



OPEN ACCESS

EDITED BY

Ambika Rajendran,
Indian Agricultural Research Institute
(ICAR), India

REVIEWED BY

Anjana Rustagi,
University of Delhi, India
Ankita Rajendra Parab,
University of Science Malaysia (USM),
Malaysia
Ahmed Madi Waheed Al-Mayahi,
University of Basrah, Iraq
MuraliKrishna Narra,
National Research Council Canada (NRC),
Canada
Jessica Antony,
Penerbit UPM, Malaysia

*CORRESPONDENCE

Aveek Samanta
✉ aveekbot@gmail.com

RECEIVED 07 November 2025

REVISED 20 February 2026

ACCEPTED 28 February 2026

PUBLISHED 17 March 2026

CITATION

Maity TR, Datta S, Bhowmik A,
Samanta S, Paria K and Samanta A (2026)
Development of a portable plant tissue
culture box: an eco-friendly solution for
tissue culture laboratories.
Front. Hortic. 5:1741481.
doi: 10.3389/fhort.2026.1741481

COPYRIGHT

© 2026 Maity, Datta, Bhowmik, Samanta,
Paria and Samanta. This is an open-access
article distributed under the terms of the
[Creative Commons Attribution License
\(CC BY\)](https://creativecommons.org/licenses/by/4.0/). The use, distribution or
reproduction in other forums is
permitted, provided the original
author(s) and the copyright owner(s) are
credited and that the original publication
in this journal is cited, in accordance
with accepted academic practice. No
use, distribution or reproduction is
permitted which does not comply with
these terms.

Development of a portable plant tissue culture box: an eco-friendly solution for tissue culture laboratories

Tilak Raj Maity¹, Siraj Datta², Atanu Bhowmik^{1,3}, Subir Samanta³,
Kislaya Paria⁴ and Aveek Samanta^{3*}

¹Department of Biotechnology, Haldia Institute of Technology, Haldia, West Bengal, India,

²Subarnarekha Mahavidyalaya, Jhargram, West Bengal, India, ³Department of Botany, Prabhat Kumar College, Contai, West Bengal, India, ⁴Department of Biotechnology, Oriental Institute of Science and Technology, Midnapore, West Bengal, India

Plant tissue culture (PTC) traditionally requires a specialized culture room with racks, controlled temperature, and regulated light and dark cycles to support *in vitro* plant growth. Establishing and maintaining such a facility is expensive and demands skilled personnel. To address these challenges, we have developed a portable PTC box that provides an artificially controlled environment within a compact chamber. This system maintains a temperature range of 20–30 °C and can simulate a cool environment suitable for plant *in vitro* culture. Its simple infrastructure and eco-friendly design make it an accessible alternative to conventional culture rooms. The portable PTC box eliminates the need for dedicated laboratory space and can be easily adapted to specific requirements. Unlike traditional air-conditioning systems that rely on harmful gases such as carbon monoxide or chlorofluorocarbons, this device offers an eco-friendly cooling mode. Light-emitting diodes (LEDs) provide efficient illumination with minimal energy consumption while maintaining the required light and dark cycles. Preliminary evaluations indicate that plantlets grown in this box exhibit chlorophyll content, fresh weight, dry weight, moisture content, and molecular levels comparable to those grown in standard *in vitro* culture laboratories. This innovation demonstrates a sustainable, cost-effective, and portable solution for PTC, making it suitable for small laboratories, educational institutions, and field applications. The portable box has significant potential to expand access to PTC techniques while reducing environmental impact and operational costs.

KEYWORDS

controlled environment, cost effective, genetic fidelity, *in vitro* morphogenesis, Peltier-based cooling

1 Introduction

The growth and multiplication of plant cells, tissues, and organs are facilitated by the application of plant tissue culture (PTC) techniques under strictly maintained aseptic conditions and within a precisely controlled environment (Dutta Gupta and Jatothu, 2013; Agarwal and Dutta Gupta, 2016; Reddy, 2024). A regulated environment is established to provide essential growth parameters, including optimized light intensity, temperature

stability, humidity control, and a synthetic medium with precisely adjusted pH and gas exchange. Plant tissue culture is an emerging technology in both agricultural and industrial sectors in recent years (Agarwal and Dutta Gupta, 2016; Gulzar et al., 2020; Twaij et al., 2020; Altaf et al., 2024).

PTC techniques are fundamental to numerous academic studies as well as various applied fields within plant science. Culture tubes, jars, plates equipped with racks, a lighting system (to maintain light-dark cycles), temperature monitoring devices, and an air conditioning system are normally kept in a culture room (Dutta Gupta and Jatothu, 2013; Trigiano and Gray, 2016; Reddy, 2024). Plant tissue culture techniques have been used in academic investigations of totipotency and the roles of hormones in cyto-differentiation and organogenesis. PTC techniques are also central to innovative areas of applied plant science, including plant biotechnology and agriculture (Altman, 2003; Trigiano and Gray, 2016). Plants can be cloned; cultured and genetically engineered cells can be produced to form transgenic whole plants through tissue culture procedures. The establishment of a conventional plant tissue culture facility involves significant capital investment and the recruitment of highly skilled personnel. For many academic and research institutions, the substantial financial burden associated with both the construction and the long-term maintenance of such laboratories often proves prohibitive (Trigiano and Gray, 2016).

Plant tissue culture has emerged as a critical tool in modern plant science, offering controlled *in vitro* conditions for studying plant growth, development, and genetics. In research, it facilitates the investigation of gene expression, metabolic pathways, and plant-pathogen interactions, while enabling the production of phytochemicals and secondary metabolites for pharmaceutical and industrial applications (Bhatia et al., 2015; Gunaseelan et al., 2025). Tissue culture allows the generation of pathogen-free plants, ensuring reliable experimental materials. Plant tissue culture plays a pivotal role in *ex situ* preservation of rare, endangered, and threatened plant species. It allows for the maintenance of germplasm, rapid regeneration of species that are difficult to propagate conventionally, and restoration of degraded ecosystems through the production of healthy plantlets. By supporting biodiversity protection and sustainable resource management, tissue culture techniques have become indispensable tools in conservation biology (Reed et al., 2011).

