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***Corresponding Author**

Musarrat Sharif

E-mail

musarratsharif388@gmail.com

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Anti-microbial, antioxidant potential of *Cassia fistula* and study their phytochemical assessments

Musarrat Sharif^{1*}, Farheen Ansari¹, Najam Ul Hassan²

¹Institute of Molecular Biology and Biotechnology, The University of Lahore, Lahore, Pakistan

²Department of Pharmacy, The University of Lahore, Lahore, Pakistan

Abstract

Cassia fistula (*C. fistula*) is a flowering plant and a member of *Fabaceae* family. This study was designed to examine antibacterial, antioxidant and phytochemical activity of ethanolic extract of *C. fistula* plant. The microbial inhibitory effect of ethanolic extracts of *C. fistula* was tested against Gram positive isolates such as *Bacillus cereus*, *Staphylococcus aureus* and Gram-negative isolates such as *Salmonella typhi*, *Klebsiella pneumoniae*, *Escherichia coli*, *Pseudomonas aeruginosa* using disc diffusion method and well diffusion method. The 25 mg extract of *C. fistula* leaves (CF-05) showed more zone of inhibition against *Salmonella typhi* i.e. (21mm) and in 50 mg extract of CF-13 fruit showed best zone of inhibition against *Salmonella typhi* i.e. (17mm). Qualitative analysis and antioxidant activity at various concentrations was also measured. The phytochemical analysis showed the presence of alkaloids, carbohydrates, fats, tannins, flavonoids, saponins, terpenoids, and sterols. The antioxidant activity in 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity revealed the distinguished antioxidant activity of *C. fistula*.



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Introduction

C. fistula is an extremely regular plant known for its restorative properties and is semi wild in nature. Its habitat includes Asia, China, Brazil and Indonesia [1]. *C. fistula* is a flowering plant in the family *Fabaceae*, local to southern Asia, from southern Pakistan east through India to Myanmar and south to Sri Lanka. *C. fistula* is a species from genus *Cassia*. It belongs to division Magnoliophyta of Kingdom Plantae. *C. fistula* from class Magnoliopsida and belongs to subclass and order Rosidae and Fabales respectively. It is a flowering plant in the family and subfamily as *Caesalpinaceae*. It has conspicuous racemes, up to 2” long with brilliant, yellow, fragrant blossoms [2]. Medicinally significant plants have been utilized from ages particularly in Asia and have been cleaned for the cure of particular disease. Poisonous free radicals from our body are identified and emptied by regular cancer prevention agents present in plants. The therapeutic properties of plants are being explored all through the world [3]. *C. fistula* fruit color is dark brown, and the covering of natural product is dry and hard. Fruit length is approximately 12 inches, and the diameter are 1 inch. *C. fistula* fruit shape is pod or pod like, elongated and does not attract wildlife, showy is the characteristics of *C. fistula* fruit. *C. fistula* seeds are poisonous in nature and oval in shapes which are attached with sticky brown pulp [4].

C. fistula bark have tonic and anti-dysentric properties, the powder of the bark is controlled in uncleanliness, jaundice and heart ailments. *C. fistula* is broadly utilized for its restorative properties, its appropriate property being that of gentle diuretic reasonable for kids and pregnant ladies. Many biologically important compounds were segregated and see from different parts of the plant [5]. The plant extracts were appeared as strong antibacterial, antifungal and antioxidant properties and the discoveries were done using ethanol solvent extracts and parts of the plant [6].

The chemical investigation of various parts of *C. fistula* has been reported. It was found to contain flavonoids, phenolic compounds and proanthocyanidins [7]. *C. fistula* extracts have been reported for various activities wound healing properties [1] and anticancer activity. The plant has fantastic therapeutic quality, and it applies a pain relieving and antipyretic effect [8]. Medications are produced utilizing *C. fistula*, which is valuable to cure high blood pressure, fever, joints pain, itching, skin burst and blood vomiting. For treatment of some overpowering disease, *C. fistula* is being utilized as

antimicrobial operators. This plant has high volume of phenolic compounds [9]. *C. fistula* can be utilized for antimalarial treatment [10]. Physicians used the flowers of *C. fistula* for the preparation of herbal medicines for diabetes [11]. Phytochemicals are the chemicals that present normally in plants. Phytochemicals are more typical because of their medicinal uses and the phytochemicals assume vital part against diseases like tumor and asthma. Because of their medicinal use’s phytochemicals are considered as “man-friendly medicines”. Phytochemicals contain flavonoids, alkaloids, saponins, tannins, crystals and minerals that have cancer prevention agent movement [12].

Materials and Methods

Sample collection and extract preparation

Different parts of *C. fistula* plant (stems, leaves, fruits and flowers) were freshly collected from Model Town Lahore, Pakistan (**Fig. 1**).

