



In vitro propagation of valuable Bromeliad's: A review.

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Abstract

Bromeliad is a plantlet having ornamental, medicinal and commercial valued. This review briefly summarizes the research study conducted on *in vitro* tissue culturing of bromeliads. Numerous factors are key for successful micropropagation such as period of explant collection, size of explants, concentration and combination of Plant growth regulators, culture medium and cultural conditions. Sequential study of experiments helpful for understanding biological, physical and physiochemical aspects that govern the *in vitro* morphogenesis of bromeliad. *In vitro* propagation is an alternative way of propagation that can working in bulk multiplication of plants in lesser time and required low space which is can be grown in any condition.

Keywords: *in vitro*, Bromeliad's, morphogenesis, problems.

Introduction

Bromeliads are flowering plants which have 77 genera with 3629 species native to tropical areas distributed throughout the world (Butcher and Gouda, 2020). About 40% of the bromeliads are found in brazil (Anonymous, 2023). In vascular epiphytes it is a biggest family in biome (Kersten, 2010). Bromeliads are known for colored leaves and their flower spikes. They are plant species which can be grown anywhere such indoor as home decorative and also outside in the environment.

The bromeliads leaf colour, leaf shape and growth can be affected by the amount of light given to the plant, considering water and moisture they belong to the tropical areas so must avoid over watering the bromeliads plantlets. The bromeliads are the plants which are free from pests. i.e. they are not usually affected by pests but mealy bugs can create problem to the plantlets.

Bromeliads provide a range of ecosystem services. It maintains biodiversity, nutrient cycling, community structure, and provisioning of food and water. Bromeliads can grow all over the year, it is not season specific. Bromeliads flowers last for last for many months, depending on the age of the plant, they require minimal care. Most bromeliads are native to tropical areas so indoor plants does best when kept away from drafts and at temperature above 55 degrees. Bromeliads can get their nutrients from their leaves.

Bromeliads even grow on rocks or trees as they use their roots for balance rather than transferring nutrients. These plants feature rosettes of spiny stiff leaves and thrive in consistent warm indoor temperatures. They require less care. They are easy to grow and produce long lasting colorful leaf crowns. These plants add significant intrigue and beauty to your home décor.

To preserve the bromeliads there are techniques like biotechnological techniques from seed germination to slow growth and it can give the ideal condition for conservation (Simao *et al.*, 2016). To minimize the extraction of high number of bromeliads which are rare and disappearing varieties there are techniques used which are *in vitro* techniques can be used for the propagation. *In vitro* propagation plays an important role for the bromeliad. By using *in vitro* technique's multiplication of endangered species easily achieves. Using traditional method's we can produce less number of plants. Ornamental bromeliad such as *Aechmea*, *Tillasanda*, *Alcantarea nahoumii*, *Aechmea fasciata*, etc. can be produced in larger number for ornamental use such as decorative purpose. By traditional methods we can produce only limited number of plantlets but by using *in vitro* techniques we can produce larger number of copies at a time.

There are some problems in *in vitro* establishment of bromeliads such as contamination of fungal and bacterial agents etc. They can largely affected by bacteria. Fungus and bacteria can destroy whole culture.

In vitro establishment problems and solutions

While establishment there are some problems are faced. The main problem faced while *in vitro* establishment is contamination in the culture medium. The contamination can be fungus, bacteria, yeasts and molds. Browning of media can caused due to oxidation of phenolic compounds it slows down the rate of the cell division and it can lower down capacity of regeneration of a explant that results death. Acclimatization of tissue culture plantlets, the tissue cultured plant should adaptable to external environment otherwise the plants will get shock and will die if proper environment not given. Recalcitrance – it is defined as the plants which are not able to grow in *in vitro* culture environment. Vitrification- It is a physical disorder or malformation can cause effect on growth of plants and its development. The factors by which it causes are Higher salt concentrations, light intensity, relative humidity, type of explants.

Table 1: Globally well-known Bromeliads literatures and their outcomes.

