P-ISSN: 2349–8528
E-ISSN: 2321–4902
www.chemijournal.com
IJCS 2020; 8(3): 2205-2210

Received: 20-03-2020
Accepted: 24-04-2020

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

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DOI: https://doi.org/10.22271/chemi.2020.v8.i3ae.9537

Abstract
Uraria picta Desv. is one of the ten species of Dashmoolarishta, 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 (20:80) extracts of stem of *U. picta*. Th results showed the presence of 4-Carboxy cyclohexanone (RT- 11.868, 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 Stigmast-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 aliments 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]. Phytocompounds 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 (McNeill *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 antidote 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 antianxiety 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.

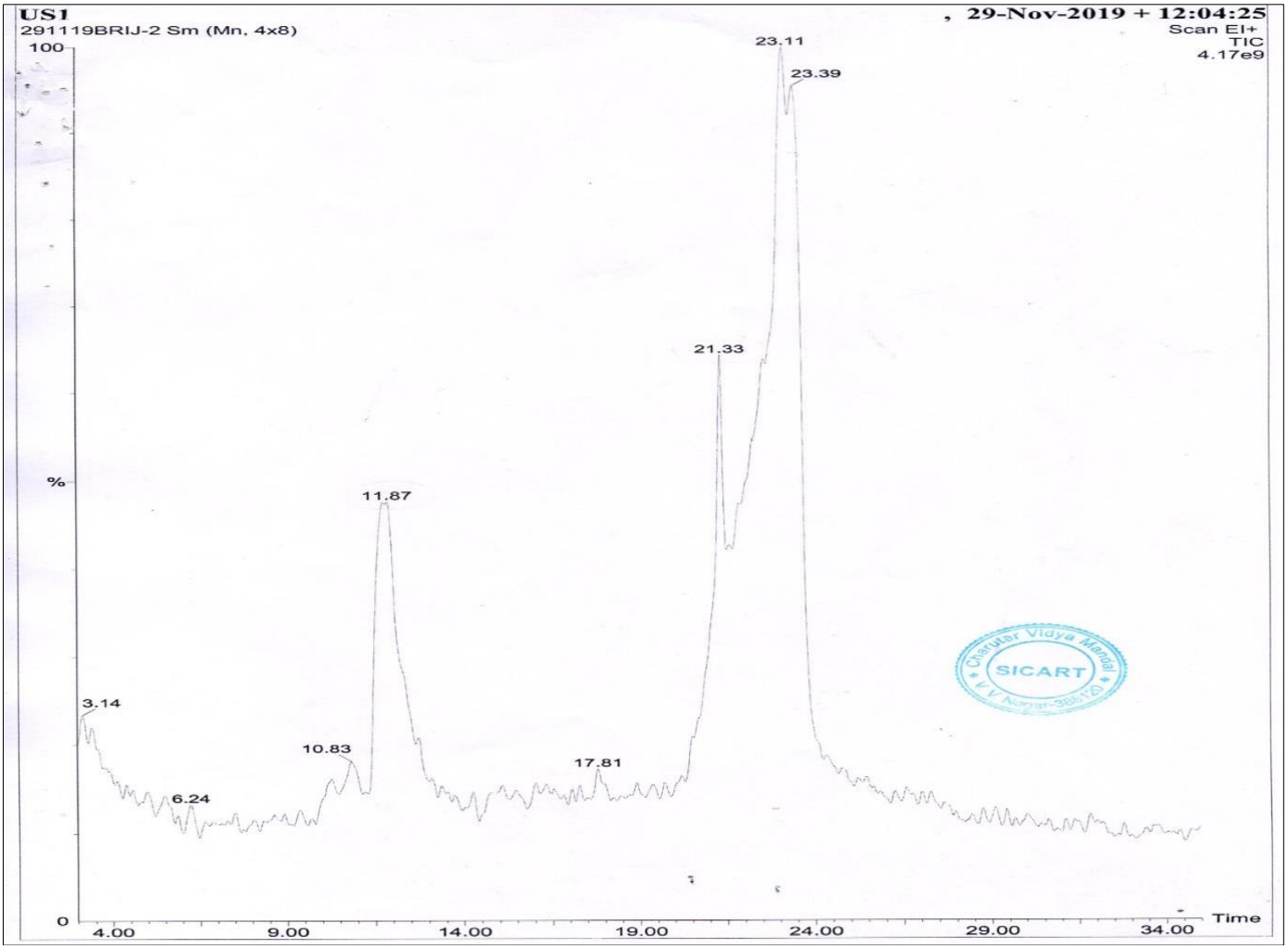


Fig 1: GC-MS chromatogram of ethanolic extract of *U. picta* stem

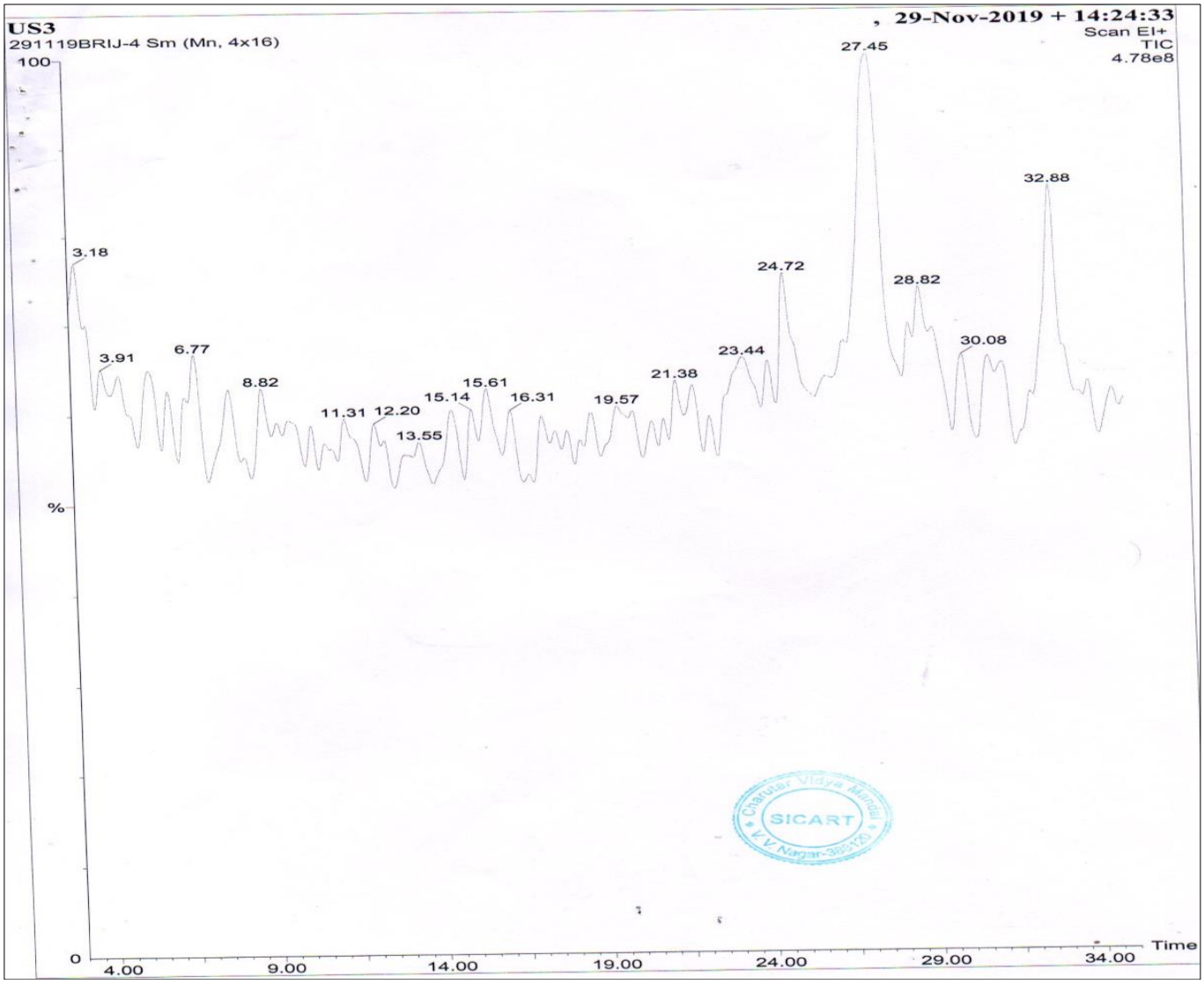


Fig 2: GC-MS chromatogram of aqua-ethanolic extract of *U. picta* stem

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)