C. fistula plant (leaves, fruits, stems, flowers) were washed with tap water and shade dried for few days and powdered by with the assistance of grinder. After grinding then dissolved in solvent 500 mL ethanol and then place them for 6-7 days and shake the bottles twice in a day. After 6-7 days the blend was separated with the assistance of funnel and Whatman filter paper. At that point the concentrate was evaporated till the sticky concentrate was obtained with the assistance of rotary evaporator to evacuate the dissolvable totally. At that point pour the concentrate in falcon tubes and place them in water bath for evaporation. At last save the extract in freezer for against anti-bacterial, antioxidant and phytochemical activity.

Bacterial culture

The bacterial isolates such as *Escherichia coli* (*E. coli*), *Bacillus cereus* (*B. cereus*), *Klebsiella pneumoniae* (*K. pneumoniae*), *Staphylococcus aureus* (*S. aureus*), *Salmonella enterica* (*S. enterica*) and *Pseudomonas aeruginosa* (*P. aeruginosa*). These microorganisms were accessible in the Microbiology Laboratory, University of Lahore.

Preparation of inoculum

In normal saline inoculation of bacterial culture was completed. 6 test tubes were taken, and each test tube was loaded with 5 mL normal saline.



Fig. 1: *C. fistula* (A) seed (B) leaves, (C) flower and (D) fruit.

Disc diffusion method

For microorganism testing Muller-Hinton agar sterilized in autoclave at 121°C for 15 min and cooled at room temperature than poured into sterilized petri plates. The test bacterial inoculums were swabbed on the media. The discs were set on the surface of media with the help of sterilized forceps. The tests were led at various concentrations of extracts (10 μ L, 20 μ L). The extract was loaded with the help of 100 μ L pipette. The diameter of disc was 6 mm. The plates were incubated for 24 hours. The zones were observed in millimeter (mm) [13].

Well diffusion method

In well diffusion technique, 20 mL Muller-Hinton agar poured in sterilized petri plates. The arranged inoculums were swabbed on to the surface of agar plates and disinfected cotton swabs were utilized for appropriation of the inoculums. Wells were set up in the agar plates with the sterilized borer of 3 mm diameter. Wells were loaded with plant extract with the help of 100 μ L pipette. The petri plates were put in incubator for 24 hours at 37°C to watch the zones [14].

Phytochemical activity

There are number of tests for the identification of phytochemicals compounds in the plants. There is some important test such as Molisch test, Ninhydrin test, Wagner's test, Alkaline reagent test, Froth's test, Salkowski test, Triterpenoid test, Keller Kilianis test, Braymer's test and Opened looped and closed loop response test are described below [15].

Molisch's test

One mL of filtrate solution is treated with 2 drops of alcoholic-naphthol solution in a test tube. 2 mL of concentrated sulfuric acid was put on the side of the test tube. Improvement of the violet ring demonstrates the presence of carbohydrates. The test reagent gets dried out pentoses to form furfural (top reaction) and dries out hexoses to form 5-hydroxymethyl furfural (bottom reaction). The furfurals additionally respond with alpha-naphthol display in the test reagent to create a purple item. 1 mL of filtrate solution is treated with 2 drops of alcoholic alpha-naphthol solution in a test tube. Two mL of concentrated sulfuric acid is included the side of the test tube. Development of the violet ring at the intersection shows the presence of carbohydrates.

Alkaline reagent test

Alkaline reagent test comprises of reducing sugars being heated in the presence of an alkali get converted to powerful reducing species known as enediols. Enediols reduce the cupric compounds (Cu²⁺) present in the Benedict's reagent to cuprous compounds (Cu⁺) which get precipitated as insoluble red copper (I) oxide (Cu₂O). Extract sample was treated with a few drops of sodium hydroxide solution. Formation of intense yellow color, which became colorless on addition of dilute acid, indicated the presence of flavonoids.

Wagner's test

It is a watery solution of iodine and potassium iodide; utilized for micro chemical investigation of alkaloids. Wagner's test gives a reddish dark colored encourage that confirms the presence of alkaloids. Another filtrate portion is treated with Wagner's reagent (iodine in potassium iodide). Development of brown/reddish precipitate shows the presence of alkaloids.

Froth test

Froth's test includes froth being shaped on the water surface within the sight of saponin. 10 mL refined water is the reagent utilized as a part of the said test. On the off chance that the honeycomb foam is more noteworthy than 2 cm, range from the surface of the fluid perseveres following 10 minutes; the specimen is viewed as positive for saponin. Crude dry powder of extract is vigorously shaken with 2 mL of refined water and is permitted to remain for 10 min. If stable froth appears, it demonstrates the presence of saponins.

Opened loop-closed loop response test

Two drops of 1% sodium hydroxide solution was included in the test tube, and was heated in bubbling water for 3 min to get an unmistakable arrangement. 4 drops of 2% hydrochloric corrosive was added to this arrangement. At that point the arrangement ends up cloudy it demonstrates the presence of coumarins.