Sr. no.	Varieties	Medium combinations			Results		Other additives	Culture condition	Reference
		Medium	Cytokinin	Auxin	Cytokinin	Auxin			
1.	<i>Aechmea setigera</i>	Murashige and Skoog's (MS, 1962) medium	BAP 0.0 to 8.0 mg l ⁻¹	-	BAP 8.0 mg L ⁻¹	-	-	Silva <i>et. al.</i> , 2022	
2.	<i>Alcantarea nahoumii</i>	MS medium	BAP 0.0 to 6.6 µM	NAA 0.5 µM	BAP 2.2 µM	NAA 0.5 µM	-	Temp. 25°C Silva <i>et. al.</i> , 2020	
3.	<i>Chapada Diamantina</i>	MS medium	BAP 0.0 to 13.20 µM	NAA 0.0-5.20 µM	BAP 5 µM	NAA 3 µM	Charcoal	Temp 25±3°C and light 60 µmol.m ⁻² s ⁻¹ Lima <i>et. al.</i> , 2019	
4.	<i>Vriesea gigantea and philippicoburgii</i>	MS medium	BAP 2.0 mg/l	NAA 0.5 mg/l	BAP 2.0 mg/	NAA 0.5 mg/l	-	Temp 26±1°C Light 22.5 µEm ⁻² s ⁻¹ Droste <i>et. al.</i> , 2005	

5.	<i>Aechmea fasciata</i>	MS medium	KIN 1.0 mgL ⁻¹	IAA 1 mg/l ⁻¹ NAA 0-2.0 mgL ⁻¹	KIN 1.0 mgL	IAA 1mgL NAA 2 mgL	Charcoal	Temp 25±2°C	Vinterhalter <i>et. al.</i> , 1992
6.	<i>Neoglaziovia variegata</i>	MS medium	BAP 2.2 to 4.4 µM KIN 2.2 to 4.4 µM	NAA 0.05- 0.50 µM	BAP 4.4 µM KIN 2.2 µM	NAA 0.5 µM	-	Temp 22±1°C Light 22 µE.m ⁻² .s ⁻¹	Silveira <i>et. al.</i> , 2009
7.	<i>Ananas comosusl. merr</i>	MS medium	BA 0-7 mgL ⁻¹	NAA 2 mgL ⁻¹	BA 5 mgL ⁻¹	NAA 2 mgL ⁻¹	-	Light 16/8hrs Temp 25±2°C	Farahani <i>et. al.</i> , 2014
8.	<i>Aechmea fasciata (lindl)</i>	MS medium	BA 5.0 mgL ⁻¹ ZEA 5.0 mgL ⁻¹ KIN 0 to 0.5 mgL ⁻¹	2-4 D 2 mgL ⁻¹ IAA 0-0.4 mgL ¹ NAA 2.0 mgL ⁻¹	BA 5.0 mgL ⁻¹ ZEA 5.0 mgL ⁻¹	NAA 2.0 mgL ⁻¹ 2-4 D 2.0 mgL ⁻¹	-	-	Villanueva <i>et. al.</i> , 2022
9.	<i>Vriesea reitzii</i>	MS Medium	KIN 1.0 µM BAP 4.0 µM	2-4-D - 20.0 µM 2 iP – 2.5 µM NAA 0.5 µM	KIN 1.0 µM BAP 2.5 µM	2-4-D - 20.0 µM NAA 0.5 µM iP- 2.5 µM	-	Temp 25±1°C Light 37µE/m ² /s 16hr	Alves <i>et. al.</i> , 2006
10.	<i>Vriesea fosteriana and Hiaroglyphi ca</i>	Knudson medium (1946)	-	NAA 0.2 mgL ⁻¹ IBA 5.0 mgL ⁻¹	-	NAA 0.5 mgL ⁻¹ IBA 2.0 mgL ⁻¹	-	-	Mercier <i>et. al.</i> , 1995
11.	<i>Ananas comosus L.</i>	MS medium	BA 5 to12.5 µM	NAA 2 to 3 µM	BA 5.0 µM	NAA 3.0 µM	Lysergic acid dieethylam ide	Temp 27±2°C	Usman <i>et. al.</i> , 2013
12.	<i>Cryptanthus</i>	MS medium	-	NAA 0.1 to 0.2 mgL ⁻¹ IBA 2.0 mgL ⁻¹	-	IBA 2.0 mgL ⁻¹ NAA 0.1 mgL ⁻¹	Phlorogluc i-nol, ancymidol, fusicoccin, riboflavin, ascorbic acid	Light 6/8hrs Temp 25±2°C Irradianc e 5.0- 7.2Wm ⁻²	Todorovic <i>et. al.</i> , 1995

