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In vitro regeneration from nodal explants of *Kedrostis foetidissima* - A medicinal climber

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Abstract: ***Kedrostis foetidissima*** **is an important medicinal climber belonging to the family cucurbitaceae. It is widely used in folklore and traditional systems of medicine for various disorders like seborrheic dermatitis, measles, diarrhea, urinary tract infections and asthma. The availability of plant decreased in its natural habitats due to high medicinal value and over usage. To enhance plant multiplication of this plant an efficient *in vitro* regeneration protocol was established using nodal explants through direct regeneration. MS medium amended with BAP, TDZ (0.5-2.5 mg/l) alone and together with IAA/NAA (0.2-1.0 mg/l) were used. Of these BAP (2.0 mg/l) induced the highest percentage response (73%) with 17.2 ± 0.24 mean number of shoots per explant after the first subculture. The shootlets were rooted on half strength MS medium augmented with IBA (1.0 mg/l) with an average of 16.3 ± 0.47 roots per shoot and mean root length (cm) is 1.31±0.07 after four weeks of culture. The regenerated plantlets were hardened in polycups, then acclimatized in greenhouse and transferred to the field with 68% survival rate.**

Index Terms: Kedrostis foetidissima, **MS Medium, Multiple shoots, Nodal explant, Shoot bud initiation.**

I. INTRODUCTION

Kedrostis foetidissima (Jacq.) Cogn. is an important medicinal climber belonging to the family Cucurbitacece. It is a monoecious scandant herb with perennial root stock. It is commonly known as "Kunkuma donda" in Telugu. It is distributed in Tropical and South Africa, Asia through India to Burma (Britto, 2019). In India it is distributed in Andhra Pradesh, Gujarat, Karnataka, Madhya Pradesh, Maharashtra, Tamilnadu (Renner & Pandey, 2013) and Telangana (Reddy & Reddy, 2016).

The medicinal importance of cucurbitaceae members attributed to its presence of cucurbitacins. *Kedrostis foetidissima* has great medicinal value and uses in traditional and folklore systems of medicine (Siddha) since many years (Kunthavi *et al*., 2018). Leaves used to treat skin diseases, measles, chest pain, urinary tract infections (Giday, 2003), common cold in children (Kuruppuswamy, 2007), diarrhea and seborrheic dermatitis, eczema (Jaganathan *et al.*, 2016). The tuber and fruit are demulcent is advised for asthma (Dogra *et al.*, 2015). Phytoconstituents like quercetin-rhamnoside, cucurbitacin glycoside and rutin in leaves and stems are responsible for antimicrobial, anti-diarrheal, antioxidant and anticancer activities (Amutha, 2017). The most interesting feature of this plant is the presence of cucurbitacins. Cucurbitacins B, D, E and I (Rios, 2005) and toxic cucurbitacins C from stem, I from leaves were reported (Njoroge & Newton, 1994). These compounds are responsible for anticancer activity. Further investigations proved that crude extract of leaves showed anti-proliferative activity against breast cancer cell lines (Choene & Motadi, 2012) and Hepatocellular liver carcinoma cell lines (Elavazhagan, 2016). Leaf and stem extracts have antibacterial and antifungal properties (Priyavardini *et al.*, 2008; 2012) and anti-diarrheal property (Shivaprakash, 2016) and due to having high amount of antioxidants, it is used to free radical induced diseases as curative agent (Pavithra & Vadivukarasi, 2015).

Tubers also possess nutritional values such as high protein content, starch and niacin vitamin (Mohan & Kalidas, 2010).

Tissue culture techniques successfully employed for conservation of many crop and wild Cucurbitaceae members such as *Momordica dioica* (Mustafa *et al*., 2013), *Momordica charantia* (Thiruvengadam *et al*., 2012), *Coccinia indica* (Borah *et al.*, 2019), *Trichosanthes cucumerina* (Kawale & Chowdary, 2009), *Bryonopsis laciniosa* (Shastree *et al*., 2012).

