# PHENOLIC, FLAVONOID CONTENT AND ANTHELMINTIC ACTIVITY DETERMINATION OF Cassia fistula L. LEAVES HYDRO ALCOHOLIC EXTRACT

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## **ABSTRACT**

**INTRODUCTION:** Most plant species in the genus *Cassia* L. are used extensively in traditional medicine by herbal medicine practitioners. Because of its many yellow blooms, *Cassia fistula* is often grown as an ornamental plant in the tropics.

AIM: To determine the hydroalcoholic extract's phenolic, flavonoid, and anthelmintic activities.

**MATERIALS & METHODS:** *Pheretima Posthuma* test worms were used to assess the anthelmintic efficacy of three different doses (25, 50, and 100 mg/ml) of hydroalcoholic extract of Cassia fistula L.

**RESULTS:** Results were examined in terms of the lengths of time it took for worms to die and become paralyzed, and activity levels are compared with that of albendazole, the standard drug. It was observed that there was a dose-dependent reduction in the paralyzing and death times.

**CONCLUSION:** There was a noticeable anthelmintic effect from the hydroalcoholic extract. The research findings suggest that Cassia fistula L. has potential application as an anthelmintic agent.

Keywords: Cassia fistula, flavonoid content, anthelmintic activity, albendazole.

## INTRODUCTION

Plants are vital to human survival. Nature's pharmacy has all the answers to human ailments.<sup>1</sup> India possesses a plethora of well-researched and useful data regarding conventional herbal medicine.<sup>2</sup> Herbal medicines are used by traditional medical practitioners all over the world to treat a variety of illnesses.<sup>3</sup> The manufacture of synthetic medications has advanced significantly in recent years, but these medications are found to have one or more side effects; in contrast, plants continue to hold a distinct place in medicine because they do not have any negative effects.<sup>4</sup>

Because of its beautiful yellow petals, Cassia fistula L. (Leguminosae) is widely grown as an attractive plant in tropical climates and is also utilized in many traditional medical systems, such as Chinese and Ayurvedic medicine. Reaching a height of 6 to 9 meters, it has a straight trunk, pale grey bark that is smooth while young, and rough, brown bark when it ages. The branches are thin and dispersed. The tree, which goes by the name Yellow Shower, is a medium-sized deciduous tree with elongated, rod-shaped fruits that are packed with pulp and vivid yellow blooms. Cassia fistula grows best in full sun and well-drained soils, although it also does well in dry microclimates. In addition to being somewhat tolerant of salt and dehydration, the plant can sustain little damage from frost if it continues to freeze.

Cassia fistula is used in the treatment of a variety of illnesses, including fever, heart-related problems, ringworm, leprosy, and asthma symptoms, among others. Extracts from C. fistula are used as a laxative and to cure constipation; the leaves are used to treat edema, pain, and skin irritation caused by swelling, while the root is used to treat colds and flus. Furthermore, fruit and stem bark preparations are utilized to remove toxins from the blood.<sup>5</sup>

Globally, parasitic worm infections affect around 2 billion individuals. Anthelmintics are drugs that either kill or immobilize parasitic worms, enabling the body to expel them. There are other terrible names for them: vermifuges and vermicides. Unfortunately, gastrointestinal helminths are not treatable by the current helminth drugs. These days, using certain synthetic anthelmintics can be hazardous to people. It is safe to utilize anthelmintic plants, such as *Centratherum anthelminticum, Ocimum sanctum, and Carica papaya*.



Figure 1: Cassia fistula L. leaves



Figure 2: Cassia fistula fruits

## MATERIALS AND METHODS

#### Collection and authentication

Leaves of Cassia fistula L. were gathered from the surroundings of Nalgonda, Telangana State, India. Botanist Dr. K. Srinivasa Reddy, Assistant Professor, Department of Botany, Govt. Degree College for Women, Nalgonda, Telangana, authenticated the authenticity of the plant specimen. After being shade-dried, the plant material was pulverized and stored in an airtight container. The material that had been pulverized was used in the extraction process.

# **Extract preparation**

The leaf powder was extracted using an orbital shaker macerating 100 grams of the powdered sample in 800 mL of n-hexane and 70% ethanol in water for 72 hours at room temperature. After extraction, the extract is concentrated and removed from plant debris with a twelve-fold-folded muslin cloth. It was determined that the hue, consistency, and yield % were. The extracted product was used to anthelmintic activity, phytochemical screening, and quantitative phytochemical estimation.<sup>6</sup>

# **Phytochemical screening**

Using standardized procedures, the extract of Cassia fistula L. leaves was subjected to phytochemical screening to determine the content of tannins, phenols, flavonoids, glycosides, and anthracene glycosides.<sup>7</sup>

#### **Total Phenol content determination**

Using gallic acid as a standard, the Folin-Ciocalteu test was used to quantify the total phenol concentration. 1.5% sodium carbonate solution was added after 5 minutes to 0.5 ml of plant extracts and 1.5 ml of diluted Folin-Ciocalteu's reagent (FCR) diluted 1:10 v/v. After adding distilled water to raise the tubes' final capacity to 10 ml, they were left to stand at room temperature for 90 minutes. Using a spectrophotometer, the sample's absorbance was measured at 750 nm in relation to the blank. To ensure accuracy, the experiment was conducted three times. The results are expressed as mean + standard deviation for the phenol content (also known as gallic acid equivalent, or GAE) per gram of dry weight.<sup>8</sup>

