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IMPACT OF SODIUM CHLORIDE AND SORBITOL IN RISING PRODUCTION OF SOME STEROIDAL SAPONINS FROM FENUGREEK CALLUS

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ABSTRACT : Steroidal saponins often had pharmacological effects. The aim at this study is to increase some steroidal saponins in fenugreek. Callus induction occurred on cotyledon segments 5 mm long. Murshige and Skoog medium supplied with 1 mg/l from2, 4-dichlorophenoxy acetic acid inaddition to 0.4 mg/l of kinetin as well as the best mediumprepared to maintenance callus was MS medium accomplished by 0.5 mg/l of 2, 4-D and 0.5 mg/l kin. To permit a callus formation leave for a period of thirty days with darkness under the temperature of 25±2°C then analyzed the Callus using high performance liquid chromatography (HPLC).Methanol callus extract showed high concentrations of some steroidal saponins compared to the methanol extract of cotyledons. For increasing, the concentration of secondary metabolites, NaCl was added at concentrations 0, 1, 1.5 or 2 g/l, sorbitol at concentrations 0, 7, 8 or 9 g/l. NaCl at 2 g/l led to significant increase in Diosgenin and Smilagenin reached (230.28), (285.95) µg per 100 mg fresh weight of callus, respectively. NaCl at 1.5 g/l increased Tigogenin reached 80.64 µg.ml^{-1} per 100 mg. Treatment with 9 g/l sorbitol increased Diosgenin reached 385.98 µg per 100 mg, while treatment 8 g/l sorbitol significantly increased in Smilagenin and Tigogenin recorded (253.68), (73.05) µg per 100 mg, respectively.

Key words : Fenugreek, steroidal saponins, NaCl, sorbitol.

INTRODUCTION

Fenugreek (*T. foenum-graecum* L.) is an important crop belonging to the leguminosaefamily and cultivated in semi-arid regions of the world. It is a native of Asia, India and Southern Europe (Acharya *et al*, 2008). Is utilized as a Cooking flavor with spice, feed additional, defensive or eat as vegetable and Medical plant from Old Time. It contains important pharmaceutical compounds such as amino acids, alkaloids, carbohydrates. In addition to organic and inorganic compounds and minerals (Aasim *et al*, 2018) as well asa rich source of steroidal sapogenins, including (diosgenin, smilagenin, tigogenin) and diosgenin, which is one of the most important compounds (Lohvina *et al*, 2012) was utilized as a substrate for around 60% of steroid, hormone and cortisone syntheses in the drug at world (Acharya *et al*, 2010). New medicine has been appears that fenugreek to possess antimicrobial, antiparasitic, anti-ulcer activity (Baliga *et al*, 2017) anti-inflammatory, anti-oxidant, anti-diabetic and discovered as strong anti-cancer agent (Sethi *et al*, 2018). Raju *et al* (2004) mentioned which diosgenin inhibition tumor growth in human colon cancer cells with stimulated apoptosis in a dosage-subordinate method.

The studies have shown that it was possible to increase the production of callus for active substances when using abiotic stress and represent this processis adding of sodium chloride and sorbitol. Many nitrogen compounds accumulate in the plants that had been exposed to salt stress. These are contain quaternary ammonium compounds, amino acids, amides, proteins and polyamine, where they vary from plant type to control osmotic potential, protect macromolecules, storage components, maintain pH, anti-toxicity, and eliminate free radicals of the cell (Budhiraja, 2004).

Pile sugars and compatible solutes in the cytoplasm of cell as reaction to osmotic stress (Chen and Murata, 2002). Esam and Esam (2011) clarify that there was increased in total phenolics and flavonoid when media supplemented with minimum concentration of NaCl decrease growth with raise in total phenols, flavonoids and tannins whereas the high level from sodium chloride significantly reduce growth and active compound quantities.

AL-Marsoomi (2010) showed that in the study of the *Salvia officinalis* plant, the MS medium of 40 g/l sorbitol produced the highest quantity of Thujone, Camphor and