In agriculture, tissue culture techniques such as micropropagation enable the rapid multiplication of high-yielding, uniform, and disease-free planting material. This supports the large-scale production of elite crop varieties and contributes to crop improvement programs by facilitating the introduction of traits such as pest resistance, drought tolerance, and enhanced nutritional quality (Mondal et al., 2016). Additionally, tissue culture allows the propagation of seedless fruits, ornamental plants, and the clonal propagation of superior genotypes, ensuring consistency and productivity in agricultural systems.

Traditional PTC laboratories, while essential for *in vitro* propagation, are associated with several inherent limitations. High establishment and operational costs represent a major barrier, as these laboratories require sophisticated infrastructure, including climate-controlled culture rooms, autoclaves, laminar airflow

cabinets, and specialized shelving systems. Maintaining such facilities demands continuous financial investment and skilled technical personnel (Reddy, 2024).

Energy consumption is another significant concern. Conventional tissue culture rooms rely heavily on air conditioning, artificial lighting, and humidity control systems, which not only increase operational costs but also contribute to environmental impacts through high electricity usage and greenhouse gas emissions. Additionally, contamination risk remains a persistent challenge. The *in vitro* environment is highly susceptible to microbial contamination from air, instruments, or personnel, which can compromise culture success rates, reduce plantlet quality, and increase labor and resource expenditure for repeat experiments. These limitations highlight the need for cost-effective, energy-efficient, and portable alternatives that can maintain controlled environmental conditions, minimize contamination, and reduce dependence on extensive infrastructure, thereby making plant tissue culture more accessible and sustainable (Shukla et al., 2017; Samanta et al., 2023). The present study proposes a low-cost portable plant tissue culture box that requires less maintenance. The primary aim of this study is to develop and validate a cost-effective, eco-friendly portable culture box equipped with a Peltier-based cooling system, thereby bypassing the high costs of traditional laboratory infrastructure. The objective is to evaluate the system's efficacy by comparing the morphological, biochemical, and molecular parameters of *in vitro* grown plantlets against those maintained in standard culture facilities.

2 Materials and methods

2.1 Design and construction of the portable plant tissue culture box

The article describes a novel design of a low-temperature portable plant tissue culture box (PPTCB) (Figure 1). The detailed technical specifications of the PPTCB are summarized in Table 1. This system can create an environment suitable for callus and tissue culture that have specific temperature requirements (Samanta et al., 2023). The temperature can be controlled between 20°C and 30°C. The portable culture unit was constructed with dimensions of 18 × 18 × 24 inches. These specific proportions were selected to optimize the surface-area-to-volume ratio, ensuring that the Peltier cooling module could maintain a stable internal temperature of 20°C to 30°C with minimal power consumption. It has an environment-friendly cooling mode, as it does not need an air-conditioning system that uses harmful gases during operation. In the system, the *in vitro* plants and callus culture can be maintained properly. The system contains a Peltier cooler (TEC1-12706 Thermoelectric Peltier Cooler Module 12V 6A), a thermoelectric module that is used for creating low temperature. With fixed voltage and current, the Peltier module can be heated and cooled. The thermal regulation assembly operates within a closed-loop system, utilizing a custom-

