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Somatic embryogenesis in mono and poly embryonic cultivars

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Somatic embryogenesis in mono and poly embryonic cultivars

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Introduction

• Mango (Mangifera indica L.) is the most important fruit crop because of its wide adaptability, high nutritive value, richness in variety, delicious taste, excellent flavour, attractive appearance and commercial utility in India as well as in many part of the world.

• Conventional breeding in perennial crops is difficult and time consuming.

• A standard uniform protocol of regeneration is the prime and foremost prerequisite for not only improve the productivity, the production from the existing area but also development of transgenics in mango for various traits.
Why rootstocks?

• Day-by-day, breeders are introducing various new mango varieties but availability of good rootstock is still lacking.

• Most of the commercial mango varieties were sensitive to various biotic and abiotic stress.

• The quality of fruits and plant growth is affected by rootstock.

• According to various reports, there is a stagnation in the area and production of mango with an average productivity of 6 t/ha.

• In India, soil salinity is becoming a major problem and nearly 6.73 million hectares of area is under salt affected soils.

• Salinity is a serious limitation for bringing large geographical areas under mango cultivation particularly at early stages of growth.
• Polyembryonic cultivars are good rootstock because of moderate canopy, tolerant to draught, salt, environmental stress (wind, high temperature, air pollution), fungal disease (anthracnose, powdery mildew, gummosis) and hoppers.

• Polyembryonic mango genotypes (e.g. Kurakkan, 13-1) are reported to have salt tolerance.

• The mango variety '13-1' is a promising salt tolerant rootstock from Israel where the average productivity is 30-35 t/ha.

• Salt tolerant rootstocks need to be identified for increasing the area, production and productivity of mango.

• Besides, tissue culture can also help in clonal multiplication of root stock through nucellar embryogenesis.

• We report here genotype response (mono and poly embryonic) of mango in terms of nucellar embryogenesis.
Material and Methods

• 3.5 cm long immature fruits of mango were harvested and brought to laboratory.

• Fruits were opened and ovules were isolated under aseptic conditions and the ovules were bisected longitudinally.

• Intact ovular halves containing nucellar tissues were carefully scooped out and placed on induction medium.

• The developed embryos were kept under different temperature regime (5, 15 and 25 °C) to assess conversion efficiency of embryos.

• The complete regeneration medium used during the course of studies as follows:
## Nutrient formulation for somatic embryogenesis in mango

<table>
<thead>
<tr>
<th>Basal media</th>
<th>Induction and Proliferation</th>
<th>Conversion</th>
<th>Maturation</th>
<th>Germination</th>
<th>Root promotion</th>
</tr>
</thead>
<tbody>
<tr>
<td>B-5 Major salts + MS minor salts</td>
<td>0.05% Malt extract, 2736.9µM L-Glutamine, 4.52µM 2,4-D, 2.22µM BAP</td>
<td>0.05% Malt extract, 2736.9µM L-Glutamine</td>
<td>0.75µM Abscisic acid, 0.57µM IAA, 30.30 µM PEG</td>
<td>2.68µM NAA, 11.60µM Kinetin, 2736.9µM L-Glutamine, 2.88µM GA3</td>
<td>9.80µM IBA, 2.68µM NAA, 2.5 µM PVP, 13.78µM Spermidine</td>
</tr>
</tbody>
</table>
Monoembryonic cultivars of mango

Dashehari  Amrapali

Polyembryonic cultivars of mango

Bapakkai  Kurukkan  Moovandan
Results

• Nucellar embryogenesis was induced in different monoembryonic and polyembryonic cultivars of mango (*Mangifera indica* L) viz., Dashehari, Amrapali, Bapakkai, Kurukkan and Moovandan.

• Nucellus tissue excised from 3.5 cm long fruits of these cultivars developed pro-embryonic calli on modified MS medium supplemented with 4.52μM 2,4-D, 0.05% malt extract and 13.78μM spermidine.

• Among all the cultivars, polyembryonic cultivars gave higher level of somatic embryogenesis in comparison to monoembryonic.
• Among all polyembryonic cultivars, Bappakai produced 187.33 embryos per explant followed by Kurukkan 158.33 embryos per explant and Movandan 146.45 embryos per explant, whereas monoembryonic cultivars show comparatively lower level of somatic embryogenesis.

• Dashehari gave rise to 97.25 embryos per explant followed by Amrapali 82.33 embryos in 100 days under dark culture conditions.

• However, all the differentiated embryos proliferated on medium having low level of sucrose (4% w/v) and auxin (2.26μM 2, 4-D).

• Most of the proembryonic calli converted into heart shaped and cotyledonary embryos by reducing temperature to 15°C.
• Somatic embryos were matured on modified MS medium fortified with 0.38μM ABA, 0.57μM IAA and 30.30μM PEG.

• Matured somatic embryos germinated on MS medium supplemented with 2.68μM NAA, 11.60μM kinetin and 2736.9μM glutamine.

• Among all cultivars, Bappakai showed higher germination (39.33) followed by Kurukkan (29.97 %), Movandan (28.25%), Deshahari (26.45%) and Amrapali (25.25%).
Different stages of somatic embryogenesis in mango (*Mangifera indica* L).

A-B Somatic embryo induction from nucellar tissues, C-Proliferation and development of globular embryos, D- conversion of SE into heart shaped, E- early cotyledonary stage, F- late cotyledonary shaped embryo.

Cont...
Different stages of somatic embryogenesis in mango

Different stages of somatic embryogenesis in mango (*Mangifera indica* L). G-rooting and conversion into early stage of plants and H-rooted plants growing vigorously in liquid culture medium and I- plant in polyhouse during acclimatization.
Influence of Spermidine on production of somatic embryo in Mango cv. Kurrukan.
**Effect of temperature on conversion of embryos**

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>% embryos converted to heart shaped</th>
<th>% embryos necrosed</th>
<th>% embryos survived</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>0.00</td>
<td>36.33</td>
<td>63.67</td>
</tr>
<tr>
<td>15</td>
<td>40.94</td>
<td>8.66</td>
<td>91.34</td>
</tr>
<tr>
<td>25</td>
<td>34.76</td>
<td>10.66</td>
<td>89.34</td>
</tr>
<tr>
<td>SEM±</td>
<td>0.75</td>
<td>1.02</td>
<td>1.32</td>
</tr>
<tr>
<td>CD (P=0.05)</td>
<td>1.46</td>
<td>1.98</td>
<td>2.57</td>
</tr>
</tbody>
</table>

Effect of temperature on conversion of early heart shaped embryo into late heart shaped embryo after 30 days.
Conclusion

- Rootstocks offer the possibility of improving yields and managing tree size without increasing input costs.

- This is despite the availability of a diverse range of polyembryonic cultivars that could be used as rootstocks.

- By using good quality rootstocks, we can produce export quality mango even in waste/barren land.
Publications


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Thank you