

To Determine the Effectiveness of Cassia tora Plant (Charota) Against Vaginal Microflora Amongst Rural and Urban Populations.

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Abstract

Vaginal infections are mainly caused by various microorganisms that infect the vaginal tract and are a more common condition in women that can lead to various diseases such as vaginitis, urinary tract infections, bacterial vaginosis, trichomoniasis, etc. *Cassia tora* (family Leguminosae/ Fabaceae), is a plant with tremendous medicinal properties, possessing many pharmacological activities including antibacterial, antifungal, anti-inflammatory, and antioxidant properties. The chloroform and aqueous extract of leaves of *Cassia tora* exhibited antibacterial activity (50-250 mg/ml) against six bacterial strains respectively out of 56 various bacterial strains. The MIC values range from (12.5-100 mg/ml) against those bacterial strains. Phytochemical studies have shown that the leaf extract contains flavonoids, saponins, alkaloids, glycosides, terpenoids, tannins, oils, and reducing sugars, as well as antioxidant activity was performed using a DPPH assay against chloroform and an aqueous extract of *Cassia tora* leaves. The traditional medicinal properties of leaves of *Cassia tora* were evaluated to determine an antibacterial property of leaf extracts against vaginal microflora which cause vaginal infection, to analyse the phytochemical constituents and antioxidant properties of this plant, as in future, the plant extracts of this plant will be incorporated with the commercial intimate washes and in cosmetics products.

Keywords: Cassia tora; Antibacterial; Antioxidant; Pharmacological Properties; Vaginal infection; Intimate washes.

Introduction:

The vagina is the most sensitive part of a woman. Any neglect of this can weigh heavily on the woman. Improper hygiene can cause infections such as itching, burning, irritation, pain, white discharge, and discomfort. Maintaining vaginal cleanliness with intimate wash has become an important part of a woman's lifestyle. Intimate hygiene is still frowned upon [Agrawal A, Sehrawat H 2021].

Despite increasing literacy among rural and urban areas, women of all ages continue to practice poor hygiene during menstruation or for other reasons. Such topics are either treated softly with metaphors or ignored entirely. There's nothing wrong with being ashamed, but people are ashamed to talk about the need for proper treatment because a little carelessness can exaggerate the infection into a serious form [Agrawal A, Sehrawat H 2021]. About 90% of

women will experience vaginitis at least once in their lifetime [Yadav SK, Jain GK, Mazumdar A, Khar RK 2019]. It is caused by other factors such as poor hygiene, sweating, hormonal imbalance, menstruation, pH changes, pregnancy, sexual intercourse, hair loss techniques, etc. The microorganisms that dwell inside the vagina are known as vaginal flora or vaginal microbiota or vaginal microbiome. Human vaginal flora plays an intense role in reproductive health and disease. The vaginal flora contributes to a variety of microorganisms, many of which play an important role in keeping the vagina healthy and protected through multiple mechanisms that prevent other microorganisms from colonizing the vaginal epithelium. Lactobacillus sp. dominates vaginal floral diversity; these species contribute to vaginal health by producing lactic acid, hydrogen peroxide, and other chemicals that inhibit the growth of yeast and other undesirable organisms [Gupta A 2021]. Pathogenic microorganisms such as bacteria, archaea, protists, fungi, and viruses exist in the vaginal microflora environment [Kim JM, Park Y J 2017]. The vaginal microflora is a mixture of bacteria, including gramnegative and gram-positive bacteria, and an imbalance in the microflora composition leads to vaginal infections and diseases. The vaginal microflora is important in preventing vaginal diseases by protecting against pathogens and maintaining vaginal health. Even though the vagina is a self-cleaning organ, using harsh chemicals in cleansers can irritate it. But, using intimate washes can help in lessening the burning sensation, rashes, and itching and removing the unwanted odour. Using soap can make things worse by washing away the beneficial bacteria known as lactobacillus, which can upset the pH balance of your vagina Many efforts have been made over the past decades to investigate the mechanism of plant compounds against pathogens of vaginal infections. Cleansers and other intimate hygiene products made from plant extracts can help balance vaginal pH and reduce vaginal infections. Women prefer herbal intimate hygiene products because they are free of harsh chemicals, have fewer side effects, are inexpensive, and are environmentally friendly.

