International Journal of Engineering and Science Invention (IJESI)
ISSN (Online): 2319-6734, ISSN (Print): 2319-6726
www.ijesi.org ||Volume 14 Issue 12 December 2025 || PP 65-72

Synergistic Effects of Indole-3-Acetic Acid and Gibberellic Acid on *In Vitro* Growth of Potato Plantlets for Seed Production

Dhanorkar Shravani Shankarrao[1], Dr. Naveen Joshi[2]
[1, 2]Department of Biotechnology, Mansarovar Global University, Sehore, MP

Abstract
The present study evaluates the physiological response and multiplication efficiency of Solanum tuberosum L. microshoots under varying concentrations of plant growth regulators during in vitro culture. The effects of Indole-3-acetic acid (IAA) and Gibberellic acid (GA3), applied individually and in combination, were assessed with respect to shoot elongation, nodal proliferation, and biomass accumulation. Particular emphasis was placed on understanding the synergistic interaction between these phytohormones in enhancing micropropagation efficiency.
Results demonstrated that specific concentrations of IAA and GA3 significantly influenced growth parameters, with combined treatments exhibiting a pronounced synergistic effect compared to individual applications. Enhanced internode elongation, increased nodal development, and improved root initiation were observed under optimized hormonal conditions. Additionally, the study highlights the role of these growth regulators in accelerating dormancy break and promoting sprout development, which are critical for efficient seed tuber production.
Overall, the findings indicate that the synergistic application of GA3 and IAA provides an effective strategy for improving in vitro clonal propagation of potato. This approach can be employed to facilitate large-scale production of uniform, healthy plantlets, thereby supporting sustainable seed production systems and agricultural productivity.
Keywords: *Solanum tuberosum L.; in vitro propagation; Indole-3-acetic acid (IAA); Gibberellic acid (GA3); plant growth regulators; microshoots; hormonal synergy; nodal proliferation; shoot elongation; seed tuber production.*

I. Introduction

The use of *in vitro* propagation techniques, particularly those utilizing growth regulators like gibberellins, has become a standard approach for the rapid multiplication of high-quality, virus-free potato seed stock (Mohapatra & Batra, 2017). These growth regulators, such as GA3 and IBA, play a critical role in modulating physiological processes including node formation, shoot elongation, and root development (Kumari, 2023). Furthermore, the precise optimization of these hormonal concentrations in Murashige and Skoog media remains essential for enhancing the survival rates and overall biomass accumulation of diverse potato cultivars (Makau et al., 2022). Additionally, the synergistic application of auxins like indole-3-acetic acid in combination with benzylaminopurine has proven effective in maximizing multiple shoot production during the micropropagation phase (José & Sivaprasad, 2025). Moreover, studies investigating the interaction between gibberellic acid and synthetic auxins such as indole-3-butyric acid have demonstrated significant improvements in the uniformity of shoot proliferation and subsequent rhizogenesis in specific potato cultivars (Kumari, 2023). Beyond hormonal optimization, recent inquiries have explored the integration of bio-organic amendments, such as mycorrhiza and vermicompost, as sustainable strategies to further enhance nutrient uptake and tuberization efficiency (Boubaker et al., 2023). In hydroponics use of plant tissue culture plantlets offers a viable pathway to circumvent land limitations and high costs associated with traditional soil-based seed propagation (Nasution et al., 2025). These advancements in micropropagation methodologies, specifically the standardization of sucrose and gibberellic acid concentrations, are particularly vital for scaling the production of resilient local potato varieties in emerging agricultural sectors (Makau et al., 2022). In the present study, we investigate the distinct physiological responses of widely cultivated potato variety to varying concentrations of gibberellic acid and indole-3-acetic acid to determine the optimal hormonal balance for accelerating shoot development and enhancing root biomass under controlled in vitro conditions (Stupko & Lugovtsova, 2023), (Abdella et al., 2023).

