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**Original article**

# Tea polyphenols extracted from fresh young tea leaves exhibits antibacterial activity against some bacterial species: An *in vitro* study

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***Background:*** Natural products are rich sources of therapeutic agents for remedy, prevention and cure of diseases. Currently, many strains of bacteria have become resistant to many of the conventional antibiotics. So, in-depth researches should be focused on the antibacterial activity of natural compounds derived from natural sources.
***Objectives:*** The purposes of this study were to determine the antibacterial activity of polyphenols extracted from fresh young tea leaves against *Escherichia coli*, *Pseudomonas* spp., *Klebsiella* spp. and *Staphylococcus aureus*.
***Methods:*** Antibacterial activity of young and fresh leaves of tea was evaluated by disc diffusion method and well diffusion method. The results were reported in zone diameter in millimeter as mean ± standard deviation of triplicates (n = 3).
***Results:*** We found that polyphenols extracted from fresh young tea leaves showed potential inhibitory activity against bacterial growth. The diameters of the inhibition zone found for extracted polyphenols from young fresh tea leaves showed satisfactory antibacterial activity against the selected bacterial isolates. In disc diffusion method, polyphenol extracts have shown effective results against *Pseudomonas* spp. (19.33 ± 2.52 at 100 mg/ml concentration) and *Staphylococcus aureus* (18.00 ± 3.61 at 75 mg/ml concentration). In well diffusion method, the polyphenol extracts have shown effective results against *Pseudomonas* spp. (17.00 ± 2.65 at 100 mg/ml) and *E. coli* (15.00 ± 2 at 100 mg/ml).
***Conclusion:*** Polyphenols extracted from fresh young tea leaves have potential inhibitive effects against growth of the selected bacterial isolates and can be used as an antibacterial agent.

***Keywords:*** Antibacterial activity, polyphenols, tea leaves, inhibitive effects, bacterial isolates.

As many strains of bacteria are becoming resistant to the conventional antibiotics due to climate change, many researchers are now focusing their investigations on the antibacterial activities of natural products as an emerging source of antibacterial compounds.[1] Tea is the most widely consumed beverage all over the world. It is refreshing, highly stimulating, has a wide range of beneficial physiological and pharmacological effects as well as considerable nutritional value. Drinking green tea regularly has been associated with numerous health benefits.[2] Consumption of green tea is linked to lower incidences of cardiovascular disease, strokes, obesity and cancer. Many of the flavanols compounds found in green tea have a direct antibacterial effect on human health.[3] Numerous investigations have been conducted on the chemical constituents and biological properties of tea

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because of the potential health benefits of compounds present in tea.[4 – 7] Tea can inhibit and kill a wide range of pathogenic bacteria at or slightly below typical concentrations of brewed tea. Tea polyphenols are reported to prevent cancer and cardiovascular diseases and have antibacterial, antiviral, antifungal, anti-mutagenic and anti-carcinogenic activities. [8, 9] The anti-mutagenic and anti-carcinogenic activities of tea polyphenols are mostly due to their powerful scavenging and antioxidant activity, which inactivates direct carcinogens and inhibits the activation of indirect carcinogens extracellularly. Polyphenols are used to treat a wide range of diseases such as diabetic retinitis, ecchymosis, gingivitis, pyorrhea, hemorrhoids, menorrhagia, Rh incompatibility, influenza etc. [10] The polyphenols present in tea leaves constitute the catechin group of the flavanols. There are six flavanol compounds present in fresh tea leaves in high concentrations. They are (+) catechin, (-) epicatechin, (+) gallocatechin, (-) epigallocatechin, (-) epicatechingallate, and (-) epigallocatechingallate. [11] The catechin components of tea are responsible for the observed antibacterial activity. Epigallocatechin, epicatechingallate and epigallocatechingallate constitute the most important antibacterial agents present in tea. [12] Epigallocatechingallate present in young fresh tea leaves can induce apoptotic cell death, can prevent cell cycle arrest in tumor cells and favorably affects several signal transduction pathways and is efficacious in animal models of tumor induction.[13] Aqueous extracts of different types of tea from various sources inhibited a wide range of pathogenic bacteria, including methicillin-resistant bacteria. Epicatechingallate and epigallocatechingallate markedly lowers the minimum inhibitory concentrations of methicillin, oxacillin and other □-lactam antibiotics in clinical isolates of methicillin-resistant *Staphylococcus aureus*, given the fact that *Staphylococcus aureus* is one of the major causes of both nosocomial and community-acquired infections worldwide.[14] The 'anti-methicillin-resistant *Staphylococcus aureus'* properties of naturally occurring, semi-synthetic catechins are suitable as topical agents for the treatment of superficial bacterial infections at minimum inhibitory concentrations ranged from 64 - 256 mg/l.[15] The bactericidal action of catechins against methicillin-resistant *Staphylococcus aureus* can be notably improved by substitution of the gallate moiety of epicatechingallate with 3-*O*-acyl chains of various lengths. [16] Tea extracts were bactericidal to *Staphylococcus* and *Yersinia enterocolitica*at well below 'cup of tea' concentrations. The extraction of tea polyphenols inhibited and killed *Staphylococcus epidermidis, Staphylococcus mutanas*, *Salmonella typhi, Salmonella typhimurium, Shigella flexneri* and *Vibrio* spp. [8, 10, 17 – 20]

