

Research Article

www.ijrap.net



(ISSN Online:2229-3566, ISSN Print:2277-4343)

GAS CHROMATOGRAPHIC-MASS SPECTROMETRIC ANALYSIS OF K.G. 101 (KSHEERAGULOOCHI 101): AN AYURVEDIC PATENT PRODUCT

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Received on: 06/02/23 Accepted on: 23/03/23

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DOI: 10.7897/2277-4343.140238

ABSTRACT

K.G.101 (Ksheeraguloochi 101) is an Ayurvedic patent product manufactured by Sitaram Ayurveda Private Ltd. Thrissur, Kerala. This is widely used by practitioners in the management of joint disorders. To explain this mechanism of action, the knowledge of biomedical compounds in the product is mandatory. The present study deals with the Gas chromatographic-mass spectrometric analysis of K.G.101 to explain its medicinal value. This type of study is the need of the hour for better scientific validation and a new approach to the treatment. The Gas chromatographic-mass spectrometric profile indicated the presence of nearly 39 molecules. Most compounds show anti-inflammatory potential, which explains their action in inflammatory joint disorders. The antioxidant property of the compounds explains their rejuvenating property. Some compounds show anti-cancerous activity also, which yet has to be explored.

Keywords: Ksheeraguloochi 101, Gas chromatographic-mass spectrometric analysis, joint disorders

INTRODUCTION

K.G.101 (Ksheeraguloochi 101) is an Ayurvedic patent drug. K.G.101 is medicated oil presented in the form of a soft gelatin capsule. Each soft gelatin capsule contains 175 mg of medicated oil. It is widely used by practitioners in managing inflammatory and degenerative joint disorders.

The ingredients of K.G.101 are milk, sesame oil and *Tinospora cordifolia* (*Guloochi*). This *Tinospora cordifolia* (*Gulocchi*) paste is processed in milk and sesame oil for 101 times. *Tinospora cordifolia* is said to have anti-arthritic, anti-osteoporotic, immunomodulatory and antioxidant effects¹.

Most traditional medicines contain more than one herb, and the preparation is elaborate. The final product may have only a few molecules in the individual compounds. New compounds may be formed in the process. The knowledge of biomedical compounds in such medicines helps to understand the mechanism of action.

Gas chromatographic-mass spectrometric (GC-MS) analysis was done to determine the type of molecule present in the Ksheeraguloochi 101 and explain its medicinal value. The present study demonstrates scientific validation and a new approach to the treatment.

MATERIALS AND METHODS

GC-MS Analysis

GC-MS Analysis was performed using the instrument model - 7890 A GC with 5975C with triple axis detector. The equipment has a DB 5MS 30 m \times 0.250 mm Diameter \times 0.25 Micrometer Thickness.

The sample was dissolved in Hexane, taken into vials and injected into GCMS. Analysis was performed by injecting one microliter of the sample with a split ratio of 150: 1. Helium gas (99.9995 %) was used as the carrier gas at a flow rate of 1 mL/min. The analysis was performed in the EI (electron impact) mode with 70eV ionization energy. The injector temperature was maintained at 280 $^{\circ}$ C (constant), and the oven temperature was programmed as follows: 50 $^{\circ}$ C for 10 minutes, then gradually increased to 280 $^{\circ}$ C at 15 minutes.

The compounds were identified after comparing the spectral configurations obtained with the available mass special database (NIST -08 SPECTRAL DATA).

RESULTS AND DISCUSSION

GC-MS Analysis of K.G.101 revealed the presence of 39 compounds in the chromatogram. Out of these, three molecules showed the highest peak. The possible medicinal role of the three significant compounds identified is explained in detail.

1. Diosgenin (Spirost - 5- en- 3-ol)

Diosgenin is a steroidal sapogenin. It occurs abundantly in *Dioscorea alata, Smilax china* and *Trigonella foenum graecum*. The medicinal properties explained for Diosgenin are detailed.

Anti-cancer effect of Diosgenin

Several preclinical studies suggest the effect of Diosgenin as a chemopreventive/therapeutic agent against cancers of several organs. Diosgenin also has antimetastatic effects.

Table 1: Compounds and their properties

SI No	R.T. min	Peak area %	Name of the compound	Properties of the compound
1	50.677	12.839	Diosgenin	Anti-inflammatory, Anti-cancerous, Immunological, Anti-thrombotic ²
2	52.150	11.100	n- Hexadecanoic acid	Anti-inflammatory ³
3	34.197	10.998	Digin	Anti-inflammatory, Antiproliferative ⁴
4	47.683	6.712	Hexadecanoic acid	Anti-inflammatory ³
5	37.424	4.332	cis-13-Octadecenoic acid	Anti-inflammatory ⁶
6	40.011	4.249	Docosanoic anhydride	-
7	50.426	3.561	Octadecane	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁷
8	39.217	3.027	Hexadecanoic acid	Anti-inflammatory ³
9	30.256	2.946	Tetradecanoic acid	Anti-inflammatory ³
10	53.286	2.395	Cholesterol	-
11	43.623	2.386	Hexadecanoic acid,	Anti-inflammatory ³
12	37.847	2.335	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
13	44.707	2.237	Tetradecanoic acid	Anti- inflammatory ³
14	36.421	1.937	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
15	32.915	1.921	4,8,12,16-Octadecatetraen-1-ol, 4,9,13,17- tetramethyl-	-
16	42.183	1.841	Oleic acid	Anti-inflammatory ⁹
17	32.719	1.831	Squalene	Anti-inflammatory, Antioxidant ¹⁰
18	44.520	1.619	Octadecane	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
19	36.563	1.521	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
20	42.608	1.491	Stearic anhydride	Anti-inflammatory. Hepatoprotective ¹¹
21	44.004	1.284	(22R)-6α,11β,21-Trihydroxy-16α,17α- propylmethylenedioxypregna-1,4-diene- 3,20-dione	-
22	33.289	1.265	Hexadecanoic acid,	Anti-inflammatory ³
23	43.347	1.219	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
24	26.260	1.214	Dodecanoic acid	Anti-microbial ¹²
25	35.000	1.046	Tetradecanoic acid	Anti-inflammatory ³
26	28.222	0.942	Ar-turmerone	-
27	22.463	0.939	Dodecanoic acid	Anti-microbial ¹²
28	20.011	0.905	Tumerone	-
29	45.999	0.783	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
30	32.564	0.669	Hexadecanoic acid,	Anti- inflammatory ³
31	39.583	0.635	1H-Indene	-
32	28.522	0.521	Decanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
33	28.990	0.462	Curlone	Antioxidant, Anti-inflammatory, Anti- nociceptive ¹³
34	37.047	0.401	Octadecanoic acid	Anti-bacterial, Anti-cancer, Anti-asthmatic ⁸
35	31.762	0.344	Dodecanoic acid	Anti microbial ¹²
36	29.310	0.264	Methyl tetradecanoate	-