Ninhydrin test

Ninhydrin is a compound used to distinguish the smelling salts or essential and optional amines. Ninhydrin test is utilized to distinguish the presence

of α L-amino acid. Every amino corrosive that have a free amino gathering will give positive outcome (purple shading), while not free amino gathering proline will give a (yellow color). Ninhydrin (tri ketohydrindene hydrate) corrupts amino acids into aldehydes, smelling salts and CO₂ (on pH extend 4-8) however a progression of responses. The net outcome is ninhydrin in an in part lessened from hydrindantin. Ninhydrin at that point gathers with smelling salts and hydrindantin to deliver a seriously blue or purple shade, now and then called ruhemann's purple. For the preparation of ninhydrin reagent, 8 g of ninhydrin by weight was dissolved in 100 mL of acetone to get ready ninhydrin reagents.

Keller-Kiliani's test

A part of dry concentrate was blended with 1 mL of FeCl₃ reagent in test tube. At that point few drops of concentrated H₂SO₄ were incorporated. The greenish blue color inside a few minutes demonstrates the existence of cardiac glycosides.

Salkowski test

The extract sample is dissolved in chloroform and equivalent volume of concentrated sulphuric acid is included. Bluish red, cherry red and purple color in chloroform layer shows the presence of sterols while development of reddish-brown color of the interface shows the presence of triterpenoidal nucleus.

Braymer's test

The water extract of the crude dry powder of the plant is treated with 10 % alcoholic FeCl₃. The blue-black or green color indicates the presence of tannins.

Antioxidant Activity

DPPH, referred to formally as 2, 2-diphenyl-1-picrylhydrazyl, is a cell-porous, stable free radical that is normally used to assess the capacity of compounds to go about as free radical scavengers or hydrogen benefactors and to measure the cancer prevention agent activity of tissue separates. The response of DPPH with cell reinforcement or decreasing compound delivers the relating hydrazine DPPH₂, which can be trailed by shading change from purple (absorbance at 515-528 nm) to yellow. Test tube contained 50 μ L of concentrates in fixations from 1 to 5 mg/mL and 5 mL 0.1 mM DPPH solution (4 mg/100 mL ethanol) was included. The acquired blend was

vortexed and incubated for 30 minutes in room temperature in a moderately dim place and after that was perused utilizing spectrophotometer at 517 nm. The clear was 80 % (v/v) ethanol. Ascorbic acid (10 mg/mL dimethyl sulfoxide (DMSO) was utilized for correlation [16]. DPPH scavenging impact was computed utilizing the accompanying equation:

$$\text{DPPH scavenging effect (\%)} = [(AB-AA)/AB] \times 100$$

Results

Ethanol extract of *C. fistula* presented anti-bacterial activity against *E. coli* was CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems), CF- 13 (fruits) dissolved in DMSO. CF- 09 (21 mm) demonstrated the greatest impact of zones of inhibition with antibiotic Ciprofloxacin when contrasted with CF- 01 (18 mm), CF- 05 (20 mm) and CF- 13 (19 mm). The extract utilized against *S. typhi* was CF- 01, CF- 05, CF- 09, CF- 13 disintegrated in DMSO. CF- 05 (21 mm) demonstrated the most astounding effect of zones of inhibition with Ciprofloxacin as compared to CF- 01

(17 mm), CF- 09 (20 mm) and CF- 13 (19 mm). The extract utilized against *K. pneumoniae* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (19 mm) showed the most extreme impact of zones of inhibition with Ciprofloxacin when contrasted with CF- 01 (17 mm), CF- 05 (18 mm) and CF- 09 (15 mm). The concentrate utilized against *S. aureus* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (16 mm) demonstrated the greatest impact of zones of restraint with antibiotic Ciprofloxacin as compared to CF- 01 (12 mm), CF- 05 (14 mm) and CF- 09 (10 mm). The extract utilized against *P. aeruginosa* was CF- 01, CF- 05, CF- 09, CF- 13 disintegrated in DMSO. CF- 13 (21 mm) indicated maximum impact of zones of inhibition with Ciprofloxacin as compared to CF- 01 (16 mm), CF- 05 (15 mm) and CF- 09 (9 mm). The extract used against *B. cereus* was CF- 01, CF- 05, CF- 09 and CF- 13 dissolved in DMSO. CF- 01 (18 mm) and CF- 13 (18 mm) showed the highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 05 (17 mm), CF- 09 (10 mm). The results are shown in **Table 1, Fig. 2, Table S1** and **Fig S1**.

