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Phytochemical screening and antimicrobial activity of stem extract of *Coleus forskohlii* L. collected from Al-Baha Area, Saudi Arabia

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Abstract

This study aimed to investigate the phytochemical screening, total phenolic content, antioxidant and antimicrobial activities of *Coleus forskohlii* L. stem extract in Al-Baha area, Saudi Arabia. Stem samples were collected from Al-Baha area and air-dried followed by extraction with ethanol, petroleum ether, chloroform, ethyl acetate, and n-butanol. The extracts were then subjected to phytochemical screening, determination of total phenolic content, antioxidant and antimicrobial activities. Results showed the presence of flavonoids, alkaloids, tannins, terpenoids, steroids, saponins, and reducing sugars. Total phenolic content was significantly ($P<0.001$) higher in n-butanol extract (274.33±3.29 mg GAE/gm), followed by ethyl acetate extract (182.94±1.82 mg GAE/gm), ethanol extract (79.63±2.02 mg GAE/gm) and petroleum ether extract (73.38±3.07 mg GAE/gm), while the lowest content was in chloroform extract (60.06±2.12 mg GAE/gm). The antioxidant activity was significantly ($P<0.001$) higher in n-butanol extract (67.68±1.55%), followed by ethyl acetate extract (43.38±1.27%), ethanol extract (36.02±1.29%), petroleum ether extract (20.71±0.59%) and chloroform extract (19.73±0.74%). The antimicrobial activity showed that all microorganisms tested were resistant at the concentration of 25 and 50 mg/ml of plant extracts, whereas the concentrations of 100, 150 and 200 mg/ml showed varying activities against gram-negative (*Escherichia coli, Pseudomonas aeruginosa*), gram-positive (*Staphylococcus aureus, Bacillus cereus*) and *Candida albicans*. The study concluded that the stem extracts of *C. forskohlli* have promising pharmacological and biological activities that could be beneficial in pharmaceutical as well as food and medicinal industries.

Keywords: *Coleus forskohlii*; Stem extract; Phytochemical screening; Antimicrobial activity

1. Introduction

There is a growing interest in using natural antibacterial compounds such as plant extracts of herbs and spices for the preservation of foods because these extracts possess a characteristic flavour and sometimes show antioxidant and antimicrobial activities [1]. Plants are the major source of medicines and foods which play a vital role in the conservation of human health. The importance of plants in medicine is of significance with the current global trends to obtain drugs from plant sources [2]. Medicinal plants are important for pharmacological research and drug development, not only when constituents are used directly as therapeutic agents, but also as starting materials for the synthesis of drugs or as models for pharmacologically active compounds [3]. Medicinal plants and plant-based products that contain a wide variety of free radical scavenging molecules such as phenolic compounds, nitrogen compounds, vitamins, terpenoids, and some other endogenous metabolites are rich in antioxidant activity [4].

Nearly ninety percent of the world's population relies on conventional as well as traditional medicines for their primary health care, the majority of which involves the use of plant extract drugs and secondary metabolites [5]. *C. forskohlii* has been used for treating heart diseases, respiratory disorder, insomnia, epilepsy, bronchitis, burning sensation,

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constipation, intestinal disorder, and angina, and this plant constitutes thousands of natural bioactive compounds such as phenolics, tannins, terpenoids, alkaloids, saponins, that may produce health beneficial effect by scavenging free radicals [5]. With the advancement in science and technology, remarkable progress has been made in the field of medicine with the discoveries of many natural and synthetic drugs, and antibiotics are indisputably one of the most important therapeutic discoveries that had effectiveness against serious bacterial infections [6]. Antibiotic resistance has increased substantially in recent years and is posing an ever-increasing therapeutic problem, and one of the methods to reduce the resistance to antibiotics is by using antibiotic resistance inhibitors from plants [7]. The development of drug resistance in human pathogens against commonly used antibiotics has necessitated a search for new antimicrobial substances from other sources [8].

