A Comparison Study On Anti-Microbial Activities Of Green Tea And Black Tea Leaves (*Camellia Sinensis L)* Extract

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ABSTRACT:
Tea comes loaded with antioxidants and compounds like polyphenols and catechins that can fend off free radicals and reduce the risks of chronic diseases. Many tea leaves also come stuffed full of vitamins and minerals along with anti-inflammatory properties and immune system boosters. Tea is not only a popular drink but also a drink with refreshing and functional properties. Thus, the green and black tea leaves were collected and used for preparing green and orthodox black tea to study Anti-microbial activity. The contemporary scientific community has presently recognized flavonoids to be a unique class of therapeutic molecules due to their diverse therapeutic properties. Of these, rutin, also known as vitamin P or rutoside, has been explored for a number of pharmacological effects. Tea leaves, apples, and many more possess rutin as one of the active constituents. Today, rutin has been observed for its nutraceutical effect. The present study highlights the anti-microbial effects of green and black tea leaves extract.

Keywords: Green and Black tea leaves, Antimicrobial, E.Coli, S.aureus, S.typhi, K.pneumoniae.

INTRODUCTION:
Tea plant (*Camellia sinensis L.*) is such a source to provide us a tea brew that's so refreshing and further, it is a popular drink in the world. It has been well defined as an infusion aqueous and as well as hot of dried leaves. That is to say, it is all an extract of leaves, leaf nodes, and further as of inter-nodes of the plant (*Camellia sinensis L.*) (1). They have not only the stimulating effect but also therapeutic properties because of polyphenolic compounds present in them. It seems drinking black tea has indeed benefits equal to those of drinking green tea in terms of their antioxidant capacities and such is the case, because the flavins that are available in black tea leaves possess at least the same antioxidant potency as catechins available in green tea (2).

Fig 1: Tea leaves

In this study, it is introduced as to a conclusion such as that both green and black tea leaves possess a marked antibacterial activity, *in- vitro* and further, green tea is found to be more effective than black tea, most plausibly because of the higher catechincontents of green tea (5). It seems apparently that green tea has more health benefits than an equal volume of black tea when deemed in terms of antioxidant capacity and such a statement can be explained by the fact that each tea is different in terms of composition as well as concentration of antioxidant compounds(6). Tea is one of the most-widely consumed beverages in the world with a number of different beneficial health effects, mainly ascribed to the polyphenolic content of the tea catechins (7). No doubt, rutin has performed many pharmacological activities, including antioxidant, cytoprotective, vasoprotective, anticarcinogenic, neuroprotective and cardioprotective activities.

Through cellular, animal, and also through human experiments, it has already been shown that the green tea leaf and its major component such as epigallocatechin-3-gallate (EGCG) have anti-inflammatory effects (6). Antibacterial activity is generally governed by total phenolics, flavanoids and antioxidant ability of an extract.

The bioactivity of green tea, its benefits is well established and documented. Antibacterial activity of green tea has also been investigated in details. Green tea has been known to prevent dental caries for decades. The active antibacterial component in green tea has been nailed to be EGCG. EGCG has received much attention for its effects on the inhibition of HIV infection and multidrug-resistant *Staphylococcus aureus* infections. The most favorable effects of green tea are accredited to the green tea polyphenols, predominantly the catechins, which make up, 25–35% of the dry weight of green tea leaves[7,8,9]. The tea catechins belong to the family of flavanoids and possess two benzene rings referred to as the A- and B-rings and notorious for their bactericidal effect. The bactericidal action of catechin is due to its hydrogen peroxide generation. The highest antimicrobial activity of tea is due to presence of catechins polyphenols which damage the bacterial cell membrane[10]. The highest antimicrobial activity of tea is due to presence of catechins polyphenols which damage the bacterial cell membrane. In terms of antimicrobial acitivity, EGC and EGCG have been shown to exhibit highest antimicrobial effect. EGCG is the most established in terms of bactericidal activity[11]. In the present study one of the promising bioactivities of green and black tea leaves, namely, antimicrobial activity was tested against Escherichia coli, Staphylococcus aureus, Salmonella typhi and Klebsiella pneumoniae in leaves extract.