Table 1: Anti-bacterial activity of *Cassia fistula* plant by disc diffusion method (25mg/1 mL DMSO) (10 μ L bacteria, 20 μ L extract)

Bacteria	Positive Control	Negative Control	Zones of inhibition (mm)			
			CF-01 (flowers)	CF-05 (leaves)	CF-09 (stems)	CF-13 (fruits)
<i>E. coli</i>	25	0	18	20	21	19
<i>S. typhi</i>	20	0	17	21	20	19
<i>K. pneumoniae</i>	20	0	17	18	15	19
<i>S. aureus</i>	15	0	12	14	10	16
<i>P. aeruginosa</i>	35	0	16	15	9	21
<i>B. cereus</i>	20	0	18	17	10	18

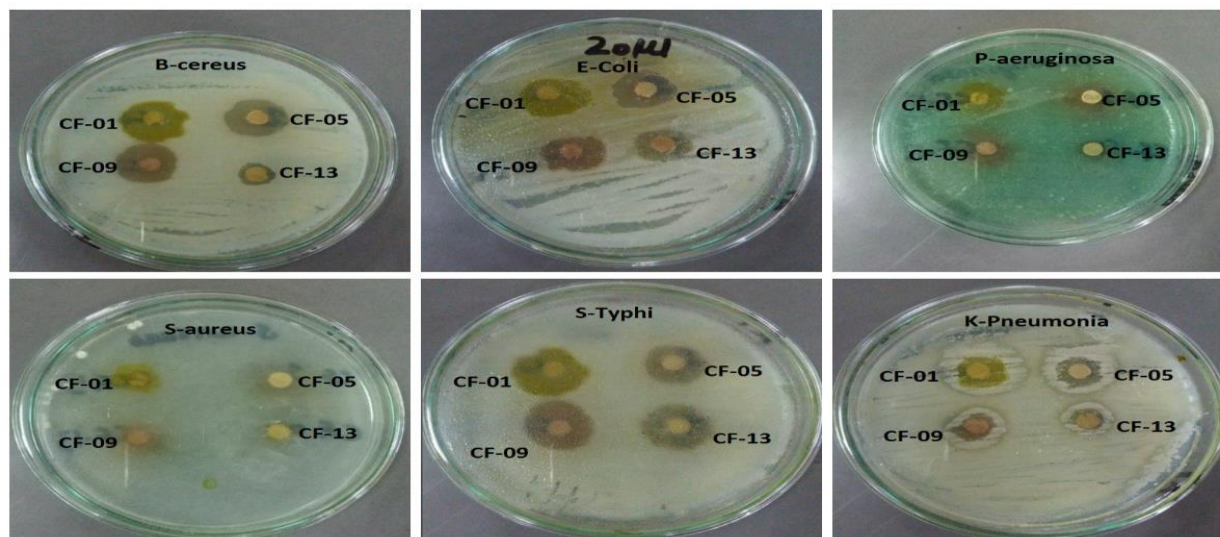


Fig. 2: The effect of *Cassia fistula* plant extracts against bacterial isolates by disc diffusion method at 25 mg/1 mL DMSO concentration with 10 μ L bacterial culture and 20 μ L extract.

The concentrate utilized against *S. typhi* was CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems), CF- 13 (fruits) dissolved in DMSO. CF-01 (21 mm) indicated the greatest impact of zones of restraint with Ciprofloxacin when contrasted with CF- 05 (14 mm), CF- 09 (15 mm) and CF-13 (14 mm). The extract utilized against *E. coli* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF-13 (19 mm) showed the most extreme impact of zones of hindrance with Ciprofloxacin as compared to CF- 01 (12 mm), CF- 05 (14 mm) and CF- 09 (16 mm). The concentrate used against *P. aeruginosa* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (21 mm) indicated the maximum effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (15 mm), CF- 05 (14 mm) and CF- 09 (12 mm). The extract

utilized against *B. cereus* was CF- 01, CF- 05, CF- 09, CF-13 disintegrated in DMSO. CF- 01 (17 mm) indicated the maximum effect of zones of inhibition with Ciprofloxacin as compared to CF- 05 (11 mm), CF- 09 (15 mm) and CF- 13 (14 mm). The concentrate utilized against *K. pneumoniae* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (17 mm) indicated the highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (12 mm), CF-05 (15 mm) and CF- 09 (13 mm). The extract utilized against *S. aureus* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 01 (23 mm) showed the highest effect of zones of inhibition with ciprofloxacin as compared to CF- 05 (15 mm), CF- 09 (20 mm) and CF-13 (18 mm). The results are shown in **Table 2**, **Fig. 3**, **Table S2** and **Fig. S3**.

Table 2: Anti-bacterial activity of *Cassia fistula* plant by disc diffusion method (50 mg/1 mL DMSO) (10 μ L bacteria, 20 μ L extract)

Bacteria	Positive Control	Negative Control	Zones of inhibition (mm)			
			CF-01 (flowers)	CF-05 (leaves)	CF-09 (stems)	CF-13 (fruits)
<i>E. coli</i>	25	0	12	14	16	19
<i>S. typhi</i>	20	0	21	14	15	14
<i>K. pneumoniae</i>	20	0	12	15	13	17
<i>S. aureus</i>	15	0	23	15	20	18
<i>P. aeruginosa</i>	35	0	15	14	12	21
<i>B. cereus</i>	20	0	17	11	15	14

