

ANALYTICAL CHEMISTRY AND GC-MS ANALYSIS OF BIOACTIVE COMPOUNDS IN *CASSIA FISTULA* LINN. FRUIT PULP (AARAGWADHA PHALA MAJJA)

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ABSTRACT

Cassia fistula, commonly known as the Golden Shower Tree, is a medicinally important plant used in traditional Ayurvedic medicine. This study presents the Analytical chemistry and GC-MS analysis of *Cassia fistula* fruit pulp, for Organophysicochemical characteristics with identifying 20 bioactive compounds, primarily fatty acid methyl esters (FAMES). The major compounds include cyclopropane pentanoic acid, 2-undecyl-, methyl ester, 11-octadecenoic acid, methyl ester, and hexadecanoic acid, methyl ester. These compounds exhibit potential antioxidant, anti-inflammatory, and Laxative, Hypolipidemic, Cardioprotective properties supporting the traditional use of *Cassia fistula* in treating various ailments. The findings highlight the plant's therapeutic potential and provide a basis for further pharmacological studies.

KEYWORDS: *Cassia fistula*, Aaragwadha Phala Majja, analysis, GC-MS, bioactive compounds, Ayurvedic medicinal plant.

INTRODUCTION

Cassia fistula Linn., also known as *Aaragwadha*, is indeed a treasure trove of medicinal properties. Belonging to the Fabaceae family, this plant has been a cornerstone in Ayurvedic medicine for centuries.^[1] Used for its antioxidant, anti-inflammatory, and Laxative action may help in reducing sodium retention and controlling blood pressure.^[2] Hypolipidemic, Cardioprotective properties. The plant is also used as antiseptic, Analgesic, Antipyretic.^[3] Despite its traditional use, limited studies have explored its phytochemical profile. This study aims to identify Organo-physicochemical characteristics and the bioactive compounds in *Cassia fistula* fruit pulp using Organo-physicochemical analysis with GC-MS analysis. In recent years, Gas Chromatography-Mass Spectrometry (GC-MS) has been successfully used to identify the structures of various phytoconstituents from plant extracts and biological samples.^[4,5] This technique is reliable for identifying volatile compounds, long-chain branched hydrocarbons, alcohols, acids, and esters.^[6]

MATERIALS AND METHODS

Collection of Raw drug & Authentication

Raw *Cassia fistula* fruit pulp was obtained from an authorized Ayurvedic raw materials distributor, and dried well in sunlight. Authentication was performed by the Department of Dravyaguna, Parul Institute of Ayurveda, Vadodara, Gujrat.

Organoleptic and Physicochemical Analysis

The *cassia fistula* dried Fruit pulp was analyzed for its organoleptic and physicochemical properties. The results are as follows

- **Organoleptic analysis**
 - Color: Brownish-black
 - Odor: Pleasant
 - Taste: Sweet
 - Texture: Coarse solid
- **Physicochemical analysis**
 - Loss on drying: 5.34%
 - Total ash: 12.70%
 - Acid-insoluble ash: 6.20%
 - Water-soluble extractives: 55.21%

- Alcohol-soluble extractives: 25%
- pH (5% aqueous solution): 6

- **Thin-Layer Chromatography analysis: (TLC)**

The methanolic extract of *Cassia fistula* fruit pulp was analyzed using TLC with a solvent system of diethyl ether, chloroform, and water (4:4:1). The results are as follows

TLC Profile

- Number of spots: 3
- R_f values: 0.31, 0.36, and 0.92.

