



ANTIBACTERIAL EFFECT OF ETHANOL EXTRACT OF CAMELLIA SINENSIS L AGAINST ESCHERICHIA COLI

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ABSTRACT

The aim study was carried out to determine the potential antibacterial effect of ethanol extract of *Camellia sinensis* L against *E. coli*. All 30 strains of *E. coli* isolated from urine culture of hospitalized patients Hospital (Zabol, south-eastern Iran) suffered from urinary tract infections during the years 2011- 2012 and the minimum inhibitory concentrations were investigated by microdilution method. The result show that the resistant antibiotics against the isolates were nalidixic acid (41.17%), ceftazidime (41.17%) and ampicillin (58.92%). To determine the lowest concentration that completely inhibited visible growth, the minimum inhibitory concentration (MIC) was used. The MIC of *Camellia sinensis* against *E. coli* were shown in Table 1, showing that the highest MIC of *Camellia sinensis* against *E. coli* was 10mg/ml.

KEYWORDS

Antibacterial activity, camellia sinensis, antibiotic resistant, escherichia, coli.

1. INTRODUCTION

Biological Since ancient times, plants have been an exemplary source of medicine. Tea is one of the most popular beverages consumed worldwide. Plants contain chemical constituents that have great potential for medicinal use and both traditional healers and pharmaceutical drug companies make use of these plants [1]. *Camellia sinensis* is the species of plant whose leaves and leaf buds are used to produce Chinese tea. *Camellia sinensis*, is consumed in different parts of the world as green, black, or Oolong tea. Among all of these, however, the most significant effects on human health have been observed with the consumption of green tea [2]. Tea from the leaves of plant *Camellia Sinensis* has a wide range of antioxidant, anti-inflammatory and anticarcinogenic activity. Based on a study, green tea extracts are more stable than pure epigallocatechin gallate, one of the major constituents of green tea, because of the presence of other antioxidant constituents in the extract [3]. Tea contains many different compounds that grant it health-promoting properties, including a group of polyphenolic compounds called flavonoids. The most abundant class of these flavonoids is the flavonols. According to a research, the plant materials have shown the antimicrobial activities against various pathogenic microorganisms therefore consumption of tea has been associated with reduced risk of major diseases [4]. Urinary tract infections are serious health problem affecting millions of people each year. Generally there is an agreement among the authors in the literature that the predominant uropathogens acquired from any source are gram negative bacteria with *Escherichia coli* accounting for the highest prevalence in most instances [5]. The aim study was carried out to determine the potential antibacterial effect of ethanol extract of *Camellia sinensis* L against *E. coli*.

2. ISOLATION OF E. COLI

In this descriptive research, a total number of 30 non repetitive clinical isolates *E. coli* were collected between May 2011 and December 2012 from hospitalized patient in three major hospitals in Zabol, south-eastern Iran. The isolates were obtained from the cultures of urine. Each sample was streaked on the blood and MacConkey agar medium and incubated at 37°C for 24 hour after incubation, *E. coli* isolates were detected by standard biochemical tests such as indole, methyl red, Voges-Proskauer, and citrate.

2.1 Plant materials

The leaf of *Camellia sinensis* was collection in the region of Iran and plant in Zabol university received approval and dried at room temperature. Samples were crashed and transferred into glass container and preserved until extraction procedure was performed in the laboratory.

2.2 Minimum Inhibitory Concentration (MIC)

The broth microdilution method was used to determine MIC. Briefly, serial doubling dilutions of the silver nanoparticles produced in the plant *Camellia sinensis* leaf extract were prepared in a 96-well microtiter plate ranged from 12.5 µl/ml to 200 µl/ml. To each well, 10 µl of indicator solution and 10 µl of Mueller Hinton Broth were added. Finally, 10 µl of bacterial suspension (10⁶ CFU/ml) was added to each well to achieve a concentration of 10⁴ CFU/ml. The plates were wrapped loosely with cling film to ensure that the bacteria did not get dehydrated. The plates were prepared in triplicates, and then they were placed in an incubator at 37°C

for 18-24 hours. The color change was then assessed visually. The lowest concentration at which the color change occurred was taken as the MIC value. The MIC is defined as the lowest concentration of the extract at which the microorganism does not demonstrate the visible growth. The microorganism growth was indicated by turbidity.