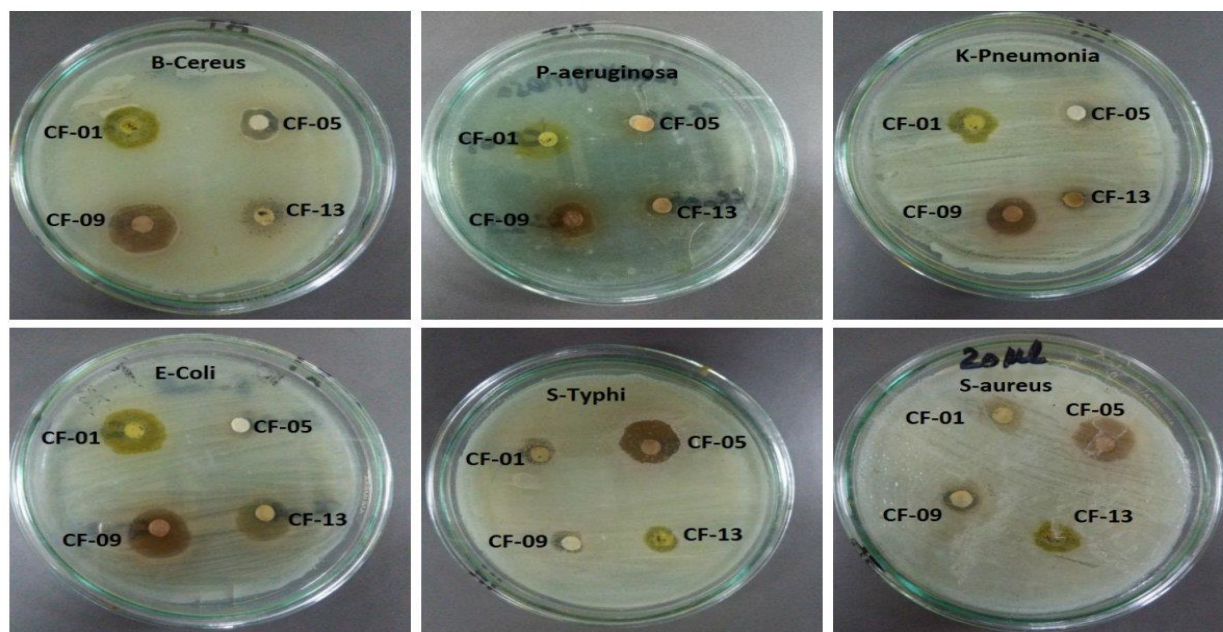


Fig. 3: The effect of *Cassia fistula* plant extracts against bacterial isolates by disc diffusion method at 50 mg/1 mL DMSO concentration with 10 μ L bacterial culture and 20 μ L extract.

The concentrate utilized against *S. typhi* was CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems), CF- 13 (fruits) dissolved in DMSO. CF- 09 (17 mm) demonstrated the highest impact of zones of inhibition

with Ciprofloxacin as compared to CF- 01 (0 mm), CF- 05 (15 mm) and CF- 13 (15 mm). The extract used against *E. coli* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (15 mm) showed the

highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (8 mm), CF- 05 (10 mm) and CF- 09 (13 mm). The extract used against *P. aeruginosa* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 05 (19 mm) indicated the highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (10 mm), CF- 09 (14 mm) and CF- 13 (13 mm). The extract utilized against *B. cereus* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 09 (23 mm) showed the highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (0 mm), CF- 05

(0 mm) and CF- 13 (21 mm). The extract used against *K. pneumoniae* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (16 mm) showed the highest effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (0 mm), CF- 05 (10 mm) and CF- 09 (13 mm). The extract used against *S. aureus* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 09 (20 mm) showed the maximum effect of zones of inhibition with Ciprofloxacin as compared to CF- 01 (0 mm), CF- 05 (11 mm) and CF- 13 (18 mm). The results are shown in **Table 3**, **Fig. 4**, **Table S3** and **Fig. S3**.

Table 3: Anti-bacterial activity of *Cassia fistula* plant by well diffusion method (50 mg/1 mL DMSO) (10 μ L bacteria, 20 μ L extract)

Bacteria	Positive Control	Negative Control	Zones of inhibition (mm)			
			CF-01 (flowers)	CF-05 (leaves)	CF-09 (stems)	CF-13 (fruits)
<i>E. coli</i>	25	0	8	10	13	15
<i>S. typhi</i>	20	0	0	15	17	15
<i>K. pneumoniae</i>	20	0	0	10	13	16
<i>S. aureus</i>	15	0	0	11	20	18
<i>P. aeruginosa</i>	35	0	10	19	14	13
<i>B. cereus</i>	20	0	0	0	23	21