The techniques of micro propagation provide a good alternative for the plant species that shows resistance to practices of conventional bulk propagation (Bhatia *et al*, 2015). Increased demand for high yield and disease resistant plants, plant tissue culture aids in large scale production of plants in stipulated time using an explant and light as a source (Patil *et al.*, 2021). As this plant has been widely used in traditional systems of medicine, its availability is getting decreased in its natural habitats and it is considered as least concerned species (Nirmala & Pandian, 2013). There is an urgent need to conserve this plant species. So this study has been carried out for regeneration of *Kedrostis foetidissima* through *in vitro* techniques.

II. Materials and Methods

A. Plant material collection and surface sterilization

Plant material was collected from Warangal district and grown in the botanical garden of the Department of Botany, Kakatiya University, Warangal. The specimen was identified by Dr. Md. Mustafa, Department of Botany, Kakatiya University, Warangal and deposited in Herbarium (Accession no. 1124).

Healthy and juvenile shoots were collected from field grown plant, they were dipped in water and brought to the laboratory immediately. The stem material was cut into single node pieces of 1.cm.length. They were washed under running tap water for 20 minutes. After that they were dipped in NaOCl solution for surface disinfection and followed by rinsed with normal water, next with distilled water to remove excess disinfectant. Further sterilization was performed under laminar-air-flow hood with 1.0 % (W/V) $HgCl_2$ for 3-4 minutes, immediately 5 times rinsed with sterilized double distilled water. Then the nodal segments were blotted dried and tips were cut slant and inoculated on MS medium supplemented with various combinations of hormones for multiple shoot induction.

B. Composition of media and maintenance of culture conditions

The explants were inoculated on MS medium supplemented with 3% sucrose and 0.8% agar-agar was added for solidification. The P^H of the medium was balanced to 5.6 to 5.8 after that 20 ml of medium dispensed in 25 x150 mm borosil culture tubes. All the culture tubes along with medium were closed with non-absorbent cotton, then autoclaved at 121^0 C temp and 15 lb pressure for 20 minutes for disinfection. Inoculated culture vessels were kept in a growth chamber maintained at 25 ± 2^0 C temp, photoperiod of 8/16 hrs dark and light under cool and white fluorescent tubes.

Table I. Effect of BAP, TDZ alone and in combination with IAA and NAA on multiple shoot induction from nodal explants of *Kedrostis foetidissima*

MS Medium + Hormone (mg/l)	(%) of response	Mean number of shoots/explant ± S.E	Mean length of shoot (cm) ± S.E
BAP			
0.5	43	1.5 ± 0.16^{ooo}	2.55 ± 0.08^{ddd}
1.0	57	6.1 ± 0.23^{fff}	6.79 ± 0.32^{aaa}
1.5	67	8.9 ± 0.17^{ccc}	3.47 ± 0.14^{bbb}
2.0	73	17.2 ± 0.24^{aaa}	3.15 ± 0.23^{bbb}
2.5	60	10.1 ± 0.23^{bbb}	2.11 ± 0.14^{ddd}
TDZ			
0.5	0	NR	NR
1.0	57	3.5 ± 0.16^{lll}	1.71 ± 0.25^{fff}
1.5	60	5.1 ± 0.23^{hhh}	2.39 ± 0.06^{ddd}
2.0	53	2.9 ± 0.31^{mmm}	1.25 ± 0.06^{hhh}
2.5	0	NR	NR
BAP+IAA			
1.0+0.2	47	1.4 ± 0.16^{ooo}	1.38 ± 0.12^{ggg}
1.0+0.4	53	5.2 ± 0.20^{hhh}	1.79 ± 0.13^{fff}
1.0+0.6	57	4.9 ± 0.23^{iii}	2.47 ± 0.08^{ddd}
1.0+0.8	53	3.1 ± 0.31^{lll}	2.01 ± 0.09^{eee}
1.0+1.0	60	7.1 ± 0.31^{ddd}	3.49 ± 0.13^{bbb}
BAP+NAA			
1.0+0.2	40	2.5 ± 0.16^{nnn}	1.85 ± 0.11^{fff}
1.0+0.4	47	5.5 ± 0.16^{ggg}	2.92 ± 0.12^{ccc}
1.0+0.6	53	3.7 ± 0.27^{kkk}	2.55 ± 0.07^{ddd}
1.0+0.8	50	2.6 ± 0.16^{mmm}	2.03 ± 0.06^{eee}
1.0+1.0	57	6.3 ± 0.45^{fff}	3.23 ± 0.25^{bbb}
TDZ+IAA			
1.0+0.2	45	3.6 ± 0.23^{kkk}	1.46 ± 0.06^{ggg}
1.0+0.4	63	6.6 ± 0.22^{eee}	2.61 ± 0.05^{ccc}
1.0+0.6	53	5.7 ± 0.21^{ggg}	1.36 ± 0.16^{ggg}
1.0+0.8	50	5.2 ± 0.35^{hhh}	1.15 ± 0.56^{hhh}
1.0+1.0	47	2.9 ± 0.23^{mmm}	1.03 ± 0.23^{iii}
TDZ+NAA			
1.0+0.2	40	1.3 ± 0.15^{ooo}	1.32 ± 0.13^{ggg}
1.0+0.4	60	5.6 ± 0.29^{ggg}	2.82 ± 0.25^{ccc}
1.0+0.6	57	4.0 ± 0.14^{jjj}	2.36 ± 0.21^{ddd}
1.0+0.8	50	3.3 ± 0.13^{lll}	1.33 ± 0.53^{ggg}
1.0+1.0	53	3.8 ± 0.46^{kkk}	1.23 ± 0.45^{hhh}

