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Short Communication

Saponin, total tannin, and anthraquinone contents in *Alhagi maurorum* from the Rajasthan desert

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The present paper deals with the secondary metabolite contents such as saponin, total tannin and anthraquinone during three different developmental stages such as vegetative, flowering and fruiting in *Alhagi maurorum* from the Rajasthan desert. The study of this plant is needed due to its higher medicinal values. The aim of the study is the quantitative estimation of selected secondary metabolites. The quantitative amounts of saponin, total tannin and anthraquinone contents were evaluated by HPLC, titrimetric and GC/MS/MS methods, respectively. The maximum amounts of total tannins and saponin were observed during the flowering stage, while anthraquinone was in the vegetative stage. The most suitable stage for obtaining the maximum amount of the total tannins and saponin is flowering, but vegetative for anthraquinone.

Keywords: Anthraquinone, Rajasthan desert, Saponin, Secondary metabolites, Total tannins

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Introduction

Plants are valued as an important source of human food and animal feed and are esteemed as natural remedies to cure many health disorders. Traditionally, plants have a long history of folk medicinal uses that can be supported because they contain many bioactive commonly classed as secondary metabolites, such as phenolic, possessing multiple medicinal properties and health benefits[1]. The plants with therapeutic values are known as medicinal plants and contain secondary metabolites, which possess a variety of structural arrangements and properties. The practice of the use of herbs for healthcare delivery is a phenomenon that is handed down from one generation to another. This practice is believed to be a sum of knowledge, skills and practices based on the indigenous people's beliefs, theories and experiences. Apart from the use of medicinal plants for therapeutic purposes, some of them possess phytoconstituents, which can aid in the maintenance and improvement of health and also can prevent sicknesses and diseases[2].

The Fabaceae family consists of approximately 550 genera and more than 13,000 species, of which most of the plants have medicinal properties, and the plants of this family are used in many pharmaceutical industries because of the essential source of raw material[3]. *Alhagi maurorum* is a promising medicinal plant due to the presence of flavonoids and phenolic compounds as major contents[4]. Different parts of the plant, including its leaves, are rich in various phytochemicals such as polysaccharides, sitosterol, glycosides, terpenoids, coumarins, saponins, carotenoids, vitamins, tannins, phenolics and flavonoid compounds that act as natural antioxidants[5-6]. The antioxidant activity of this plant has been proved in previous by numerous research; accordingly, it can be applied as a source of natural antioxidants in the food and pharmaceutical industries[7]. *A. maurorum* contains fats, carbohydrates, proteins, fibres, and energy values, and it also contains some trace elements like iron, lead, calcium, nickel, sodium, magnesium, zinc, potassium and copper[8]. The plant contains many secondary metabolites like fatty acids, alkaloids, triterpenes, carbohydrates, vitamins, coumarins, steroids, resins, sterols, tannins, flavonoids, unsaturated sterols, etc[9].

A. maurorum Medikus (Family: Fabaceae), commonly known as Camelthorn, is restricted to northwest Rajasthan's rocky and gravelly soil. It is a small erect shrub armed with sharp and long spines. The plant grows with a massive rhizome system extending up to 5-6 feet into the ground. The rhizome system grows horizontally and has the potential to allow new shoots to grow upwards. Studying the plant from different growth stages is important for identifying the most suitable stage to obtain the maximum amount of desired secondary metabolites. The plant is used to treat numerous diseases such as gastro-entroenteritis[10], headache, toothache and cancer[11], liver disorders, kidney stones, urinary tract infections, etc[12]. *A. maurorum* has been used in folk medicine for a long time as a laxative, diaphoretic, expectorant, and diuretic, and the aqueous extracts of

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seeds relax the ureter and remove kidney stones[13]. Ahmad *et al*.[14], isolated numerous secondary metabolites from the methanolic extract of aerial parts of *A. maurorum*, which included β-sitosterol, trans-cinnamic acid, *p*-coumaric acid, 4-hydroxy benzoic acid, etc.

