PRODUCTION OF CANTALOUPE SEEDLINGS USING TISSUE CULTURE TECHNIQUE

By

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B.Sc. Agric. Sc. (Horticulture), Ain Shams University, 1998 M.Sc. Agric. Sc. (Vegetable Crops), Ain Shams University, 2004

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Approval Sheet

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ABSTRACT

Nesreen Ahmed Sabry Mohamed Mostafa Helal: Porduction of Cantaloupe Seedlings Using Tissue Culture Technique. Unpublished Ph. D. Thesis, Department of Horticulture, Faculty of Agriculture, Ain Shams University, 2009.

This work was carried out in tissue culture laboratory and greenhouses of the Arid Land Agricultural Research and Services Center, Horticulture Dept., Faculty of Agriculture, Ain Shams University, during 2007 – 2008 to investigate the effect of explant type, growth regulators and hybrids on cantaloupe seedling production through tissue culture technique, which can be planted in green house in comparison with *in vivo* (transplants from seeds) in order to evaluate the total production and yield under the prevailing conditions. Treatments included the usage of two types of explants (shoot tip and stem node) ,two cantaloupe hybrids: Sun Up F1 hybrid (Galia type) and NS-89 F1 hybrid (Charanty type) , two different cytokinins , i.e., benzyladenine (BA) and kinetin (Kin) at 0.0 (control), 0.25, 0.5, and 1.0 ppm added separately to the medium and the proliferated shoots were subcultured for 4 times.

The study was conducted in four main steps:-

Firstly: - Plant material preparation

Secondly: - *In vitro* propagation in four stages as follows

- > Establishment stage.
- ➤ Multiplication stage.
- > Rooting and acclimatization stage.

Thirdly: - *In vivo* evaluation.

Fourthly: DNA fingerprint using RAPD-PCR.

The obtained results can be summarized as follows:

A- Micropropagation of cantaloupe hybrids.

Adding the two tested cytokinins at 0.00, 0.25,0.5,1.0 mg/l to the media for culturing shoot tip gave 93% of direct shoot proliferation .But at the same time the two tested cytokinins at any concentration suppressed the development of root production .

The lowest concentration induced the less number of shoot and the gradual increase in concentration of both forms of cytokinins was correlated with an increment in number of shoot produced. Moreover, there was no significant effect in relation to the number of subculture within each concentration of the tested cytokinin on the number of shoots proliferated from cantaloupes either NS-89 F1 or Sun Up F1 hybrids ,also no significant differences were existed in between the four subcultures during multiplication stage.

B- Rooting and acclimatization

There is an increase in rooting percent with the increase in IBA concentration. The highest level (1.0 mg/l) produced the highest mean number of roots of the two examined hybrids.

C- Vegetative growth

The complete plants were transferred to the pags filled with sand, peat moss and vermiculate at 1:1:1(v: v: v) for acclimatization stage and all the plants were transferred into greenhouses for further vegetative and yield evaluation.

The micro propagated plants produced vigorous vegetative growth than the other plants produced from via seeds .

D- Early and total yield

The tissue culture plants were significantly superior than the conventional plants (seed, source) in producing higher early and total yield per plant. Also, showed that the tissue culture derived plants were significantly superior and produced significantly a higher average of fruit weight than the conventional derived plants of the two tested hybrids.

E-Fruit characters

A significant increase was observed in the TSS % and total sugar% of cantaloupe plants produced from conventional methods as compared with those obtained from tissue culture in the two tested hybrids. A significant increase was observed in the total acidity , vitamin (C) of cantaloupe plants produced from tissue culture as compared with those obtained from conventional methods in Charanty hybrid , that there were no significant differences between cantaloupe plants produced from the two methods in Galia hybrid.

From the previous results, it could be concluded that tissue culture derived plants were morphologically stable at establishment stage. It was observable that plants practice assimilation with efficient water and nutrient uptake, the root system grew quickly, leaf area expanded and the dry mass production increased. Thus, all these factors enable tissue culture plants to double the vegetative growth and increase yield more than those which were derived from conventional method.

F- DNA fingerprint using RAPD-PCR.

The consensus fingerprint profiling using the randomly amplified polymeric DNA (RAPD) marker is a useful and reliable method for establishing genetic identities of the two melon hybrids.

Finally, there was no genetic difference in plants propagated through plant tissue culture and conventional methods. The propagated plants from tissue culture technique were true to type to mother plant, there were no genetical differences found in morphogical and fruit characters.

Key Words: Tissue Culture, Propagation, Multiplication, Shoot Tip, Stem Node, Cytokinins, *Cucumis melo*, Cantaloupe

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ABREVIATION

BA 6-benzyladenine iP Isopenteny adenine TDZ Thidiazuron **IAA** Indole- 3- acetic acid Indole- 3- butyric acid **IBA** Gibberellic acid GA3 kinetin Kin \mathbf{CH} Casein hydrolysate **NAA** α-naphthalene acetic acid Deoxyribonucleic acid **DNA** l Liter MS Murashige and Skoog **PCR** Polymerase chain reaction Centimeter cm Gram g \mathbf{C} Centigrade **CON** Conventional TC Tissue culture

1-INTRODUCTION

(*Cucumis melo L.*) is commonly called melon, muskmelon or cantaloupe melon. It belongs to the Cucurbitaceae family, known as the ground family or by the more popular name cucurbits. The genus *Cucumis* consists of 32 species, several of which are cultivated. Thirty of the 32 species within *Cucumis*, belong to *Cucumis melo.L.*

Cantaloupe (*Cucumis melo* L.) has become one of the popular and favorite fruit to the Egyptian consumer due to its high nutritional value, reasonable price, availability in the markets and the wide utility either fresh or processed. Moreover, it has become of strong potentialities as an export commodity to the Arab and European markets. Therefore, cantaloupe is considered of great economic importance as a source of hard currency that is needed for our country.

Traditionally, cucurbits are propagated sexually by seed. The reat of propagation has depended on seed amount and quality. In addition, breeding systems using traditional method has taken long time to produce and multiply the new local hybrids.

Nowadays, tissue culture technique can replace the traditional nursery methods and was considered the prime method for reproducing all asexually propagated cultivars. Cloning *in vitro* can also be a tool to obtain disease- free plants, increase the genetic manipulation of separate chimeras and store plant material in gene bank. However, this technique still has not been developed so far in a large number of horticulture plants, specially vegetables. Therefore, further research efforts would have to be directed to such crops to gain the advantage of *in vitro* technique to produce mass of plantlets in a short period of time and to minor cost accounts (El Zeiny, 1997).

Due to the increasing the price of hybrid seeds year after year, the asexual propagation of *cucumis* ssp by tissue culture methods became of great benefit. However, a good micropropagation protocol could reduce the cost of hybrid seed production, which can account for 30 % of the total seedling cost (**Ahmad and Anis**, 2005).

The aim of this research was , therefore, to study the influence of explant types, growth regulators and hybrids on cantaloupe seedlings production through tissue culture technique, which can be planted in green house in comparison with *in vivo* (transplants from seeds) in order to evaluate the total production and yield under the prevailing conditions.