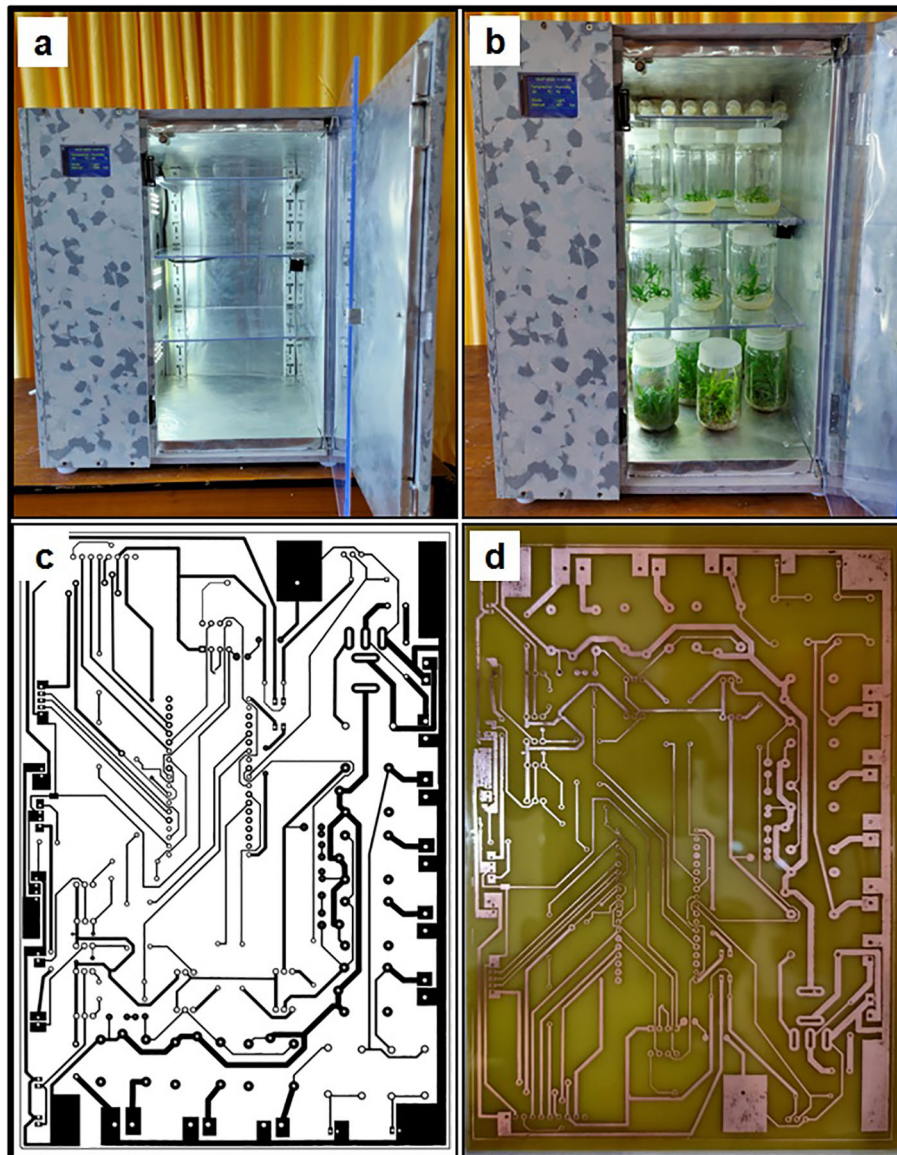


FIGURE 1

Development of the plant tissue culture (PTC) system: (a, b) external and internal views of the assembled portable PTC box prototype; (c, d) stages of printed circuit board (PCB) design and fabrication using the chemical etching technique.

configured thermoelectric (Peltier) module for primary temperature control. The Peltier modules are arranged such that the heat-rejecting junction constitutes the upper section, while the heat-absorbing junction forms the lower, cold-sink interface. This thermoelectric assembly is integrated with a custom-fabricated hollow aluminium thermal manifold that functions as a heat exchanger. During the cooling phase, ambient air is channelled through lateral openings in this manifold, passing over the cold-sink surface. The resulting cooled air is subsequently directed into the internal chamber using direct current (DC) fans. To manage latent heat removal, condensate forming on the cold-sink surface is collected via integrated piping and drained from the chamber. Heat dissipation from the upper hot side of the Peltier modules is managed by active exhaust fans. These fans exhaust thermal energy through purpose-built aeration channels located in the top lid structure. To supplement the cooling system when ambient

temperatures drop below the required setpoint, auxiliary resistive heating elements are incorporated. These heaters are thermally coupled to the heat-sink assembly to provide necessary thermal energy on demand. To minimize thermal losses and ensure operational stability, the walls are lined with thermal insulation material. The light source within the chamber are light-emitting diodes operating at 5 to 12 V in blue, red, and white (Maity et al., 2016). A digital temperature controller thermostat module is used to maintain the desired temperature. The whole system can be powered by solar energy and electricity.

2.2 Design and fabrication of PCB via chemical etching

The printed circuit board (PCB) was fabricated using a chemical etching process (Zakai et al., 2021). Initially, the circuit layout was

TABLE 1 Technical specifications of portable plant tissue culture box (PPTCB).

Type	Table top, portable
Gross volume & Chamber volume	130 L & 60 L
LED temperature range	20 °C to 30 °C
Temperature controller	Digital PID controller
Heating	Tubular air heaters
Air circulation	Forced air circulation through axial fans
Illumination	Blue, Red & White LED
Intensity of light	100–3000 Lux
Light timer	24x7 weekly digital timer
Interior	Aluminium inner walls Height adjustable shelves
Inner door	Transparent door
Outer door	Insulated solid door with lock & key and magnetic gasket
Body material	Recycled plastic
Power supply	220 Volts/Solar
Additional	Digital display, Camera & UV

designed using computer-aided design (CAD) software and printed onto a transparent sheet (Figure 1). A copper-clad board was cleaned thoroughly with fine abrasive material and solvent to remove grease and oxides, ensuring proper adhesion of the photoresist. A light-sensitive photoresist layer was applied uniformly on the copper surface. The prepared circuit transparency was then aligned over the board, and the assembly was exposed to ultraviolet (UV) light. UV exposure transferred the circuit pattern onto the photoresist, hardening the regions corresponding to the desired conductive pathways. The board was subsequently developed using an appropriate developer solution, which removed the unexposed unpolymerized photoresist, thereby exposing the underlying copper in areas to be etched. The board was then immersed in a chemical etchant, such as ferric chloride (FeCl₃), which selectively dissolved the unprotected copper, leaving behind the intended circuit pattern. After etching, the remaining photoresist was stripped using a solvent, and the board was rinsed and dried to remove residual chemicals. The final PCB revealed the completed conductive pathways, ready for component assembly and testing.

2.3 Explants and *in vitro* culture establishment and maintenance

The 'Queen' variety of pineapple (*Ananas comosus* L. family Bromeliaceae) was collected from the experimental garden of the Department of Biotechnology, Nagaland University, Dimapur, Nagaland, India, and grown in the laboratory experimental greenhouse for further use. After aseptic sterilization, apical meristems were inoculated on solid Murashige and Skoog (MS) medium containing 3% (w/v) sucrose and 0.8% (w/v) agar. These media were supplemented with 3 mg L⁻¹ BAP (6-benzylaminopurine) and 0.5 mg L⁻¹ IAA (indole-3-acetic acid) for

establishment and multiplication of pineapple plantlets (Murashige and Skoog, 1962). The pH was maintained at 5.6–5.8 using KOH (0.1 N) and HCl (0.1 N) before autoclaving at 121 °C, 15 psi for 15 minutes.

2.4 Experimental setup

Control and box-based *in vitro* experimental setup as follows:

1. Control = Conventional plant tissue culture system
2. PPTCB = Portable plant tissue culture box

The *in vitro* grown pineapple plantlets were planted on MS (Murashige and Skoog, 1962; Dutta et al., 2013) medium containing 30 g L⁻¹ sucrose, 0.75 mg L⁻¹ IAA (indole-3-acetic acid), and 0.8% (w/v) agar. All *in vitro* cultures were maintained at 25 ± 2 °C under a 14 h light/10 h dark photoperiod (long-day condition) with a light intensity of 50 ± 5 μmol m⁻² s⁻¹. A total of 25 plantlets were cultured under each treatment condition, with the conventional plant tissue culture system serving as a control group and the portable PTC box as the treatment group.