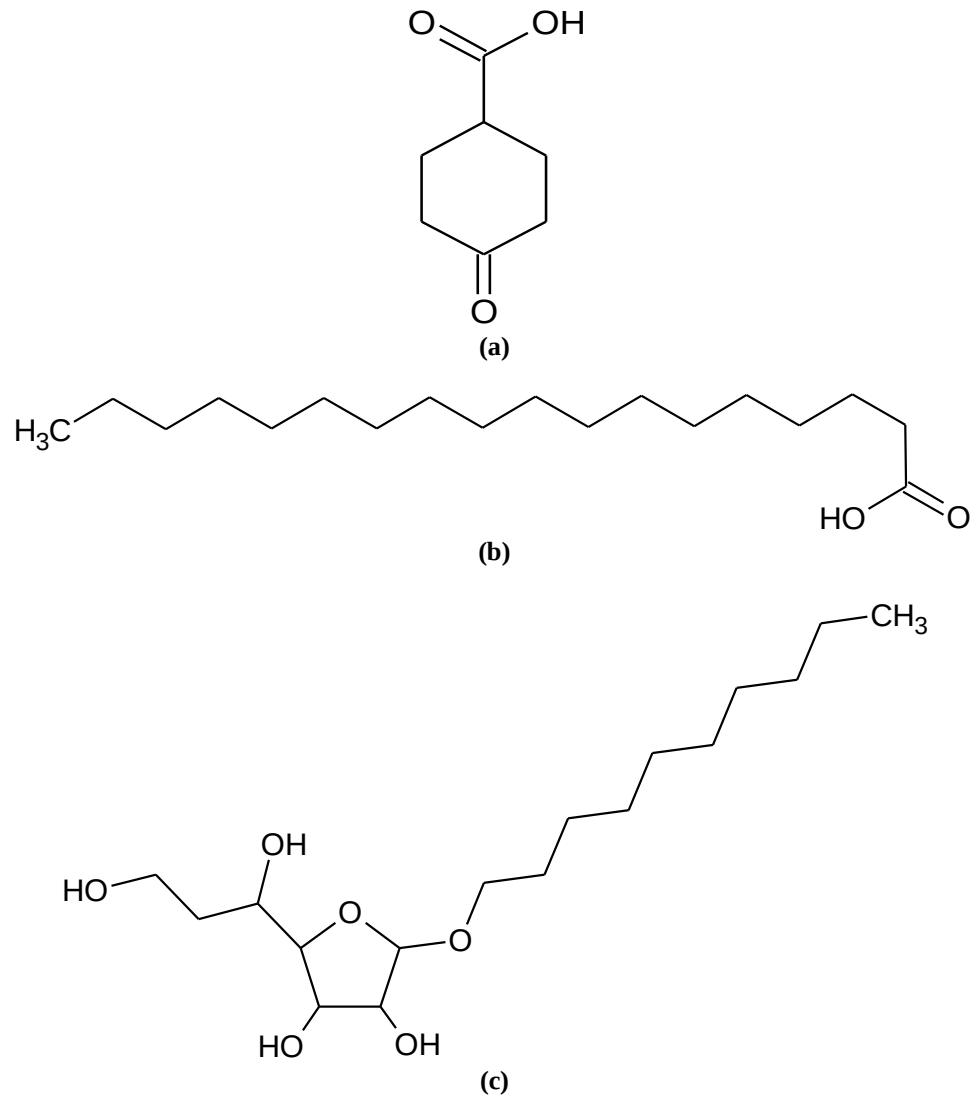
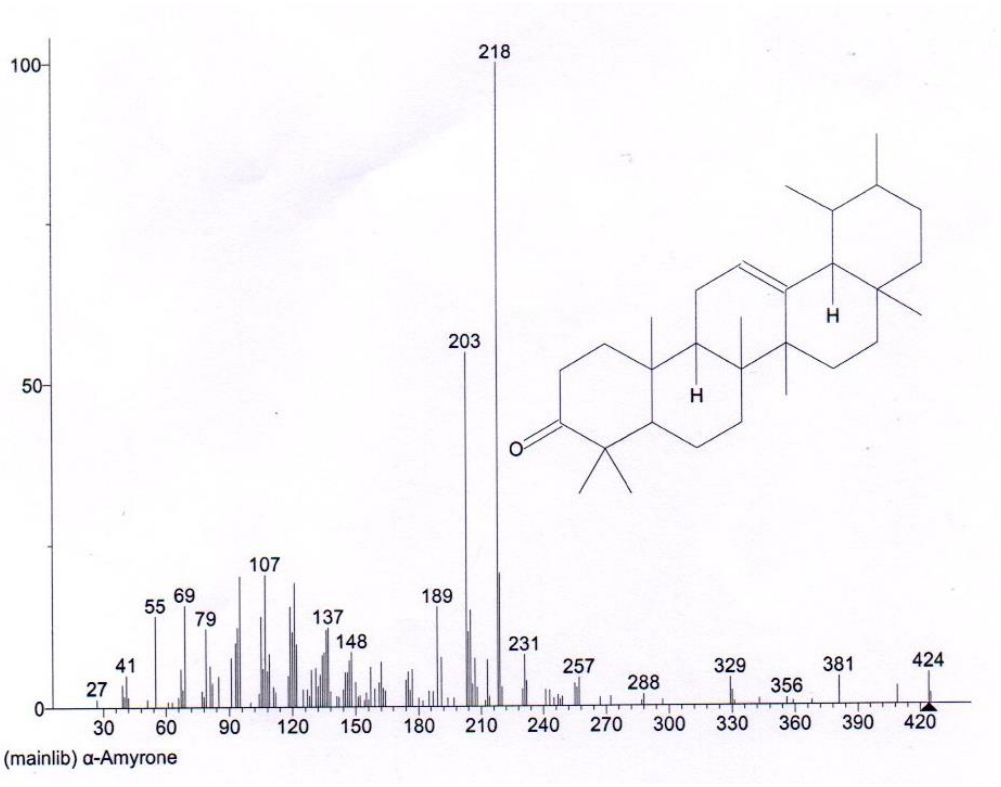