13.	<i>Succulent plants</i>	MS medium	BAP 1 to 2 mg ^l ⁻¹	NAA 0.01 to 1.0 mg ^l ⁻¹	BAP 1 mg ^l ⁻¹	NAA 0.1 mg ^l ⁻¹	Chlorine, sodium hypochloride	21°C at day and 16 °C at night	Gratton <i>et. al.</i> , 1999
14.	<i>Tiliandsia Eizii</i>	Knudson medium (1946)	BA 0.5 mg ^l ⁻¹	NAA 1mg ^l ⁻¹	BAP 0.5 mg ^l ⁻¹	NAA 1mg ^l ⁻¹	Glycine	-	Pickens <i>et. al.</i> , 2005
15.	<i>Naoregela cruenta</i>	MS medium	BA 4.4 to 8.8 μM	NAA 2.5μM	BA 4.4 μM	NAA 2.5μM	-	Temp 28±2°C Light 16 hrs Irradiation 46 μmol m ² s ⁻¹	Carneiro <i>et. al.</i> , 1999
16.	<i>Vriesea Bromeliads</i>	MS medium	BAP 0.5 mg ^l ⁻¹	-	BAP 0.5 mg ^l ⁻¹	-	-	Temp 27±2°C Light 16 hrs intensity - 40 μmol m ² s ⁻¹	Aranda-Peres <i>et. al.</i> , 2009
17.	<i>Pineapple</i>	MS medium	BAP 4 μl	NAA 2μl	BAP 4 μl	NAA 2μl	-	Temp 27±2°C Light 16 hrs 37 μmol m ² s ⁻¹ 60% relative humidity	Vesco <i>et. al.</i> , 2001
18.	<i>Vriesea reitzii</i>	MS medium	BAP 2 to 4 μM	NAA 2 to 4 μM IAA 4μM IP-2 μM	BAP 4 μM	NAA 4 μM IAA 4 μM IP-2 μM	Gibberellic acid	Temp 25±2°C Light 16 hrs. Luminous intensity 50-60 μmol m ² s ⁻¹	Vesco <i>et. al.</i> , 2015

19.	<i>Passiflora edulis Sims</i>	MS medium	BAP 1.0 mg ^l ⁻¹	-	BAP 1.0 mg ^l ⁻¹	-	-	Temp 25±2°C	Vesco, <i>et. al.</i> , 2021
20.	<i>Dyckia Distachya</i>	MS medium	-	IP 12 µM NAA 0.5 µM	-	IP 12 µM NAA 0.5 µM	Giberalic acid	Temp 25±2°C Humidity 60±5 Light 16hrs Irradiance 60 µmol m ² s ⁻¹	Pompelli, <i>et. al.</i> , 2005
21.	<i>Dyckia Distachya</i>	MS medium	BA 4 µM BAP 2 µM	NAA 2 µM	BA 4 µM BAP 2 µM	NAA 2 µM	Paclobutrazol	-	Pompelli, <i>et. al.</i> , 2005
22.	<i>Bromiloides</i>	MS medium	BAP 4 µM	NAA 2 µM	BAP 4 µM	NAA 2 µM	Gibberellic acid	Temp 25±2°C Irradiance 50 - 60 µmol m ² s ⁻¹	Guerra <i>et. al.</i> , 2014
23.	<i>Aechmea fasciata</i>	MS Medium	-	2-4-D 1.0 to 1.5 mg ^l ⁻¹ NAA 1.0 to 0.5 mg ^l ⁻¹	-	2-4-D 1.0 mg ^l ⁻¹ NAA 1.0 mg ^l ⁻¹	Arginine, Aspergin, Citric acid	Temp 25±2°C	Huang <i>et. al.</i> , 2011

