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Potential Regeneration Capacity of *Jatropha Curcas* L Leaf Explants after Chemical Treatment for Aseptic In vitro Regeneration

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suggest that the incorporation of two or more growth regulators in the culture medium results in its further proliferation and multiplication (Deore AC et al 2005). In the present study, we report an efficient and reproducible method for large-scale propagation of *Jatropha curcas* L using leaf explants from one month old plantlets grown in green house.

autoclaved distilled water for about 2-3 times, finally the explants were blotted on autoclaved Whatmanns paper. Subsequently, the leaves were cut into small pieces (5-7 mm) and placed with abaxial side on the MS medium.

3. Media and Culture Conditions

The medium used for callus development of leaf explants was CI-1 along with 30 g/L sucrose and 5.0 g/L agar. The

media was adjusted to pH 5.8 prior to adding agar and autoclaving. Explants of leaf were cultured in jam bottles containing 20mL of culture medium CI-1 for about 4 weeks for callus development. Then the callus generated in CI-1 were transferred to SR-1 which generated embryoids from callus and further developed to shoots within a period of 8 weeks through repeated subculture at regular intervals of time, approximately once in 15 days. Then the shoots were transferred to half strength MS medium along with 4.4µM IAA for rooting. Cultures were incubated at 25 ± 2ºC with a 16-h photoperiod of 35- 40 µ mol $.m^{-2}.s^{-1}$ provided by cool white fluorescent lights for callus induction and shoot regeneration while for root development the tissues were provided with 8-h light and 16-h dark photoperiod.

4. Results and Discussion:

4.1 Effect of Mercuric chloride on leaf explants and generation of callus on CI-1

Leaf explants treated with 0.1% Mercuric chloride resulted in absence of contamination, and those not treated with Mercuric chloride resulted in contamination. However when treated with 0.1% Mercuric chloride most part of the explant resulted in chlorosis because of harsh treatment of Mercuric chloride. However CI-1 was effective in generation of callus at certain specific locations of explant where chlorosis was not observed. Lesser concentration of BAP in CI-1 was responsible for generation of callus in leaf explants.

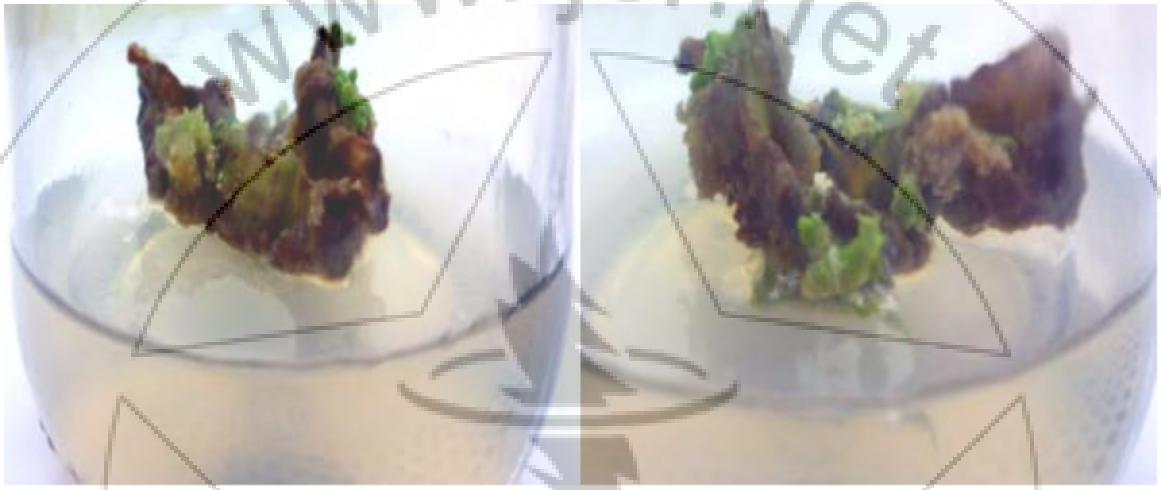

Figure 1: Development of callus from leaf explants of *Jatropha curcas* L on CI-1

4.2 Effect of SR-1 on callus for development of multiple embryoids and shoots.

Callus generated from leaf explant on CI-1 was transferred to SR-1 and cultured for about 8 weeks. The SR-1 was effective for generation of embryoids from callus. An average of 6 to 7 embryoids was generated out of each callus. Then the embryoids were sub cultured on to SR-1 for development of individual shootlets and later transferred to half strength MS medium along with 4.4µM IAA for rooting. Higher concentration of BAP in SR-1 was responsible for regeneration of embryoids from callus which later developed in to shootlets.Leaf has higher proportion of merismatic tissue which was responsible for generation of more number of embryoids from each explant.

Figure 2: Development of embryoids from callus of leaf explants *Jatropha curcas* L on SR-1.

