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Research Article

Phenolic Acids and Biological Activities of *Coleus forskohlii* and *Plectranthus barbatus* as Traditional Medicinal Plants

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Abstract

Background and Objective: Importance of traditional medicines cannot be over emphasized, as they are not only widely used in Saudi Arabia but worldwide. This study was designed to determine phenolic acids contents, antitumor, antioxidant and antimicrobial activities of traditional medicinal plants *Coleus forskohlii* (*C. forskohlii*) and *Plectranthus barbatus* (*P. barbatus*) in Saudi Arabia.

Materials and Methods: Phenolic acids were detected by HPLC, antioxidant activity determined by 2,2-Diphenyl-1-picrylhydrazyl (DPPH) scavenging system. Antitumor activity performed on HCT-116 cell line by antioxidant enzymes catalase, polyphenoloxidase and peroxidase enzymes were estimated in plant extracts. Poisoned food technique and disc diffusion method for antifungal and antibacterial assay were used respectively. **Results:** The HPLC analysis of the *Coleus forskohlii* and *Plectranthus barbatus* extracts indicated the presence of different types of phenolic acids. Ferulic acid and gallic acids were detected in highest concentrations among the other phenolic acid, where the concentration of ferulic acid in *C. forskohlii* and *P. barbatus* was 27.55 and 23.26 $\mu\text{g g}^{-1}$, respectively, while gallic acid concentration in *C. forskohlii* and *P. barbatus* was 25.42 and 22.58 $\mu\text{g g}^{-1}$, respectively. *Coleus forskohlii* and *P. barbatus* species have different antioxidant enzymes which include catalase, polyphenoloxidase and peroxidase enzymes. The IC_{50} value of the antioxidant activity of *C. forskohlii* was lower than in *P. barbatus*. *Coleus forskohlii* and *P. barbatus* have promising effect in tumor activities that appeared in its ability in inhibiting cell proliferation of HCT-116 cell line. *Coleus forskohlii* but not *P. barbatus* has antibacterial activities against *S. aureus*, *E. coli* and *S. typhi* while weak antifungal activity was observed with using *C. forskohlii* and *P. barbatus* extracts. **Conclusion:** *Plectranthus barbatus* has potential antioxidant activity than *C. forskohlii* whereas, *C. forskohlii* has antimicrobial properties which are not shown by other plant so, current comparative results support the folk use of this medicinal plants.

Key words: Phenolic acids, *Coleus forskohlii*, *Plectranthus barbatus*, biological activities, folk medicine

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Now, the plant species that contributing in the development of modern medicines reached to more than 85,000 species. Natural compounds from medicinal plants hold great promise for the detection and advancement of novel pharmaceuticals in a diverse human ailments¹. *Plectranthus* species are used economically in Saudi Arabia as traditional medicine for their biological activities in the primary health care system². *Plectranthus* including numerous species but certain species are known and excellent not only in traditional medicines but used as antiseptic for wounds, treat for stomach, liver disturbance, respiratory diseases and heart disorders³. Different phytochemicals including diterpenes, phenolics and triterpenoids were detected as a plant secondary metabolites from certain species of *Plectranthus*⁴.

One of the family members of the Lamiaceae is *Coleus forskohlii*. *Coleus forskohlii* is a large family that is common, widely spread and mostly distributed in the Mediterranean region⁵. Phytochemical analysis of *C. forskohlii* explained showed that it contains of terpenoids, flavonoids, tannins, reducing sugars and alkaloids. Flavonoids belong to the group of polyphenolic compounds and are mostly known by health promoting properties such as antioxidant, anti-allergic, anti-inflammatory, antimicrobial and anti-cancer properties^{6,7}. *Plectranthus barbatus* and *P. ornatus* in folk medicine was possess antioxidant, antibacterial activities and are capable of inhibiting the oxidation forming products⁸ and therefore these plants extracts can be used as a good antioxidant.

Ammon and Muller⁹ reported that there were many diseases that can be treated by *Coleus forskohlii* such as heart problems, abdominal colic, respiratory disorder, insomnia, convulsion, asthma, bronchitis, intestinal disorder, burning sensation, constipation, epilepsy and angina. Furthermore, Rout *et al.*¹⁰ showed this plant constitutes different natural bioactive compounds such as phenolics, terpenoids, alkaloids, saponin, tannins and may produce health beneficial effect by scavenging free radicals. GC-MS result clearly indicated the plant of *C. forskohlii* root ethanol extract have following compound, n-hexadecanoic acid have antibacterial and antifungal properties¹¹.

