In-Vitro Propagation of Cinnamon (*Cinnamomum verum* Presl.) Using Embryos and *in Vitro* Axillary Buds

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***Abstract*—Embryo culture was developed in half strength MS medium in order to optimize the culture condition for axillary buds. 15% Clorox® for 20 min was very effective in minimizing pathogenic contaminants (100% non-contaminants) embryos as well as minimum browning. Embryonic axis with ½ of cotyledon portion was used as explants for *in-vitro* establishment and inoculated into the half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA to initiate *in-vitro* seedlings, giving maximum culture initiation (90%). 1g L^{-1} activated charcoal was effective for establishment of *in-vitro* culture, recording minimum browning effect (34.9 mean rank value on non-browning appearance), enhancing stem elongation (19.5mm height) and leaf initiation (2.06 leaves/plantlet) after 14 days of culturing. Treatment combination of 0.1mg L^{-1} NAA + 4.0m L^{-1} BAP + 1.0g L^{-1} activated charcoal in full strength MS medium was effective for adventitious root elongation on *in-vitro* micro-stem cuttings and given the highest root length (6.7cm) after 6 weeks of incubation period. Coir dust was the best potting medium for acclimatization giving maximum survival (90%). Therefore, the findings of the research could be used as a protocol for *in-vitro* propagation of Cinnamon (*Cinnamomum verum* Presl.) through *in vitro* axillary buds.**

***Index Terms*—axillary BUDS, Cinnamomum verum Presl., in-vitro propagation**

I. Introduction

Cinnamon (*Cinnamomum verum* Presl.) which belongs to the family Lauraceae, known as true Cinnamon or Ceylon cinnamon, is an economically important indigenous plant to Sri Lanka. Sri Lanka is the largest Cinnamon producer in the world and accounts for 65-70 percent of global production. Cinnamon cultivation in Sri Lanka is facing several constraints such as, unavailability of improved varieties, lack of suitable propagation techniques etc. Therefore to overcome these problems, a suitable propagation technique should be established which maintains the quality of the planting materials.

Conventional propagation is through seeds has several constraints and problems such as seeds are seasonal, recalcitrant [1] and unevenly matured [2] etc. Above all, homogenous plantation of Cinnamon would not be established with saplings of cross-pollinated seeds [3], [4]. Further, as other vegetative propagation methods are also seemed to be not much success [1] and therefore, *in-vitro* propagation would be good alternative for mass scale production of saplings for large scale plantations**.**

Enhancing of roots through air layering is claimed to be easy compared with stem cuttings [5], but this method has not been recommended for subsequent multiplication of selected materials on a mass scale production as the rate of multiplication is limited [6]. At this stage, clonal micro-propagation and germplasm conservation methods would be required [7]. Successful *in-vitro* propagation method for cinnamon has not been reported yet. Therefore the present study was conducted to develop a protocol for *in-vitro* micro-propagation of Cinnamon through *in vitro* grown axillary buds.

II. Materials and Methods

The research was carried out under two main streams; *in-vitro* propagation through embryos and isolated axillary buds grown *in vitro*.

A. Experiment 1: Establishment of In-Vitro Embryo Culture

The research was initiated through embryos excised from mature fresh fruits. Six months old evenly matured fruits were collected from a selected mother plant in the faculty farm, University of Ruhuna, Sri Lanka to use for producing initial explants of embryo culture. The experimental designs used for the parametric data were Completely Randomized Design (CRD) and two-factor factorial in Completely Randomized Design (CRD-Factorial). Data was analyzed using 'Statistical Analysis System' (SAS) computer package through 'Analysis of Variance' (ANOVA) procedure. Non-parametric data (ranked or count data) were analyzed using Kruskal Wallis rank test or Chi-square test.

Seeds with the removed pericarps were washed with Teepol® and running tap water. Then they were agitated slowly with Clorox® solution with different concentrations (15, 20 and 25%) and different exposure times (10, 15 and 20 minutes) and replicated 10 times. Then the treated seeds were cut into half of the cotyledons with the embryonic axis and inoculated into

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the half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA to initiate *in-vitro* seedlings. All the cultures were given total dark condition for first three days and then transferred to 16hrs photoperiod light regime of 1220 lux using fluorescence lamps. After 2 and 7 days of culture establishment, mean rank value of the browning appearance of plantlets and percentage of non-contamination were taken. Criteria for scores given according to the appearance (intensity of browning) of the explants that is increased browning appearance with more number of plus marks (No Browing – 1, + 3, ++ 5, +++ 7, ++++ 9).

