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Aziza M. Taj ALdeen & Mona S.Abd El-Aal

Sana'a University

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Chinese evergreen (Aglaonema commutatum) is being cultivated recently as a decorative plant in Egypt. In the present study trials were done to micropropagate it in vitro. For this purpose nodal segments were excised from shoots and cultured on Murashige and Skoog's (MS) culture media augmented with naphthalene acetic acid (NAA) 0.1 and 0.5 mg/l in combination with thiadiozhuros (TDZ) O.5 , 1.0 and 2 mg/l, followed by transfer to hormone –free culture media added to it active coal, for four weeks and regularly subcultured four successive times . Results of the present study have shown that 0.5 mg/l NAA + 2 mg/l TDZ for eight weeks were superior to the other hormonal combinations used. This treatment led to significant increases over the other treatments in number of shoots obtained per explant, the number of leaves per shoot and number of roots per plantlets 4.67±0.58, 2±0.00, 7.00±1.00 respectively, the average length of shoot and roots 8.50±0.10, 7.33±0.15 cm respectively, and the average fresh weight per regenerant 9.99±0.22 gram with a corresponding dry weight 0.30±0.01 gram. The obtained regenerates were easily acclimatized and transferred to pots.

Keywords: chinese evergreen, decorative plant, nodal segments, tissue culture.

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Chinese evergreen (Aglaonema commutatum) is being cultivated recently as a decorative plant in Egypt. In the present study trials were done to micropropagate it in vitro. For this purpose nodal segments were excised from shoots and cultured on Murashige and Skoog's (MS) culture media augmented with naphthalene acetic acid (NAA) 0.1 and 0.5 mg/l in combination with thiadiozhuros (TDZ) 0.5 , 1.0 and 2 mg/l, followed by transfer to hormone –free culture media added to it active coal, for four weeks and regularly subcultured four successive times . Results of the present study have shown that 0.5 mg/l NAA + 2 mg/l TDZ for eight weeks were superior to the other hormonal combinations used. This treatment led to significant increases over the other treatments in number of shoots obtained per explant, the number of leaves per shoot and number of roots per plantlets 4.67±0.58, 2±0.00, 7.00±1.00 respectively, the average length of shoot and roots 8.50±0.10, 7.33±0.15 cm respectively, and the average fresh weight per regenerant 9.99±0.22 gram with a corresponding dry weight 0.30±0.01 gram. The obtained regenerates were easily acclimatized and transferred to pots.

The obtained results may facilitate production of A. cammutatum on the commercial level in our country.

Keywords: chinese evergreen, decorative plant, nodal segments, tissue culture.

Author α: Biology Department, Faculty of Science, Sana'a University, Sana'a, Yemen.

σ: Botany Dept., Fac. of Agriculture. Ain Shams University, Cairo, Egypt.

I. INTRODUCTION

Aglaonema commutatum is a monocotyledonous plant belonging to family Araceae, known as aroids, it is an ornamental plant important in interior landscaping due to its attractive brightly colored leaves. Vegetative methods are the traditionally propagation means for Aroid plants, such as division or cuttings (Chen and Stamps, 2006). Vegetative propagation is associated with diseases spread between plants (Norman and Yuen, 1998). Tissue culture techniques, are known to be faster than traditional methods of propagation and many reach commercial production levels within 2–3 years (Henny and Chen, 2003).

Compared to other plant growth regulators (PGRs), TDZ is a more powerful and potent synthetic growth regulator exhibiting both auxin and cytokinin (CK)-like effects on plants. Despite this unique and dual effect, TDZ's action is often overgeneralized and referred to as a cytokinin. It is therefore important to note that although TDZ can mimic the effects of auxins and CKs. Structurally it differs from both of these PGR groups, possessing both phenyl and thiadiazole functional groups. Both groups are required for biological activity (Mok et al, 1987). TDZ can be used for regeneration at lower concentrations making it a valuable commercial agrochemical (Guo et al, 2011).