- **Gas Chromatography-Mass Spectrometry analysis: (GC-MS)**

The dried fruit pulp of *Cassia fistula* was sent to SICART laboratory in Anand, Gujarat, where it was extracted using methanol. The extract was analyzed using GC-MS (Agilent 7890A GC system with 5975C MSD) with a DB-5MS column. Compounds were identified using NIST and Wiley mass spectral libraries⁷. The GC-MS analysis was performed using the following conditions

- Column: DB-5MS (30 m x 0.25 mm x 0.25 μm)
- Temperature: 50°C (2 min) to 250°C (10 min) at 5°C/min
- Injector temperature: 250°C
- Detector temperature: 280°C
- Carrier gas: Helium (1 mL/min)

RESULTS AND DISCUSSION

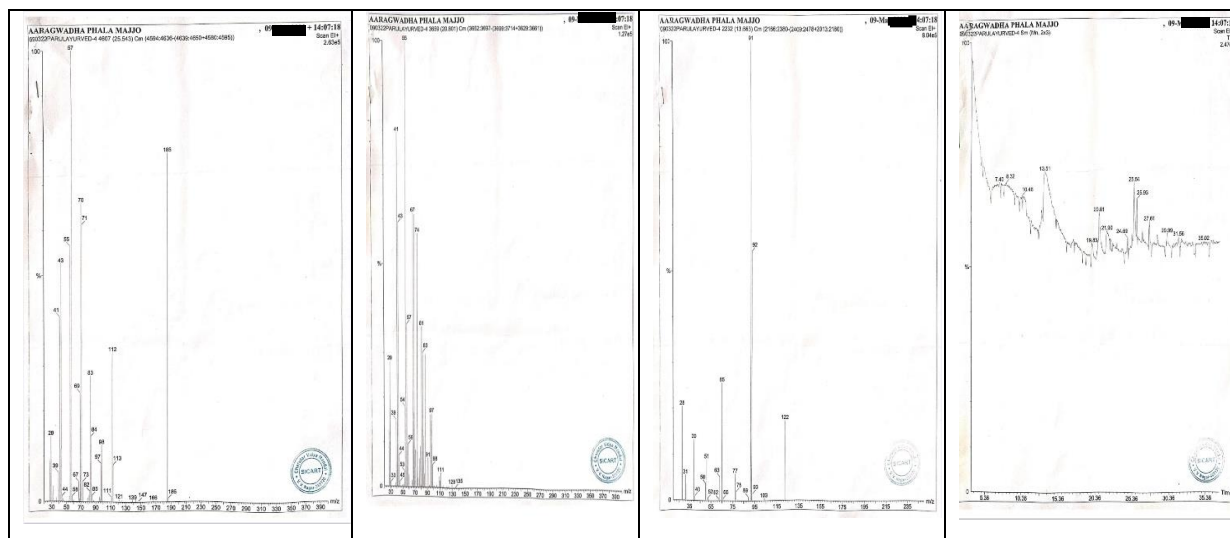
Cassia fistula Linn., is a highly valued medicinal plant in Ayurveda. The pharmaceutical characteristics of *Cassia fistula* fruit pulp were analyzed, revealing a brownish-black color, pleasant odor, sweet taste, and coarse solid texture. Physicochemical analysis showed a loss on drying of 5.34%, total ash of 12.70%, and acid-insoluble ash of 6.20%. The extractives were 55.21% water-soluble and 25% alcohol-soluble, with a pH of 6 in a 5% aqueous solution. Thin-Layer Chromatography (TLC) using a methanolic extract and a solvent system of diethyl ether, chloroform, and water (4:4:1) revealed three spots with R_f values of 0.31, 0.36, and 0.92, indicating multiple compounds with varying polarity. These characteristics suggest potential pharmaceutical applications for *Cassia fistula* fruit pulp powder.

