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GC-MS ANALYSIS ON *PIPPALI RASAYANA*

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ABSTRACT

Pippali Rasayana, an Ayurvedic formulation known for its rejuvenating properties, was subjected to Gas Chromatography-Mass Spectrometry (GC-MS) analysis to identify its chemical constituents. The analysis was carried out using a GC-MS system equipped with a non-polar capillary column, coupled with a mass spectrometer. The sample preparation involved extraction of *Pippali Rasayana* in a suitable solvent followed by evaporation to obtain the extract. GC-MS analysis revealed the presence of diverse chemical compounds including alkaloids, terpenoids, flavonoids, and fatty acids. Major compounds identified included piperine, sesquiterpenes, and volatile oils known for their pharmacological activities such as antioxidant, anti-inflammatory, and immunomodulatory effects. This study provides valuable insights into the chemical composition of *Pippali Rasayana*, supporting its traditional use and highlighting its potential therapeutic benefits in modern medicine.

Keywords: *pippali, pippali rasayana,* GC-MS analysis, bioactive compounds, phytochemicals

INTRODUCTION

Since the 12th Century B.C., India has the holistic, ancient medicinal knowledge known as ayurveda. The eight *angas* or *Chikitsa* that make up the Ayurvedic therapeutic modality each define a distinct field of study. *Rasayana Chikitsa* includes all methods of rejuvenation used to treat ageing **[1]**. Longevity, memory, intelligence, freedom from disorders, youth, excellence of luster, complexion, and voice, oratory, optimum strength of body and sense organs, respectability, and brilliance are all provided by *Rasayana* medications and formulations **[2]**.

Rasayana is the means of attaining excellent qualities of rasa and other dhatu i.e., body cells and tissues. It is not only for promotive but also for curative purpose. Acharya Charaka has mentioned many formulations of *Rasayana* for different diseases in Charaka *Samhitha Chikitsa*sthana *Rasayana* adhyaya. One such is *Pippali Rasayana*. Where, it is mentioned that three-three *Pippali* fruits should be taken in the morning, before and after breakfast. The fine powder of *Pippali* should be given bhavana with *Palasha Kshara jala* (alkaline liquid obtained after processing ash of *Palasha* with water) and then the triturated product is fried in *Ghrita* (ghee) **[3]**. The final product should be taken with honey to get *Rasayana* effect, particularly in order to alleviate cough, wasting, phthisis, dyspnea, hiccup, throat disorders, piles, grahani (IBS), paleness, and intermittent fever, disorders of voice, chronic rhinitis, swelling, tumour.

Many phytochemicals have been identified till date, including alkaloids as its major secondary metabolites (piperine and piperlongumine), essential oil, flavonoids and steroids. These exhibit a wide range of activities including anti-inflammatory, analgesic, anti-oxidant, anti-microbial, anti-cancer, anti-parkinsonian, anti-stress, nootropic, anti-epileptic, anti-hyperglycemic, hepatoprotective, anti-hyperlipidemic, anti-platelet, anti-angiogenic, immunomodulatory, anti-arthritic, anti-ulcer, anti-asthmatic, anthelmintic action, anti-amebic, anti-fungal, mosquito larvicidal and anti-snake venom **[4]**.

Pharmacological actions like hepatoprotective, antifertility, antifilarial, anti-diabetic, antiviral, anthelmintic, anticonvulsant, antifungal, antimicrobial, antiestrogenic, anticancer, antiinflammatory, antioxidant, antiulcer, wound healing, anti-diarrhoeal, anti-implantation, antidopaminergic, antimycobacterial, osteogenic and osteoprotective activity. These medicinal properties may provide potential active principles with higher efficacy **[5]**.

Out of all the *Sneha Dravya* (uncting subsatances) *Ghrita* is par excellence

because of its special property *Samskarasya anuvartana* ie., as a media it aids in incorporating the lipid soluble active principles from the drugs into it when drugs are added to it **[6]**.