II. Methodology:-

Plant collection:-

In this study, potato plants (*Solanum tuberosum* L.) were propagated via plant tissue culture to obtain disease-free plantlets for use in an aeroponic system for minituber production. To initiate the process, potato tubers were collected and used for tissue culture.

Preparation of tuber for inoculation :-

Healthy tubers were first washed three times with sterile distilled water and then surface-sterilized with 70% ethyl alcohol for 4 minutes, followed by three additional washes with distilled water. The tubers were subsequently treated with 0.1% $HgCl_2$ for 2 minutes with vigorous shaking and then washed again with sterile distilled water to yield surface-sterilized tubers, which were transferred to the plant tissue culture medium.

Preparation of tuber germination media:-

Since plant tissue culture provides the healthiest and disease-free plantlets for potato planting, tubers were used as a germination source for shoot and root induction in plant tissue culture medium with the nutrient composition given below.

The medium contained plant growth regulators, namely indole acetic acid and gibberellic acid, which were tested for successful plantlet germination using tubers as explants.

During media preparation, 1 N NaOH was used to adjust the pH to 5.8. Additionally, 0.2% (w/v) phytagel was used as a solidifying agent. The solution was then heated in a microwave oven before being dispensed into tubes and autoclaved for 15 minutes at 121°C and 15 psi pressure. The tubes were allowed to cool and solidify before inoculating the tubers.

Plant tissue culture conditions:-

Once the tubers were successfully transferred to the culture medium, the light intensity in the lab was maintained at 3000 lux and the temperature was set at 26 ± 1°C. During maintenance, the photoperiod was set to 16 hours of light and 8 hours of dark.

Transplanting of plantlets to polyhouse:-

Upon successful plantlet germination, the plantlets were carefully separated from the half MS medium without damaging their roots. These plantlets were then transferred to pots filled with moist cocopeat and covered with transparent polythene bags to maintain necessary moisture and prevent external infection. The pots were subsequently transferred to the polyhouse for hardening. After 10 days of hardening, the plantlets were washed with water to remove cocopeat traces and transferred to the net pods of the aeroponics system for survival screening.

III. Result And Discussion:-

Collection of Potato tubers and surface sterilization:-

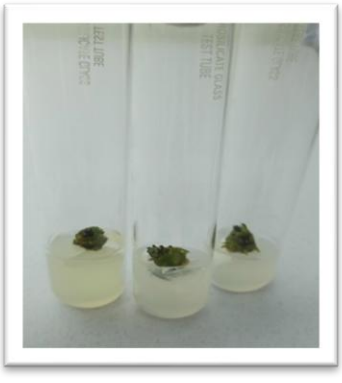
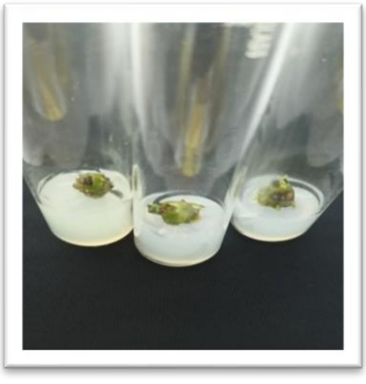
In this study, disease-free potato plantlets were germinated using half MS medium supplemented with plant growth regulators, namely IAA and GA3. Initially, healthy potato tubers were collected. These were successfully surface-sterilized first with 70% ethyl alcohol for 4 minutes, followed by 0.1% $HgCl_2$ for 2 minutes (Fig. 1). This sterilization procedure effectively eliminated microbial contaminants, ensuring that the subsequent inoculation of nodal explants into the Murashige and Skoog medium remained axenic (Omar, 2017). The use of given sterliants facilitated a sterile initiation process, similar to protocols utilizing 0.1% $HgCl_2$ for surface sterilization of primary explants (Borna et al., 2019).

