

Short communication**Hot water treatment for improving germination in *Melochia corchorifolia***

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ABSTRACT

Melochia corchorifolia seeds collected from Odisha were tested for germination before conserving in genebank, exhibited physical dormancy due to hard seededness. In order to determine the most suitable method to reduce the hard seeds and promote rapid and uniform germination, seeds were subjected to hot water treatment at different temperatures and duration. Soaking seeds in water at 70°C for 10 min. was found most effective in breaking dormancy and on an average, germination increased from 7 to 96% in two genotypes. Besides germination, seedling vigour, vigour index and electrolyte leakage were also noticed significantly higher in the treated seeds than in untreated seeds.

Key words: *Melochia corchorifolia*, wild species, dormancy, hot water treatment, seed germination.

Wild vegetables are an important component of traditional food systems across the world. *Melochia corchorifolia* (Syn *M. Concatenata*) belongs to the family Sterculiaceae, which is commonly found on the bunds of upland and lowland rice fields. It is used as leafy vegetable by the Santhal tribe habitating in Jharkhand, Odisha and West Bengal (Sinha and Lakra, 11). Seeds collected from Nayagarh and Bargarh districts of Odisha were processed for conservation in National Genebank (NGB). It was observed that seeds of two genotypes remained hard for more than 40 days and showed physical dormancy due impermeable seed coat. Physical dormancy is prevalent in as many as 17 plant families including Sterculiaceae, which is mainly due to structural and chemical characteristics of epidermal cells and restriction of water and oxygen movement inside the seed. In general, germination of such seeds can be enhanced by making their seed coat permeable to water through physical and chemical treatments. However, precise information regarding dormancy breaking regimes are lacking in *M. corchorifolia* and moreover treatment devised to break the dormancy is highly specific to species, therefore, the objective of this experiment was to investigate the effect of hot water treatments in breaking dormancy and to improve uniform and rapid germination for seed testing and for better field establishment under normal conditions during multiplication and regeneration of this species.

An experiment was carried out in two genotypes of *M. corchorifolia* (G1: IC610692, G2: IC610794) collected from Odisha during September 2014, seeds of which were subjected to hot water treatment at

varying duration. Three hundred seeds from each seed lot were wrapped in muslin cloth and immersed in water bath at 60, 70 and 80°C for 10 and 20 min. each. Accordingly, the seeds were subjected to seven treatments prior to germination: T1- Hot water 60°C for 10 min; T2-Hot water 60°C for 20 min; T3-Hot water 70°C for 10 min; T4-Hot water 70°C for 20 min; T5-Hot water 80°C for 10 min; T6-Hot water 80°C for 20 min and untreated seeds were used as the control (T7). After the treatments, seeds were allowed to cool at room temperature for 2 h before testing for germination. Three replicates of 50 seeds from each treatment were placed between papers and kept at a temperature of 30 ± 1°C in a germinator without light for 14 days. At the end of germination test, seeds were evaluated according to ISTA Rules (5) and the percentage of seeds in each category was calculated. The vigour indices were calculated according to Abdul-Baki and Anderson (1). Electrolyte leachate of treatments were measured after 24 hrs and expressed as per the method given by Matthews and Bradnock (6) and mean germination time (MGT) was calculated as per the protocol suggested by Ellis and Roberts (4). The data were analysed using R Core software developed by R Core Team (9). The data in percent were Arcsine transformed and then subjected to ANOVA and least significance differences (LSD) test for comparison among the treatments at P = 0.05 level using Agricolae and Car package.

All treatments applied to improve the germination were statistically significant. The initial germination percentage of the genotypes ranged from 5 to 9% with an average of 7%. Seeds immersed in water at 70°C for 10 min significantly reduced the percent hard seed and increased the germination up to 96% (average)

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in both the genotypes, followed by 60, 70 and 80°C for 20 and 10 minutes, respectively (Table 1). Hot water treatment reduced the proportion of hard seeds to zero but longer exposures at high temperature (80°C for 20 minutes) decreased germination to an average of 40% in both the genotypes and increased the number of abnormal seedlings and dead seeds because of excessive damage to the embryo or tissue inside the seed coat. On the other hand, soaking seeds at 60°C for 10 min was not found to be significantly effective to stimulate the germination, probably due to softening of seed coat and making it permeable to water. Eastin (3) also reported that scarification of freshly harvested *M. corchorifolia* seeds for 15-30 min in an electric scarifier resulted in higher germination index (662) as compared to non-scarified seeds (25) indicating that the softening of coat improves germination, whereas failure of non-scarified seeds to attain full germination seems to be mediated through interference in water uptake by the seed coat as reported in different leguminous species (Bewely and Black, 2).

