Standardization of seed vigour tests in seven medicinal plants

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Abstract : A lab experiment was conducted during 2007-2008 at the Seed Research and Technology centre of Acharya N.G. Ranga Agricultural University, Rajendra Nagar, and Hyderabad to standardize the seed vigour tets in seven medicinal plants. Evaluation of various vigour tests showed first count (in BP method) and final count (in BP method) for Dulagandi and Kalmegh; Brick gravel for Dulagandi and Senna; cool germination test for Isabgol and Dulagandi; paper exhaustion test for Dulgandi and Isabgol; speed of germination for Senna and Isabgol; mean germination time for Senna, Isabgol and Dulgandi and filed emergence for Dulgandi and kalmegh; were very effective for obtaining higher germinatin percentage and many number of vigourous seedlings which were superior over methods.

Key Words : Medicinal plants, Vigour tests, Germination percentage

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INTRODUCTION

In medicinal plants, the productivity and expansion for large scale production is restricted due to non-availability of quality seeds. A good quality seed is a pre-requisite to obtain healthy plant. Hence, standardized seed testing procedures are essential for obtaining accurate and reproducible seed testing results and to know the planting value of seed lots offered for sale. The procedures for testing the seeds of medicinal plants viz., Ashwagandha (Withania somnifera), Dulagandi (Mucuna pruriens), Isabgol, (Plantago ovata), Kalmegh (Andrographis paniculata), Podapatri (Gymnema sylvestre), Senna (Cassia angustifolia) and Tulasi (Ocimum sanctum), are not available in the international rules for seed testing (ISTA, 2006) and national rules provided in the seed testing manual. Hence, this study was undertaken to standardize the procedures for seed vigour tests for the seven medicinal plants and to find out the influence of various vigour tests on germination percentage of seven medicinal plants.

MATERIAL AND METHODS

A lab experiment was conducted during 2007-2008 at

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the Seed Research and Technology centre of Acharya N.G. Ranga Agricultural University, Rajendra Nagar, Hyderabad to standardize seed vigour tests and to assess the effect of various vigour tests on seed germinability of seven medicinal plants. In the present investigation, nine stress tests have been employed to evaluate their suitability in seven medicinal plants viz., Ashwagandha (Withania somnifera), Dulagandi (Mucana pruriens), Isabgol (Plantago ovata), Kalmegh (Andrographis paniculata), Podapatri (Gymnema sylvestre), Senna (Cassia angustifolia) and Tulasi (Ocimum sanctum).

First count and final count :

Germination test was conducted following between paper (BP) method and first count was taken on fourteenth day for Ashwagandha, seventh day for Dulagandi,Isabgol and Senna,ninth day for Kalmegh, thirteenth day for Podapatri and fifth day for Tulasi.The final count was taken on that day when there is no increase in germination percentage i.e when the seedlings attain maximum germination percentage *i.e.* on eighteenth day for Ashwagandha, tenth day for Dulagandi and Isabgol, on fourteenth day for Kalmegh, on sixteenth day for Podapatri, on eleventh day for Senna and Tulasi, respectively.

Brick gravel test (Hiltner and Ihssen 1911):

Four hundred seeds in four replications of hundred seeds each were planted on 2 cm thick layer of 2 to 3 cm size porous brick gravel. Again a 2 cm layer of moist brick gravel was laid above the seed and wetted up to 60 per cent water holding capacity. The seedlings that emerged through the layer of brick gravel were counted and are considered to be vigorous.

Cool germination test:

Seeds from each variety were subjected to germination test in four replications of 100 seeds each following between paper method in a seed germinator by maintaining an optimum temperature of 18°C, and germination percentage was calculated based on normal seedlings on the day of final count (Fiala, 1981).

Paper exhaustion test:

Two sheets of paper towel (15 x 37 cm) placed on top of each other and rolled, were kept in cylindrical glass containing 30 ml of water. After absorption of water, the roll was taken out and spread on glass plate and unrolled. 50 Seeds were placed on moist paper in straight lines at a spacing of 5 cm below the upper and lower edge of the paper and 3 cm from left and right of the moist paper towel. The rolled paper was placed in a glass container covered with glass plate and placed in dark at 10°C for ten days and the seedlings are evaluated (Agrawal, 2006). The seedlings that had shoot and root exceeding more than two inches and above were considered vigorous and expressed as number of vigorous seedlings.

Speed of germination index:

Germination rate (or) speed of germination can be used as a tool for evaluation of seedling vigor. Speed of germination is one of the oldest concepts of seedling vigor. Seed lots with similar total germination often vary in rate of seedling emergence and rate of growth. Speed of germination estimated by taking daily counts in germination test using top of paper method under controlled conditions of temperature ($25 \pm 1^{\circ}$ C) and relative humidity ($90 \pm 2\%$)in four replications of hundred seeds each. Germination is considered to have occurred when radical has appeared and speed of germination was calculated using the formula (Maguire, 1962)

Speed of germination = $\frac{\text{Number of normal seedlings}}{\text{Days of first count}} + \dots + \frac{\text{Number of normal seedlings}}{\text{Days of final count}}$ $\mathbf{X} = \Sigma\left(\frac{\mathbf{n}}{\mathbf{t}}\right)$ where, N = no. of seeds germinated on day 't' t = time in days.

