

## Effect of various pre-treatments for breaking the dormancy of *Helicteres isora* Linn.

Mudasir Qadir and Fatima Khan\*

Govt. College of Science and Commerce Benazeer, Bhopal-462008 (India)

### Abstract

In the present investigation, seeds of *Helicteres isora* Linn. were subjected to various treatments to achieve early germination by breaking dormancy.

It was found that on 17<sup>th</sup> day of sowing, the germination percentage was 16, 81, 61, 9, 20, 17, 18, 18, 16, 40, 53, 27, 50, 32, 68 and 75 respectively in the seeds kept as control, and those treated with hot water, subjected to scarification, stratification, alternate high and low temperature, KNO<sub>3</sub>, thiourea, kinetin, GA<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, pre-soaking for 6 hours, electric current, mechanical injury, coumarin, brassinolide and IAA. Pre-treatment with hot water not only induced 65% germination on 3<sup>rd</sup> day but was also responsible for 81% germination on the 17<sup>th</sup> day after sowing. Thus, the best option for the germination of *Helicteres isora* seeds is pre-treatment with hot water.

The seeds of some plants easily germinate after sowing in nature but the seeds of a number of plants do not germinate easily and exhibit dormancy for varying period of time. The dormancy may be due to internal factors or may be due to external factors. Certain plants may immediately germinate after the harvest, it can be best exemplified by the seeds of *Pisum sativum*, which sometimes germinate in the fruit itself which is still on the plant, a phenomenon known as vivipary. However, sometimes the dormancy period is very prolonged and can take months together for germination. This is true for the seeds of *Malus domestica* which has a hard

seed coat and *Entada gigas* which has a very thick seed coat and do not germinate easily.

It is a straggling shrub of the family Sterculiaceae. The plant grows wild on hilly tracts in Bhopal and surrounding areas. It is commonly met with on the slopes of Shyamla Hills in Vanvihar Bhopal. The fruits of this plant are known as *Maror phali* and are taken in high esteem in indigenous medicine as a remedy for gastrointestinal disorders. The germination of seeds under natural conditions is often very poor.

So, in the present study, seeds of this

plant were tested for their germination potential and shortening of dormancy period. Initial studies exhibited that there was only 16% germination till 17<sup>th</sup> day of sowing recorded under untreated seeds. Therefore, it was thought imperative to undertake this investigation to find out the substance that can break the dormancy of this plant. The seeds were subjected to various treatments which are mentioned in table-1.

According to Berlyn<sup>3</sup>, germination is a sequential series of morphogenetic events that result in the transformation of an embryo into a seedling. The seeds of every plant have the capability to germinate but their germination is affected due to some factors, such as seed coat, hard seed coat, rudiment embryo, over-ripening, presence of plant growth inhibitors, due to absence of water, oxygen and due to unfavourable conditions. Dormancy of seeds is due to external factors or due to internal factors. When it is caused due to internal factors, it is called as true dormancy or innate dormancy or primary dormancy. And when it is caused due to external factors, it is called as imposed dormancy or quiescent dormancy or secondary dormancy. Both of these primary and secondary dormancy influences are mutually dependent and can not be singled out. True dormant seeds do not germinate even if they are provided with suitable environmental factors. Secondary dormant seeds may germinate immediately after shed off. After some storage, they fail to germinate and thus exhibit secondary dormancy. Some seeds such as *Brassica alba*, *Ambrosia tripolia* and *Xanthium pennsylvanicum* exhibit secondary dormancy. Secondary dormancy is opposite to after ripening.

Presence of high carbondioxide concentration, absence of light and very high or low temperature induce the secondary dormancy.

A number of techniques are available for breaking the dormancy of seeds, such as; scarification, exposure to light, alternating high & low temperatures, stratification, impaction, pressure, electric current, pretreatment with coumarins, kinetin, GA<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, thiourea, KNO<sub>3</sub> and hot water.

Studies on germination and dormancy of seeds have been carried out by various workers on different types of species. These include; the studies of Shul,<sup>34</sup> on the oxygen minimum and the germination of *Xanthium* seeds. A detailed account of seed dormancy mechanics was given by Crocker<sup>8</sup>. Influence of low temperature in improving germination percentage was found out by Conville, (1920). Similarly, alternating temperatures to break the dormancy was used by Harrington<sup>19</sup>. Morinaga, (1926) has studied the germination of seeds under water.

