The Pharma Innovation Journal 2019; 8(8): 438-441

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.03
TPI 2019; 8(8): 438-441

www.thepharmajournal.com
Received: 23-06-2019
Accepted: 25-07-2019

Anshu Dhaka
Department of Botany, D.N. (P.G.) College, Meerut, Utter Pradesh, India

Pradeep Chaudhary
Department of Statistics, Ch. Charan Singh University, Meerut, Utter Pradesh, India

Correspondence
Anshu Dhaka
Senior Lecturer, Department of Department of Botany, D.N. (P.G.) College, Meerut, Utter Pradesh, India

Effect of some pre-sowing treatments on *Sapindus trifoliatus* seed germination

Anshu Dhaka and Pradeep Chaudhary

Abstract
The seeds of Sapindus trifoliatus showed 100 percent viability as tested with Triphenyl tetrazolium chloride (TZ test). The electrical conductivity of the seed lechate was also observed to find out the integrity of membranous system of seed. The low ions leakage value (0.045 – 0.067µmho cm-^{1}) of EC also indicated the high level of intactness of membranous system and viability of the seeds. *Sapindus* genus usually undergoes physical or physiological dormancy. The seeds of *Sapindus trifoliatus* exhibited complete dormancy i.e. no germination when placed on wet filter paper under favourable condition showing the high level of seed dormancy. Seed germination increased through various seed treatments such as mechanical scarification, hot water, acid treatment and chipping plus chemical treatments (GA_3, KNO_3 and Thiourea). The exposure of seeds to hot water at 70 ^{0}C for 60 minutes recorded 10 - 12% germination, seedling vigour (10.0 – 11.0), vigour index (100-132.0) and speed of germination (1.60 – 1.81) as compared to 10, 30 min. and control. The seeds treated with 25% conc. H_2SO_4 recorded higher germination percentage (70-75) as compared to other scarification treatments. The maximum germination percentage (85-95), vigour index (1394.0-1463) and speed of germination (19.7–20.2) were recorded with 0.5% KNO_3 soaked seeds followed by the mechanical scarification (chipping +200 ppm GA_3 (24 hr soaking).

Keywords: Seed dormancy, germination, scarification, seed viability

Introduction
Sapindus trifoliatus of the family sapindaceae, commonly known as Reetha in Hindi and Soapnut in English. Plant commonly growing in south India and also cultivated in W. Bangal, Bihar, M.P. and U.P. The fruits contain saponin in their pericarp. According to Sharma *et al*., 2011 the fruits of *Sapindus trifoliatus* have been considered as a tonic, stomachic alexipharmic, astringent and fruitful in chronic dysentery, diarrhea, cholera, hemicranias, tubercular glands, paralysis and epileptic fits of children. The seeds of *S. trifoliatus* have stimulatory effect on uterus and are used in childbirth as well as to increase menstruation (pelegrini *et al*., 2008) [9]. The pericarp of *S. trifoliatus* are also used for washing hair, silks, woolens and delicate fabrics. Hence there is the need to conserve the genus *Sapindus* due to its multifunctional benefits. The seed germination of *Sapindus mukorossi, Sapindus emarginatus* and other species were known to be low and uneven (Dobhal *et al*., 2012, Swaminathan and Revathy 2013) [5, 11].
Physical dormancy is caused by structural barriers to germinate, such as hard seed coat and water proof and mechanical resistance of the seed coat to the growth of embryo. Physiological dormancy is caused by a number of physiological factors or immature embryos. *Sapindus* Genous has the type of physical and physiological dormancy (Cook *et al.*, 2008) [4]. Physical dormancy is caused by the existence of the hard seed coats (testa) which interfere the absorption of water. Lerak seed also has structure of the hard testa as well as Kepel (*Stelechocarpus burahol*) that can lead to the difficulty of germination (Isnaeni and Habibah, 2014) [7].
Therefore, in the present study an attempt has been made to study the effect of some pre-sowing treatments on seed germination including physical and chemical scarification in order to test, identify and workout the suitable pre-sowing treatment for *Sapindus trifoliatus.*

Material and Methods
The mature seeds of *Sapindus trifoliatus* were collected from NBPGR, Pusa Campus, New Delhi and other reliable sources for this study. The study on imbibition of water by the intact seeds, Quick viability test (TTC test) the seeds were imbibed for 8-10 hr. in water to soften the

seeds. The cotyledons were opened to expose the embryo and were soaked in colorless 0.5 - 1% solution of 2, 3, 5-triphenyl tetrazolium chloride (TTC) solution at 20^0C in the dark for 6-8 hr. The seeds were then washed 3-4 times with distilled water to permit direct observation of the embryo. The viable seeds appeared reddish in colour. The electrical conductivity was also observed to find out the integrity of membranous system of seed with a view to workout seed germination and viability test. The seeds (1g) were soaked in 20 ml of distilled water and incubated at 25 ^{0}C for 4 hr. Seed lechates was collected and electrical conductance (EC) was measured using a conductivity bridge (Elico-Ch-82T). Measurements were recorded in 3 replication each, EC of distilled water was taken as control. The seeds were subjected to the following treatments:
The pre-sowing treatments were given to *S. trifoliatus* seeds so that to break dormancy through the physical scarification.