In general, immersing seeds in hot or boiling water have been reported by many researchers as an effective method of softening seed coat, thus allowing a greater permeability of water and overcoming hard seededness in several tree species. The results obtained in the present study were on similar lines as reported by Srinivasan *et al.* (12) in *Corchorus*

olitorius and Rehman *et al.* (8) in *Acacia salicina*. Results of the present experiment showed significant effect on mean germination time (MGT) due to hot water treatment, irrespective of the genotypes studied. Although, all the treatments were effective in reducing the MGT over that of control, but only the brief exposure (10 minutes) to hot water at 70°C increased the germination up to 96% in a very short period of 1.12 days after imbibition as compared to control (5.09 days; Table 1). This could be because of rapid activation of enzymatic activities in the seeds treated with hot water at 70°C for 10 min. Similar results have been reported by Sharma *et al.* (10) in *Albizia lebbek*, *Albizia procera*, *Peltophorum pterocarpum*, *Acacia auriculiformis* and *Leucaena leucocephala*, where seeds immersed at 100°C for 1-10 minutes increased the germination 7-9 folds (94-100%) and decreased the mean germination time from 17-20 days to 4-6 days in treated seeds.

Evaluation of the seedling characters revealed that seedling length, fresh and dry weight of seedlings showed no significant difference between seeds immersed at 60 and 70°C as well as for 20 and 10 min., respectively, even though both the treatments did differed significantly in their percent germination, indicating that hot water treatment causes minimum damage to the seeds (Table 2). In general, hot water treatment significantly increased the vigour index I and II compared to the control (Fig. 1). Electrical

Table 1. Effect of hot water treatments on germination percent and mean germination time in different *M. corchorifolia* genotypes.

Hot water treatment	Normal seedling (%)		Abnormal seedling (%)		Hard seed (%)		Mean germination time (days)	
	G1	G2	G1	G2	G1	G2	G1	G2
60°C for 10 min	49.33 (44.62) ^d	48.00 (43.85) ^d	0.00 (4.05) ^e	0.00 (4.05) ^e	50.67 (45.38) ^b	52.00 (46.15) ^b	1.61 ± 0.06 ^{de}	1.67 ± 0.16 ^b
60°C for 20 min	82.67 (65.45) ^{bc}	82.00 (64.92) ^{bc}	3.33 (10.4) ^{de}	0.00 (4.05) ^e	14.00 (21.94) ^c	18.00 (25.08) ^c	1.22 ± 0.02 ^e	1.28 ± 0.05 ^{bc}
70°C for 10 min	95.33 (77.58) ^a	96.67 (79.61) ^a	0.67 (5.41) ^e	0.00 (4.05) ^e	4.00 (11.54) ^d	3.33 (10.4) ^{de}	1.07 ± 0.06 ^e	1.17 ± 0.04 ^{de}
70°C for 20 min	88.00 (69.77) ^b	80.00 (63.45) ^{bc}	9.33 (17.77) ^c	10.00 (18.38) ^c	0.00 (4.05) ^e	4.67 (12.42) ^d	2.29 ± 0.24 ^{cd}	1.52 ± 0.12 ^{de}
80°C for 10 min	85.33 (67.53) ^{bc}	78.00 (62.04) ^c	8.00 (16.35) ^{cd}	14.00 (21.94) ^{bc}	0.00 (4.05) ^e	0.00 (4.05) ^e	1.4 ± 0.02 ^{de}	1.71 ± 0.27 ^{de}
80°C for 20 min	34.00 (35.65) ^e	38.00 (38.05) ^{de}	20.67 (27.02) ^{ab}	28.67 (30.19) ^a	0.00 (4.05) ^e	0.00 (4.05) ^e	3.27 ± 0.13 ^b	3.14 ± 0.27 ^e
Control (untreated seeds)	5.33 (13.30) ^f	8.67 (17.11) ^f	0.00 (4.05) ^e	0.00 (4.05) ^e	94.67 (76.70) ^a	91.33 (72.91) ^a	4.92 ± 0.36 ^a	3.86 ± 0.26 ^e

G1: Genotype 1 (IC 610692); G2: Genotype 2 (IC 610794)

Data in parenthesis are Arc Sin transformed values. Values superscripted with different letter(s) in same column are significantly different at 0.05 level.