Davies,<sup>11</sup> used high pressure to achieve higher seed germination. Denny & Stanton<sup>13</sup> suggested chemical treatments for breaking the seed dormancy. Joseph,<sup>21</sup> investigated the germination and vitality of birch seeds. Barton,<sup>2</sup> investigated on coniferous seeds. In 1936, Crocker investigated the effect of visible spectrum upon the germination of seeds and fruits. In 1938, Crocker<sup>10</sup> also gave an account of life-span of seeds.

Chouard,<sup>7</sup> has investigated vernalization and its relation to dormancy. Experimental

induction of dormancy in *Betula pubescens* was investigated by Eagles & Wareing<sup>16</sup>. Evanari,<sup>17</sup> has studied the physiology of seed dormancy, after ripening and germination. Ribosome and enzyme changes during maturation and germination of the castor bean seeds was investigated by Marre<sup>26</sup>. Effects of light, temperature and their interaction on the germination of seeds was investigated by Toole<sup>38</sup>.

Hayes & Klein,<sup>20</sup> investigated special quality influence of light during development of *Arabidopsis thaliana* plants in regulating seed germination. Bewley and Black<sup>4</sup>, studied the physiology and biochemistry of seeds. Isoenzymes of sugar phosphate metabolism in endosperm of germinating castor beans were studied by Nishimura<sup>31</sup>. Seed germination and dormancy have been studied by Bewley<sup>5</sup>. Improvement of seed germination in *Asparagus racemosus* has been reported by Gupta, *et al.*,<sup>18</sup>.

Effect of pre-sowing treatment on seed germination of *Babchi* (*Psoralea corylifolia*) and *Senna* (*Cassia angustifolia*) in nursery has been reported by Koppad and Umarbhadsha<sup>24</sup>. Seed germination behaviour of *Asparagus racemosus* (*Shatavari*) under *in-vivo* and *in-vitro* conditions has been investigated by Raghav and Kaser<sup>32</sup>. Siva,<sup>36</sup> *et al.*, have studied the enhanced seed germination of *Psoralea corylifolia* L. by heat treatment. Musara, *et al.*,<sup>29</sup> have investigated the evaluation of different seed dormancy breaking techniques on Okra (*Abelmoschus esculentus* L.) seed germination. Asha and Illa<sup>1</sup> have studied the effect of seed direction

and growth media on *in vitro* seed germination and seedling establishment of *Pterocarpus marsupium*.

Cantoro, *et al.*,<sup>6</sup> have reported seed dormancy QTL identification across a *Sorghum bicolor* segregating population. Dave, *et al.*,<sup>11</sup> have investigated the regulation of *Arabidopsis thaliana* seed dormancy and germination by 12-oxo-phytodienoic acid. *Entada phaseoloids* seed dormancy and germination: implications for conservation and restoration has been reported by Deepa and Shinde<sup>13</sup>. The effect of the use of temperature on the breakage of dormancy and the subsequent performance of rice (*Oryza* spp.) has been investigated by Doku, *et al.*,<sup>15</sup>. Transcriptome analysis of seed dormancy after rinsing and chilling in ornamental peaches (*Prunus persica*) has been investigated by Kanjana, *et al.*,<sup>22</sup>.

Effect of different pretreatments and seed coat on dormancy and germination of seeds of *Senna obtusifolia* has been studied by Mensah and Ekeke<sup>27</sup>. Mishra,<sup>28</sup> has investigated the effect of temperature and light on the seed germination of *Sida cordifolia*. Redwood, *et al.*,<sup>33</sup> have reported seed longevity and dormancy state in a disturbance-dependent forest herb, *Ageratina*. Germination pretreatments to break hard-seed dormancy in *Astragalus cicer* L. has been studied by Statwick<sup>37</sup>.

Effect of various dormancy breaking treatments on seed germination, seedling growth and seed vigour of medicinal plants has

investigated by Warghat, *et al.*,<sup>40</sup>. Zohra, *et al.*,<sup>41</sup> have reported the effect of salicylic acid on germination of *Ocimum gratissimum* seeds induced into dormancy by chlormequat. The release of dormancy, a wake-up call for seeds to germinate has reported by Nee, *et al.*,<sup>30</sup>.

Healthy seeds of *Helicteres isora* were collected from the seed market (Bhopal). The seeds were washed with running tap water three to four times and once surface sterilized with 0.1% H<sub>2</sub>CL<sub>2</sub> solution for 5 minutes to remove the surface adhering microbes. After

surface sterilization, the seeds were again washed with double distilled water. Uniform sized seeds were then transferred to sterilized Petri Plates provided with filter paper pads. Three replicates of treated and control seeds were kept for germination studies. The filter paper pads were moistened as and when needed. The emergence of radical was taken as germination.