Mechanical Scarification: In mechanical scarification the surface of the seed were scarified by creaking, chipping or rubbing on the rough surface or sand paper.

Hot water treatment: The seeds were dipped in hot water (70 ^{0}C) for 10, 30 and 60 Minutes duration and then cooled down to room temperature.

Acid treatments: The seeds were soaked in different concentrations of H_2SO_4 (100, 50 and 25%) for (5 minutes) according to the nature of their seed coat. After treatment the seeds were removed and washed thoroughly with water to remove the remaining acid.

Chemical treatment: The seeds were treated with different concentrations of GA_3, KNO_3 and Thiourea for 24 hr. soaking duration.

1. **Gibberellic acid:** chipped seeds were soaked in 100,200 and 300 ppm GA_3 concentration for 24 hr and then plated them on the moist filter / tissue paper for germination.
2. **KNO_3:** chipped seeds were soaked in 0.1%, 0.5% and 1% concentration of KNO_3 for 24 hr.
3. **Thiourea:** chipped seeds were soaked in 0.1%, 0.5% and 1% thiourea concentration for 24 hr.

Seed germination test: Germination test was conducted with four replicates of 25 seeds each, following the ISTA method at 27^0C.The germinated seeds were categorized in to normal, abnormal, dead and hard seeds counted every day. Germination percentage was recorded on the basis of normal seedling only. Seeds were considered to have germinated when the redicle has emerged out. The germination percentage (g) was calculated by using the formula

$$g = Ng / Nt \times 100.$$

Where (Ng) is the number of seeds germinated, (Nt) is the total number of seeds sown.

Vigour measurement: Vigour index was calculated as the product of seedling vigour (root + shoot length) and germination percentage.

Speed of germination: For calculating the speed of germination the germination counts were taken every 24 hr. and seeds were considered germinated when 1 mm radicle emerged. An index was computed for each treated lot by dividing the number seedlings removed each day by the day after planting on which they were removed.

$$\text{Speed of germination} = \frac{\text{No of seedling removed daily}}{\text{Days after planting}}$$

Statistical analysis: The data recorded for different parameters were analysed statically by method of analysis of variance as described by Cochran and Cox (1967) under completely randomized block design.

Effect of different pre - showing seed treatments on germination, root/shoot length (Seedling Vigour), vigour Index and speed of germination in different seeds lot of *Sapindus trifoliatus.*

Table 1: The shoot length cm root length cm total vigour index speed of germination

Treatments	Germination %		Shoot length cm		Root length cm		Total		Vigour index		Speed of germination	
Seeds lot	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Control	0	0	0	0	0	0	0	0	0	0	0	0
Scarification (Mechanical)	60	65	7.0	7.9	4.2	5.1	11.2	13.0	672.0	845.0	8.73	8.11
Hot water treatment (70 ^{0}C)												
10 min	0	0	0	0	0	0	0	0	0	0	0	0
30 min	0	0	0	0	0	0	0	0	0	0	0	0
60 min	10	12	6.1	6.7	3.9	4.3	10.0	11.0	100.0	132.0	1.60	1.81
Acid treatment (H_2SO_4)												
25%	75	70	6.3	6.0	3.5	3.8	9.8	9.8	735	686.0	15.2	14.9
50%	70	60	6.5	6.2	4.0	4.3	10.5	10.5	735	630.0	15.8	14.4
100%	60	50	3.5	4.5	3.5	3.5	7.0	8.0	420	400.0	21.0	20.0
Chipping + Chemical												
GA_3 100 ppm	70	70	8.1	8.9	5.5	5.9	13.6	14.8	952	1036.0	20.3	19.0
200 ppm	80	75	8.5	9.1	5.9	6.6	14.4	15.7	1152	1177.5	18.9	19.4
300 ppm	60	50	9.0	9.5	6.0	6.5	15.0	16.0	900	800.0	9.5	8.42
KNO_3 0.1%	90	80	8.8	9.1	6.1	6.5	14.9	15.6	1341	1248.0	20.3	19.0
0.5%	95	85	8.9	9.5	6.5	6.9	15.4	16.4	1463	1394.0	19.7	20.2
1.0%	80	70	9.2	8.5	6.0	6.1	15.2	14.6	1216	1022.0	13.0	12.3
Thiourea 0.1%	60	55	6.0	7.5	4.0	5.0	10.0	12.5	600	687.5	11.3	10.8
0.5%	80	75	6.5	7.8	4.2	5.5	10.7	13.3	856	997.5	13.7	14.4
1.0%	70	60	6.9	8.2	4.5	5.2	11.4	13.4	798	804.0	14.7	15.2
CD at 5%	5.11	5.50	0.43	0.55	2.98	0.41	0.71	0.937	56.56	77.20	0.736	0.940

