Full Length Research Article

© 2024, Centre for Advanced Research in Agricultural Sciences
Research Journal of Agricultural Sciences
Volume 15; Issue 05 (Sept–Oct 2024); pp 1196–1203

ISSN: 0976-1675 (P)
ISSN: 2249-4538 (E)

Phytochemical Screening, Thin layer Chromatographic Profiling and Antimicrobial Activities of *Hedychium coronarium*

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Received: 29 Aug 2024; Revised accepted: 14 Oct 2024

Abstract

The present study was aimed to preliminary phytochemical screening, thin layer chromatographic profiling and antimicrobial activities of *Hedychium coronarium*. Phytochemical screening was performed to investigate bioactive constituents of dried flowers by utilizing various solvent system (petroleum ether, dichloromethane, ethyl acetate, methanol and water) to confirm the presence of alkaloid, flavonoids, saponins and glycosides etc. Total phenol and flavonoid content were examined spectrophotometrically. The crude extracts (dichloromethane, ethyl acetate, methanol and aqueous) which shows good phytochemical result was subjected to TLC profiling and separation of spots were observed under normal day light, shorter wavelength (254nm) and longer wavelength (365nm) of light by using different solvent systems with Rf values compared to standard drugs of gallic acid (0.28) and quercetin (0.50). The antimicrobial activity was determined by well diffusion method along with standard antibiotic against selected bacterial and fungal strains. In preliminary phytochemical screening methanol and aqueous extracts of flower contain higher bioactive constituents than other solvent extract. Total phenol and flavonoid contents were ranged from (0 to 1.57±0.01) mg GAE/g and (1.76±0.02) to (4.13±0.01) mg QE/g. Among all the solvent extracts, dichloromethane, methanol and aqueous extracts showed well separation of spots. The retardation factor was found to be 0.22 and 0.50 to 0.62 which is similar to standard gallic acid and quercetin respectively. The methanol extract demonstrated effective antimicrobial activity against the tested pathogenic species with minimum inhibitory concentrations values ranging from 25-100 mg/mL for bacteria and fungi respectively. Methanolic extract of *Hedychium coronarium* flower exhibit the presence of various phytoconstituents and display significant antimicrobial activity.

***Key words*:** *Hedychium coronarium*, Phytochemical screening, TPC, TFC, Thin layer chromatography, Antimicrobial activity

Herbal medicines are in great demand for primary healthcare due to their vast medical and biological importance as well as their higher safety margins [1]. *Hedychium coronarium* has the greatest reservoirs of drugs of traditional systems of medicine [2]. *Hedychium coronarium J. Koening* (family Zingiberaceae) the white ginger lily or Dolanchampa or kapur kachri is a perennial, erect branched annual weed of 3-6 feet height. Traditionally various part of this plant was used in treatment of irregular menstruation, piles bleeding, stone in urinary tract, cancer, eye problems, diabetes headache, lancinating pain, contusion, inflammation, hypertensive disease and rheumatic pain [3-5]. Rhizome extract of *Hedychium coronarium* possess better antifungal property than standard drug such as nystatin and griseofulvin [6]. The rhizome extract of *Hedychium coronarium* has indeed been reported to exhibit strong antifungal properties, often surpassing standard antifungal drugs like nystatin and griseofulvin in effectiveness. This can be attributed to the presence of potent bioactive compounds, such as flavonoids, terpenoids, and essential oils, particularly compounds like coronarin D, which have been identified for their antifungal activity. The aroma ingredients of this plant include 13 compounds, some of them are α-pinene; ocimene; 1,8-cineole; L-linalool; caryophyllene and farnesene [7]. *Hedychium coronarium J. Koening* (family Zingiberaceae), the white ginger lily or Dolanchampa or kapur kachri is an erect branched perennial aromatic rhizomatous plant of 3-6 feet height. The genus Hedychium firstly described by Jhon Koenig in 1783 include about 47 genus and 1400 species and is mainly distributed in tropical as well as subtropical regions [8-9]. In Central region of India, it is one of the endemic species of Amarkantak region of Madhya Pradesh and Chhattisgarh. In the Amarkantak region *Hedychium coronarium* flower extract is known as "Gulbakawali Ark" which is world famous as an eye tonic and prevents "Motia bind" (Cataract) [10]. The therapeutic potential of *Hedychium coronarium* is attributed to its bioactive compounds, including essential oils, flavonoids, and phenolic compounds, which possess anti-inflammatory, antioxidant, and antimicrobial properties. These properties could help reduce oxidative stress and inflammation in the eye, which are factors in cataract formation.

