academicJournals

Vol. 8(24), pp. 3172-3176, 27 June, 2013
DOI: 10.5897/AJAR2013.7288
ISSN 1991-637X ©2013 Academic Journals
http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Standardization of germination media for the endangered medicinal tree, bael (*Aegle marmelos*)

B. Venudevan[1]*, P. Srimathi[2], N. Natarajan[3] and R. M. Vijayakumar[4]

[1]Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore-3, India.
[2]Seed Centre, Tamil Nadu Agricultural University, Coimbatore-3, India.
[3]Department of Nano Science and Technology, Tamil Nadu Agricultural University, Coimbatore-3, India.
[4]Department of Medicinal and Aromatic Crops, Tamil Nadu Agricultural University, Coimbatore-3, India.

Accepted 14 June, 2013

Bael is an endangered medicinal tree, highly propagated through seeds. Either on trade or before sowing evaluation of seed germination is essential. International rules for seed testing recommended different techniques for seed quality evaluation. One such requirement is the selection of media for germination to assure actual germination as media provide proper platform for full expression of seedling emergence and growth. Hence, the studies were conducted at the Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore, on standardization of media and methodologies for germination test under germination room conditions in line with ISTA for Bael seeds. The results revealed that, either river sand or paper media could be used for obtaining reproducible and complete expression of germination of seed. In river sand, in-sand method (seed are to be sown at depth of 2 cm) and in paper, between paper (Roll towel) method had better expression for germination (83 and 78%) respectively, recommending it as the best media for evaluation of germination percentage of Bael seeds in seed testing.

Key words: Bael, endangered, sand or paper media, seedling emergence, germination.

INTRODUCTION

Aegle marmelos (L.) Corr. is a medicinal tree which belongs to the family Rutaceae and its various parts are used in Ayurvedic and Siddha medicines to treat a variety of ailments. Bael is highly habitated to tropical and subtropical climate of India, Burma, Pakistan, Bangladesh, Sri Lanka, Northern Malaya, Java and Philippine (Islam et al., 1995). It is a medium sized tree having profuse dimorphic branches, alternate, trifoliate and deep green leaves, membranous leaflets, large sweet scented, greenish white flowers, large and globose fruits (Purohit and Vyas, 2005). Mazumder et al. (2006) revealed that, approximately 200 to 250 kg of fruits could be obtained per tree. All parts of the tree are highly useful in preparation of herbal medicines (Kala, 2006). The roots are useful for treating diarrhea, dysentery and dyspepsia. The aqueous stem and root bark extracts are used as medicine for malaria, fever, jaundice, cancer, ulcers, urticaria and eczema (Nadkarni, 1954).The fruit and root of the plant have antiamoebic and hypoglycaemic activities (Ponnachan et al., 1993). Goel et al. (1997) revealed that, crop is rich with the alkaloids aegline, marmesin, marmin and marmelosin. Rana et al. (1997) revealed that, the seed is rich in luvangetin and pyranocoumarin compounds, which has antiulcer activity. They also revealed that, essential oil isolated from the leaf has antifungal activity. The foundation for revitalization of local health traditions (FRLHT), Banglore, India listed bael *(Aegle marmelos*) as rare, endangered

*Corresponding author. E-mail: venudevan005@gmail.com.

Table 1. Seeds sown in different media adopting different methodologies.

Media	Methodologies for germination test	
Germination paper	✓	Top of the paper (TP)
	✓	Between the paper (Roll towel)
River sand	✓	sowing at 2 cm depth
Quartz sand	✓	sowing at 2 cm depth
Vermiculite	✓	sowing at 2 cm depth
Inclined plate		

and threatened (RET) species specifically endangered species. This underutilized tree is generally propagated through seeds. ISTA (1993) formulate, procedures for testing the physical, physiological and health status of seed, which differ from seed to seed and newer crops are added based on necessity. Among the seed quality characters, evaluation of germination is the prime and most important reliable character that explores the relative planting value of the seed lot. It should be evaluated in a correct media, to give accurate and reproducible results. The objective of the germination test is to express the maximum germination potential of seed which is the most important than any other quality parameters.

