



## EFFECT OF DORMANCY BREAKING TREATMENTS ON SEED GERMINATION OF *STYLOSANTHES* SPECIES

R.K. BHATT\*, R.K. TRIPATHI, H.S. TIWARI, D.S. RAJPUT AND AMARESH CHANDRA

Seed Technology Division, Indian Grassland and Fodder Research Institute, Jhansi-284 003, U.P.

Received on 20 Sept., 2007, Revised on 13 March, 2008

### SUMMARY

In different genotypes of *Stylosanthes* species maximum germination was recorded in the seeds scarified through coarse sand paper followed by concentrate  $H_2SO_4$  (for 5 minutes) treatment and minimum germination was observed in the control. The effect of  $GA_3$  (300 ppm),  $KNO_3$  (0.2 %) and hot water (60 °C) treatments were at par with respect to germination. Comparing the genotypic difference maximum germination was recorded in *S. seabrana* 104710 followed by *S. seabrana* 2523, *S. scabra* 93116, *S. hamata* 110123 and *S. seabrana* 105546 B in the scarification treatment and minimum germination was recorded in *S. viscosa*. The total seedling length, seedling dry matter and vigour index were also improved by these dormancy breaking treatments and more significantly enhanced by the scarification treatments. The data revealed that variability exists with respect to the germination and seedling vigour among the genotypes of different *Stylosanthes* species.

**Key words:** Dormancy breaking treatments, germination, *Stylosanthes hamata*, *S. scabra*, *S. seabrana*, vigour index.

### INTRODUCTION

The genus *Stylosanthes* has been recognized for its adaptability, growth and biomass production under different land use systems and thus, has attracted many studies in different countries on its biology and utilization. Seed germination in *Stylosanthes* has been a problem because of physical dormancy. *Stylosanthes* is a small seeded crop having high proportion of hard seed coat which is described as exogenous dormancy and can reduce the establishment of this species resulting in to an uneven establishment. The seeds, which were sown in the month of July showed only 20 to 30% germination in the first year whereas rest of the seeds remained in the dormant stage and germinated during the second year. Seeds of *Stylosanthes* developed different degrees of hardness but when seed moisture fall below (7%) all

seeds showed exogenous dormancy (Burin *et al.* 1987). Cameron (1967) also noted difference in the degree of exogenous dormancy in consecutive years for 25 lines of *Stylosanthes* grown in the same locality and attributed these differences to environmental factors. The seed coat hardness may be because of seed polymorphism due to genetic as well as environmental factors. Several treatments, chemical and or physical have been developed to rupture, remove or dissolve the water impermeable seed coat of legumes or hard and stiff lemma and palea of grasses for getting synchronized germination and also for assessing the true germination percentage in the seeds of commercial importance (Hopkinson and Paton 1993). Therefore, a study was undertaken using different dormancy breaking seed treatments in various genotypes of *Stylosanthes* for enhancing the percent of germination and vigour index.

\*Author for Correspondence, E-mail: researcher\_rkb@yahoo.com

## MATERIALS AND METHODS

The mature seeds of different genotypes of *Stylosanthes* (viz., *S. hamata* 110123, *S. hamata* 110135, *S. hamata* 61670, *S. scabra* 36260, *S. scabra* 93116, *S. scabra* cv. Fitzroy, *S. scabra* cv. Seca, *S. scabra* q 10042, *S. scabra* RRR 94-100, *S. scabra* RRR 94-86, *S. scabra* RRR 94-93, *S. scabra* RRR 94-97, *S. scabra* RRR 94-96, *S. seabrana* 104710, *S. seabrana* 105546 B, *S. seabrana* 110372, *S. seabrana* 2523, *S. seabrana* 2534, *S. seabrana* 2539 and *S. viscosa*) were harvested during November-December, 2003 and stored in polythene bags (700 gauge) at seed storage room at Indian Grassland and Fodder Research Institute, Jhansi, India. These seeds were tested for germination after eight months of storage in the laboratory condition in the seed germinator at 30 °C and 75 % RH. The dormancy breaking seed treatments of GA<sub>3</sub> (300ppm), KNO<sub>3</sub> (0.2%), hot water (60 °C initial temperature and then left for 12 hours), concentrate H<sub>2</sub>SO<sub>4</sub> (for 5 minutes and then washing in running tap water for 15 minutes) and scarification through coarse sand paper were applied. Twenty five seeds of each genotypes were placed in each Petri plate (6 cm. diameter) lined with 2 layers of filter paper moistened with distilled water. Three replications for each treatment were taken in this study. Seed germination was recorded on alternate days upto completion of germination (21 days of experiment). Seedling length and dry weight of seedling were also recorded at the end of germination. The seedling weight was recorded by drying ten seedlings in oven at 80°C for 48 hours. The vigour index was calculated by multiplying percentage germination with seedling length.