Phytochemicals are non- nutritive bioactive compounds naturally synthesized by plant for diseases protection and they

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Citation: Pandey S, Awasthi A. 2024. Phytochemical screening, thin layer chromatographic profiling and antimicrobial activities of *Hedychium coronarium*. *Res. Jr. Agril. Sci*. 15(5): 1196-1203.

can be used for treatment of many diseases [11]. These bioactive compounds include alkaloid, flavonoid, diterpenes, carbohydrate, phenols and tannins [12]. Due to the presence of several bioactive compounds, Plant products have been used in formation of phytomedicine since several years. These compounds have therapeutic properties like anti-inflammatory, antioxidant, antimicrobial, and anticancer effects. The use of plant-based medicines is rooted in traditional systems, such as Ayurveda and Traditional Chinese Medicine, and continues to play a key role in modern drug discovery and development. Advances in biotechnology and phytochemistry have also expanded the scope of phytomedicines by enabling the identification and extraction of these compounds with higher precision and efficacy. The present study was aimed to phytochemical screening, TLC profiling and antimicrobial analysis of *Hedychium coronarium* flower in different solvent system such as petroleum ether, dichloromethane, ethyl acetate, methanol and water.

MATERIALS AND METHODS

Collection of plant material

The plant material used in this study was collected from Amarkantak region, Anuppur District (23.1°N, 81.68°E) of Madhya Pradesh, India. The Plant was authenticated by plant identification cell, department of botany, Gurughasidas university Bilaspur (Chhattisgarh) using standard reference with a voucher specimen No. Bot/GGV/2023/58. Collected plant parts were properly clean from any foreign organic matter, dried under shade and powdered. Plant sample was stored at 4° C until needed for the various analysis.

Chemical and reagents

All solvents and chemicals used for this study were of analytical grade and purchased from Merk and Himedia lab. Pvt. Limited. Silica gel 60 F-254 (Merk) was used for preparation of TLC plate.

Preparation of plant extract

The dried flowers (54 g) sample was extracted separately in a Soxhlet apparatus with 200 ml of solvents such as petroleum ether, dichloromethane, ethyl acetate, methanol and distilled water successively for 24 hours each. The crude extracts were concentrated separately using rotary evaporator at 40°C. After solvent evaporation, each solvent extracts were stored at 4°C for extraction yield, phytochemical analysis, TLC profiling and antimicrobial analysis.

Determination of extraction yield (% yield)

Percentage yield is defined as quantity of plant extracts recovered in mass after solvent extraction when compared with the initial quantity of plant samples. Following formula was adopted for determination of percentage yield of selected plant materials:

$$\text{Percentage yield} = \frac{\text{Weight of extract } (W_1) \times 100}{\text{Weight of powdered plant sample taken } (W_2)}$$

Phytochemical screening

Flower extracts were tested for the presence of bioactive constituents like carbohydrate, protein, flavonoids, alkaloids, saponins, diterpenes and tannins by using standard methods [13-19].

Test for protein

Biuret's test: 2 - 3 ml of crude extracts was mixed with 1 ml of 40% NaOH solutions and 2 drops of 1% $CuSO_4$ solution. Purplish - violet or pinkish - violet color appeared that indicate presence of protein.

Xanthoproteic test: Few drops of concentrated HNO_3 were added in small amount of extract. After few minutes, it forms white colour precipitate which turns to yellow colour on heating. after sequential cooling, 20% NaOH solution was added to extract. The orange colour observed in test solution confirmed aromatic amino acid in plant sample.

Test for carbohydrates

Molisch's test: 2 ml of crude Plant extracts was mixed with two drops of alcoholic solution (20%) of α- naphthol followed by addition of 2 ml of concentrated H_2SO_4 along the sides of the test tube. Appearance of a purple colour ring formed at the intersection between two liquids indicated presence of carbohydrate.

Fehling's test (Reducing sugars): 2 ml of crude extract was mixed with equal volume of Fehling's solution A and B then boiled the solution for few minutes in water bath. Formation of brick red coloured precipitate indicated the presence of reducing sugars.

Test for alkaloids

50 mg of extract was dissolved in 5 ml of dilute HCl and then filtered. The filtrate was used for the detection of alkaloid using various reagents-

Dragendorff's test

To 2-3 ml of test solution was mixed with 1ml of Dragendorff's reagent (potassium bismuth iodide solution). Formation of orange or orange reddish precipitates indicated the presence of alkaloids in sample.