Table 2. Effect of hot water treatments on seedling length, fresh and dry weight of different genotypes of *M. corchorifolia*.

Hot water treatment	Genotype 1 (IC610692)			Genotype 2 (IC610794)		
	Seedling length (cm)	Fresh weight (mg)	Dry weight (mg)	Seedling length (cm)	Fresh weight (mg)	Dry weight (mg)
60°C for 10 min	10.56 ± 0.15 ^{abc}	134.6 ± 5.66 ^{ab}	12.9 ± 0.65 ^{sd}	9.44 ± 0.44 ^{bcd}	133.9 ± 7.37 ^{ab}	12.3 ± 0.55 ^{de}
60°C for 20 min	10.94 ± 0.57 ^{ab}	151.3 ± 9.53 ^a	16.07 ± 0.38 ^{abc}	11.98 ± 0.97 ^a	138.27 ± 3.63 ^{ab}	14.4 ± 0.67 ^{abcd}
70°C for 10 min	11.79 ± 0.34 ^{ab}	151.4 ± 3.64 ^{ab}	16.47 ± 0.52 ^a	12.02 ± 0.24 ^a	163.4 ± 8.76 ^a	16.3 ± 0.53 ^{ab}
70°C for 20 min	10.99 ± 0.39 ^{ab}	150.57 ± 6.54 ^{ab}	14 ± 0.25 ^{abcd}	12.27 ± 0.29 ^a	129.47 ± 24.51 ^{ab}	13.17 ± 0.58 ^{bcd}
80°C for 10 min	7.78 ± 0.23 ^{de}	132.8 ± 10.5 ^{ab}	12.1 ± 1.31 ^{de}	8.32 ± 0.28 ^{cde}	133.7 ± 10.6 ^{ab}	13.4 ± 0.67 ^{abcd}
80°C for 20 min	7.18 ± 0.22 ^{de}	124.67 ± 7.31 ^b	7.13 ± 0.29 ^f	6.7 ± 0.18 ^e	102.83 ± 6.02 ^b	9.17 ± 0.26 ^{ef}
Control (untreated seeds)	8.16 ± 0.41 ^{de}	38 ± 1.73 ^c	0.27 ± 0.03 ^g	7.73 ± 0.26 ^{de}	31 ± 1.73 ^c	0.27 ± 0.03 ^g

Data in parenthesis are Arc Sin transformed values of the original percentages. Values superscripted with different letter in same column are significantly different at 0.05 level.