Green tea can reduce the teeth plaque index significantly and can be used as mouth rinse. [6, 21] Currently, many strains of bacteria have become resistant to many of the conventional antibiotics. So, in-depth researches should be focused on the antibacterial activity of natural compounds derived from natural sources such as tea leaves. Several studies have determined the antibacterial activity of tea polyphenols extracted from made tea [8 - 10, 15, 18, 19, 22, 23], but no study has been conducted till date on the antibacterial activity of tea polyphenols extracted from young fresh tea leaves to the best of our knowledge. Keeping these in mind, the current study was conducted to determine the antibacterial activity of polyphenol extracted from fresh young tea leaves against some gram negative bacteria such as *E. coli, Klebsiella* spp*., Pseudomonas* spp. and gram positive bacteria *Staphylococcus aureus*.

## Materials and methods

This experiment was conducted in the laboratories of Department of Food Engineering & Tea Technology and Department of Genetic Engineering & Biotechnology, Shahjalal University of Science and Technology, Sylhet-3114.

### *Sample collection*

Young fresh tea leaves were collected from Malnichara Tea Estate the oldest tea garden of Bangladesh.

### *Sterilization of glassware*

The required glassware was washed thoroughly, cleaned, dried and wrapped with paper and plugged with cotton wool where necessary and sterilized in a hot air oven at 180°C for 60 min before using.

### *Sample preparation*

After collection, the young fresh tea leaves were washed thoroughly in running water to remove dirt and other unwanted materials. Then, they were rinsed with 70.00% ethanol and spread under a net at room temperature (30°C) with ceiling fan running for

overnight to reduce the moisture content up to the minimal level. The young fresh tea leaves were kept in this condition until brittle like structures were found on the texture of the tea leaves. When the tea leaves have dried, they were grounded by using a mechanical blender to obtain powder. The powder of young fresh tea leaves were then stored into a air tight container at room temperature (30°C).

***Polyphenols extraction***

The extraction of polyphenol from the powder of young fresh tea leaves was carried out by using 80.00% hot methanol. 30 grams dried tea leaf powder was weighed and kept into a clean, dried and sterilized 250 ml conical flask. 90 ml of 80.00% hot methanol was added to the flask in 1:3 ratio (g/ml). The flask was then swirled with vortex machine for 3 times to mix the powder. Then the mixture was centrifuged at 4,000 rpm for 15 minutes. The liquid phase was filtered with Whatman number 1 filter paper. The supernatant crude extract was collected while the pellet being discarded. The crude extract was then condensed with vacuum rotary evaporator at temperature of 45°C- 50°C in a reduced pressure to extract bioactive compounds. The extracted bioactive compounds were further separated into free flavonoid and bound flavonoid by liquid separation.

***Water-hexane separation***

In a 250 ml of separation funnel, 150 ml of distilled water was added with dried crude extract which contained numerous non-phenolic compounds like fats, terpenes, and pigments etc. addition of 60 ml of hexane in the flask separated with all non-phenolic compounds from the water as they are immiscible solvent. The hexane was discarded and remaining portion was collected into a conical flask.