No reports are available on secondary metabolites analyses like saponin, total tannins and anthraquinone in *Alhagi maurorum*, an important medicinal plant of the Rajasthan desert. Thus, the present study was conducted to investigate the estimation of secondary metabolites such as saponin, total tannins and anthraquinone during three developmental stages, *i.e.*, vegetative, flowering and fruiting for evaluating suitable stages to harvest *A. maurorum* for obtaining the maximum amount of secondary metabolites.

Materials and Methods

For chemical analyses of the plant, leaf samples of *A. maurorum* were collected from Kanana village of the Barmer district (25° 49' 17.5944" N and 72° 25' 59.5416" E), which is located 105 km away in the south-west direction from the University Campus, Jodhpur during three developmental stages, *viz.* vegetative (August to February), flowering (March to May) and fruiting (April to July) during 2020 and 2021. The Botanical Survey of India, Jodhpur, confirmed the identification of the plant, and specimens have been deposited in the BSI herbarium. For plant analyses, fresh leaf samples were randomly collected from the fixed 4th to 5th nodes, washed with distilled water to remove adherent soil particles and allowed to dry in the oven at 80^{o}C for a minimum of 48 h for further chemical analyses. Saponin contents were estimated per gravimetric analysis methods suggested by Ezeabara *et al.*[15], and the HPLC method as per Köse and Bayraktar[16]. Total tannin contents were estimated by the titrimetric method as per Garg and Katara[17], while anthraquinone by GC/MS/MS as per Latimer[18]. The experiment for each parameter was performed in triplicate and confirmed twice.

The data collected during both the years (2020 and 2021) were subjected to analyses of variance (ANOVA) as suggested by Gomez and Gomez[19].

Results and Discussion

Data on various important secondary metabolic products such as saponin, total tannins and anthraquinone in leaves of *A. maurorum* are presented in Fig. 1a-c. Saponin is a kind of natural secondary metabolite, which is composed of sapogenin and a sugar chain[20]. Saponins have many important biological activities and pharmacological actions such as immunity enhancement, antitumor, anti-inflammatory, antifungal, and antiviral actions, blood glucose and lipid reduction, antioxidation, cardiovascular function improvement, etc.[21]. In recent years, it has been widely used in medicines, health foods, animal feed, cosmetics and other items. In addition, saponins are used as plant growth regulators and insect repellents in agriculture. Therefore, saponins have excellent research value and broad developmental prospects[22]. Saponin production is part of a plant's natural growth and developmental process and is a significant chemical obstacle to pathogenic fungal and insect resistance mechanisms[23]. They are a heterogeneous group distinguished by a structure with a steroid or triterpenoid aglycone and one or more