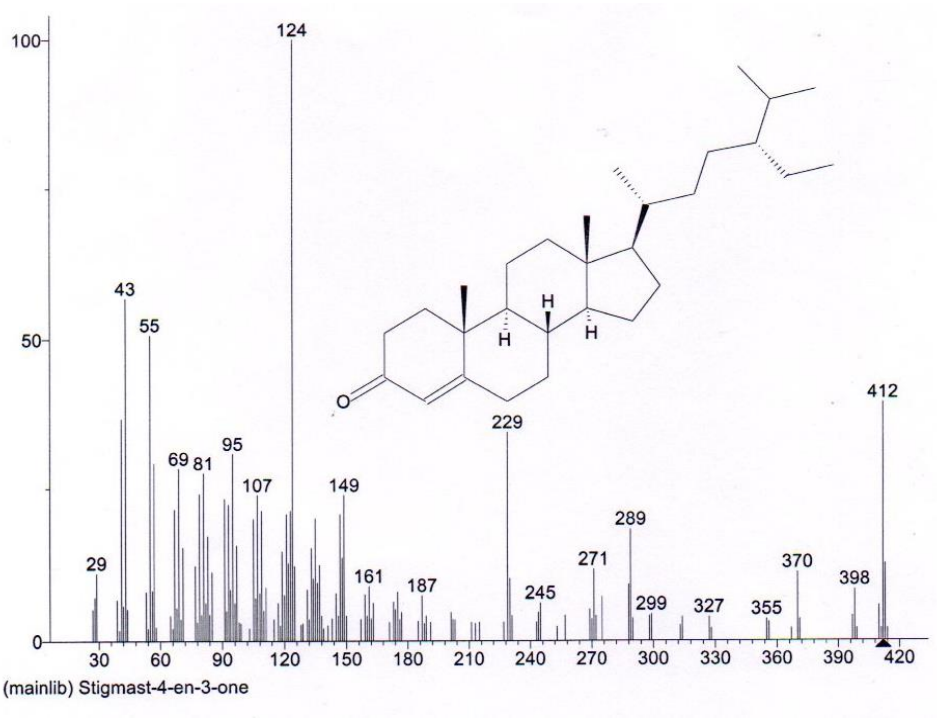