2.5 Measurement of dry weight and moisture content

The growth performance of *in vitro* cultured plantlets was evaluated based on fresh weight, dry weight, and moisture content after 30 days of treatment under both the conventional plant tissue culture (PTC) system and the developed portable PTC box. The plants were dried in a hot-air oven at 80 °C. After two hours, plantlets were taken from the oven. The percentage of dry weight and moisture content were measured according to Van De Sande-Bakhuyzen (1928) using the following equations:

$$\text{Dry weight} = \text{Dry weight} / \text{Fresh weight}$$

$$\text{Moisture content} = [(\text{Fresh weight} - \text{Dry weight}) / \text{Fresh weight}] \times 100$$

2.6 Spectroscopic analysis of chlorophylls and carotenoids

Leaf samples were collected from the *in vitro* cultured plantlets and washed thoroughly with distilled water. Leaf sample from each of the tested plants weighed 0.5 gm. They were homogenized using a mortar and pestle with 10 ml of 80% acetone (Maity et al., 2014). The sample was then centrifuged at 5000 rpm for 5 minutes at room temperature. The supernatants were carefully collected to determine Chlorophyll a, Chlorophyll b, and carotenoids contents in a spectrophotometer (SYSTRONICS Double Beam Spectrophotometer). The amounts of the photosynthetic pigments were calculated using the formula according to Maity et al. (2019). The concentrations are expressed as mg/g fresh weight of the leaf. The equations used to calculate the quantity are as follows:

$$\text{Total chlorophyll } a \text{ (per gram tissue)}$$

$$= [12.7 (A663) - 2.69 (A645)] \times [V / (W \times 100)]$$

$$\begin{aligned} &\text{Total chlorophyll } b \text{ (per gram tissue)} \\ &= [22.9 (A645) - 4.68 (A663)] \times [V / (W \times 100)] \end{aligned}$$

$$\begin{aligned} &\text{Total carotenoids (per gram tissue)} \\ &= [1000 \times A470 - 3.27 (\text{chl } a) - 104 (\text{chl } b)] \div [V / (W \\ &\quad \times 100)] \end{aligned}$$

(Where, A = Absorbance at specific wavelength; V = Final volume of chlorophyll extract in 80% acetone; W = Fresh weight of tissue extracted; chl = Chlorophyll).

2.7 Study of genetic fidelity

DNA was extracted from leaves of different light-treated *in vitro* plantlets using the CTAB method (Rogers and Bendich, 1988). The quality and quantity of DNA were inspected by gel electrophoresis and a spectrophotometric assay using a UV-visible spectrophotometer (Genesys 10S UV-vis). PCR (Polymerase Chain Reaction) amplifications were performed according to the method of Bera et al. (2015). A set of 20 oligonucleotide primers (10 bases long), OPA1 to OPA10 and OPB1 to OPB10, were obtained from Operon Technologies Inc. Amplification of DNA was done using a thermal cycler (Applied Biosystem, USA) with an initial denaturation of 3 min at 95°C, followed by 45 cycles of 1 min at 94°C (denaturation), 1 min at 30°C to 34°C (annealing), and 1 min 30 s at 68°C (extension). After completion of cycles, the PCR mix was held for 5 min at 72°C for primer extension. A 25 µl PCR reaction mixture was prepared containing 1X standard Taq reaction buffer, 2 mM MgCl₂, 100 ng DNA, 0.2 µM primer, 100 µM dNTPs, and 1 µl Taq DNA polymerase (3 units). Agarose gel electrophoresis of the DNA was performed (apparatus, TARSON, India) following Ogden and Adams (1987). A 1% agarose gel was prepared using 1X TAE buffer, followed by the addition of 1 µg/ml ethidium bromide, and then poured into the gel casting tray and allowed to solidify. After solidification, the gel was submerged in running buffer (1X TAE buffer) in the horizontal electrophoresis chamber. The PCR-amplified product was loaded into the well with 6X gel loading dye, and a voltage of 100 V and a current of 100 mA were applied to the electrophoresis chamber. After electrophoresis, the gel was observed under UV light as suggested by Ogden and Adams (1987).

2.8 Statistical analysis

All the data were analyzed as mean ± standard deviation, and least significance difference ($p \leq 0.05$) was performed with the help of ANOVA test using Microsoft Excel data analysis tools according to Duncan's multiple range test.

3 Results and discussion

3.1 Electricity consumption and environmental impact of cooling systems

The energy requirements of conventional air-conditioning systems contribute significantly to operational costs and

greenhouse gas emissions (Li and Strezov, 2015; Dong et al., 2021). For instance, a standard 1.5-ton, 5-star-rated air conditioner consumes approximately 1500 W per hour (1.5 kWh). Over a 24-hour period, this corresponds to a total electricity consumption of 36 kWh per day. Assuming electricity generation from bituminous coal, approximately 1.12 pounds (0.5 kg) of coal is required to produce 1 kWh of electrical energy. Consequently, generating 36 kWh of electricity necessitates 40.32 pounds (18.28 kg) of coal daily. Bituminous coal contains roughly 66% carbon by mass, and complete combustion releases carbon dioxide (CO₂) at a rate of 3.67 kg CO₂ per kg of carbon. Therefore, burning 1 kg of coal produces approximately 2.42 kg of CO₂. Using this calculation, 18.28 kg of coal required for a conventional AC system emits nearly 44.24 kg of CO₂ per day (Li and Strezov, 2015; Dong et al., 2021).