MATERIALS AND METHODS

Green Tea Leaves

The fresh tea leaves were collected from the hills of Nelyampathy tea estate located in Palakkad, Kerala. The leaf buds used for green tea sampling were properly cut from the tea plants, washed and dried well in shadow. Once the leaves were dry, they were grinded in mortar and pestle to a fine powder.

Fig: 2. Green tea Leaves

The solvents used were Aqueous, Ethanol and N – Hexane. Dissolved 2g of powdered sample in 20ml of each solvents, shaken overnight. The next day filtered the final supernatant (the sample or the extract) which was used for further phytochemical studies.

Black Tea Leaves

The solvents used were Aqueous, Ethanol and N – Hexane. Dissolved 2g of powdered sample in 20ml of each solvents, shaken overnight. The next day filtered the final supernatant (the sample or the extract) which was used for further phytochemical studies.

Fig: 3. Black Tea Leaves

Fig: 4. Extract of Green Tea Leaves:

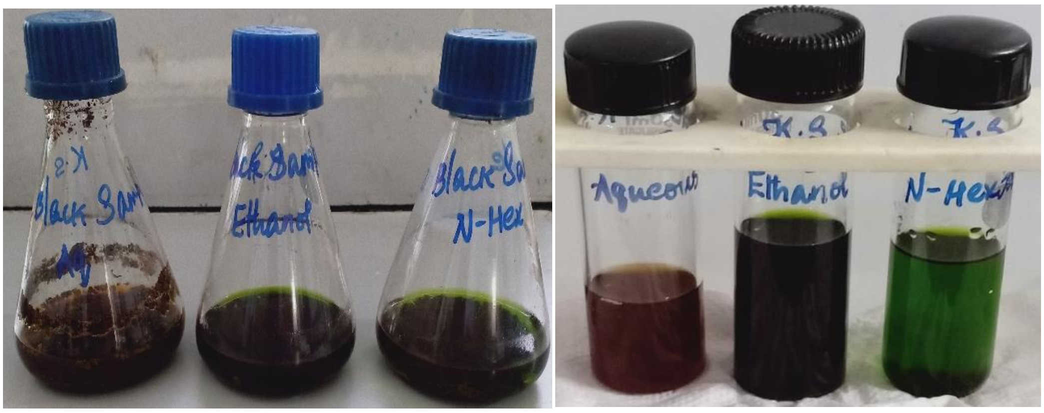


Fig: 5. Extract of Black Tea Leaves:

Fine grinded tea leaves were soaked in solvents and the extracts were obtained after overnight soaking.

ANTI-MICROBIAL STUDIES

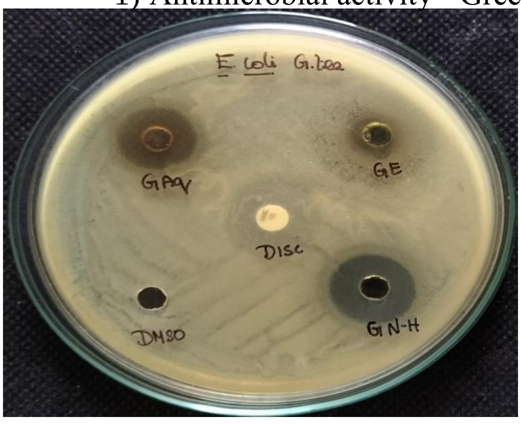
Antimicrobial activity for all the extracts in the following organisms;

Escherichia coli, Staphylococcus aureus, Klebsiella pneumonia, Salmonella typhi

The agar well diffusion method was used to study the antibacterial activity of the sample. Nutrient broth medium was used to subculture the bacteria and was incubated at 37 °C for 24 h, afterwards, 70µl cultures of E.Coli, S.Aureus, K.Pneumoniae, and S.Typhi were taken and spread on the Mueller hinton agar plates (39g of Mueller hinton agar was dissolved in 1000 ml of distilled water and sterilized under autoclave at 1210C for 15 minutes) to cultivate a uniform microbial growth plates. Followed by wells were made with cork borer and the samples were added to the respective wells along with negative (DMSO) and positive control (Antibiotic disc-AMC-Amoxyclav 10mcg) and Finally, the petri dishes were incubated for 24 h at 37 °C. In order to evaluate the antibacterial activity of the samples, the diameter of the inhibition zone was measured and noted.

