seed weight	Pod no./ plant	Shelling percentage
Replication	1	0.04	0.11	6358.3*	3043.2*	81.9*	8.8	0.8
Cross	24	0.75	0.08	4353.5**	1581.9*	125.7**	20.8 [†]	46.6
Male (M)	4	0.57 [†]	0.03	5380.2*	1742.7 [†]	60.2*	26.1 [†]	89.7 [†]
Female (F)	4	1.74*	0.17 [†]	13535.6**	3437.3*	648.2**	27.0 [†]	60.7
M x F	16	0.55	0.07	1801.2	1077.9	11.5	17.9	32.3
Error	24	0.52	0.06	1281.8	700.9	20.8	10.5	38.5
GCA/SCA		4:1	3:1	10:1	5:1	62:1	3:1	5:1
CV (%)		58.5	49.0	15.2	18.7	10.2	23.8	10.2

[†], *, ** Significant at 0.10, 0.05 and 0.01 probability levels, respectively

evaluation was conducted in the F_3 generation when dominance contribution, if any, should be substantially reduced after two generations of inbreeding. SCA mean squares should be insignificant in the F_3 generation, except that they were very large. Wynne and Rawlings (1978) and Holbrook (1990) pointed out that, if SCA mean squares were significant in the F_3 or later generations, this might be the result of additive x additive epistatic gene effect that can be fixed in homozygous lines.

GCA effects clearly identified at least one best parent with good combining ability for most traits under study, but failed to identify the best male parent for nodule dry weight (Table 3). Among male parents, PI 269109 had high GCA effects for nodule fresh weight, pod yield and shelling percentage. PI 243574 was the best general combiner for pod yield and seed yield, whereas PI 244602 was the best general combiner for 100-seed weight. Among female parents, Lampang had high and positive GCA effects for nodule fresh weight, 100-seed weight and shelling percentage. KK 60-1 was the best general combiner for 100-seed weight and pod number per plant. KK 60-2 was the best general combiner for nodule dry weight and KK 60-3 had high and positive GCA effects for nodule fresh weight pod yield and seed yield.

For nodule traits, the difficulty to recover nodules from soil might cause large environmental variation, and result in low GCA difference. Genetic contribution of female parents to total variation was larger than that of male parents, possibly due to the fact that all male parents were selected for high nodulation, whereas female parents, which were Spanish and Virginia peanuts, were selected mainly for high yield.

For yield and yield-related traits, Wynne *et al.* (1975) reported that estimates of GCA and SCA were significant for pod weight and pod number, and GCA were greater than SCA. Jogloy *et al.* (1999) found that GCA mean squares were important for pod length and seed size, but SCA mean squares were less important for these traits. Green *et al.* (1983) and Swe and Branch (1986) found that GCA and SCA mean squares were significant for yield and yield component traits. Similar results were also reported. Holbrook (1990) found that GCA and SCA were also significant for yield and yield components. Although SCA remained significant in the F_3 generation, it accounted for a relatively small portion of total variation. In contrast, Isleib *et al.* (1978) found persistent significance of SCA mean squares for yield and seed traits in the F_1 through F_5 generation from the crosses of diverse origins.

Table 3. General combining ability (GCA) effect of 10 parents of peanut for nodule parameters and agronomic traits

Cultivars	Nodule parameter		Agronomic trait				
	Fresh weight	Dry weight	Pod yield	Seed yield	100-seed weight	Pod no. /plant	Shelling percentage
Male parent							
PI 152 133	0.23	-0.07	-0.08	-0.06	-0.46	-1.90	0.39
PI 268 770	-0.15	0.06	-0.06	-0.01	-3.36	0.20	-3.82
PI 269 109	0.29	-0.04	0.06	0.01	2.04	2.30	4.07
PI 244 602	-0.20	-0.01	0.01	-0.01	2.74	0.50	1.15
PI 243 574	-0.17	0.06	0.08	0.06	-0.96	-1.10	-1.79
LSD (0.10)	0.55	0.19	0.09	0.07	3.50	2.49	4.76
LSD (0.05)	0.66	0.23	0.11	0.08	4.20	2.99	5.71
Female parent							
Tainan 9	-0.22	-0.03	0.05	0.02	2.14	-0.20	0.89
Lampang	0.37	-0.11	-0.07	-0.06	4.64	-0.90	1.67
KK 60-1	0.12	0.05	0.07	0.04	5.34	2.90	0.12
KK 60-2	-0.61	0.20	-0.17	-0.06	-14.16	-1.10	-4.26
KK 60-3	0.34	-0.11	0.12	0.06	2.04	-0.70	1.59
LSD (0.10)	0.55	0.19	0.09	0.07	3.50	2.49	4.76
LSD (0.05)	0.66	0.23	0.11	0.08	4.20	2.99	5.71

High and positive GCA effects were desirable in all traits. Therefore, among male parents, PI 269109 was identified as the best general combiner for nodule fresh weight and PI 243574 was the best general combiner for pod yield and seed yield. Among female parents, KK 60-3 had the highest GCA effects for nodule fresh weight, pod yield, seed yield and 100-seed weight.