In comparison, the developed portable PPTCB operates at only 300 W per hour (0.3 kWh), corresponding to a daily electricity consumption of 7.2 kWh. To generate this energy from coal, only 3.65 kg of bituminous coal is required, producing 8.83 kg of CO₂ per day. This represents a fivefold reduction in CO₂ emissions compared with conventional air-conditioning systems, highlighting the energy efficiency and environmental advantage of the developed PTC box.

This reduction in energy consumption and greenhouse gas emissions demonstrates not only the sustainability of the portable system but also its potential for adoption in resource-limited settings or locations with limited grid electricity. The data highlight the dual benefits of the system: reduced operational costs and significant environmental impact mitigation. By lowering both electricity demand and CO₂ output, the portable system contributes to sustainable laboratory practices while supporting global efforts to reduce fossil fuel dependence.

3.2 Measurement of dry weight and moisture content

The growth performance of *in vitro* cultured plantlets (Figure 2) was evaluated based on fresh weight, dry weight, and moisture content after 30 days of treatment under both the conventional plant tissue culture (PTC) system and the developed portable PTC box (Table 2). At the initiation of the experiment (Day 0), the plantlets exhibited a mean fresh weight of 102 ± 12 mg and a dry weight of 127 ± 4 mg g⁻¹ fresh tissue, with a moisture content of 87.25%, indicating that the tissues were in their initial developmental phase with high water content and low biomass accumulation. After 30 days of culture, a significant increase ($p \leq 0.05$) in both fresh and dry weights was observed in plantlets grown under both systems, demonstrating active growth and biomass accumulation during the *in vitro* period. Plantlets cultured under the conventional PTC system recorded a fresh weight of 347 ± 17 mg and dry weight of 201 ± 8 mg g⁻¹, while those grown in the developed portable PTC box showed comparable values of 341 ± 15 mg and 196 ± 7 mg g⁻¹, respectively. The negligible difference between the two systems indicates that the portable PTC box effectively supports similar physiological growth responses to those achieved under conventional culture conditions (Samanta et al., 2023).

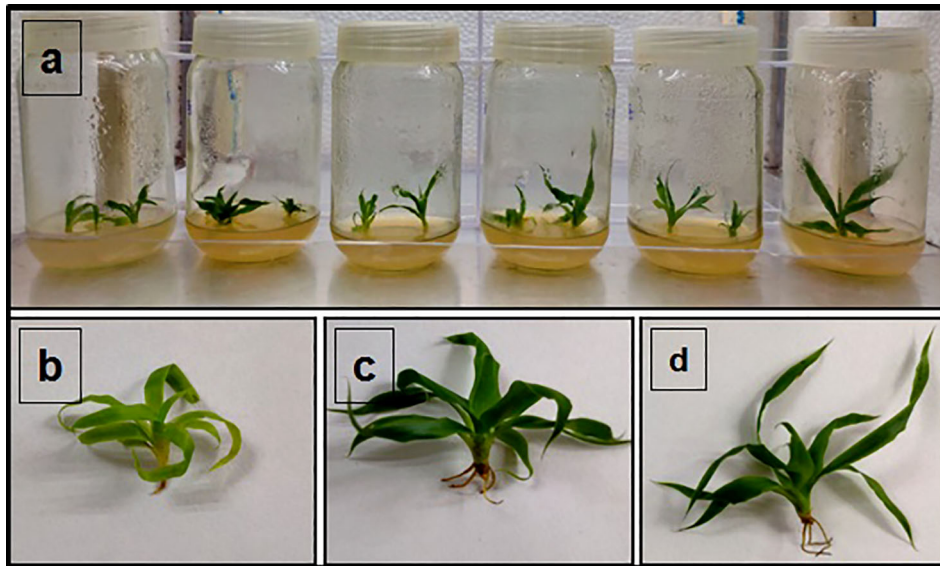


FIGURE 2 Vegetative growth of *in vitro* treated pineapple plantlets under two different systems. (a) Plantlets under PTC box (b) Initial days of treatment (c) After 30 days of treatment under conventional PTC system (d) After 30 days of treatment under portable PTC box.

Moisture content decreased slightly in both treatments (79.83% and 80.35%) after 30 days, suggesting enhanced tissue maturation and biochemical activity associated with chlorophyll synthesis and cell wall development. The similarity in moisture percentage further confirms the ability of the portable system to maintain optimal humidity and temperature conditions conducive to normal plant growth.

3.3 Spectroscopic analysis of chlorophylls and carotenoids

The photosynthetic pigment content, including chlorophyll *a*, chlorophyll *b*, total chlorophyll, and carotenoids, was analyzed in plantlets grown under conventional plant tissue culture (PTC) conditions and those maintained in the developed portable PTC box for 30 days (Figure 3).

At the initial stage (Day 0), pigment concentrations were comparatively low, reflecting limited chlorophyll biosynthesis in

freshly cultured plantlets. After 30 days of growth, a significant increase ($p \leq 0.05$) in chlorophyll *a*, chlorophyll *b*, total chlorophyll, and carotenoids was observed in both the conventional system and the portable PTC box, indicating improved photosynthetic capacity and physiological development under both controlled environments.

Notably, the chlorophyll *a* and total chlorophyll contents of plantlets grown in the portable PTC box were statistically comparable ($p \leq 0.05$) to those grown in the conventional system. Similarly, chlorophyll *b* and carotenoid levels showed no significant difference between the two systems, suggesting that the portable PTC box effectively supports pigment synthesis and overall photosynthetic function. The maintenance of comparable pigment profiles demonstrates that the portable system provides adequate light intensity, temperature regulation, and humidity control necessary for normal chloroplast development (Samanta et al., 2023).

These results indicate that the PPTCB can successfully replicate the physiological conditions of a conventional culture room, ensuring healthy plantlet growth while reducing energy consumption and environmental impact. The comparable pigment accumulation also confirms that the portable system can serve as an efficient, low-cost, and eco-friendly alternative for *in vitro* culture without compromising plant quality.

