Genetic variability for fresh seed dormancy in Spanish bunch advanced breeding lines of groundnut (*Arachis hypogaea* L.)

NARENDRA KUMAR*, B C AJAY, A L RATHANAKUMAR, T RADHAKRISHNAN, CHUNI LAL, M Y SAMDUR, R K MATHUR, P MANIVEL AND B M CHIKANI

ICAR-Directorate of Groundnut Research, Junagadh-362 001, Gujarat

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ABSTRACT

A study was carried out to identify genotypes possessing fresh seed dormancy in Spanish bunch genetic background. Analysis of variance revealed highly significant differences among the genotypes for fresh seed dormancy at all the stages. Among the three parameters of fresh seed dormancy, only intensity of fresh seed dormancy at weekly interval instead of only at 7 days after sowing was found very important parameter to identify ideal sources of fresh seed dormancy in Spanish types. Two years results revealed that seven genotypes *viz.,* PBS-12190, PBS-12191, PBS-12192, PBS-12171, PBS-12187, PBS-12189 and TPG-41 were the best genotypes which recorded average more than 90 per cent intensity of fresh seed dormancy for 21 days and these also had 100 per cent fresh seed dormancy at 7 DAS except PBS-12191, PBS-12171 and PBS-12189. However three genotypes PBS-12200, PBS-12200B and PBS-12201 also had more than 90 per cent fresh seed dormancy for 14 days. Therefore, these genotypes were identified as new sources of fresh seed dormancy with high intensity and high degree of fresh seed dormancy for over 2-3 weeks. These genotypes could be used as donor parents in breeding programmes to develop high yielding Spanish bunch varieties with 3-4 week fresh seed dormancy in groundnut.

Keywords: Advanced breeding lines, Fresh seed dormancy, Genetic variation, Groundnut, Spanish bunch

Groundnut (*Arachis hypogaea* L.) is an important self-pollinated oilseed crop grown in 117 countries with different agro-climatic conditions between 40°N to 40°S latitudes. It is cultivated globally in 26.2 million ha area with a production of 43.6 million tonnes and productivity of 1666 kg/ha during 2012 to 2014 (triennial average) (FAO, 2017). India is second largest area and production of groundnut after China. In India, it is cultivated in about 4.87 million ha area with the production and productivity of 7.22 million tonnes and 1543 kg/ha respectively during 2014-15 to 2016-17 (Anonymous, 2017). Groundnut is valued as a rich source of oil (48-50%), protein (25-28%), dietary fiber, minerals, vitamins and energy (Mondal and Badigannavar, 2016). Groundnut haulms and cake are important sources of animal feed. Groundnut belonging to Spanish (subsp. *fastigiata* var. vulgaris) and Valencia (subsp. *fastigiata* var. *fastigiata*) types have short life cycle and are generally lack of seed dormancy while Virginia (subsp. *hypogaea* var. *hypogaea*) types have longer life cycle with seed dormancy (Upadhyaya and Nigam, 1999). The physiological basis of dormancy in groundnut is due to hormonal balance between abscisic acid and ethylene, which is produced by the embryo through the action of cytokinin during seed imbibition (Ketring and Morgan, 1971; 1972). Nautiyal *et al.* (1994) has been reported that genetic constitution and different seed parts like seed coat, cotyledons and embryo have a role in imparting dormancy in groundnut. Lack of seed dormancy in the Spanish bunch varieties have a major problem resulting in 20-50 per cent loss in pod yield due to *in-situ* germination resulting from unpredictable rainfall at the time crop maturity (Reddy, 1982; Nagarjun and Radder, 1983). In India, groundnut is cultivated mainly in the *kharif, rabi* and summer seasons. To fit groundnut in these cropping seasons, short duration cultivars are required. Spanish and Valencia type varieties have short duration but have a major problem of *in-situ* germination due to unpredictable rains, prolonged rainy season and irrigation at the time of crop maturity to enable easy harvest and to avoid pod loss. Hence under such conditions at least 2-3 week fresh seed dormancy would be required to avoid yield losses. Therefore, present investigation was to study genetic variability among Spanish bunch advanced breeding lines for fresh seed dormancy and to identify genotypes with 2-3 week seed dormancy.