		
Potato having tubers	**Tuber collection**	**After Surface sterilization**
Fig. 1 : Process of potato tuber collection, sorting and surface sterilisation		

Shoot and Root induction of half MS media:-

Upon transferring potato tubers to half MS medium supplemented with sucrose, IAA, and GA3, positive effects on shoot and root induction were observed using potato tuber explants. A photoperiod of 16 hours light and 8 hours dark at 26 ± 1°C proved promising with this supplemented half MS medium.

Maintenance studies recorded initial shoot induction at 7-10 days and root induction by 10-14 days under tissue culture conditions. Well-developed plantlets were ready by the 20^{th}-25^{th} day and transplanted into cocopeat by the 28^{th}-30^{th} day (Fig.2). Following this acclimatization period, plantlets with well-proliferated roots were suitable for transfer to the aeroponic unit, where environmental parameters such as temperature and nutrient delivery could be precisely controlled (Borna et al., 2019; Miriam et al., 2018). The use of IAA, and GA3 significantly enhanced the vegetative growth phase and subsequent stolon development, which is critical for maximizing minituber yield within the closed aeroponic environment (Bharath & B, 2024). Here sucrose served as the primary carbon source at a concentration of 30 g/l, providing the necessary energy for cellular metabolism and robust plantlet development (Buckseth et al., 2020). Furthermore, the integration of such controlled micropropagation techniques is essential for the subsequent mass production of high-quality propagules in specialized aeroponic systems (Tang et al., 2024).

		
Shoot induction on tubers	**Shoot elongation**	**Development of root and shoot**

Well-developed plantlets

Fig 2: Plant tissue culture used to generate seedling from the potato showing the stages of shoot induction, elongation and development of shoot and root

Hardening of potato plantlets:-

The plantlets were successfully transferred to cocopeat and, with proper bagging, hardened without any traces of acclimatization issues. By the 12th day of acclimatization, they were ready for transplanting to the aeroponics system for disease-free potato seed production (Fig. 3 and Fig. 4). This approach ensures a high survival rate of the plantlets upon transition to the controlled environment, as consistent moisture and nutrient management are vital for successful establishment (Kulus & Tymoszuk, 2024), (Kafle et al., 2023). Furthermore, the implementation of an aeroponic growth chamber facilitates optimal root development and tuber initiation, significantly increasing the yield of virus-free seed potatoes (Buckseth et al., 2020). Hence use of hardened plants serves as a robust foundation for achieving higher uniformity in minituber production while mitigating the risk of pathogen transmission during the transition to soil-free cultivation (Toma, 2022).

Hardening of *in vitro* grown plantlets

After hardening 3 days

After hardening 6 days

Fig. 3: Hardening of the plant upon 6th day

After 12 days planlet transferred into the system

Washing

Fig. 4 : Plant lets transferred into the aeroponics setup upon 1 days by washing and made ready for furhter process

Growth performance of tissue culture plantlets for potato seed production:-

The tissue-cultured plantlets, previously hardened in the polyhouse, were successfully transferred to the aeroponics system, where they grew well and produced potato seeds by 75 to 90 days, as shown in the setup and techniques used (Fig. 5). Quantitative analysis revealed that these aeroponically grown plantlets exhibited superior tuberization metrics compared to traditional soil-based cultivation, largely due to the precise delivery of nutrient solutions (Abuhena et al., 2022). Use of plant tissue culture plants facilitates the rapid generation of healthy, disease-free materials that serve as an ideal foundation for high-density aeroponic production (Sadawarti et al., 2020). Earlier also studies have demonstrated that utilizing microplants for seed production systems under insect-proof net house conditions provides a secure pathway for mass multiplication prior to aeroponic transfer (Sadawarti et al., 2020). By optimizing root zone aeration and maintaining independence from seasonal constraints, this aeroponic integration maximizes the yield of high-quality minitubers while significantly reducing pathogen exposure compared to conventional soil-based methods (Buckseth et al., 2022; Sugiyono et al., 2021). Moreover, the deployment of such soilless cultivation technologies effectively addresses critical challenges, including water scarcity and environmental degradation, by enabling efficient potato production in non-arable areas. Hence use of plant tissue culture along with Hydroponics for potato seed production represents a paradigm shift toward sustainable agriculture, offering a viable solution for the massive production of potato mini-tubers with high efficiency (Rajendran et al., 2024).