Hager's test: To 2-3 ml of the plant extract filtrate, few drops of Hager's reagent (solution of picric acid) was added. Formation of yellow precipitates indicated the presence of alkaloids in sample.

Wagner's test: 2-3 ml of the plant extract filtrate was mixed with few drops of Wagner's reagent (iodine potassium iodide solution). Formation of reddish-brown precipitate confirmed the presence of alkaloids in sample.

Test for saponins

Foam test: 0.5gm of crude extract was dissolved with 20 ml distilled water and shaken vigorously for 15 min. formation of foam to a length of 1 cm indicates the presence of saponin.

Test for flavonoid

Lead acetate test: Small amount of plant extract was mixed with few drops of 10% Lead acetate solution. Formation of intense yellow precipitates indicated the presence of flavonoid in plant sample.

Alkaline reagent test: Small amount of Plant extract was dissolved in 2ml of 2% NaOH solution. An intense yellow colour was produced which turns colourless on adding few drops of diluted acid indicates presence of flavonoids.

Test for phenolics

Ferric chloride test: 3-4 drops of 5% ferric chloride solution were mixed with 1ml of extract. Appearance of blue colour indicates the presence of hydrolysable tannins, while the green colour indicates the presence of condensed tannins.

Test for diterpene

Copper acetate test: 50 mg of crude extracts was dissolved in 2ml of water and 3-4 drops of copper acetate solution. Formation of emerald green colour indicates the presence of diterpenes.

Determination of total phenolic content

Total phenolic content was determined spectrophotometrically using Folin-Ciocalteu's method with some modified [20]. Briefly, 2ml each of various crude extracts (1 mg/mL) and standard gallic acid (10– 50µg/mL) was dispensed in different test tube and was mixed with 1 ml of Folin-Ciocalteu reagent (1:10 v/v diluted with distilled water) followed by 1 ml Na_2CO_3 (1 M). Then, mixture was vortexed for 15s and allowed to stand 10 min in the dark for colour development. After incubation, the absorbance was measured at 765 nm using a spectrophotometer (Epoch2, BioTek, Instruments, Inc., USA). The phenol content was extrapolated from gallic acid standard/calibration graph equation:

$$y = 0.014x - 0.013, R^2 = 0.999$$

Where, X is the Gallic acid equivalent (GAE) and Y is the absorbance

TPC was calculated using following formula:

$$C = c \times V/m$$

Where, C was total phenol content in mg/g plant extract in mg GAE/g extract, c was the concentration of gallic acid from calibration curve in mg/mL, V was the volume of extract (mL) and m was the weight of extract used in the assay in g.

Determination of total flavonoid content

Total flavonoid content of crude extracts was determined by using aluminium chloride colorimetric method [21]. This method is based on the quantification of the yellow-orange colour produced by the interaction of flavonoid with $AlCl_3$. Briefly, 3 ml each of extracts (1mg/mL) and standard quercetin (5–25 µg/mL) was dispensed in separate test tube and was mixed with 1ml of 2% $AlCl_3$. The mixture was allowed to stand for 15 min. Absorbance was measured at 420 nm with a UV-visible spectrophotometer (Epoch2, BioTek, Instruments, Inc., USA). Flavonoid content was calculated using a quercetin calibration curve equation:

$$y = 0.036x + 0.015, R^2 = 0.999$$

Where, X is the Quercetin equivalent (QE) and Y is the absorbance

Results were expressed as mg of quercetin equivalent (QE)/g using the formula CV/m in the same manner as described for total phenol.

Thin layer chromatography analysis

Qualitative Detection of gallic acid and Quercetin in all extracts of *Hedychium coronarium* was done by standard TLC method [22]. Various flower extracts of *Hedychium coronarium* was subjected to thin layer chromatography (TLC) as per conventional one-dimensional ascending method using silica gel 60F254, 7X6 cm (Merck). The test sample (1µl) and standard test were loaded on base line at 1.0 cm interval at 5 tracks and then allowed to dry at room temperature. After saturation with the solvent vapour for 30 minutes, the TLC plate loaded with test sample and the reference. TLC plates was kept in a TLC twin trough developing chamber with Mixture of solvent system containing toluene: ethyl acetate: formic acid (5:4:1 V/V) for Quercetin and toluene: ethyl acetate: formic acid (7:5:1 V/V) for gallic acid. The developed plates were dried and kept in a photo-documentation chamber. Images of the TLC plates were captured in normal light, short UV light (254nm), and long UV light (365nm). Then, the numbers of spotted were noted and Rf values were calculated.