***Water-chloroform separation***

The separated extract might contain chlorophyll and caffeine along with polyphenols. 60 ml of chloroform was added with the extract into a separation funnel which then separated with caffeine and chlorophyll as water and chloroform are two immiscible mixtures. The portion of chloroform was discarded. The remaining portion contained only polyphenols.

***Water-ethyl acetate separation***

The extracted polyphenols were purified by addition of 60 ml, 2.00% acidic ethyl acetate into separation funnel. In this case only 8.00% ethyl acetate dissolved in water while leaving content separated from water. The portion of ethyl acetate was discarded. Finally, the remaining extract was evaporated with rotary vacuum evaporator at the temperature of 45°C - 50°C. Thus, extracted pure polyphenols was taken into a biker covered with aluminum foil tightly and preserved it into the freeze.

***Collection of the sub-cultures of bacterial isolates***

The sub-cultures of gram negative bacteria *Escherichia coli*, *Pseudomonas* spp., *Klebsiella* spp., and gram positive *Staphylococcus aureus* were collected from Laboratory of Department of Food Engineering and Tea Technology.

***Preparation of medium for the assessment of antibacterial activity***

Nutrient broth is a liquid medium formulation used for cultivation and optimization of a wide variety of microorganism. Nutrient broth was freshly prepared and autoclaved before use. Muller-Hinton agar (MHA) is considered to be the best for routine susceptibility testing of non-fastidious bacteria. Muller-Hinton agar was prepared from a commercially available dehydrated base according to the manufacturer's instructions.

***Assessment of antibacterial activity***

Disc diffusion technique (also known as Kirby Bauer method) and agar well diffusion technique was used for the *in vitro* investigation of antibacterial activity of young fresh tea leaves against selected isolates. All the identified isolates were subjected to various concentrations of crude extracts of young fresh tea leaves for the evaluation of antibiotic activity pattern. The bacterial culture was streaked on MHA plate. Blank discs containing crude extract were then placed on the inoculated plate surface and incubated at 37°C for 24 hours. After incubation the zone of inhibition around each disc was measured in millimeters.

***Disc diffusion method***

From an agar plate of pure culture, 5 colonies of the same morphological type were selected. The top of each colony was touched with a loop and the growth was transferred Into a tube containing 4 - 5 ml of nutrient broth. The broth culture was then incubated at 37°C to attain the preferable turbidity. A sterile cotton swab was dipped into the suspension. Excess

fluid was removed by pushing and rotating the swab firmly against the wall just above the fluid level. The entire dried surface of MHA plate was streaked by the swab 2 - 3 times resulting in an even distribution of the inoculum over the entire surface. For preparation of test solution of appropriate concentration, crude extracts were solubilized to distilled water and diluted to desired concentration by adding distilled water.

For disc preparation, blank discs prepared by filter paper were used. Using a micropipette, dried and sterilized blank discs were treated separately with desired concentration of previously prepared test solution. Under aseptic condition, these prepared discs were air dried. After preparation, discs of different test concentration were then stored in vials at 4°C. The predetermined battery of anti-microbial discs was dispensed on to the surface of the inoculated agar plates evenly so that they were no closer than 24 mm from center to center. Each disk was pressed down to ensure complete contact with the agar surface.

The plates were then inverted and placed in an incubator at 35°C within 15 minutes after the discs were applied. After 16 - 18 hours of incubation, each plate was examined. There were uniformly circular zones of inhibition on the surface. The diameter of the discs around zones were measured using a ruler to nearest whole millimeter.

### *Agar well diffusion method*

In agar well diffusion test, crude extracts were allowed to diffuse into the medium. The antibacterial compounds present in the extracts interacted with the freshly seeded test organisms. This interaction resulted in a uniform circular zone of inhibition. The diameter of the inhibition zone was measured in millimeters. From an agar plate of pure culture, 4 colonies of the same morphological type were selected. The top of colony was touched with a loop and the growth was transferred into a tube containing 4 - 5 ml of nutrient broth. The broth culture was then incubated at 37°C to attain the preferable turbidity. A sterile cotton swab was dipped into the suspension. Excess fluid was removed by pushing and rotating the swab firmly against the wall, just above the fluid level. The entire dried surface of MHA plate was streaked by the swab 2 - 3 times resulting in an even distribution of the inoculum over the entire surface.