*Corresponding author E-mail: narendrapb09@gmail.com

MATERIALS AND METHODS

Plant material and field experiment: The experimental material consisted of 27 Spanish advanced breeding lines and three high yielding popular Spanish bunch varieties *viz.,* TG 37A, Dh 86 and TPG 41. These genotypes were harvested at maturity as indicated by blackening of inner parenchyma of the pod (Miller and Burns, 1971). To study fresh seed dormancy, a sample of mature pods were randomly collected and shelled immediately after harvesting from each genotype. Enough care was taken to prevent any damage of the seed testa, cotyledons and embryo while removing seeds from

pods. Before sowing the seeds were treated with carbendazim (3g/kg of seed) to protect from soil-borne diseases. A total of 30 genotypes were evaluated during 2016 and 2017 (June-July) at ICAR-Directorate of Groundnut Research, Junagadh, Gujarat (Latitude 21°31' N, Longitude 70°36' E) in medium black calcareous soil. The experiment was laid out in a randomized complete block design with three replications. Each replication consisted of 20 fresh harvested seeds sown at 2 to 3 cm deep for each genotype. The seeds of each genotype were sown at 45 cm spacing between rows and 10 cm between plants. The soil moisture was maintained at field capacity during the growth period up to 35 days after sowing (DAS). The observations were recorded on number of seeds germinated at weekly interval until the end of experiment.

Estimated parameters: Fresh seed dormancy is characterized by its duration and intensity. These two parameters were studied in the present investigation for all the genotypes for two seasons. The percentage of germinated seeds for entry at a given date was calculated by the following formula:

$$\text{Germination (\%)} = \frac{\text{Number of germinated seeds}}{\text{Total number of sown seeds}} \times 100$$

Duration of fresh seed dormancy was measured by days taken to attain 50 per cent germination by a genotype and intensity of fresh seed dormancy was measured as percentage of non-germinated seed at seven days after sowing. These parameters were estimated using the method suggested by Kumar *et al.* (1991). Degree of dormancy was classified using 1-8 scale according to the scale devised by Landfort *et al.* (1965) where scale 1 = 0-10%, 2=11-20%, 3=21-40%, 4=41-60%, 5=61-70%, 6=71-80%, 7=81-99% and 8=100% non-germinated seeds.

Statistical analysis: Analysis of variance was performed using the statistical package DSAASTAT (Onofri, 2007). The partitioning of means was made with Duncann's multiple range Test at 5% probability level.

RESULTS AND DISCUSSION

Analysis of variance for germination per cent at weekly intervals revealed that highly significant genotypic differences for fresh seed dormancy at all the stages while genotype × year interaction was highly significant at 7 and 21 DAS was highly significant (Table 1). Significant interaction effects indicated that germination per cent varies from one year to other which could be attributed to environmental conditions like temperature, moisture and other non-genetic factors (Toole *et al.*, 1964). It was observed that there was sufficient genetic variability was observed among all the genotypes studied for germination per cent at different weekly intervals during both the year. Germination percentage of genotypes averaged over two seasons is presented in Table 2. At 7th day an average germination per cent ranged from 0 to 64.2 per cent and genotypes *viz.,* PBS-12187, PBS-12190, PBS-12192, PBS-12200 and TPG-41 had no germination during both the years and these genotypes also recorded less than 10 per cent germination up to 21 days after sowing. Hence, these genotypes can be recommended for areas where an unpredictable rain is a common feature.