Fig 3: Chemical structures of compounds identified by GC-MS in ethanolic extract (a) 4- Carboxy cyclohexanone (b) Octadecanoic acid (c) α - D-Mannofuranoside, 1- O-decyl-



(a)



(b)

Fig 4: Mass spectrum and chemical structures of (a) α- amyrone and (b) Stigmast-4-en-3-one identified by GC-MS in aqua-ethanolic extract

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 5-α-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 Govindharajalu, 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.

Acknowledgements

Authors are grateful to the Principal of Govt. Science College, Jabalpur and Director of Tropical Forest Research Institute, Jabalpur for providing necessary facilities to conduct the research work.

References

1. Kokate CK, Purohit AP, Gokhale SB. Textbook of pharmacognosy. Nirali Prakasan, Pune. 2002; 18:1-4.
2. Harborne JB. Phytochemical methods. London. 3rd ed., Chapman and Hall, Ltd, 1998, 1-302.
3. Anand SP, Jayakumar E, Jeyachandran R, Nandagobalan V, Doss A. Plant Tissue Culture Biotechnology. 2012; 22(1):87-91.
4. Julsing KM, Quax JW, Kayser O. The Engineering of Medicinal Plants: Prospects and Limitations of Medicinal Plant Biotechnology. In: Medicinal Plant Biotechnology (eds Oliver K. and Wim J.Q.) WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim, 2007.
5. Akinmoladun AC, Ibukun EO, Afor E, Obuotor EM, Farombi EO. Phytochemical constituent and antioxidant activity of extract from the leaves of *Ocimum gratissimum*. Scientific Research and Essays. 2007; 2(5):163-166.
6. Mohan B, Saxena HO, Kakkar A, Mishra MK. Determination of antioxidant activity, total phenolic and flavonoid contents in leaves, stem and roots of *Uraria picta* Desv. Environment Conservation Journal. 2019; 20(3):1-8.
7. McNeill J, Barrie FR, Burdet HM, Demoulin V, Hawksworth DL, Marhold D *et al*. International Code of Botanical Nomenclature (Vienna Code). Koeltz Scientific Books, Konigstein – Germany, 2006.
8. Ohashi H, Iokawa Y. A Revision of *Uraria* (Leguminosae) in Taiwan. Taiwania. 2007; 52:177-183.
9. Yadav AK, Yadav D, Shanker K, Verma RK, Saxena AK, Gupta MM. Flavone glycoside based validated RP-LC method for quality evaluation of Prishniparni (*Uraria picta*). Chromatographia. 2009; 69:653-658.
10. Pathak JM, Krishnamurthy R, Chandorkar MS, Gulkari VD, Gupta R. Identification of high yielding genotypes