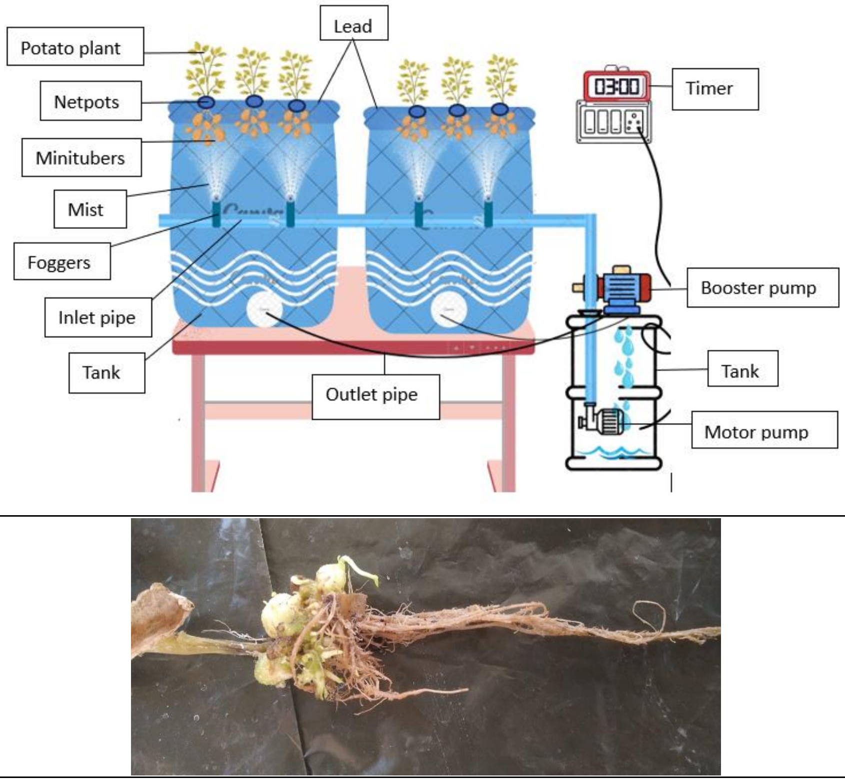


Fig. 5: The setup of the aeroponics used for tissue cultured potato plantlets showing success of disease-free potato seed production

IV. Conclusion:-

Study highlights the importance of disease free plantlets production of potato once used for aeroponics systems. Here study showcased that potato tubers can be used as a source of explant and upon successful surface sterilization using ethyl alcohol (70%) and HgCl2 (0.1%) the tubers can induce shoot and root and resultant complete plantlet once maintained with half MS (4.4g/L), sucrose (30g/L), IAA(0.1g/L) and GA3 (0.25 g/L) by the 25 to 30 days of maintenance study also showcased that plantlets successfully abled to grow in aeroponics set and produced potato seed production successfully .Hence we proposed to use the plant tissue cultured plantlets by the given MS media for future aeroponics system which provides better controlled , low cost and disease free plantlets for potato aeroponics farming.