Cassia tora commonly known as charota is a name that is familiar to everyone. This plant belongs to the Fabaceae family which includes the largest genera of about 2500 species. Cassia tora is an annual shrub that is widely distributed in tropical countries (like India, Pakistan, Bangladesh, and China) and grows well in the wasteland as a rainy-season weed [Pawar H A, MD Mello 2010]. Cassia tora is one of the most important sources of traditional medicinal plants containing important phytochemicals and is widely used in Ayurveda. Cassia tora plant is Composed of several phytochemicals such as flavonoids, alkaloids, steroids, terpenoids, anthraquinones, tannins, saponins, glycosides, etc. which have many Pharmacological actions. These plants have been found to have therapeutic value in solving major health problems and are of great advantage in treating and eliminating various ailments, including asthma, gastrointestinal symptoms, skin diseases, respiratory and urinary problems, vaginal infection, and hepatic, and cardiovascular disease [Bhandirge SK, Patel V, Patidar A, Pasi A, Sharma V 2016]. Phytochemical compounds of the medicinal plant have antibacterial, antifungal, antiinflammatory, and antioxidant properties that can be used to treat many genital and urinary tract infections or diseases. The future prospects of this study are to isolate and diagnose the major pathogenic bacteria of the female genital tract (vaginal infection) as well as the inhibitory effect of plant extracts on these bacteria to evaluate the efficacy of these extracts and to estimate the possibility of using these extracts as alternatives to commercial vaginal washes. The main aim of this study is to check the antibacterial activity of Cassia tora against isolated bacteria from vaginal flora in both rural and urban populations.

1. Plant extract preparation

1.1 Plant Sample Collection

Leaves of *Cassia. tora* plants were collected from the nearby local field of Kharora Village in Tilda Tehsil in Raipur District of Chhattisgarh State, India in the month of September 2021.

1.2 Preparation of Extracts

Leaves were collected, shed dried, and made powder. Extraction was done using the Soxhlet apparatus (12 grams leaf powder for 200 ml solvent). Distilled water and chloroform were used for extraction. The obtained extracts were concentrated under a vacuum at 40°C. The residual semisolid extracts were stored in a refrigerator at 4°C. Organoleptic properties of the plant extracts (colour, texture, and odour) were determined in respective solvents under dried and wet conditions.

2. Isolation of bacteria from vaginal sample

2.1 Vaginal Sample Collection

Fifty Vaginal mucus samples were collected from healthy women of age 18 to 50 years from rural and urban areas, from Kharora village in Raipur district at Chhattisgarh state, India, respectively. A questionnaire developed based on the literature was used for data collection. Written consent was taken from the women by giving information about the study. After administering the questionnaire. Vaginal samples were taken from the posterior wall of the vagina without touching the vulva using a sterile cotton swab by themselves.

2.2 Statistical analysis

The presence of pathogens that can cause vaginal infection and women's health condition were considered dependent variables. Factors are-

Based on the area they belong (Rural and Urban)

Based on Age group (18-25, 26-35 & 36-45)

Based on Marital Status (Married & Unmarried)

2.3 Inoculation of Samples

Cotton swabs were streaked on the Luria Bertani, Muller Hinton Agar plates out of which fifty-six selected individual colonies were streaked on the fresh media to obtain pure cultures, and pure cultures were stored at 4°C at the refrigerator. Out of 56 bacterial strains, 47 bacteria were Gram-positive, and 9 bacteria were Gram-negative bacteria. Out of these bacterial strains, only 6 isolates were selected for the analysis of antibacterial activity, and MIC, the selection was done based on morphological characterization like colony size, shape, color, elevation, etc. A loop full of bacterial strains was inoculated into 5ml of Luria Bertani-Muller Hinton broth. The turbidity of the actively growing bacterial suspension was used when equivalent to approximately 1×10⁶ CFU/ml.