For preparation of test solution of appropriate concentration, crude extracts were solubilized to distilled water and diluted to desired concentration by adding distilled water. Using a micropipette, 100 µl of test samples of different concentrations were poured into the well. The plates were kept in refrigerator for 30 minutes for diffusion of test solution to the surrounding media. The plates were then inverted and placed in an incubator at 35°C. After 18 hours of incubation, each plate was examined. There were uniformly circular zones of inhibition around the well on the surface. The diameter of the complete zone of inhibition was measured by unaided eye. The diameter of the discs and zones were measured to nearest whole millimeter using a ruler.

### *Statistical analysis*

Data were analyzed using SPSS software (SPSS Inc., Chicago, IL, USA), version 25 for Windows. The results are reported as mean ± standard deviation (SD) of triplicates (n = 3). One-way analysis of variance (ANOVA) followed by Duncan Multiple Range Test (DMRT) (multiple comparison post-hoc test) were used to analyze the statistical difference. Differences with $P < 0.05$ were considered statistically significant.

## Results

The mean flavanol content of polyphenols extracted from fresh young tea leaves was found to be 13.05 ± 0.17% w/w as given in Table 1. The estimation of flavanol content was carried out by taking 10 gm, 20 gm and 30 gm of tea leaf powder. The flavanol content (as % w/w) for 10 gm, 20 gm and 30 gm of tea leaf powder was found to be 13.21% w/w, 12.87% w/w and 13.07% w/w respectively and their mean was calculated to find out the mean flavanol content of the fresh young tea leaves.

**Table 1.** Total flavanol content of young fresh tea leaves.

| Experiment No. | Amount of powder (gm) | Flavanol content (gm) | Flavanol content (% w/w) | Mean flavanol content (% w/w) |
|---|---|---|---|---|
| 1 | 10 | 1.32 | 13.21 | 13.05 ± 0.17 |
| 2 | 20 | 2.57 | 12.87 | |
| 3 | 30 | 3.92 | 13.07 | |

The antibacterial activities of polyphenols extracted from fresh young tea leaves against selected bacterial species in well diffusion method has been represented in Figure 2 and given in Table 2. 50 mg/ml, 75 mg/ml and 100 mg/ml concentrations of polyphenols extracted from fresh young tea leaves were prepared. 100 µl of each concentration was kept into their corresponding well. At 100 mg/ml concentration, the polyphenol extracts showed better zone of inhibition against *Escherichia coli* (15.00 ± 2.00 mm), *Pseudomonas* spp. (17.00 ± 2.65 mm), *Staphylococcus aureus* (12.00 ± 1.00 mm) and *Klebsiella* spp. (13.33 ± 1.53 mm). Polyphenols extracted from fresh young tea leaves inhibited the bacterial growth of all of the selected bacterial isolates. Growth inhibition of *Pseudomonas* spp. was highest 17.00 ± 2.65 mm zone of inhibition) at 100 mg/ml of polyphenol concentration.