Mean time of germination :

It was estimated by taking daily counts in germination test using top of paper method under controlled conditions of temperature ($25 \pm 1^{\circ}$ C) and relative humidity ($90 \pm 2\%$) in four replications of hundred seeds each and mean time of germination was calculated using the following formula (Nichols and Heydecker, 1968).

$$\mathbf{t}^{-} = \frac{(\mathbf{t.n.})}{\mathbf{N}}$$

where, t = time in days

n = no. of seeds completing germination on day't'

N = total no. of seeds germinated.

Field emergence (ISTA 2006):

Field emergence potential of seeds was measured by sowing 400 seeds of each sample in four rows each in raised beds of sandy loam soils. The seedling was considered emerged when the plumule was just visible on the soil surface. The number of seedlings emerged were counted on the day of final count and percentage field emergence were calculated using the following formula :

 $Filed emergence (\%) = \frac{Number of seeds germinated}{Total no. of seeds} \times 100$

Tetrazolium(Tz) test :

After pre conditioning the Dulagandi, Podapatri and Senna seeds by soaking them in distilled water for 16 hours on top of moist blotters and then they were cut longitudinally to expose the embryo (ISTA 1985). They were placed in a Petridish having 0.5% aqueous solution of 2, 3, 5 triphenyl tetrazolium chloride and kept in dark at $30 \pm 1^{\circ}$ C for 16 hours. After staining, the excess solution was drained out and seeds were washed thoroughly in distilled water. The stained seeds were examined for determination of viability.

RESULTS AND DISCUSSION

The germination percentages of seven medicinal plants significantly influenced by different vigor tests.Dulagandi followed by Kalmegh recorded better germination than other crop seeds in both first and final counts, which were superior to other crops(Table 1).The germination percentage was examined by using brick gravel method which showed significant variations among the seven medicinal crop plants(Table 1).Dulagandi showed maximum germination percentage (83.3%) when compared to Senna (75.3%), Kalmegh (60.3%), Ashwagandha (39.3%), Tulasi(30.0%), Podapatri (20.0%) and Isabgol(17.6%).The seed germination percentage in cool germination test(Table 1) were better in Senna(73.3%), Isabgol(69.6%) and Dulagandi (68.8%) as compared with Tulasi(50.0%), Ashwagandha (46.0%), Kalmegh (43.6%) and Podapatri (27.2%).

STANDARDIZATION OF SEED VIGOUR TESTS IN SEVEN MEDICINAL PLANTS

Table 1: Germination percentages of seven medicinal crops subjected to various vigour tests									
Medicinal crops	First count (in BP method)	Final count (in BP method)	Brick Gravel test	Cool germination test	Paper exhaustion test	Speed of germination index	Mean time of germination (days)	Field emergence (%)	Tetrazolium test (percentage of viability)
Ashwagandha	28.2	56.7	39.3	46.0	36.0	1.6	13.7	43.0	-
Dulagandi	73.4	85.0	83.3	68.8	82.0	2.6	4.3	77.2	70
Isabgol	66.3	74.0	17.6	69.6	80.0	6.5	3.6	58.0	-
Kalmegh	70.6	78.7	60.3	43.6	43.0	4.3	5.2	62.6	-
Podapatri	32.0	53.7	20.0	27.2	39.0	2.5	5.3	16.5	60
Senna	51.3	76.3	75.3	73.3	64.0	8.6	3.1	72.5	77
Tulasi	30.2	66.0	30.0	50.0	28.0	3.7	4.3	19.0	-
Mean	50.30	70.08	46.56	55.40	53.14	4.26	5.63	49.77	
S.E. <u>+</u>	1.47	1.80	2.44	1.93	1.02	0.10	0.62	0.72	
C.D. (P=0.05)	4.37	5.36	7.52	5.97	3.14	0.33	1.92	2.24	
C.V. (%)	5.99	5.15	9.00	6.05	3.30	4.44	3.46	2.53	

Speed of germination was recorded by using top of paper method (Table 1).Maximum speed of germination was recorded in Senna(8.6) which was significantly more than Isabgol (6.5), Kalmegh (4.3), Tulasi(3.7), Dulagandi (2.6), Podapatri (2.5) and Ashwagandha(1.6). Mean time to complete the total germination was differed in all medicinal crops (Table 1). Ashwagandha showed maximum mean time of germination (13.7 days), followed by Podapatri(5.3 days), Kalmegh (5.2 days), Tulasi(4.3 days), Dulagandi(4.3 days), Isabgol (3.6 days) and Senna (3.1 days). Field emergence exhibited significant variation in all the seven medicinal crops(Table 1).Dulagandi recorded maximum field emergence (77.2%), which was superior to Senna(72.5%), Kalmegh (62.6%), Isabgol(58.0%), Ashwagandha(43.0%), Tulasi (19.0 %) and Podapatri(16.5%). Tetrazolium test was conducted in three medicinal crops namely Dulagandi, Senna and Podapatri. These crops showed 77.0%, 70.0% and 60.0% viable seeds, respectively.

Based on the results obtained it is concluded that Dulagandi recorded higher germination in all vigour tests particularly in first count followed by brick gravel, paper exhaustion test, and field emergence index.Cool germination test was effective in recording higher germination for Senna followed by Isabgol.Vigour tests of speed of germination for Senna and Isabgol, mean germination test for Ashwagandha and Podapatri and field emergence test for Kalmegh were very effective to obtain optimum vigorous seedlings.Irrespective of medicinal crop seeds, first count, field emergence index and mean germination time were effective for obtaining maximum germination and gave reproducible results.

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