24.	<i>Guzmanie</i>	MS medium	-	2-4-D 1.0 to 2.0 mg ⁻¹ NAA 0.5 to 1.0 mg ⁻¹ IAA 0.1 to 1.0 mg ⁻¹	-	2-4-D 2.0 mg ⁻¹ NAA 1.0 mg ⁻¹ IAA 1.0 mg ⁻¹	-	Temp 25±2°C Light 16 hrs Irradiance 50 μmol m ² s ⁻¹	Huang <i>et. al.</i> , 2011
26	<i>Sphaerant- hus amarantho- ides</i>	MS medium	BAP 1.5 to 5.0 mg ⁻¹ KN 1.0 to 5.0 mg ⁻¹	IBA 1.0 to 5.0 mg ⁻¹	BAP 4.0 mg ⁻¹ KN 2 mg ⁻¹	IBA 2.0 mg ⁻¹	-	-	Devika <i>et. al.</i> , 2012
27	<i>Huarlia hystrix</i>	MS medium	BA 4.44 to 22.19 μM	NAA 0.00 to 8.06 μM	BA 22.19 μM	NAA 5.37 μM	-	Temp 25°C Light 30 μmol m ² s ⁻¹	Amoo <i>et. al.</i> , 2009
28	<i>Cryptanthus spp.</i>	MS medium	-	IBA 0.0 to 10 mg ⁻¹	-	IBA 2 mg ⁻¹	Citric acid	-	Davidson <i>et. al.</i> , 1977
29.	<i>Dyckia Agudensis</i>	MS medium	BAP 0.0 to 2.0 mg ⁻¹	IBA 0.0 to 2.0 mg ⁻¹	BAP 1.0 mg ⁻¹	IBA 1.0 mg ⁻¹	-	Temp 25±2°C Light 16 hrs . intensity 14.3 μEm ² s ⁻¹	Silva <i>et. al.</i> , 2007
30.	<i>Nidularium procerum</i>	MS medium	BAP 0.0 to 8.0 μM	NAA 0.0 to 5.4 μM	BAP 4.0 μM	NAA 2.0 μM	Naphthale ne acid	Temp 25±2°C Light 16 hrs. Intensity 28 μMm ² s ¹	Silva <i>et. al.</i> , 2012

31.	<i>Nidularium Immoentii</i>	MS medium	BAP 0.0 to 8.0 μM	NAA 0.0 to 5.4 μM	BAP 8.0 μM	NAA 2.0 μM	-	Temp 25±2°C Light 16 hrs Intensity 28 $\mu\text{Mm}^2\text{s}^{-1}$	Silva <i>et. al.</i> , 2012
33.	<i>Acacia auriculiformis</i>	MS medium	BA 0.44 μM	IBA 0 to 9.84 μM NAA 0.0 to 10.74 μM	BA 0.44 μM	IBA 9.84 μM NAA 5.37 μM	-	Temp 25±2°C Light 16hrs	Ismail <i>et. al.</i> , 2016
34.	<i>Billbergia zebrina</i>	MS medium	BA 13 μM	-	BA 13 μM	-	-	-	Martins <i>et. al.</i> , 2022
35.	<i>Dyckia brevifolia</i>	MS medium	BA 0.5 to 1.0 mg l^{-1} KIN 0.5 to 1.0 mg l^{-1}	IP 1.0 mg l^{-1}	BA 1.0 mg l^{-1} KIN 1.0 mg l^{-1}	IP 1.0 mg l^{-1}	-	Temp 25°C Light 16hrs Intensity 37.5 $\mu\text{mol m}^2\text{s}^{-1}$	Bertsouklis <i>et. al.</i> , 2022
36.	<i>Bromelia balansae mez</i>	MS medium	BAP 0 to 4 μM	NAA 1 to 4 μM	BAP 4 μM	NAA 2 μM	-	-	Faria Souza <i>et. al.</i> , 2023
37.	<i>Aechma vulnerable and Aechmea distinchantha</i>	MS medium	BAP 2.2 μM	NAA 0.5 μM	BAP 2.2 μM	NAA 0.5 μM	-	Temp 25±2°C 16hrs Light Intensity 30 $\mu\text{mol m}^2\text{s}^{-1}$	Santa-Rosa, <i>et. al.</i> , 2012

Conclusion:

It is concluded that, most of aseptic cultures are initiated from newly emerging shoot propagules, apical buds and leaves. Therefore, these explants from healthy mother plant are chosen. During surface sterilization of explants, polysorbate Tween-20 treatment given to the explant to removal of proteins and which also reduces surface tension of explants, which enhances the sterilant to act more deeply on explants. Use of the contact fungicides at 200 mg/l ex. Mancozeb and systemic carbendaxim can clear the fungal contamination. Similarly, systemic bactericide can play important role in bacterial control. H₂O₂ solution play role in the removal of the waxy layer of the explant. Sodium hypochloride with reduced concentration can be used for sterilization of explant for 20 mns to 2 hours, depending upon the type of explants.

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