## RESULTS AND DISCUSSION

On average of the seed germination percentage of different genotypes under various treatments, maximum germination (79%) was recorded in the seeds scarified through sand paper followed by concentrate H<sub>2</sub>SO<sub>4</sub> treatment (55.45%) over the control (19.65%) (Table 1). The effect of GA<sub>3</sub> (300 ppm), KNO<sub>3</sub> (0.2 %) and hot water (60 °C) treatments were at par with respect to seed germination. Comparing the genotypic difference of different genotypes of *Stylosanthes*, maximum germination was recorded in *S. seabrana* (104710)

(92%) followed by *S. seabrana* 2523, *S. scabra* 93116, *S. hamata* 110123 and *S. seabrana* 105546 B in the scarification treatment. In control maximum seed germination was observed in *S. scabra* (cv. seca) followed by *S. scabra* (RRR 94-100, RRR 94-86), *S. hamata* 110135 and *S. seabrana* 105546 b and minimum in *S. scabra* RRR 94-96, 93116, RRR 94-97 and *S. viscosa*. In some of the genotypes the seed treatments of GA<sub>3</sub> (300 ppm), KNO<sub>3</sub> (0.2 %) and hot water (60 °C) has also promoted germination up to 55 % (*S. scabra* cv. seca). Mechanical scarification was most effective in breaking dormancy of seeds of forage legume *Pachecoa venezuelensis* passing through a blender increasing germination percentage after 28 days from 2% in untreated seeds to 53.5% (Diaz *et al.* 1995).

The germination of seeds has also significantly improved by the treatment of concentrated H<sub>2</sub>SO<sub>4</sub> (for 5 minutes and then washing in tap water for 15 minutes) but it is difficult to be practiced by the common farmers. The result revealed that the germination percentage in various genotypes of *Stylosanthes* can be improved by the scarification of seeds. Nan *et al.* (1998) reported that the most effective treatment for reducing fungal infection without reducing seed viability in *Stylosanthes hamata* and *S. scabra* were immersion in sulphuric acid for 6 minute. In general all the genotypes of *Stylosanthes* species exhibited poor seed germination because of hard seed coat. Seeds from various genotypes of different species of *Stylosanthes* varied in response of dormancy breaking seed treatments on germination percentage. The scarification pretreatments improved the germination significantly in all the genotypes of *Stylosanthes* species. H<sub>2</sub>SO<sub>4</sub> (5 minutes) pre treatments have also improved the germination as compared to other pre treatments (GA<sub>3</sub>, KNO<sub>3</sub>, hot water). Airi *et al.* (2005) has also reported similar effects on germination of *Cornus capitata*. The effect of sulphuric acid treatments may be due to softening of seed coat resulting in to water influx (Hortmann *et al.* 1989) and breaking hard coat seed dormancy in other plant species as reported by various workers (Airi *et al.* 1998, Li *et al.* 1999, Bhatt *et al.* 2000). In *Stylosanthes humilis* the organic acids pretreatments stimulated the germination of scarified dormant seeds (Pelacani *et al.* 2005).