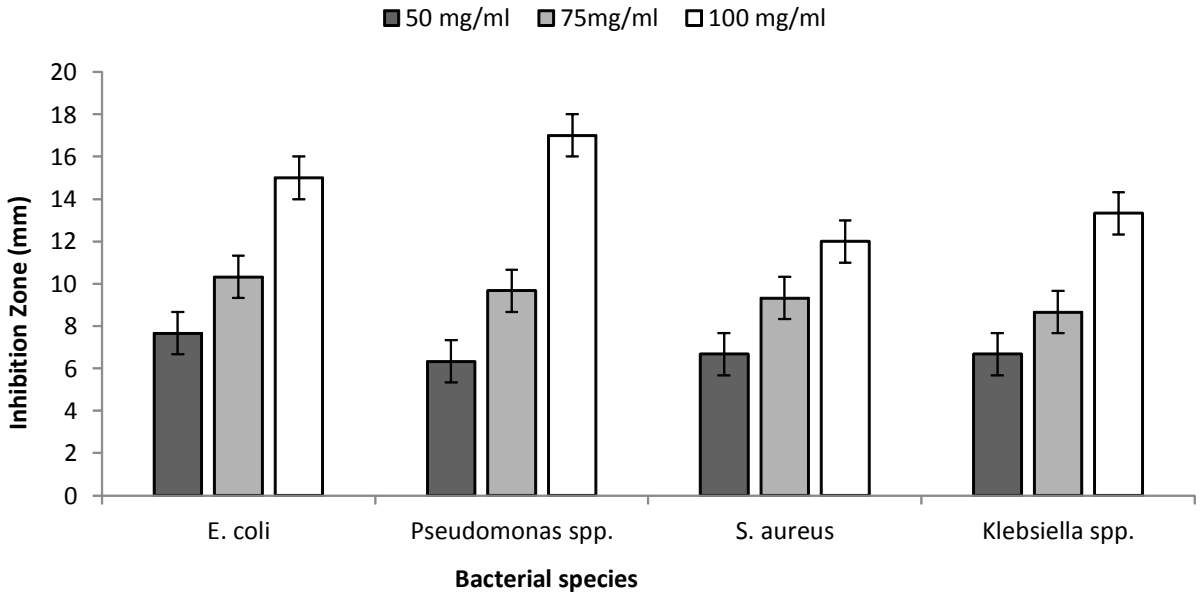



**Figure 1.** Antibacterial activity of polyphenols extracted from fresh young tea leaves against selected bacterial species in well diffusion method.

**Table 2.** Zone of inhibition (mm) of selected bacterial species in well diffusion method.

| Bacterial isolates | Concentrations of polyphenol extracts (mg/ml) | Experiment 1 | Experiment 2 | Experiment 3 | Zone of inhibition (mm) |
|---|---|---|---|---|---|
| *E. coli* | 50 | $8 \pm 1.0^{bc}$ | $7 \pm 1.0^{ab}$ | $8 \pm 1.0^{bc}$ | $7.67 \pm 0.58^{abc}$ |
| | 75 | $11 \pm 1.0^{ef}$ | $10 \pm 1.0^{c}$ | $10 \pm 1.0^{de}$ | $10.33 \pm 0.58^{cd}$ |
| | 100 | $15 \pm 1.0^{gA}$ | $17 \pm 1.0^{fB}$ | $13 \pm 1.0^{gA}$ | $15 \pm 2^{fgAB}$ |
| *Pseudomonas* spp. | 50 | $6 \pm 1.0^{a}$ | $6 \pm 1.0^{a}$ | $7 \pm 1.0^{ab}$ | $6.33 \pm 0.58^{a}$ |
| | 75 | $10 \pm 1.0^{deB}$ | $7 \pm 1.0^{abA}$ | $12 \pm 1.0^{fgB}$ | $9.67 \pm 2.52^{cdAB}$ |
| | 100 | $16 \pm 1.0^{gA}$ | $20 \pm 1.0^{gB}$ | $15 \pm 1.0^{hA}$ | $17 \pm 2.65^{gA}$ |
| *Staphylococcus aureus* | 50 | $6 \pm 1.0^{a}$ | $7 \pm 1.0^{ab}$ | $7 \pm 1.0^{ab}$ | $6.67 \pm 0.58^{ab}$ |
| | 75 | $11 \pm 1.0^{ef}$ | $8 \pm 1.0^{b}$ | $9 \pm 1.0^{cd}$ | $9.33 \pm 1.53^{bcd}$ |
| | 100 | $12 \pm 1.0^{f}$ | $13 \pm 1.0^{d}$ | $11 \pm 1.0^{ef}$ | $12 \pm 1^{de}$ |
| *Klebsiella* spp. | 50 | $7 \pm 1.0^{ab}$ | $7 \pm 1.0^{ab}$ | $6 \pm 1.0^{a}$ | $6.67 \pm 0.58^{ab}$ |
| | 75 | $9 \pm 1.0^{cdAB}$ | $10 \pm 1.0^{cB}$ | $7 \pm 1.0^{abA}$ | $8.67 \pm 1.53^{abcAB}$ |
| | 100 | $12 \pm 1.0^{fA}$ | $15 \pm 1.0^{eB}$ | $13 \pm 1.0^{gAB}$ | $13.3\,3 \pm 1.53^{efAB}$ |

Values represent the mean ± standard deviation of 3 replicates (n = 3); $^{abcdef}$column - means within the different superscript letter were significantly different ($P < 0.05$) through one-way ANOVA followed by DMRT; $^{ABCDEF}$row - means within the different superscript letter were significantly different ($P < 0.05$) through one-way ANOVA followed by DMRT.