Numerous studies indicated that phenolic products of plants plays an important role in human health such as minimize the risk of cardiovascular disorders^{12,13}. Aliyu *et al.*¹⁴ mentioned that the great antioxidant activities of plants due to phenolic compounds. Certain amino acids L-phenylalanine or L-tyrosine, phenolic are considered precursors of the synthesis of the natural phenolic products in plants¹⁵. Phenolic

compound such as gallic acid has many biological activities including antibacterial, antineoplastic, antimelanogenic and antioxidant activities¹⁶. This molecule showed anticancer properties in prostate carcinoma cells¹⁷, while Kratz *et al.*¹⁸ described its promising activity as an anti-HSV-2 (Herpes simplex virus) agent. Furthermore, Lu *et al.*¹⁹ mentioned that gallic acid may use for brain tumors treatment due to its ability to suppress cell viability, proliferation, invasion and angiogenesis in human glioma cells. p-hydroxybenzoic acid, has been reported to have antioxidant activity against free radicals¹⁵, antimicrobial activity against pathogenic bacteria and fungi²⁰. Likewise ferulic acid, which can be isolated from different food such as cereals, fruits, vegetables and coffee, demonstrates apparent antimicrobial activity against *Escherichia coli*. This antimicrobial activity is obvious as well in other chlorogenic acid CGA and related compounds such as isoferulic, benzoic and hydroxybenzoic acids²¹. Additionally, ferulic acid has activity against cancer cell and inflammatory cells and has anti-thrombogenic activity as well²²⁻²⁴. Its activity against cancer cells through inhibition of N-nitroso compounds seems to decrease many types of cancers such as stomach, colon, breast, prostate, liver, lung and tongue cancers. Furthermore, FA has effect on cardiovascular diseases through reduction in cholesterol and triglycerides levels^{25,26}.

Chlorogenic acid (CGA) and related compounds exhibited specific antimicrobial activity and corresponding reduction in log survival ratio, in which ferulic, isoferulic, benzoic and hydroxybenzoic acids exhibited obvious antimicrobial activity against *E. coli*²¹. Ferulic acid is isolated from many staple foods, such as fruits, vegetables, cereals and coffee. It and its derivatives exhibited diversity activity, such as anticancer, antiatherogenic, anticarcinogenic and antibacterial agents, as well as anti-inflammatory activity^{22-24,27}. The FA decreases the level of cholesterol and triglycerides, thereby reducing the risk of heart disease. Moreover, FA seems to reduce the risk of many cancers, including stomach, colon, breast, prostate, liver, lung and tongue cancers by inhibition the formation of N-nitroso compounds^{25,26}. Biological and pharmacological activities of caffeic acid have been reported earlierly such as anticancer with using human cancer cell lines, antioxidant, anti-inflammatory, anti-thrombosis, antimicrobial and antihypertensive^{25,26}. Most traditional medicinal plants in use today have no scientific data on their bioactivity and levels of safety or even how they are likely to affect each other when used as combinations in medicines, therefore, our study aimed to determine phenolic acids contents, antitumor, antioxidant and antimicrobial activities of traditional medicinal plants *Coleus forskohlii* and *Plectranthus barbatus* in Saudi Arabia.

MATERIALS AND METHODS

Plant material and extract preparation: *Coleus forskohlii* and *Plectranthus barbatus* as traditional medicinal plants were collected from South region, Saudi Arabia. The air-dried leaves of each plant (250 g) were grinded to fine powder and extracted separately with methanol. The collected methanol extracts were dried and kept frozen until usage.

Phenolic acids detection by HPLC: *Coleus forskohlii* and *P. barbatus* extracts were subjected to qualitative and quantitative determination of phenolic acids using the HPLC. The extracts according to Shaheen *et al*² were subjected to solid phase extraction using silica gel finger column eluted with water, then the aliquot samples were filtered prior to injection in high performance liquid chromatography (HPLC). An HPLC (Waters 600E System controller) with a fluorescence detector (Waters 470) and an auto-sampler (Waters 712 WISP) were used at the regional center for Mycology and Biotechnology AL-Azhar University, Cairo, Egypt. Identification of peaks was achieved by congruent retention times and UV-PDA profile against standards.