Germination per cent results revealed that seven genotypes *viz.,* PBS-12190, PBS-12191, PBS-12192, PBS-12171, PBS-12187, PBS-12189 and TPG-41 had average less than 10 per cent germination up to 21 days and also had no germination at 7 DAS except PBS-12191, PBS-12171 and PBS-12189 which had less than 5 per cent germination at 7 DAS. However three genotypes PBS-12200, PBS-12200B and PBS-12201 also exhibited less than 10 per cent germination for 14 days and except PBS 12200 had no germination at 7 DAS. Hence these were the genotypes recommended to be used as donors in hybridization programme. These findings are in agreement with the results of Kumar *et al.* (1991) and Faye *et al.* (2009).

Table 1 Analysis of variance for germination percentage at weekly intervals during 2016 and 2017

Sources of variation	DF	7 DAS	14 DAS	21 DAS	28 DAS	35 DAS
Year	1	19858.2	1190.0	62.5	161.9	747.0
Rep (Year)	4	111.5	177.8	187.9	121.9	199.6
Genotype	29	2981.5**	6082.1**	5880.1**	2871.0**	1857.5**
Genotype × Year	29	785.3**	198.4	342.9**	426.9	434.3
Residual	116	101.3	188.0	152.7	273.2	282.0
Total	179	789.3	1149.9	1111.7	715.0	562.7

*Significance at P< 0.05 level, **Significance at P< 0.01 level

Table 2 Germination percentages of genotypes tested at weekly intervals in the field after harvesting during 2016 and 2017