The antibacterial activities of polyphenols extracted from fresh young tea leaves against selected bacterial species in disc diffusion method has been depicted in Figure 3 and given in Table 3. 50 mg/ml, 75 mg/ml and 100 mg/ml concentrations of polyphenols extracted from fresh young tea leaves were prepared. Better zones of inhibition were found against *E. coli* (12.33 ± 1.53 mm) at 50 mg/ml concentration, *Pseudomonas* spp. (19.33 ± 2.52 mm) at 100 mg/ml concentration, *S. aureus* (18.00 ± 3.61 mm) at 75 mg/ml concentration and *Klebsiella* spp. (15.67 ± 5.03 mm) at 50 mg/ml concentration. Polyphenols extracted from fresh young tea leaves inhibited the bacterial growth of all of the selected bacterial isolates. Growth inhibition of *Pseudomonas* spp. was highest (19.33 ± 2.52 mm zone of inhibition) at 100 mg/ml concentration of polyphenol.

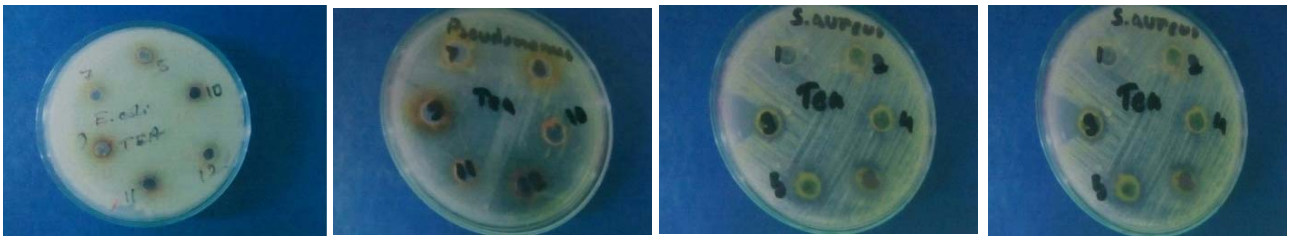



**Figure 2.** Bacterial colony formation of **(a)** *Escherichia coli* **(b)** *Pseudomonas* spp. **(c)** *Staphylococcus aureus* **(d)** *Klebsiella* spp. in well diffusion method against polyphenol extracts of fresh young tea leaves.