*NR= No response, Data represents average of three replicates; each replicate consists of 30 cultures and data recorded after 4 weeks of first subculture. Values are Mean ± S.E. Mean followed by different letters within hormone alone and combinations are significantly different at p=0.05, as per Duncan's Multiple Range Test.

C. Shoot induction, Shootlet elongation and Multiplication

MS medium supplemented with various concentrations of 6-Benzyl amino purine (BAP) and Thiadiazuron (TDZ) alone

(0.5- 2.5mg/l) and along with auxins like Indole 3-acetic acid (IAA) and Napthalene acetic acid (NAA) each (0.2-1.0mg/l) were used to elicit morphogenic response from nodal explants. *In vitro* raised shoots were sub cultured on fresh MS medium of same concentration of hormones with 4-5 weeks of intervals. The mean number of shoots and mean length of shoots were recorded after 4 weeks of each subculture.

D. Root induction and acclimatization of plantlets

The *in vitro* grown shootlets of 5-6 cm. length were cut out from the cultures and transferred on half strength MS medium fortified with Indole 3-butyric acid (IBA), IAA and NAA (0.5-2.5mg/l) alone. Results were reported after 4 weeks of culture. *In vitro* rooted plantlets were shifted to poly cups containing sand and black soil (1:1) for hardening. They were kept in a culture room under 16/8 dark and light period for 2 weeks. After that they were shifted to the greenhouse at 80-90% RH and 28±2^{o} C temp before shifting into the field.

E. Statistical analysis

Thirty replicates for each treatment were used for multiple shoot induction and root initiation individually and experiments were repeated for thrice. The details for percentage of response, mean number of shoots per explant, shoot length (Table-I), mean number of roots and mean length of root per plantlet (Table-II) were recorded. Data was statistically presented as the mean ±standard error variance (ANOVA) using SPSS software and means were compared using Duncan`s multiple range test at 5% probability.

III. RESULTS AND DISCUSSION

A. Effect of phytohormones on multiple shoot induction

In the present study BAP, TDZ alone and in combination with auxins i.e. IAA and NAA were used to investigate *in vitro* multiple shoot induction from nodal explants of *Kedrostis foetidissima*. Shoot bud induction was observed in all concentrations of plant growth regulators tested, but response of percentage varied in different concentrations was shown in Table-I. Nodal explants cultured on MS medium augmented with BAP, TDZ (0.5mg/l -2.5mg/l) alone, and along with auxins NAA and IAA (0.2mg/l -1.0mg/l) responded within one week and proliferation of axillary shoot bud noticed from nodes and it is also noticed that swelling of lower cut ends to produce callus after 12 days of culture (Fig-1A). Similar results were achieved in *Woodfordia fructicosa* (Mallesham *et al.*, 2012). The developed shoots from nodal explants were subcultured after 4 weeks on fresh MS medium of same concentration.