DHA along with fat-soluble vitamins, anti-oxidants, and conjugated linoleic acid (CLA) of ghee are responsible for health benefits[7]

METHODOLOGY

The Clarus 680 GC was used in the analysis employed a fused silica column, packed with HP-5MS (5% biphenyl 95% dimethylpolysiloxane, 30 m × 0.25 mm ID × 250µm df) and the components were separated using Helium as carrier gas at a constant flow of 2 ml/min. The injector temperature was set at 250°C during the chromatographic run. The 1µL of extract sample injected into the instrument the oven temperature was as follows: 50 °C (2 min); followed by 150 °C at the rate of 15 °C min−1; and 150 °C, where it was held for 1min and then followed by 240°C at the rate of 25°C min−1; it was held for 12.00 min. The mass detector conditions were: Inlet line temperature 250 °C; ion source temperature 230 °C; and ionization mode electron impact at 70 eV, a scan time 0.2 sec and scan interval of 0.1 sec. The fragments from 40 to 600 Da. The spectrums of the components were compared with the database of spectrum of known components stored in the GC-MS NIST (2014) library.

OBSERVATIONS AND RESULTS

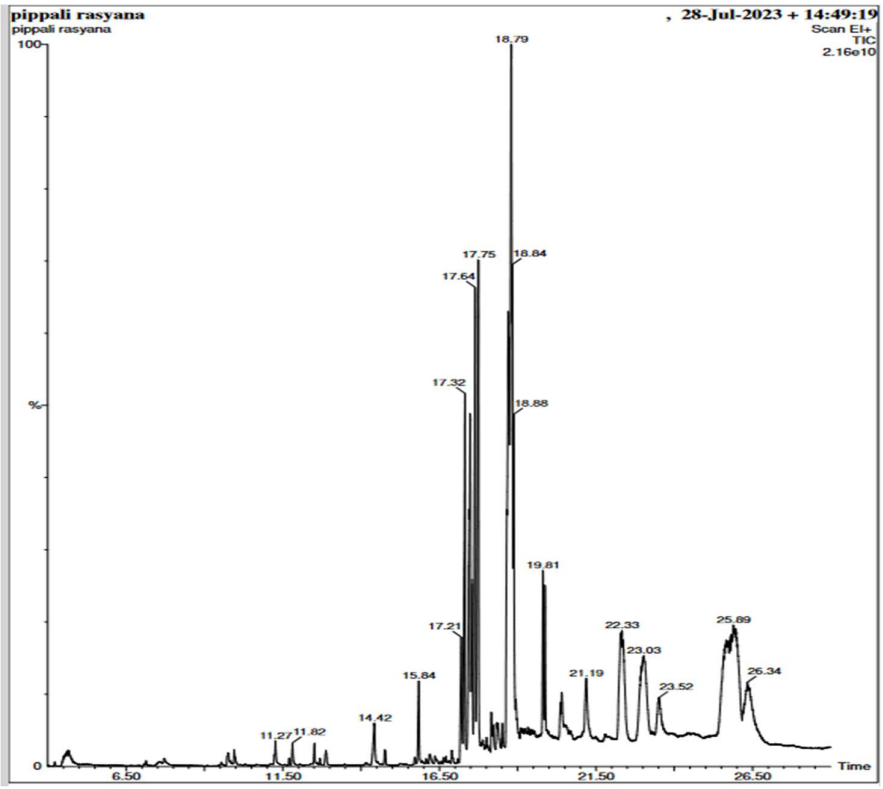

Figure 1: TIC Plot showing GC-MS results of *Pippali Rasayana*

Table 1

Sl. No.	Retention Time	No. Of Components
1.	11.276	Z,Z-6,28-HEPTATRIACTONTADIEN-2-ONE
		CYCLOHEXANEETHANOL
		4-METHYL-.BETA.-METHYLENE
		1R,2R,5S)-5-METHYL-2-(PROP-1-EN-2-YL)CYCLOHEXANOL
		CYCLOHEXYLMETHYLNOL
		4-METHYL-.BETA.-METHYLENE-, TRANS-
2.	11.826	3-METHYL-HEPT A-1,6-DIEN-3-OL
		2-BUTEN-1-OL, 3-METHYL-, FORMATE
		LINALOOL
		LINALOOL
		TERPINEN-4-OL
		TERPINEN-4-OL
		(1R,2R,5S)-5-METHYL-2-(PROP-1-EN-2-YL)CYCLOHEXANOL
		TERPINEN-4-OL
		(1R,2R,5R)-5-METHYL-2-(PROP-1-EN-2-YL)CYCLOHEXANOL
		3-CYCLOHEXEN-1-OL, 4-METHYL-1-(1-METHYLETHYL)-, (R)-
		LINALOOL
		TERPINEN-4-OL
		CYCLOHEXANOL, 5-METHYL-2-(1-METHYLETHENYL)-
		ISOPULEGOL
		ISOPULEGOL
		2(3H)-FURANONE, 4,5-DIHYDRO-3-METHOXY-4-(2-METHYL-1-PROPEN-3-YL)-, TRAN
		3-CYCLOHEXEN-1-OL, 4-METHYL-1-(1-METHYLETHYL)-, (R)-
		CYCLOHEXANOL, 5-METHYL-2-(1-METHYLETHENYL)-
		ISOPULEGOL
		BICYCLO[2.2.1]HEPT AN-2-OL, 2,3,3-TRIMETHYL-
3.	12.522	DODECANOIC ACID
		N-DECANOIC ACID
		DODECANOIC ACID
		DODECANOIC ACID
4.	12.892	HENTRIACONTANE
		TRIACONTANE
		DOTRIACONTANE
		DOTRIACONTANE, 1-IODO-
5.	14.422	TETRADECANOIC ACID
		TETRADECANOIC ACID
		DODECANOIC ACID
		TETRADECANOIC ACID
6.	15.848	TETRADECANOIC ACID