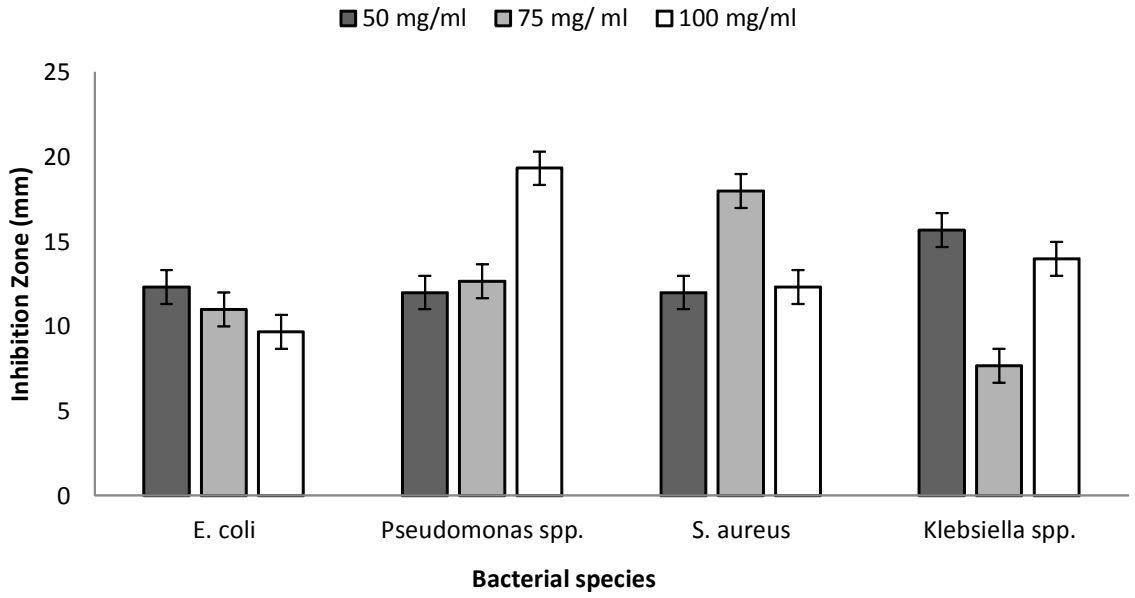
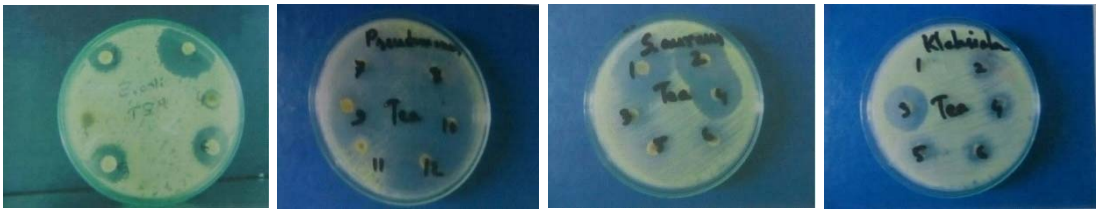



**Figure 3.** Antibacterial activity of polyphenols extracted from fresh young tea leaves against selected bacterial species in disc diffusion method.

**Table 3.** Zone of inhibition (mm) of selected bacterial species in disc diffusion method.

| Bacterial isolates | Concentrations (mg/ml) | Experiment 1 | Experiment 2 | Experiment 3 | Zone of inhibition (mm) Mean ± SD |
|---|---|---|---|---|---|
| *E. coli* | 50 | $12 \pm 1.0^{cAB}$ | $14 \pm 1.0^{deB}$ | $11 \pm 1.0^{bA}$ | $12.33 \pm 1.53^{abcAB}$ |
| | 75 | $9 \pm 1.0^{bA}$ | $12 \pm 1.0^{cB}$ | $12 \pm 1.0^{bcB}$ | $11 \pm 1.73^{abcAB}$ |
| | 100 | $10 \pm 1.0^{bAB}$ | $8 \pm 1.0^{aA}$ | $11 \pm 1.0^{bB}$ | $9.67 \pm 1.53^{abAB}$ |
| *Pseudomonas* spp. | 50 | $9 \pm 1.0^{bA}$ | $12 \pm 1.0^{cAB}$ | $15 \pm 1.0^{eB}$ | $12 \pm 3^{abcAB}$ |
| | 75 | $12 \pm 1.0^{c}$ | $14 \pm 1.0^{de}$ | $12 \pm 1.0^{bc}$ | $12.67 \pm 1.15^{abc}$ |
| | 100 | $22 \pm 1.0^{eB}$ | $17 \pm 1.0^{fA}$ | $19 \pm 1.0^{gA}$ | $19.33 \pm 2.52^{eAB}$ |
| *Staphylococcus aureus* | 50 | $15 \pm 1.0^{dB}$ | $7 \pm 1.0^{aA}$ | $14 \pm 1.0^{deB}$ | $12 \pm 4.36^{abc}$ |
| | 75 | $22 \pm 1.0^{eB}$ | $15 \pm 1.0^{eA}$ | $17 \pm 1.0^{fA}$ | $18 \pm 3.61^{deA}$ |
| | 100 | $10 \pm 1.0^{bA}$ | $12 \pm 1.0^{cA}$ | $15 \pm 1.0^{eB}$ | $12.33 \pm 2.52^{abcAB}$ |
| *Klebsiella* spp. | 50 | $21 \pm 1.0^{eB}$ | $15 \pm 1.0^{eA}$ | $11 \pm 1.0^{bA}$ | $15.67 \pm 5.03^{cdeA}$ |
| | 75 | $5 \pm 1.0^{aA}$ | $10 \pm 1.0^{bB}$ | $8 \pm 1.0^{aB}$ | $7.67 \pm 2.52^{aAB}$ |
| | 100 | $16 \pm 1.0^{dB}$ | $13 \pm 1.0^{cdA}$ | $13 \pm 1.0^{cdA}$ | $14 \pm 1.73^{bcdAB}$ |