The aim of the present study is to micropropagate Aglaonema commutatum in vitro using NAA And TDZ.

II. MATERIAL AND METHODS

Mother cultures of *Aglaonema commutatum* were obtained from tissue culture lab, EL-Zohria Botanical Garden, Cairo, Egypt. The experiment was carried out in Plant Tissue Culture Lab of Agric. Botany Dept., Fac. of Agric. Ain Shams Univ., during the years of 2019–2020.

Explants: Nodal segments of three month old sterile plantlets.

Culture media: Murashige and Skoogs basal medium (1962) containing sucrose 30 g/l and solidified with Agar 7 gram/l. pH adjusted to 5.7 prior autoclaving, the medium was divided into glass jars (200 ml) containing 40 ml of the testing medium. The culture medium was autoclaved at 121°C and 1.1Kg cm^{-2} for 20 min.

Growth regulators: Naphthalene acetic acid (NAA) and Thidiazuron (TDZ).

Table 1: Various concentrations of Growth regulators used

Treatments	Growth regulators (mg/l)	
	NAA	TDZ
1	0	0
2	0.1	0.5
3	0.5	1.0
4	0.5	2.0

Culture conditions : Cultures were incubated in a growth room 25 ± 2°C under illumination intensity of 1500 lux day light located 40 cm above the top of cultures (40 watts white fluorescent lamp). The photoperiod was 16 hours light and 8 hours dark that is automatically controlled.

Statistical analyses: Data represent mean ± standard deviation of 3 different values. The experiment was arranged in a complete randomized design (Gomez and Gomez, 1984) with ten replicates (jars), each replicate has four explants. The obtained results were subjected to statistical analysis of variance (ANOVA) in statistics (8^{th} edition analytical software, USA) by (Steel *et al*, 1997). Differences between means were contracted by LSD meth.

II. RESULTS AND DISCUSSION

Ornamental plants are an important element of indoor decorating and coordination. *Aglaonema commutatum* is a beautiful indoor plant, distinguished by the beauty of its pied leaves, but there are great difficulties in propagating it by traditionally methods. Traditionally methods of vegetative propagation of plant have many disadvantages such as infection with bacterial and fungal diseases as stated by (Ranjan Kumar Tarai et al, 2020). It has been mentioned by_(Ajit Kumar Sahoo. *et al*, 2019) that growth and development of plants is controlled by two sets of internal factors, such as nutrition and hormonal constituents.

In this experiment, plant growth regulators were used *in vitro* with success and positive results were achieved in multiplication and production of a lot of plants during a short period as indicated in figure (1) and table (2). In addition it was observed that there is a direct relationship between cytokinin concentration and the percentage of segments that gave shoots, such results were observed by (Ahmed *et al*, 2008).

Table 2: Effect of different growth regulators treatments on % of bud multiplication of *Aglaonema commutatum in vitro*

Treatments	growth regulators		% of bud multiplication
	NAA (mg/l)	TDZ (mg/l)	
Treat. 1	0.0	0.0	0.0
Treat. 2	0.1	0.5	67
Treat. 3	0.5	0.1	73
Treat. 4	0.5	2.0	88

