biology and life sciences forum

MDPI

*Proceeding Paper*

# The Phenotypic Reactivity of *Passiflora incarnata* L. on Various Content of Mineral Salts and Regulators during Micropropagation and Acclimatization †

**Marcin Ożarowski [1,*], Elżbieta Bilińska [2], Mariola Dreger [1], Milena Szalata [1], Tomasz M. Karpiński [3], Artur Adamczak [2] and Douglas Siqueira de Almeida Chaves [4]**

[1] Department of Biotechnology, Institute of Natural Fibres & Medicinal Plants National Research Institute, Wojska Polskiego 71b, 60-630 Poznan, Poland; mariola.dreger@iwnirz.pl (M.D.); milena.szalata@iwnirz.pl (M.S.)
[2] Department of Botany, Breeding and Agricultural Technology for Plants, Institute of Natural Fibres & Medicinal Plants National Research Institute, Kolejowa 2, 62-064 Plewiska, Poland; elzbieta.bilinska@iwnirz.pl (E.B.); artur.adamczak@iwnirz.pl (A.A.)
[3] Chair and Department of Medical Microbiology, Poznań University of Medical Sciences, Wieniawskiego 3, 61-712 Poznan, Poland; tkarpin@ump.edu.pl
[4] Department of Pharmaceutical Sciences, Institute of Biological and Health Sciences, Federal Rural University of Rio de Janeiro, BR-465, Km 7, Seropédica, Rio de Janeiro 23897-000, Brazil; gnosy.ufrrj@gmail.com
* Correspondence: marcin.ozarowski@iwnirz.pl
† Presented at the 2nd International Electronic Conference on Plant Sciences—10th Anniversary of Journal Plants, 1–15 December 2021; Available online: https://iecps2021.sciforum.net/.

**Abstract:** *Passiflora incarnata* is ornamental and medicinal plant that contains a valuable active chemical derivatives of apigenin and luteolin. Conventional cultivation of this plant in Poland is a very problematic, caused by the low percentage of seed germination, viability of seedlings, and plant diseases which can seriously reduce the productivity of *P. incarnata*. An alternative and promising way to solve these problems may be used the technique of micropropagation, which may have applied for the plant multiplication under controlled conditions and have offered the production of healthy, pathogen-free and true-to-type plants. The aim of this study was to detrmine (1) the influence of IAA (0.1–1.0 mg $L^{-1}$), and IBA (0.1–1.0 mg $L^{-1}$) on Brasilian seed germination, and (2) the influence of various concetrations of mineral salts in Murashige and Shoog (MS), Gamborg (B-5), Shenk-Hildebrandt (SH) and Phytamax media on growth, development and condition of plant in vitro, (3) induction of adventitious shoots using nodal fragments under influence of BAP (0.1–1.0 mg $L^{-1}$), TDZ (0.1–1.0 mg $L^{-1}$), KIN (0.1–1.0 mg $L^{-1}$) with IAA (0.1 mg $L^{-1}$). Results showed that (1) MS medium with IAA (1.0 mg $L^{-1}$) has been most efective in induction of seed germination (60%); (2) Gamborg (B-5) medium has been more favorable for plant growth and development, and (3) SH with BAP (1.0 mg $L^{-1}$) and TDZ (0.1 $L^{-1}$) with IAA induced more adventitious buds and new regenerated plantlets. After rooting, 100% obtrained plants have been actimatizared to ex vitro conditions and have been observed in greenhaouse.

**Keywords:** *Passiflora incarnata* in vitro cultures; seeds; micropropagation

check for updates

**Citation:** Ożarowski, M.; Bilińska, E.; Dreger, M.; Szalata, M.; Karpiński, T.M.; Adamczak, A.; de Almeida Chaves, D.S. The Phenotypic Reactivity of *Passiflora incarnata* L. on Various Content of Mineral Salts and Regulators during Micropropagation and Acclimatization. *Biol. Life Sci. Forum* **2022**, *11*, 44. https://doi.org/10.3390/IECPS2021-11992

Academic Editor: Feibo Wu

Published: 30 November 2021



## 1. Introduction

The *Passifloraceae* family (the Passion flower) consists of 16 genera and 650 species which grow in range from large woody lianas longer than 35 m to delicate climbers [1]. These plants are distributed in the tropical regions of the South America (e.g., in Argentina, Brazil, Colombia, Ecuador, Peru, Venezuela); they are much rarer in Asia, Australia, and tropical Africa [2,3]. In Europe, plants from *Passifloraceae* family are popular in Spain and Italy. In Poland, they are collectible ornamental plants that are grown indoors or outdoors.

*Passiflora incarnata* L. is ornamental and medicinal plant that contains a valuable active chemical derivatives of apigenin and luteolin [4]. Many studies have reported that *Passiflora incarnata* is valuable source of pharmacopoeial raw material exerted mainly anxiolytic and sedative effects [5,6].

Conventional cultivation of this plant in Poland is a very problematic, caused by the low percentage of seed germination, viability of seedlings, and plant diseases which can seriously reduce the productivity of *P. incarnata*. An alternative and promising way to solve these problems may be used the technique of micropropagation, which may have applied for the plant multiplication under controlled conditions and have offered the production of healthy, pathogen-free and true-to-type plants [7]. The aim of this study was to determine (1) the influence of IAA (0.1–1.0 mg $L^{-1}$), and IBA (0.1–1.0 mg $L^{-1}$) on Brazilian seed germination, and (2) the influence of various concentrations of mineral salts in Murashige and Shoog (MS), Gamborg (B-5), Shenk-Hildebrandt (SH) and Phytamax media on growth, development and condition of plant in vitro, (3) induction of adventitious shoots using nodal fragments under influence of BAP (0.1–1.0 mg $L^{-1}$), TDZ (0.1–1.0 mg $L^{-1}$), KIN (0.1–1.0 mg $L^{-1}$) with IAA (0.1 mg $L^{-1}$).