Antioxidants are compounds that inhibit or delay the oxidation of other molecules by inhibiting the initiation or propagation of oxidizing chain reactions and can protect the human body from free radicals and retard the progress of many chronic diseases as well as lipid peroxidation [9, 10]. Most natural plants containing a wide variety of phytochemical constituents are a major source of antioxidants that affect the decrease in the possible stress caused by reactive oxygen species [11]. *C. forskohlii* is one of the family members of the Lamiaceae; a large common family, widely spread and mostly distributed in the Mediterranean region, and it has been prescribed in folk medicine to treat various medical conditions including stomach, liver, intestine disorders, heart, and respiratory problems [12]. This study is conducted to investigate the phytochemical constituents, total phenolic content, and antioxidant and antimicrobial activities of *C. forskohlli* stem extract collected from Al-Baha area, Saudi Arabia.

2. Material and methods

2.1. Sample collection and preparation

C. forskohlii stems were collected from Al-Baha area, Kingdom of Saudi Arabia. The plant was taxonomically identified and authenticated by Dr. Haidar Abd Algadir, Department of Biology, Faculty of Science and Al-Baha University. The voucher specimens were identified according to Collenette [13] and Chaudhary [14], after comparing with voucher specimens deposited at the Herbaria of King Saud University. The stems were washed with fresh water to remove soil and dust particles and cut into small pieces.

2.2. Preparation of ethanolic extract of samples

Two hundred grams (200 gm) of the powdered sample were weighed into a clean dried flask (2-3 L size). Two thousand milliliters (2000 ml) of 80% ethanol (800 ml ethanol+200 ml distilled water) were added. The mixture was well mixed and soaked for 2 days at room temperature. The mixture was then filtered with filter paper (Whatman no. 4). This procedure was repeated three times to ensure that all contents were extracted with ethanol. The filtrates were collected and allowed to air dry for 10 days, and the extract was stored in a coloured bottle at 4-6°C till analysis.

2.3. Fractionation of the ethanolic extract by liquid-liquid extraction

The ethyl alcohol extract was soaked in 200 mL distilled water and extracted consecutively using different solvents (petroleum ether, chloroform, ethyl acetate, and *n*-butanol, respectively) for 7 days at room temperature to produce extracts. The solvents were chosen depending on their polarity difference. The filtrate was dried by using a rotary evaporator at room temperature and stored at 4-6°C till used.

2.4. Determination of total phenolic content (TPC)

The concentration of phenolics in the stem extracts was determined using a spectrophotometric method [15], with some modifications. Sample solutions of the ethanolic extracts in the concentration of 1 mg/ml were used in the analysis. The reaction mixture was prepared by mixing 0.5 ml of sample solutions of fractions, 2.5 ml of 10% Folin-Ciocalteu's reagent dissolved in water, and 2.5 ml of 7.5% $NaHCO_3$. Blank was concomitantly prepared, containing 0.5 ml methanol, 2.5 ml 10% Folin-Ciocalteu's reagent dissolved in water, and 2.5 ml of 7.5% of $NaHCO_3$. The samples were thereafter incubated at 30°C for 90 min. The absorbance was determined using a spectrophotometer at λmax = 765 nm. The samples were prepared in triplicate for each analysis. The same procedure was repeated for the standard solution of gallic acid and the calibration line was constructed. Based on the measured absorbance, the concentration of phenolics was read (mg/ml) from the calibration line; then the content of phenolics in extracts was expressed in terms of gallic acid equivalent (mg of GAE/gm of extract).

2.5. Determination of total antioxidant activity

2.5.1. Free radical scavenging assay

The antioxidant assay used was based on the scavenging ability of the antioxidant (s) in plant extracts towards the stable free radical 1, 1-diphenyl-2-picrylhydrazile (DPPH), which is deep purple, to form the corresponding hydrazine with the accompanying change of colour to light purple or golden yellow.