MS medium supplemented with 2.0mg/l BAP showed maximum percentage response of explants (73%) with highest mean number of shoots 17.2 ± 0.24 after 4 weeks of first subculture (Fig -1C). Results were on par with *Corollocorpus epigaeus* (Vemula *et al.*, 2019) *Cucumis trigonus* (Aruna & Niranjana, 2016). But previous reports of tissue culture studies in *Kedrostis foetidissima* achieved high frequency regeneration (80%) from intermodal derived callus on MS medium supplemented with 10 mg/l BAP (Suganthi & Poornima, 2014). However higher mean length of shoots about 6.79±0.32 (cm) achieved at 1.0mg/l BAP. Length of shoot was also affected by concentration of BAP. An increased concentration of BAP in culture medium resulted in shorter length of shoot. These results are similar with previous reports in *Limnophila aromatica* (Mohemad & Asim, 2015) and in *Cucumis sativus* (Alam *et al.*, 2015)

Nodal explants cultured on MS medium supplemented with 1.0mg/l BAP and 1.0mg/l IAA have shown maximum response percentage (60%) with maximum number of shoots 7.1±0.31 and higher shoot length (cm) 3.49±0.13 were recorded. Explants were cultured on 1.0mg/l BAP and 1.0mg/l NAA showed maximum response percentage (57%) with a good number of shoots 6.3±0.45 and shoot length (cm) 3.23±0.25. Remarkable

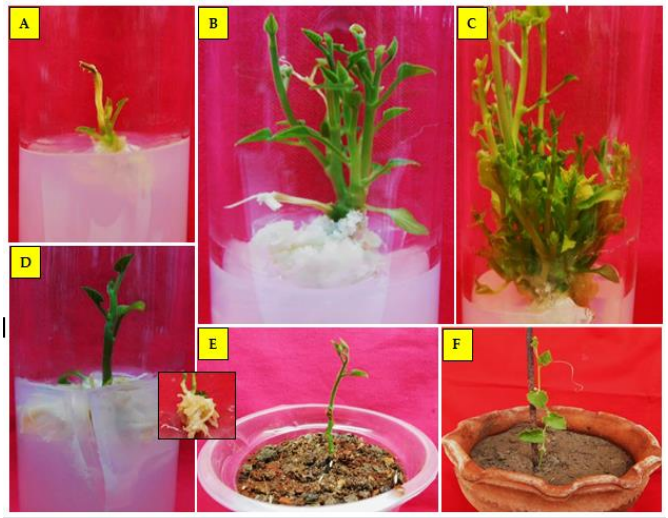

Fig.1. Morphogenic response of *Kedrostis foetidissima* on MS medium supplemented with various combinations and concentrations of hormones.

- **A.** Shoot bud initiation after 1 week of inoculation on MS medium with 2.0mg/l TDZ from nodal explants
- **B.** Multiple shoot induction after 4 weeks of inoculation on MS medium with 1.5 mg/l TDZ from nodal explants
- **C.** High frequency multiple shoot induction on MS medium with 2.0mg/l BAP after first subculture
- **D.** *In vitro* rooting of shootlets on MS medium with 1.0 mg/l IBA
- **E.** Hardening of plantlet in cups containing soil and sand
- **F.** Growth of *in vitro* regenerated plants on soil in the green house.

Table II. Effect of auxins on root induction in *in vitro* raised shootlets of *Kedrostis foetidissima*

Half strength MS Medium + Hormone (mg/l)	(%) of response	Mean number of roots/explant ± S.E	Mean length of root (cm) ± S.E
IBA			
0.5	67	13.4± 0.30^{ddd}	0.85±0.06^{fff}
1.0	93	16.3 ± 0.47^{aaa}	1.31±0.07^{ddd}
1.5	83	14.5±0.46^{ccc}	1.80±0.06^{bbb}
2.0	70	8.3±0.47^{fff}	2.86±0.29^{aaa}
IAA			
0.5	63	8.9±0.23^{eee}	1.04±0.04^{eee}
1.0	90	15.1±0.36^{bbb}	1.85±0.12^{bbb}
1.5	77	13.1±0.50^{ddd}	1.46±0.07^{ccc}
2.0	0	NR	NR
NAA			
0.5	0	NR	NR
1.0	67	5.9±0.37^{ggg}	1.64±0.09^{bbb}
1.5	63	4.9±0.04^{hhh}	1.47±0.07^{ccc}
2.0	0	NR	NR

