

Assessment of Anti-diabetic Activity of *Cassia Siamea* Lam Leaves Ethanol Extract

B. Gawade¹, M. Farooqui^{2*}

¹Dept. of Chemistry, Aandrao Dhonde Alias Babaji Mahavidyalaya, Kada (M.S.), India

²Dept. of Chemistry, Maulana Azad College of Arts, Science and Commerce, Aurangabad (M.S.), India

Available online at: www.isroset.org

Received: 04/Apr/2020, Accepted: 23/April/2020, Online: 31/April/2020

Abstract- The species of the genus *Cassia* has been reported for various pharmacological activities and their rich contents of phytochemicals use to treat health related disorders. The different parts of *Cassia siamea* Lam plant has been traditionally used as a medicine to treat diabetes mellitus related disorders. The assessment study aimed to investigate anti-diabetic activity of locally used medicinal plant *Cassia siamea* Lam leaves ethanol extract by porcine pancreatic α -amylase enzyme inhibition in vitro assay. A potent enzyme inhibitory activity was showed by the leaves of *Cassia siamea* Lam ethanol extract. The enzyme assay-guided leaves ethanol extract led to the inhibition activity of enzyme and has potent α -amylase inhibitor content of bioactive phytochemicals. This assessment study that describes the potency of *Cassia siamea* Lam medicinal plant leaves has been used an alternative medicine to treat disorders of diabetes mellitus.

Keywords- *Cassia siamea* Lam, diabetes mellitus, α -amylase, phytochemicals, α -amylase inhibitor.

I. INTRODUCTION

Diabetes mellitus disease is a metabolic disorder of irregular secretory action of hyperglycaemia regulating organ in the human beings and becomes a serious threat to mankind health. There are many therapeutic treatment exists for the regulate diabetes related disorders. In the treatment of diabetes mellitus especially in the developing countries medicinal plants has been plays important role due to their cost effectiveness. Plant based drugs are generally considered safe, easily available and are much effective [1]. Enzyme activity inhibitors also have been plays potential role for to control and treatment many diseases.

Artificially prepared enzyme inhibitory agents can produce serious side effects and are not suitable for use during a pregnancy [2]. Therefore, more effective and safer enzyme inhibitory agents has been searching of new area to active research, and after the recommendations made by WHO on diabetes mellitus [3] research on hypoglycaemic agents from medicinal plants has become an important aspect of study.

A medicinal plant *Cassia siamea* Lam has been shown different pharmacological activities like antimicrobial, antimalarial [4], anti-diabetic, anticancer, hypotensive, antipyretic, laxative, analgesic, diuretic, anti-inflammatory, sedatives antioxidant, anxiolytic and antidepressant. The

main constituents were reported in this plant as chromone, chromone alkaloids, anthraquinones, bianthraquinones, flavonoids and phenolics compounds are barakol was identified as the major chemical constituents of leaves and flowers [5].

The reviews of literature found that the traditionally used medicinal plants were possessing outstanding anti-diabetic property, which could be possibly investigated further for the presence of α -amylase inhibitor contents [6]. The literature searches were provided updated information of various medicinal plants use in the treatment of Diabetes mellitus disease related disorders and afforded no more information on the α -amylase inhibition activity of the *Cassia siamea* Lam leaves ethanol extract investigated [7].

Therefore, we design our study to assess potency of *Cassia siamea* Lam plant leaves as a source of α -amylase inhibitor for assessing their anti-diabetic activity and bioactive contents.

II. MEDICINAL PLANTS IN USE

The different species of the genus *Cassia* has also been reported various pharmacological activities and rich contents of novel phytochemical constituents useful for the treatment of many health related disorders [8].

The root extract of *Cassia siamea* Lam showed pancreatic lipase enzyme inhibition activity and its bioassay guided

fractionation provided important information of cassiamin-A and a bianthraquinone, as most active compound of pancreatic lipase inhibition [9]. This inhibitory activity of plant extensively used for the evaluation of potency of

natural products as antiobesity agents [10].

Due to previously reported various pharmacological potencies of different parts of *Cassia siamea* Lam medicinal plant have been growing their therapeutic demands.

III. MATERIAL AND METHODS

Collection of plant material: The leaves of *Cassia siamea* Lam plant were collected from local area and identified with the help of our college botanists.