		TETRADECANOIC ACID
		N-DECANOIC ACID
		TETRADECANOIC ACID
7.	17.324	2,4-DECADIENAMIDE, N-ISOBUTYL-, (E,E)-
		(2E,4E)-N-ISOBUTYLUNDECA-2,4-DIENAMIDE
		1-SILACYCLOHEXA-2,5-DIENE
		10,12-OCTADECADIENOIC ACID, 9-OXO-
8.	17.484	N-HEXADECANOIC ACID
		DODECANOIC ACID
		TETRADECANOIC ACID
		N-DECANOIC ACID
9.	17.649	2,4-DECADIENAMIDE, N-ISOBUTYL-, (E,E)-
		(2E,4E)-N-ISOBUTYLUNDECA-2,4-DIENAMIDE
		4H-1,3-BENZODIOXIN-4-ONE, 5-BUTYL-2-(1,1-DIMETHYLETHYL) HEXAHYDRO-4A-M
		NAPHTHALENE, DECAHYDRO-1,1-DIMETHYL-
10.	17.754	N-HEXADECANOIC ACID
		TETRADECANOIC ACID

		L-(+)-ASCORBIC ACID 2,6-DIHEXADECANOATE
		DODECANOIC ACID
11.	18.799	CIS-13,16-DOCASADIENOIC ACID
		17-OCT ADECYNOIC ACID
		(R)-(-)-14-METHYL-8-HEXADECYN-1-OL
		EICOSEN-1-OL, CIS-9-
		11,14-OCTADECADIENOIC ACID, METHYL ESTER
		2-CHLOROETHYL LINOLEA TE
		9,12-HEXADECADIENOIC ACID, METHYL ESTER
		1,7-HEXADECADIENE
		Z,E-3,13-OCT ADECADIEN-1-OL
		13-DOCOSEN-1-OL, (Z)-
		9-OCT ADECEN-1-OL, (Z)-
		Z,Z-3,13-OCT ADECADIEN-1-OL
		9,17-OCT ADECADIENAL, (Z)-

DISCUSSION

Components which support therapeutic activity and exhibits *Rasayana* effect has been discussed below

The chemical compound 3-methyl-hepta-1,6-dien-3-ol is commonly known as geraniol.

Medicinal Properties: Geraniol has shown potential health benefits, such as anti-inflammatory, antioxidant, and anticancer properties. Studies suggest that geraniol may have therapeutic effects against certain types of cancer cells and could be used in the development of anticancer drugs.

Preservative: Due to its antimicrobial properties, geraniol can act as a natural preservative in various products, extending their shelf life and preventing the growth of bacteria and fungi.

The chemical compound mentioned, 2-buten-1-ol, 3-methyl-, formate, is also known as geranyl formate.

Antimicrobial Properties: Linalool has shown some antimicrobial properties, making it useful in controlling the growth of certain bacteria and fungi. It is sometimes used in natural cleaning products and as a preservative in cosmetics and personal care items.

Analgesic and Anti-inflammatory Effects: Linalool may have mild analgesic (pain-relieving) and anti-inflammatory properties. Some studies suggest it could potentially help with pain management and reducing inflammation.

Terpinen-4-ol is a terpene alcohol that is naturally present in various essential oils, with one of the most well-known sources being tea tree oil.