3. Analysis

3.1 Antimicrobial activity

3.1.1 Agar Disc Diffusion Assay

Antibacterial assays were performed by Agar Disc Diffusion Method using Luria Bertani-Muller Hinton agar media. Plates were inoculated by bacterial broth cultures (inoculum size of 1×10^6 CFU/ml). 6mm sterilized disc were impregnated in the respective concentrations (200mg/ml, 150mg/ml, 100mg/ml, 50mg/ml) of both the plant extracts (Aqueous extract and Chloroformic extract) that was prepared by dissolving in DMSO. The discs were kept over the agar plates. Amoxicillin was used as a positive control and DMSO as a negative control. Antimicrobial activity was observed and calculated, and the diameter of the zone of inhibition was indicated by a clear area that was devoid of the growth of bacteria.

3.1.2 *MIC* (*Minimum Inhibitory Concentration*)

Plant extracts that gave a positive result for the agar disc diffusion assay were used to determine the minimum inhibitory concentration (MIC) of the aqueous extract and chloroformic extract by broth dilution method. Two-fold serial dilutions of the extracts were prepared in the DMSO (200, 100, 50, 25, and 12.5µg/ ml). 3ml broth was taken in each tube containing different concentrations of plant extracts and inoculated with a loop full culture of bacteria. Media only with bacterial culture were used as positive control and plant extracts in broth were used as a negative control after overnight, any visible growth in the incubation tubes was observed. The MIC was defined as the lowest concentration that did not show any visible growth when compared to control tubes containing only the extracts. The absorbance at 600nm wavelength was used to calculate antimicrobial activity.

3.2 Antioxidant activity (DPPH Scavenging activity)

0.1mM solution of DPPH in methanol was prepared (i.e., 33mg in 1L). Different concentrations (100mg/ml, 200mg/ml, and 300mg/ml) of the plant extracts were prepared. The mixture (1ml plant extracts + 4ml DPPH + methanol) was shaken vigorously and incubated in dark for 30 minutes at RT. DPPH + methanol solution was taken as blank and ascorbic acid was taken as standard for comparison. The UV-Vis Spectrophotometer was used to measure the absorbance at 517nm. The free radical scavenging activity of the extracts was measured by a decrease in absorbance. The reaction mixture with the lowest concentration has the highest DPPH scavenging activity. The following equation was used to calculate the DPPH scavenging activity:

DPPH scavenging activity (%) = $[(A_b - A_a) / A_b] \times 100$

Where the absorbance of the blank was A_b and the absorbance of the extracts or standard solution was A_a [Kumar A, Anand V, Dubey R C, Goel K K 2019].

3.3 Qualitative analysis of Phytochemicals

Extracts were prepared for the analysis of each phytochemical according to the respective method used. Qualitative tests for phytochemical screening were performed for Proteins and Amino acids, Anthraquinones, Terpenoids, Steroids, Alkaloids, Flavonoids, Saponins, Tannins, Glycosides, Reducing sugars, Fats and oils and, Lignin.

3.4 Bacterial identification using 16S RNA gene sequencing

In this study, the 56 vaginal bacteria strains were isolated out of which 4 pure cultures of bacteria were sent for identification (16S RNA gene Sequencing method) at NCIM (National Collection of Industrial Microorganisms), Pune, Maharashtra.

Results

1.1 Plant extract: Organoleptic properties of both the aqueous and chloroform extracts were determined including weight, colour, texture, and odour.