Poisoned food technique and disc diffusion method for antifungal and antibacterial assay: Potato dextrose agar medium (PDA) supplemented with plant extract at different concentrations 100, 200 300 mg/100 mL medium. The PDA medium (25 mL) was poured into plate. After medium solidify, 5 mm disc of 5 days old fungal cultures was placed at the center of the plate, then incubated for 6 days at 25°C, the growth represented by colony radius was measured. The PDA without plant extract used as control. Inhibition of the fungal growth in relation to the control treatment was calculated as:

$$I (\%) = \left(\frac{C - T}{C} \right) \times 100 \quad (1)$$

where I is the fungal growth inhibition (%), C is the radial fungal growth with the control (cm) and T is the radial fungal growth with the plant extract treatment (cm).

Growth inhibition of bacteria by the plant extracts was determined by disc-diffusion assay. Three bacterial species including *Staphylococcus aureus*, *Escherichia coli* and *Salmonella typhi* were cultivated on nutrient agar plates. Bacterial suspension (10^5 cell mL⁻¹) was poured over the plates forming a homogenous top layer. Filter paper disc (5 mm diameter) was emergen in 100, 200 and 300 μ L of extract equal 10, 20 and 30 mg of extract. The discs were

air-dried and placed onto the seeded top layer of the agar plates. Antibacterial activity was expressed as the zone of inhibition (mm) after incubation period 24 h at 37°C. Methanol saturated discs (air dried) and commercial antibiotic were used as negative controls and positive control.

Antioxidant enzymes measurements

Peroxidase activity: Reaction mixture of peroxidase activity estimate was prepared by mixing 0.1 mL of plant extract with 1 mL reagent containing 40 mM guaiacol, 8 mM H₂O₂ and 50 mM sodium acetate buffer, pH 5.5²⁸, then the result of reaction after 1 min was measured at absorbance 470 nm. The amount of enzyme which increases the O.D. 1 per min under standard assay conditions equal one unit of peroxidase activity.

Polyphenol oxidase activity: Reaction mixture of polyphenol oxidase activity estimate was prepared by mixing 0.1 mL of plant extract with 2.5 mL of 20 mM catechol reagent prepared in 1 mM of phosphate-buffered saline (PBS), pH 6.8²⁹. The absorbance of reaction mixture was measured at 400 nm. The amount of enzyme which increases the O.D. One per min under standard assay conditions equal one unit of polyphenol oxidase activity.

Catalase activity: Reaction mixture of catalase activity estimate was prepared by mixing 0.5 mL of plant extract with 2 mL of 25 mM H₂O₂ prepared in a 75 mM PBS pH 7.0³⁰. The absorbance of reaction mixture was measured at 240 nm. The amount of enzyme which increases the O.D. One per min under standard assay conditions equal one unit of catalase activity.

Antioxidant activity by DPPH radical scavenging activity:

The antioxidant activity of the plant extract was determined by the DPPH free radical scavenging assay freshly prepared (0.004% w/v) methanol solution of 2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical was prepared and stored at 10°C in the dark. A methanol solution of the extract was prepared. A 40 μ L aliquot of the methanol solution was added to 3 mL of DPPH solution. Absorbance measurements were recorded immediately with a UV-visible spectrophotometer (Milton Roy, Spectronic 1201). The decrease in absorbance at 515 nm was determined continuously, with data being recorded at 1 min intervals until the absorbance stabilized (16 min). The absorbance of the DPPH radical without antioxidant (Control) and the reference compound ascorbic acid were also measured.

All the determinations were performed in 3 replicates and averaged. The percentage inhibition (PI) of the DPPH radical was calculated according to the formula:

$$P (\%) = \left(\frac{AC - AT}{AC} \right) \times 100 \quad (2)$$

Where:

AC = Absorbance of the control at $t = 0$ min

AT = Absorbance of the sample+DPPH at $t = 16$ min³¹

Antitumor evaluation MTT assay: Human colon carcinoma (HCT-116) was obtained from the American Type Culture Collection (ATCC, Rockville, MD) and used as a test cells. The HCT-116 at level 5×10^4 cell/well in Corning® 96-well tissue culture plates were suspended and incubated for 24 h in the medium. Different concentrations of plant extracts were added into 96-well plates (three replicates) to perform cytotoxicity test. With using MTT [3-(4,5-Dimethylthiazol-2yl)-2,5-Diphenyltetrazolium bromide] test, the numbers of viable cells were detected as following: The media were discarded from the 96 well plate and substitute with 100 μ L of fresh culture RPMI 1640 medium without phenol red then 10 μ L of the 12 mM MTT stock solution (5 mg of MTT in 1 mL of PBS) was added to each well including the untreated controls (6 vehicle controls with media or 0.5% DMSO were run for each 96 well plate). At 37°C and 5% CO₂ the 96 well plates were then incubated for 4 h. Media (85 μ L aliquot) media were discarded from the wells and 50 μ L of DMSO were added to each well and mixed thoroughly with the pipette and incubated at 37°C for 10 min. Then, the absorbance was measured in triplicates at 490 nm using ELISA reader system (SunRise TECAN, Inc., USA) to determine the number of viable cells and the percentage of viability was calculated by the following equation:

$$\left[1 - \frac{OD_t}{OD_c} \right] \times 100\%$$

where, OD_t is the mean optical density of wells treated with the tested sample and OD_c is the mean optical density of untreated cells. The relation between surviving cells and treatments concentration is plotted to get the survival curve of each tumor cell line after treatment with the specified compound. The 50% inhibitory concentration (IC₅₀), the concentration required to cause toxic effects in 50% of intact cells, was estimated from graphic plots of the dose-response curve for each concentration using Graphpad Prism software (San Diego, CA. USA)³².

Statistical analysis: The results are reported as mean \pm standard deviation SD of three independent replicates. Statistical analyses of data were carried out by computer using SPSS ver. 22.0 software.

RESULTS AND DISCUSSION

Importance of traditional medicines cannot be over emphasized, as they are not only widely used in Saudi Arabia but worldwide. The persistence of traditional medicine depends on the diversity and knowledge on medicinal properties of the plant. In the current study, HPLC analysis of the plant extracts showed 9 peaks (Fig. 1) indicated the presence of different types of phenolic acids in *Coleus forskolii* and *Plectranthus barbatus* with different retention times (Table 1, Fig. 2). Ferulic and gallic acids were detected in highest concentrations among the other phenolic acid. Generally, the extract of *C. forskolii* was found to be rich source of phenolic acids as compared to the *P. barbatus*. These results came in agreement with many other species of *Plectranthus* taxa from which these acids were previously identified by Shaheen *et al*², who stated that for example caffeic acid was detected only in *P. arabicus* (7.3 μ g g⁻¹). Trans-ferulic acid was present in *P. arabicus*, *P. hijazensis* and *P. asirensis* with concentrations of 12, 55 and 35 μ g g⁻¹

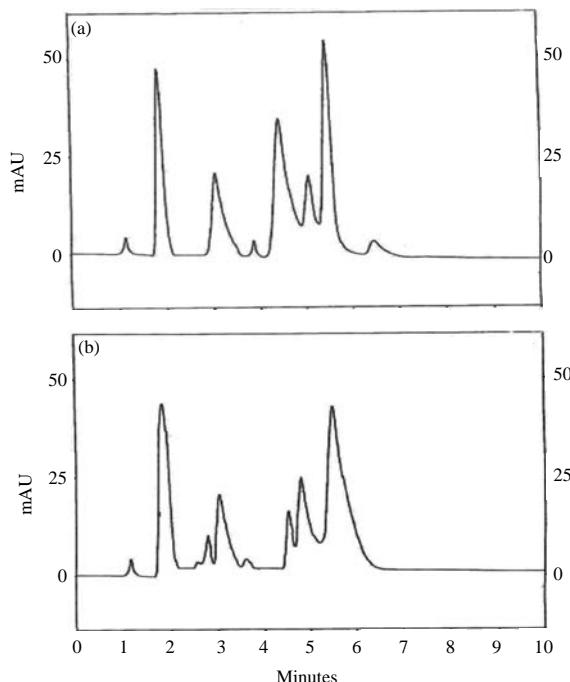


Fig. 1(a-b): HPLC chromatograms of the detected phenolic acids of (a) *Coleus forskolii* and (b) *Plectranthus barbatus*