Antimicrobial activity

The bacterial and fungal pathogen used in this study were selected on the basis of their role as opportunistic pathogens of humans and animals and their association with eye infections.

Microorganism strains

The selected bacterial strains were *Escherichia coli* (MTCC- 40), *P. aeruginosa* (MTCC-1688) and *K. pneumoniae* (MTCC- 432). The fungal strains used for this investigation were *Candida albicans* (MTCC-183) and *Aspergillus niger* (MTCC-281).

Bacterial inoculum preparation

The test bacterial strains that were originally maintained on nutrient agar slants were recovered in sterile nutrient broth media and incubated overnight at 37°C. In order to obtain distinct colonies, the 24 h old cultures were diluted 1:100 v/v in fresh sterile nutrient broth and cultured on nutrient agar medium overnight at 37°C. For the fungal inoculum preparation, fungal strains were freshly sub-cultured on potato dextrose Agar media and incubated at 25°C for 72 h.

Well dilution assay

In this study, well diffusion method was used to determine the antimicrobial activity of extract using standard procedure [23]. The nutrient Agar medium and potato dextrose agar medium were prepared for antibacterial and antifungal screening. The medium was autoclaved at 121°C for 15 min and allowed to cool to 50°C in a water bath. After sterilization, the media in flask was immediately poured (20 ml/ plate) into sterile Petri dishes on plane surface and allowed to cool and solidify. About 10 mL each from both the bacterial and fungal inoculum was delivered individually on the solidified agar surfaces to give the desired final inoculum of 1×10^4 CFU/spot. The extracts concentrations for the antibacterial and antifungal evaluation ranged from (25-100) mg/ml. The ciprofloxacin concentration ranged from (0.01–0.03) mg/ml, while fluconazole (antifungal standard) was also ranged from (0.01–0.03) mg/ml. Bacteria plates were incubated at 37°C for 16–24 h while fungi plates were incubated at 30°C for (48–72) h. The minimum inhibitory concentrations (MICs) were determined as the lowest concentration of extracts inhibiting the visible growth of each organism on the agar plate.

Extract preparation

A 100 mg/mL stock solution prepared in a little amount of DMSO and made up with either nutrient broth or potato dextrose broth for anti-bacterial and anti-fungal activity respectively. Three concentrations of extracts i.e. 25, 50 and 100 mg/ml were used in this study. Standard antibiotics (ciprofloxacin and Fluconazole for antibacterial and antifungal standard respectively) were also prepared.

RESULTS AND DISCUSSION

Percentage of yield extract

The yield of sequential extracts (g) is shown in (Table 1). The percentage yield of crude extract obtained from petroleum ether, dichloromethane, ethyl acetate, methanol and water were ranged from 0.911% to 30.948%. The wide range in yield (0.911% to 30.948%) indicates that different bioactive compounds present in the plant material are more soluble in certain solvents, affecting the extraction efficiency.

Table 1 Results of percentage yield (%) of *Hedychium coronarium* flower extract

S. No.	Name of extract	Colour of the extract	Percentage yield (%w/w)
1	Petroleum ether	Brown	0.911
2	Dichloromethane	Black	4.024
3	Ethyl acetate	Brown	1.944
4	Methanol	Brown	1.035
5	Distilled water	Black	30.948

Phytochemicals screening

In the present study, qualitatively phytochemical investigation of flower extracts of *Hedychium coronarium* reveals the presence of various bioactive constituents and the results are presented in (Table 2). In this screening process alkaloids, carbohydrate, saponins, phenol, flavonoid, proteins and diterpenes shows different types of results in different solvents extracts. Among these phytochemical screening, alkaloids, carbohydrate, saponins, flavonoid, proteins and diterpenes, were present in methanolic solvent extracts whereas carbohydrate, saponins, phenol, flavonoid and diterpenes were present in aqueous extract. Phenol was only seen in aqueous extract and flavonoid are present in its dichloromethane, methanol and aqueous extracts.