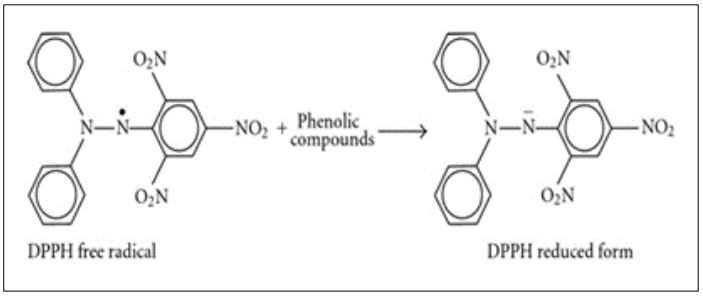

Figure 1 DPPH-free radical and its reduced form

2.5.2. Free radical scavenging procedure

This method was carried out according to Shyur et al. [16] with some modifications. A stock solution was prepared by dissolving 1mg of the sample in 1ml of absolute ethanol (98%). The stock solution was diluted to final concentrations of 100, 50, 25, 12.5, 6.25, 3.125, 1.5625 µg/ml in ethanol. 0.9ml of Tris-HCl and 1ml of 0.1 mM DPPH in methanol solution were added to each concentration and incubated at room temperature in the dark for 30 minutes. The absorbance of the resulting mixture was measured at 517 nm and converted to percentage antioxidant activity using the formula below: -

$$\text{Scavenging activity (DPPH scavenged) (\%)} = \frac{A_C - A_S}{A_C} \times 100$$

Where: Ac= Absorbance of control; As = Absorbance in the presence of the sample of extract. A solution of 0.9 ml Tris-HCl+ 0.1ml absolute ethanol+ 1ml absolute ethanol was used as a blank, while the solution of 0.9 ml tris-HCl+0.1ml absolute ethanol+1ml DPPH was used as a control. Freshly prepared DPPH solution exhibits a deep purple colour with a maximum absorbance at 517 nm. The purple colour disappears when an antioxidant is present in the medium. Thus, the change in the absorbance of the reduced DPPH was used to evaluate the ability of the compound to act as a free radical scavenger.

2.6. Determination of antimicrobial activity

2.6.1. Microbial organisms

Bacterial strains of *B. subtilis* (NCTC 8236), *S. aureus* (ATCC 25923), *E. coli* (ATCC 25922), and *P. aeruginosa* (ATCC 27853), in addition to a fungal strain *C. albicans* (ATCC 7596) were used in this study for antimicrobial activity test.

2.6.2. Preparation of bacterial suspensions

Aliquots (1 ml) of a 24 hr broth culture of the bacteria were aseptically distributed onto nutrient agar slopes and incubated at 37°C for 24 hr. The bacterial growth was harvested and washed off with 100 ml sterile normal saline to produce a suspension containing about 10^8- 10^9cfu/ ml, which was stored at 4°C till used. The average number of viable organisms/ml of the stock suspension was determined employing the surface viable counting technique [17]. Serial dilutions of the stock suspension were made in a sterile normal saline solution and 0.02 ml volumes of the appropriate dilution were transferred by micropipette onto the surface of dried nutrient agar plates, which were allowed to stand for 2 hr at room temperature for the drops to dry and then incubated at 37°C for 24 hr. After incubation, the number of developed colonies in each drop was counted. The average number of colonies per drop (0.02 ml) was multiplied by 50 and the dilution factor to get the viable count of the stock suspension, expressed as colony forming units (cfu)/ml suspension. Each time a fresh stock suspension was prepared. All the above experimental conditions were maintained constant so that suspensions with very close viable counts would be obtained.

2.6.3. Preparation of fungal suspension

The fungal cultures were maintained on Sabouraud dextrose agar, incubated at 25°C for 4 days. The fungal growth was harvested and washed with sterile normal saline and finally suspended in 100 ml sterile normal saline and stored at 4°C until used.