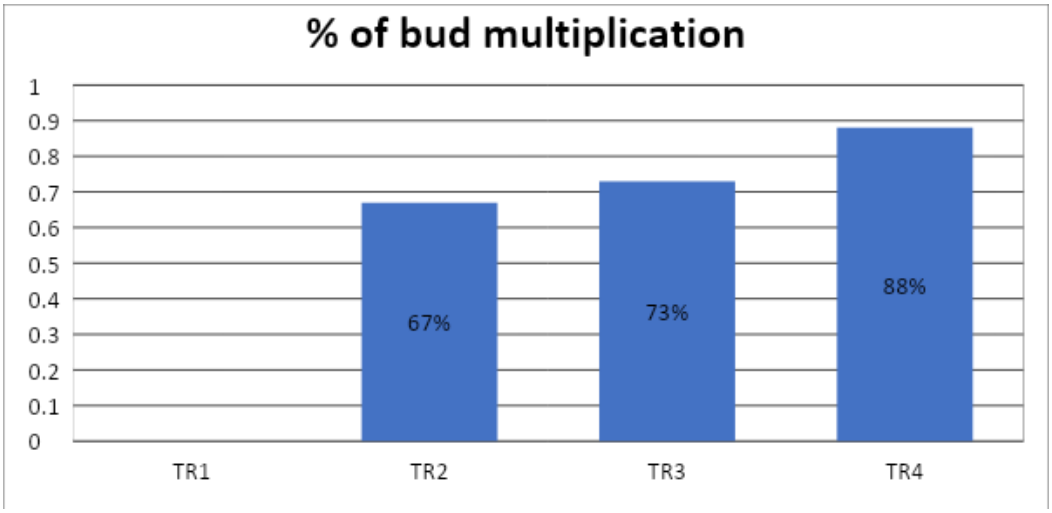

Figure 1: Effect of different growth regulators treatments on % of bud multiplication of *Aglaonema commutatum in vitro*

Table 3: Effect of different growth regulators treatments on different morphogenic parameters of *A. commutator in vitro*

Treatments		Growth parameters						
NAA (mg l^{-1})	TDZ (mg l^{-1})	No of shoots/ explant	No of leaves/ shoot	Shoot length (cm)	Root length (cm)	No of roots/ plantlet	F.W shoot system	D.W shoot system
0.0	0.0	0.00 ^{d}	0.00 ^{c}	0.00 ^{d}	0.00 ^{d}	0.00 ^{c}	0.00 ^{d}	0.00 ^{c}
0.1	0.5	1.00±00 ^{c}	1.00±00 ^{b}	3.50±0.10 ^{c}	3.03±0.21 ^{c}	4.33±0.58 ^{b}	2.26± 0.06 ^{c}	0.05±00 ^{c}
0.5	1.0	2.00±00 ^{b}	1.00±00 ^{b}	5.77±0.06 ^{b}	5.03±0.15 ^{b}	5.33±0.58 ^{b}	4.57± 0.05^{b}	0.12±0.01 ^{b}
0.5	2.0	4.67±0.58 ^{a}	2.00±00 ^{a}	8.50±0.10 ^{a}	7.33±0.15 ^{a}	7.00±1.00 ^{a}	9.99± 0.22 ^{a}	0.30±0.01 ^{a}
LSD		0.54	0.00	0.14	0.62	1.21	0.22	0.055
Means followed by different letters are significantly different.								

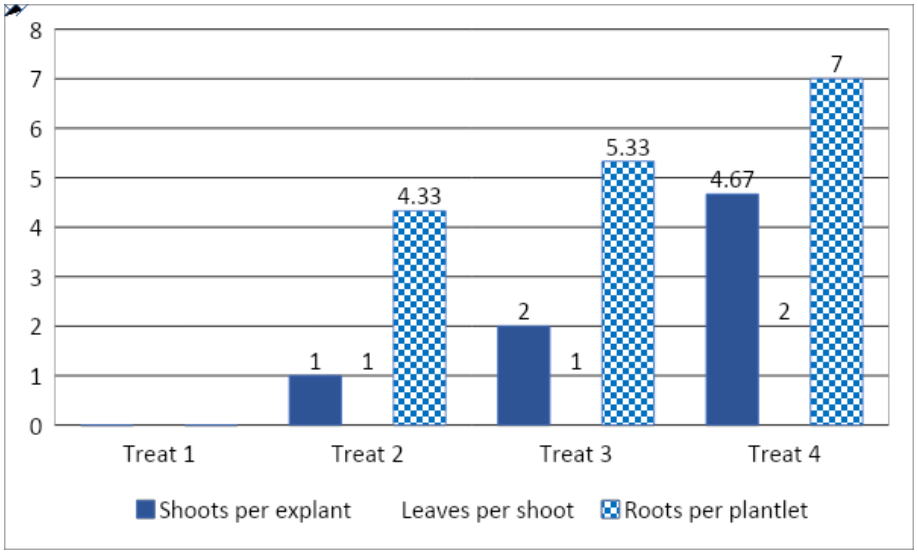

Figure 2: Effect of growth regulators treatments on number of shoots per explant and number of leaves and roots per plantlet