At the end of 17<sup>th</sup> day, the germination percentage was 16, 81, 61, 9, 20, 17, 18, 18, 16, 40, 53, 27, 50, 32, 68 and 75 respectively in

Table-1 showing the effect of various treatments on the germination percentage of *Helicteres isora*

D A S → Treatment ↓	3 <sup>rd</sup> day	5 <sup>th</sup> day	7 <sup>th</sup> day	9 <sup>th</sup> day	11 <sup>th</sup> day	13 <sup>th</sup> day	15 <sup>th</sup> day	17 <sup>th</sup> Day
<b>Control</b>	0	0	8	8	11	16	16	16
Hot water	65	68	72	75	79	81	81	81
Scarification	31	37	45	51	57	60	61	61
Stratification	0	3	3	6	7	9	9	9
Alt. high & low temp.	6	9	10	10	14	19	20	20
KNO <sub>3</sub>	7	9	11	11	13	16	17	17
Thiourea	8	8	11	14	15	18	18	18
Kinetin	10	12	15	18	18	18	18	18
GA <sub>3</sub>	4	9	9	13	14	15	15	16
H <sub>2</sub> SO <sub>4</sub>	16	19	23	27	34	39	40	40
Presoaking	25	31	38	43	47	51	53	53
Coumarin	3	11	16	21	28	32	32	32
Electric current	3	4	9	17	21	26	27	27
Brassinolide	11	27	38	47	56	65	67	68
Mechanical injury	26	31	37	44	49	50	50	50
IAA	15	39	43	54	68	73	75	75

the seeds kept as control, and those treated with hot water, subjected to scarification, stratification, alternate high and low temperature, KNO<sub>3</sub>, thiourea, kinetin, GA<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, pre-soaking for 6 hours, electric current, mechanical injury, coumarin, brassinolide and IAA. Pre-treatment with hot water not only induced 65% germination on 3<sup>rd</sup> day but was also responsible for 81% germination on the 17<sup>th</sup> day after sowing. It was followed by 75% germination of IAA treated seeds and 68% germination was attained in the seeds which were treated with brassinolide. Thus, the best option for the germination of *Helicteres isora* seeds is pre-treatment with hot water as under natural condition it is very poor as is evident from table 1 which shows only 16% germination on 17<sup>th</sup> day after sowing in the seeds which were untreated and kept as control.

The extent of the seed dormancy can be guessed from table 1 which shows that even after 17 days of sowing, only 16% seeds kept as control could germinate. Among all the treatments to break the seed dormancy of *H. isora*, pretreatment with hot water facilitated the germination to the tune of 65% on the 3<sup>rd</sup> day and 81% on the 17<sup>th</sup> day from the day of sowing. By hot water pretreatment, there is softening of the seed coat which becomes more permeable to water and air for stimulating germination. There was 75% and 68% germination recorded respectively for the seeds treated with IAA and brassinolide on 17<sup>th</sup> day after sowing. There was 15% and 11% germination respectively under IAA and brassinolide treatment on the 3<sup>rd</sup> day from sowing. Thus, for achieving maximum germination of *H. isora* seeds, pretreatment of seeds with hot water is the best option which is also in conformity with Venudevan *et al.*,<sup>39</sup>,

Khakpor *et al.*,<sup>23</sup> and Kumar *et al.*,<sup>25</sup>. Moreover, pretreatment with hot water is not expensive and therefore could be used for achieving higher germination percentage of this medicinally important plant.

#### References :

1. Asha, P. and P. Illa (2016). *International Journal of Plant, Animal and Environmental Sciences*, 6 (2): 139-144.
2. Barton, L.V. (1930). *An. J. Bot.*, 17 : 88-115.
3. Berlyn, G.P. (1972). Seed germination and morphogenesis pp. 223-313 in T.T. Kozlowski (ed.) *Seed Biology* Vol. I. Academic Press In.
4. Bewley, C. C. and M. Black (1978). *Physiology and biochemistry of seeds in relation to germination. Vol I Development, Germination and growth. Springer-verlag Berlin. Heidelberg, New York.*
5. Bewley, J. D. (1997). *The Plant cell*. 9: 1055-1060.
6. Cantoro, R., L. G. Fernandez, G. D. L. Cervigni, M.V. Rodriguez, J.O. Gioco, N. Paniego, R. A. Heinz and R.L.B. Arnold (2016). *Euphytica*, 211 (1): 41-56.
7. Chouard, P. (1960). *Ann. Rev. Plant Physiol.*, 11 : 191-238.
8. Crocker, W. (1916). *Amer. Journal of Botany*, 3: 99-120.
9. Crocker, W. (1936). Effect of visible spectrum upon the germination of seeds & fruits In : *Bio. Effect of Radiation*, New York 2 : 79-827
10. Croker, W. (1938). *The Botanical Review* 4 : 235-274.
11. Dave, A., F. E. Vaistij, A. D. Gilday, S. D. Penfield and I.A. Graham (2016). *Journal of Experimental Botany*, 67 (8): 2277-2284.