2.6.4. Agar disc diffusion method

The disc diffusion method was used to screen the antimicrobial activity of plant extracts and performed by using Mueller Hinton agar (MHA) and Sabouraud dextrose agar (SDA). The experiment was carried out according to Boudjema et al. [18]. Bacterial and fungal suspensions were diluted with a sterile physiological solution to 10^8cfu/ ml (turbidity = McFarland standard 0.5). One hundred microliters of bacterial and fungal suspensions were swabbed uniformly on the surface of MHA and SDA and the inoculum was allowed to dry for 5 min. Sterilized filter paper discs (Whatman No.1, 6 mm in diameter) were placed on the surface of MHA and SDA and soaked with 20 µl of a solution of each plant extract. The inoculated plates were incubated at 37°C for 24 hr in an inverted position. After incubation, the antimicrobial activity was determined by measuring the diameter of the inhibition zone surrounding each disc. The antimicrobial activity results were expressed in terms of the diameter of the inhibition zone as follows: <9 mm zone (resistant strain); 9-12 mm (partially sensitive strain); 13-18 mm (sensitive strain); >18 mm (very sensitive strain).

2.7. Statistical analysis

The statistical analysis was performed using Statistical Analysis Systems (SAS, Ver. 9, SAS Institute Inc., Cary, NC, USA) and the results were presented as the mean ± standard deviation (SD) of three replicates. All the data were statistically assessed using the General Linear Model (GLM) and the significant difference was performed using Duncan multiple range test at P≤0.05.

3. Results and discussion

3.1. Phytochemical screening of *C. forskohlii* stem extract

The phytochemical screening revealed the presence of all phytochemicals in the stem extracts except flavonoids and saponins which were absent in petroleum ether and chloroform extracts, terpenoids were absent in ethyl acetate extract, and tannins and saponins were absent in n-butanol extract (Table 1). *C. forskohlii* is one of the most important potential medicinal plants of the future with its therapeutic properties being authenticated recently [19]. Phytochemical analysis of *Coleus species* (whole plant) extracts showed the presence of alkaloids, flavonoids, tannins, cardiac glycosides, carbohydrates, steroids, glycosides, proteins, and anthracene glycosides, while reducing sugars and saponins were not detected in any of the plant extracts [20]. Phytochemical-screening results of *C. forskohlii* water, hexane, chloroform, methanol, and 80% methanol extracts showed the presence of all the constituents in aqueous and methanol extracts, while proteins, carbohydrates, and cardiac glycosides were found in chloroform extract, and cardiac glycosides were found in hexane extracts, indicating that water and methanol were able to extract most of the phytochemicals present in the shoot [19].

Table 1 Phytochemical constituents of *C. forskohlii* stem extracts

Phytochemicals	Extracts				
	Ethanol	Petroleum ether	Chloroform	Ethyl acetate	n-butanol
Flavonoids	+	-	-	++	++
Alkaloids	+	++	+	+++	+
Tannins	++	+	+	++	-
Terpenoids	++	+	+	-	+
Steroids	++	+	+	++	+
Saponins	+	-	-	+	-
Reducing sugars	++	+	+	+	+

+: Present; ++: Present; +++; Present; -: Absent

The presence of alkaloids, flavonoids, phenolic compounds, terpenoids, protein, and cardiac glycosides in the aqueous, ethanol, acetone, and chloroform extracts of leaf, stem, and root of *C. forskohlii*, indicated that these solvents are effective to isolate active biological compounds due to their high polarity [2]. It has been suggested that phytochemical extracts from plants hold promises for use in allopathic medicine as they are potential sources of antiviral, antitumoral, and antimicrobial agents [21].

