

IMPROVEMENT OF ALKALOIDS YIELD USING PHENYLALANINE AS A PRECURSOR SUPPLEMENTED TO *MORINA OLEIFERA* L. CALLUS CULTURES

Huda E. Mahood^{1*}, Bushra M. Alwash² and Kadhim M. Ibrahim³

¹College of Sciences, University of Al-Qadisiyah, Diwaniyah, Iraq.

²College of Sciences for Women, Baghdad University, Baghdad, Iraq.

³College of Biotechnology, AL-Nahrain University, Baghdad, Iraq.

*e-mail: huda7713@yahoo.com

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ABSTRACT : *Moringa oleifera* has been utilized as a source for bioactive compounds. Thus, this study aimed to increase alkaloids yield in callus cultures supplied with Phenylalanine (Phe) as a precursor. Seeds of *M. oleifera* were surface sterilized and cultured *in vitro* on Murashige and Skoog (MS) medium then used as a source of explants. Explants (leaves and stems) were cultured on MS medium supplemented with combinations of Benzyl adenine purine (BAP) and Indole -3- butyric acid (IBA). The best fresh weight for callus formation was achieved at 1.0 mg.L⁻¹ BAP and 1.5 mg.L⁻¹ IBA. Quantitative analyses with High-performance liquid chromatography (HPLC) showed alkaloids presence such as Niazirin, Benzylcarbamate and Vincosamide at low levels in leaves of mother plant, (7, 9.6, 11.1 ppm respectively). The addition of different concentrations of Phe as a precursor to the medium led to increase the levels of alkaloids in callus tissue. Phe at low concentration (3.0 mg.L⁻¹) enhanced production of Niazirin to 1000%, Benzylcarbamate 566.6% and Vincosamide 440.5% in comparison with mother plant. In contrast, the highest concentration of Phe (10.0 mg.L⁻¹) was found to be inhibitory for alkaloids yield in comparison with control and other concentrations.

Key words : *Moringa oleifera*, niazirin, benzylcarbamate, vincosamide, phenylalanine.

INTRODUCTION

Moringa oleifera Lam belongs to Moringaceae family. It has been called the Miracle tree or tree of life, in reference to its potential medicinal use. The tree is highly nutritive, drought resistant, grows very fast. It also offers many benefits for the third world countries particularly India, Pakistan, Philippines, Hawaii and many parts of Africa. It has been utilized in cleaning water (Agarwal and Mehta, 2009). Reports indicated that the plant contains various amino acids, fatty acids, vitamins, and nutrients (Nesamani, 1999) and its parts have been used frequently as herbal medicine in treatments of anti-inflammatory, paralysis and hypertension (Pari and Kumar, 2002).

The most important alkaloids derived from *M. oleifera* including Niazirin, Benzyl carbamate and Vincosamide, which have many pharmacological effects such as antitumor, anti-blood sugar and anti-inflammatory (Panda *et al*, 2013). Plant tissue culture techniques are used as an alternative method for the production of secondary metabolites when plant material is scarce or

difficult to obtain and when synthesis of these metabolites is low in intact plants (Alfen, 2014).

Several attempts have been reported to promote biosynthesis of secondary metabolites in *in vitro* cultures of different plant species such as alkaloids in *Stemona* sp. (Chaichana and Dheeranupattana, 2012), terpenoids in *Erycomal longifolia* (Parikrama and Esyanti, 2014) and phenols in *Verbascum thapsus* (AL-jibouri *et al*, 2016).

Many researchers suggest that precursors seem to be a promising approach to improve the yield of plant cells. Therefore, this study is conducted to test the potential of phenylalanine as a precursor promotes the accumulation of alkaloids in *in vitro* cultures.

MATERIALS AND METHODS

Plant source and callus induction

Seeds of *Moringa oleifera* were purchased from Agricultural Seeds and Medicinal Plants Company Cairo, Egypt. Seeds were washed under running tap water for five minutes. Surface sterilization of seeds were carried