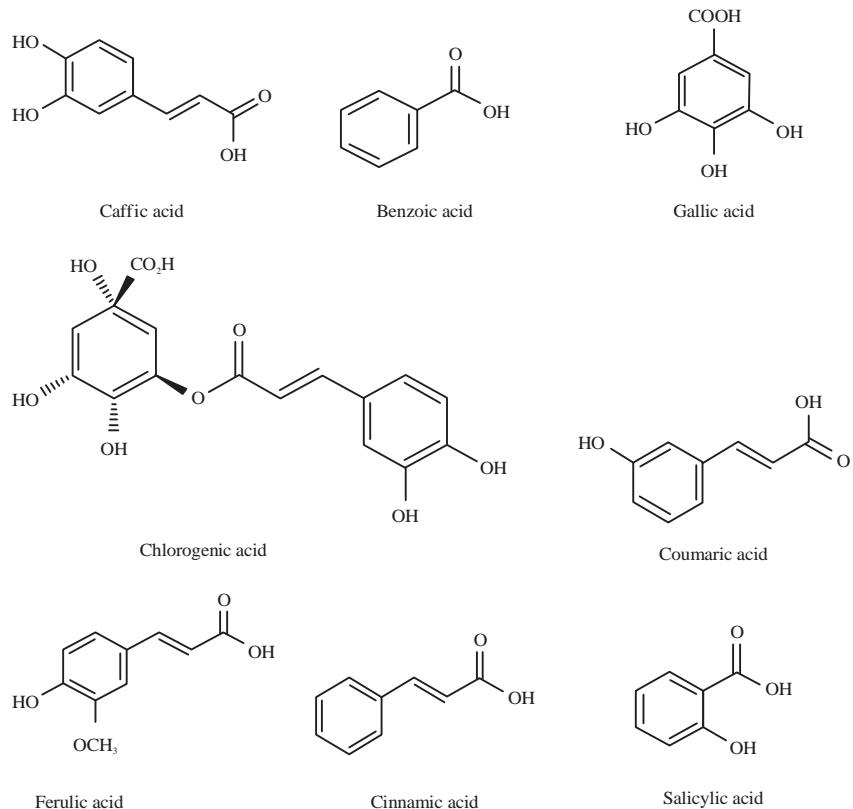


Fig. 2: Structural formula of detected phenolic acids

Table 1: HPLC report for phenolic acids detection in *Coleus forskohlii* and *Plectranthus barbatus*

Phenolic acids	<i>Coleus forskohlii</i> ($\mu\text{g g}^{-1}$)	<i>Plectranthus barbatus</i> ($\mu\text{g g}^{-1}$)	Retention time	Molecular formula
Benzoic	7.24	7.37	1.1	$\text{C}_7\text{H}_6\text{O}_2$
Gallic	25.42	22.58	1.8	$\text{C}_7\text{H}_6\text{O}_5$
Unknown	ND	9.06	2.8	-
Chlorogenic	15.9	13.98	3.0	$\text{C}_{16}\text{H}_{18}\text{O}_9$
Caffic	5.56	6.06	3.6	$\text{C}_9\text{H}_8\text{O}_4$
Coumaric	19.51	9.99	4.5	$\text{C}_9\text{H}_8\text{O}_3$
Salicylic	11.12	15.15	5.0	$\text{C}_7\text{H}_6\text{O}_3$
Ferulic	27.55	23.26	5.5	$\text{C}_{10}\text{H}_{10}\text{O}_4$
Cinnamic	2.19	ND	-	$\text{C}_9\text{H}_8\text{O}_2$

Table 2: Antioxidant enzymes activities of *Coleus forskohlii* and *Plectranthus barbatus*

Plant species	Antioxidant enzyme activities (units g^{-1})		
	Catalase	Polyphenoloxidase	Peroxidase
<i>Coleus forskohlii</i>	621.21 ± 0.20	140.52 ± 0.06	89.30 ± 0.020
<i>Plectranthus barbatus</i>	540.50 ± 0.05	120.06 ± 0.30	60.43 ± 0.02

extract, respectively. In this study cinnamic acid was detected in *C. forskohlii* but not in *P. barbatus*. Therefore, the results confirmed data of Shaheen *et al.*² where cinnamic acid was not detected in several species including, *P. arabicus*, *P. asirensis*, *P. pseudomarrubiooides*, *P. barbatus*, *P. hijazensis* and *P. aegyptiacus*, growing in Saudi Arabia. Gallic acid was detected in *Plectranthus* sp., growing in Saudi Arabia². Present result confirmed the presence of gallic acid in *P. barbatus* and *C. forskohlii*.