Presence of contamination was considered as Yes or No. Collected rank data (browning) under non-parametric scores were analyzed using 'Kruskal Wallis' rank test, while the count data (contamination) were analyzed using 'Chi-square' test under categorical data analysis technique.

B. *Experiment 2: Effect of Different Treatments on Growth Parameter of In-Vitro Raised Shoots*

Shoots excised from the *in-vitro* seedings were cultured into the half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA and tested different antoxidents; 1g L^{-1} Polyvinylpyrolidone (PVP), 1g L^{-1} of Activated Charcoal (AC), 1g L^{-1} Ascorbic Acid (AA), 1g L^{-1} of Citric Acid (CA) and control (without any antioxidant). Since AA and CA are heat liable, they were filter sterilized and add to the autoclaved culture media. Browining appearence, number of shoots per plantlet, height of the main stem and number of leaves per plantlet were taken as growth parameters. Each treatment was replicated 10 times. The completely randomized design was used to analyse the data except non browining. Data were converted to square root transformation for analysis. Collected rank data under non-parametric scores were analysed using Kruskal Wallies' rank test.

C. *Experiment 3: Adventitious Root Induction/Elongation of In-Vitro Micro Shoots*

Embryonic axis with half of cotyledons of surface sterilized seeds were used to raise *in-vitro* seedlings on half strength MS hormone-free medium. After two months of establishment, the basal root portion of the *in-vitro* raised seedlings was removed and approximately 3cm height stem cuttings were taken for induction of adventitious roots. Then they were cultured in full strength MS basal medium incorporated with 1.0g L^{-1} activated charcoal with 16 different combinations of NAA (0.1, 0.2, 0.3 and 0.4mg L^{-1}) and BAP (1, 2, 3 and 4mg L^{-1}) concentrations.

The cultures were kept at 23±2℃ for 16hrs photoperiod light regime (1220 lux) and sub culturing was not practiced. Each treatment was replicated 20 times. Length of the roots was recorded at 4, 6 and 8 weeks of culturing. The two factor factorial with Completely Randomized Design (CRD-factorial) was used to analyze the data.

D. *Experiment 3: Acclimatization of Cinnamon Plantlets*

Successfully rooted *in-vitro* raised micro shoots were transferred into three different potting media (coir dust only, coir dust: sand 1:1 and top soil only) for acclimatization. Transparent plastic pots containing sterilized potting media covered with the same empty pots were used to maintained >80% RH for at least 2 weeks and then gradually acclimatized for the field condition. Each treatment was replicated 10 times. After 6 weeks, survival rate was recorded.

III. Results and Discussion

A. *Experiment 1: Establishment of In-Vitro Embryo Culture*

There were no significant differences among the means of the treatments after 2 days of culturing (Fig. 1). In the 7 days of culturing, means were significantly different from one another. Among them, the lowest mean rank value/minimum browning effect (23.5) was observed in 15% Clorox® with 10 minutes and 15 minutes exposure times, while the highest mean rank value/maximum browning effect (69.9) was observed in 20% and 25% Clorox® for 20 minutes exposure time (Fig. 1).

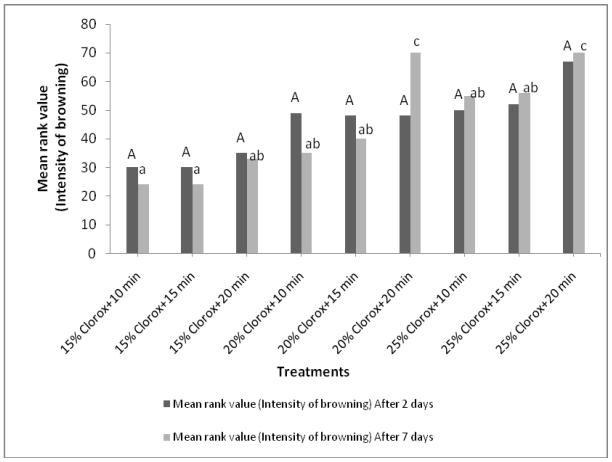

Figure 1. Effect of different concentration (15, 20, 25%) and exposure time (10, 15, 20 min) of surface sterilent (Clorox®) on browning of isolated embryos after 2 and 7 days of culture period in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA. Means on the collumn represent by the same letter are not significantly different at 5% probability level

Plate 1. Browning of isolated embryos after 7 days of culture period in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA

It was revealed [8] that the effect of browning is increased with increase concentration of the surface sterilent used and with the exposure time. Results of the present experiment also showed similar behavior. Browning of cinnamon explants was increased with increasing concentration of surface sterilent and exposure time (Fig. 2).