Table 2 Phytochemical screening of *Hedychium coronarium* flower extracts

S. No.	Phyto-constituents	Petroleum ether	Dichloromethane	Ethyl acetate	Methanol	Aqueous
1.	Alkaloid	-	+ ve	-	+ ve	-
2.	Carbohydrate	+ve	-	+ ve	+ ve	+ ve
3.	Saponins	-	-	-	+ ve	+ ve
4.	Phenol	-	-	-	-	+ ve
5.	Flavonoid	-	+ ve	-	+ ve	+ ve
6.	Proteins	-	-	-	+ ve	-
7.	Diterpenes	+ ve	-	-	+ ve	+ ve

- Absent; + ve Presence

Total phenolic content (TPC)

The total phenol concentrations in flower extracts of *Hedychium coronarium* are shown in Table 3. The results showed that the content of total phenols in extracts, expressed as gallic acid equivalents (GAE)/g dry weight (DW) of plant. among all extracts, phenolic content was only recorded in aqueous extract *Hedychium coronarium* flower (1.57 mg GAE/g). Pharmacists usually targets the plant with high phenolic content to treat different diseases [24]. High amount of phenolic content indicates the ability of plant to treat inflammatory diseases and can be implicated in wound healing. Phenolic compounds, which include flavonoids, tannins, and phenolic acids, possess significant antioxidant, anti-inflammatory, and antimicrobial properties.

Table 3 Total phenol content and total flavonoid content of various solvent extracts of *Hedychium coronarium* (mean ± SD)

S. No.	Solvent extract	Total phenol (mg GAE/g)	Total flavonoids (mg QE/g)
1.	Dichloromethane	-	1.76 ± 0.02
2.	Ethyl acetate	-	-
3.	Methanol	-	4.13 ± 0.01
4.	Aqueous	1.57 ± 0.01	2.97 ± 0.01

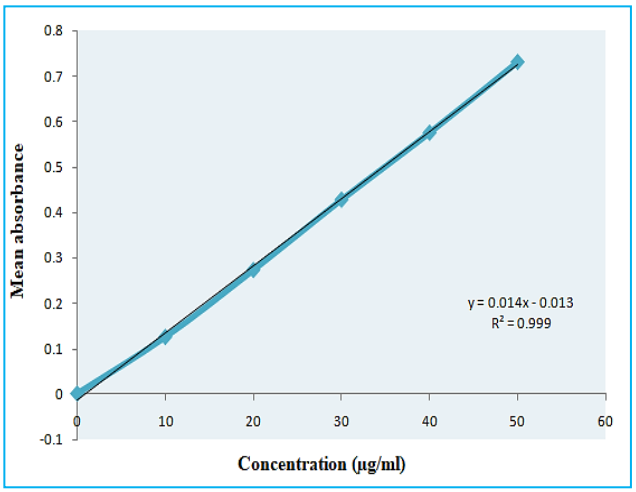

Fig 1 Standard curve of gallic acid

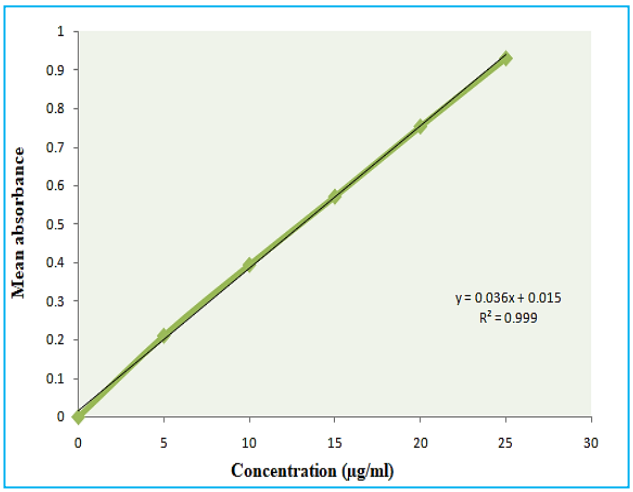

Fig 2 Standard curve of quercetin

Total flavonoid content (TFC)

TFC was calculated as quercetin equivalents (QE) (y = 0.036x + 0.015, R^2 = 0.999) as shown in (Table 3). Flavonoids are important because of their ability to inhibit enzymes, anti-inflammatory activity and antimicrobial activity. The difference in TFC among studied extracts varied significantly, ranging from 1.76 mg QE/g of sample for dichloromethane extracts and 4.13mg QE/g of sample for methanol extract.

Table 4 TLC profiling of flavonoid content in flower extract of *Hedychium coronarium*

Type of extract	Solvent system and its ratio	Wave length (nm)	Spot	Rf Values
Quercetin	toluene: ethyl acetate: formic acid (5:4:1)	365nm (Longer Wavelength)	1	0.50
		254nm (Shorter Wave length)	1	0.50
		Visible Light	1	0.50
Dichloro-methane	toluene: ethyl acetate: formic acid (5:4:1)	365nm (Longer Wavelength)	3	0.62, 0.84, 0.96
		254nm (Shorter Wavelength)	3	0.56, 0.68, 0.92
		Visible Light	2	0.62, 0.98
Methanol	toluene: ethyl acetate: formic acid (5:4:1)	365nm (Longer Wave length)	1	0.92
		254nm (Shorter Wave length)	2	0.50 0.80
		Visible Light	0	0