Extraction of plant leaves: The leaves of *Cassia siamea* Lam plant were dried under shade and then powdered. 10 g of powdered leaves material was dissolved in 100 mL of ethanol and kept on a magnetic stirrer for 1 hrs. Thereafter, it was extracted sequentially using a soxhlet apparatus in ethanol solvent. The extract was collected and the solvent was evaporated out to dryness. The obtained material was stored at 4°C in airtight bottles for further assessment study.

In vitro α -amylase Inhibitory Assay: α -amylase inhibition activity assessment study was adopted by using a modified 3,5-dinitrosalicylic acid (DNS) *in vitro* assay method to quantify reducing sugar maltose liberated under the assay conditions. The enzyme inhibitory activity was expressed as a decrease in units of maltose liberated [11,12,13].

Phytochemical Analysis: The extract was preliminary qualitatively analysed for the active phyto-constituents such as phenols, protein and amino acids, glycoside, steroids, carbohydrates, tannins, flavonoids, alkaloids, saponins, triterpenoids etc. according to the standard protocol [14,15].

GC-MS analysis: GC-MS analysis was carried out on Shimadzu GC-MS model number QP 2010S. The phyto-constituents were identified by comparison of mass spectra with the help of NIST-11 and WILEY-8 inbuilt libraries.

Statistical analysis: The experiments were performed out in triplicate and the results were expressed in mean \pm SD.

IV. RESULTS AND DISCUSSION

The results of assessment study showed that the *Cassia siamea* Lam plant leaves ethanol extract exhibited dose dependent α -amylase inhibitory activity by *in vitro* assay using potato starch as substrate. The detected biochemical compounds in the leaves of *Cassia siamea* Lam find themselves in the traditional preparation with various pharmacological activities.

In vitro α -amylase inhibitory assay: The anti-diabetic activity was assessed through the inhibition of α -amylase enzyme that made the digestion of starch and so reduced the glucose absorption. Acarbose is used as a standard drug at a concentration range of 20-100 μ g/mL (Table 1) and *Cassia siamea* Lam leaves ethanol extract (20-100 μ g/mL) exhibited α -amylase inhibitory activity in a dose dependent manner (Table 2).

Table 1. α -Amylase inhibitory Activity of Acarbose (Standard Drug)

$\text{Absorbance of the sample at 540nm} / \text{Absorbance of Control} = 0.513$

Sr. No.	Concentration In (μ g/mL)	Absorbance	% Inhibition	IC ₅₀ Value (μ g/mL)
1	20	0.271	47.17	28.35
2	40	0.234	54.38	
3	60	0.208	59.45	
4	80	0.183	64.32	
5	100	0.160	68.81	

Table 2. α -Amylase inhibitory activity of *Cassia siamea* Lam leaves extract

$\text{Absorbance of the sample at 540nm} / \text{Absorbance of Control} = 0.513$

Sr. No.	Concentration In (μ g/ml)	Absorbance	% Inhibition	IC ₅₀ Value (μ g/ml)
1	20	0.274	46.58	40.89
2	40	0.257	49.90	
3	60	0.236	53.99	
4	80	0.213	58.48	
5	100	0.200	61.01	

The experimental data of leaves ethanol extract and acarbose as a standard were represents inhibition activity at higher concentration tested. Leaves of *Cassia siamea* Lam. exhibited higher activity i.e. it inhibits α -amylase enzyme activity by about IC₅₀ values of ethanol extract 40.89 μ g/mL and acarbose as a standard 28.35 μ g/mL at concentrations ranging from 20 μ g/mL to 100 μ g/mL as shown in fig. 1.

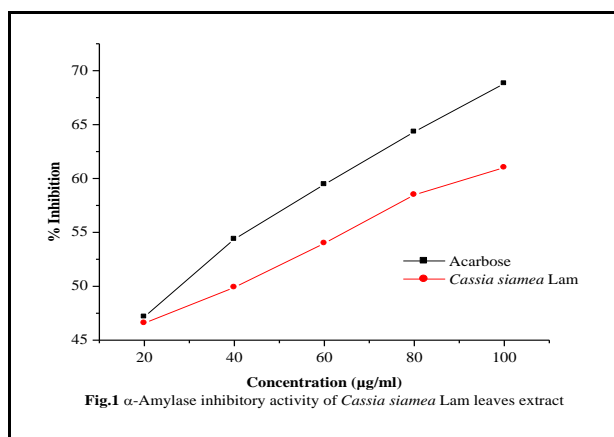


Fig.1

Phytochemical Analysis of Extracts: The ethanol extracts of *Cassia siamea* Lam leaves were investigated qualitatively for the active phytochemical components by using standard protocols. The analysis result of the ethanol

extract were showed the presence of alkaloids, protein and amino acids, saponin, flavonoids, steroids, triterpenoids and phenolic [16] as shown in table 3.