Coleus forskohlii and *P. barbatus* species have different antioxidant enzymes which include catalase, polyphenoloxidase and peroxidase enzymes (Table 2). The antioxidant activities of these enzymes, in general, are higher in *C. forskohlii* species compared to *P. barbatus* species. According to Khatun *et al.*³³ superoxide dismutase, peroxidase, polyphenol oxidase and catalase were founded in different parts of *C. forskohlii* including roots, stem, leaves and tubers. Antioxidant enzymes serve as an intrinsic defense tool to resist

oxidative damage in plants³⁴. Bandeira *et al.*³⁵ stated that the antioxidant activity of *P. barbatus* is mainly related to reducing properties and chemical structure of the phenolic compounds that play an essential role in neutralizing "reactive oxygen species" and "reactive nitrogen" species acting both as the initiation step in the propagation of the oxidative process. Antioxidant properties of many plants play an important role in food processing industry, since their possible use as natural additives has emerged from a growing tendency to replace synthetic antioxidants with natural ones³⁶⁻³⁸.

The results of the antioxidant activity of *C. forskolii* and *P. barbatus* extracts (Table 3) showed a dose-dependent response particularly at concentration up to 80 μ L. The IC₅₀ value of *C. forskolii* obtained in this study was lower than in *P. barbatus*. A correlation between the antioxidant activity and the phenolic acids detected of the extracts indicated that compounds were the dominant contributors to the antioxidant activity of both plants. In previous study of Rasineni *et al.*³⁹ suggested that *C. forskolii* can be used as an important source of phenolic compounds with significantly high antioxidant activity. *Plectranthus barbatus* and *P. ornatus* possess antioxidant properties and are capable of inhibiting the oxidation forming products and also have potential effectiveness against *S. aureus*⁹. The use of traditional medicine is wide spread and plants are still a large source of natural antioxidants that might serve as leads for the development of novel drugs⁴⁰.

Table 3: Antioxidant activity *Coleus forskohlii* and *Plectranthus barbatus* using DPPH scavenging

Extract concentration (μ L)	DPPH scavenging (%)	
	<i>Coleus forskohlii</i>	<i>Plectranthus barbatus</i>
160	95.60	83.15
80	92.43	80.32
40	80.21	75.23
20	62.41	60.56
10	58.52	54.61
5	52.05	51.24
0	0.00	0.00
IC ₅₀ (μ L)	5.09	5.40

Coleus forskolii and *P. barbatus* have promising effect in tumor activities that appeared in its ability in inhibiting cell proliferation (Table 4). These antitumor activities against HCT-116 cell line are profound when exposing these cells to the higher extracted concentrations and as expected the effect decreases with using lower concentration of any of these species. *C. forskolii* exhibit 83.18% cell proliferation inhibition at 320 μ L, while *P. barbatus* produces 55.55% cell proliferation when it was used at the same concentration with the IC₅₀ 89.13 and 81.50 of *C. forskolii* and *P. barbatus* extracts, respectively. Findings of Krishnamoorthy and Ramaswamy⁴¹ suggested that root ethanol extract of *C. forskolii* possesses potent anticancer properties against gastric cancer cell lines through the apoptosis induction. Gurgel *et al.*⁴² demonstrated that extract of *P. amboinicus* possesses anti-inflammatory and antitumor activities. Ferulic acid has an essential role as effective antioxidant and anticancer compound^{43,44}. Therefore present results in this study supporting the folk use of this medicinal plants.

Coleus forskohlii has antibacterial activities against *S. aureus*, *E. coli* and *S. typhi* and these effects were more prominent when using higher concentration of this species (Fig. 3). These findings are in accordance with the previous studies by Malleswari *et al.*⁴⁵, which indicated that the extracts of *C. forskohlii* roots, shoots and leaves have a potential broad spectrum antibacterial activity against *Bacillus subtilis*, *Pseudomonas fluorescence*, *P. aeruginosa*, *Klebsiella pneumoniae*, *Escherichia coli*, *Staphylococcus aureus* and *Streptococcus pneumoniae*. On the other hand, the antibacterial activity of *P. barbatus*, was negative even in higher concentration (Fig. 3, Table 5). A similar report has been made by Mwitari *et al.*⁴⁶. Although *P. barbatus* was not shown as an antimicrobial in this study, Lukhoba *et al.*⁵ reviewed that several species including *P. barbatus*, *P. amboinicus*, *P. fruticosus*, *P. ecklonii*, *P. lexfilorus*, *P. glandulosus* and *P. parviflorus* have antimicrobial and antiviral activities. Therefore, treatment of gastrointestinal, genitourinary and eye or ear infections was reported earlier by Wellsow *et al.*⁴⁷. Also,