#### **Total Flavonoid content determination**

Using quercetin as a standard, the total flavonoid quantity was determined using the aluminum chloride method. A volumetric flask (10 ml volume) was filled with 1 ml of the test material and 4 ml of water. After five minutes, add 0.3 ml of 10% aluminum chloride and 0.3 ml of 5% sodium nitrite. 1 milliliter of 1 M sodium hydroxide was added to the reaction mixture after a 6-minute room temperature incubation period. At once, distilled water was added to bring the final volume to 10 ml. With a spectrophotometer set at 510 nm, the sample's absorbance was measured against the blank. To ensure accuracy, the entire experiment was conducted three times. The results were expressed as mean  $\pm$  standard deviation for the flavonoid content (also known as quercetin equivalent, or QE) per gram of dry weight.<sup>8</sup>

# **Biological study**

Because *Pheretima postuma* bears morphological and physiological similarities to human intestinal roundworm parasites, healthy adult Indian earthworms were employed in this work. Every earthworm had roughly the same size. They were gathered from a nearby location, cleaned, and stored in water.

# **Drugs**

Each group tested different dosages of Cassia fistula hydroalcoholic extract. The control was the normal saline solution. The standard medication was albendazole.

## **Experimental method**

Using adult *Pheretima postuma* Indian earthworms, anthelmintic activity was evaluated. By using bioassay, different concentrations of each extract and drug (25, 50, and 100 mg/ml) were evaluated to determine the worms' paralysis and death times. As a standard, albendazole was utilized, while saline water served as the control. The earthworms were taken out of the damp soil, cleaned with normal saline to get rid of all the feces, and then utilized for the study of anthelmintic. The earthworms were divided up into three groups, with each group having six earthworms. Before experiments started, all of the extract and the standard drug solution were prepared from fresh in normal saline. Standard drug solutions and extract were placed in different Petri dishes. The earthworms were all discharged into the 20 ml formulation in the following manner: Three different concentrations of hydroalcoholic extract and albendazole. The amount of time it took for each worm to become paralyzed and die was noted. When there was no movement at all, with the exception of the worms shaking violently, it was noted that the time for paralysis had arrived. After being submerged in heated water (50°C), the worms lost their ability to move and their body colors faded, signaling their death.

## **RESULTS AND DISCUSSION**

The powdered leaves of Cassia fistula were extracted using a maceration process and a hydroalcoholic solvent. Table 1 contains information on colour, consistency, and yield percentage.

Table 1: Yield and physical appearance of extract

S. No	Property	Report
1	Colour	Greenish black
2	Odour	Characteristic
3	Consistency	Pasty
4	%Yield	3.5 g

Table 2's tannins, phenols, flavonoids, glycosides, and anthracene glycosides have been detected during preliminary phytochemical screening. Table 2: Preliminary phytochemical screening

S. No	Constituents Report	
1	Carbohydrates Present	
2	Tannins &phenols Present	
3	Proteins	Present
4	Alkaloids	Absent
5	Glycosides	Present
6	Anthracene glycoside	Present
7	Flavonoids	Present
8	Saponins	Absent
9	Amino acids	Present

Table 3: Total phenol content

Total Phenol content (mg of Gallic acid equivalent/g of dry weight)
21.41 <u>+</u> 0.65

Mean ± Standard Deviation

Table 4: Total flavonoid content

Total flavonoid content (mg of Quercetin equivalent/g of dry weight)
18.41 <u>+</u> 0.85

Mean ± Standard Deviation

The background knowledge for some of the bioactivities is provided by quantifying the phytochemicals, such as total phenolic and flavonoid concentrations. Tables 3 and 4 display the extract's phenolic and flavonoid content concentrations. Numerous studies in the past have suggested that one of the key elements for the antioxidant activity of medicinal plants is phenolic compounds. Another group of physiologically active substances that have long been used to treat a variety of illnesses in humans, including cardiovascular disease, are flavonoids. Tannins have been linked to several health-promoting properties, including antioxidant, antimicrobial, and anti-mutagenic properties. Thus, our study clearly shows that the high concentration of phenol and flavonoids is responsible for some of the medical benefits of these leaves.

Cassia fistula hydro alcoholic extract showed dose-dependent anthelmintic action Table 5 displays that the standard medicine Albendazole exhibits paralysis at 17 minutes and death at 21 minutes, while the hydroalcoholic extract exhibits paralysis at 29 minutes and death at 37 minutes. Tannins were found to have anthelmintic action based on phytochemical screening. Tannins are polyphenolic substances chemically. When tannins attach to free proteins in the host animal's gastrointestinal tract or to glycoprotein on the parasite's cuticle, they can be fatal. The necessity to isolate the active ingredients that cause activity will be a future focus.

Table 5: Anthelmintic activity

S.No	Groups	Conc.(mg/ml)	Response		
			Paralysis time (min)	Deathtime (min)	
1	Normal control				
2	Hydro alcoholic	25	73	80	
		50	41	52	
		100	29	37	
3	Albendazole	25	41	45	
		50	24	27	
		100	17	21	

All values represent Mean +SD; n= 6 in each group.

#### **CONCLUSION**

The results of the phytochemical screening indicated the presence of flavonoids, phenols, tannins, and glycosides in the *Cassia fistula* leaf extract. This plant might be useful as a natural vermicide source, according to the extract's strong anthelmintic activity, total phenolic content value, and flavonoid content value. Based on these results, it is recommended that the extract be further tested to identify and isolate phytochemicals with vermicide properties.

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