Table 3. Phytochemical Tests Performed for *Cassia siamea* Lam leaves extract

Phytochemicals	Result
1. Alkaloid	+
2. Carbohydrate	-
3. Protein and amino acids	+
4. Glycoside	-
5. Tannin	+
6. Saponin	+
7. Flavonoids	+
8. Steroids	+
9. Triterpenoids	+
10. Phenolic compounds	+

(+) for present and (-) for absent

GC-MS analysis of ethanol extract: The GC-MS analysis of *Cassia siamea* Lam leaves ethanol extract were detected 15 bioactive phytochemicals, out of which mome inositol and squalene abundance are predominant.

The phytochemicals were detected in analysis contributes to α -amylase inhibitory and other pharmacological activities as shown in chromatogram (TIC) fig. 2.

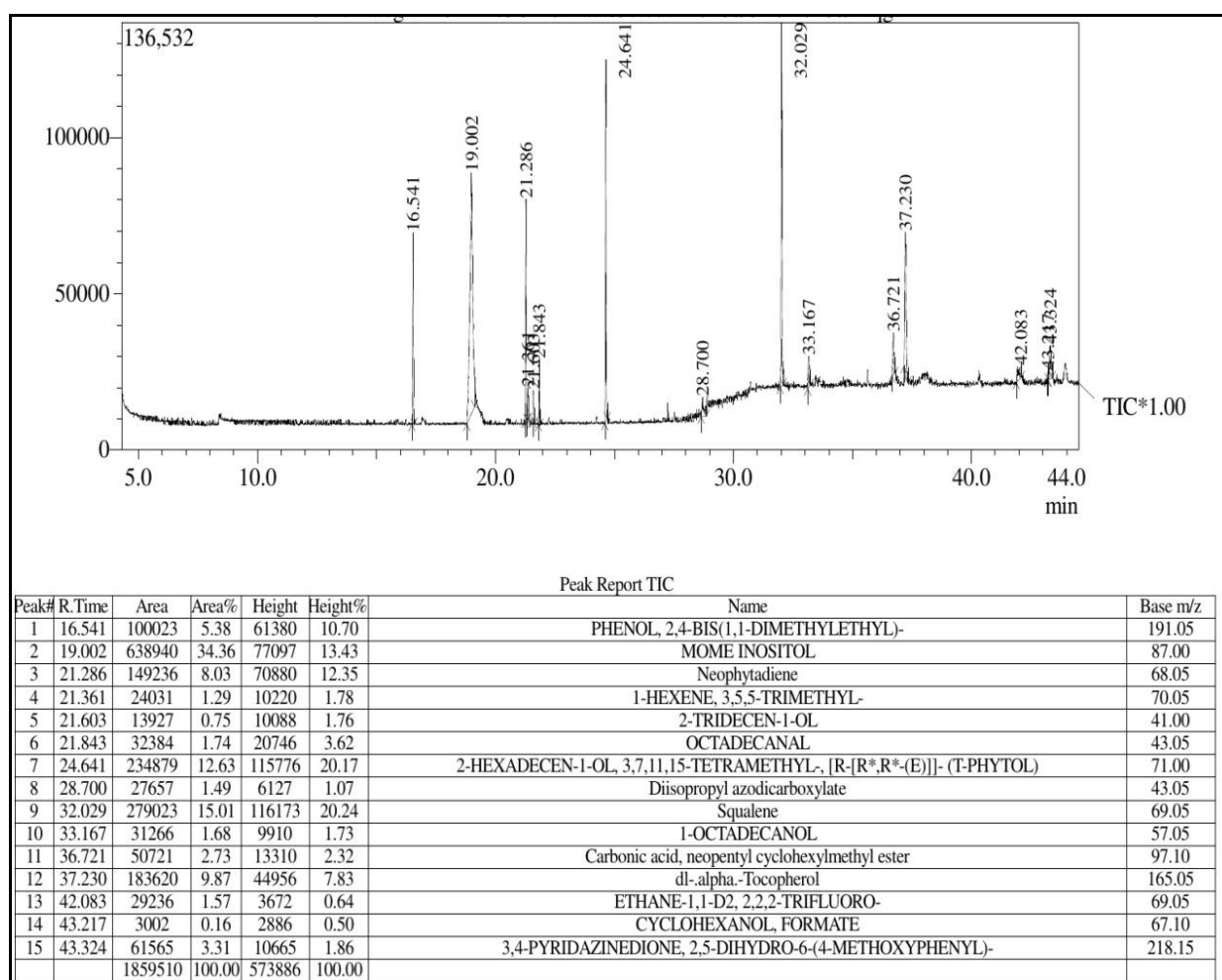


Fig. 2 GC-MS chromatogram (TIC) and chemical constituents detected in *Cassia siamea* Lam leaves extract

The naturally occurring health products contains phytoconstituents from plant origin were clearly indicates as a promising avenue for the prevention and treatments of chronic disorders. The leaves extracts of *Cassia siamea* Lam. were tested for anti-diabetic activity in alloxan induced diabetes of diabetic rats; different extract doses produced significantly decreased the plasma blood glucose level as well as improving lipid metabolism and body weight in rats with induced diabetes problems [17,18]. In silico molecular docking studies has shown good docking score, emodin and chrysophanol are present in *Cassia siamea* Lam leaves to be good inhibitors of angiotensin II receptor type 2 and possess good antidiabetic activity [19].

The leaves of *Cassia siamea* are rich source of minerals [20] and contains different phytochemical compounds like cassiamin, siameadin, lupeone, lupeol, chrysophanol, cassiamin A, chrysophanol-antrone, barakol, rhein, cassia chromone (5-acetyl-7-hydroxy-2-methylchromone), β -sitosterol, p-coumaric acid, apigenin-7-o-galactoside, cassia chromone and cassiadine [21,22,23,24]; whereas cassiarin A [25], bischromone, chrobisiamone-A [26], were isolated from the *Cassia siamea* Lam leaves and displayed promising antiplasmodial activity.