The use of standardized ideal techniques in the laboratory as prescribed by ISTA is warranted as it ensures that, results obtained for a given seed lot in one laboratory would be identical with those obtained from any other laboratory in the same or other countries (Willan, 1985). In the laboratory, the environmental conditions, including moisture, temperature, aeration and light, must not only be specific enough to indicate germination but also favorable for the development of the seedlings to a stage where interpretation as normal and abnormal types were possible. Medium plays an important role in germination testing, because seeds have characteristic requirements of moisture and oxygen for germination. Thus, minute seeds germinate well on top of paper (TP) rather than sand. Best suited medium depends upon the physical condition of the seeds. Generally, sand as medium is best suited for large sized seeds and paper for small sized seeds (Nawabahar, 2008). Hence, studies were initiated to standardize suitable media and methodology for evaluation of seed germination of Bael seeds at the Department of Seed Science and Technology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu during 2012.

MATERIALS AND METHODS

Bael seeds were collected from different places of Coimbatore district (76°57 E, 11°8 N and 320 MSL) which are; Ram Nagar, Perur, Karamadai, and Saibaba colony. To evaluate the germination potential and seedling growth rate, the seeds were sown in different media adopting different methodologies as follows (Table 1 and Figure 1).

In each of the media/methodologies, 100 seeds in four replications were sown and kept under the germination room maintained at 25°C and 95 ± 2% RH. Data on daily germination was recorded in each of the media until no further germination was observed that is, up to 23 days of germination period. Then the germination test was terminated and the resultant seed and seedlings were categorized into normal, abnormal, and dead seeds. In randomly selected all normal seedlings, the seedlings were measured for their root and shoot length and the mean reported as root and shoot length per seedlings. The seedlings were dried for 48 h in an oven maintained at 85°C and weighed in a top pan balance and the mean dry matter per seedlings in milligram. Vigour index values, the totality expressions of seed quality characters, were also computed as per Abdul and Anderson (1973) adopting the following formula and the results reported as whole number.

Vigour index = Germination (%) × Total seedling length (cm)

The data gathered from various sources for all the seed quality parameters were subjected for analysis of variance (ANOVA) as per Panse and Sukhathme (1995) to determine the level of significance (5%).

RESULTS AND DISCUSSION

Highly significant variations were observed for the evaluated seed quality parameters obtained from different growing media, methodologies and locations (Table 2). The results revealed that, germination was initiated 11 days after sowing in river sand on adopting in-sand method while it took 12 days for initiation of germination in roll towel (RT) and modified RT method. The initiation of germination was observed between 13 to 15 days. The seed germination recorded based on normal seedlings was the highest germination (83%) in sand media sown at 2 cm depth and was followed by the paper; RT 78%, quartz sand 76%, inclined plate 73% and vermiculite 67%. The root 12.8 cm and shoot length 11.6 cm, dry matter production 47.9 mg, and vigour index (1866) also recorded the highest value in sand media sown at 2 cm depth and was followed by between the paper (roll towel) (Tables 3 and 4). It was also observed that, expression of seedlings as normal seedling was more in sand and paper media on adoption of modified RT method and the lowest germination percentage (56%), root (5.0 cm) and shoot length (5.4 cm), dry matter production (20.0 mg) and vigour index (504) recorded with top of the paper (TP) method expressing

Figure 1. Standardization of different germination media.

that, it could not be used for evaluation of germination of bael seeds, might be due to the insufficiency of water and space requirement for development of the seedlings as normal seedlings for this medium sized seeds.

Seeds sown in sand at 2 cm depth produced the maximum germination of 83% irrespective of seed sources. However, Zhe et al. (2009) reported that, seeds in quartz sand seemed to grow faster and the seedlings would die in shorter durations, but in the present study, though there is no death in quartz sand, germination was easier in river sand than quartz sand.

Based on easy availability and economic utility, for Bael the river sand could be used as the testing media, but in that too, in-sand method (sowing of the seed at 2 cm) should be adopted for obtaining reproducible results in the germination test. It was followed by the other media that is, between the paper (Roll towel) and quartz sand. According to Nawabahar (2008), the choice of media depends on the species being tested, as species seems to have a distinctive preference for a particular media. Minute seeds are best germinated on TP while large seeds are best germinated in sand media or RT. Towel paper is more commonly used for medium sized seed because it is easier to handle than sand and permits

Table 2. Standardization of germination media on days to first emergence and germination percentage in Bael (*Aegle marmelos*) seeds .