**(a)** **(b)** **(c)** **(d)**

**Figure 4.** Bacterial colony formation of **(a)** *Escherichia coli* **(b)** *Pseudomonas* spp. **(c)** *Staphylococcus aureus* **(d)** *Klebsiella* spp. in well diffusion method against Polyphenol extracts of fresh young tea leaves.

Values represent the mean ± standard deviation of 3 replicates (n = 3); $^{abcdef}$ column - means within the different superscript letter were significantly different ($P < 0.05$) through one-way ANOVA followed by DMRT; $^{ABCDEF}$ row - means within the different superscript letter were significantly different ($P < 0.05$) through one-way ANOVA followed by DMRT.

## Discussion

Infections caused by drug resistant bacteria has become a major public health issue both in developing and developed countries.[24] Conventionally, synthetic antibiotics are being used for a long time to treat infections and inhibit pathogenic organisms. But synthetic antibiotics can cause serious side effects.[25] Currently, a large number of life threatening pathogenic bacterial species have associated with resistivity towards synthetic antibiotics. [26] Treatment of infections are becoming more and more difficult due to the resistance of bacteria with a large group of synthetic antibiotics or simply because of high cost of antibiotics. [27] So, more and more studies should be focused to find new source of natural compounds with antibacterial activity.[28] In our study, we found that flavanol content of polyphenols present in fresh young tea leaves was 13.05 ± 0.17% (w/w). This finding indicates that there is a potentiality for fresh young tea leaves to be used as an antibacterial therapeutic agent. We assessed the antibacterial activity of polyphenol extracted from fresh young tea leaves based on the measurement of the inhibition zones formed around the disc and wells. In well diffusion method, comparatively better bacterial inhibition was observed against *Pseudomonas* spp. (zone of inhibition: 17.00 ± 2.65 mm) and *E. coli* (zone of inhibition: 15.00 ± 2.00) mm Although previous studies have reported good performance of tea extract to inhibit the growth of *Staphylococcus aureus* [14 - 17, 20], bacterial inhibition of polyphenol

extracts was less effective against *Staphylococcus aureus* (zone of inhibition: 12.00 ± 1.00 mm). This difference in results could be due to different concentrations and types of extracts investigated. Study on disc diffusion method showed potential antibacterial activity of polyphenol extracts against *Pseudomonas spp.* (zone of inhibition: 19.33 ± 2.52 mm) and *Staphylococcus aureus* (zone of inhibition: 18.00 ± 3.61 mm). Comparatively less effective antibacterial activity but yet potential inhibition effect was observed against *Klebsiella* spp. (zone of inhibition: 15.67 ± 5.03 mm) and *E. coli* (zone of inhibition: 12.33 ± 1.53 mm). Similar findings were reported by Jazani NH, *et al.* [29] and Amarowicz R, *et al.* [23] for *Pseudomonas aeruginosa* and *Escherichia coli* K12 respectively. The results of our study suggest that the polyphenol extracted from fresh young tea leaves showed better antibacterial activity than that of green tea seed isolated saponins, green tea extract, oolong tea extract or black tea as previously reported. [5, 8 - 10, 15, 18 – 23] Polyphenols extracted from fresh young tea leaves showed promising antibacterial activity against several multi-drug resistant bacteria namely *Pseudomonas* spp., *Staphylococcus aureus*, *E. coli and Klebsiella* spp.

## Conclusion

In our study, we observed potential antibacterial activity of polyphenols extracted from fresh young tea leaves against some selected gram negative and gram positive bacterial species such as *Escherichia coli, Pseudomonas* spp., *Klebsiella* spp. *and Staphylococcus aureus*. The findings of our study indicate that polyphenols extracted from fresh young tea leaves may be used as a therapeutic agent against these bacteria.

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## Conflicts of interest

No authors have any potential conflict of interest to disclose.

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