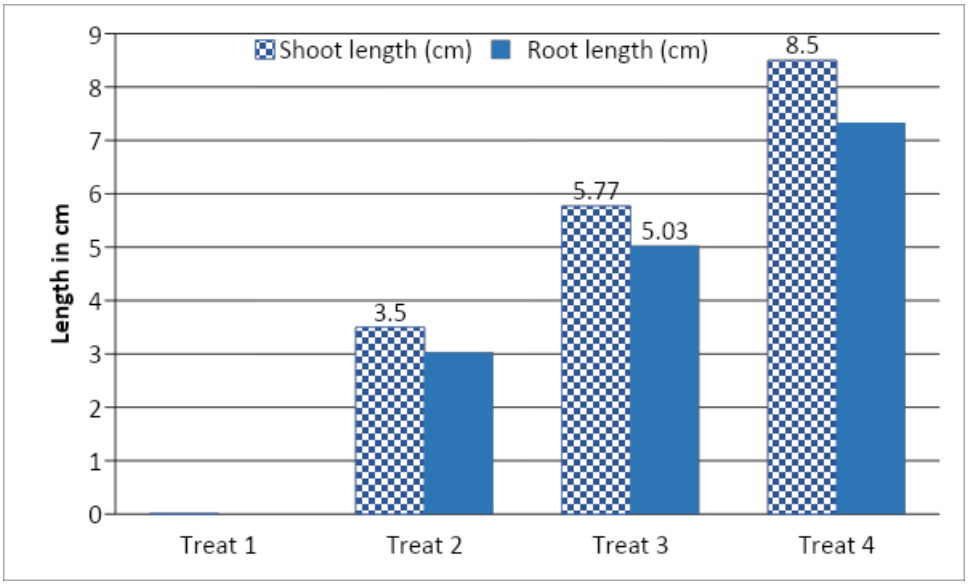

Figure 3: Effect of different growth regulators treatments on shoot and root lengths

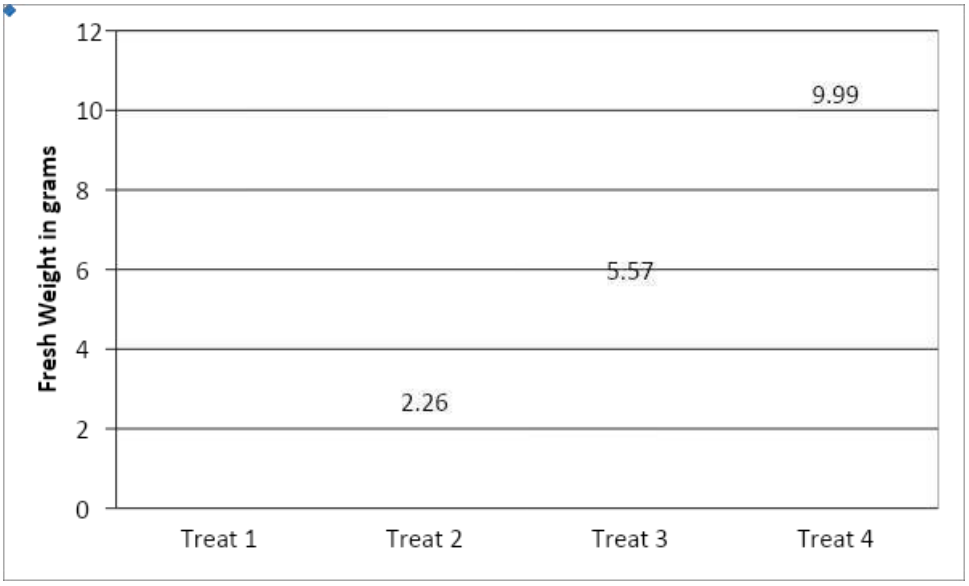

Figure 4: Effect of different growth regulators treatments on the fresh weight of shoot system