*NR= No response. Data represents average of three replicates; each replicate consists of 30 cultures and data recorded after 4 weeks of culture. Values are Mean ± S.E. Mean followed by different letters within hormone alone are significantly different at p=0.05, as per Duncan's Multiple Range Test.

increase in the number of shoots was observed with the increasing concentration of auxins IAA and NAA with stable concentration of BAP. Thus synergistic effect of auxins with cytokinins given good results. Similar results are reported in *Momordica dioica* at NAA (1.0mg/l) along with BAP (1.0mg/l) (Kapadia *et al.*, 2018). In contrary to this low concentration of auxin (0.1mg/l IAA) along with 0.5mg/l BAP were given best shoot proliferation in *Momordica dioica* (Chowdary *et al*., 2017). and 2.0mg/l IAA + 0.5mg/l BAP in *Corollocarpus epigaeus* (Narayan, 2016).

Nodal explants cultured on 1.5 mg/l TDZ have shown maximum response (60%) with high number of shoots 5.1 ± 0.23 and shoot length (cm) 2.39 ± 0.06 (Fig-1B). It is observed that when TDZ was used, more callus production and reduction of number of shoots emergence was observed. It is proved that TDZ is not more effective in multiple shoot induction in *Kedrostis foetidissima* than BAP. Results are similar with shoot tip explants of *Ficus carica* var. Black Jack, in which TDZ drastically reduced the number of shoots (Rajendra parab *et al*., 2021). The MS medium supplemented with1.0 mg /l TDZ and 0.4 mg/l IAA given higher percentage (63%) of response with mean number of shoots 6.6 ±0.022 and shoot length (cm) 2.61 ±0.05. The synergistic effect of TDZ with auxin given more effective result than alone TDZ. High response of multiple shoot production was observed with TDZ+IAA when compared to alone TDZ used in *Luffa acutangula* (Yasodhara *et al.*, 2016). The MS medium enriched with 1.0mg /l TDZ and 0.6mg /l NAA induced a more response (60%) with a 5.6±0.29 mean number of shoots and shoot length (cm) 2.82 ± 0.25. Similarly the combination of TDZ+NAA given better result in case of *Pterocarpus marsupium* (Tippani *et al.*, 2021).

Our study has proved that present results among the different treatments of cytokinins and auxins used either singly or in combinations, MS medium with BAP alone found highly effective in shoot bud initiation and multiplication in *Kedrostis foetidissima*. The MS-medium supplemented with cytokinin alone (BAP) induced multiple shoot formation was observed in nodal and hypocotyl explants of *Lycium barbarum* (Karakas, 2020), *Bupleurum distichophylum* (Karuppuswamy & Pullaiah, 2007), *Marsdenia bruniana* (Ugraiah *et al*.,2010).

B. In vitro rooting and acclimatization of cloned plantlet

Well grown shoot lets were shifted to half strength MS medium fortified with various auxins IAA, NAA, and IBA (0.5-2.0mg/l). After 3 weeks of incubation root formation was observed. Varied response to the rooting in the auxins tested in our study is shown in Table-II. The data for percentage of formation of roots per shoots recorded after 4 weeks. Shootlets cultured in MS half strength medium with 1.0mg/l IBA responded to maximum percentage (93%) with highest mean number of roots 17.9±0.31 (Fig-1D). However, the highest mean root length (cm) 2.86± 0.29. was achieved in IBA 2.0mg/l. Shoot lets cultured on IAA 1.0mg/l have given maximum percentage of response (90%) with optimum mean number of roots 15.1±0.36, maximum root length (cm) of 1.85± 0.12. Shootlets cultured on NAA 0.5, 2.0mg/l did not respond for rooting but in 1.0, 1.5mg/l of NAA roots were produced. IAA, IBA produced thin thread like roots whereas NAA produced short thick roots. IBA was more effective in induction of *in vitro* rooting of shoot lets in *Kedrostis foetidissima* with highest mean number of roots (17.9±0.3). Similar to these results 1.0mg/l IBA more effective in rooting in *Trichosanthes cucumerina* (Devendra *et a.l*, 2008), *Coccinia grandis* (Mohammad *et al.*, 2020). In contrary to this NAA proved effective in rooting in *Philodendron bipinnatifidum* (Alawaadh *et al.*, 2020), *Momordica balsamina* (Thakur *et al.*, 2011). But in *Costus pictus* 1.5mg/l IAA given higher rooting response (Ahmed & Arunkumar, 2009).