Antimicrobial Properties: Terpinen-4-ol is renowned for its powerful antimicrobial activity. It has been found to be particularly effective against a wide range of bacteria, fungi, and other microorganisms.

Anti-inflammatory Effects: Terpinen-4-ol has demonstrated anti-inflammatory properties, which can help reduce redness, swelling, and discomfort

Antioxidant Activity: As an antioxidant, terpinen-4-ol helps neutralize free radicals in the body, which can contribute to cellular damage and aging. By combating oxidative stress, it may contribute to overall health and well-being.

The chemical compound (1R,2R,5S)-5-methyl-2-(prop-1-en-2-yl) cyclohexanol is commonly known as α-terpineol.

Antimicrobial Properties: α-Terpineol exhibits antimicrobial activity and can act as a natural disinfectant.

Analgesic and Anti-inflammatory Effects: Some studies suggest that α-terpineol may have analgesic (pain-relieving) and anti-inflammatory effects, potentially making it useful in managing pain and reducing inflammation.

The chemical compound cyclohexanol, 5-methyl-2-(1-methylethenyl)- is known as p-menthan-3-ol or more commonly as menthol.

Pain Relief: Menthol's cooling sensation also acts as a mild analgesic, helping to reduce pain and discomfort.

Respiratory Relief: Inhaled menthol can help relieve congestion and ease breathing, making it a common ingredient in cough drops, lozenges, and inhalers for respiratory relief.

Isopulegol is a terpene alcohol that can be found in the essential oils of certain plants, including mint and eucalyptus.

Respiratory Relief: Like menthol, isopulegol can help relieve congestion and facilitate easier breathing when inhaled. It is used in cough drops, lozenges, and inhalers for respiratory relief.

The chemical compound 3-cyclohexen-1-ol, 4-methyl-1-(1-methylethyl), (R)- is known as (R)-carvone. It is a chiral molecule, meaning it exists in two mirror-image forms known as enantiomers, with (R)-carvone being one of them. (R)-carvone is commonly found in essential oils of certain plants, most notably in spearmint and dill.

Medicinal Uses: In traditional medicine, (R)-carvone has been used for its potential medicinal properties. It is believed to have digestive and carminative effects, aiding in digestion and reducing gas and bloating.

Cyclohexanol, 5-methyl-2-(1-methylethenyl)- is also known as p-menthan-5-ol or simply as p-menthol

Respiratory Relief: Inhaled p-menthol can help relieve congestion and facilitate easier breathing. It is found in cough drops, lozenges, and inhalers for respiratory relief and to alleviate symptoms of common colds and respiratory conditions.

Dodecanoic acid, also known as lauric acid,

Antimicrobial Properties: Lauric acid's potential antimicrobial properties make it useful in natural health and wellness products, as well as in certain medical applications

10,12-octadecadienoic acid, 9-oxo-, also known as 9-oxo-10,12-octadecadienoic acid, is a type of fatty acid with potential therapeutic applications, particularly in the field of nutrition and health. This compound is often classified as a metabolite derived from linoleic acid, an essential fatty acid.

Anti-Inflammatory Properties: Some research suggests that certain oxylipins, which include compounds like 10,12-octadecadienoic acid, 9-oxo-, might have anti-inflammatory effects. Inflammation is associated with various chronic diseases, so compounds with anti-inflammatory properties could potentially be used to manage or prevent such conditions

N-hexadecanoic acid is a straight-chain saturated fatty acid, commonly known as palmitic acid.

Inflammation: High levels of saturated fats, including palmitic acid, in the diet have been linked to inflammation. Chronic inflammation is associated with various chronic diseases, so dietary balance is important.

L-(+)-ascorbic acid 2,6-dihexadecanoate is a derivative of ascorbic acid, which is also known as vitamin C.

Antioxidant Potential: Vitamin C is a well-known antioxidant, and its derivatives may also possess antioxidant properties. Antioxidants help protect cells from damage caused by free radicals, which are molecules that contribute to aging and various diseases.

CONCLUSION

In the present work prepared pippali rasayana was subjected for GC-MS analysis and the obtained results were tabulated and discussed with respect to its properties. Many constituents were found with 11 different retention times. It helps to predict the formula and structure of bio-molecules which can be used as drugs. The GC-MS results found in *pippali rasayana* can be considered as the preliminary standards for further study

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