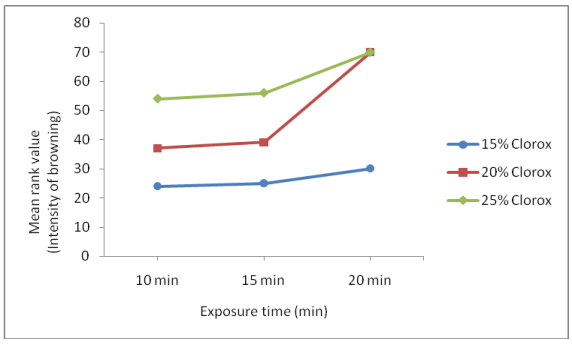

Figure 2. Change of browning level of isolated embryos against the concentrations of Clorox® and its exposure times after 7 days of culturing in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA

According to the Fig. 3, all the treatments, except 15% Clorox® with 10 minutes and 15 minutes exposure time, have given the maximum percentage (100%) of non-contaminants. Only these two treatments were given lower percentage (80%) of non-contaminants or in other words 20% of bacterial and fungal contaminations. It may be due to use of surface sterilent in mild concentrations with limited exposure time. These concentrations and exposure times may not be optimum to sterilize the material.

Considering browning and contamination of explants, 15% Clorox® with 20 minutes exposure time was the most promising treatment for surface sterilization of zygotic embryos of cinnamon. It was given reasonable mean rank value (33.1) of browning appearance after 7 days of culturing with no contamination. Therefore, it would be appropriate to use 15% Clorox® with 20 minutes exposure time for minimizing browning effect of tissues and to achieve 100% aseptic cultures ensuring subsequent *in-vitro* establishment of cinnamon embryo cultures.

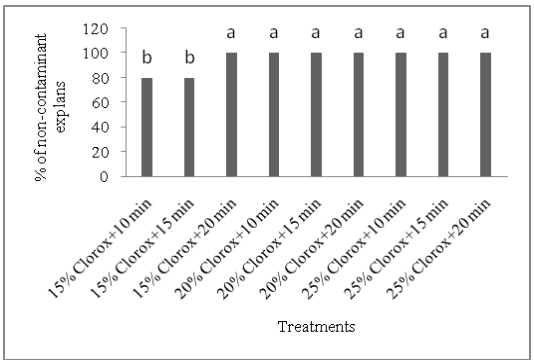

Figure 3. Effect of different levels of Clorox® and its exposure times on % of non-contaminated isolated embryos after 7 days of incubation period in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA.
Means on the collumn represent by the same letter are not significantly different at 5% probability level.

Plate 2. Bacterial contamination in isolated embryo after 7 days of culture period in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA.

Plate 3. *In-Vitro* cinnamon seedlings raised from isolated embryos in half strength MS basal medium supplemented with 1.5mg L^{-1} BAP + 0.2mg L^{-} IAA.

Similarly, It was reported [9] that the most common cleaning agent used to disinfect woody explants in 'bleach' in various concentrations (pure house hold bleach is 5% sodium hypochlorite), used alone or in combination with other disinfectants. Furthermore, it was mentioned that the most commonly used concentrations of bleach solutions are 5%, 10% and 20% concentrations. It was reported [10] that 2% sodium hypochlorite (30 min) can be used for surface sterilization of Cashew seeds which can be used for *in-vitro* embryo culture and on the other hand the use of domestic bleach at high concentrations (30-50%) for 10-30 min, is beneficial for surface sterilizing Cashew seeds, and it resulted 66% of the cultures, free from contamination [11]. This proves that cinnamon also respond similarly like other woody plants in terms of surface sterilization of embryos using Clorox.

B. Experiment 2: Effect of Different Treatments on Growth Parameters of In-Vitro Raised Shoots

Embryonic axis with ½ of cotyledon portion was used as explants for *in-vitro* establishment and inoculated into the half strength MS basal medium supplemented with autoclaved 1.5mg L^{-1} BAP + 0.2mg L^{-1} IAA to initiate *in-vitro* seedlings, giving maximum culture initiation (90%). 1g L^{-1} activated charcoal was effective for establishment of *in-vitro* culture, recording minimum browning effect (34.9 mean rank value on non-browning appearance), enhancing stem elongation (19.5mm height) and leaf initiation (2.06 leaves/plantlet) after 14 days of culturing.