TABLE 2 The growth performance of *in vitro* cultured pineapple plantlets after 30 days of treatment under both the conventional plant tissue culture system and the developed portable PTC box.

Treatments	Fresh weight (mg)	Dry weight (mg/g fresh tissue)	Moisture content (%)
Day 0	102 ± 12	127 ± 4	87.25
After 30 days of treatment under Conventional PTC System	347 ± 17	201 ± 8	79.83
After 30 days of treatment under Developed PTC Box	341 ± 15	196 ± 7	80.35

The data are represented as mean ± standard deviation.

3.4 Study of genetic fidelity

The genetic fidelity of *in vitro* cultured plants is a critical factor determining the success and reliability of large-scale micropropagation programs. In the present study, Random Amplified Polymorphic DNA (RAPD) analysis was employed to assess the genetic stability of tissue-cultured plantlets. RAPD markers were selected as they provide a rapid, cost-effective, and highly sensitive method for detecting somaclonal variations that may arise during *in vitro* propagation.

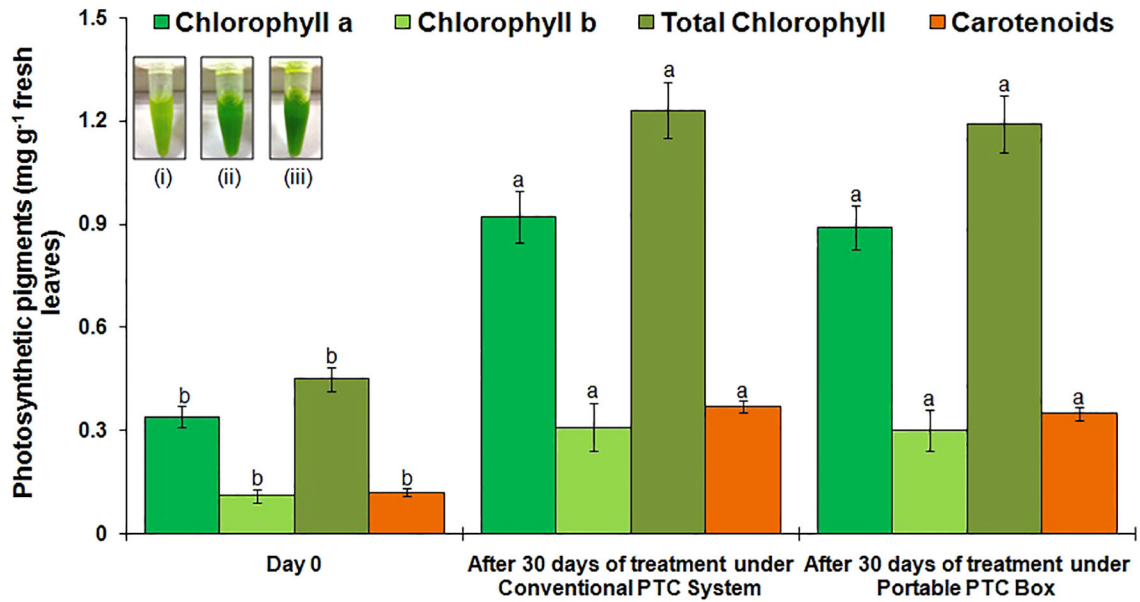


FIGURE 3 Quantification of photosynthetic pigment content in *in vitro* grown pineapple leaves treated under two different systems. The data are represented as mean ± standard deviation, and the data indicated with different lower-case letters are significantly different ($p \leq 0.05$) according to Duncan's multiple range test ($n = 20$). [Inset represents the pigments intensity (i) early day, (ii – iii) after 30 days of treatment under conventional PTC system & portable PTC box respectively].