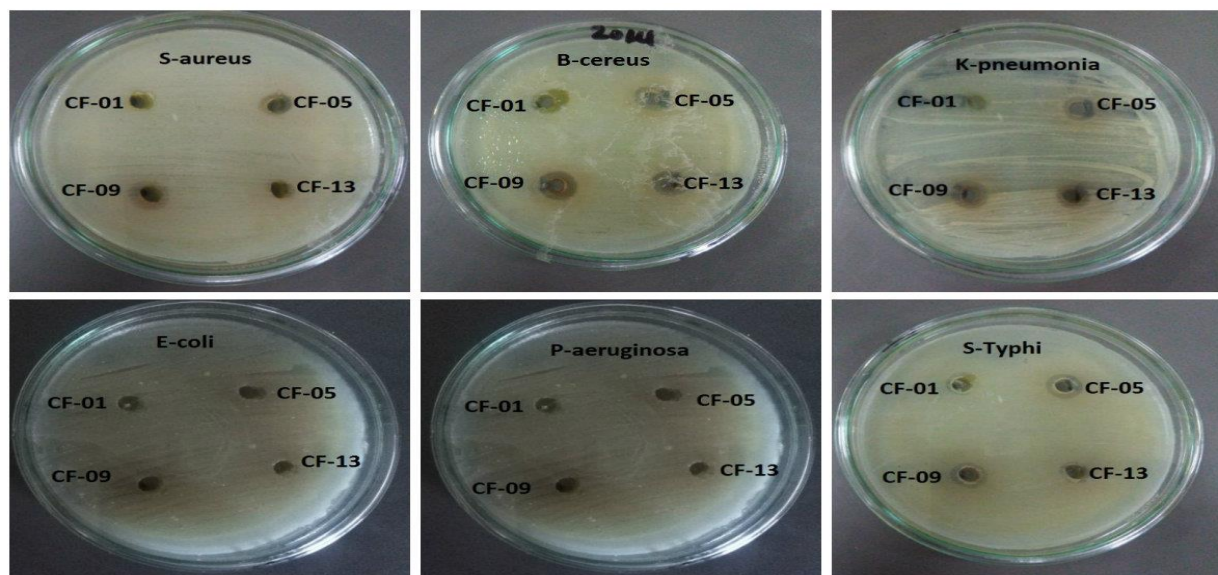


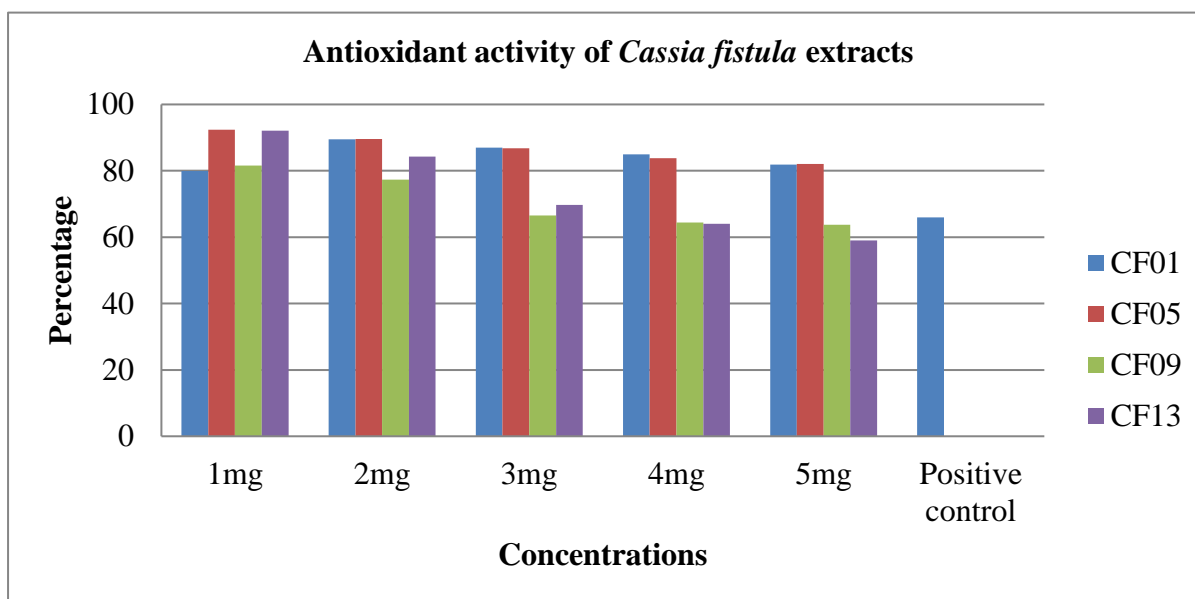
Fig. 4: The effect of *Cassia fistula* plant extracts against bacterial isolates by well diffusion method at 50 mg/1 mL DMSO concentration with 10 μ L bacterial culture and 20 μ L extract

The Molisch's test, alkaline test, Ninhydrin test, Braymer's test and Wagner's test gave positive results for CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems) and CF- 13 (fruits) extracts. The froth test demonstrated the positive results for CF- 01, CF- 05 and CF- 13 however negative result for CF- 09. The CF- 01 and CF- 05 extracts indicated positive results but negative results for CF- 09 and CF- 13 after applying opened loop-closed loop test. The crude extract indicated the positive results for CF- 01, CF-

09 and CF- 13 but CF- 05 indicated negative result after applying Keller-Kiliani's test. The crude concentrate demonstrates the positive result for CF- 01, CF- 05 and CF- 09 however CF- 13 showed the negative result after applying Salkowski test. The crude extract indicated the negative results for CF- 01, CF- 05 and CF- 09 but the CF- 13 demonstrated the positive result after applying Triterpenoid's test (**Table 4**). *C. fistula* extracts indicated that all parts exhibited good antioxidant potential in comparison to

Table 4: Phytochemical Analysis of *Cassia fistula* Plant Extracts.