Organoleptic Properties Determination

PLANT EXTRACTS	WEIGHT	TEXTURE	COLOUR	ODOUR
Aqueous Extract	4.85 gm	Foamy when shaken	Brown	Smoky
Chloroformic extract	4.13 gm	Non-foamy	Greenish brown	Pungent

Table 1. Organoleptic properties



FIGURE 1. CASSIA TORA PLANT



FIGURE 2. CASSIA TORA PLANT LEAF POWDER



FIGURE 3.EXTRACTION OF LEAF EXTRACT



FIGURE 4. LEAF CRUDE EXTRACT

1.2 VAGINAL SAMPLE

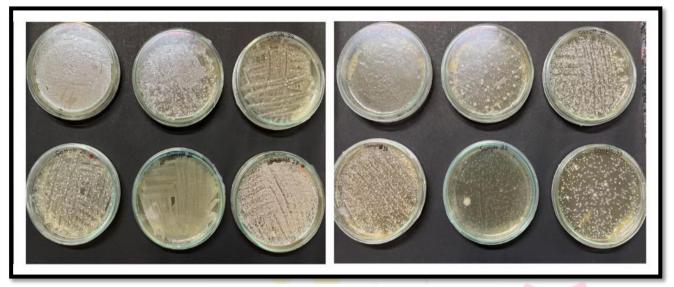


FIGURE 5. VAGINAL SAMPLES COLLECTIONS



FIGURE 6. PURE CULTURES OF BATERIAL STRAIN

1.3 Bacterial identification:

Out of 50 samples, 9 pure cultures were obtained and Gram staining was done for all the 9 isolates out of which 7 bacteria were Gram-positive and 2 bacteria were Gram-negative and out of that 9 we worked on 6 isolates that were not identical.

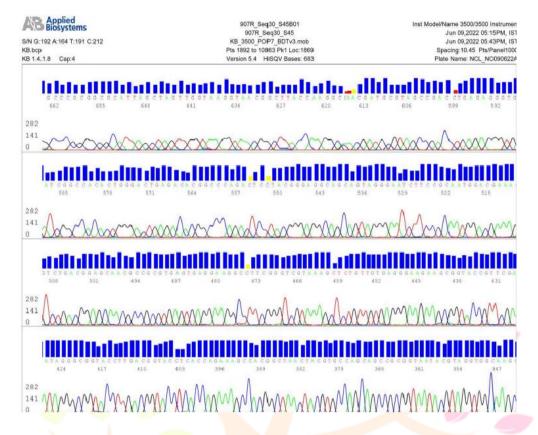
NAME OF SAMPLE	Bacterial sample 3	Bacterial sample 4	Bacterial sample 1	Bacterial sample 2	Bacterial sample 5	Bacterial sample 6	Bacterial sample 7	Bacterial sample 8	Bacterial sample 9
MORPHOLOGY									
I. Shape of Colony	Circular	Circular	Circular	irregular	circular	circular	Circular	Circular	Circular
II. Gram's Nature	Positive	Positive	Positive	negative	positive	Positive	positive	Negative	Positive
III. Colony Color	White	Creamish white	White	Off-white	White	White	Creamish white	White	White
VI. Elevation	Raised	Convex	Flat	Flat	circular	Convex	raised	Raised	Umbonate
V. Microscope view									
VI. Microscopic Shape	Spherical	Rod	Rod	Spherical	Curved	Rod	Spherical	Curved	Rod

Molecular identification of Bacterial species 3:

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Sr.	Strain	Aim	Primer	NCBI BLAST	Remarks
No.				(Type Strain)	
1	Seq02	Identification of	907R	Sequence ID:	Strain showed closest
	S45_	16S rRNA gene	(651bp)	NR_026140	homology with
	NC09062	++ Strain received		Taxon ID:	Alkalihalobacillus
	2_	from Dr. Arunima		Alkalihalobacillus	sp. (Closer to
		Sur, Amity		clausii	Multiple species)
		Institute Of		Identities:	++
		Biotechnology,		646/651(99.23%)	This genus is
		Amity University		Ref: FEMS Microbiol.	comprised of large
		Chhattisgarh.		Lett. 117, 61-66 (1994)	number of species,
				++	hence 16S rRNA may
				Sequence ID:	not resolve species
				NR_108311	identity. Genome
				Taxon ID:	sequencing and
				Alkalihalobacillus	detailed phenotypic
				Rhizosphaerae	tests might resolve
				Identities:	species level identity.
				645/651(99.08%)	Kindly refer latest
		III		Ref: Antonie Van	literature related to
		Ke/earc		Leeuwenhoek 100	this genus taxnomy.
				(3), 437-444 (2011) ++	
				Sequence ID:	
				NR_025741	
				Taxon ID:	
				Alkalihalobacillus	
				Patagoniensis	
				Identities:	
				624/654(95.41%)	
				Ref: Int. J. Syst. Evol.	
				Microbiol. 55 (PT 1),	
				443-447 (2005)	