Phenotypic correlation

Nodule fresh weight was positively correlated with nodule dry weight ($r = 1.00^{**}$). Both nodule fresh weight and nodule dry weight were negatively correlated with shelling percentage, but they were not correlated with pod yield, seed yield, 100-seed weight and pod number per plant. Pod yield, seed yield and 100-seed weight were positively correlated with each other, but not correlated with pod number per plant. Seed yield was also positively correlated with shelling percentage, but pod yield, 100-seed weight and pod number per plant were not correlated with shelling percentage (Table 4).

Significant and positive correlation of fresh weight with nodule dry weight indicated that it is not necessary to evaluate fresh and dry weight of nodules simultaneously. Evaluation of either fresh or dry weight of nodules is sufficient to identify the relative performance of peanut lines for nodule weight. Arrendell *et al.* (1985) found that nodule number and nodule weight were positively correlated with each other and with nitrogenase activity (measured by acetylene reduction assay), and nitrogenase activity and shoot dry weight also positively correlated. Nigam *et al.* (1985) reported that nitrogenase activity was significantly and positively correlated with nodule number, nodule mass, total nitrogen, top dry weight and root dry weight. They also suggested the possibility of breeding for increased nitrogen fixation and yield in peanut. In this experiment, lack of association between nodule parameters and yield components such as pod yield, seed yield and 100-seed weight suggested that nodule traits have more contribution to shoot dry weight than to yield. Unfortunately, shoot dry weight was not recorded in this experi-

Table 4. Phenotypic correlation in the F₃ generation of 25 peanut crosses for nodule parameters and agronomic traits

Nodule dry weight	Agronomic trait				
	Pod yield	Seed yield	100-seed	Pod num ber/plant	Shelling percentage
Nodule fresh weight	1.00**	0.28	-0.14	0.02	0.06
Nodule dry weight		0.03	-0.14	0.20	0.06
Pod yield			0.92**	0.53**	-0.12
Seed yield				0.39**	-0.08
100-seed weight					-0.15
Pod number/plant					0.14

** Significant at 0.01 probability level

ment. For nitrogen-starving soils, many reports suggested that peanut yield depended mainly on nitrogen derived from air. Fixed nitrogen can be drastically reduced when soils were supplied with nitrogen fertilizer. Arrendell *et al.* (1985) also reported low correlation of nitrogenase activity with yield, ranging from 0.36 to 0.57, indicating that simultaneous selection for high nitrogen fixation and high yield might be possible. Negative correlation between nodule parameters and shelling percentage indicated that fixed nitrogen might adversely affect seed/shell ratio.

Pod yield, seed yield, and 100-seed weight positively correlated with each other, but they did not correlate with pod number per plant, indicating that seed size contributed to yield greater than did number of pods. Positive association between seed yield and shelling percentage indicated possibility for simultaneous improvement of these traits. Waranyuwat and Tongsri (1990) reported highly significant associations between pod and seed yield, pod yield and number of mature seeds per plant, and seed yield and number of mature seeds per plant, but shelling percentage had variable correlation with pod and seed traits in different generations. They also suggested selection based on high number of pods per plant for high yield. The difference in populations with different genetic backgrounds and environmental fluctuation might affect association among traits, and thus yield of each peanut genotype may have unique com-

ponents. In our study, selection based on large seed size should increase yield, while that based on high pod number would not.

Conclusion

Additive gene effects were found governing the inheritance of fresh weight and dry weight of nodules, pod yield, seed yield, 100-seed weight, pod number per plant and shelling percentage. Non-additive gene effects were not important for these traits. Selection for these traits through conventional breeding is possible. PI 269109 was identified as the best male parent for nodule fresh weight and PI 243574 was the best male parent for pod yield and seed yield. KK 60-3 was the best female parent for nodule fresh weight, pod yield, seed yield and 100-seed weight.

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