C. Experiment 3: Adventitious Root Induction/Root Elongation of In-Vitro Micro Shoots

Results revealed that (Fig. 4) the individual effect of BAP on root elongation of *in-vitro* micro shoots was not significant (P>0.05). However, the treatment combination of highest BAP level (4mg L^{-1}) with lowest level of 0.1mg L^{-1} NAA in the medium was given the highest root length. It was mentioned that BAP, the most frequently used cytokinins, inhibit root formation and therefore left out from rooting media [12]. But, in this experiment, the reason for the effectiveness of highest concentration of BAP on root elongation may not be considered as a main effect but as a combined effect with NAA with the presence of 1.0g L^{-1} activated charcoal in the medium.

Similarly, low auxin concentration in the medium improved rooting in *Betula species* [13]. Likewise, it was reported [14] that, rooting of nodal and shoot tip explants of Cashew was observed in MS basal medium containing 0.3% activated charcoal and NAA in the culture medium. Moreover, they have mentioned that cultures in rooting media often benefit from the addition of charcoal, which appears to help to remove residual cytokinine. Further, the darkness of charcoal in the rooting area may also be a beneficial factor. Likewise, it was reported that, rooting of Cashew nodal segments was effective in MS medium containing 2.0mg L^{-1} NAA + 2.0mg L^{-1} IBA + 1g L^{-1} activated charcoal [15].

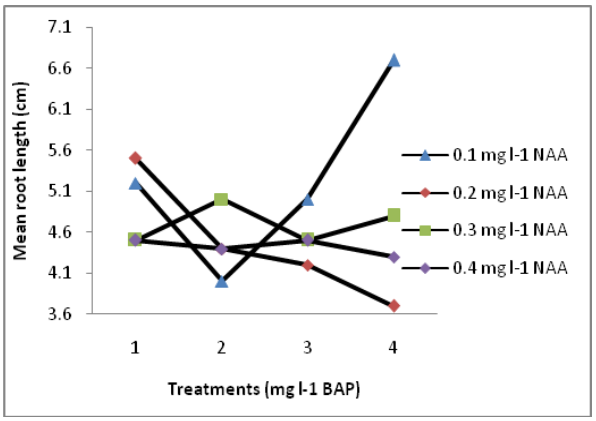

Figure 4. Combination effect of NAA and BAP on elongation of adventitious roots of *in-vitro* micro shoots after 8 weeks of culturing in full strength MS basal medium incorporated with 1.0g L^{-1} activated charcoal.

Initiation and elongation of rooting have been reviewed by Torres in 1988. According to him, initiation of roots is possible only in presence of auxins. The auxins added media to initiate rooting are NAA (0.05-1.0mg L^{-1}), IAA (0.1-10.0mg L^{-1}) and IBA (0.5-3.0mg L^{-1}) or combination of these.

Further, according to him incorporation of activated charcoal into the medium also beneficial for rooting [16]. It has also noted that auxins are only required during the initial phases of adventitious root formation and after that they become inhibitor and block the out-growth of newly formed organs [16].

Plate 4. Elongated adventitious root on *in-vitro* raised micro shoot (01) and rooted micro shoot prepared for data collection (02)

D. Experiment 4: Acclimatization of Cinnamon Plantlets

Acclimatization is the most crucial stage of tissue culture technology on which the ultimate success depends. Ref. [17] also reported that the establishment of tissue culture derived plants especially woody perennials is difficult. Further, Ref. [18] have mentioned that transfer of *in-vitro* propagated woody species from test tube to soil, requires careful monitoring of humidity and control of pathogens in plantlets and is a very difficult process. Therefore, this experiment is conducted to identify the effect of different potting media on the rate of survival and acclimatization of regenerated cinnamon plantlets.

