

Elicitation Effect on Alkaloid Production in *Polyalthia bullata* Callus at Different Growth Incubation Time

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Introduction



Family : Annonaceae
Genus : *Polyalthia*
Vernacular name : Tongkat Ali Hitam
(Chee & Nik, 2013)

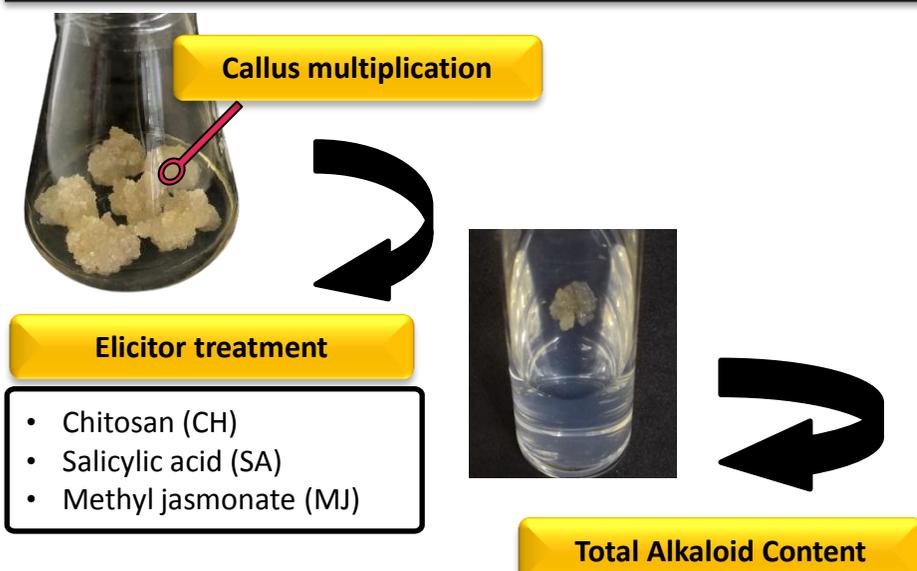
Problem Statement

- ❖ Massive collection of *P. bullata* from the forest can lead to extinction.
- ❖ The low of alkaloid production in intact plants for commercialization purposes

Objectives

To study the effect of elicitors on *P. bullata* callus at different concentrations and incubation time towards production of alkaloids and callus growth

Methodology



Total alkaloid content of *P. bullata* crude extract and treated callus was determined based on Dragendorff's reaction according to method described by Mishra, Srivastava & Akhtar (2018).

Results and Discussion

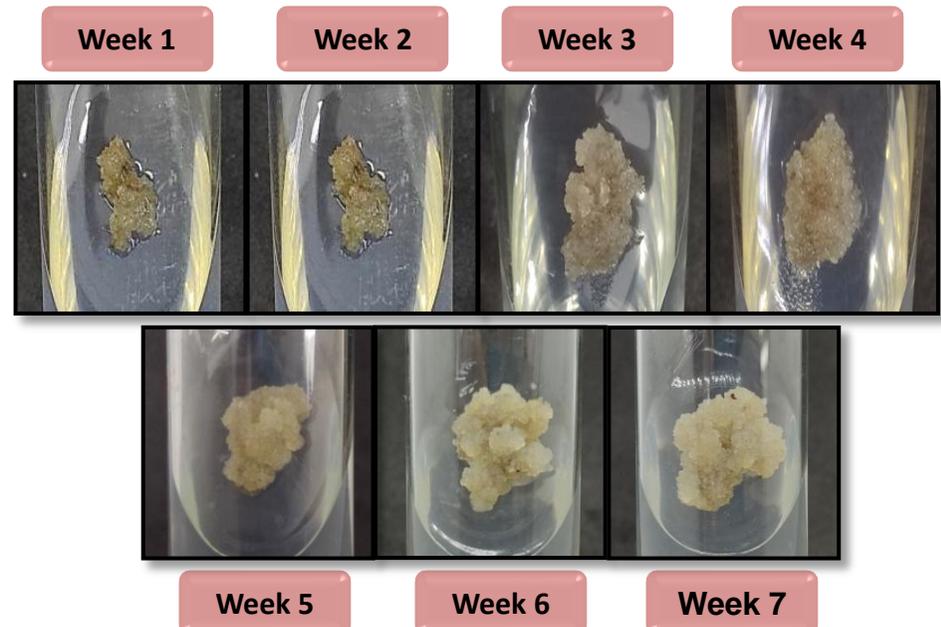
Callus Morphology

- ❑ The morphogenic responses of callus were identified to be improved as the friability of callus was enhanced with the combination of elicitors with 2,4-D.
- ❑ The best callus morphology was represented by the callus treated with 30µM 2,4-D + 50µM salicylic acid at week 7 as friable and whitish callus were obtained.
- ❑ Salicylic acid facilitates cell proliferation of callus suggesting the role of salicylic acid as endogenous growth regulator, which regulates cell division (Govindaraju & Arulselvi, 2018; Abdelnasser, 2012).

References

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- Subramaniam, S., Sundarasekar, J., Sahgal, G., & Murugaiyah, V. (2014). Comparative analysis of lycorine in wild plant and callus culture samples of *Hymenocallis littoralis* by HPLC-UV method. *The Scientific World Journal*, 2014, 1-6

Appearance of Callus Grown on MS + 30 µM 2,4-D + 50 µM Salicylic Acid



Callus Biomass

- ❑ The callus treated with MS + 30 µM 2,4-D + 150 µM SA exhibited the highest fresh weight (1.280 ± 0.003 FW) and dry weight (0.063 ± 0.003 mg DW), respectively at week 7.
- ❑ The callus growth was enhanced when cultured on media supplemented with the combination of 2,4-D and elicitors instead of elicitors alone.
- ❑ 2,4-D is an auxin-like herbicide which able to mimic auxin at low concentration by promoting cell division and elongation (Song, 2014).

Total Alkaloid Content

- ❖ Total alkaloid content of methanolic leaf extract was 4.61 ± 0.04 µg/mg DW.
- ❖ Callus grew on medium supplemented with 30µM 2,4-D + 50µM Chitosan recorded the highest total alkaloid content at Week 7 (8.290 ± 0.137 µg/mg DW).
- ❖ The addition of elicitors alone did not significantly affect the accumulation of alkaloid in *P. bullata* callus. Combination of 2,4-D and elicitor demonstrated better accumulation of alkaloid.
- ❖ Plant growth regulators (PGRs) such as 2,4-D are known to have effects not only on cell differentiation and proliferation but also on the secondary metabolite biosynthesis pathway of the cell (Subramanian et al., 2014). There could be a probability that 2,4-D and chitosan produce synergistic effect in enhancing the production of alkaloid.

Conclusion

- ✓ Elicitation of callus and addition of auxin-like herbicide, the 2,4-D was able to enhance the production of alkaloid in *P. bullata* callus.
- ✓ The combination between elicitors and 2,4-D might activate the expression of alkaloid biosynthetic genes, therefore enhancing the accumulation of alkaloid content in *P. bullata* callus.