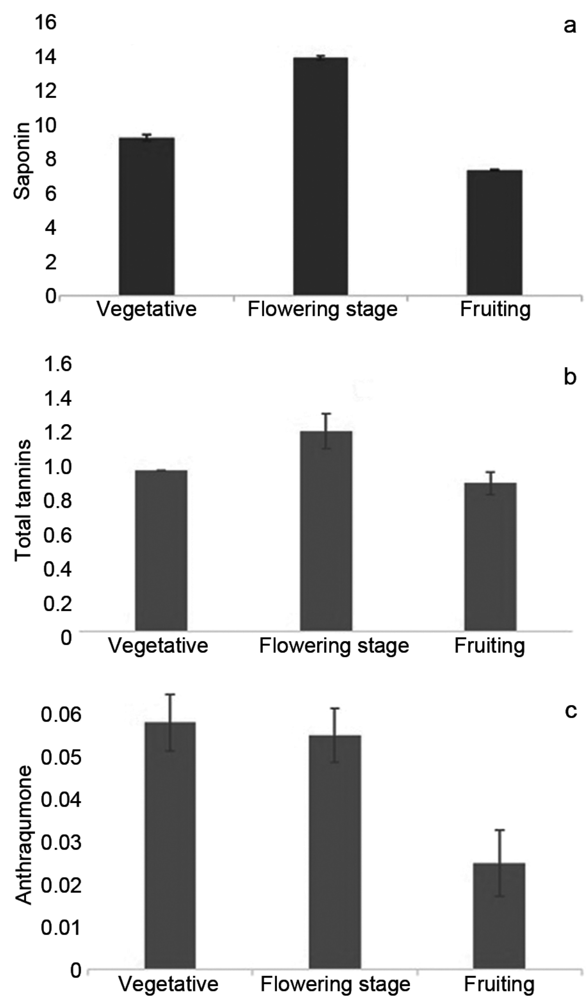

Fig. 1 — a) Saponin (%); b) Total tannin (%); and c) Anthraquinone (mg/kg) contents in *Alhagi maurorum* leaves during three growth stages (Mean values and standard error are presented).

sugar chains[24]. It is evident from Fig. 1a that values of saponin ranged from 7.31 to 13.86%, maximum during the flowering stage. Adegbaju *et al*.[25] and Rad-Sharifi *et al*.[26], reported the highest amount of saponin contents during the flowering stage in *Celosia argentea* and *Teucrium polium,* respectively, which is in accordance with present investigations.

Tannins are a heterogeneous group of water-soluble polyphenolic compounds of high molecular weight (500–3000 Daltons) with as many as 20 hydroxyl groups and are present in plants, foods and beverages[27]. Being a phenolic compound, tannins are chemically reactive and form inter- and intra-molecular hydrogen bonds that can interact with and precipitate macromolecules such as proteins and carbohydrates. They are also responsible for the astringent taste of many fruits and vegetables[27-28]. Tannins might be associated with adverse effects as antinutritional factors, causing lower dry matter intake and reduced digestion of protein and fiber[29]. In the present studies, its values ranged from 0.91 to 1.23% during three stages, being the highest in flowering and the lowest in the fruiting stage (Fig. 1b). Jugran *et al*.[30], reported the highest value of total tannin in *Valeriana jatamansi* during the flowering stage, which support present findings.

Anthraquinone can be found in all plant parts, such as roots, rhizomes, fruits, flowers and leaves. Most of these compounds are derivatives of the basic structure 9,10-anthracenedione, a tricyclic aromatic organic compound with the formula $C_{14}H_8O_2$. The latter is a yellow solid crystalline powder that absorbs visible light and is the basic structure of a large class of dyes and pigments[31]. A wide body of literature has demonstrated that natural anthraquinones possess a broad range of bioactivities such as anticancer, anti-inflammatory, immunosuppressive, antimicrobial, diuretic, cathartic, laxative, vasorelaxant, antioxidant, and phytoestrogen activities[32]. Anthraquinones are an important group of natural products occurring in higher plants as glycosides. They act as antioxidants and cholinesterase inhibitors, which decrease degeneration in Alzheimer's patients[33]. The maximum anthraquinone content was reported during the vegetative stage, while the minimum was in the fruiting stage. The values ranged from 0.025 to 0.058 mg/kg (Fig. 1c). Kallenberger *et al*.[34], observed the maximum anthraquinone before the flowering stage in *Fallopia* spp. The present findings also support the above observations.

Statistical analyses revealed that saponins in *A. maurorum* were significant at P <0.5 level, whereas the remaining parameters were non-significant.

Conclusions

It is concluded from the present investigations that the maximum amount of saponin and total tannin contents were accumulated during the flowering stage, while anthraquinone was during the vegetative stage in *A. maurorum.* Therefore, the flowering stage was found to be the most favourable for obtaining the maximum production of saponin and total tannins, whereas the vegetative stage for anthraquinone. Thus, based on the present findings, we can determine the most suitable developmental stage to obtain the maximum amount of these products for commercial use.

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Conflict of interest

The authors declare no conflict of interest.

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