Effect of various pre-treatments for breaking the dormancy of *Mucuna pruriens* Bak.

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Abstract

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

It was found that on 17^{th} day after sowing, the germination percentage was 4, 32, 40, 17, 23, 27, 18, 28, 31, 35, 40, 17, 28, 74 and 82 respectively under untreated seeds and those subjected to hot water, scarification, stratification, alternating high and low temperature, KNO₃, thiourea, kinetin, GA₃, H₂SO₄, pre-soaking, electric current, coumarin, brassinolide and IAA. In this way, mechanical injury of the seeds of *M. pruriens* is the best option for achieving higher germination percentage. IAA is also a useful method as it induced 82% germination in the present study (table-1).

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.

M. pruriens is a shrub which uses its vines as support to climb and grow. When M. pruriens are young, they are covered with hair which disappears as the plant gets old. Its leaves are of various shapes like ovate, tripinnate and rhombus shape. The tips of the leaves are pointed and the sides are grooved. This plant grows well in tropical areas of India, West Indies and Africa. It is common to find purple, white and lavender flowers on Mucuna pruriens. Its seed pod causes severe itch by touching it. The pod is about 10 cm long and is covered with orange hairs. The itch caused by the pod is due to the presence of proteins; Serotonin and Mucunain. This plant is used as natural fertilizer and manure for the crops. It is also used as fodder because it has high fiber and protein content. Its beans are used on

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sportspersons to increase their muscle mass. These seeds also help to gain weight which is lost due to excessive exercise. In some countries, its beans are used as a substitute for coffee. It has been long used to increase the sexual libido in both men and women. Oral intake helps in promoting fertility and improves erection. Its seed are also used for treating intestinal gas, diarrhoea, cough, rheumatic disorder, muscular pain, diabetes, menstrual pain and tuberculosis.

Seeds of this plant were tested for their germination potential and shortening of dormancy period. Initial studies exhibited that there was only 4% germination till 17th 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 (1972), 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, overripening, 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, GA₃, H₂SO₄, thiourea, KNO₃ 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 $Shul^{28}$ on the oxygen minimum and the germination of *Xanthium* seeds. A detailed account of seed dormancy mechanics was given by Crocker⁷.

Davies¹⁰ used high pressure to achieve higher seed germination. Barton² investigated on coniferous seeds. In 1936, Crocker⁸ investigated the effect of visible spectrum upon the germination of seeds and fruits. Chouard,⁶ has investigated vernalization and its relation to dormancy. Experimental induction of dormancy in *Betula pubescens* was investigated by Eagles & Wareing,¹³. Evanari,¹⁴ 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,¹⁹. Effects of light, temperature and their interaction on the germination of seeds was investigated by Toole³¹.

Hayes & Klein,¹⁶ investigated special quality influence of light during development of *Arabidopsis thaliana* plants in regulating seed germination. Bewley and Black,³ studied the physiology and biochemistry of seeds. Isoenzymes of sugar phosphate metabolism in endosperm of germinating castor beans were studied by Nishimura,²⁴. Seed germination and dormancy have been studied by Bewley,⁴. Improvement of seed germination in *Asparagus racemosus* has been reported by Gupta, *et al.*,¹⁵.

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,¹⁸. Seed germination behaviour of *Asparagus racemosus* (*Shatavari*) under *in-vivo* and *in-vitro* conditions has been investigated by Raghav, and Kasera,²⁵. Siva, *et al.*,²⁹ have studied the enhanced seed germination of *Psoralea corylifolia* L. by heat treatment. Musara, *et al.*,²² have investigated the evaluation of different seed dormancy breaking techniques on Okra (*Abelmoschus esculentus* L.) seed germination. Asha and Illa¹ have studied the effect of seed direction and growth media on *in vitro* seed germination and seedling establishment of *Pterocarpus marsupium*.

Cantoro, et al.,⁵ have reported seed dormancy QTL identification across a Sorghum bicolor segregating population. Dave, et al.,⁹ 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,¹¹. 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., 12. Transcriptome analysis of seed dormancy after rinsing and chilling in ornamental peaches (Prunus persica) has been investigated by Kanjana, et al..¹⁷.