The well-developed plantlets of *Kedrostis foetidissima* were successfully transferred into plastic cups containing sand and black soil 1:1 ratio (Fig-1 E). They were kept in an incubation room for 2 weeks under 16/8 light and dark period. After that they were shifted to green house with a survival rate of 75%. After 2 months hardened plants were shifted from greenhouse to field with survival rate of 68% (Fig-1 F). The acclimatized plants

were typical, healthy and morphologically alike to parent plants.

CONCLUSION

The results thus proved that auxiliary shoot buds of nodal segments have the potential to regenerate and act as a source of propagule for regeneration in a short span of time. The present study provides reliable *in vitro* regeneration protocol from nodal explants of *Kedrostis foetidissima*. Over exploitation of medicinal plants will result in extinction of species in future. As this plant is a potential source of phytoconstituents like cucurbitacins, which has the capacity of anti-inflammatory and anti-cancer properties. For sustainable production and conservation of this plant, this standardized protocol could help in *in vitro* regeneration of this plant.

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REFERENCES

Alam M, Amin R, Uddin M, Biswas S, Islam M. (2015). Regeneration of shoot from nodal explants of *Cucumis sativus* considering different hormonal concentration. International Research Journal of Biological Sciences, 4(7),48-52

Alawaadh, A. A., Dewir, Y. H., Alwihibi, M. S., Aldubai, A. A., El-Hendawy, S., & Naidoo, Y. (2020). Micropropagation of lacy tree philodendron (*Philodendron bipinnatifidum* Schott ex Endl.). *HortScience*, *55*(3), 294-299.

Amutha M. (2017). Phytochemical Biological Pharmacological Evaluation of *Kedrostis foetidissima* Jacq. (Cogn) and Plant Mediated synthesis of metal nanoparticles., Ph.D.dessertation ,Avinashilingam Deemed University For Women, http://hdl.handle.net/10603/184784

B.Kunthavi , M.Malarvizhi , A. S. Poongodi Kandhimathi, (2018). Retrospective study of leaves of *Kedrostis foetidissima* in siddha medicine , Int. J. Curr. Res. Biol. Med. 3(5): 18-21

Bakrudeen, A. A. A., & Arun, K. R. (2009). In vitro propagation of Monocot (*Costus pictus* D. Don)—an antidiabetic medicinal plant. *Journal of Agricultural Technology*, *5*, 361-369.

Bhatia, S., Bera, T., Dahiya, R., Bera, T., Bhatia, S., & Bera, T. (2015). Classical and nonclassical techniques for secondary metabolite production in plant cell culture. *Modern Applications of Plant Biotechnology in Pharmaceutical Sciences*, 231-291.

Borah AR, Anbumalarmathi J, Sharmili SA. (2019). In vitro propagation of Coccinia indica (L.) Voigt. from internodal segments. Indian Journal of Agricultural Research, 53(2), 202-207.

Bulle, M., Kota, S., Rathakatla, D., Aileni, M., Kokkirala, V. R., Gadidasu, K. K., & Abbagani, S. (2012). An efficient in vitro leaf-based regeneration and evaluation of genetic Fidelity using ISSR markers in *Woodfordia fruticosa* (L.) Kurz. *Journal of herbs, spices & medicinal plants*, *18*(2), 178-190.

Choene, M., & Motadi, L. R. (2012). Anti-proliferative effects of the methanolic extract of Kedrostis foetidissima in breast cancer cell lines. *Mol Biol*, *1*(2), 1-5.

Choudhary, S. K., Patel, A. K., Shekhawat, S., & Shekhawat, N. S. (2017). An improved micropropagation system, ex vitro rooting and validation of genetic homogeneity in wild female *Momordica dioica*: an underutilized nutraceutical vegetable crop. Physiology and Molecular Biology of Plants, *23*(3), 713-722.

Devendra, N. K., Rajanna, L., Sheetal, C., & Seetharam, Y. N. (2008). In vitro Clonal Propagtion of *Trichosanthes cucumerina* L. var. *cucumerina*. *Plant Tissue Culture and Biotechnology*, *18*(2), 103-111.