References

[1]. Abdella, B., Yusuf, Z., & Petros, Y. (2023). Optimization of Hormonal Compositions of Media for In vitro Propagation of Apple (Malus ×domestica Borkh.) Cultivars. *The Open Biotechnology Journal*, *17*(1). https://doi.org/10.2174/18740707-v17-e230202-2022-15

[2]. Abuhena, M., Al-Rashid, J., Azim, Md. F., Khan, Md. N. M., Kabir, Md. G., Barman, N. C., Rasul, N. M., Akter, S., & Huq, Md. A. (2022). Optimization of industrial (3000 L) production of Bacillus subtilis CW-S and its novel application for minituber and industrial-grade potato cultivation. *Scientific Reports*, *12*(1). https://doi.org/10.1038/s41598-022-15366-5

[3]. Bharath, S. R., & B, M. R. (2024). Effect of Varied Levels of Gibberellin, Anti-Gibberellin and Cytokinin on Growth and Tuberization in Potato. *Madras Agricultural Journal*, *111*. https://doi.org/10.29321/maj.10.000ma9

[4]. Borna, R. S., Hoque, M., & Sarker, R. (2019). In vitro Microtuber Induction and Regeneration of Plantlets from Microtuber Discs of Cultivated Potato (Solanum tuberosum L.). *Plant Tissue Culture and Biotechnology*, *29*(1), 63. https://doi.org/10.3329/ptcb.v29i1.41979

[5]. Boubaker, H., Saadaoui, W., Daşgan, H. Y., Tarchoun, N., & Gruda, N. S. (2023). Enhancing Seed Potato Production from In Vitro Plantlets and Microtubers through Biofertilizer Application: Investigating Effects on Plant Growth, Tuber Yield, Size, and Quality. *Agronomy*, *13*(10), 2541. https://doi.org/10.3390/agronomy13102541

[6]. Buckseth, T., Singh, R. K., Tiwari, J. K., Sharma, A. K., Singh, S., & Chakrabarti, S. K. (2020). A novel sustainable aeroponic system for healthy seed potato production in India – An update. *The Indian Journal of Agricultural Sciences*, *90*(2), 243. https://doi.org/10.56093/ijas.v90i2.98995

[7]. Buckseth, T., Tiwari, J. K., Singh, R. K., Kumar, V., Sharma, A. K., Dalamu, D., Bhardwaj, V., Sood, S., Kumar, M., Sadawarti, M. J., Challam, C., Naik, S., & Pandey, N. K. (2022). Advances in innovative seed potato production systems in India. *Frontiers in Agronomy*, *4*. https://doi.org/10.3389/fagro.2022.956667

[8]. José, S. C. B. R., & Sivaprasad, A. (2025). Influence of gibberellic acid (GA3) on in vitro seed germination of Dendrocalamus strictus (Roxb.) Nees: Exploring its effect under varied storage periods and micropropagation potential. *Advances in Bamboo Science*, *11*, 100141. https://doi.org/10.1016/j.bamboo.2025.100141

[9]. Kafle, D. P., Parajuli, S., Upreti, A., & Gauchan, D. P. (2023). Effect of Coconut Water and GA3 Concentrations on in vitro Clonal Propagation of Potato Cultivars from Nepal. *Nepal Journal of Biotechnology*, *11*(1). https://doi.org/10.54796/njb.v11i1.274

[10]. Kulus, D., & Tymoszuk, A. (2024). Advancements in In Vitro Technology: A Comprehensive Exploration of Micropropagated Plants. *Horticulturae*, *10*(1), 88. https://doi.org/10.3390/horticulturae10010088

[11]. Kumari, M. (2023). Gibberellic acid (GA3) alone and in combination with indole 3 butyric acid (iba) modulation during in vitro propagation of potato (*Solanum tuberosum* L.) microplants. *Plant archives*, *23*, 122. https://doi.org/10.51470/plantarchives.2023.v23.no1.021

[12]. Makau, F. M., Mwangi, M., Oyoo, M. E., Kibe, A. M., & Oggema, J. (2022). Effects of Sucrose and Gibberellic Acid on Growth and Survival of Local Potato (Solanum tuberosum L.) Varieties in vitro in Kenya. *European Journal of Biology and Biotechnology*, *3*(4), 13. https://doi.org/10.24018/ejbio.2022.3.4.372