Thin layer chromatographic studies

Thin layer chromatographic analysis provides a chromatographic drug fingerprint. The solvents system used in this study are toluene: ethyl acetate: formic acid (5:4:1) for Quercetin (flavonoid) and toluene: ethyl acetate: formic acid (7:5:1) for gallic acid (phenol) [25]. The Thin layer chromatographic (TLC) study showed that among five solvent (petroleum ether, dichloromethane, ethyl acetate, methanol and aqueous) that are used for extraction, dichloromethane and methanol extracted higher metabolites of medicinal important from flower of *Hedychium coronarium*. The Number of spots and corresponding Rf value of various secondary metabolites were recorded in (Table 4-5). The corresponding TLC is presented in (Fig 4-5).

Table 5 Result of TLC fingerprinting of phenolic content in flower extract of *Hedychium coronarium*

Type of extract	Solvent system and its ratio	Wave length (nm)	Spot	Rf Values
Gallic acid	toluene: ethyl acetate: formic acid (7:5:1)	365nm (Longer Wave length)	1	0.28
		254nm (Shorter Wave length)	1	0.28
		Visible light	1	0.28
Aqueous	toluene: ethyl acetate: formic acid (7:5:1)	365nm (Longer Wave length)	1	0.22
		254nm (Shorter Wave length)	0	0
		Visible light	0	0

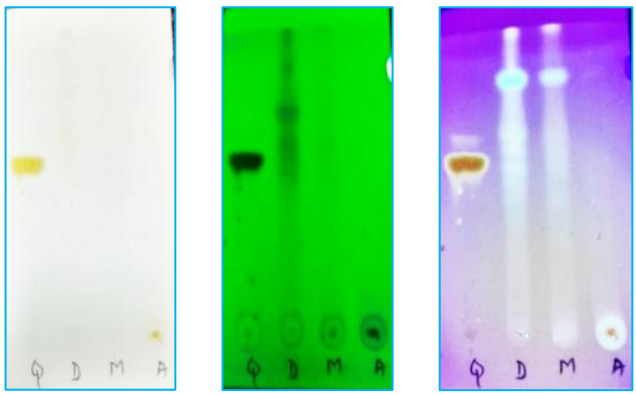

Fig 4 TLC profiling of flavonoid content in dichloroacetate, methanol and aqueous extract of *H. coronarium* flower

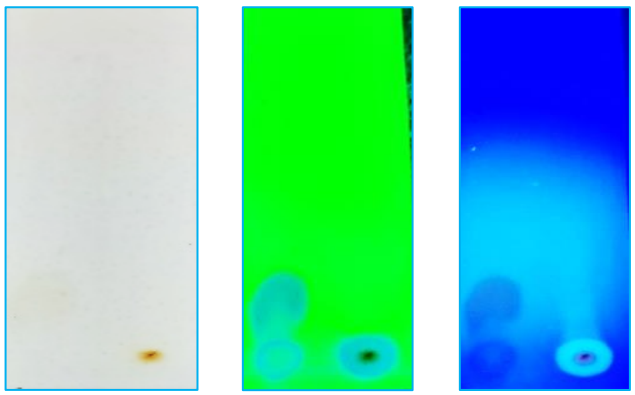

Fig 5 TLC profiling of phenol content in aqueous extract of *H. coronarium* flower

Antimicrobial activities

The plant extracts showed varying degree of antimicrobial potential due to different chemical composition. In general methanol extract of flower inhibited the growth of bacterial and fungal strains more efficiently than other extracts. Data revealed that maximum inhibition was seen in the growth of *P. aeruginosa* by methanolic extract (50 mg/ml and 100 mg/ml concentration). Methanolic extract was also effectively inhibited the growth of *Escherichia coli. Klebsiella pneumoniae*, *Candida albicans* and *Aspergillus niger*. The antibacterial and antifungal standard used in this study was ciprofloxacin (a broad-spectrum antibacterial drug) and fluconazole respectively. They showed higher inhibitory properties towards all the studied bacterial and fungal species.

The inhibitory activity of all methanolic extract (25, 50 and 100 mg/ mL) varied against the tested bacterial isolates *E. coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* with DIZ values ranging from 6.0 to 25 ± 0.47 mm whereas the inhibitory activity against fungal isolates i.e. *Candida albicans* and *Aspergillus niger* with DIZ value ranging from 6.0 to 22±0.47 mm as indicated in (Fig 6-7). However, other flower extracts (petroleum ether, dichloromethane, ethyl acetate and aqueous extracts) did not show significant zone of inhibition.