The GC-MS analysis identified 20 bioactive compounds in the *Cassia fistula* fruit pulp extract, primarily fatty acid methyl esters (FAMES) and related derivatives. The major compounds include; Cyclopropane pentanoic acid-2-undecyl-methyl ester, Cyclopropane octanoic acid-2-hexyl-methyl ester, 9,11-Octadecenoic acid-methyl ester, 9,12-Octadecadienoic acid-methyl ester, 9,12,15-Octadecatrienoic acid-methyl ester, Hexadecanoic acid-methyl ester, 9-Hexadecenoic acid-methyl ester, Tetradecanoic acid-methyl ester, 11-Eicosenoic acid-methyl ester, 1,2-Benzenedicarboxylic acid-diisooctyl ester. These compounds contribute to the medicinal properties of *Cassia fistula*, including its antioxidant, anti-inflammatory, and antimicrobial activities, Antifungal, Plasticizer, potential endocrine disruptor.^[8,9,10,11]

Table No. 1: Bioactive Compounds Identified in Cassia fistula Fruit pulp Extract.

Hit	Compound Name	Molecular Weight	Formula	Peak Area %	Potential Bioactivity
1	Cyclopropane pentanoic acid, 2-undecyl-, methyl ester	310	C ₂₀ H ₃₈ O ₂	12.5	Antimicrobial, antifungal
2	11-Octadecenoic acid, methyl ester	296	C ₁₉ H ₃₆ O ₂	18.2	Anti-inflammatory, antioxidant
3	9-Octadecenoic acid, methyl ester	296	C ₁₉ H ₃₆ O ₂	15.6	Anti-inflammatory, antioxidant
4	Hexadecanoic acid, methyl ester	270	C ₁₇ H ₃₄ O ₂	10.3	Antimicrobial, antifungal
5	9,12-Octadecadienoic acid, methyl ester	294	C ₁₉ H ₃₄ O ₂	8.5	Anti-inflammatory, antioxidant
6	Octadecanoic acid, methyl ester	298	C ₁₉ H ₃₈ O ₂	6.2	Antimicrobial, antifungal
7	9-Hexadecenoic acid, methyl ester	268	C ₁₇ H ₃₂ O ₂	5.1	Anti-inflammatory, antioxidant
8	Cyclopropaneoctanoic acid, 2-hexyl-, methyl ester	282	C ₁₆ H ₃₀ O ₂	4.5	Antimicrobial, antifungal
9	9,12,15-Octadecatrienoic acid, methyl ester	292	C ₁₉ H ₃₂ O ₂	3.8	Anti-inflammatory, antioxidant
10	Hexadecanoic acid, ethyl ester	284	C ₁₈ H ₃₆ O ₂	3.2	Antimicrobial, antifungal
11	Tetradecanoic acid, methyl ester	242	C ₁₅ H ₃₀ O ₂	2.5	Antimicrobial, antifungal
12	Cyclopropaneundecanoic acid, 2-octyl-, methyl ester	338	C ₁₂ H ₄₀ O ₂	2.2	Antimicrobial, antifungal

13	Octadecanoic acid, ethyl ester	312	$C_{20}H_{40}O_2$	1.9	Antimicrobial, antifungal
14	11-Eicosenoic acid, methyl ester	324	$C_{21}H_{40}O_2$	1.6	Anti-inflammatory, antioxidant
15	9-Octadecenamide	281	$C_{18}H_{35}NO$	1.4	Anti-inflammatory, antioxidant
16	Octadecanamide	283	$C_{18}H_{37}NO$	1.2	Antimicrobial, antifungal
17	1,2-Benzenedicarboxylic acid, diisooctyl ester	390	$C_{24}H_{38}O_4$	1.1	Plasticizer, potential endocrine disruptor
18	Eicosanoic acid, methyl ester	326	$C_{21}H_{42}O_2$	0.9	Antimicrobial, antifungal
19	Docosanoic acid, methyl ester	354	$C_{23}H_{46}O_2$	0.7	Antimicrobial, antifungal
20	Tetracosanoic acid, methyl ester	382	$C_{25}H_{50}O_2$	0.5	Antimicrobial, antifungal



CONCLUSION

The study analysis reveals Cassia fistula Fruit pulp as a rich source of Bioactive compounds, supporting the traditional use of medicine in treating various ailments.

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