[13]. Miriam, W. M., Charles, L. M., Susan, A. O., Moses, W. N., Margaret, N. M., & Judith, N. O. (2018). Performance of five potato varieties with regards to growth and production of mini-tubers under an aeroponic system in central highlands of Kenya. *African Journal of Agricultural Research*, *13*(8), 366. https://doi.org/10.5897/ajar2017.12762

[14]. Mohapatra, P. P., & Batra, V. K. (2017). Tissue Culture of Potato (Solanum tuberosum L.): A Review [Review of *Tissue Culture of Potato (Solanum tuberosum L.): A Review*]. *International Journal of Current Microbiology and Applied Sciences*, *6*(4), 489. Excellent Publishers. https://doi.org/10.20546/ijcmas.2017.604.058

[15]. Nasution, M. W. A., Siregar, L. A. M., & Basyuni, M. (2025). Optimization medium at the propagation and formation stage of micro tubers some varieties of potato. *Indonesian Journal of Agricultural Research*, *8*(1), 9. https://doi.org/10.32734/injar.v8i1.16798

[16]. Omar, G. (2017). Growth Responses of Potato Plantlets Cultured In Vitro under Different Colors Light-Emitting Diodes (LEDs). *Hortscience Journal of Suez Canal University*, *6*(1), 65. https://doi.org/10.21608/hjsc.2017.6397

[17]. Rajendran, S., Domalachenpa, T., Arora, H., Li, P., Sharma, A., & Rajauria, G. (2024). Hydroponics: Exploring innovative sustainable technologies and applications across crop production, with Emphasis on potato mini-tuber cultivation. *Heliyon*, *10*(5). https://doi.org/10.1016/j.heliyon.2024.e26823

[18]. Sadawarti, M. J., Samadhiya, R. K., Singh, R. K., Singh, S., Buckseth, T., Rawal, S., Singh, V., Katare, S., ROY, S., & Chakrabarti, S. K. (2020). Standardization of agro-techniques for aeroponic potato (Solanum tuberosum) minitubers under generation-0. *The Indian Journal of Agricultural Sciences*, *90*(3), 616. https://doi.org/10.56093/ijas.v90i3.101499

[19]. Sadawarti, M. J., Singh, S. P., Singh, R. K., Buckseth, T., Katare, S., Chakrabarti, S. K., & Khambalkar, P. A. (2020). In vivo performance of hi-tech planting materials for seed potato production: A review [Review of *In vivo performance of hi-tech planting materials for seed potato production: A review*]. *International Journal of Chemical Studies*, *8*(5), 1151. AkiNik Publications. https://doi.org/10.22271/chemi.2020.v8.i5p.10450

[20]. Stupko, V. Yu., & Lugovtsova, S. Yu. (2023). The use of phytohormones to increase the efficiency of potato propagation in nodal cuts culture. *BIO Web of Conferences*, *66*, 1006. https://doi.org/10.1051/bioconf/20236601006

[21]. Sugiyono, S., Prayoga, L., Proklamasiningsih, E., Faozi, K., & Prasetyo, R. (2021). The Improvement of Mini Tuber Production of Granola Potato Cultivar in Aeroponics System. *Biosaintifika Journal of Biology & Biology Education*, *13*(1), 77. https://doi.org/10.15294/biosaintifika.v13i1.27714

[22]. Tang, L., Syed, A.-A., Otho, A. R., Junejo, A. R., Tunio, M. H., Li, H., Ali, M. N. H. A., Brohi, S. A., Otho, S. A., & Channa, J. A. (2024). Intelligent Rapid Asexual Propagation Technology—A Novel Aeroponics Propagation Approach. *Agronomy*, *14*(10), 2289. https://doi.org/10.3390/agronomy14102289

[23]. Toma, R. S. (2022). Minitubers production of four potato (solanum tuberosum l.) cultivars by tissue culture technique. *Iraqi journal of agricultural sciences*, *53*(5), 1058. https://doi.org/10.36103/ijas.v53i5.1619