Genotypes	7 DAS		14 DAS		21 DAS		28 DAS		35 DAS	
	2016	2017	2016	2017	2016	2017	2016	2017	2016	2017
PBS12029B	61.7b-e	31.1a-c	83.3a	71.1a-b	86.7a-b	71.1a-e	93.3a	73.3	95.0a	75.6a-c
PBS-12171	1.7i	0.0h	1.7i	14.4d-f	5.0h	17.8j-k	16.7d	53.3	25.0g	64.4a-c
PBS-12187	0.0i	0.0h	3.3i	0.0f	6.7h	4.4k	18.3d	33.3f-g	35.0e-g	46.7c-d
PBS-12189	1.7i	0.0h	1.7i	2.2e-f	10.0g-h	6.7k	18.3d	40.0e-g	31.7f-g	62.2a-c
PBS-12190	0.0i	0.0h	1.7i	0.0f	6.7h	2.2k	13.3d	13.3g	25.0g	15.6d
PBS-12191	5.0h-i	0.0h	3.3i	6.7e-f	1.7h	6.7k	13.3d	51.1d-f	18.3g	68.9a-c
PBS-12192	0.0i	0.0h	1.7i	2.2e-f	1.7h	2.2k	20.0d	51.1d-f	46.7d-g	66.7a-c
PBS-12199	46.7d-e	31.1a-c	75.0a-b	71.1a-b	86.7a-b	77.8a-d	88.3a	75.6a-d	73.3a-d	77.8a-c
PBS-12200	0.0i	0.0h	10.0i	8.9e-f	18.3g-h	46.7f-h	81.7a-b	73.3a-d	83.3a-c	88.9a
PBS-12200B	11.7g-i	2.2g-h	13.3h-i	6.7e-f	20.0g-h	31.1h-j	50.0b-c	64.4a-f	51.7c-g	68.9a-c
PBS-12201	5.0h-i	0.0h	5.0i	6.7e-f	8.3h	20.0i-k	61.7a-c	66.7a-e	65.0a-e	75.6a-c
PBS-12202	6.7g-i	0.0h	20.0g-i	15.6d-f	45.0e-f	37.8g-j	51.7b-c	60.0b-f	60.0b-f	75.6a-c
PBS-12203	58.3c-f	4.4f-h	66.7a-d	77.8a-b	76.7a-c	82.2a-c	78.3a-b	84.4a-c	75.0a-d	80.0a-c
PBS-12204	15.0g-i	6.7e-h	48.3c-f	37.8c-d	60.0c-e	68.9b-f	75.0a-b	80.0a-d	76.7a-d	77.8a-c
PBS-12204B	11.7g-i	0.0h	38.3f-g	13.3d-f	48.3d-f	46.7f-h	66.7a-c	77.8a-d	68.3a-d	82.2a-b
PBS-12205	24.4g	24.4b-e	48.2c-f	53.3b-c	61.7c-e	73.3a-e	73.6a-b	80.0a-d	75.3a-d	88.9a
PBS-12206	6.7g-i	22.2b-f	36.7f-h	37.8c-d	31.7f-g	71.1a-e	78.3a-b	82.2a-d	85.0a-b	86.7a
PBS-12207	71.7a-c	40.0a-b	81.7a	91.1a	81.7a-c	91.1a-b	75.0a-b	86.7a-b	83.3a-c	88.9a
PBS-12208	88.3a	40.0a-b	88.3a	82.2a	90.0a	82.2a-c	90.0a	80.0a-d	90.0a-b	80.0a-c
PBS-12209	75.0a-c	20.0c-h	81.7a	68.9a-b	90.0a	66.7e-f	93.3a	64.4a-f	91.7a-b	66.7a-c
PBS-12210	50.0d-f	8.3d-h	76.7a-b	70.7a-b	73.3a-c	76.0a-d	70.0a-c	73.7a-d	73.3a-d	78.2a-c
PBS-12211	80.0a-b	26.7b-d	88.3a	88.9a	91.7a	93.3a	91.7a	93.3a	91.7a-b	93.3a
PBS-12212	71.7a-c	6.7e-h	75.0a-b	73.3a-b	91.7a	86.7a-c	93.3a	86.7a-b	93.3a-b	91.1a
PBS-12213	43.3d-f	2.2g-h	65.0a-e	20.0d-f	68.3a-d	42.2g-i	78.3a-b	73.3a-d	80.0a-c	73.3a-c
PBS-12214	83.3a	20.3c-g	73.3a-c	74.9a-b	90.0a	72.5a-e	86.7a	77.3a-d	91.7a-b	77.3a-c
PBS-13003	21.7g-h	4.4f-h	43.3d-g	19.2d-f	65.0b-e	40.8g-i	90.0a	64.7a-f	90.0a-b	71.4a-c
PBS-18035	11.7g-i	2.2g-h	40.0e-g	28.9c-e	45.0e-f	51.1e-h	71.7a-c	77.8a-d	80.0a-c	84.4a-b
TG-37A	81.7a	46.7a	85.0a	73.3a-b	88.3a-b	73.3a-e	86.7a	73.3a-d	88.3a-b	73.3a-c
Dh 86	61.7b-c	26.2b-d	55.0b-f	45.2c	80.0a-c	54.8d-g	80.0a-b	54.8	81.7a-c	76.2a-c
TPG-41	0.0i	0.0h	11.7i	6.7e-f	11.7g-h	8.9k	41.7c-d	37.8e-g	60.0b-f	51.1b-c

The data reveled that an average germination per cent increased from 23 to 42, 51, 66 and 71 per cent at 7, 14, 21, 28 and 35 days after sowing respectively during both the years. Highest percent increase in germination was observed from 7 to 14 DAS (19%) followed by 21 to 28 DAS (15%), 14 to 21 DAS (9%) and 28-35 DAS (5%). It implying that seed of genotypes studied in the experiment were viable and having minimum germination percentage standard criteria (70%) at 35 DAS of groundnut. Hence genotypes identified under study had only fresh seed dormancy. The variability in fresh seed dormancy among Spanish advanced breeding lines in the present study was in agreement with earlier works like Yaw *et al.* (2008), Rathanakumar *et al.* (2009), Faye *et al.* (2009; 2010), Wang *et al.* (2012) and Gaikwad and Bharud (2016).

