VARIATION IN SYMBIOTIC PERFORMANCE OF BRADYRHIZOBIUM JAPONICUM STRAINS AND SOYBEAN CULTIVARS UNDER FIELD CONDITIONS

C. APPUNU C*, D. SEN, M. K. SINGH & B. DH

Microbial Genetics Laboratory, Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi- 221005, India, Phone: 0542 2307123; Fax: 0542 2368174, e-mail: cappunu@yahoo.com

*Present Address: Scientist, Division of Microbiology, M. S. Swaminathan Research Foundation, Taramani Institutional Area, Third Cross Street, Chennai- 600 113, Tamil Nadu, India. Phone: 044 22541229 (Extn. 303); Fax: 044 22541319, e-mail: cappunu@yahoo.com

Manuscript received: January 21, 2007; Reviewed: March 17, 2008; Accepted for publication: March 25, 2008

ABSTRACT

In this study the symbiotic interactive effect of different Bradyrhizobium japonicum strains with six soybean cultivars were evaluated under field conditions. The rhizobial strains ASR011, USDA123 and CB1809 respectively showed host-cultivar specificity with JS335, Lee and Bragg. B. japonicum ASR011 recorded the highest nodulation and nitrogenase activity with all the studied cultivars. Generally, plants inoculated with strain ASR011 produced higher plant dry matter accumulation and seed yield over all other cultivars. On the basis of analysis of correlation coefficients, it was found that plant dry matter accumulation emerged as best criterion for selection of most effective legume-Rhizobium associations for given physical and biological conditions.

KEY WORDS: B. japonicum, host specificity, soybean, symbiotic effectiveness

INTRODUCTION

Soybean [Glycine max (L.) Merrill.] has played a significant contribution to yellow revolution in India and as a food plant it forms an important part of the routine diet of the people in India [3]. Today, soybean belongs to one of the most important economic crops in the subcontinent. Symbiotic nitrogen fixation (SNF) resulting from mutual beneficial interaction between soybean and soil nodule bacteria (rhizobia) provides a significant boost to N fertilization and additionally, does not cause any hazard to the environment. Thus soybean depends on its symbionts for a large part of its nitrogen requirements for effective growth and dry matter production. Therefore, the success of this crop in the country lies on its efficient symbiosis with N_2-fixing bacteria. Sridhara et al. [15] had reported that N_2 fixation in soybean ranges from 35-56 kg / ha / season in India, as against the estimated 103 to 313 kg / ha / season in Australia [10]. The variable extent of nitrogen fixation by soybean cultivars is probably due to differences in symbiotic effectiveness of rhizobial strains and their compatibility. Selections of host cultivar-compatible inoculant have been recognized as an important method for increasing nitrogen fixation in soybean [6]. It has been reported that the plant dry matter is the best parameter to evaluate symbiotic activity of legume-Rhizobium associations [14]. It was observed that the SNF in legume depends upon co-selection of plant and bacterium genotypes [1, 4, 9]. Therefore, the present study was undertaken to identify soybean cultivars and B. japonicum strains, which are superior in nodulation and plant development under field conditions.

MATERIALS AND METHODS

Four Indian (JS335, PK416, Pusa20 and NRC37) and two American (Bragg and Lee) soybean cultivars were used in this study. Five B. japonicum strains, CB1809 (Australia), USDA123 (USA), and ASR011, ASR031 and ISR076 (India) [1] were used to inoculate the soybean cultivars. Surface sterilized bold healthy seeds were coated by mixing the seeds with gum arabic and B. japonicum strain inoculated peat carrier. The coated soybean seeds were planted in a field experiment conducted at Agriculture farm, Banaras Hindu University, Varanasi during June-October, 2004 and June-October, 2005. The experimental layout was a randomized complete block design with treatments in a split block arrangement. Whole plots were six soybean cultivars and the sub-plots were treatments (five different B. japonicum strains and one control). Three sets of four replicates were used. Each plot was made up of three rows of 5 m long and 0.5 wide with seeds planted 10 cm apart, into each row. At the full-bloom stage, five plants from each plot were uprooted. The nodulation and total dry matter accumulation data were recorded as described previously [2]. At maturity, all remaining plants from the middle row were harvested and the seed yield determined.