Table 2. Sequencing results

>907RC Seq30 S45(Page2)



Graph of sample no 3 which showed Gram-positive bacteria.

1.4 Antimicrobial activity.

The antibacterial activity of the aqueous and chloroform extracts of the leaves of the *cassia Tora* plant was compared with the standard antibiotic Amoxicillin (control) by measuring the zone of inhibition diameter. The aqueous and chloroform extracts were tested for antibacterial activity against a number of both gram-negative and gram-positive bacteria. The highest zone of inhibition (20mm) was recorded with bacterial sample no.2 and found that both crude extracts of cassia Tora were active against bacterial samples no.1, 2, and 4 and were not able to inhibit the growth of bacterial sample no. 3 and 5. It is widely known that lactic acid bacteria inhibit the growth of pathogenic microorganisms through the production of different inhibiting substances (Arutcheva et al. 2001, Matu et al. 2010). The summarized results are presented in Tables 3 and 4 respectively.

1.4.1 Agar disc diffusion method:

Zone of inhibition of aqueous plant extract:

Contraction	50mg/ml	100mg/ml	150mg/ml	200mg/ml	Amoxicillin
BS 1	-	11mm	12mm	13mm	17mm
BS 2	7mm	15mm	17mm	20mm	25mm
BS 3	-	-	-	-	-
BS 4	6mm	10mm	12mm	15mm	20mm
BS 5	-	7mm	9mm	15mm	20mm
BS 6	-	-	-	-	-

Table 3. Antibacterial activity of aqueous extract (*BS stands for bacterial species).

Zone of inhibition of chloroform extract:

Contraction	50mg/ml	100mg/ml	150mg/ml	200mg/ml	Amoxicillin
BS 1	-	-	13mm	15mm	19mm
BS 2	-	10mm	12mm	18mm	20mm
BS 3	-	-	-	-	-
BS 4	-	11mm	13mm	17mm	22mm
BS 5	-		9mm	15mm	18mm
BS 6	-	-	-	-	-

Table 4. Antibacterial activity of chloroformic extract

Antibacterial activity was done against 6 bacteria isolates from the vaginal sample in which bacterial sample no. 5 and bacterial sample no. 3 showed no zone of inhibition in both

aqueous and chloroform extracts. A larger inhibition zone was shown by sample no. 4 i.e., 20 mm at 200mg/ml in aqueous extracts and 18 mm at 200mg/ml in chloroform.

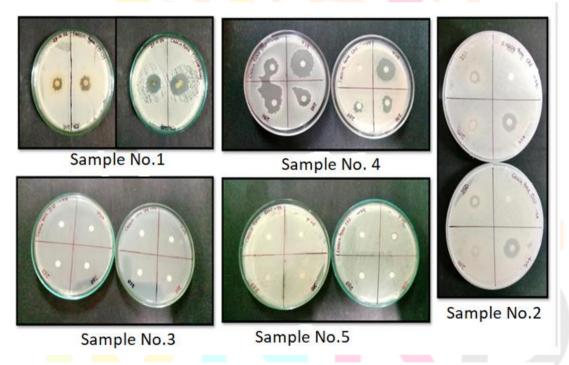


FIGURE 7. ANTIBACTERIAL ACTIVITY OF AQUEOUS AND CHLOROFORM EXTRACTS

MIC (minimum inhibitory concentration):