Duration of fresh seed dormancy: Duration of fresh seed dormancy was measured by days taken to attain 50 per cent germination by a genotype. Genotypes showed different durations of dormancy and it ranged from 7 to >35 days during both the year. Results of durations of fresh seed dormancy showed that two advanced breeding lines PBS-12187 and PBS-12190 had more than 35 days duration of fresh seed dormancy (Table 3). Five advanced breeding lines PBS-12171, PBS-12189, PBS-12191, PBS-12192, PBS-12200B and one cultivar TPG-41 had 28 days duration of fresh seed dormancy. Five advanced breeding lines *viz.*, PBS-12200, PBS-12201, PBS-12202, PBS-12204B and PBS 18035 had 21 days duration of fresh seed dormancy while PBS-12204, PBS-12205, PBS-12206, PBS-12213 and PBS-13003 had 14 days duration of fresh seed dormancy. These genotypes were thus identified as new sources of fresh

seed dormancy of 2-5 weeks in Spanish bunch groundnut genotypes. These results were in agreement with the findings of Kumar *et al.* (1991) and Faye *et al.* (2009). Mathur *et al.* (2000) also observed that two advanced breeding lines PBS-12115 and PBS-12126 possessed fresh seed dormancy of 21-28 and 14-21 days, respectively in groundnut.

Intensity of fresh seed dormancy: Intensity of dormancy is defined as the percentage of seeds that not germinated even seven days after the harvest (Kumar *et al.* 1991). It ranged from 11.7 to 100 per cent and 53.3 to 100 per cent during 2016 and 2017, respectively (Table 3). The results showed that five genotypes *viz.,* PBS-12187, PBS-12190, PBS-12192, PBS-12200 and TPG-41 had 100 per cent intensity followed by eight advanced breeding lines *viz.,* PBS-12171, PBS-12189, PBS-12191, PBS-12201, PBS-12202, PBS-12200B, PBS-12204B, PBS-18035 with an average more than 90 per cent fresh seed dormancy during both the year, while the advanced breeding lines PBS-12204, PBS-12206, PBS-13003 had 80-89 per cent intensity of fresh seed dormancy during both the year at 7 DAS. Intensity of dormancy was found very important at all the weekly intervals instead of only at 7 DAS from practical point of view. In the present investigation we have considered only those genotypes having 90 per cent dormancy for at least 2-3 weeks. It was observed that four genotypes *viz.,* PBS-12187, PBS-12190, PBS-12192 and TPG-41 had 100 per cent fresh seed dormancy at 7 DAS along with more than 90 per cent fresh seed dormancy for 21days and while three genotypes PBS-12191, PBS-12171 PBS-12189 also had more than 95 per cent fresh seed dormancy at 7DAS along with more than 90 per cent dormancy for 21days. Three genotypes PBS-12200, PBS-12200B and PBS-12201 had more than 90 per cent fresh seed dormancy for 14 days and except PBS-12200 (Table 4). This large variation in intensity of dormancy could be due to genotypic differences among the genotypes. These findings are in agreement with the results of Kumar *et al.* (1991) and Faye *et al.* (2009).

Table 3 Duration, intensity and scale of fresh seed dormancy at 7 days after sowing during 2016 and 2017