3.2. Total phenolic content of *C. forskohlli* stem extract

The results in Table 2 have shown that the total phenolic content was significantly (P<0.001) higher in n-butanol extract (274.33±3.29 mg GAE/gm), followed by ethyl acetate extract (182.94±1.82 mg GAE/gm), ethanol extract (79.63±2.02 mg GAE/gm) and petroleum ether extract (73.38±3.07 mg GAE/gm), while the lowest content was in chloroform extract (60.06±2.12 mg GAE/gm). Studies showed that many flavonoids and related polyphenols contribute significantly to the total antioxidant activity of many plants [22]. Phenols contain good antioxidant, antimutagenic and anticancer properties, therefore, the highest content of total phenol in tubers is held responsible for better antioxidant characteristics [22]. Flavonoids are considered the major group of phenolic compounds for their antiviral, antimicrobial, and spasmolytic properties, while alkaloids are commonly found to have antimicrobial properties [23]. Various plants have already been proved to have high antioxidant activity with high amounts of phenolics and flavonoids [21]. The total phenol content was significantly higher (P<0.05) in the tubers (27.05 µg catechol equivalents/gm dry tissue) compared to roots (24.22 µg catechol equivalents/gm dry tissue) and stem (21.26 µg catechol equivalents/gm dry tissue) [22]. The stems of *C. aromaticus* showed higher content of total polyphenols (62.12 mg/gm fw) compared with *C. forskholii* (31.32 mg/gm fw) and *C. zeylanicus* (48.5 mg/gm fw) [24]. The ethanolic extract of *C. forskohlii* tubers had maximum phenolic content of 38.82±0.22 mg GAE/gm [25].

Table 2 Total phenolic content (mg GAE/gm) of *C. forskohlii* stem extract

Extract	Total phenolic content (mg GAE/gm)
Ethanol	79.63±2.02^{c}
Petroleum ether	73.38±3.07^{d}
Chloroform	60.06±2.12^{e}
Ethyl acetate	182.94±1.82^{b}
n-butanol	274.33±3.29^{a}

P<0.001; n = 3

3.3. Antioxidant activity of *C. forskohlli* stem extract

Results in Table 3 show that the antioxidant activity, measured as DPPH scavenging, of stem extract of *C. forskohlii*, was significantly (P<0.001) higher in n-butanol (67.68±1.55%), followed by ethyl acetate (43.38±1.27%), ethanol (36.02±1.29%), petroleum ether (20.71±0.59%) and chloroform (19.73±0.74%). DPPH is a stable free radical compared to other antioxidants and considered an important method to assess the potential of natural compounds to scavenge toxic oxygen molecules [25]. Antioxidants are compounds that inhibit or delay the oxidation process by preventing the initialization or propagation of oxidizing chain reactions [26]. A direct linear relationship between the total phenolic content and total antioxidant activity was reported in leaf extracts of medicinal plants indicating that the phenolic compounds might be the major contributors to the antioxidant activities of these extracts [27]. *C. forskholii* stem tissue had relatively higher antioxidant content (12.6 mM/gm fw) compared to *C. aromaticus* (11.6 mM/gm fw) and *C. zeylanicus* (11.8 mM/gm fw) [24]). The ethanolic rhizome extract of *C. forskohlii* collected from different regions of India showed antioxidant activity ranging from 62.1% to 87.6% [25]. The antioxidant activity of ethanolic extracts of root, stem, leaves, and tubers of *C. forskohlii* was evaluated and it was reported that the radical-scavenging activity of the tubers (90.32% at 1.0 mg/ml) was significantly higher (P<0.05) followed by leaves (87.34% at 1.0 mg/ml), roots (85.34% at 1.0 mg/ml) and stem (69.40% at 1.0 mg/ml), respectively [22]. The methanolic extracts from the leaves exhibited a strong antioxidant activity than the stems, and the correlation coefficient between the phenolic concentration of leaves, stem, and DPPH scavenging activity was found to be 0.946 and 0.979, respectively indicating a strong correlation [12]. The antioxidant activity of *C. forskohlii* and *Plectranthus barbatus* showed a dose-dependent response, and the correlation between antioxidant activity and phenolic acids indicated that compounds were the dominant contributors to the antioxidant activity of both plants [28].