RESULTS AND DISCUSSION

1) Antimicrobial activity - Green tea leaves extract



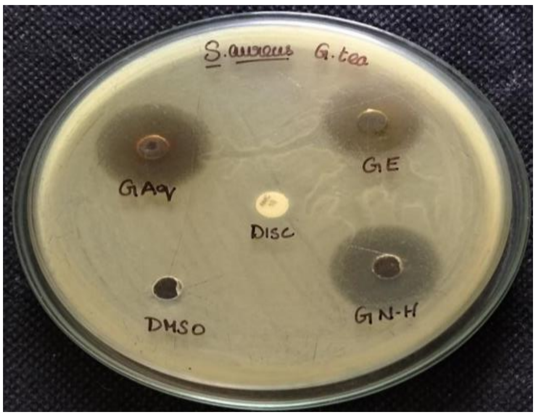
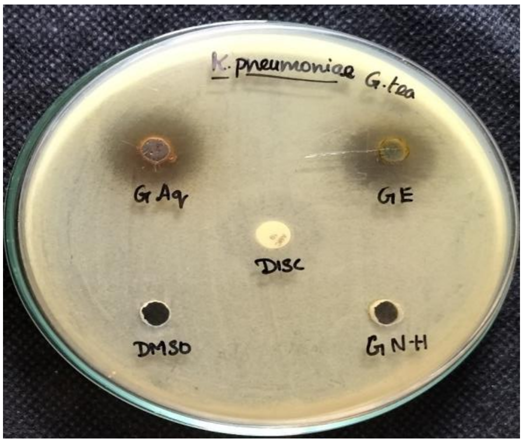


Fig: 6. ***Escherichia coli*** **Fig: 7.** ***Staphylococcus aureus***



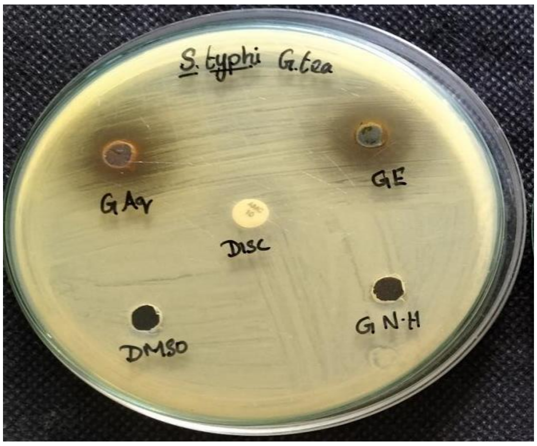


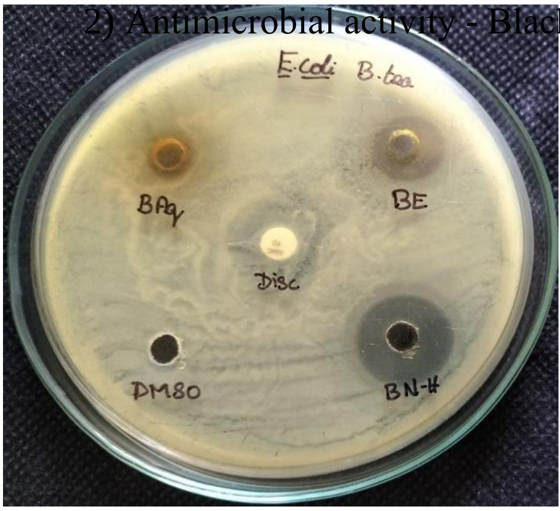
Fig: 8. *Klebsiella pneumoniae* **Fig: 9. *Salmonella typhi***

Name of the orgainsms	Zone of inhibition in mm				
	G Aq	G E	G NH	DISC	DMSO
E.coli	15	10	16	Nil	Nil
S.aureus	21	18	20	Nil	Nil
K.pneumoniae	18	15	Nil	Nil	Nil
S.typhi	11	15	Nil	Nil	Nil