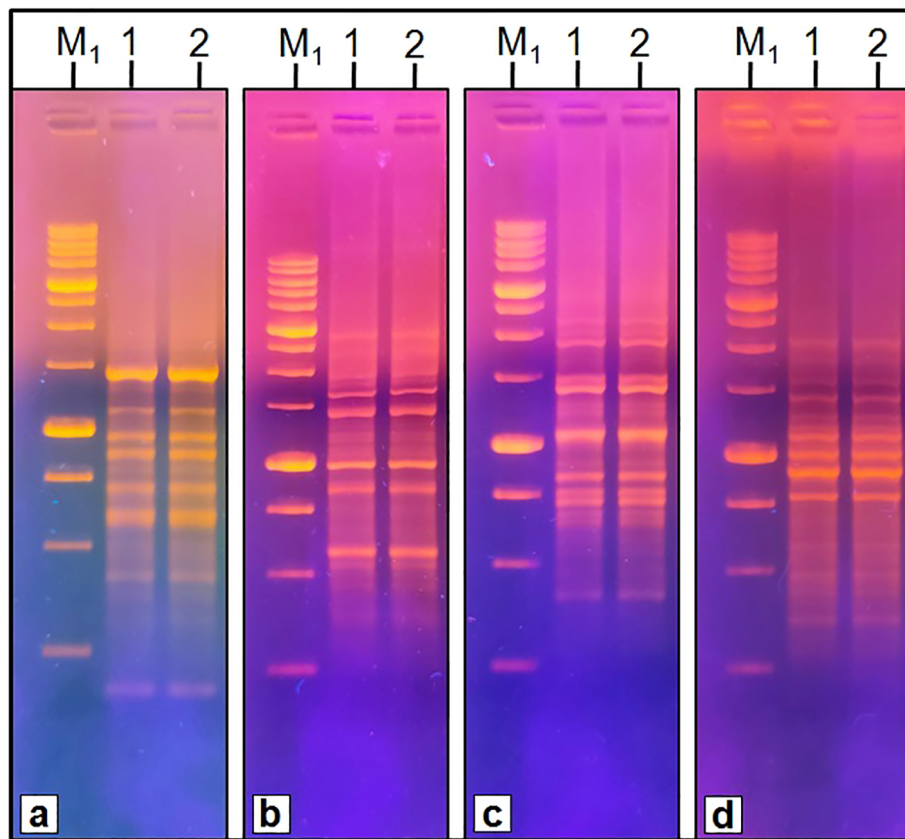


FIGURE 4 Randomly Amplified Polymorphic DNA (RAPD) profiles of *in vitro* treated pineapple (*Ananas comosus*) leaf samples. Electrophoretic patterns of DNA amplification products generated using primers (a) OPA5, (b) OPA10, (c) OPB5, and (d) OPB7. Lane M1: 1 kb DNA ladder (size range: 250 bp – 10,000 bp). Lanes 1 and 2 represent samples from the conventional PTC system and the developed portable PTC box, respectively.

A set of random primers was used for amplification, and all the primers produced clear bands across both conventional plant tissue culture system plantlets and PPTCB plantlets (Figure 4). The RAPD profiles revealed identical banding patterns for all samples tested, showing the absence of polymorphic bands (Bera et al., 2015). The uniformity in amplification products indicates that no detectable genetic variation occurred among the regenerated plantlets. These results confirm the genetic homogeneity and stability of the *in vitro* propagated plants, suggesting that the culture conditions and regeneration protocols employed did not induce somaclonal variation.

Moreover, if the prototype system is powered entirely by solar energy, CO₂ emissions can be eliminated completely, offering a zero-emission alternative for maintaining controlled environmental conditions in plant tissue culture laboratories. Furthermore, the portable plant tissue culture box presents a viable alternative to cryopreservation for the short-to-medium term maintenance of live germplasm. Due to its modular and lightweight design, the unit facilitates the secure transport of endangered or medicinally significant species while maintaining physiological integrity (Li et al., 2022). Additionally, the unit's specialized environmental control system offers significant potential for aerospace research, enabling the study of plant development under varying gravitational or atmospheric conditions (Zabel et al., 2016; Jia et al., 2024).

4 Conclusion

The plantlets grown in the portable plant tissue culture box are morphologically similar to conventional *in vitro* grown plants. The Chlorophyll pigment content, dry weight, and moisture content are similar to *in vitro* grown plants. Therefore, the PTC box can serve as an alternative to a conventional *in vitro* culture laboratory. The portable tissue culture box is designed to accommodate various *in vitro* stages, including the maintenance of callus cultures, the induction of multiple shoots, and the storage of regenerated plantlets under controlled environmental conditions. Overall, these results demonstrate that the developed portable plant tissue culture box can replicate the growth efficiency of a standard culture room, providing a cost-effective and energy-efficient alternative for *in vitro* plant propagation without compromising plantlet quality or physiological performance.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

TM: Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. AS: Conceptualization, Data

curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Validation, Visualization, Writing – original draft, Writing – review & editing. SD: Conceptualization, Formal analysis, Methodology, Writing – original draft, Writing – review & editing. AB: Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing. SS: Investigation, Resources, Supervision, Validation, Writing – original draft, Writing – review & editing. KP: Investigation, Methodology, Validation, Writing – original draft, Writing – review & editing.

Funding

The author(s) declared that financial support was not received for this work and/or its publication.

Acknowledgments

The authors acknowledge the financial support received from the Carbon Zero Challenge programme, IIT Madras, to construct the system. The authors are thankful to the Department of Biotechnology and IDEA lab, Haldia Institute of Technology, for providing the necessary facilities to carry out the experiments of this work.

Conflict of interest

The author(s) declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Generative AI statement

The author(s) declared that generative AI was not used in the creation of this manuscript.

Any alternative text (alt text) provided alongside figures in this article has been generated by Frontiers with the support of artificial intelligence and reasonable efforts have been made to ensure accuracy, including review by the authors wherever possible. If you identify any issues, please contact us.