Dogra, K. S., Chauhan, S., & Jalal, J. S. (2015). Assessment of Indian medicinal plants for the treatment of asthma. *Journal of Medicinal Plants Research*, *9*(32), 851-862.

Giday, M., Asfaw, Z., Elmqvist, T., & Woldu, Z. (2003). An ethnobotanical study of medicinal plants used by the Zay people in Ethiopia. *Journal of ethnopharmacology*, *85*(1), 43-52.

Jaganathan, G. K., Hoa, T. H. T., & Liu, B. L. (2016). Ethnobotanical survey of Irular tribes in Pillur valley, Coimbatore, Tamil Nadu (India). *International Journal of Herbal Medicine*, *4*(1), 1-11.

John Britto S. (2019). The Flora of Central and North Tamil Nadu, The Rapinat Herbarium, St.Joseph`s college (Autonomous), Tiruchirapalli (India); Part-2,

Kapadia, C., Patel, N., Patel, N., Ahmad, T. (2018). Optimization of Culture Medium for Higher Multiplication and Efficient Micropropagation of Spine Gourd (Momordica dioica Roxb.). J. of Experimental Biol. and Agric. Sci., 6(3), 599– 605.

Karakas, F. P. (2020). Efficient plant regeneration and callus induction from nodal and hypocotyl explants of goji berry (*Lycium barbarum* L.) and comparison of phenolic profiles in calli formed under different combinations of plant growth regulators. *Plant Physiology and Biochemistry*, *146*, 384-391.

Karataş, M., & Asim, M. (2015). In vitro whole plant regeneration of medicial aquatic plant-*Limnophilla aromatica*. *Fresenius Environmental Bulletin*, *24*, 1-4.

Karuppusamy S, Pullaiah T. (2007). In vitro Shoot Multiplication of *Bupleurum disticho- phyllum* Wight-A Native Medicinal Plant of Southern India. Plant Tissue Culture and Biotechnology, 17(2),115-24.

Karuppusamy, S. (2007). Medicinal plants used by Paliyan tribes of Sirumalai hills of southern India.

Kawale, M. V., & Choudhary, A. D. (2009). In vitro multiple shoot induction in *Trichosanthes cucumerina* L. Indian J Plant Physiol. *14*(2), 116-123.

Mali, A. M., & Chavan, N. S. (2016). In vitro rapid regeneration through direct organogenesis and ex-vitro establishment of *Cucumis trigonus* Roxb.—An underutilized pharmaceutically important cucurbit. *Industrial Crops and Products*, *83*, 48-54.

Mohammad Amin, A .V. D. Dorajee Rao, K. Ravindra Kumar and Salomi suneetha, D.

Mohan, V. R., & Kalidass, C. (2010). Nutritional and antinutritional evaluation of some unconventional wild edible plants. *Tropical and subtropical Agroecosystems*, *12*(3), 495-506.

Mustafa, M., Swamy, T. N., Raju, S., & Mohammad, S. P. (2013). Multiple shoot induction from the nodal cultures of teasle gourd (*Momordica dioica* Roxb.). *Int. J. Biosci*, *3*, 8-12.

Narayan, J. P. (2016). Ex-situ conservation of the rare and threatened medicinal climber Corallocarpus epigaeus Rottler through in vitro regeneration method. *Biotechnology Journal International*, 1-10.

Nirmala, J., & Pandian, R. (2013). Morphological and systematic studies on *Kedrostis foetidissima* (Jacq.) Cogn. Int. J. Curr. Microbiol. App. Sci. *2*(10), 560-571.

Njoroge, G. N., & Newton, L. E. (1994). Edible and poisonous species of Cucurbitaceae in the Central Highlands of Kenya. *Journal of East African Natural History*, *83*(2), 101-115.

Parab, A. R., Chew, B. L., Yeow, L. C., & Subramaniam, S. (2021). Organogenesis on apical buds in common fig (Ficus carica) var. Black Jack. *Electronic Journal of Biotechnology*, *54*, 69-76.

Patil SM, Kumari VC, Sumana K, Sujay S, Tejaswini M, Shirahatti PS, Ramu R. (2021).Sustainable development of plant tissue culture industry: The Indian scenario. Journal of Applied Biology & Biotechnology, 9(2), 18-27.