**Table 1.** Germination (%) and vigour index of different genotypes of *Stylosanthes* under various treatments.

Genotypes	Germination %					Vigour index					
	Control	GA <sub>3</sub> (300ppm)	KNO <sub>3</sub> (0.2%)	Hot water (60 °C)	H <sub>2</sub> SO <sub>4</sub> (5 min.)	Control	GA <sub>3</sub> (300ppm)	KNO <sub>3</sub> (0.2%)	Hot water (60 °C)	H <sub>2</sub> SO <sub>4</sub> (5 min.)	Scarification
<i>S. hamata</i> 110123	21	25	25	22	60	95.40	112.50	117.60	102.0	357.5	351.0
<i>S. hamata</i> 110135	28	26	25	35	55	142.50	148.40	108.00	170.2	310.0	459.0
<i>S. hamata</i> 61670	15	25	18	18	70	49.40	105.60	39.00	102.6	403.2	330.0
<i>S. scabra</i> 36260	24	30	30	30	60	84.00	100.80	64.40	67.20	249.1	263.2
<i>S. scabra</i> 93116	10	20	12	22	50	16.10	79.20	35.00	100.0	263.2	607.2
<i>S. scabra</i> cv. fitzroy	20	24	22	20	32	64.60	110.40	85.10	106.0	128.8	413.4
<i>S. scabra</i> cv. seca	32	38	40	55	75	171.00	197.40	144.00	324.0	408.8	481.4
<i>S. scabra</i> q 10042	20	15	18	20	40	33.00	66.00	71.40	82.50	248.0	439.2
<i>S. scabra</i> RRR 94-100	30	30	25	20	40	114.80	120.00	73.60	96.90	173.9	403.2
<i>S. scabra</i> RRR 94-86	28	18	36	35	70	155.00	108.00	188.70	220.5	595.2	585.0
<i>S. scabra</i> RRR 94-93	16	18	12	20	40	29.00	64.50	19.00	74.80	151.7	349.4
<i>S. scabra</i> RRR 94-97	10	12	15	15	76	16.80	38.00	28.00	84.00	606.8	500.5
<i>S. scabra</i> RRR 94-96	10	25	15	18	65	20.00	127.50	43.20	87.00	448.5	648.0
<i>S. seabrana</i> 104710	20	18	20	22	62	63.00	96.00	54.00	95.40	353.8	762.6
<i>S. seabrana</i> 105546 B	25	38	32	25	75	117.30	199.80	129.00	151.8	474.5	612.0
<i>S. seabrana</i> 110372	16	25	18	25	70	32.40	110.70	48.00	111.3	325.0	360.4
<i>S. seabrana</i> 2523	16	20	18	20	66	50.70	88.20	81.60	90.00	352.8	693.0
<i>S. seabrana</i> 2534	22	18	18	20	38	57.00	92.00	54.00	95.20	180.0	565.8
<i>S. seabrana</i> 2539	20	15	20	20	35	122.40	90.10	66.60	107.1	174.0	518.3
<i>S. viscosa</i>	10	15	36	20	30	12.80	57.00	57.50	57.60	118.8	197.6
<b>Average</b>	<b>19.65</b>	<b>22.75</b>	<b>22.75</b>	<b>24.10</b>	<b>55.45</b>	<b>72.36</b>	<b>105.60</b>	<b>75.38</b>	<b>116.30</b>	<b>316.18</b>	<b>477.01</b>
<b>CD at 5%:</b>	<b>Genotypes (G) -NS, Treatments (T) -NS, G x T - NS</b>					<b>79.05</b>	<b>105.60</b>	<b>75.38</b>	<b>116.30</b>	<b>316.18</b>	<b>477.01</b>

**Table 2.** Seedling length (cm) and dry weight (g) of ten seedlings of different genotypes of *Stylosanthes* under various treatments.