The antimicrobial susceptibility pattern of bacterial and fungal isolates was shown in (Table 6-7). The antibiotic ciprofloxacin exhibited the maximum zone of inhibition against the strain *Escherichia coli* (30±0.47 mm) and the minimum against the strain *Pseudomonas aeruginosa* (18±0.47 mm) even

at a lower concentration (10-30 µg/mL) compared to the flower extracts. Diameter of inhibitory zone (DIZ) of *Hedychium coronarium* flower extracts towards the studied bacterial species were increased with the concentrations. However, the DIZ of *Hedychium coronarium* flower extracts towards the studied fungal strains had no significant variation.

Hedychium coronarium exhibited the maximum zone of inhibition against *Pseudomonas aeruginosa* (25 ± 0.47) and the minimum against *Escherichia coli* (6.0 mm) bacterial isolates. On the contrary, Fluconazole antifungal compound exhibited the maximum zone of inhibition against the strain *Candida albicans* (32±0 mm) and the minimum against the strain *Aspergillus niger* (8±0.74 mm) even at their lower concentration (10-30 µg/mL) compared to the flower extracts. Candida albicans and *Aspergillus niger* both have same zone of inhibition (22 ± 0.47 mm) with methanolic extract of *Hedychium coronarium.* Since *Hedychium coronarium* extracts exhibited maximum antibacterial activity against *Pseudomonas aeruginosa* and *K. pneumoniae* than *E. coli* although maximum antifungal activity was seen in both tested fungal isolates.

Table 6 Zone of inhibition affected by *H. coronarium* flower extract using the well diffusion method

S. No.	Plant extract [a] / Standard	Zone of inhibition (DIZ)/(mm)			
		Conc.	*Escherichia coli*	*Pseudomonas aeruginosa*[b]	*Klebsiella pneumoniae*[c]
1.	Ciprofloxacin (µg/ml)	10	22±0.47	18±0.47	23±0.47
		20	26±0.47	25±0.47	26±0.47
		30	30±0.47	27±0.47	28±0.47
3.	Methanol (mg/ml)	25	06 ± 0	15 ± 0.47	10 ± 0.47
		50	08± 0	20 ± 0.47	13 ± 0.47
		100	12±0.47	25 ± 0.47	16 ± 0.47

a- The flower extract which did not show antibacterial and antifungal activity were excluded from above table. Test was performed in triplicate and data show the mean ±=standard error.

b- *Pseudomonas aeruginosa* *c- Klebsiella pneumoniae* DIZ- Diameter of inhibitory zone

Table 7 Zone of inhibition affected by *H. coronarium* flower extract

S. No.	Plant extract [a] / Standard	Zone of inhibition (DIZ)/(mm)		
		Conc.	*Candida albicans* [d]	*Aspergillus niger* [e]
2.	Fluconazole (µg/ml)	10	26±0.47	8±0.74
		20	30±0.47	10±0.5
		30	32±0	14±0.5
3.	Methanol (mg/ml)	25	12± 0.47	06 ± 0
		50	16 ± 0.47	22 ± 0.47
		100	22 ± 0.47	15 ± 0

a- The flower extract which did not show antibacterial and antifungal activity were excluded from above table. Test was performed in triplicate and data show the mean ±=standard error.

d- Candida albicans *e- Aspergillus niger* DIZ- Diameter of inhibitory zone

Fig 6 Images of antimicrobial activity of standard drug Ciprofloxacin and Fluconazole against selected bacterial and fungal strains