of Dashmool Shalparni (*Desmodium gangeticum*) drug plant and its cultivation under high density planting. Indian Journal of Horticulture. 2005; 62:378-384.
11. Kirtikar KR, Basu, BD. Indian Medicinal Plants, Vol 2. International Book Publishers Dehradun, India, 1993.
12. Garg N, Garg M, Maan AS, Sandhu BS, Mittal S, Goyal S. Phytochemical studies and anti-anxiety activity of *Uraria picta* leaves. Journal of Pharmaceutical Research and Opinion. 2012; 2(5):39-40.
13. Yusuf M, Chowdhury JU, Wahab MA, Begum J. Medicinal plants of Bangladesh. BSCIR Laboratories, Chittagong, Bangladesh, 1994.
14. Saxena HO, Soni A, Mohammad N, Choubey SK. Phytochemical screening and elemental analysis in different plant parts of *Uraria picta* Desv. A Dashmul species. Journal of Chemical and Pharmaceutical Research. 2014; 6(5):756-760.
15. Saxena HO, Soni A, Mohammad N, Kakkar A, Singh N. HPLC analysis of rhoifolin in different plant parts of *Uraria picta*: a dashmool species. Indian Journal Tropical Biodiversity. 2014; 22(2):199-201.
16. Saxena HO, Mohan B, Kakkar A, Pawar G, Choubey SK. Phytochemical screening and HPTLC finger print analysis of aerial parts of *Uraria picta* Desv. – a dashmool species. Journal of Pharma Research. 2016; 5(5):87-93.
17. Saxena HO, Mohan B, Kakkar A. Assessment of variation in rhoifolin content in aerial parts of *Uraria picta* Desv. from different locations of Madhya Pradesh. Journal of Pharmacy Research. 2016; 10(5):185-190.
18. Anonymous. WHO guidelines on good agricultural and collection practices (GACP) for medicinal plants. World Health Organization, Geneva, 2003.
19. Varghese S, Narmadha R, Gomathi D, Kalaiselvi M, Devaki K. Phytochemical screening and HPTLC finger printing analysis of *Citrullus lanatus* (Thunb.) seed. Journal of Acute Disease. 2013; 2(2):122-126002E.
20. Tohru M, Teruyuki N, Hideki M. Process for the preparation of 4-hydroxybphenyl-4-carboxyl acid. U.S. Patent. 1998; 4:617-755.
21. Miura T, Yamashitamachi OF, Nagata T, Shiraganemachi OF, Mizuta H, Miike OF. Process for the preparation of 4'-hydroxybiphenyl-4-carboxylic acid. E.U. Patent, 1991, 0240362B1.
22. Mathavi P, Nethaji S, Velavan. GC-MS analysis of phytocomponents in the methanolic extract of *Shorea robusta*. International Journal of Science and Research. 2015; 4:1935-1938.
23. Markkas N, Govindharajalu M. Determination of phytocomponents in the methanolic extract of *Mollugo cerviana* by GC-MS analysis. International Journal of Research in Biological Sciences. 2015; 5(4):26-29.
24. Arora S, Kumar G. Gas Chromatography-Mass Spectrometry (GC-MS) determination of bioactive constituents from the methanolic and ethyl acetate extract of *Cenchrus setigerus* Vahl (Poaceae). The Pharma Innovation Journal. 2017; 6(11):635-640.
25. Almeida PDO, Boleti APA, Rüdiger AL, Lourenço GA, Junior VFV, Lima ES. Anti-Inflammatory Activity of Triterpenes Isolated from *Protium paniculatum* Oil-Resins. Evidence-Based Complementary and Alternative Medicine, 2015, 1-10.
26. Alexander-Lindo RL, Morrison EYSA, Nair MG. Hypoglycaemic effect of stigmast-4-en-3-one and its corresponding alcohol from the bark of *Anacardium occidentale* (Cashew). Phytotherapy Research. 2004; 18(5):403-7.
27. Jamaluddin F, Mohameda S, Lajis N. Hypoglycaemic effect of Stigmast-4-en-3-one, from *Parkia speciose* empty pods. Food Chemistry. 1995; 54(1):9-13.