GC-MS analysis of phytocomponents in the ethanolic and aqua-ethanolic extracts of *Uraria picta* Desv

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Abstract

*Uraria picta* Desv. is one of the ten species of *Dashmoolarista*, a well-established ayurvedic drug of Indian system of medicine. The aim of the present study is to determine the phytoconstituents in ethanolic and aqua-ethanolic (2:80) extracts of stem of *U. picta*. The results showed the presence of 4-Carboxy cyclohexanone (RT- 11.686, 16.11%), Octadecanoic acid (RT- 21.332, 11.27%) and α-D-Mannofuranoside, 1-O-decyl-(RT - 23.108, 72.62%) compounds in ethanolic extract while α-amyrone (RT- 27.45, 58.65%) and Stignast-4-en-3-one (RT- 32.883, 28.78%) compounds in aqua-ethanolic extract. The identified phytocompounds have biological and commercial significance.

Keywords: *Uraria picta*, stem, ethanolic and aqua-ethanolic extracts, GC-MS analysis

Introduction

Since ancient era, nature functions as a complete store house of remedies to cure all ailments of mankind (Kokate et al., 2002) [1] and provides us drugs in the form of herbs, plants and algae to cure diseases without any toxic effect (Harborne, 1998) [2]. In present time also, more than 80% of world population are still relying on traditional system of medicines to cure their diseases (Anand et al., 2012; Julsing et al., 2007) [3, 4]. It is an established fact that the medicinal value of plants lies in their bioactive phytochemical constituents that produce specific physiological action on the human body (Akinmoladun et al., 2007) [5]. Phytoconstituents such as alkaloids, essential oils, flavonoids, tannins, terpenoids, saponins, phenolics, cardiac glycosides etc form the backbone of the modern medicine (Mohan et al., 2019) [6].

*Uraria picta* Desv. (Syn. *Doodia picta* Roxb., *Hedysarum pictum* Jacq., Family: Fabaceae) is commonly known as Prishnaparni or Pithvan and widely distributed throughout India, Bangladesh, Sri Lanka, Tropical Africa, Malay Islands, Philippines, Australia, Africa and almost all parts of Asia (McNeil et al., 2006; Ohashi and Iokawa, 2007) [7, 8]. It is one of the important constituents of “Dashmoolarista”, a well-established ayurvedic drug of Indian system of medicine, prepared from the roots of 10 medicinal plants and used for treating general fatigue, oral sores and several gynecological disorders (Yadav et al., 2009) [9]. Dashmool is also used as basic ingredient in manufacture of over 109 drug formulations (Pathak et al., 2005) [10]. Traditionally, the plant is used as an antitode to the venom of a dangerous Indian snake, *Echis carinata* (Kirtikar and Basu, 1993) [11]. Its leaves are a good antiseptic and are used against gonorrhea. Leaves of *U. picta* also showed antiinxiety activity (Garg et al., 2012) [12]. The fruits, pods are effective against oral sores in children, roots are being used against cough, chills and fever (Kirtikar and Basu, 1993; Yusuf et al., 1994) [11, 13]. Moreover, research works such as phytochemical screening and elemental analysis in different plant parts (Saxena et al., 2014) [14], HPLC analysis of rhoifolin in different plant parts (Saxena et al., 2014) [15], phytochemical screening and HPTLC finger print analysis of aerial parts (Saxena et al., 2016) [16] and assessment of variation in rhoifolin content in aerial parts from different locations of Madhya Pradesh (Saxena et al., 2016) [17] have been carried out earlier in our laboratory. In further investigation, we have identified the phytocompounds in ethanolic and aqua-ethanolic extracts of stem of *U. picta* through GC-MS analysis.
Materials and Methods

Chemicals and reagents
AR grade chemical and solvents and distilled water were utilized in the experiments.

Collection of plant materials

*U. picta* species was collected by following the guidelines of good agricultural and collection practices (GACP) for medicinal plants (Anon, 2003) [18] from the Khandwa region of Madhya Pradesh, India during December month.

Processing of plant materials

Plant materials were washed thoroughly in running water to remove soil and other foreign particles. Stem were separated, cut into small pieces and dried in shade. Shade dried material was powdered using pulverizer. The powdered material was utilized for making extracts.

Preparation of extracts

Powdered stem sample was subjected to successive extraction with Ethanol and Ethanol: Water (Aqua-alcoholic) (80:20) (Varghese *et al.*, 2013) [19]. A total of 20g of dried powder was extracted in 250 ml of each solvent in successive manner for 12 hrs. Solvents were evaporated to dryness to yield the respective extracts which were used for GC-MS analysis.

GC-MS analysis

Ethanolic and aqua-ethanolic extracts were subjected to chemical analysis by using GC-MS instrument, Perkin Elmer, USA & Model - Auto system XL with Turbo Mass. Compounds were separated on PE-5MS 30m x 0.250mm x 0.250µm column. Oven temperature was programmed as follows: isothermal temperature of 75°C for min and then increased up to 280°C at the rate of 10 °C/ min and held for 15 min. Injection temperature was 250 °C and injection volume was 1µl. EI source temperature was set as 220 °C. Helium gas was used as carrier gas at 1 ml/ min flow rate. MW range was set at 22 to 620 amu.

Identification of compounds

Interpretation of mass spectrum of GC-MS was conducted using the database of NIST. The spectrum of investigated components was compared with spectrum of known components stored in NIST. Molecular weight, molecular formula and number of hits were used to identify the name of compounds from NIST.