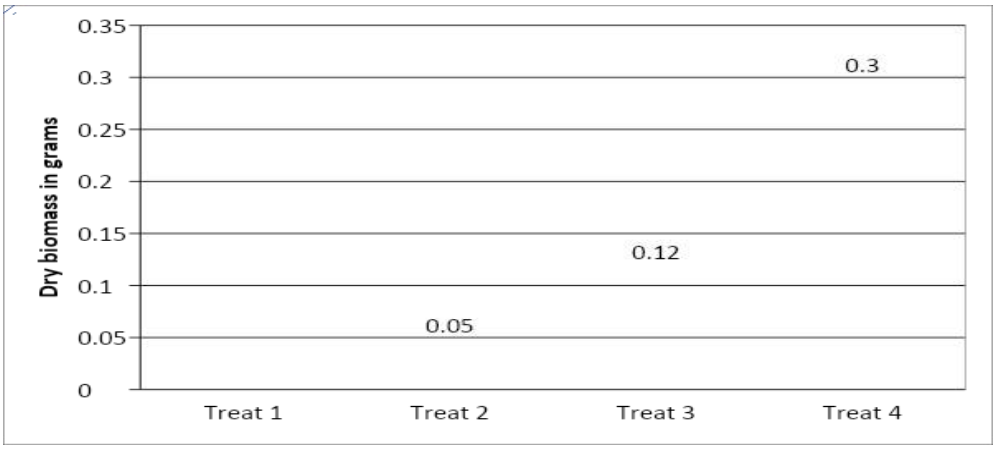

Figure 5: Effect of different growth regulators treatments on the shoot dry biomass

Data clarified in table (2) and figure (1) showed the effect of various NAA & TDZ concentrations on the shoot induction percentage. The data revealed that TDZ treatments promoted and significantly increased all growth parameters.

In table (3) fig (2), (3), (4) and (5).The highest and significant number of shoots obtained per explant, leaves per shoot and roots per plantlets 4.67±0.58, 2±0.00, 7.00±1.00 respectively, the average length of shoot and roots 8.50±0.10, 7.33±0.15 cm respectively, and the average fresh and dry weight per plantlets 9.99±0.22, 0.30±0.01gram respectively were recorded with 0.5 mg/l NAA+ 2 mg/l TDZ.

In many studies e.g. (Asma *et al*, 2020) indicated that both auxins and cytokin is were necessary for maximum response *in vitro*, but in other studies like (Mariani *et a,* 2011) reported that TDZ is CK-like compounds that can promote shoot proliferation and had important role for induction of shoots, and play role of both auxins and cytokin is. Direct shoot organogenesis, can be limited by the availability of preexisting meristems on the explants and a low multiplication rate. Also, (Fang *et al,* 2013) reported that when the cytokinin were used alone, it failed to induce adventitious shoots, while in contrast using of an auxin with a cytokinin may often prove useful.

Diverse factors may affect the capability of TDZ to induce shoot bud initiation and growth including: concentration of TDZ, cultivar, type and source of explant, age or phase of growth, presence of other PGRs, balance of endogenous growth regulators and presence of light (Sanikhani *et al,* 2006; Visser *et al,* 1992).