File :D:\GCMSD\2022\DECEMBER\17.12.2022\I_2047R.D Operator : Acquired :17 Dec 2022 12:04 using AcqMethod GCMS_PROFILING2022.M Instrument : GCMS Sample Name: KG Misc Info : Vial Number: 2

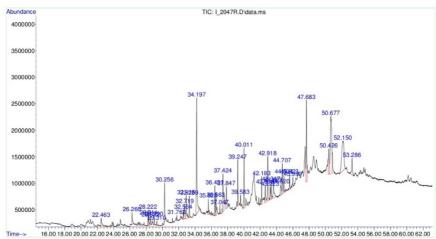


Figure 1: Chromatogram analysis of K.G.101

Anti-inflammatory and immunological activity

Diosgenin has proven anti-inflammatory effects. Studies suggest that Diosgenin can suppress inflammation within the atherosclerotic lesion and modulate the immune response. Due to this property, it can also treat rheumatoid and osteoarthritis.

Effect on diabetes, dyslipidemia and obesity

In vitro and *in vivo* studies show the protective effect of Diosgenin against diabetes, obesity, metabolic syndrome, and dyslipidemia, including hypercholesteremia.

Anti-coagulant and antithrombotic effects

Diosgenin exerts antithrombotic activity and thus prevents blood coagulation.

Diosgenin shows anti- cancer, anti- inflammatory, immunological, anti- coagulant and anti- thrombotic effects.²

2. n- Hexadecanoic acid (Palmitic acid, Pentadecanecarboxylic acid, Cetylic acid)

n- Hexadecanoic acid is the most common saturated fatty acid in animals, plants and microorganisms. It is a major oil component from the fruit of oil palms, milk, cheese, butter and meat. Studies suggest that n- Hexadecanoic acid is an excellent anti-inflammatory agent³.

3. (25R)-5α-Spirostan-2α, 3β-diol (Digin, Gitogenin, Spirostan-2, 3-diol, 2α, 3β, 5α, 25R)

(25R)- 5α -Spirostan- 2α , 3β -diol belongs to the class of organic compounds known as triterpenoids. It is considered to be a sterol lipid molecule.

Studies suggest that (25R)- 5α -Spirostan- 2α , 3β -diol has antiproliferative and anti-inflammatory activities⁴.

Out of 39 compounds identified, the most prominent peak identified is Diosgenin. In rheumatoid arthritis, fibroblast-like synoviocytes are the most common cell type at the pannus cartilage junction and contribute to joint destruction. Diosgenin, the major compound identified, can inhibit the growth of structures that lead to joint destruction⁵. This explains the action of K.G.101avarthi in inflammatory and degenerative arthritis.

The second and third peak identified is n Hexadecanoic acid and Digin, which also possesses anti-inflammatory and antioxidant properties. These actions widely help in the treatment of inflammatory arthritis. The antioxidant property reveals its Rasayana (rejuvenating) property.

Out of the 39 compounds identified, most of the compounds possess anti-inflammatory action. This helps in the treatment of inflammatory conditions. Some of the compounds show antiproliferative and anti-cancerous activity. So the product can be explored in the treatment of cancers also.

CONCLUSION

The GC- MS analysis of Ksheeraguloochi 101 shows some promising molecules with anti-inflammatory, immunological, anti-cancerous and antioxidant activities. This explains its action in inflammatory and degenerative joint disorders. The antioxidant activity explains its rejuvenating action (Rasayana Guna). The product can be explored in the treatment of cancers also.

ACKNOWLEDGEMENT

The authors sincerely thank Dr D. Ramanathan (Managing Director), Dr Vignesh Devaraj (Technical Director), and Smt. Janani Ramanathan (Director), Mr Sandeep VR (Chief General Manager- Unit Head) and Mr Bijeshmon PP (Manager- Quality Control) of Sitaram Ayurveda Pvt. Ltd. for their constant support and encouragement. The authors also thank the 'CARe Keralam' team members who helped execute the GC-MS study.

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Cite this article as:

Meenu M and Sibi Narayanan. Gas chromatographic-Mass spectrometric analysis of K.G. 101 (Ksheeraguloochi 101): An Ayurvedic patent product. Int. J. Res. Ayurveda Pharm. 2023;14(2):43-46

DOI: http://dx.doi.org/10.7897/2277-4343.140238

Source of support: Nil, Conflict of interest: None Declared

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