The GC-MS analysis of leaves extract was detected 15 bioactive phytochemicals, out of which mome inositol and squalene abundance is predominant. Preliminary qualitative phytochemical analysis of leaves ethanol extract also confirmed the presence of secondary metabolites like alkaloids, carbohydrates, proteins, flavonoids, saponins, tannins, glycosides, triterpenes and steroids [27]. All the chemical groups identified a wide range of biochemical compounds in the leaves of *Cassia siamea* Lam find themselves in the traditional preparation with various pharmaceutical importances [28,29,30]. The different parts of *Cassia siamea* Lam plant also exhibited significant pharmacological activities and uses [31].

Similarly, our assessment study suggests that the ethanol extract of leaves showed significant anti-diabetic activity at higher concentration tested due to the rich contents of bioactive phytoconstituents. The dose dependent inhibition of α -amylase enzyme reported by extract than standard used in vitro assay. Therefore, *Cassia siamea* Lam herbal local medicinal plant leaves have been used as potent α -amylase inhibitor to treat and prevent metabolic chronic disorders.

V. CONCLUSION

The assessment study of *Cassia siamea* Lam leaves ethanol extract has been significantly reported potent α -amylase inhibitory activity. The overall activity depends on contribution of phytochemical contents were present in the leaves extract. It could be a main source of natural anti-diabetic agent and have more significant role as therapeutic use for prevention and management diabetes related disorders. Therefore, it was concluded that ethanol extract of *Cassia siamea* Lam leaves showed potent anti-diabetic activity due to rich contents of bioactive components. More

investigations are proposed to validate these claims by isolate and identifying bioactive components present in the leaves, which give more therapeutic benefits for to treat diabetes mellitus disorders.

ACKNOWLEDGMENT

We wish to gratefully acknowledge the enabling facilities provided by the Director, KFRI, Kerala for GC-MS facility, Research Institute, Maulana Azad College, Aurangabad, Head of Department and Principal of our college.

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