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Optimization of Sucrose Concentration, PH Level and Inoculum Size for Callus Proliferation and Anti-bacterial Potential of Stevia Rebaudiana Bertoni

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Abstract : Stevia rebaudiana B. is a shrubby perennial herb of Asteraceae family that possesses the unique ability of accumulative non caloric sweet Steviol Glycosides (SGs). The purpose of the study is to optimize sugar concentration, pH level and inoculum size for inducing the callus with optimum growth and efficient antibacterial potential. Three different experiments were conducted in which Callus explant from three-months-old already established callus of Stevia reabudiana of four different sizes were inoculated on Murashige and Skoog (MS) basal medium supplemented with five different sucrose concentration and pH adjusted at four different levels. Maximum callus induction 100, 87.5 and 85.33% was resulted in the medium supplemented with 30g/l sucrose, pH maintained at 5.5 and inoculated with 1.25g inoculum respectively. Similarly, the highest fresh weight 65.00, 75.50 and 50.53g/l were noted in medium fortified with 40g/l sucrose, inoculated 1.25g inoculum and 6.0 pH level respectively. However, the callus developed in medium containing 50g/l sucrose found highly antibacterial potent with 27.3 and 26.5mm inhibition zone against P. vulgaris and B. subtilize respectively. Similarly, the callus grown on medium inoculated with 1.00g inoculum resulted in maximum antibacterial potential against S. aureus and P. vulgaris with 25 and 23.72mm inhibition zones respectively. However, in the case of pH levels the medium maintained at 6.5pH showed maximum antibacterial activity against P. vulgaris, B.subtilis and E.coli with 27.9, 25 and 23.72mm respectively. The ethyl acetate extract of Stevia callus and leaves did not show antibacterial potential against Xanthomonas campestris and Clavebactor michiganensis. In the entire experiment the standard antibacterial agent Streptomycin showed the highest inhibition zones from the rest of the callus extract, however the pure DMSO (Dimethyl Sulfoxide) caused no inhibitory zone against any bacteria. From these findings it is concluded that among various levels sucrose at the rate of 40g L-1, pH 6.0 and inoculums 0.75g was found best for most of the growth and quality attributes including fresh weight, dry weight and antibacterial activities and therefore can be recommended for callus proliferation and antibacterial potential of Stevia rebaudiana

Keywords : Steviol Glycosides, Skoog, Murashige, Clavebactor michiganensis
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