Table 1. Antimicrobial activity - Green tea leaves extract zone of inhibition

In this study, the effect of green tea Aqueous, Ethanol and N – Hexane leaves extracts were tested by measuring the non-growth zone of Gram -ve bacteria (*E. coli ATCC25922* and *staphylococcus*) and Gram + ve bacteria (*S.klebsiella pneumonia* and salmonella). The data presented that the Aqueous , N – Hexane extract gave the highest zone of inhibition against *staphylococcus*. This is consistent with the results obtained by Dailey and Vuong, 2015 and the study of (Ikigai et al., 1993), that green tea extracts has more activity against + ve Grams than Gram -ve bacteria membranes. Data obtained were compatible too with Ababutain, 2015, Arokiyaraj et al., 2009 as well as the solvent type is an important factor which affects the antimicrobial effect of the plant extract (Table .1)

2) Antimicrobial activity - Black tea leaves extract



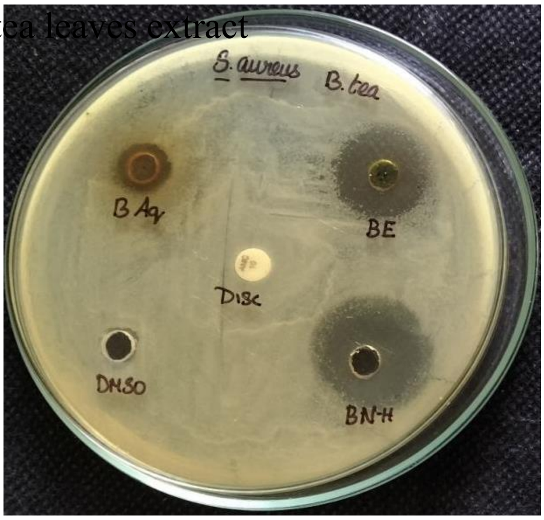


Fig: 10. *Escherichia coli* **Fig: 11. *Staphylococcus aureus***

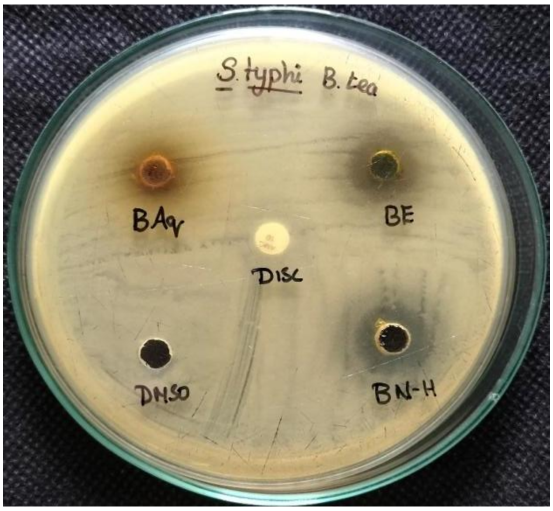


Fig:12. ***Salmonella typhi*** **Fig:13.** ***Klebsiella pneumoniae***

Name of the orgainsms	Zone of inhibition in mm				
	B Aq	B E	B NH	DISC	DMSO
E.coli	Nil	12	16	Nil	Nil
Saureus	10	17	20	Nil	Nil
K.pneumoniae	13	18	12	Nil	Nil
S.typhi	10	13	14	Nil	Nil

Table 2. Antimicrobial activity - Green tea leaves extract zone of inhibition

In this study, the effect of black tea Aqueous, Ethanol and N – Hexane leaves extracts were tested by measuring the non-growth zone of Gram -ve bacteria (*E. coli ATCC25922* and *staphylococcus*) and Gram + ve bacteria (*S.klebsiella pneumonia* and salmonella). The data presented that the N – Hexane extract gave the highest zone of inhibition against *staphylococcus* as well as the solvent type is an important factor which affects the antimicrobial effect of the plant extract (Table .2).

CONCLUSION

In the current study, we have compared the antimicrobial activities of green tea and black tea leaves extract. The results indicated that the particles played size dependant roles in moving the antibacterial activity of the extract with high percentage zone of inhibition by the green tea leaves extract compared with black tea leaves extract. It has been proved that the polyphenols and flavanoids available both in green tea leaves as well as in black tea leaves are safe and non-toxic so that these can be used in medicine rather.

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