Abstract

Tissue culture techniques coupled with genetic engineering methodologies serve as potential tools for crop improvement. Application of such techniques, however, largely depends on the availability of reliable plant regeneration protocol via organogenesis or somatic embryogenesis. Among oil seed crops, such protocol in safflower (*Carthamus tinctorius* L.) have not yet been fully achieved.

In the present study, direct adventitious shoot buds were induced on the cotyledonary leaves after two weeks of culture on MS medium containing BAP (0.5 - 4 mg/l) and 2 mg/l BAP was found to be optimum. Morphological features of regenerated shoots varied with BAP concentrations in the medium. Plant regeneration was found to be a two - step process. Regenerated shoots produced roots on half-strength MS medium supplemented with 0.8% agar, 3% sucrose and varying concentrations of auxin (NAA, IAA or IBA). Small capitula were also formed under *in vitro* conditions . *De novo* origin of shoot bud was confirmed from histological studies.

Out of the eight safflower cultivars tested, cv. S-144 proved to be the best organogenic line and optimization process was studied with this line only. Regeneration frequency varied with the age and position of explant segments. Five d old whole cotyledon or its proximal part was more responsive.

Among the other factors 0.8% agar, 3% sucrose and NO^-_3 as nitrogen source proved to enhance the process. Of the different ethylene inhibitors 25 µM $AgNO_3$ helped increasing the regeneration frequency.

Direct somatic embryogenesis was also achieved from cotyledons after 8-10 d of inoculation on a medium supplemented with auxin (NAA,IAA, IBA or p-CPA) and BAP combination. NAA at 2 mg/l along with 0.5 mg/l BAP was found to be optimum for the same. Regeneration of these embryos to complete plantlets took place on transfer to NAA containing half-strength MS nedium. The regenerated plants exhibited normal morphology and diploid chromosome complement.

Factors influencing somatic embryogenesis was also studied like that of the shoot organogenesis.Cv. Girna gave the maximum response. Histological studies showed unicellular origin of these embryos from epidermal cells. This direct origin was also confirmed through SEM studies. The embryogenic process was noted to accompany a decrease in starch and protein content in the cells involved.

Triglyceride (TG) content also showed changes and its highest amount was recorded from 18 d old culture when substantial number of cotyledonary embryos developed.