(Mariani *et a,* 2011) demonstrated that using 1.50 mg/l TDZ on *Aglaonema sp*. micropropagation was successful. This suggests that a low concentration of TDZ (0.15 mg/l) favors the tissue culture of Araceae plants. In the same trend (Fang *et al,* 2013) excised the single stem nodal segments from the elongated shoots for Aglaonema 'Lady Valentine and treated them with different combinations of NAA and TDZ, the average of adventitious shoots per stem segment was 10.9 produced with 0.5 mg/l NAA and 2 mg/l TDZ. The number of adventitious shoots induced varied greatly from one stem segment to another. Since the adventitious shoots most likely originated from the meristematic cells located on the periphery of the axillary bud, it is suspected that the number of meristematic cells present on the nodal region of each stem segment is highly variable. The variable response of stem nodal segments may due to age, size or other conditions of the plant material, as observed in *Dieffenbachia compacta* by (Azza *et al,* 2010; Chen and Yeh 2007; Zhang *et al*, 2004 and Huetteman and Preece, 1993) who observed that low concentrations of TDZ could induce shoot multiplication while the corresponding BA concentrations could not. Superiority of TDZ for the node and shoot induction was reported in *Aglaonema* sp. and a number of other ornamental plant species (Mariani *et al*, 2011). The probable reason for this may be attributed to the ability of plant tissues to absorb and use TDZ more readily than other PGRs. Adding of 0.5 mg/l BA + 0.5 mg/l Kin to the cultures saved the plantlet from stunted growth and encouraged the continuation of the growth of new shoots to some extent. It was mentioned that by (Ahmed et al. 2008) that adding cytokinins stimulate the cell division and growth of shoot.

Root morphogenesis was done on MS medium without PGR, where plantlets rooted well. The non-prerequisite for an auxin at the rooting stage shows that the plantlets may contain enough indogenous auxin (Murti *et al,* 2012) for root initiation.

(Mariani, 2011) explained in his research that the survival rate of live plants after the seedlings acclimatization stage was 100%, but the seedlings were transferred on sphagnum moss then after that transfer to soil, while in figure (6 –f,g,h,i) clearly the success of the seedling acclimatization phase, directly without gradations in pots containing regular loam soil is illustrated.

More or less similar results were carried out by (Kaviani *et al,* 2019) on *Aglaonema widuri* and

(El-Mahrouk, 2016) on *Aglaonema nalantine* where the authors could successfully enhance micropropagatio using NAA and TDZ which were used in the present study.

