PRODUKSI STEVIOSIDA PADA KALUS *Stevia rebaudiana* Bertoni M. DENGAN KOMBINASI AUKSIN, SITOKININ DAN GA_3

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INTISARI

Stevia merupakan sumber pemanis alami yang mempunyai tingkat kemanisan 300 kali lebih manis dibandingkan dengan gula tebu. Bahan pemanis tanaman ini terutama terdapat pada daun yang mengandung gula steviosida dan rebaudiosida-A (glikosida diterpen). Teknik kultur *in vitro* dapat meningkatkan produksi steviosida pada kalus dengan modifikasi medium pertumbuhan kalus. Penelitian ini bertujuan memperoleh kombinasi ZPT untuk menghasilkan kalus dengan konsentrasi steviosida terbesar. Penelitian telah dilakukan di Laboratorium Bioteknologi, Fakultas Biologi, UGM dan Laboratorium Kimia, Fakultas Mipa, Universitas Muhammadiyah Malang (UMM) sejak Juni – Oktober 2015. Penelitian dibagi menjadi 2 tahap, 1) tahap induksi kalus, eksplan daun stevia ditanam pada medium MS dengan kombinasi ZPT auksin/NAA (2 mg/L), 2,4-D (2 mg/L) dan sitokinin/BAP (0,5 dan 1 mg/L); 2) tahap produksi steviosida, kalus terbaik dari tahap induksi kalus ditanam pada medium MS dengan kombinasi ZPT GA_3 (1; 3; 5 mg/L) dan BAP (1 mg/L). Semua perlakuan dengan 3 ulangan. Hasil penelitian menunjukkan bahwa kombinasi perlakuan 2,4-D 2 mg/L dan BAP 1 mg/L merupakan konsentrasi yang terbaik untuk menghasilkan kalus *Stevia rebaudiana*, dengan laju induksi kalus yaitu 12 hari, warna kalus putih, dan tekstur kalus remah. Perlakuan kombinasi GA_3 dan BAP belum mampu meningkatkan kadar steviosida dalam kalus *S.rebaudiana*, perlakuan GA_3 3 mg/L menghasilkan kalus dengan kandungan steviosida tinggi sebesar 39,40 mg/g, sedikit lebih rendah dibanding kontrol yang menghasilkan steviosida sebesar 40,01 mg/g. Kandungan steviosida pada kalus masih lebih rendah dibandingkan kandungan steviosida pada daun stevia.

Kata kunci: kalus, auksin, sitokinin, steviosida, GA_3, *Stevia rebaudiana*

PRODUCTION OF STEVIOSIDE IN CALLUS FROM *Stevia rebaudiana* BERTONI M. WITH COMBINATION AUXIN, CYTOKININ AND GA_3

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ABSTRACT

Stevia is a plant that produces natural sweetener with sweetness level about 300 times higher than sugar. This sweetener was obtained mainly in the leaf of stevia, which contains stevioside and rebaudioside-A (diterpen glycosides). Production of stevioside can be generated in callus from leaves of Stevia through the technique of plant tissue culture. The objectives of this research were to induce proliferation of callus and increasing the content of stevioside in callus. This research was conducted in the laboratory of Biotechnology, Faculty of Biology, UGM and Laboratory of Chemistry, Faculty of Natural Sciences, Universitas Muhammadiyah Malang (UMM) from June 2015. The design of the research was CRD. This reseach was divided into two stages, 1) callus production using combination of ZPT Auxin; NAA (2 mg/L) and 2,4-D (2 mg/L) and Cytokinin; BAP (0; 0.5; 1 mg/L); 2) Inducing stevioside content by using various concentration of GA_3 (1; 3; 5 mg/L) and BAP (1 mg/L). All treatment with 3 replication. Quantitative data of stevioside production in Stevia was perfomed by High Performance Liquid Chromatography (HPLC) and the value was analyzed by ANOVA. Qualitative data such as colour and texture of callus was analyzed as describtive data. The result showed that the combination of 2 mg/L 2,4-D and 1 mg/L is the best concentration to produce callus from leaves of *Stevia rebaudiana*, indicated by the highest rate of callus induction within 12 days, white color of callus, and friable texture of callus. The combination between GA_3 and BAP in culture medium did not affect the production of stevioside Addition of GA_3 3 mg/L induced production of stevioside in the callus with concentration 39,40 mg/g, Although it still lower than .the production of stevioside in control callus (without addition of GA_3) that produced 40,01 mg/g stevioside. Stevioside content in callus was still lower than the content of stevioside in the stevia leaf *ex vitro.*

Key words: callus, auxin, cytokinin, stevioside, GA_3, *Stevia rebaudiana*