Figure 3: Development of shoot lets from embryoids of leaf explants *Jatropha curcas* L on SR-1

4.3 Effect of IAA on shootlets for rooting

The shootlets generated on SR-1 media from leaf explants were transferred to half strength MS medium along with 4.4µM IAA. The shootlets showed rooting after 5 weeks of culture.

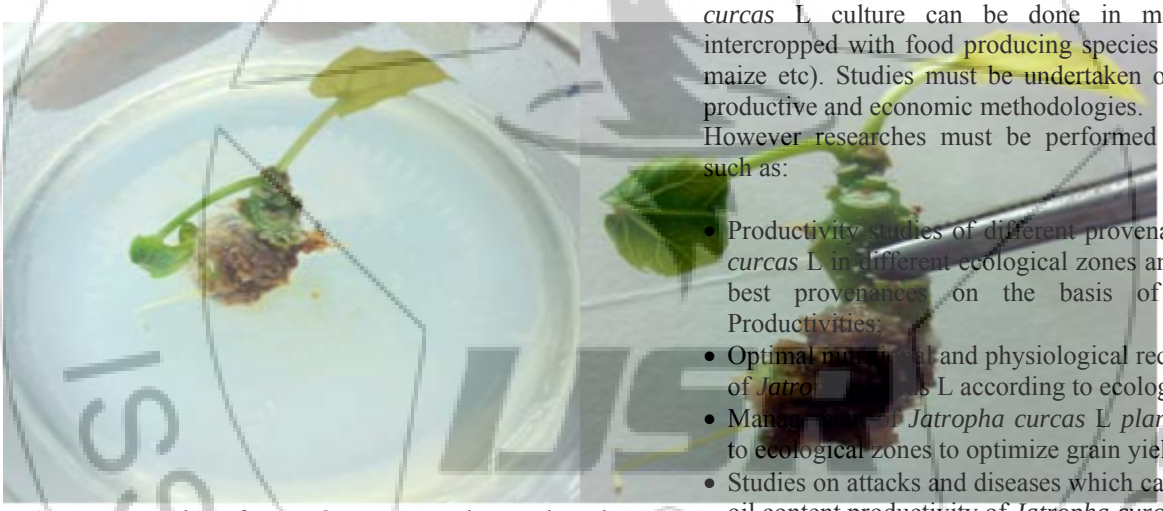

Figure 4: Rooting of *Jatropha curcas* L shoot cultured on SR-1 when transferred to half strength MS medium along with 4.4µM IAA.

The present study aims to characterize the explant that is most susceptible to chemical treatment and have maximum potential to generate shootlets for invitro regeneration of *Jatropha curcas* L plantlets .The development of appropriate techniques for *in vitro* culture and micropropagation of oil crops is necessary for germplasm collections, breeding program and mass propagation. The scope of the experiment to future is to develop a strategy for multiple embryoids production invitro from suitable explant of *Jatropha curcas* L for micropropagation.Since, *Jatropha curcas* L has gained much attention (Grimm 1996; Heller 1996; Henning 2000a; Pratt et al. 2002) for both non-oil producers and oil producers countries around the world.. The species has a high yield in oil, which can be used as fuel for diesel engines as well as for medical and insecticidal purposes (FACT Foundation, 2006).

5. Conclusion

Jatropha curcas L is becoming a new perennial cash crop for many regions of the imperative world. This makes it that special attention should be given to the way in which *Jatropha curcas* L plantations can be carried out. *Jatropha curcas* L culture can be done in marginal lands or intercropped with food producing species (millet, sorghum, maize etc). Studies must be undertaken on this feature for productive and economic methodologies.
However researches must be performed in many aspects such as:

- Productivity studies of different provenances of *Jatropha curcas* L in different ecological zones and selection of the best provenances on the basis of seeds and oil Productivities;
- Optimal nutritional and physiological requirements studies of *Jatropha curcas* L according to ecological zones;
- Management of *Jatropha curcas* L *plantations* according to ecological zones to optimize grain yield per unit area;
- Studies on attacks and diseases which can affect seeds and oil content productivity of *Jatropha curcas* L plantations;
- *In vitro* propagation using various methods (from axillary buds, tissue culture,); necessary for germplasm collection, breeding programs and mass propagation.
- Constitution of *Jatropha curcas* L gene banks (cold room, laboratory, greenhouse, fields);
- Gene viability tests and renewal of stocks.
- Disinfection of basic plant material (Samba Arona Ndiaye Samba et al).

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1. Molecular regulation of Santalol biosynthesis in *Santalum album* L published in Elsevier Science direct Gene 527 (2013) 642-648.
2. Invitro regeneration studies for multiple shoot induction through differential explants of *Jatropha curcas* L published in IJPRBS journal Volume 3 (4) : 606-612 August 2014.
3. Potential regeneration capacity of *Jatropha curcas* L leaf explants after chemical treatment for aseptic invitro regeneration accepted for publication in IJSR journal Volume 3, Issue 10 October 2014.