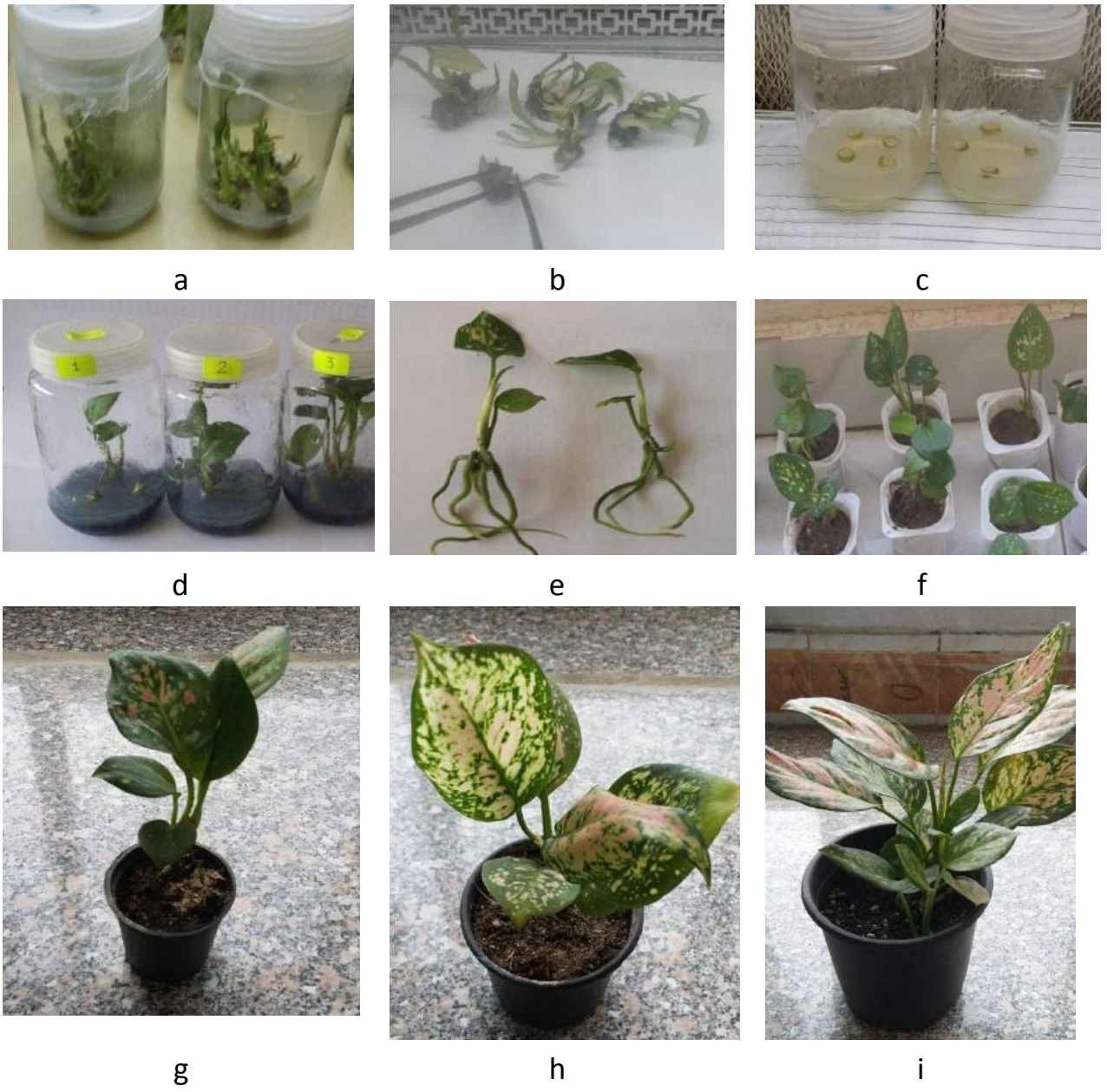

Figure 6:

a) Mother plant
b) Separated micro-shoots
c) Nodal segments
d) Shoot & root morphogenious in jars
e) Shoot & root morphogenious
f) Acclimatized plantlets 1 month old
g) Acclimatized plants 3 months old
h) Acclimatized plants 5 months old
i) Acclimatized plants 7 months old