MIC activity was checked against those same bacteria for which antibacterial activity was checked. MIC was checked at different concentrations (200, 100, 50, 25, and $12.5\mu g/ml$). MIC value of aqueous extract showed better results than chloroform extract. The MIC values obtained from the cassia Tora plants exhibited antibacterial activity ranging between 50mg/ml to 200mg/ml. Aqueous extracts of cassia Tora gave MIC values of 50mg/ml, 100mg/ml, 12.5mg/ml, and 12.5mg/ml respectively for bacterial samples 1, 2, 4, and 6 respectively and chloroform extracts gave MIC values of 100mg/ml, 100mg/ml, 50mg/ml and 50mg/ml for bacterial sample no. 1, 2, 4 and 6 respectively. And the growth of bacterial sample no. 3 and 6 were not inhibited by both crude extracts.

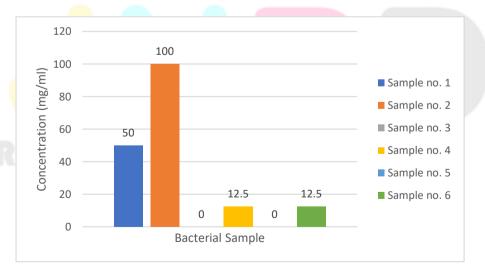
Bacterial sample	MIC Value (in
	mg/ml)
Sample no. 1	50
Sample no. 2	100
Sample no. 3	-
Sample no. 4	12.5
Sample no. 5	-
Sample no. 6	12.5

Table 5. MIC Value of the aqueous extract against bacterial samples

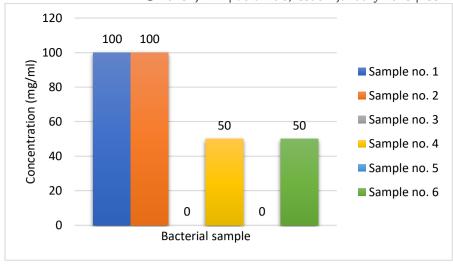
Minimum Inhibitory concentration of chloroform extract.

Bacterial sample	MIC Value
Sample no. 1	100
Sample no. 2	100
Sample no. 3	-
Sample no. 4	50
Sample no. 5	-
Sample no. 6	50

Table 6. MIC Value of chloroform extract against bacterial samples



GRAPH 1. MIC VALUE OF AQUEOUS EXTRACT



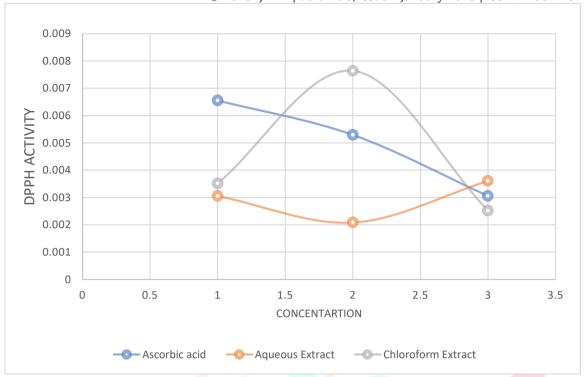
GRAPH 2. MIC VALUE OF CHLOROFORM EXTRACT

1.5 Antioxidant analysis

Antioxidant activity of aqueous extract showed better result than chloroform extract, curve of aqueous extract is closer to curve of ascorbic acid (standard). In the current study, aqueous extract of cassia tora leaves showed spotential free-radical scavenging activity, whereas chloroform extract showed less free-radical scavenging activity. The free radical scavenging property of the cassia tora plant may be one of the mechanisms by which it is effective in traditional medicines.