12. Davies, P.A. (1928). *Am. J. Botany* 15: 433-436.
13. Deepa, C. and N. W. Shinde (2016). *International Journal of Advanced Research*, 4 (5): 11-16.
14. Denny, F.E. and E.N. Stanron (1928). *Am. J. Bot.* 15: 327-336.
15. Doku, G. D., M. K. Glover, E. K. Glover and K. P. A. Dartey (2016). *International Journal of Plant Science and Ecology*, 2 (1): 1-9.
16. Eagles, C. F. and P.F. Wareing (1963). *Nature*, 199: 874-875.
17. Evanari, M. (1965). *Proc. Internat. Seed Test Assoc.*, 30: 49-71.
18. Gupta, S., A. Kumar and S. N. Sharma (2002). *Journal of Herbs, Spices & Medicinal Plants*, 9 (1): 3-9.
19. Harrington, G.T. (1923). *J. Agric Res.* 23: 295-332.
20. Hayes, R. G. and W. H. Klein (1974). *Plant cell physiol.*, 14: 643-53.
21. Joseph, H.C. (1929) *Bot. Gaz.* 87 : 127-151.
22. Kanjana, W., T. Suzuki, K. Ishii, T. Kozaki, M. Ligo and K. Yamane (2016). *BioMed Central*, 17: 575.
23. Khakpor, Alaleh, Bibalani Ghassem Habibi and Khadijeh Mahdavi (2011). *Annals of Biological Research*, 2 (5): 52-55.
24. Koppad, A. G. and N. K. Umarbhadsha (2006). *Karnataka Journal of Agricultural Science*, 19 (2): 441-442.
25. Kumar Rita, N., Sudeshna Chakraborty and J.I. Nirmal Kumar (2011). *Asian J. Exp. Biol. Sci.*, 2 (1): 143-146.
26. Marre, E. (1967). *Current Topics Devel. Biol.*, 2: 75-105.
27. Mensah, S. I. and C. Ekeke (2016). *International Journal of Biology*, 8 (2): 77-84.
28. Mishra, S. (2016). *International Journal of Scientific and Research Publications*, 6 (1): 264-266.
29. Musara, C., J. Chitamba and C. Nhuvira (2015). *African Journal of Agricultural Research*, 10 (17): 1952-1956.
30. Nee, G, Y. Xiang and W.J.J. Soppe (2017). *Current Opinion in Plant Biology*, 35: 8-14.
31. Nishimura, M and H. Beevers (1981). *Plant Physiol.*, 67: 1255-1258.
32. Raghav, A. and P. K. Kasera (2012). *Asian Journal of Plant Science and Research*, 2 (4): 1 409-413.
33. Redwood, M.E., G.R. Matlack and C.D. Huebner (2016). *Seed Science Research*, 26: 148-152.
34. Rout, J.R., R. P.A.B. Das and S.L. Sahoo (2009). *American-Eurasian J. Agric. and Environ. Sci.*, 6 (6): 689-691.
35. Shul, Charles A. (1914). *Botanical Gazette* 57 : 64-69.
36. Siva, G., S. Sivakumar, G. Premkumar, T. Baskaran, T. Senthilkumar and N. Jayabalan (2014). *World Journal of Agricultural Research*, 2 (4): 151-154.
37. Statwick (2016). *Peer J.*, 4: 2621.
38. Toole, T.K. (1973). *Seed. Sci. & Technol.*, 1: 339-396.
39. Venudevan, B., S. Sundareswaran and A. Vijayakumar (2010). *Madras Agric. J.*, 97 (1-3): 31-32.
40. Warghat, A. R., B. Kumar and O. P. Chaurasia (2016). *Tropical Plant Research*, 3 (3): 508-516.
41. Zohra, E. S. F., H. Zakaria, M. Youssef, E. E. Hassan and A. J. Khalid (2016). *Internatioal Journal of Engineering Research and Science*, 2 (9): 150-154.