Genotype	Duration of dormancy (days)		Intensity of dormancy (%)		Dormancy scale	
	2016	2017	2016	2017	2016	2017
PBS-12029B	7	14	38.3	68.9	3	5
PBS-12171	35	28	98.3	100.0	7	8
PBS-12187	35	35	100.0	100.0	8	8
PBS-12189	35	28	98.3	100.0	7	8
PBS-12190	35	35	100.0	100.0	8	8
PBS-12191	35	28	95.0	100.0	7	8
PBS-12192	35	28	100.0	100.0	8	8
PBS-12199	7	14	53.3	68.9	4	5
PBS-12200	21	21	100.0	100.0	8	8
PBS-12200B	28	28	88.3	97.8	7	7
PBS-12201	21	21	95.0	100.0	7	8
PBS-12202	21	21	93.3	100.0	7	8
PBS-12203	7	14	41.7	95.6	4	7
PBS-12204	14	14	85.0	93.3	7	7
PBS-12204B	21	21	88.3	100.0	7	8
PBS-12205	14	14	76.5	75.6	6	6
PBS-12206	21	14	93.3	77.8	7	6
PBS-12207	7	7	28.3	60.0	3	4
PBS-12208	7	7	11.7	60.0	2	4
PBS-12209	7	7	25.0	80.0	3	6
PBS-12210	7	14	50.0	92.7	4	7
PBS-12211	7	7	20.0	73.3	2	6
PBS-12212	7	7	28.3	93.3	3	7
PBS-12213	14	21	56.7	97.8	4	7
PBS-12214	7	14	16.7	79.5	2	6
PBS-13003	14	21	78.3	94.7	6	7
PBS-18035	21	21	88.3	97.8	7	7
TG-37A	7	7	18.3	53.3	2	4
Dh 86	7	14	38.3	73.8	3	6
TPG-41	28	28	100.0	100.0	8	8

Table 4 Intensity of fresh dormancy among the genotypes at weekly intervals during 2016 and 2017

Intensity of dormancy	Fresh seed dormancy (days)			
	7 DAS	14 DAS	21 DAS	28 DAS
100%	PBS-12187, PBS-12190 PBS-12192, PBS-12200, TPG-41	--	--	--
95-99%	PBS-12171, PBS-12189, PBS-12191, PBS-12201, PBS-12202	PBS-12187, PBS-12189, PBS-12190, PBS-12191, PBS-12192	PBS-12190, PBS-12191, PBS-12192	--
90-94%	PBS-12200B, PBS-12204B, PBS-18035	PBS-12171, PBS-12200, PBS-12200B , PBS-12201, TPG-41	PBS-12171, PBS-12187, PBS-12189, TPG-41	--
80-89%	PBS-12204, PBS-12206, PBS-13003	PBS-12202	PBS-12201	PBS-12190

Degree of fresh seed dormancy: Degree of fresh seed dormancy of genotypes was recorded on a 0 to 8 scales of Landfort *et al.* (1965), wherein scale 0 indicates least dormant and scale 8 indicates most dormant genotype. In the present study it ranged from 2-8 during both the years. Results revealed that five advanced breeding lines PBS-12187, PBS-12190, PBS-12192, PBS-12200 and TPG-41 had an average score 8 while nine advanced breeding lines *viz.*, PBS-12171, PBS-12189, PBS-12191, PBS-12200B, PBS-12201, PBS-12202, PBS-12204, PBS-12204B and PBS-18035 had an average score more than 7 during both the year at 7 DAS. Therefore, these genotypes were identified with high degree of fresh seed dormancy than other genotypes. The present results are in agreement with the observations made by Faye *et al.* (2009).

Present investigation showed significant genetic variation for germination percent at different weekly intervals, duration, intensity and degree of fresh seed dormancy in 30 Spainsh genotypes. While considering all the parameters, only intensity of dormancy was found very important at all the stages (weekly intervals) from practical point of view. In the present investigation we have considered only those genotypes having 90% dormancy for at least 2-3 weeks. It was concluded that seven genotypes *viz.,* PBS-12171, PBS-12187, PBS-12189, PBS-12190, PBS-12191, PBS-12192 and TPG-41 had more than 90 per cent fresh seed dormancy up to 21 days and while three genotypes PBS-12200, PBS-12200B and PBS-12201 had more than 90 per cent fresh seed dormancy for 14 days. Therefore, these genotypes were identified as new sources in groundnut for different duration, intensity and degree of fresh seed dormancy and could be used in breeding programs to develop genotypes with in-built fresh seed dormancy.

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