Cassia Tora	100μl	200μΙ	300μl
Control	0.862±0.00655		
Ascorbic Acid (280nm)	0.548±0.00655	0.678±0.00529	0.748±0.00305
DPPH Scavenging (%)	35.80	26.53	14.76
Aqueous Extract (517nm)	0.477±0.00305	0.620±0.00208	0.751±0.0036
DPPH Scavenging (%)	44.95	27.8	12.63
Chloroform Extract (517nm)	0.197±0.0035	0.309±0.0076	0.579±0.0025
DPPH Scavenging (%)	77.52	63.38	32.56

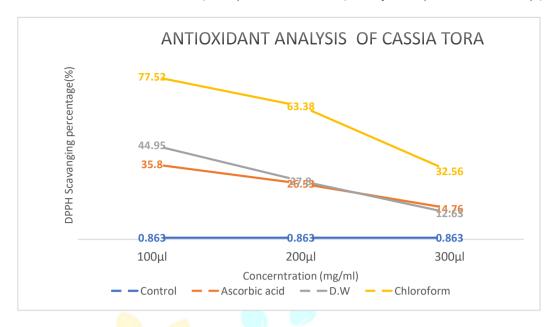
Table 7. Antioxidant analysis



Graph 3: Standard deviation graph of antioxidant analysis



FIGURE 8. ANTIOXIDANT ACTIVITY OF AQUEOUS AND CHLOROFORM EXTRACTS



GRAPH 4. ANTIOXIDANT ANALYSIS

1.6_Phytochemical analysis

The presence of various phytoconstituents in both solvent extracts of the *Cassia tora* plant was revealed by phytochemical analysis. Phytochemical analyses revealed differences in both extracts as listed in Table 8.

PHYTOCHEMICAL COMPOUNDS	PROCEDURE	OBSERVATION
Protein	In 2mL filtrate added 2 drops of Ninhydrin solution	A purple-coloured solution was obtained
Anthraquinones	10mg extract is dissolved in isopropyl alcohol and a drop of conc. ammonium hydroxide solution was added.	Red colour formed in solution after 2 min.
Terpenoids	In 2ml chloroform, 5mL plant extract was added then evaporated on water bath and 3mL conc. H2SO4 was added then boiled on water bath	Grey coloured solution formed.
Steroids	In 0.5mL plant extract, 2mL of acetic anhydride and 2mL conc. H2SO4 was added	Observed change in colour from violet to blue/green
Alkaloids	In 3mL extract solution few drops of iodine solution was added	A blue colour, which disappeared on boiling and reappeared on cooling
Flavonoids	In 1mL extract 2mL of 2% NaOH solution was added	An intense yellow colour, became colorless on addition of diluted acid
Saponins	5ml of extract was shaken vigorously to obtain forth and then mixed with 3 drops of olive oil	Formation of an emulsion was observed

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Tannin	In 1mL filtrated, 3mL distilled water and 3 drops 10% Ferric chloride solution were added	Blue-green colour obtained
Glycosides	In alcoholic extract was dissolved in 1mL of water and added few drops of aqueous NaOH solution	A yellow colour observed
Reducing Sugar	In 0.5mL filtrate, 0.5mL Benedict's reagent was added and boiled for 2 min.	Green/yellow/red colour obtained
Fats & Oils	Little quantity of plant extract was pressed in between to filter papers	Observed oil stain on the paper
Ligins	In extract solution, gallic acid was added	An olive-green colour obtained

 Table 8. Qualitative Tests for Phytochemical Screening

PHYTOCHEMICAL	AQUEOUS EXTRACT	CHLOROFORMIC
COMPOUNDS		EXTRACT
Anthraquinones	-	-
Terpenoids	+	+
Steroids	-	+
Alkaloids	+	
Flavonoids	+	-
Saponins	+	+
Tannins	+	+
Glycosides	+	-
Proteins	+	+
Reducing sugars	+	+
Fats & oils	+	+
Lignin	-	

