Abstract

***Ferula assa foetida* L., a species indigenous to Iran and Afghanistan, is an herbaceous and perennial medicinal herb and grows up to 2 m high. Asafetida is a very effective medicinal herb that acts mainly on the digestive system, cleansing and strengthening the gastro-intestinal tract. The pungently flavoured gum-resin that is obtained from the root is alterative, anthelmintic, antiperiodic, antispasmodic, carminative, deodorant, expectorant, laxative, sedative and stomachic. Most of the demand is being met through collection of large quantities of Asafetida from wild populations. Indiscriminate collection of this plant from the wild has posed a serious threat to its existence in the wild populations, especially when the plants are harvested well before seed set. The vegetative propagation of this species has not been described. The plant is conventionally propagated through seed. Moreover, propagation by seed is rather difficult due to seed dormancy. Consequently, Asafetida is classified as a vulnerable species in the Red Data Book of Iran. Hence there is a strong need for proactive understanding in the conservation, cultivation, and sustainable usage of this species for future use. In recent years, there has been an increased interest in tissue culture techniques which offer a viable tool for mass multiplication and germplasm conservation of rare, endangered and threatened medicinal plants. The efficacy of different treatments including various levels of GA_3, chilling, soaking with running water and combination treatment of cold stratification together GA_3 for germination improvement was tested. Analysis of variance indicated that cold stratification and GA_3 concentration had significant effects on seed germination percentage and rate germination. Combined treatments chilling (4 ℃) for both periods of 30 and 60 days with other treatments were most effective in breaking dormancy. Among the combined chilling treatments, Maximum germination was obtained at combination treatment cold stratification (60 days) with 2000 ppm of GA_3 solution and minimum germination was at socking in running water and control treatments. Callus was induced from root, hypocotyl and cotyledon explants of asafetida seedlings on a medium with 2,4-D (0-2 mg l^{-1}) or NAA (0-2 mg l^{-1}) alone or in combination with BA ((0-2 mg l^{-1}) or KIN (0-2 mg l^{-1}). The medium supplemented with 1 mg l^{-1} NAA and 2 mg l^{-1} BA was the most effective (100%) for the proliferation of callus from root explants. The scarified seeds were transferred to MS medium with various concentration 2,4-D alone or in combination with BA for somatic embryogenesis. Among the different concentrations used, maximum frequency (59.6%) was obtained with MS basal medium within 8 weeks in the culture medium. Formation of shoots could be induced from calluses by culturing on MS medium containing 1-3 mg l^{-1} BA or KIN alone and or in combination with 0.2 or 0.5 mg l^{-1} NAA. BA proved more useful compared to KIN in shoot induction. Callus produced from hypocotyl showed maximum percentage of cultures regenerating shoots (81.1%), with 7.4 shoots per callus whereas, root derived calli showed less number on MS medium containing 1 mg l^{-1} BA and 0.2 mg l^{-1} NAA. Rooting of *in vitro* raised shoots was best induced on half strength MS supplemented with 2.5 mg l^{-1} indole-3-butyric acid (IBA) with highest percentage of shoot regenerating roots (88.8 %) , the highest root number (7.23 root per explant) and the root length (5.34 cm). The in vitro raised plantlets were acclimatized and transferred to soil.**

Keywords: Callus induction; Tissue culture; Medicinal plant; *Ferula assa foetida*; Regeneration; Root formation;

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