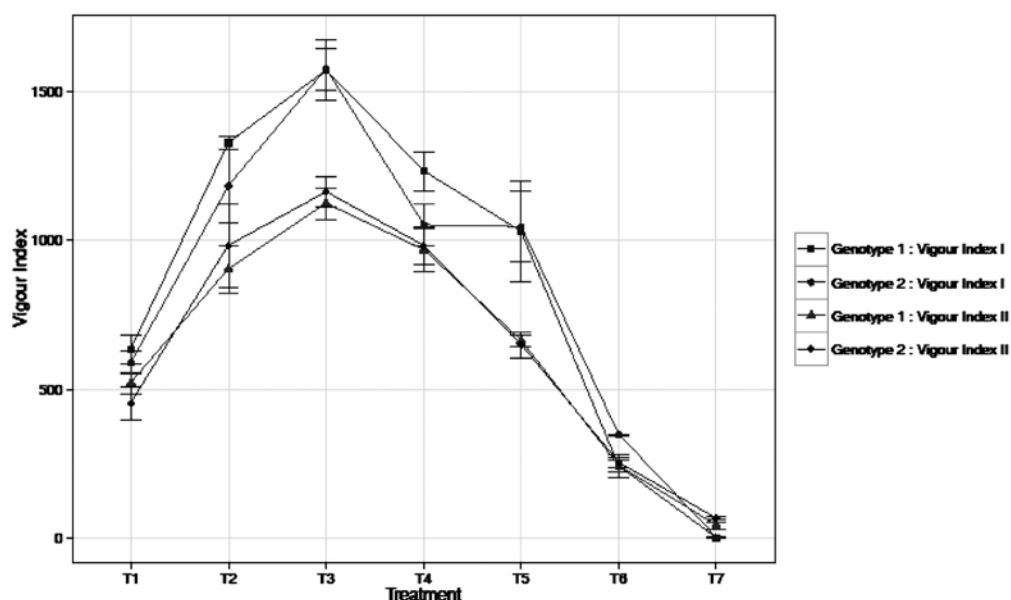


Fig. 1. Effect of temperature and immersion time on vigour indices in different *M. corchorifolia* genotypes. Vigour Index I- Seedling length (root + shoot) x germination %; Vigour Index II- Seedling dry weight x germination %

conductivity (EC) is another test to evaluate seed vigour. It was developed into a routine vigour test by measuring the leakage of electrolytes from seeds soaked in water in peas and French bean (Matthews and Bradnock, 6). In the present experiment, the extent of membrane damage due to different treatments was estimated and the differences in EC value after 24 hrs were observed to be highly significant as compared to control (Fig. 2) in both the genotypes. But the EC values recorded at 60 and 70°C for 10 and 20 min. was comparatively lesser than the seeds immersed at 80°C, irrespective of the soaking duration, which supports the hypothesis of softening the seed coat. Low EC

values are associated with high germination percent, vigour and uniform seedling growth. Therefore, the temperature and soaking duration of experiment becomes important. Similar results are reported by Pandita *et al.* (7) in fenugreek.

From the present study, it can be concluded that hot water treatment at 70°C for 10 min. is an optimum enhancer to promote maximum germination and safely overcoming the hard seed coat dormancy in *M. corchorifolia* without affecting the vigour and viability of the seeds. Results of this experiment may also serve as useful information because knowledge on seed germination requirement is an important factor for seed

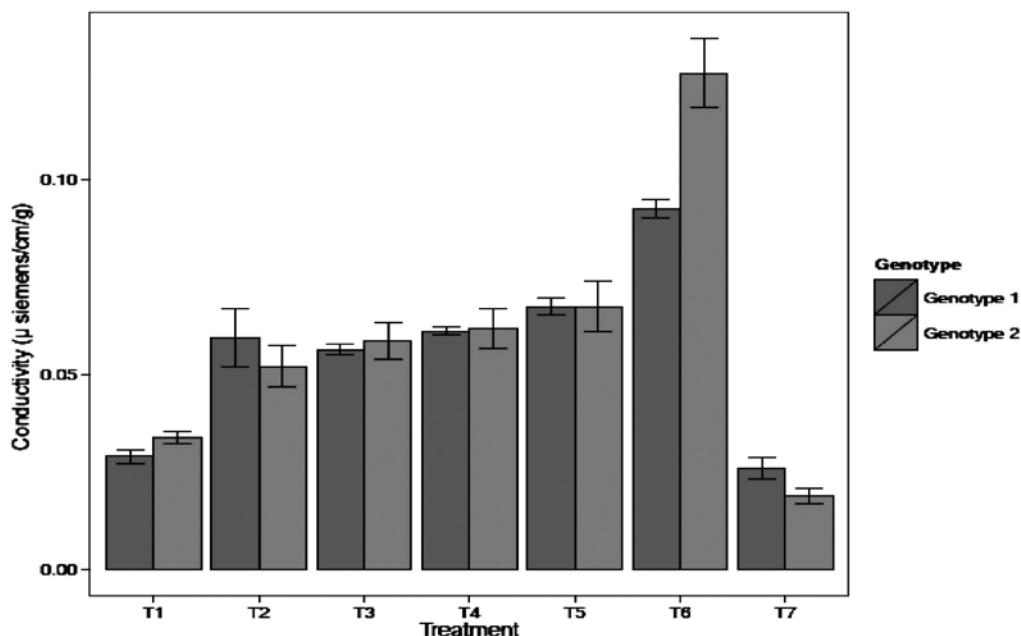


Fig. 2. Effect of temperature and immersion time on electrical conductivity of seed leachates in different *M. corchorifolia* genotypes.

testing, multiplication or regeneration and even their use in crop improvement programme.

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