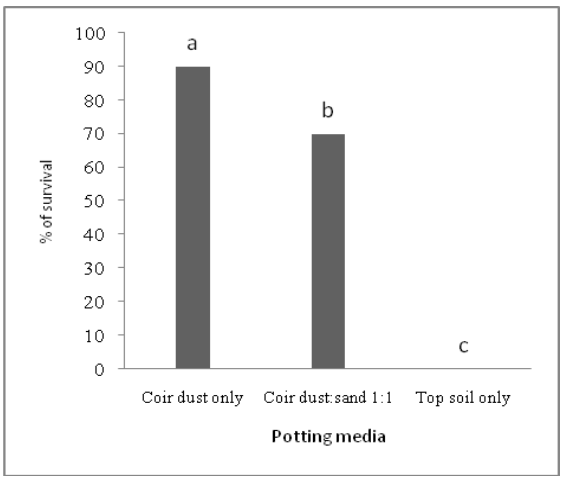

Figure 5. Survival rate of *in-vitro* rooted micro cuttings in different potting media after 6 weeks of acclimatization period

When considering the different potting media used for the acclimatization of rooted plantlets, coir dust alone gave maximum survival rate (90%) after 6 weeks of field transferring (Fig. 5). All the rooted plantlets were died in the media with the top soil alone after the 1st week of acclimatization process. However, coir dust: sand 1:1 potting medium gave comparatively better growth performance than the media containing only top soil. Therefore, it is recommended that the media alone with coir dust is the best for the acclimatization of *in-vitro* rooted micro cuttings of cinnamon.

Plate 5. Acclimatization of cinnamon plantlets in coir dust alone potting medium

The main objective of the research was to develop a suitable protocol for *in-vitro* micropropagation of cinnamon, which can be applied for most elite accessions. But, there was no any research done/limited in *in-vitro* propagation of cinnamon in elsewhere. Therefore, embryo culture was developed initially, in order to optimize the culture condition of axillary buds which may facilitate *in-vitro* micropropagation of cinnamon.

Excessive browning and systemic contaminants of explants were major drawback for culture initiation of cinnamon. Therefore, an experiment was conducted for the purpose of surface sterilization of isolated embryos using Clorox® as the surface sterilent. Different concentrations of Clorox® (15%, 20%, 25%) and exposure times (10, 15, 20 min) were tested and it was revealed that 15% Clorox® with 20 min exposure time was very effective in minimizing contaminants and browning of tissues and subsequent *in-vitro* establishment.

Surface sterilized embryos were inoculated into the half strength MS medium, but they were shown exudation of brown and black pigments and inhibited further development of tissues. Therefore, different antioxidants (0.1g L^{-1} Ascorbic Acid, 0.1g L^{-1} Citric Acid) and absorbents (1.0g L^{-1}) activated charcoal, 1.0g L^{-1} Polyvinylpyrrolidone) were incorporated into the culture medium for further reduction of browning of explants. Out of them, incorporation of 1.0g L^{-1} activated charcoal was advantageous for further development of embryo cultures of cinnamon.

A major obstacle in clonal *in-vitro* propagation of woody species is the initiation of rooting and hardening of plantlets for successful field transfer [18]. Therefore, micro shoots were taken from *in-vitro* raised seedlings and cultured in full strength MS medium incorporated with 1.0g L^{-1} activated charcoal with different NAA x BAP combinations (0.1, 0.2, 0.3 and 0.4mg L^{-1} NAA x 1, 2, 3 and 4mg L^{-1} BAP) for induction and elongation of adventitious roots. Cultures treated with 0.1mg L^{-1} NAA + 4.0mg L^{-1} BAP + 1.0g L^{-1} activated charcoal was beneficial for induction and elongation of adventitious roots of *in-vitro* stem cuttings. Therefore, combinations of NAA+BAP + activated charcoal were tested for adventitious roots induction of axillary bud cultures collected from controlled environment.

Successfully rooted micro cuttings were treated with 3 different potting media of coir dust only, coir dust: sand 1:1, top soil only and maintained >80% RH for three weeks prior to field transfer. The coir dust only was given the maximum percentage of survival. Hence, rooted axillary buds also treated with almost same potting media for acclimatization of rooted axillary bud cultures of cinnamon.

IV. Conclusions

According to the findings of the experiments conducted on *in-vitro* embryo cultures of cinnamon in half strength MS medium, following conclusions and recommendations can be made:

- For the surface sterilization of explants consists of embryonic axis with half of the cotyledon, 15% Clorox® for 20 minutes is very effective for minimizing contaminations and browning effect of tissues.
- Presence of 1.0g L^{-1} activated charcoal is effective in terms of enhancing stem elongation and leaf initiation.
- 0.1mg L^{-1} NAA + 4.0mg L^{-1} BAP +1.0g L^{-1} activated charcoal in full strength MS medium is the most effective treatment combination for adventitious root induction/elongation on *in-vitro* micro-stem cuttings of cinnamon.
- The coir dust alone in the potting medium found best for acclimatization of *in-vitro* raised rooted micro-cuttings under the controlled environment.