Result and Discussion

The seeds of *Sapindus trifoliatus* under Stuty showed 100 percent viability as tested with triphenyl tetrazolium chloride (TZ) and observed electrical conductivity of seed lechates was also noted very low (0.045 – 0.067µmho cm-^{1}) indicating seed viability. The seeds of *Sapindus trifoliatus* medicinal plants studied under two lots showed very low or nil germination under favourable germination conditions. The seeds were scarified to overcome the seed coat dormancy constraints. The scarified seeds showed 60-65% germination, seedling vigour (11.2-13.0), vigour index (672-845) and speed of germination (8.73-8.11). Highest germination percentage was also reported by Vishal R. *et al*., 2013 [13] in seeds scarified with needle pin technique (76%) followed by sand paper treatment (56%) But this technique is very laborious and time taking. Earlier authers also observed the highest percentage of germination (60%) by treatment of seed coats sanding on the side of the hilum, followed by sanding on two sides (34.44%) and untreated (31.11) respectively in *Sapindus rarak* (sulisetijono *et al*., 2016) [12].

The seeds exposed to hot water (70 ^{0}C) for 60 minutes recorded 10-12 percent germination, seedling vigour (10.0-11.0), vigour index (100-132.0) and speed of germination (1.6-1.81) which showed the detrimental effects of long duration hot water treatments. The lowest germination percentage (37.78%) was also reported at 80 ^{0}C for 40 minutes whereas hot water soaking with the temperature of 50 ^{0}C for 20 minutes gave the highest percentage of germination (81.11%), followed by temperature of 50 ^{0}C for 30 minutes (72.22%) (Sulisetijono *et al*., 2016) [12]. the germination percentage in *S. mukorossi* increased (32%) in hot water treatmens than untreated seeds (Dobhal *et al*., 2012) [5]. The seed coat is made permeable to water through the hot water imbibision and this treatment soften and open gap water in the hard seed coat (Baskin *et al*., 2004) [2].

The seeds treated with 25%, 50% H_2SO_4 resulted into higher germination percentage (70-75), (60-70) respectively and showed higher seedling vigour (9.8cm), (10.5) vigour index (686-735), (630-735) whereas seeds treated with 100% H_2SO_4 showed lower germination (50-60) percentage, seedling vigour (7.0-8.0cm), vigour index (400-420). The results are in harmony with the findings of vishal R. *et al.*, 2013 [13] in *S. laurifolius* seeds who reported the poor germination in concentrated sulphuric acid (34% / 2 min.), whereas highest germination percentage (86% / 2 min.) recorded in 2N H_2SO_4 followed by (80% / 2 min) in 1N H_2SO_4 treatments respectively. The mean maximum germination (68.52%) was also reported in concentrated H_2SO_4 for 20 minutes followed by rinsing under running tap water pre-sowing treatment of *Sapindus mukorossi* seeds. (Varun Attri *et al*., 2017) [14].

Out of the chipped seeds soaked in different concentration of GA_3 (100, 200 and 300 ppm) the concentration of GA_3 (200 ppm) was found significantly superior in term of germination percentage (75-80), seedling vigour (14.4-15.7 cm), vigour index (1152.0-1177.5) and speed of germination (18.9-19.4) followed by 100 ppm conc, where germination percentage (70), seedling vigour (13.6-14.8) vigour index (952-1036.0) and in GA_3 (300 ppm) concentration the germination percentage (50-60), vigour index (800-900) and speed of germination (8.42-9.5). Similar findings of gibberellin ($GA_3$100 ppm for 30 minutes soaking) treatment were also recorded highest germination percentage (73.33) and the lowest percentage (46.67% in GA_3 90 ppm for 30 minutes soaking) in *S. rarak* DC. (Sulisetijono *et al*., 2016) [12]. Gibberellin has been reported to be able to overcome seed dormancy in many species through the mobilization of endosperm reserves during the early stages of seed germination (Hopkins & Huner, 2008) [6]. Gibberellin can be produced by the plants, but the amount is not enough to stimulate seed germination mainly on hard testa seed (Asra, 2014) [1]. External gibberellin provided will change the level of internal gibberellin contained in the seed and this level is a trigger factor for the germination process. Hopkin & Huner (2008) [6] suggested that internal gibberellins responsible for the formation of the enzyme alpha-amylase occurred at the beginning of germination. If the internal gibberellins are in limited quantities or not active then germination will be slow.