Active Constituents	Test Names	Plant extracts			
		CF-01 (flowers)	CF-05 (leaves)	CF-09 (stems)	CF-13 (fruits)
Carbohydrates	Molisch's test	+	+	+	+
Flavonoids	Alkaline test	+	+	+	+
Alkaloids	Wagner's test	+	+	+	+
Saponins	Froth test	+	+	-	+
Coumarins	Opened loop-closed loop test	+	+	-	-
Proteins and amino acids	Ninhydrin test	+	+	+	+
Cardiac glycoside	Keller-Kiliani's test	+	-	+	+
Steroids	Salkowski test	+	+	+	-
Terpenoids	Triterpenoid's test	-	-	-	+
Tannins	Braymer's test	+	+	+	+

**Fig. 5:** Antioxidant activity of *Cassia fistula* extracts at different concentration.

ascorbic acid (vitamin C). Ascorbic acid was used as positive control (Fig. 5).

Discussion

C. fistula is an extremely regular plant known for its medicinal properties is a semi-wild in nature. Its habitat includes Asia, China and Brazil [1]. It is deciduous and blended rainstorm woodlands all through larger parts of India and it is broadly utilized as a part of conventional therapeutic arrangement of India [2]. Regular physicians utilize the flowers of *C. fistula* for the preparation of herbal medicines for diabetes [11]. Antibacterial activity demonstrates great zones of hindrance by utilizing *C. fistula* plant extract against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Bacillus cereus*, *Salmonella enterica* and *Klebsiella pneumoniae*.

In this study different concentrations of bacteria and extracts were used like 10 μ L bacteria and 20 μ L extract. The ethanolic concentrate obtained from the flowers, leaves, stems and fruits of *Cassia fistula* plant which displayed anti-bacterial activity against *E. coli* was CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems), CF- 13 (fruits) dissolved in DMSO. CF- 09 (21 mm) exhibits the greatest effect of zones of restraint with antibiotic Ciprofloxacin when accordance with CF- 01 (18 mm), CF- 05 (20 mm) and CF- 13 (19 mm). These findings are in line with Beena *et al.*, (2013) inspected that *E. coli* demonstrated more zone of inhibition 22 mm, 24 mm, 19 mm by the ethanolic concentrate of *C. fistula* plant. The extract utilized against *S. typhi* was CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems), CF- 13 (fruits) disintegrated in DMSO. CF- 05 (21 mm) demonstrated the most outstanding effect of zones of hindrance with Ciprofloxacin as compared to

CF- 01 (17 mm), CF- 09 (20 mm) and CF- 13 (19 mm). These discoveries are similar with Rizvi *et al.*, (2011) reported that Cassia species had a significant activity against Gram positive and indicated zone inhibition 18 mm, 15 mm, 10 mm by the ethanolic extract of *C. fistula* plant. The ethanolic extract of CF against *K. pneumoniae* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (19 mm) showed the most extreme impact of zones of inhibition with ciprofloxacin when contrasted with CF- 01 (17 mm), CF- 05 (18 mm) and CF- 09 (15 mm).

These discoveries are in line with [17] examined that ethanol extract exhibited high zones of inhibition 14 mm, 20 mm and 19 mm by the ethanolic extract of *C. fistula* plant. The concentrate utilized against *S. aureus* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (16 mm) demonstrated the greatest impact of zones of restraint with antibiotic Ciprofloxacin as compared to CF- 01 (12 mm), CF- 05 (14 mm), CF- 09 (10 mm). These findings are similar with [18] examined their results obtained from their study only ethanolic extract showed the inhibition zone of 10, 8 and 12 mm in three consecutive days of the *C. fistula* plant. The extract utilized against *P. aeruginosa* was CF- 01, CF- 05, CF- 09, CF- 13 disintegrated in DMSO. CF- 13 (21 mm) indicated maximum impact of zones of inhibition with Ciprofloxacin as compared to CF- 01 (16 mm), CF- 05 (15 mm) and CF- 09 (9 mm). In any case these findings are in line with [19] showed more zone of hindrance 10 mm, 14 mm and 21 mm by the ethanolic concentrate of *C. fistula*. The concentrate utilized against *S. typhi* was CF- 01, CF- 05, CF- 09, CF- 13 disintegrated in DMSO. CF- 13 (15 mm) demonstrated the most elevated impact of zones of hindrance with ciprofloxacin when contrasted with CF- 01 (11 mm), CF- 05 (12 mm), CF- 09 (10 mm). However, these discoveries are in line with [9] reported that extract can be utilized against *S. typhi* which showed zones of inhibition 15 mm, 14 mm of *C. fistula* plant. The extract used against *E. coli* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 05 (15 mm) showed the most elevated effect of zones of inhibition with ciprofloxacin as compared to CF- 01 (14 mm), CF- 09 (12 mm), CF- 13 (13 mm). These findings are similar with [20] reported that the extract of leaves and petroleum ether extract gave good result against *E. coli* and *P. aeruginosa*, like that acid content of leaves showed good activity against *E. coli*, *S. aureus*, *P. aeruginosa* and non- active against *B. subtilis*, they indicate zone of inhibition of 15, 10 and 14 mm by the extract of *C. fistula* plant. Crude ethanolic extract of *C. fistula* plant used against *S.*