Table 9. Phytochemical analysis



Discussion

FIGURE 9. PHYTOCHEMICAL ANALYSIS

Scientific research and studies on Cassia tora show that this plant has enormous biological potential. Clinical and pharmacological studies using standardized extracts and isolated phytochemical compounds are needed to explore the untapped potential of this plant. Cassia tora plants are of great benefit in the treatment of various ailments, including asthma, gastrointestinal symptoms, skin diseases, respiratory and urinary problems, vaginal infections, and liver and cardiovascular diseases. Cassia tora has great potential for study and could be used as a source of important phytochemical compounds. The phytochemical components of this plant have immense properties that can be used in the future in the pharmaceutical industry, in the production of cosmetics, and in commercial intimate cleansers. Future studies will focus on the development of new treatment strategies based on *Cassia tora's* innovative phytomedicinal approach. The main purpose of this study was to verify the antimicrobial activity of this plant against vaginal microflora isolates because in the future it will be possible to incorporate extracts from these plants into the formulation of commercial preparations for intimate hygiene. It is widely known that lactic acid bacteria inhibit the growth of pathogenic microorganisms through the production of different inhibiting substances (Arutcheva et al. 2001, Matu et al. 2010). This plant contains antioxidant properties that can be used as dietary supplements, to make cosmetic products such as anti-aging creams, lipsticks, face masks, etc. Dasari S. et al. (2014) isolated vaginal lactobacilli- producers of antimicrobial compounds, able to inhibit the growth of cervical pathogens (Dasari S. et al. 2014). Srivastava and co-workers, (2021) studied and stated the existence of numerous phytochemical compounds and therapeutic properties of Swertia chirayita plant. Further reports were also detailed by Srivastava et al., (2021) concerning the beneficial facets of Tinospora cordifolia. They swotted the phytochemistry as well as the pharmacological properties of Tinospora cordifolia. This research assisted us in comprehending the significance of traditional herbal medicine in the treatment of various women's genital diseases may be a better alternative to antibiotics as they shorten the duration of treatment, as this plant has antimicrobial properties and may be able to both target infections

and restore microflora growth. Arunima and co-worker have noticed that traditional medicinal plants like fennel and clove shows antibacterial activities and had checked the effects of fennel seed oil and clove oil against bacteria and fungi. The results showed that fennel seed oil and clove oil were more effective against bacteria in comparison with fungi [Verma S , Karkun A, Siddiqui HN (2015)].

Conclusion

The plant used for this study showed different types of phytochemical compounds that have a wide range of biological and pharmacological properties. These secondary metabolites could help females maintain intimate hygiene. Phytochemical screening of this plant revealed the presence of saponins, phenols, terpenoids, tannins, and flavonoids, which can be incorporated into the formulation of commercial preparations for intimate hygiene or creams. Phytochemicals such as saponins serve as fungicidal, anti-inflammatory, and spermicidal substances, phenolic compounds have antibacterial properties, and tannins help to tone the vagina. Antibiotics have a direct effect on the target pathogen as well as the microflora. On the other hand, medicinal herbal plants may be able to both target infections and restore microflora growth. Consequently, combination therapy, which is the interaction between medicinal plants with antimicrobial potential and antibiotics, may be a useful option to combat antibiotic-resistant microbes without destroying the normal flora. In the human body, inhaled oxygen is converted to reactive oxygen species, which include hydrogen peroxide, hydroxyl radical, superoxide anion radical, nitric oxide radical, singlet oxygen, hypochlorite radical, and other lipid peroxides. By interacting with membrane lipids, nucleic acids, proteins and enzymes, and other small molecules, they are all capable of causing cell damage. Antioxidants must be added to the dietary system as a defense mechanism against reactive oxygen species.

Future perspectives

Antioxidant supplementation is increasingly recognized as an essential technique for boosting free radical defense as part of a healthy lifestyle and a well-balanced, healthful diet. Antioxidant properties of the *Cassia tora* plant have a wide range of biological effects, including anti-inflammatory, anti-aging, anti-cancer, and detoxification properties. The properties of these plants can be incorporated into dietary supplements and taken in the form of detox water which helps in purification and skin glowing. And can be used in commercial cosmetics production like anti-aging creams, lipstick, face sheet masks, etc. This work will be helpful for the researcher for the comparative study of the *Cassia tora* plant with various other herbal medicinal plants.

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