The chipped seeds were treated with different concentration of KNO_3 (0.1, 0.5 and 1.0%) for 24 hrs duration. The synergistic effects of mechanical scarification (chipping) + 0.5% KNO_3 (24hr) soaking treatments were observed in terms of maximum germination percentage (85-95) vigour index (1394.0-1463) and speed of germination (19.7-20.2). The germination percentage (70-80) and vigour index (1022.0-1216) under 1.0% KNO_3 treatments were recorded significantly lower than 0.1, 0.5% KNO_3 conc. treatments.

The chipped seeds were soaked in different concentration of Thiourea (0.1, 0.5 and 1.0%) for 24 hrs. These all concentrations significantly increased the germination percentage ranging from 55 to 80), seedling vigour from10.0 to13.4cm, vigour index from 600 to 997.5 and speed of germination from 10.8 to 18.5 over the chipping scarification alone with germination percentage from 60 to 65 showing synergistic effect. The thiourea of 0.5% concentration was found significantly superior over 0.1 and 1.0 percent concentration in terms of germination percentage and vigour index.

References

1. Asra R. Effect of hormone gibberellin (GA3) on germination and vigoritas Calopogonium caeruleum. Biospecies. 2014; 7(1):29-33.
2. Baskin JM, Davis BH, Baskin CC, Gleason SM, Cordell S. Physical dormancy in seeds of Dodonaea viscosa (*Sapindales*, *Sapindaceae*) from Hawaii. Abstrak. Seed Science Research. 2004; 14(1):81-90.
3. Cochran WG, Cox GM. Experimental Designs, Asia Publication House, New Delhi, 1957.
4. Cook A, Turner SR, Baskin JM, Baskin C, Steadman KJ, Dixon KW. Occurrence of physical dormancy in seeds of Australian *Sapindaceae*: A Survey of species in nine genera. Annals of Botany. 2008; 101:1349-1362.
5. Dobhal U, Bhandari S, Bisht S, Bisht NS. Seed germination of *Sapindus mukorossi*. Life Sciences Leaflets. 2012; 3:60-63.
6. Hopkin WG, Huner NPA. Introduction to Plant Physiology. Fourth Edition. Hoboken, New Jersey United States of America: John Wiley & Sons, Inc, 2008.
7. Isnaeni E, Habibah E. Effectiveness of scarification and soaking temperature on seed germination Kepel [*Stelechocarpus burahol* (Blume) Hook f. & Thompson] *in vitro* and ex vitro. Jurnal MIPA. 2014; 37(2):105-114.
8. Lakon G. Topographischer nechwein derkeimgfahigkeit der getried efriichtra duch Tetrazoliumsalze. Ber. Bot. Ges. 1942; 67:299-305.
9. Pelegrini DD, Tsuzuki JK, Amado CAB, Cortez DA, Ferreira ICP. Biological activity and isolated compounds in *S. saponaria* L. and other plants of the Genus

Sapindus. Lat. Am. J. Pharm. 2008; 27(6):922-927.
10. Sharma A, Sati SC, Sati OP, Sati D, Manisha, Kothiyal SK. Chemical constituents and bioactivities of genus *Sapindus*, International Journal of Research in Ayurveda & Pharmacy. 2011; 2(2):403-409.
11. Swaminathan C Revathy R. Improving seed germination in *Sapindus emarginatus* Vahl. Pinnacle Agricultural Research & Management. Volume 2013, Article ID parm-101, 2013, 3.
12. Sulisetijono. Estri Laras Arumingtyas Retno Mastuti, Serafinah Indriyani. Physical and chemical treatments to break seed dormancy on Lerak (*Sapindus rarak* DC.) International Journal of Agriculture and Environmental Research ISSN: 2455-6939 Volume: 02, Issue: 04; 936-947, 2016.
13. Vishal Kamble R, Bazegah Sayed K, Shrinath Kavade P. Effect of some presowing treatments on *Sapindus laurifolius* seed germination journal of research in plant sciences. 2013; 2(2):205-212.
14. Varun Attri, Pant KS, Navjot Singh, Vipasha Negi. Influence of seed size and pre-sowing treatments on germination parameters of *Sapindus mukorossi* Gaertn under Laboratory condition. Int. J Curr. Microbiol. App. Sci. 2017; 6(10):2788-2799.