Table 4: Antitumor activity of *Coleus forskohlii* and *Plectranthus barbatus* against HCT-116 cell line

Extract concentration (μ L)	<i>Coleus forskohlii</i>		<i>Plectranthus barbatus</i>	
	Viability (%)	Cell proliferation inhibition (%)	Viability (%)	Cell proliferation inhibition (%)
320	16.82	83.18	44.56	55.44
160	26.45	73.55	47.28	52.72
80	52.91	47.09	75.59	24.41
40	68.49	31.51	77.24	22.76
20	82.17	17.83	94.31	5.69
10	90.24	9.76	95.72	4.25
5	100.00	0.00	100.00	0.00
0	100.00	0.00	100.00	0.00
IC ₅₀ (μ L)	89.13		81.50	

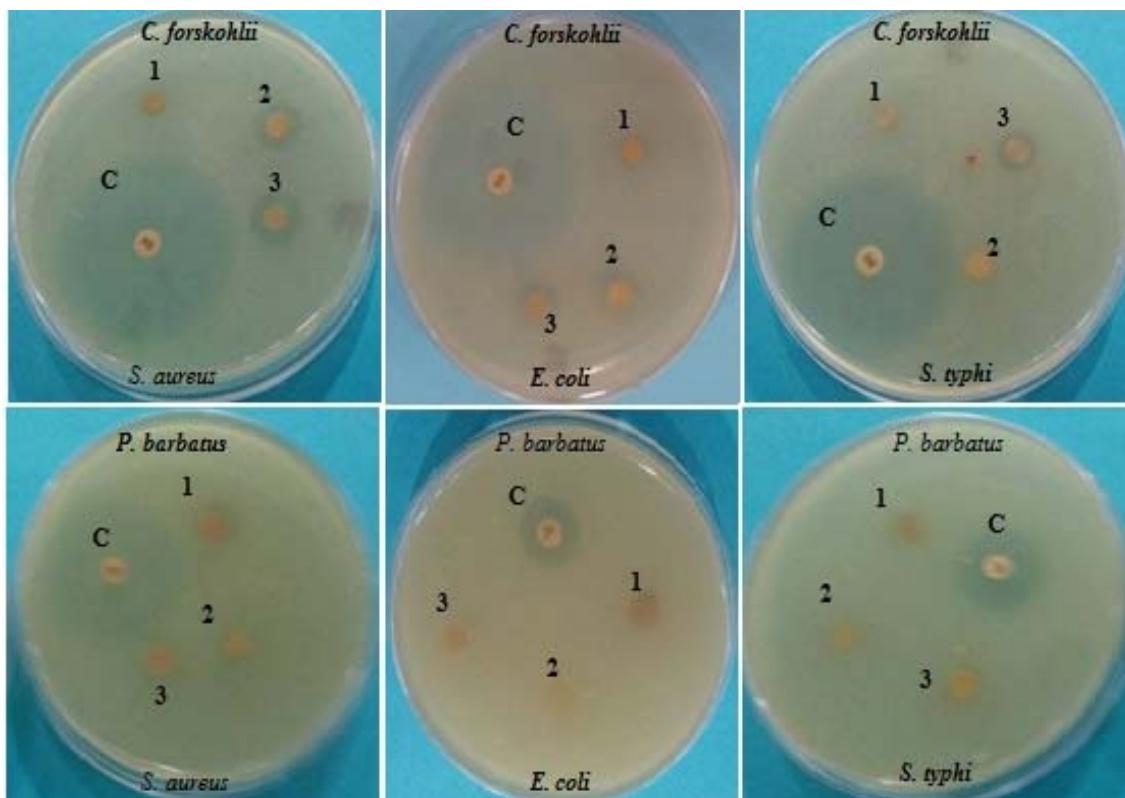


Fig. 3: Antibacterial activity of *C. forskohlii* and *P. barbatus* extracts at different concentrations on *S. aureus*, *E. coli* and *S. typhi* (1: 10, 2: 20, 3: 30 mg of extract, C: Positive control with using commercial antibiotic)

Table 5: Antibacterial activity of *C. forskohlii* and *P. barbatus* extracts on *S. aureus*, *E. coli* and *S. typhi*

Extract concentration (mg)	<i>Coleus forskohlii</i>			<i>Plectranthus barbatus</i>		
	<i>S. aureus</i>	<i>E. coli</i>	<i>S. typhi</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>S. typhi</i>
10	0.0	0.0	0.0	0.0	0.0	0.0
20	1.2	1.0	0.0	0.0	0.0	0.0
30	1.5	1.2	0.8	0.0	0.0	0.0