3. RESULT

In the study, the minimum inhibitory concentration (MIC) was used. The MIC of *Camellia sinensis* against *E. coli* were shown in Table 1, showing that the highest MIC of *Camellia sinensis* against *E. coli* was 10 mg/ml.

Table 1: Antimicrobial susceptibility and mic (mg/ml)

Bacterial	MIC(mg/ml)	Bacterial	MIC(mg/ml)
<i>E.coli</i> 1	2.5	<i>E.coli</i> 7	5
<i>E.coli</i> 2	5	<i>E.coli</i> 8	10
<i>E.coli</i> 3	1.25	<i>E.coli</i> 9	2.5
<i>E.coli</i> 4	10	<i>E.coli</i> 10	5
<i>E.coli</i> 5	10	<i>E.coli</i> 11	5
<i>E.coli</i> 6	10	<i>E.coli</i> 12	10

4. DISCUSSION

In the study the MIC of *Camellia sinensis* against *E. coli* were shown in Table 1, showing that the highest MIC of *Camellia sinensis* against *E. coli* was 10 mg/ml. Chou demonstrated that dry tea flush and green tea, in particular, are capable of killing bacteria. Dry tea proved most effective against *Bacillus subtilis*, *Proteus vulgaris* and *Staphylococcus aureus*, but green tea demonstrated better performance against *Escherichia coli* and *Salmonella* [6]. Study showed that tea extract was effective against *S. aureus* at 150 L concentration where the zone of inhibition was observed at 27 mm. 200 L and 250 L concentration of the tea extract was needed to produce a significant antibacterial effect against microorganisms like *S. typhi* and *P. aeruginosa* and the zone of inhibition was observed at 24.1 and 18.1 mm respectively [7]. Research showed the result show that the inhibition zone diameter of green tea extracts for *S. aureus* ATCC25923 and MRSA were (18.970±0.287) mm and (19.130±0.250) mm respectively [8]. While the inhibition was diameter for *P. aeruginosa* ATCC27853 and *P. aeruginosa* were (17.550±0.398) mm respectively [8]. Several previous studies have shown that green tea extract showed activity against both-MRSA and methicillin-sensitive *Staphylococcus aureus* and against MDR- *P. aeruginosa* [9-11]. In this work different concentration of leaf extracts were used against different pathogenic bacteria and highest zone of inhibition was observed against *Bacillus subtilis* and *Enterococcus* sp [12]. The study of Tiwari, chloramphenicol and tea extract in combination inhibited the growth of *S. dysenteriae* at 2.5 µg/ml chloramphenicol (MIC 5 µg/ml) and 5.094 mg/ml black tea extract (MIC 9.089 mg/ml). based on a study, tea extract showed synergistic activity with chloramphenicol and other antibiotics like gentamycin, methicillin and nalidixic acid against test strains [13]. A researcher found that extracts of tea inhibited and killed *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Salmonella typhi*, *Salmonella typhimurium*, *Salmonella enteritidis*, *Shigella flexneri*, *Shigella dysenteriae*, and *Vibrio* spp., including *Vibrio cholera* [14]. The study of Chye, extracts producing inhibition zone of more than 8.0 mm were further investigated to determine their minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC). According to a study, hydrolysed flavonoid of dried samples was the most active extract against *E. coli* O157:H7 and *L. monocytogenes* with inhibition zone of 16.0 ± 1.4 mm and 22.0 ± 1.4 mm respectively [15]. In conclusion, this experiment has shown that green tea in general does not have antimicrobial properties. Almost all of the tea samples tested do not seem to inhibit bacterial growth. However, since the loose tea does show some effectiveness in inhibiting bacterial growth, the results do support our hypothesis that the loose tea would be the most effective inhibitor.

AUTHORS' CONTRIBUTIONS

All authors had equal role in design, work, statistical analysis and manuscript writing.

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