Effect of different pretreatments and seed coat on dormancy and germination of seeds of *Senna obustifolia* has been studied by Mensah, and Ekeke,²⁰. Mishra,²¹ has investigated the effect of temperature and light on the seed germination of *Sida cordifolia*. Redwood, *et al.*,²⁶ have reported seed longevity and dormancy state in a disturbancedependent forest herb, Ageratina. Germination pretreatments to break hard-seed dormancy in *Astragalus cicer* L. has been studied by Statwick³⁰. Effect of various dormancy breaking treatments on seed germination, seedling growth and seed vigour of medicinal plants has investigated by Warghat, *et al.*,³². Zohra, *et al.*,³³ 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.*,²³.

Healthy seeds of *Mucuna pruriens* 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_gCl_2 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.

Under control, there was no germination upto 7th day from the date of sowing and so was the case with rest of the treatments upto day 3 after sowing with the exception of mechanically injured seeds and the seeds subjected to scarification, brassinolide and IAA which respectively exhibited 8%, 18%, 11% and 15% germination on day 3. There was 0, 13, 19, 7, 9, 7, 8, 10, 8, 11, 13, 9, 45, 9, 46 and 39% germination on the 7th day after sowing respectively in the seeds kept as control, those treated with hot water, scarification, stratification, alternate high and low temperature, KNO₃, thiourea, kinetin, GA₃, H₂SO₄, pre-soaking, electric current, mechanical injury, coumarin, brassinolide and IAA. On the 17th day from the date of sowing, only 4% of the seeds kept as control could germinate whereas, 88% germination of the mechanically injured seeds was exhibited in the same time span. The germination percentage under rest of the treatments varied under various other dormancy breaking factors. Thus, on 17th day after sowing, the germination percentage was 32, 40, 17, 23, 27, 18, 28, 31, 35, 40, 17, 28, 74 and 82 respectively under hot water, scarification, stratification, alternating high and low temperature, KNO₃, thiourea, kinetin, GA₃, H₂SO₄, pre-soaking, electric current, coumarin, brassinolide and IAA. In this way, mechanical injury of the seeds of *M. pruriens* is the best option for achieving higher germination percentage. IAA is also a useful method as it induced 82% germination in the present study (table-1).

Though, there is heavy production of the seeds of this species but the germination is very poor (table-1). Under control, only 4% of seeds of this plant could germinate after 17th day of sowing. The seeds have a thick seed coat and do not allow the passage of water and air. In the present investigation, germination up to 88%, 82% and 74% respectively could be achieved in mechanically injured, IAA and brassinolide treated seeds. Mechanically injured seeds make room for the entry of water and air to facilitate germination

(131)

$DAS \rightarrow$								
Treatment	3 rd day	5 th day	7 th day	9 th day	11 th day	13 th day	15 th day	17 th Day
\downarrow								
Control	0	0	0	2	4	4	4	4
Hot water	0	3	13	17	26	32	32	32
Scarification	8	12	19	25	31	39	39	40
Stratification	0	2	7	13	13	17	17	17
Alt. high &								
low temp.	0	3	9	17	21	23	23	23
KNO ₃	0	5	7	15	20	25	27	27
Thiourea	0	3	8	16	16	18	18	18
Kinetin	0	4	10	19	22	26	27	28
GA ₃	0	3	8	21	24	31	31	31
H_2SO_4	0	5	11	20	27	33	35	35
Presoaking	0	6	13	24	32	39	40	40
Coumarin	0	0	9	15	21	28	28	28
Electric current	0	4	9	9	12	16	16	17
Brassinolide	11	32	46	54	67	74	74	74
Mechanical injury	18	29	45	57	63	87	88	88
IAA	15	27	39	52	61	82	82	82

Table-1 showing the effect of various treatments on the germination percentage of *Mucuna pruriens*

and both IAA and brassinolide have been found to promote germination in many leguminous plants by bringing some chemical changes in the germination inhibitors residing in the seed coat. Thus, for achieving better germination percentage of this medicinally important plant, mechanical injury of seeds is the best option.

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