Publisher's note

All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

References

- Agarwal, A., and Dutta Gupta, D. (2016). Impact of light-emitting diodes (LEDs) and its potential on plant growth and development in controlled-environment plant production system. *Curr. Biotechnol.* 5, 28–43. doi: 10.2174/2211550104666151006001126
- Altaf, M. T., Liaqat, W., Ali, A., Jamil, A., Bedir, M., Nadeem, M. A., et al. (2024). “Conventional and biotechnological approaches for the improvement of industrial crops,” in *Industrial crop plants* (Springer Nature Singapore, Singapore), 1–48. doi: 10.1007/978-981-97-1003-4_1
- Altman, A. (2003). From plant tissue culture to biotechnology: scientific revolutions, abiotic stress tolerance, and forestry. *In Vitro Cell. Dev. Biology-Plant* 39, 75–84. doi: 10.1079/ivp2002379
- Bera, A. K., Maity, T. R., Samanta, A., Dolai, A., Saha, B., and Datta, S. (2015). Enhancement of *in vitro* corm production in Gladiolus by periodically replacement of liquid media using coir matrix. *J. Appl. Horticulture* 17, 222–224. doi: 10.37855/jah.2015.v17i03.42
- Bhatia, S., Sharma, K., Dahiya, R., and Bera, T. (2015). *Modern applications of plant biotechnology in pharmaceutical sciences* (Academic press). doi: 10.1016/b978-0-12-802221-4.00012-1
- Dong, Y., Coleman, M., and Miller, S. A. (2021). Greenhouse gas emissions from air conditioning and refrigeration service expansion in developing countries. *Annu. Rev. Environ. Resour.* 46, 59–83. doi: 10.1146/annurev-environ-012220-034103
- Dutta, I., Bhadra, J., Ghosh, P., Saha, B., and Datta, S. (2013). An efficient and cost effective protocol for *in vitro* propagation of Pineapple. *J. Ornamental Plants* 3, 229–234.
- Dutta Gupta, S., and Jatothu, B. (2013). Fundamentals and applications of light-emitting diodes (LEDs) in *in vitro* plant growth and morphogenesis. *Plant Biotechnol. Rep.* 7, 211–220. doi: 10.1007/s11816-013-0277-0
- Gulzar, B., Mujib, A., Malik, M. Q., Mamgain, J., Syeed, R., and Zafar, N. (2020). “Plant tissue culture: agriculture and industrial applications,” in *Transgenic technology based value addition in plant biotechnology* (Academic Press), 25–49. doi: 10.1016/b978-0-12-818632-9.00002-2
- Gunaseelan, R. J., Raj, A., Nagarajan, P., Perumal, S., Kumar, J., and Patil, S. J. (2025). “Biotechnological Phytochemical Synthesis: Innovations, Challenges, Advantages, Implications,” in *Biotechnology and Phytochemical Prospects in Drug Discovery* (Springer Nature Singapore, Singapore), 83–101. doi: 10.1007/978-981-96-2790-5_3
- Jia, C., Zheng, W., Liu, F., Ding, K., Yuan, Y., Wang, J., et al. (2024). Biological culture module for plant research from seed-to-seed on the Chinese Space Station. *Life Sci. Space Res.* 42, 47–52. doi: 10.1016/j.lssr.2024.04.005
- Li, J., He, M., Xu, X., Huang, T., Tian, H., and Zhang, W. (2022). *In vitro* techniques for shipping of micropropagated plant materials. *Horticultrae* 8, 609. doi: 10.3390/horticultrae8070609
- Li, X., and Strezov, V. (2015). Energy and greenhouse gas emission assessment of conventional and solar assisted air conditioning systems. *Sustainability* 7, 14710–14728. doi: 10.3390/su71114710
- Maity, T. R., Samanta, A., Jana, D., Saha, B., and Datta, S. (2014). Effect of Piper beetle leaf extract on post-harvest physiology and vascular blockage in relation to vase life and keeping quality of cut spike of tuberose (*Polianthes tuberosa* L. cv. Single). *Indian J. Plant Physiol.* 19, 250–256. doi: 10.1007/s40502-014-0110-y
- Maity, T. R., Samanta, A., Jana, D., Saha, B., and Datta, S. (2016). *In vitro* flowering of tobacco induced by light emitting diode. *Indian J. Biotechnol.* 15, 440–442.
- Maity, T. R., Samanta, A., Saha, B., and Datta, S. (2019). Evaluation of Piper beetle mediated silver nanoparticle in post-harvest physiology in relation to vase life of cut spike of Gladiolus. *Bull. Natl. Res. Centre* 43, 9. doi: 10.1186/s42269-019-0051-8
- Mondal, S., Rutkoski, J. E., Velu, G., Singh, P. K., Crespo-Herrera, L. A., Guzman, C., et al. (2016). Harnessing diversity in wheat to enhance grain yield, climate resilience, disease and insect pest resistance and nutrition through conventional and modern breeding approaches. *Front. Plant Sci.* 7. doi: 10.3389/fpls.2016.00991
- Murashige, T., and Skoog, F. (1962). A revised medium for rapid growth and bio assays with tobacco tissue cultures. *Physiologia plantarum* 15. doi: 10.1111/j.1399-3054.1962.tb08052.x
- Ogden, R. C., and Adams, D. A. (1987). “Electrophoresis in agarose and acrylamide gels,” in *Methods in enzymology*, vol. 152. (Academic Press), 61–87. doi: 10.1016/0076-6879(87)52011-0
- Reddy, J. (2024). *Plant tissue culture* (CRC Press). doi: 10.1201/9781032712611
- Reed, B. M., Sarasan, V., Kane, M., Bunn, E., and Pence, V. C. (2011). Biodiversity conservation and conservation biotechnology tools. *In Vitro Cell. Dev. Biology-Plant* 47, 1–4. doi: 10.1007/s11627-010-9337-0
- Rogers, S. O., and Bendich, A. J. (1988). “Extraction of DNA from plant tissues,” in *Plant molecular biology manual* (Springer Netherlands, Dordrecht), 73–83. doi: 10.1007/978-94-009-0951-9_6
- Samanta, S., PanChadhyae, P., Maity, T. R., Nanda, K., and Samanta, A. (2023). Development of a growth chamber for cryptogams: a step toward *ex situ* conservation. *Braz. J. Bot.* 46, 661–666. doi: 10.1007/s40415-023-00913-9
- Shukla, M. R., Singh, A. S., Piunno, K., Saxena, P. K., and Jones, A. M. P. (2017). Application of 3D printing to prototype and develop novel plant tissue culture systems. *Plant Methods* 13, 6. doi: 10.1186/s13007-017-0156-8
- Trigiano, R. N., and Gray, D. J. (2016). *Plant tissue culture, development, and biotechnology* (CRC Press). doi: 10.1201/9781439896143
- Twaij, B. M., Jazar, Z. H., and Hasan, M. N. (2020). Trends in the use of tissue culture, applications and future aspects. *Int. J. Plant Biol.* 11. doi: 10.4081/pb.2020.8385
- Van De Sande-Bakhuyzen, H. L. (1928). Studies upon wheat grown under constant conditions—II. *Plant Physiol.* 3, 7. doi: 10.1104/pp.3.1.7
- Zabel, P., Bamsey, M., Schubert, D., and Tajmar, M. (2016). Review and analysis of over 40 years of space plant growth systems. *Life Sci. space Res.* 10, 1–16. doi: 10.1016/j.lssr.2016.06.004
- Zakai, F. M., Faizan, M., and Khan, M. F. (2021). “PCB Design and Fabrication,” in *Functional Reverse Engineering of Strategic and Non-Strategic Machine Tools* (CRC Press), 79–95. doi: 10.1201/9780367808235-8