Table 6: Antifungal activity of *C. forskohlii* and *P. barbatus* extracts on *A. alternata* and *F. oxysporum*

Test fungi	<i>C. forskohlii</i> extract concentration mg/100 mL medium			<i>P. barbatus</i> extract concentration mg/100 mL medium		
	0	100	200	0	100	200
<i>A. alternata</i>						
Growth	6.5	4.20	3.80	6.5	4.50	4.30
Growth Inhibition (%)	0.0	35.38	41.53	0.0	30.76	33.84
<i>F. oxysporum</i>						
Growth	8.5	8.00	7.20	8.5	8.30	8.30
Growth Inhibition (%)	0.0	5.88	15.29	0.0	2.35	2.35

Matu and van Staden⁴⁸ stated that the methanolic extract of *P. barbatus* has potent antibacterial activity against +ve bacteria including *S. aureus* and antifungal effect against *Candida albicans*. Moreover, the fungal growth dramatically decrease with using *C. forskohlii* and *P. barbatus*, although the effect in the formal species was slightly more prominent than the later species (Table 6, Fig. 4). The growth

inhibition of *A. alternata* by *C. forskohlii* and *P. barbatus* reached as high as 41.53 and 33.84%, respectively. The results of Nidiry *et al.*⁴⁹ showed that forskolin is one of the antifungal compounds present in the *C. forskohlii*, where roots extracts exhibit mycelial growth inhibition of *Colletotrichum gloeosporioides* and spore germination inhibition of *Alternaria solani*.

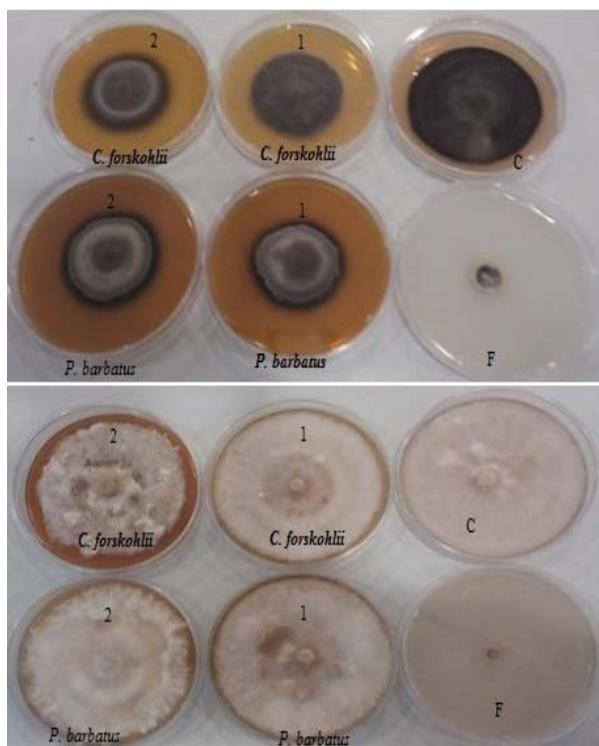


Fig. 4: Antifungal activity of *C. forskohlii* and *P. barbatus* extracts on *A. alternata* (Black colony) and *F. oxysporum* (White colony) (1: 100 mg, 2: 200 mg/100 mL medium, F: Chemical fungicide)

CONCLUSION

Antioxidant activity of *C. forskohlii* was lower than in *P. barbatus*. *Coleus forskolii* and *P. barbatus* have promising effect in tumor activities that appeared in its ability in inhibiting cell proliferation of HCT-116 cell line. *Coleus forskohlii* but not *P. barbatus* has antibacterial activities against *S. aureus*, *E. coli* and *S. typhi*. Therefore, the current results supporting the folk use of this medicinal plants. However, further study is needed to identify the other active ingredients in *C. forskohlii* and *P. barbatus* ate and action of their mechanism.

SIGNIFICANCE STATEMENT

The study evaluated the synergistic medicinal effect of *C. forskohlii* and *P. barbatus* in inhibiting the cell proliferation of HCT-116 cell line and found that *C. forskohlii* has potential antibacterial while *P. barbatus* has potential antioxidant activity in comparison of plants which is not explored before. This would help the researchers to

evaluate the comparative medicinal properties of both plants in detail but the best theory on it may be arrived at.

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