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REFERENCES

1. Ahmad, A.B.; M.N. Ling and C.T. San (2008). Plant regeneration technique from rhizome explants of *Aglaonema simplex*. J. Sustain. Sci. Manage., 3: 52-57.
2. Alawaadh, A. A, Dewir, Y. H, Alwahibi, M, Abdulhakim A.Aldubai (2020). Micropro pagation of Lacy Tree Philodendron (Philodendron bipinnatifidum Schott ex Endl.). HortScience:apublication of the American Society for Horticultural Science 55(33):294-299.
3. Azza M, El-Sheikh K, Mutasim MK (2010). *In vitro* shoot micropropagation and plant establishment of an ornamental plant dumb cane (*Dieffenbachia compacta*). Int J Curr Res 6: 27–32.
4. Chen J, Stamps RH (2006). Cutting propagation of foliage plants. In: Dole JM, Gibson JL (eds) Cutting propagation: a guide to propagating and producing floriculture crops. Ball Publishing, Batavia, pp 203–228.
5. Chen WL, Yeh DM (2007). Elimination of *in vitro* contamination, shoot multiplication, and *ex vitro* rooting of Aglaonema. HortScience 42: 629–632.
6. El-Mahrouk, M.El, Hassan, Y, Dewir, and Naidoo, Y. (2016). https:// doi.org /10.21273 /HORTSCI.51.4.398 page Count: 398-402 Volume 51:Issue4 Artic Category: Research Article Online.
7. Fang J.Y., Hsu Y.R. and Chen F.C. (2013). Development of an efficient micropropagation procedure for Aglaonema 'Lady Valentine' through adventitious shoot induction and proliferation. Plant Biotechnology 30, 423–431.
8. Gomez, K.A. and Gomez, A.A. (1984). Statistical procedures for agricultural research. 2nd ed. International Rice Research Institute, College, Laguna. 680 p.
9. Guo B, Abbasi B, Zeb A et al (2011). Thidiazuron: a multi-dimensional plant growth regulator. Afr J Biotechnol 10(45):8984–9000.
10. Henny R, Chen J (2003). Foliage plant cultivar development. Plant Breed Rev 23:245–290.
11. Huetteman, A.C. and J.E. Preece. (1993). Thidiazuron: a potent cytokinin for woody plant tissue culture. Plant Cell Tiss. Org. Cult. 33:105–119.
12. Kaviani, B. (2015). 'Some useful information about micropropagation'. Journal of Ornamental Plants, 5 (1): 29-40.
13. Kaviani1, B, Sedaghathoor, S , Motlagh, M.R .S and Rouhi, S .(2019). Influence of plant growth regulators (BA, TDZ, 2-iP and NAA) on micropropagation of Aglaonema widuri. Iranian Journal of Plant Physiology. 9 (4),2901-2909.
14. Mariani, T. S., A. Fitriani, J. A. Teixeira da Silva, A. Wicaksono and T. F. Chia. (2011). Micropropagation of Aglaonema using axillary shoot explants'. International Journal of Basic Applied Science, 11: 46–53.
15. Mok M, Mok D, Turner J, Mujer C (1987). Biological and biochemical effects of cytokinin-active phenylurea derivatives in tissue culture systems. Hort Sci 22(6):1194–1197.
16. Murashige, T. and Skoog F. (1962). A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant., 15: 473-497.
17. Murti, R.H., Debnath, S.C. and Yeoury, Y.R. (2012). Effect of high concentration of thidiazuron (TDZ) combined with 1 H-indole-3- butanoic acid (IBA) on Albion strawberry (*Fragaria x ananassa*) cultivar plantlets induction. African Journal of Biotechnology, 11(81): 14696-14702.
18. Norman DJ, Yuen JMF (1998). A distinct pathotype of Ralstonia (Pseudomonas) solanacearum race 1, biovar 1 entering Florida in pothos (Epipremnum aureum) cuttings. Can J Plant Pathol 20:171–175.
19. Sahoo, A. K, Tarai, R. K, A.K.Srivastava, Sau, S. (2019) Book: Plant Growth Regulators in Tropical & Sub-Tropical Fruit Crops (Part I) (pp.1-250). Publisher: Jaya Publishing House.Chapter Plant Growth Regulators.
20. Sanikhani M, Frello S, Serek M (2006). TDZ induces shoot regeneration in various Kalanchoe blossfeldiana Poelln. Cultivars in the absence of auxin. Plant Cell Tissue Organ Cult 85(1):75–82.
21. Steel, R.G.D., Torrie, J.H. and Dicky, D.A. (1997). Principles and procedures of statistics:

A biometrical approach, 3rd ed. McGraw Hill, Inc. Book Co., New York, USA, pp: 352-358.
22. Tarai, R. K, Naik, B, Sahoo, A. K, Mandal, P. (2020). Plant Propagation and Nursery Management. Publisher: New India Publishing Agency, New Delhi, ISBN: 9789389907247
23. Visser C, Qureshi J, Gill R et al (1992). Morphoregulatory role of thidiazuron substitution of auxin and cytokinin requirement for the induction of somatic embryogenesis in geranium hypocotyl cultures. Plant Physiol 99(4):1704–1707.
24. Wang S, Steffens G, Faust M (1986). Breaking bud dormancy in apple with a plant bioregulator, thidiazuron. Phytochemistry 25(2):311–317.
25. Zhang S, Jiang R, Zhou H (2004). Study on rapid propagation of Aglaonema commutatum cv. 'Golden Jewelry.' Chin Agr Sci Bull 20: 39–40.