Results and Discussion

GC-MS chromatograms of ethanolic and aqua-ethanolic extracts of *U. picta* stem are given as Fig. 1 and Fig. 2 respectively. On comparison of the mass spectra of the constituents with the NIST library, the three phytocompounds were characterized and identified in ethanolic and two phytoconstituents in aqua-alcoholic extracts, denoted in Table 1. The biological as well as commercial importance of chemical compounds identified from both extracts is given in Table 2. Chemical structures of compounds identified by GC-MS in ethanolic extract is given in Fig. 3. Mass spectrum and chemical structures of compounds identified by GC-MS in aqua-ethanolic extract is given in Fig. 4.

Fig 1: GC-MS chromatogram of ethanolic extract of *U. picta* stem
**Fig 2:** GC-MS chromatogram of aqua-ethanolic extract of *U. picta* stem

**Table 1:** Phytocompounds identified in ethanolic and aqua-ethanolic (80:20) extracts of stem of *U. picta* by GC-MS

| Extracts          | RT  | Name of phytocompounds          | Molecular Formula | Molecular Weight | Area  |
|-------------------|-----|---------------------------------|-------------------|------------------|-------|
| Ethanolic         | 11.868 | 4-Carboxy cyclohexanone              | C7H10O3             | 142              | 16.11 |
|                   | 21.332 | Octadecanoic acid                | C18H36O2            | 284              | 11.27 |
|                   | 23.108 | α-D-Mannofuranoside, 1-O-decyl-   | C16H32O6            | 320              | 72.62 |
| Aqua-ethanolic (20:80) | 27.45 | α-amyrone                       | C30H48O             | 424              | 58.65 |
|                   | 32.883 | Stigmast-4-en-3-one              | C29H48O             | 412              | 28.78 |

**Table 2:** Bioactivity/ importance of phytocomponents identified in ethanolic and aqua-ethanolic (80:20) extracts of stem of *U. picta* by GC-MS

| S. No. | Name of compound          | Nature of compound            | Biological Activity/ Importance                                                                 |
|--------|---------------------------|-------------------------------|-------------------------------------------------------------------------------------------------|
| 1.     | 4-Carboxy cyclohexanone   | Keto acid                     | It is used in the synthesis of 4'-hydroxybiphenyl-4-carboxylic acid, a raw material for the synthesis of polymers and liquid crystals (Tohru et al., 1998; Miura et al., 1991). |
| 2.     | Octadecanoic acid         | Saturated fatty acid          | 5-α-reductase inhibitor, hypocholesterolemic, suppository, cosmetic, lubricant, surfactant & softening agent, perfumery, propecic, flavor (Mathavi P et al., 2015; Markkas and Govindharajalu, 2015; Arora and Kumar, 2017). |
| 3.     | α-D-Mannofuranoside, 1-O-decyl- | Carbohydrate                | Not reported                                                                                     |
| 4.     | α-amyrone                 | Non-steroidal triterpenoid    | Anti-inflammatory (Patrícia et al., 2015)                                                        |
| 5.     | Stigmast-4-en-3-one       | Steroid                       | Hypoglycemic effect (Lexander et al., 2004; Fathaiya et al., 1995)                              |
**Fig 3**: Chemical structures of compounds identified by GC-MS in ethanolic extract (a) 4-Carboxy cyclohexanone (b) Octadecanoic acid (c) α-D-Mannofuranoside, 1-O-decyl-
In the ethanolic extract, the main compounds were α-D-Mannofuranoside, 1- O-decyl-, a carbohydrate (72.62%), 4-Carboxy cyclohexanone, a keto acid (16.11%) and Octadecanoic acid, a saturated fatty acid (11.27%) were the main compounds. 4-Carboxy cyclohexanone is reported to be used in the synthesis of 4-hydroxybiphenyl-4-carboxylic acid which is a very important as a raw material for the synthesis of polymers and as an intermediate for the synthesis of liquid crystals (Tohru et al., 1998; Miura et al., 1991) [20-21]. Octadecanoic acid shows 5α-reductase inhibitor activity, hypo cholesterolemic property and used in suppository, cosmetic, lubricant, surfactant and softening agent, perfumery, propecic, flavor (Mathavi P et al., 2015; Markkas and Govindharajulu, 2015; Arora and Kumar, 2017) [22-24]. Similarly, the most prevailing compounds identified in aqua-alcoholic extract were α–amyrone, a non-steroidal triterpenoid (58.65%) and stigmast-4-en-3-one, a steroid molecule (28.78%). α–amyrone is described as an anti-inflammatory agent (Almeida et al., 2015) [25] and stigmast-4-en-3-one has hypoglycemic effect (Alexander-Lindo et al., 2004; Jamaluddin et al., 1995) [26, 27]. GC-MS analysis showed the presence of important compounds in both extracts of U. picta stem which add value to the use of this plant in various ayurvedic formulations for treating several ailments by traditional practitioners. However, investigations on isolation and characterization of particular phytocompounds along with its biological activities will definitely give fruitful results.

Conclusion
From the results, it can be concluded that U. picta stem contains phytocompounds of pharmacological and other commercial importance. Therefore, this plant is utilized in numerous ayurvedic formulations. This study adds more value in the therapeutic temperament of this medicinal herb.

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