Media (M)	Days to first emergence					Germination (%)				
	Ram nagar	Perur	Karamadai	Saibaba colony	Mean	Ram nagar	Perur	Karamadai	Saibaba colony	Mean
Top of the paper (Petri plate)	16	17	13	15	15	54(47.29)	55(47.87)	58(49.60)	56(48.44)	56(48.44)
Between the paper (roll towel)	13	10	11	15	12	76(60.66)	79(62.72)	80(63.43)	78(62.02)	78(62.02)
Sand (2 cm depth)	12	10	11	12	11	80(63.43)	87(68.86)	81(64.15)	85(67.21)	83(65.65)
Vermiculate	15	14	12	14	14	68(55.55)	69(56.16)	66(54.33)	65(53.73)	67(54.94)
Quartz sand	14	15	11	13	13	74(59.34)	75(60.00)	78(62.02)	76(60.66)	76(60.66)
Inclined plate	15	11	13	15	14	72(58.05)	73(58.69)	71(57.41)	74(59.34)	73(58.69)
SEd	1.570	1.428	1.526	1.549		2.429	1.903	1.882	1.940	
CD (P = 0.05)	3.140	2.947	3.045	3.070		5.104	3.998	3.954	4.077	

Table 3. Standardization of germination media on root and shoot length in bael (*Aegle marmelos*).

Media (M)	Root length (cm)					Shoot length (cm)				
	Ram nagar	Perur	Karamadai	Saibaba colony	Mean	Ram nagar	Perur	Karamadai	Saibaba colony	Mean
Top of the paper (Petri plate)	4.7	5.7	2.7	6.7	5.0	4.6	6.6	2.6	7.6	5.4
Between the paper (roll towel)	10.4	11.4	8.4	12.4	10.7	9.2	11.2	7.2	12.2	10.0
Sand (2 cm depth)	12.5	13.5	10.5	14.5	12.8	10.8	12.8	8.8	13.8	11.6
Vermiculate	7.8	8.8	5.8	9.8	8.1	8.6	10.6	6.6	11.6	9.4
Quartz sand	8.5	9.5	6.5	10.5	8.8	9.1	11.1	7.1	12.1	9.9
Inclined plate	8.9	9.9	6.9	10.9	9.2	8.8	10.8	6.8	11.8	9.6
SEd	0.768	0.542	0.459	0.614		0.473	0.502	0.494	0.537	
CD (P = 0.05)	1.614	1.140	0.965	1.291		0.995	1.055	1.039	1.129	

easier and quicker development of seedlings. Large seeds could also be rolled in paper towel with limited seeds but is a very inconvenient method to test a large number of seeds at a time. Sand is not suitable for very small seeds but is widely used for large seeds especially for tree seeds that have longer germination period. Sand can also be sterilized easily and fungal development on sterilized sand is controlled better than paper media. It also provides good contact between the seed and moisture as seed is pressed into the medium, the bigger particle size aids in aeration that favoured the production of normal seedling in higher number. The sand was found to be the best medium for improving the forest tree seed germination as expressed by Anber (2010).

These results were in supportive of the findings of Egharevba et al. (2005) in African walnut, *Plukenetia conophorum,* Bahuguna et al. (1987a, b) in *Terminalia myricarpa* and *Adhatoda vasica,* Murugesan et al. (2008) in oil palm, Vilela and Ravette (2001) in species of Prosopis, Docker and Hubble (2008) in some Australian tree species and by Thapliyal and Rawat (1991) in *Alnus nitida* and *A. nepalensis* who, recommends sand medium for germination testing under the process of seed testing for expression as normal seedlings in germination room condition irrespective of the source seeds.

Table 4. Standardization of germination media on dry matter production and vigour index in bael (*Aegle marmelos*).

Media (M)	Dry matter production (mg seedlings $^{-10}$)					Vigour index				
	Ram nagar	Perur	Karamadai	Saibaba colony	Mean	Ram nagar	Perur	Karamadai	Saibaba colony	Mean
Top of the paper (Petri plate)	19.9	20.3	19.5	20.2	20.0	502	507	497	508	504
Between the paper (roll towel)	44.2	44.6	43.8	44.5	44.3	1411	1416	1406	1417	1413
Sand (2 cm depth)	47.8	48.2	47.4	48.1	47.9	1864	1869	1859	1870	1866
Vermiculate	42.1	42.5	41.7	42.4	42.2	1115	1120	1110	1121	1117
Quartz sand	43.9	44.3	43.5	44.2	44.0	1302	1307	1297	1308	1304
Inclined plate	39.5	39.9	39.1	39.8	39.6	1345	1350	1340	1351	1347
SEd	1.209	0.974	1.350	2.018		73.767	75.455	58.384	64.956	
CD (P = 0.05)	2.541	2.047	2.837	4.240		155.714	158.527	122.663	136.470	

Conclusion

The study highlighted that, seed germination could be tested in Bael seeds using either sand media or germination paper, on using sand media, seeds should be sown at the depth of 2 cm and paper media modified RT methods has to be adopted for obtaining reproducible results.

