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***Short Communication***

# SEED GERMINATION IN A MEDICINALLY IMPORTANT PLANT- *CASSIA AURICULALTA*

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## ABSTRACT
Being a highly medicinally important plant seeds of *Cassia auriculata* were used for plant tissue work in our lab. When normally sown the seed hardly germinated beyond 2-3%. Besides the germinated seeds when transferred to the tissue culture medium showed high degree of contamination. Therefore an alternative mechanism was devised where different percentages of sulphuric acid were used for better germination. More than 97% seeds germinated within 1 to 2 days in contrast to 2-3% when they were not treated with $H_2SO_4$. The germinated seedlings were directly used as material for tissue culture explants and proved most contamination free material for *in vitro* tissue culture work.

## INTRODUCTION
*Cassia auriculata* is a fast growing profusely branched, tall, evergreen shrub, generally 1.2-3.0 m in height. As mentioned in Wealth of India (1992) it is chiefly valued for the tannins present in the bark. The bark, known as Avaram bark or Tangeedu bark in commerce is one of the best available barks in India. It is the principal indigenous bark, used in the South Indian tanneries for crust tanning. The bark is astringent and useful for gargles in sore throat, rheumatism, in enemas, eye diseases and diabetes. Its decoction is given in stomachache and the juice of fresh bark in dysentery. The leaves are anthelmintic and good for ulcers, skin diseases and leprosy, their infusion possesses slight purgative property. Leaves ground with *Vigna mungo* and poppy seeds are applied to herpetic eruptions. The leaves contain a high percentage of nitrogen and potash and are therefore used as green manure in paddy-fields. They are also valuable for manuring alkaline lands and reclamation of soils. The pods are anthelmintic, emetic and useful in urinary discharges. The seeds are considered refrigerant and alexipharmic. They are used in chronic purulent ophthalmic and conjunctivitis, cough, asthma, gout, gonorrhoea, dysentery and diabetes. The roots are considered astringent, alexeteric and useful in skin diseases, asthma, thrist and urinary discharge. A decoction of the roots is used as a tonic. The root shows interferon like activity against Ranikhet disease virus. The flowers are used in throat troubles, urinary disorders and as astringent. An aqueous extract of the leaves and flower possesses hypoglycemic activity (Anonymous 1952). Recently it has been reported as a potent anticancer herb by Prasanna *et al.*, (2009). They reported *in vitro* anti-cancer effect of *Cassia auriculata* leaf extract in human breast adenocarcinoma MCF-7 and human larynx carcinoma Hep-2 cell lines. When normally sown the seed hardly germinate beyond 1-2%. Besides the germinated seeds when transferred to the tissue culture medium showed high degree of contamination. Therefore an alternative mechanism was devised where different combination of acid and water were used for better germination.

## MATERIALS AND METHODS
The plant, which is a small shrub, belongs to the family Leguminosae and subfamily Caesalpiniaceae. Seeds of *C. auriculata* Linn. were collected from Pushkar (Ajmer) during a field visit in 1999. Seeds were first sown in soil for germination. Seeds were also cultured on MS (Murashige and Skoog, 1962) and ½ MS media (without phytohormones). But in both the cases delayed seed germination of around 2-5% was seen. Explants taken from such seedlings and plantlets, after sterilization with 0.01% $HgCl_2$ showed poor and delayed morphogenetic response. Therefore a protocol was developed to get contamination free,

quickly germinating and better responding seedlings. About 200 seeds (taken in 500 ml autoclaved conical flask) were treated with varying percentages (10% to 70%) of $H_2SO_4$ for 1 to 15 minutes and then washed thoroughly with autoclaved distilled water for 7 to 10 times. Seeds were then completely dipped in water. Only double distilled autoclaved water was used. The flasks were plugged with cotton and kept overnight. On the next day, the seeds got swelled and after pouring off the water, seeds were sterilized with 0.1% $HgCl_2$ for about 4-5 minutes. Seeds were than washed thoroughly for 5-6 times with double distilled autoclaved water and kept for germination in preautoclaved conical flasks of 500ml capacity under aseptic conditions.

*In Vitro Study*

After 2 to 3 days of sprouting, explants including cotyledonary node having intact epicotyl and a portion of hypocotyl, were taken from the seedlings and cultured on MS media supplemented with different combinations of auxins and cytokinins under aseptic conditions. Both the cotyledons were excised and removed from the cotyledonary nodal explants. In few explants however the cotyledonary leaves were left intact along with the cotyledonary node.

## RESULTS AND DISCUSSION

Seed sown directly into the soil showed 2-3% germination after 2-5 days of sowing. Seed cultured on MS (Murashige and Skoog, 1962) and ½ MS media (without phytohormones) also showed 2-3% of seed germination. But in case of 70% sulphuric acid treatment (for 10 minutes), more than 95% seeds germinated within 1 to 2 days of treatment in contrast to 2-3% when they were not treated with sulphuric acid. Since the seed coat of the plant was very hard and impermeable to water that's why sulphuric acid treatment was found very effective. Similarly the strong acid sterilized the seed surface simultaneously. That's why we saw least contamination during the tissue culture work (Negi *et al.,* 2011). Using $HgCl_2$ for sterilization also helped the process. Further removal of possible chemical inhibitors present in the seed coat of the Cassia seeds might be another reason for the increased seed germination as reported by Morris *et al.,* (2000) in *Grevillea* species. So, for species with hard seed coats, acid treatment can be used for better germination and getting least contaminated seedlings.

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