## 2. Matherial and Methods

Seeds were obtained from the Federal Rural University of Rio de Janeiro. This primary plant culture was obtained from disinfected seeds by surface-sterilization with soaked in a sterilizing solution (20% NaOCl) for 20 min. under agitation and then seeds were washed three times with sterile deionized water in horizontal laminar airflow cabinet. These seeds were placed in basal medium MS with auxins: IAA and IBA (0.1; 0.5; 1.0 mg $L^{-1}$). After 4 weeks, shoot tips and nodal explants were excised from in vitro germinated plants and were cultured within 60 days on four kind of media such as: Murashige and Shoog (MS), Gamborg (B-5), Shenk-Hildebrandt (SH) and Phytamax media. All media have been supplemented with BAP (0.1–1.0 mg $L^{-1}$), TDZ (0.1–1.0 mg $L^{-1}$), KIN (0.1–1.0 mg $L^{-1}$) with IAA (0.1 mg $L^{-1}$). They were incubated under a 16-h photoperiod in plastic containers placed in phytotron. Control explants were cultured on a MS medium devoid of plant growth regulators. Elongated shoots obtained from the nodal explants have been transferred to MS medium with (or without) one auxin in various concentrations for rooting. Next, regenerated plants have been placed plastic pots with sterile soil in order to acclimatization of plants. After this stage, plants have been transferred to greenhouse.

## 3. Results and Discussion

Results showed that (1) MS medium with IAA (1.0 mg $L^{-1}$) has been most effective in induction of seed germination (60%); (2) Gamborg (B-5) medium has been more favorable for plant growth and development, and (3) SH with BAP (1.0 mg $L^{-1}$) and TDZ (0.1 $L^{-1}$) with IAA induced more adventitious buds and new regenerated plantlets. (4) All (100%) elongated shoots have been rooting on MS with IAA (0.5 mg $L^{-1}$). (5) After rooting, 100% obtained plants have been acclimatized to ex vitro conditions and have been observed in greenhouse. Morphological examinations showed that in vitro regenerated plants fast grew with normally developed leaves, without signs of disease. The plants were able to effective photosynthesis. In the process of micro-propagation of *P. incanata*, not only Murashige and Shoog (MS), but also Gamborg (B-5), Shenk-Hildebrandt (SH) media can be used, despite the differences in the amount of mineral salts.

## 4. Conclusions

Various techniques used in plant biotechnology, especially plant in vitro cultures, are an effective way to obtain healthy plants of *Passiflora incarnata* by vegetative reproduction. Studies have shown that both the nodal parts and the leaf blades are a promising source of adventitious shoots that effectively rooting and acclimatizing to ex vitro conditions.

**Supplementary Materials:** The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/IECPS2021-11992/s1.

**Author Contributions:** Conceptualization, M.O.; methodology, M.O., D.S.d.A.C., M.D., E.B., A.A.; formal analysis, M.O.; M.S.; investigation, M.O., E.B.; resources (seeds), D.S.d.A.C.; writing—original draft preparation, M.O., T.M.K.; writing—review and editing, M.O., T.M.K.; visualization, M.O.; supervision, M.O.; project administration. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research received no external funding.

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** Not applicable.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

1. Wiart, C. Medicinal plants classified in the family *Passifloraceae*. In *Medicinal Plants of Asia and the Pacific*; Taylor & Francis CRC: Boca Raton, FL, USA; London, UK; New York, NY, USA, 2006; pp. 101–106.
2. Dhawan, K.; Dhawan, S.; Sharma, A. *Passiflora*: A review update. *J. Ethnopharmacol.* **2004**, *94*, 1–23. [CrossRef] [PubMed]
3. Perez, J.O.; d'Eeckenbrugge Restepo, M.; Jarvis, A.; Solazar, M.; Caetano, C. Diversity of Colombian *Passifloraceae* biogeography and an updated list for conservation. *Biota Colomb.* **2007**, *8*, 1–45.
4. Ożarowski, M.; Piasecka, A.; Paszel-Jaworska, A.; Chaves, D.S.; Romaniuk, A.; Rybczynska, M.; Gryszczynska, A.; Sawikowska, A.; Kachlicki, P.; Mikolajczak, P.L.; et al. Comparison of bioactive compounds content in leaf extracts of *Passiflora incarnata*, *P. caerulea* and *P. alata* and in vitro cytotoxic potential on leukemia cell lines. Rev. *Bras. Farmacogn.* **2018**, *28*, 179–191. [CrossRef]
5. *European Pharmacopoeia 7.0.*, 7th ed.; *Passifloreae herba*; European Directorate for the Quality of Medicines & HealthCare: Strasbourg, France, 2011; pp. 1209–1210.
6. EMA, European Medicines Agency. *Assessment Report on Passiflora incarnata L., herba. EMA/HMPC/669738/2013*; Committee on Herbal Medicinal Products (HMPC): London, UK, 2014.
7. Ożarowski, M. Influence of the physico-chemical factors, plant growth regulators, elicitors and type of explants on callus culture of medicinal climbers of *Passiflora* L. *Herba Pol.* **2011**, *57*, 58–75.