Pavithra, K., & Vadivukkarasi, S. (2015). Evaluation of free radical scavenging activity of various extracts of leaves from *Kedrostis foetidissima* (Jacq.) Cogn. Food Science and Human Wellness. *4*(1), 42-46.

Priyavardhini, S., Gowri, S. S., Vasantha, K., & Umadevi, M. (2008).Antibacterial activity of stem and leaf extract of *Kedrostis foetidissima* (Jacq.) Cogn. Ancient science of Life. *28*(2), 10.

Priyavardhini, S., Vasantha, K., Tresina Soris, P., & Mohan, V. R. (2012) . Antifungal activity of *Kedrostis foetidissima* (Jacq.) Cogn. International Journal of Pharm Tech Research, 4(1),44-48.

R. (2020). Effect of in vitro Rooting in *Coccinia grandis* (L.) through Nodalsegments.*Int.J.Curr.Microbiol.App.Sci.* 9(7), 2266-2272.

Reddy, K. N., & Reddy, C. S. (2016). *Flora of Telangana State, India*. Bishen Singh Mahendra Pal Singh.

Renner, S. S., & Pandey, A. K. (2013). The Cucurbitaceae of India: Accepted names, synonyms, geographic distribution, and information on images and DNA sequences. *PhytoKeys*, (20), 53.

Ríos, J. L., Escandell, J. M., & Recio, M. C. (2005). New insights into the bioactivity of cucurbitacins. *Studies in natural products chemistry*, *32*, 429-469.

Shasthree, T., Caroline, V. J. E., Savitha, R., & Mallaiah, B. (2012). Micropropagation of *Bryonopsis laciniosa* (L) Naud an endangered and medicinally important cucurbit. *Advances in Plant Sciences*, *25*(2), 373-376.

Sivaprakash, V. (2008). *Antidiarrhoeal Activity of Kedrostis Foetidissima Leaf Extract on Experimentally Induced Diarrhoea in Mice* (Doctoral dissertation, Nandha College of Pharmacy, Erode) [Internet] Available from

Suganthi.Aand S.Poornima (2014). In vitro plantlet regeneration from internode explants of *Kedrostis foetidissima*(Jacq) Cogn –An Important rare anticancer medicinal climber. International Journal of Innovative Pharmaceutical Sciences and Research, 2(4), 802 -811.

T. Elavazhagan (Doctoral thesis),Studies On Phytochemical Antimicrobial And Anticancer Activity Against *Kedrostis Foetidissima* Jacq Cogn http://hdl.handle.net/10603/230788

Thakur, G. S., Sharma, R., Sanodiya, B. S., Pandey, M., Baghel, R., Gupta, A., & Bisen, P. S. (2011). High frequency in vitro shoot regeneration of *Momordica balsamina*, an important medicinal and nutritional plant. *African Journal of Biotechnology*, *10*(70), 15808-15812.

Thiruvengadam, M., Chung M-III, and Se-chun Chun. (2012).Influence of polyamines on in- vitro organogenesis in bitter melon (*Momordica charantia* L.). J. of Medicinal Plants Research, 6(19), 3579-3585.

Tippani, R., Thammidala, C. (2021).TDZ induced plant regeneration from immature cotyledons of *Pterocarpus marsupium* Roxb. and validation of genetic homogeneity using ISSR markers. *Vegetos,* **34,** 144–152

Ugraiah, A., Karuppusamy, S., & Pullaiah, T. (2010). Micropropagation of *Marsdenia brunoniana* Wight & Arn. - A Rare Antidiabetic Plant. *Plant Tissue Culture and Biotechnology*, *21*(1), 89-93.

Velivela, Y., Narra, M., Ellendula, R., Kota, S., & Abbagani, S. (2016). Establishment of in vitro regeneration from petiole explants and assessment of clonal fidelity by ISSR markers in Luffa acutangula L. Roxb. *J Appl Biol Biotechnol*, *4*, 41-45.

Vemula, S., Koppula, T., Jogam, P., & Mohammed, M.(2020). In vitro high frequency multiplication and assessment of genetic fidelity of *Corallocarpus epigaeus*: an endangered medicinal plant. Vegetos, *33*(1), 63-73.

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