Genotypes	Seedling length (cm)					Dry weight (g)						
	Control	GA <sub>3</sub> (300ppm)	KNO <sub>3</sub> (0.2%)	Hot water (60 °C)	H <sub>2</sub> SO <sub>4</sub> (5 min.)	Scarification	Control	GA <sub>3</sub> (300ppm)	KNO <sub>3</sub> (0.2%)	Hot water (60 °C)	H <sub>2</sub> SO <sub>4</sub> (5 min.)	Scarification
<i>S. hamata</i> 110123	5.20	4.50	4.10	5.30	6.50	3.90	0.026	0.017	0.017	0.013	0.013	0.037
<i>S. hamata</i> 110135	5.60	5.10	4.10	4.80	6.30	5.40	0.023	0.010	0.017	0.010	0.013	0.037
<i>S. hamata</i> 61670	3.90	4.60	2.90	5.70	5.80	4.40	0.013	0.017	0.010	0.013	0.013	0.030
<i>S. scabra</i> 36260	4.30	3.80	2.40	2.70	4.80	4.80	0.013	0.007	0.017	0.010	0.010	0.020
<i>S. scabra</i> 93116	2.60	4.40	3.50	5.20	5.80	6.90	0.007	0.010	0.010	0.007	0.013	0.023
<i>S. scabra</i> cv. fitzroy	3.90	4.90	3.80	5.40	4.80	5.40	0.017	0.013	0.017	0.010	0.013	0.037
<i>S. scabra</i> cv. seca	4.00	4.80	3.30	5.70	5.80	5.90	0.017	0.017	0.013	0.013	0.013	0.023
<i>S. scabra</i> q 10042	2.40	5.20	4.30	5.60	6.30	6.00	0.017	0.006	0.007	0.010	0.017	0.027
<i>S. scabra</i> RRR 94-100	4.30	4.10	3.10	5.80	4.80	5.80	0.013	0.013	0.013	0.013	0.020	0.023
<i>S. scabra</i> RRR 94-86	6.10	5.90	5.30	6.20	6.40	7.20	0.023	0.013	0.020	0.013	0.017	0.033
<i>S. scabra</i> RRR 94-93	2.90	4.40	2.00	4.40	4.30	5.60	0.010	0.010	0.013	0.013	0.010	0.020
<i>S. scabra</i> RRR 94-97	2.20	3.80	2.90	6.70	7.40	7.80	0.023	0.010	0.020	0.013	0.020	0.027
<i>S. scabra</i> RRR 94-96	4.10	5.10	3.80	5.90	6.90	8.00	0.010	0.013	0.010	0.013	0.013	0.027
<i>S. seabrana</i> 104710	4.20	6.40	3.20	5.40	6.00	8.10	0.013	0.010	0.007	0.010	0.013	0.030
<i>S. seabrana</i> 105546 B	5.00	5.40	4.40	6.50	6.50	7.40	0.023	0.020	0.017	0.013	0.017	0.037
<i>S. seabrana</i> 110372	2.80	4.30	3.30	5.40	5.20	5.40	0.010	0.013	0.013	0.010	0.010	0.027
<i>S. seabrana</i> 2523	4.00	4.90	4.90	6.00	5.80	7.80	0.030	0.006	0.010	0.013	0.013	0.023
<i>S. seabrana</i> 2534	4.00	4.80	3.80	5.80	6.00	6.90	0.013	0.013	0.020	0.017	0.013	0.023
<i>S. seabrana</i> 2539	5.40	5.20	3.80	6.40	5.90	7.20	0.020	0.017	0.007	0.013	0.010	0.027
<i>S. viscosa</i>	1.80	3.90	2.70	3.60	4.70	3.90	0.007	0.007	0.010	0.007	0.017	0.023
<b>Average</b>	<b>3.90</b>	<b>4.70</b>	<b>3.50</b>	<b>5.40</b>	<b>5.80</b>	<b>6.19</b>	<b>0.016</b>	<b>0.012</b>	<b>0.013</b>	<b>0.012</b>	<b>0.014</b>	<b>0.028</b>
<b>CD at 5%</b>	<b>Genotypes (G) – 0.326</b>	<b>Treatments (T) – 0.596</b>	<b>G x T – 0.231</b>	<b>Genotypes (G) – 0.00028</b>			<b>Treatments(T) – 0.00056</b>			<b>G x T – 0.00028</b>		

Highest seedling length (Table 2) was recorded in the seed scarification treatment (3.9 cm to 8.10 cm) followed by concentrated H<sub>2</sub>SO<sub>4</sub> and lowest seedling length was recorded in the treatment of KNO<sub>3</sub> (0.2%), which was at par with the control (1.8 cm to 6.10 cm). The genotypic variation in the seedling length was also observed among the different genotypes of *Stylosanthes*. Maximum seedling length was recorded in *S. seabrana* 104710 (8.10 cm) and *S. scabra* RRR 94-96 (8 cm) followed by *S. scabra* RRR 94-97 (7.80 cm) and minimum in *S. hamata* 110123 (3.90 cm) and *S. viscosa* (3.90 cm). In control highest seedling length was recorded in *S. scabra* RRR 94-86 (6.10) followed by *S. hamata* 110135 (5.60) and *S. seabrana* 2539 (5.40 cm) and minimum seedling length was recorded in *S. viscosa* (1.8 cm). Maximum dry weight of seedlings was recorded in the scarification treatments, which ranged from 0.020 to 0.037 g per ten seedlings (Table 2). On average the dry weight of seedlings was at par in the KNO<sub>3</sub> (0.2%) and concentrate H<sub>2</sub>SO<sub>4</sub> treatments. The genotypic variation in dry weight has also been observed.