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References

[1] D. N. Samaraweera, "Propagation of cinnamon," in *Revitalization of Cinnamon Industry in Sri Lanka: Constraints and Remedies*, R. Senaratne and G. A. Dayatilake, Eds., Matara, 2000, pp. 38-45.

[2] J. Joseph and S. Wisweshwara, "Floral biology and variation in cinnamon. Genetics, plant breeding and horticulture," in *Proc. 4th Annual Symposium on Plantation Crops*, Mysore, 1982, pp. 431-434.

[3] E. Vadivel, V. Pannuswami, I. Irulappan, and G. Dharmaraj, "Vegetative propagation in cinnamon (*Cinnamomum zeylanicum*)," *South Indian Horticulture*, vol. 29, no. 4, pp. 231-232, 1981.

[4] K. G. G. Wijesinghe and R. Pathirana, "Germplasm screening and varietal development of cinnamon," in *Revitalization of Cinnamon Industry in Sri Lanka: Constraints and Remedies*, R. Senaratne and G. A. Dayathilake, Eds., Matara, 2000, pp. 15-25.

[5] K. R. Hedge, G. S. Sulikeri, and N. C. Hulamani, "Effect of growth regulator and pre-gridline treatments on rooting of cinnamon (*Cinnamomum verum* Presl.) air layers," *South Indian Horticulture*, vol. 3, no. 6, pp. 329-332, 1989.

[6] G. A. Dayathilake, "Possibilities of cloning cinnamon in-vitro and in-vitro," in *Revitalization of Cinnamon Industry in Sri Lanka: Constraints and Remedies*, R. Senaratne and G. A. Dayatilake, Eds., Matara, 2000, pp. 26-35.

[7] R. Senaratne, "The work shop – An overview," in *Revitalization of Cinnamon Industry in Sri Lanka: Constraints and Remedies*, R. Senaratne and G. A. Dayatilake, Eds., Matara, 2000, pp. 2-7.

[8] S. S. Bhojwani and V. Dhawan, "Acclimatization of tissue culture raised plants for transplantation to the field," in *Application of Biotechnology in Forestry and Horticulture*, V. Dhawan, Ed., New York: Plenum Pree, 1989, pp. 249-256.
[9] R. M. Skirvin, "Stone fruits," in *Hand Book of Plant Cell Culture*, W. R. Sharp, D. A. Evans, P. V. Ammirato, and Y. Yamada, Eds., New York: Macmikkon Publishing Con., 1984, pp. 349-368.
[10] M. Hedge, M. Kulasekaran, K. G. Shanmugavelu, and S. Jayasankar, "Cashew," in *Biotechnology of Horticultural Crops*, V. A. Parthasarathy, T. K. Bose, and P. Das, Eds., India: Naya Prokash, 1991, pp. 609-629.
[11] R. M. A. Sardinha, A. M. S. Bessa, J. Blake, D. Guyyer, C. Cassami, and P. Tamba-Bungue, "Cashew," in *Biotechnology of Horticultural Crops*, V. A. Parthasarathy, T. K. Bose, and P. Das, Eds., India: Naya Prokash, 1993, pp. 609-629.
[12] P. J. Davies, "Guava," in *Biotechnology of Horticultural Crops*, V. A. Parthasarathy, T. K. Bose, and P. Das, Eds., India: Naya Prokash, 1985, pp. 277-282.
[13] M. R. Ahuja, *Micropropagation of Woody Plants*, Dordrecht, Netherlands: Kluwer Academic Publishers, 1993, vol. 41, pp. 236-256.
[14] S. M. S. D. Ramanayake and A. Kovoor, "Cashew and coconut," in *Proc. Intl. Conf.*, Dar-es-Salaam, Tanzania, 1997, pp. 17-21.
[15] S. R. Nair and N. Mohanakumaran, "Cashew," in B*iotechnology of Horticultural Crops*, V. A. Parthasarathy, T. K. Bose, and P. Das, Eds., India: Naya Prokash, 1993, pp. 609-629.
[16] K. C. Torres, *Tissue Culture Techniques for Horticultural Crops*, New York: Van Nostrand Reienhold, 1988, pp. 87-152.
[17] S. C. Das, T. S. Barman, and R. Singh, "Morphogenesis in clonal propagation of woody plants," *J. Am. Soc. Hort. Sci*., vol. 4, pp. 151-174, 1990.
[18] A. Kumar and V. Kumar, *Clonal Tissue Culture of Important Fruit Crops*, India: International Book Distributing Company, 1998, pp. 106-107.

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