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***Research note :***

# *In vitro* Studies on callus induction and plantlet regeneration in Safflower (*Carthamus tinctorius* L.)

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Callus induction and regeneration in safflower (*Carthamus tinctorius* L.) cv. Sharda and PBN-12 was optimized using various explants such as hypocotyls, cotyledonary leaves roots and apical shoot buds. The cotyledonary leaves exhibited better response for callus induction as compare to roots and hypocotyls on MS supplemented with 5mgl$^{-1}$ NAA, 0.25mg$^{-1}$ BAP and 1% sucrose. The calli were maintained on the same medium. Of the two cultivars, Sharda showed better regeneration response for on MS medium supplemented with 1.8 mgl$^{-1}$ BAP, 0.5 mgl$^{-1}$ NAA and 2% sucrose. The carbohydrate concentration exhibited vital role in regeneration efficiency of the two cultivars. The regeneration protocol may be useful for engineering the plant for novel characters.

Safflower (*Carthamus tinctorius* L.) is an important oilseed crop of semiarid regions. It belongs to family compositae and is locally known as Kardi, Kusumbha and kusum. It has originated from Abyssinia and Afghanistan. The seeds of safflower yield oil, which is used for cooking. Since it is drying oil, it has industrial uses in the manufacture of paints, varnishes, waterproofing materials and plastic adhesives for Tissue culture is an important approach for improvement of plant variety. Safflower offers a tremendous scope for genetic improvement through the exploitation of tissue culture techniques. The circumstances obtaining for varietal improvements in conventional breeding method are solved by tissue culture techniques. Similarly it has been reported that the regeneration frequency in safflower is very high and it is possible through embryogenic and organogenic pathways (Sujatha, 2002). Several authors reported the mode of regeneration in safflower is through direct or indirect organogenesis (Tejovathi and Anwar, 1984; Orlikowska and Dyer, 1995; Sujatha and Suganya, 1996). Nikam and Shitole (1998) had conducted study on callus induction and *in vitro* plantlet regeneration of safflower (*Carthamus tinctorius* L.) cv. Bhima by using root, hypocotyl, cotyledon and leaf as explants. The earlier studies conclude that the regeneration protocols are cultivar specific, therefore the present investigation was designed to optimize the regeneration protocol for safflower cultivars namely Sharda (B.S.F.168-4) and PBN-12 (Parbhani kusum).

The seeds of both varieties were washed with detergent Tween-20 for 15 min. followed by thrice washing with tap water. The seeds were surface sterilized with sequential treatment of 70 % ethyl alcohol for 3 min. and 0.1 % mercuric chloride for 5 min. After both the treatments, the seeds were rinsed with sterile double distilled water to remove the traces of surface sterilizing agent. The surface sterilized seeds were subjected for germination on basal seed germination ½ MS medium in culture tubes, each containing 2 seeds. Culture conditions were 25°C temperature, relative humidity more than 50 % and 16 hrs photoperiod followed by 8 hrs darkness. The explants mainly cotyledonary leaf disc, hypocotyls, root and apical shoot bud were prepared and cultured for callus induction.

The explants were inoculated on modified MS (Murashige and Skoog, 1962) supplemented with NAA (3, 4, 5 mgl$^{-1}$), BAP (0.25 mgl$^{-1}$) and 1 % sucrose. The medium was gelled with 0.7 % agar after adjusting pH to 5.8. The media were sterilized by autoclaving at 121°C for 15 min. at 15 psi. The explants cultured were incubated in darkness for callus induction initiation.

The calli induced were cultured on MS supplemented with BAP (0.2, 1.5, 1.8 mgl$^{-1}$), NAA (0.4, 0.5 mgl$^{-1}$) and 2 % sucrose. The pH was optimized at 5.8 and media

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