REFERENCES

Abdul BA, Anderson JD (1973). Vigour determination in soybean seed by multiple criteria. Crop Sci. 13:630-633.

Anber MA (2010). Improving seed germination and seedling growth of some economically important trees by seed treatments and growing media. J. Hortic. Sci. Ornamental Plants 2(1):24-31.

Bahuguna VK, Rawat MMS, Joshi SR, Maithani GP (1987a). Studies on the viability, germination and longevity of *Terminalia myriocarpa* seed. J. Trop. Forest. 3(IV):318-323.

Bahuguna VK, Sood OP, Rawat MMS (1987b). Preliminary studies on the germination behaviour of *Adhatoda vasica* seeds. An important shrub for regeneration of Sub-Himalayan wastelands. Indian For. 113(6):256-261.

Docker BB, Hubble TCT (2008). Quantifying root reinforcement of river bank soils by four Australian tree species. Geomorphology 100(3-4):401-418.

Egharevba RK, Ikhatua MI, Kalu C (2005). The influence of seed treatments and growing media in seedling growth and development of African walnut, *Plukenetia conophorum.* Afr. J. Biotechnol. 4(8):808-811.

Goel RK, Maiti RN, Manickam M, Ray AB (1997). Antiulcer activity of naturally occurring pyranocoumarin and isocoumarins and their effect on prostanoid synthesis using human colonic mucosa. Ind. J. Expt. Biol. 35:1080-1083.

Islam R, Hossain M, Karim MR, Joarder OI (1995). Regeneration of *Aegle marmelos* (L.) Corr., plantlets *in vitro* from callus cultures of embryonic tissues. Cur. Sci. 69:494-495.

ISTA (1993). International Rules for Seed Testing. Seed Sci. and Technol. 21:1-288(suppl.).

Kala CP (2006). Ethnobotany and ethnoconservation of *Aegle marmelos* (L.) Correa. Indian J. Trad. Knowl. 5:541-550.

Mazumder R, Bhattacharya S, Mazumder A, Pattnaik AK, Tiwary PM, Chaudhary S (2006). Antidiarrhoeal evaluation of *Aegle marmelos* (Correa) Linn. root extract. Phytother. Res. 20:82-84.

Murugesan P, Bijimol G, Haseela H (2008). Effect of different substrates on growth of germinated oil palm hybrid seeds. Indian J. Hortic. 65(4):477-480.

Nadkarni KM (1954). Indian material Medica, 3rd edn. Popular Book Depot, Bombay, India. pp. 45-49.

Nawabahar (2008). Effect of media on seed germination of *Cupaniopsis anacardioides* (A. Rich.) Radlk. Indian J. For. 31(1):137-139.

Panse VS, Sukhatme PV (1995). Statistical Methods for Agricultural Workers. Indian Council for Agricultural Research, New Delhi, India.

Ponnachan PTC, Paulose CS, Panikar KR (1993). Effect of the leaf extract of *Aegle marmelos* (L.) Corr. In diabetic rats. Indian J. Exp. Biol. 31:345-347.

Purohit SS, Vyas SP (2005). Medicinal Plant Cultivation-A Scientific Approach. Agrobion. India. P. 282.

Rana BK, Sing UP, Taneja V (1997). Antifungal activity and kinetics of inhibition by essential oil isolated from leaves of *Aegle marmelos* (L.) Corr. J. Ethnopharmacol. 57:29-34.

Thapliyal RC, Rawat MMS (1991). Studies on the germination and viability of seed of two species of Himalayan Alders (*Alnus nitida* and *A. nepalensis*). Ind. For. 117(4):256-261.

Vilela AE, Ravetta DA (2001). The effect of seed scarification and soil media on germination, growth, storage and survival of seedlings of five species of Prosopis L. (Mimosaceae). J. Arid Environ. 48:171-184.

Willan RL (1985). A guide to forest seed handling with special reference to the tropics. FAO Forestry Paper, 20/2. FAO, Rome.

Zhe J, Paolo P, Robert F, Peter M, Paolo B (2009). An experimental comparison of silica gel and quartz sand grains as sediment media for growing vegetation at the laboratory scale. Aquat. Sci. 71:350-355.