Average data recorded over two years was subjected to ANOVA using the statistical analysis systems [11]. Means of all treatments were calculated and the differences tested for significance using the least significant differences (LSD) test at 0.05 probability (P) level. Correlation co-efficient were calculated to study the associative relations among the measurement traits

RESULTS AND DISCUSSION

In the experiment, six soybean cultivars were evaluated for their symbiotic performances with five B. japonicum strains. All strains were shown to induce nodules on the tested cultivars. However, it was observed that host cultivars and inoculation treatment as well as interactions had significant ($P< 0.05$) effect on nodulation, plant growth and seed yield (Table 1). Soybean cultivars differed in the average nodule dry weight per plant induced. The highest nodule dry weight was noted with cultivar NRC37. Comparison of rhizobial strains revealed that the strain CB1809 was most effective in nodule production. Maximum nitrogenase (N_2-ase) activity was expressed in soybean cv. Lee with association of strain USDA123 followed by cultivar JS335 with ASR011. On the basis of N_2-ase activity expressed per plant, the efficacy of strains in decreasing order was ASR011, USDA123, CB1809, ASR031 and ISR076. Analysis of data also revealed that among the six cultivars, JS335, Bragg and Lee recorded higher dry matter accumulation in that order after inoculation with different B. japonicum strains. The lower average dry matter production observed for other cultivars. The mean symbiotic performance of control was significantly lower than those of the plant relying on symbiotically fixed N_2. In soybean, significant differences between rhizobial strains for parameters such as nodule dry weight and N_2-ase, and total plant dry weight have been reported under growth room, green house and phytotron conditions [1, 7]. Maximum seed yield at harvest was recorded in cv. JS335 in association with inoculant strain ASR011. Overall seed yield was found to be highest with ASR011 followed by USDA123 inoculated plants whereas, lowest was with cultivar PK416.

Genetic variation in biomass production and seed yield has already been reported [5, 12]. When B. japonicum strains were compared for symbiotic performance on all studied cultivars, the best performer was the Indian strain

Table 1. Symbiotic performance of *B. japonicum* strains with soybean cultivars under field conditions.

Inoculation treeatments		Cultivars					
		Bragg	Lee	JS335	Pusa20	NRC37	PK416
Seed yield (g plant^{-1})							
Control		14.0	13.23	16.98	12.04	12.86	11.35
USDA 123		15.10	17.21	17.01	12.89	13.57	12.98
CB1809		18.01	13.81	16.11	13.41	13.16	12.49
ASR011		14.50	13.86	21.23	13.02	13.01	12.25
ASR031		14.82	14.14	18.32	13.31	14.21	13.16
ISR076		14.91	14.01	17.15	12.91	12.99	12.63
	Cultivars	1.60					
LSD	Inoculations	1.30					
	Interactions	2.91					
Total dry matter accumulation (g plant^{-1})							
Control		5.32	5.01	4.01	4.52	3.67	3.02
USDA 123		7.32	8.23	6.83	6.52	6.91	6.93
CB1809		8.75	6.02	5.92	7.11	6.83	6.09
ASR011		6.73	7.01	12.23	6.02	5.88	5.62
ASR031		7.13	7.25	9.26	6.35	6.68	6.95
ISR076		6.92	6.98	9.01	6.52	6.71	6.01
	Cultivars	3.67					
LSD	Inoculations	1.56					
	Interactions	5.22					
Nodule nitrogenase activity (μmol C_2H_2 reduced h^{-1} g^{-1} fresh nodule)							
Control		7.31	8.16	4.61	5.23	6.12	3.20
USDA 123		18.36	53.91	17.23	23.16	18.36	16.13
CB1809		48.13	17.19	16.16	17.01	20.13	22.13
ASR011		17.29	19.63	52.36	32.15	26.16	19.63
ASR031		25.21	36.21	46.13	38.31	28.93	22.16
ISR076		22.13	27.83	20.61	25.89	22.10	16.23
	Cultivars	4.21					
LSD	Inoculations	4.72					
	Interactions	9.01					
Nodule dry weight (mg plant^{-1})							
Control		20.78	13.21	32.13	18.36	10.91	15.71
USDA 123		62.31	183.12	67.41	48.93	43.67	43.61
CB1809		191.16	55.61	43.53	70.13	44.51	48.39
ASR011		58.01	50.26	180.91	61.61	49.67	56.18
ASR031		90.09	98.31	130.25	70.35	105.13	61.26
ISR076		66.17	69.18	68.17	49.01	36.81	42.81
	Cultivars	20.22					
LSD	Inoculations	23.15					
	Interactions	43.41					

LSD at 0.05 level of probability

ASR011 (Table 1). The plants inoculated with this strain maintained highest average symbiotic effectiveness with regards to plant dry matter accumulation and seed yield compared with plants inoculated with other strains. This study also confirms the symbiotic specificity of strains ASR011, USDA123 and CB1809 respectively with cultivars JS335, Lee and Bragg under laboratory conditions [1]. Under field conditions, the supporting effectiveness of strains ASR011, USDA123 and CB1809 on percent improvement in total plant dry matter accumulation was slightly lower than that observed under laboratory conditions [1]. This clearly indicates that various climatic and edaphic factors might adversely affect the efficiency of Rhizobium-legume symbiosis when grown under field conditions. Further, native rhizobial strains were symbiotically more effective on

Table 2. Correlation coefficients between symbiotic parameters in soybean–*Bradyrhizobium* interaction under field conditions

Symbiotic parameters	N_2-ase activity[a]	Total plant dry weight (g)	Seed yield (g pl^{-1})
Nodule dry weight (mg pl^{-1})	0.612*	0.692*	0.647*
N_2-ase activity[a]		0.756**	0.692*
Total plant dry weight (g)			0.918**

Data from uninoculated control was included in analysis, (μmol C_2H_2 reduced h^{-1} g^{-1} fresh nodule, * and ** are significant at 5% levels and 1% levels, respectively.