aureus was CF- 01, CF- 05, CF- 09, CF-13 dissolved in DMSO. CF-01 (16 mm) demonstrated the highest impact of zones of restraint with Ciprofloxacin when contrasted with CF- 05 (15 mm), CF- 09 (10 mm), CF- 13 (13 mm). These results demonstrated that the tested crude extract indicated antibacterial activity towards the bacteria. However, these findings are similar with [21] analyzed that ethanolic extract gave good result against *S. aureus* they showed zone of inhibition 20, 14 and 12 mm of *C. fistula* plant just, however no activity against other bacterial species. The extract utilized against *P. aeruginosa* was CF- 01, CF- 05, CF- 09, CF- 13 dissolved in DMSO. CF- 13 (12 mm) showed the most extreme effect of zones of restraint with ciprofloxacin when contrasted with CF- 01 (10 mm), CF- 05 (11 mm), CF- 09 (10 mm). These findings are similar with [22] inspected that the leaf concentrate of *C. fistula* showed antibacterial activity against all the microorganisms tested. Among the ethanol extract displayed higher activity than alternate extracts and petroleum ether extract indicated least activity. Ethanol (18, 20 mm inhibition zone), ethyl acetate (14, 22 mm inhibition zone), chloroform (13, 16 mm inhibition zone) and petroleum ether (12, 14 mm inhibition zone) extracts of the leaf showed marked activity against all the tested organisms for example, *B. cereus*, *S. aureus*, *E. coli*, *K. pneumoniae*, *P. aeruginosa*.

In this study the qualitative analysis of *C. fistula* revealed the presence of different phytochemicals like terpenoids, flavonoids, tannins and phenolic compounds. The rough concentrate indicates positive results for CF- 01 (flowers), CF- 05 (leaves), CF- 09 (stems) and CF- 13 (fruits). The unrefined concentrate demonstrates positive results for CF- 01, CF- 05, CF- 09 and CF- 13. The crude extract showed the positive results for CF- 01, CF- 05, CF- 09 and CF- 13. The unrefined concentrate demonstrates the positive results for CF- 01, CF- 05 and CF- 13 however negative result for CF- 09. The unrefined extract indicates the positive results for CF- 01 and CF- 05 but negative results for CF- 09 and CF- 13. The rough extract demonstrates the positive results for CF- 01, CF- 05, CF- 09 and CF- 13. The unrefined extract indicates the positive results for CF- 01, CF- 09 and CF- 13 but CF- 05 indicates negative result. The crude concentrate demonstrates the positive result for CF- 01, CF- 05 and CF- 09 however CF- 13 shows the negative result. The rough extract indicates the negative results for CF- 01, CF- 05 and CF- 09 but the CF- 13 demonstrates the positive result. The unrefined concentrate indicates the positive results for CF- 01, CF- 05, CF- 09 and CF- 13. In any case these findings

are in line with [23] qualitative examination of *C. fistula* revealed the presence of various phenolic compounds like tannins, phenolic compounds, flavonoids, terpenoids, etc. and these active compounds are notable for their pharmacological activities, tannins, flavonoids and terpenoids are notable for their antimicrobial properties. But that as it may, these discoveries are in line with high performance liquid chromatography (HPLC) based correlation demonstrated that there is a variety concerning maintenance times inside various concentrates showed the presence of various phytochemicals.

Conclusion

These bacteria *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Bacillus cereus*, *Salmonella typhi* and *Klebsiella pneumoniae* are pathogenic and create various skin diseases and inflammation hence from this study tried to investigate medicinal value of plant *Cassia fistula*. Results have proved the importance of plant extracts to inhibit growth of bacteria, thus plant extracts may be useful to fight against bacterial causing diseases. The phytochemical analysis showed the presence of alkaloids, carbohydrates, fats, tannins, flavonoids, saponins and terpenoids in traditional medicine; hence it can be used in the treatment of intestinal disorders, diabetes and as antipyretic and analgesic.

Conflict of interest

The authors declare no conflict of interest.

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