As evident from the data (Table 1) highest value of vigour index was recorded in the scarification treatment followed by concentrated H<sub>2</sub>SO<sub>4</sub> and lowest in control whereas in other treatments it was at par. On average of the data of all the genotypes the vigour index improved by 6 fold by the scarification treatments over the control. The variability among the genotypes of different species of *Stylosanthes* was also indicated by the vigour index traits. Highest vigour index was recorded in *S. seabrana* (104710) followed by *S. scabra* (105546 B) and minimum in *S. viscosa*. The vigour index also improved to four folds by pre treatment of concentrate H<sub>2</sub>SO<sub>4</sub>. However, genotypic differences were observed in case of vigour index. Under the control, the highest vigour index was recorded in *S. scabra* cv.seca.

The difference in seed germination rate, seedling length and dry weight and vigour index among the various genotypes of *S. hamata*, *S. scabra* and *S. seabrana* clearly indicates the genotypic variability. The germination is positively and significantly correlated with vigour index ( $r = 0.9567$ ) (Fig. 1). Apart from these parameters the germination is also positively correlated

with seedling length ( $r = 0.5853$ ) and seedling dry weight ( $r = 0.4194$ ). The data revealed that scarification through coarse sand paper and conc. H<sub>2</sub>SO<sub>4</sub> (5 minute) are suitable dormancy breaking treatments for improving the germination in *Stylosanthes* species, however, there is difficulty in handling of concentrated sulphuric acid at farmers level. Therefore, scarification of seeds through coarse sand paper is the easiest and feasible method to remove the hard seed coat dormancy in different genotypes of *Stylosanthes* hence, recommended the most economical and practical dormancy breaking method for getting better germination and field establishment.

**Fig. 1. Relationship between vigour index and germination (%) in different genotypes of *Stylosanthes***

## ACKNOWLEDGEMENTS

The authors are highly thankful to the Director of the Institute and head of the Division for providing facilities.

## REFERENCES

- Airi, S., Rawal, R.S., Samant, S.S. and Dhar, U. (1998). Treatments to improve germination of four multipurpose trees of central sub Himalaya. *Seed Sci. Tech.* **26**: 347-354.
- Airi, S., Rawal, R.S. and Dhar, U. (2005). Presowing treatment effects on germination of *Cornus capitata* seeds. *Seed Sci. Tech.* **33**: 77-86.

SEED GERMINATION OF *STYLOSANTHES* SPECIES

- Bhatt, I.D., Rawal, R.S. and Dhar, U. (2000). Improvement in seed germination of *Myrica esculenta* Buch.-Ham. Ex. D. Don-a high value tree species of Kumaun Himalaya, India. *Seed Sci. Tech.* **28**: 597-606.
- Burin, M.E., Barros, R.S. and Rena, A.B. (1987). Chemical regulation of endogenous dormancy in seed of stylo. H.B. K.- Turrialba **37**: 281-285.
- Cameron, D.F. (1967). Hardseededness and seed dormancy of Townsville lucerne (*Stylosnathes humilis*) selections. *Aust. J. Exp Agri. & Anim. Hus.* **7**: 237-240.
- Diaz, Y., Viera, J. and Escobar, A. (1995). Effect of different methods of scarification on seed germination in *Pachecoa venezuelensis* Burkart. *Agronomia Tropical* (Maracay) **45**: 561-570.
- Hartmann, H.T., Kester, D.E. and Davies, E.T. (1989). Plant Propagation: Principals and Practices, Fifth edition. Prentice Hall, USA.
- Hopkinson, J.M. and Paton, C.J. (1993). Treatment of Seca stylo seed to reduce hard seed content. *Tropical Grassld.* **27**: 327-334.
- Li, X., Baskin, J.M. and Baskin, C.C. (1999). Anatomy of two mechanisms of breaking physiological dormancy by experimental treatments in seeds of two North American *Rhus* species (Anacardiaceae). *American J. Bot.* **9**: 237-245.
- Nan, Z.B., Hanson, J. and Yeshi, W.M. (1998). Effect of sulphuric acid and hot water treatments on seed born fungi and germination of *S. hamata*, *S. guianensis* and *S. scabra*. *Seed Sci. Tech.* **26**: 33-43.
- Pelacani, C.R., Ribeiro, D.M., Barros, R.S. and Frigeri, R.B.C. (2005). Germination of dormant seeds of *Stylosanthes humilis* as affected by organic acids. *Seed Sci. Tech.* **33**: 105-113.