Indian cultivars than strains of foreign origin. Symbiotic compatibility between native rhizobia and soybean cultivars both under laboratory conditions [1, 13] and field conditions [8, 12] has been observed.

Correlation coefficients among symbiotic traits for soybean-Rhizobium system are presented in Table 2. Seed yield correlated positively significantly with total dry matter accumulation, nodule N_2-ase activity and dry weight in decreasing order. Based on our results and similar such reports on symbiotic performances of N_2-fixing plants [1, 4, 5, 12], it can be concluded that plant dry weight is the best and generally accepted criterion for selection of the most effective legume-Rhizobium associations. Additionally, it was found that Indian soybean cultivars could constitute good parental genotypes in breeding programmes for improved N_2-fixation capability.

REFERENCES

[1] Appunu C., Dhar B., Differential symbiotic response of Bradyrhizobium japonicum phage-typed strains with soybean cultivars, J. Microbiol. (2006) 44(3): 363-368.

[2] Appunu C., Sen, D., Dhar B., Acid and aluminium tolerance of Bradyrhizobium isolates from traditional soybean growing areas of India, Indian J. Agri. Sci. (2005) 75(12): 826-827.

[3] Chauhan G. S., Joshi, O. P., Soybean (Glycine max) – the 21st century crop, Indian J. Agri. Sci. (2005) 75(8): 461-469.

[4] Hefny M., Dolinski, R., Malek W., Variation in symbiotic characters of alfalfa cultivars inoculated with Sinorhizobium meliloti strains, Biol. Fertil. Soils. (2001) 33: 435-437.

[5] Hungria M., Bohrer T. R. J., Variability on nodulation and dinitrogen fixation capacity among soybean cultivars. Biol. Fertil. Soils. (2000) 31: 45-52.

[6] Israel D. W., Mathis, J. N., Barbour, W. M., Elkan G. H., Symbiotic effectiveness and host-strain interactions of Rhizobium fredii USDA191 on different soybean cultivars, Appl. Environ. Microbiol. (1986) 51: 898-903.

[7] Kucey R. M. V., Snitwongse, P., Chaiwanakupt, P., Wadisirisuk, P., Siripaipool, C., Arayangkool, T., Boonkerd, N., Rennie, R. J., Nitrogen fixation (^{15}N dilution) with soybean under Thai field conditions. I. Developing protocols for screening Bradyrhizobium japonicum strains, Plant Soil. (1988) 108: 33-41.

[8] Musiyiwa K., Mpepereki, S., Giller, K. E., Symbiotic effectiveness and host ranges of indigenous rhizobia nodulating promiscuous soybean varieties in Zimbabwean soils, Soil Biol. Biochem. (2005) 37: 1169-1176.

[9] Mytton L. R., Skot, L., Breeding for improved symbiotic nitrogen fixation, p. 451-472. In M. D. Hayward, N. O. Bosemark and I Romagosa (eds.), Plant Breeding: Principles and Prospects. Chapman and Hall, London (1993).

[10] Peoples M. B., Herridge, D. F., Ladha, J. K., Biological Nitrogen Fixation: An efficient source of nitrogen for sustainable agricultural production, Plant Soil. (1995) 174: 3-28.

[11] SAS Institute, SAS/STAT user's guide. SAS, Cary, N.C (1988).

[12] Shivananda T. N., Rudraswamy, P., Viswanath, R. S., Viswanath, D. P., Siddaramappa, R., Field evaluation of soybean (Glycine max) genotypes for root nodulation, biomass accumulation and seed yield in different soils, Indian J. Agri. Sci. (2000) 70 (8): 534-535.

[13] Shutsrirung A., Thampibool, S., Bhromsiri, A., Senoo, K., Tajima, S., Hisamatus, M., Symbiotic efficiency and compatibility of native rhizobia in northern Thailand with different soybean cultivars II. Laboratory experiment using native isolates from irrigated soybean-growing areas, Soil Sci. Plant Nutri. (2002) 48: 501-510.

[14] Somasegaran P., Hoben, H. J., Handbook for Rhizobia, Methods in Legume-Rhizobium Technology,

p.1-450. Springer-Verlag, Heidelberg, Germany (1994).

[15] Sridhara S., Thimmegowda, S., Prasad, T. G., Effect of water regimes and moisture stress at different growth stages on nodule dynamics, nitrogenase activity and nitrogen fixation in soybean [Glycine max (L.) Merr.], J. Agron. Crop Sci. (1995) 174: 111-115.