Table 3 Antioxidant activity (%) of *C. forskohlii* stem extract

Extract	Antioxidant activity (%)
Ethanol	36.02±1.29[c]
Petroleum ether	20.71±0.59[d]
Chloroform	19.73±0.74[d]
Ethyl acetate	43.38±1.27[b]
n-butanol	67.68±1.55[a]

P<0.001; n = 3

3.4. Antimicrobial activity of *C. forskohlli* stem extract

The results in Tables 4-8 showed that all microorganisms tested were resistant at the concentration of 25 and 50 mg/ml of plant extracts, whereas the concentrations of 100, 150 and 200 mg/ml showed varying activities against gram-negative (*E. coli, P. aeruginosa*), gram-positive (*S. aureus, B. cereus*) and *C. albicans*. At the concentration of 100 mg/ml, all bacteria were sensitive to petroleum ether extract (10±0.23 mm, 11±0.01 mm, 10±0.00 mm, and 12.5±0.00 mm inhibition zone for *E. coli*, *P. aeruginosa*, *S. aureus,* and *B. cereus*, respectively), while *C. albicans* was sensitive to ethanol and n-butanol extracts (14±0.1 mm and 13±0.00 mm IZ, respectively) at 100 mg/ml. At 150 mg/ml, *E. coli* and *B. cereus* were resistant to all extracts except petroleum ether, while *P. aeruginosa* was partially sensitive to all extracts, and *B. subtilis* and *C. albicans* were resistant to ethanol and ethyl acetate extracts, respectively. At the concentration of 200 mg/ml, all microorganisms under study were sensitive to all extracts. Many studies suggested that different solvent extracts of various plants have tremendous biological activity, and such effective extracts can be subjected to isolation of the therapeutic compounds and antimicrobials for further pharmacological studies [21]. Plants play an important role in human health because they produce a wide range of bioactive molecules with medicinal value including antimicrobial properties [29]. A previous study showed that all the tested pathogens (*Bacillus subtilis*, *Pseudomonas fluorescence*, *P. aeruginosa, Klebsiella pneumoniae, E. coli, S. aureus,* and *Streptococcus pneumoniae*) were highly susceptible to the crude leaf, stem, and root extracts of *C. forskohlii*, although the antibacterial activity was more pronounced against gram-negative bacterial strains than gram-positive ones [3]. It was reported that the 50% methanolic extract of *Coleus species* inhibited the growth of the test fungi, namely, dermatophytes, *Basidiobolus haptosporus, Rhodotorula species, C. albicans*, species of *Aspergillus, Mucor*, *Rhizopus,* and *Geotricum* for ≥14 days, and the absolute methanolic extract, as well as the aqueous extract, inhibited the fungal growth for ≥12 days [20]. At the concentration of 100 mg/ml, the leaf extract of *C. forskohlli* was most effective against *S. aureus* (zone of inhibition of 14.56 mm), followed by *Streptococcus mitis* (13 mm), but in terms of percentage of relative inhibition zone diameter, the extract produced the highest inhibition percentage against *Streptococcus mitis*, followed by *S. aureus* [30]. *C. forskohlii* leaf extract showed powerful antimicrobial activity against *K. pneumoniae* and *E. coli* (inhibition zone of 10 and 8, respectively), and moderate activity against *S. aureus* (7 mm), and no activity against *C. albicans* [29]. Ethanol, methanol, ethyl acetate, acetone, and chloroform extracts of *C. forskohlii* root showed varying activities against *B. cereus*, *Micrococcus luteus*, S. *aureus*, *Klebsiella pneumoniae*, *E. coli*, *C. albicans*, *Aspergillus flavus*, *Aspergillus niger*, and *Cryptococcus neoformans* [23].

