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Variations in the cytotoxic glycoalkaloids solamargine and solasonine in different parts of the *Solanum incanum* plant during its growth and development in Oman

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Abstract

In addition to several important traditional medicine applications of *Solanum incanum*, the plant is a rich source of important cytotoxic glycoalkaloids, such as solamargine and solasonine. Because *S. incanum* is a potential source of compounds for steroid synthesis, it is worthwhile to study the content of these important glycoalkaloids during plant developmental stages. Therefore, an attempt has been made to quantitatively estimate solasonine and solamargine content using an optimized isolation process and a newly developed and validated HPTLC method using different parts of *S. incanum* plants at different developmental stages and comparing changes in the whole-plant GAs profile during the growth and development of *S. incanum* plants in Oman. Solamargine and solasonine produced well-separated compact bands with R_f values of 0.26 and 0.14, respectively, on silica gel HPTLC plates using chloroform:methanol:5% ammonia (7:3:0.5, v/v/v) after visualization using anisaldehyde sulphuric acid reagent. The chromatograms were scanned at 530 nm wavelength and the simultaneous method was linear ($r^2 \geq 0.9962$) in concentrations ranging from 50 to 2000 ng/spot for both of the drugs. The validated method was applied to analyse solamargine and solasonine in small, young, immature and mature leaves as well as stem and root parts up to the 40th week of plants' growth, and showed a rich concentration of glycoalkaloids with large variations at different stages of plant development. Hence, this study highlights the importance of developmental stages of particular organs and the overall age of the plant when harvesting for these GAs from *S. incanum* plants.

Keywords: HPTLC; Steroidal glycoalkaloids; Solamargine; Solasonine; *Solanum incanum*

1. Introduction

The genus *Solanum* (Family: Solanaceae), with more than 1700 species, is widespread in the temperate and tropical regions of the world [1,2]. It is characterized by the presence of steroidal glycoalkaloids (SGAs), which are of great interest from both ecological and human health perspectives [3–6]. Approximately 20 *Solanum* species have been recorded in Arabia. In Oman, there

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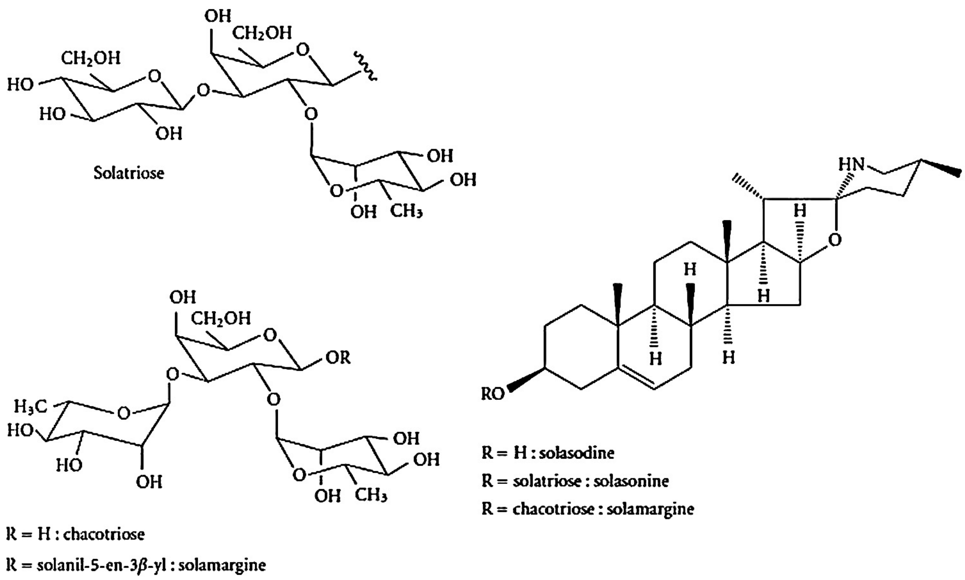

Fig. 1. Chemical structure of solasodine and its respective glycoalkaloids.

are seven known species of *Solanum*, including *S. cordatum*, *S. incanum*, *S. melongena*, *S. nigrum*, *S. surattense*, *S. tuberosum*, and *S. villosum* [7]. *Solanum incanum*, commonly known as thorn apple, is an important medicinal plant. In Oman, the leaves, fruits (berries) and roots of *S. incanum* are used as a traditional medicine to treat bruised fingers, dyspepsia, earache, and haemorrhoids [8]. Glycoalkaloids consist of two structural components, which account for their amphiphilic nature. The aglycone unit consists of a hydrophobic 27-carbon skeleton of cholestane with nitrogen incorporated into the F ring. The second unit is a hydrophilic carbohydrate side chain attached to the 3-OH position [5]. Generally, the glycoalkaloids are referred to as α-compounds; cleavage (by acid or enzymatic hydrolysis) of the individual sugars of the glycoside leads to β-, γ-, or δ-compounds, depending on the number of sugars in the side chain [9]. The majority of plants utilize glycoalkaloids as the main aglycone is solasodine in the form of water soluble triglycosidessolasonine (SN) and solamargine (SM), which occurs in approximately 200 species of *Solanum* plants [10,11]. These two compounds bear the same aglycone, solasodine, and differ from each other only in the nature of the trioses involved, namely, solatriose for solasonine and chacotriose for solamargine (Fig. 1) [5,9]. Solamargine and solasonine are economically beneficial because their chemical structures are very similar to steroidal hormones. Therefore, they have been proposed for their use as important sources in the production of medicines, such as contraceptives and steroidal anti-inflammatory drugs [2,12]. These glycoalkaloids have been studied for their antidiabetic, antifungal, antiparacetic, antibiotic, antimicrobial, antiviral and, most importantly, anticancer properties [12,13].

Biological investigations of solamargine and solasonine showed significant cytotoxicity against several human cancer cell lines and skin tumours [14]. Because *S. incanum* is a potential source of compounds required for steroid synthesis, it is worthwhile to study the content of these important glycoalkaloids during plant developmental stages.

Steroidal glycoalkaloids (SGAs) lack chromatographic groups that can be analysed in the common operating range of UV spectrophotometry and absorb only at the low wavelength end of the UV spectrum. This means that they have low UV sensitivity and can be detected and identified by UV diode array detection (DAD) only when present in relatively high amounts (5–10 ng/injection) [15–17]. The absence of chromophores in SGAs makes their detection a major challenge in the assay of a biological sample [18]. A useful analysis method consists of three major steps: extraction of the alkaloids with aqueous or non-aqueous solvents, pre-purification or removal of interfering substances (impurities), and chromatographic procedures [4,10]. Different methodologies have been proposed to analyse Solanaceae glycoalkaloids in foodstuffs and plant materials, including colorimetric, chromatographic, or immunoassay methods [9,19]. The chromatographic methods, such as high-performance thin-layer chromatography (HPTLC), high-performance