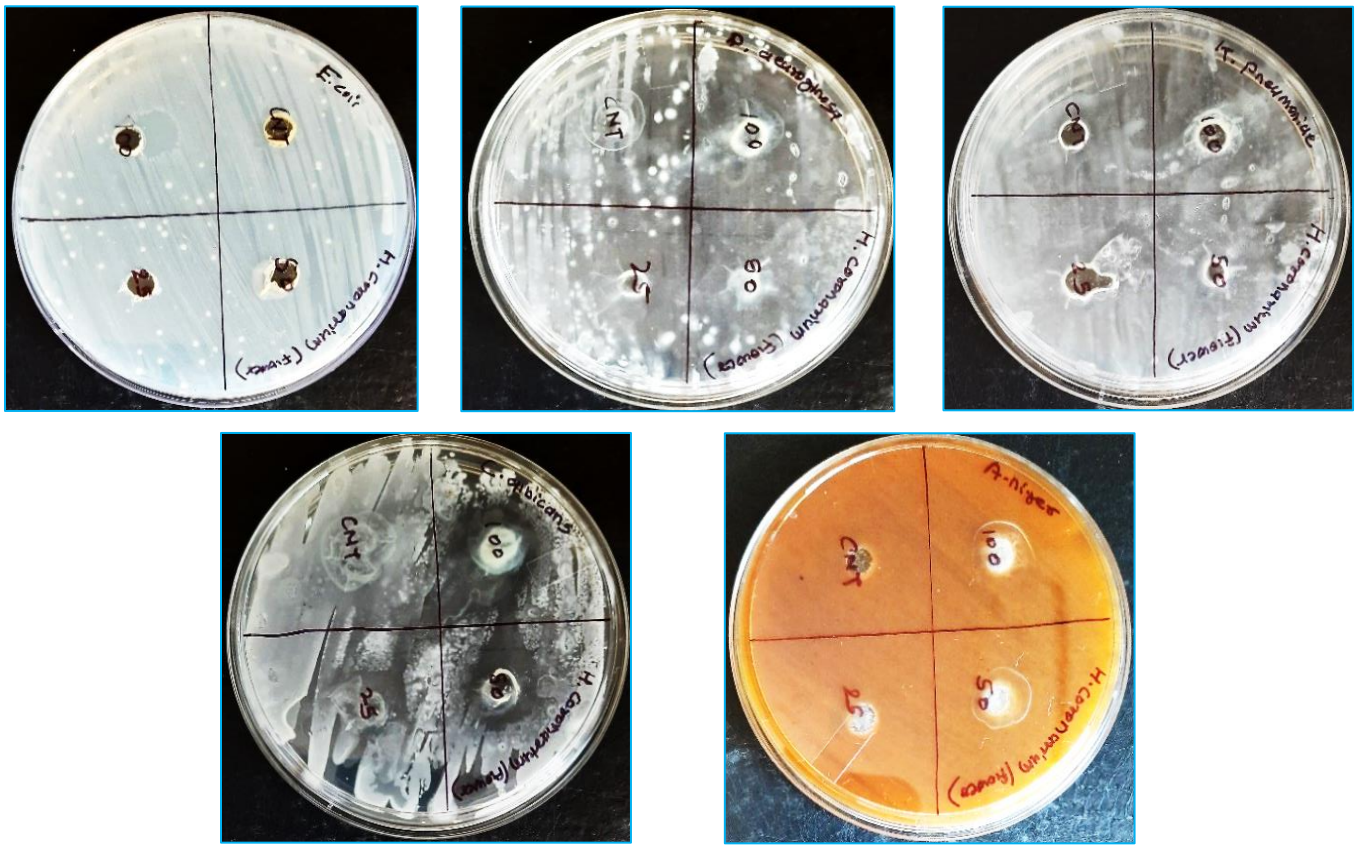

Fig 7 Agar plate for antimicrobial activity of *H. coronarium* (Flower) extract with concentration 25 mg/ml, 50 mg/ml and 100mg/ml against selected bacterial and fungal strains

CONCLUSION

The present study revealed that this medicinally important plant *Hedychium coronarium* has potential to act as a source of useful drugs and also to improve the health status of the consumers due to the presence of various phytoconstituents confirmed by various conducted assays such as Total phenolic content and Total flavonoid content (TPC and TFC). The protective activity of this plant could be attributed to the presence of saponins, diterpenes, flavonoids, phenols, and alkaloids, as confirmed by the preliminary phytochemical screening of different solvent extracts. Particularly, methanolic extract of flower showed more dominant activities in the assays conducted compared to other solvent extracts. The TLC profiling showed most phyto-compounds such as gallic acid and quercetin were found in dichloromethane, methanol and aqueous extracts that confirming to be a better supplement for nutrients. The methanol extract showed significantly antimicrobial activity than other solvent extracts. The highest bacterial and fungal inhibitory activity was observed in *H. coronarium* towards *Pseudomonas aeruginosa*, *Candida albicans* and *Aspergillus niger* respectively. Therefore, *Hedychium coronarium* flower can be explored as a promising source of phytocompounds and antimicrobial compounds in pharmaceutical industry. The crude extract of *Hedychium coronarium* can provide lead molecules which could be useful substrate for the synthesis of new drugs for the treatment of eye diseases, hepatic diseases and kidney related diseases. Further purification, identification and characterization of the active compounds would be our priority in future studies.

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