Table 4 Inhibition zones (mm) of different concentrations of *C. forskohlli* stem extract against *E. coli*

Extract	Extract concentration (mg/ml)				
	25	50	100	150	200
Ethanol	-	-	-	8±0.01	12±0.00
Petroleum ether	-	-	10±0.23	15±0.02	17±0.02
Chloroform	-	-	-	7±0.01	13±0.01
Ethyl acetate	-	-	-	9±0.01	12±0.02
n-Butanol	-	-	-	5±0.01	11±0.01

The ethanolic extract of *C. aromaticus* roots and leaves was reported to be highly active against *E. coli*, *S. aureus,* and *P. mirabilis* and moderately active against *K. pneumonia* and *P. aeruginosa* [31].

Table 5 Inhibition zones (mm) of different concentrations of *C. forskohlli* stem extract against *P. aeruginosa*

Extract	Extract concentration (mg/ml)				
	25	50	100	150	200
Ethanol	-	-	-	9±0.02	10±0.00
Petroleum ether	-	-	11±0.01	12±0.00	13±0.02
Chloroform	-	-	-	10±0.02	12±0.03
Ethyl acetate	-	-	-	8±0.03	9±0.04
n-Butanol	-	-	-	9±0.05	14±0.01

Table 6 Inhibition zones (mm) of different concentrations of *C. forskohlli* stem extract against *S. aureus*

Extract	Extract concentration (mg/ml)				
	25	50	100	150	200
Ethanol	-	-	-	7±0.00	13±0.03
Petroleum ether	-	-	10±0.00	13±0.02	16±0.07
Chloroform	-	-	-	5±0.03	11±0.02
Ethyl acetate	-	-	-	6±0.01	14±0.01
n-Butanol	-	-	-	8±0.02	15±0.05

Table 7 Inhibition zones (mm) of different concentrations of *C. forskohlli* stem extract against *B. cereus*

Extract	**Extract concentration (mg/ml)**				
	25	**50**	**100**	**150**	**200**
Ethanol	-	-	-	7±0.02	15±0.01
Petroleum ether	-	-	12.5±0.00	15±0.01	18±0.03
Chloroform	-	-	-	10±0.00	13±0.02
Ethyl acetate	-	-	-	13±0.01	15±0.04
n-Butanol	-	-	-	12±0.02	16±0.00

Table 8 Inhibition zones (mm) of different concentrations of *C. forskohlli* stem extract *C. albicans*

Extract	**Extract concentration (mg/ml)**				
	25	**50**	**100**	**150**	**200**
Ethanol	-	-	14±0.1	17±0.01	20±0.02
Petroleum ether	-	-	-	10±0.02	15±0.01
Chloroform	-	-	-	9±0.04	13±0.21
Ethyl acetate	-	-	-	8±0.03	12±0.25
n-Butanol	-	-	13±0.00	15±0.00	15±0.51

The crude ethanolic rhizome extract of *C. forskohlii* had a maximum inhibitory activity against *B. subtilis* followed by *B. cereus, P. aeruginosa,* and *S. aureus*, while it was not inhibitory against *E. coli* [32].

4. Conclusion

Solvent extract (ethanol, petroleum ether, chloroform, ethyl acetate, and n-butanol) of *C. forskohlii* stem revealed the presence of flavonoids, alkaloids, tannins, terpenoids, steroids, saponins, and reducing sugars. The total phenolic content of stem extract had phenolic compounds in varying concentrations depending on the solvent used in extraction. The n-butanol extract had the highest antioxidant activity, followed by ethyl acetate, ethanol, petroleum ether, and chloroform. All microorganisms tested were resistant at the concentrations of 25 and 50 mg/ml of plant extracts, whereas the concentrations of 100, 150 and 200 mg/ml showed varying activities against gram-negative (*E. coli, P. aeruginosa*), gram-positive (*S. aureus, B. cereus*) and *C. albicans*. The study concluded that *C. forskohlii* stem extract used in this study had pharmaceutical and biological activities and can be used in the pharmaceutical as well as the food industry.

Compliance with ethical standards

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