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RESEARCH **P**APER

Antibacterial effect of green tea on oral bacterial species

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In the present study the samples were collected from mouth for isolation of bacterial species using spreading and streaking techniques. The isolated bacterial species were identified by performing various biochemical tests. Different extracts of green tea were prepared in different solvents like ethanol, methanol, propanol of different concentrations to check the antibacterial activity of green tea on bacteria isolated from mouth. The maximum zone of inhibition was observed in green tea (Tetley) extract in methanol 90 per cent (3.5cm) and minimum was observed in propanol 30 per cent (1.4 cm) in Tetley green tea. Thus, from present study it was found that the green tea extracts exhibited remarkable antibacterial properties. The green tea extract helped in destroying the pathogenic bacteria present in mouth, thus preventing diseases.

Key words : Green tea, Inhibition zone, Antibacterial activity, Biofilm

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INTRODUCTION

The dark, wet and warm environment of the mouth, with the occasional meal running through it, makes it an excellent niche for microbes to live. From birth to around age 12, when the permanent dentition is complete, the local oral conditions are continuously changing as teeth are shed and new ones erupt (Skinner and Carr, 1974). The bacteria within the mouth not only have to communicate with all the different species living within the biofilm, but they must obtain a strong adherence to a surface in order to not be washed away by saliva (Gibbons and Houte, 1975 and Kuramitsu , 2007). Salivary flora does not necessarily represent the microbial composition of the different components of the mouth, it does impact which microbes can live within the oral cavity and has recently been the target of research in early disease detection (Wilson, 2008). Green tea extract containing polyphenols and caffeine has been shown to induce thermogenes is and stimulate fat oxidation, boosting the metabolic rate 4 per cent without increasing the heart rate (Dulloo *et al.*, 1999; Cabrera *et al.*, 2006 and Heiss and Heiss, 2007). The content of flavonoids may vary dramatically amongst different tea products (Lambert *et al.*, 2007). The investigation was performed for the identification of bacterial species in human saliva by Gram's staining and biochemical tests and to check the antibacterial effect of green tea on oral bacterial species.

RESEARCH METHODOLOGY

Sample used :

Different types of green tea (Tetely green tea, Lipton green tea, Tata green tea) and saliva were used for the present investigation.

Solvent used :

Ethanol, methanol and propanol were used as solvent at different concentrations 30 per cent, 60 per cent and 90 per cent, respectively.

Sample collection :

Sample of saliva was collected from different people and different varieties of green tea were purchased from local market nearby Cytogene Research and Development, Lucknow, Uttar Pradesh.

Isolation of single bacterial colonies :

The principle of this technique is to streak a suspension of bacteria until single cell are separated on the plate. Each individual cell then grows in isolation to produce a clone of identical cells known as a "colony". The majority of these cells are genetically identical. However, during growth, mutation of a single colony can give rise to low level of mutant's cells.

Identification of unknown bacteria species :

The bacterial species was identified by Gram's staining and by various biochemical tests.

Preparation of different extracts :

Various extracts for testing the antibacterial activity of bacterias isolated from mouth were prepared using solutions of ethanol, propanol, methanol and normal tea. Various solutions were prepared at different percentages of 30 per cent, 60 per cent and 90 per cent. Green tea was weighed and dissolved in these solutions. After preparing these solutions they were kept in bottles and bottles was wrapped, sealed and kept at drank places. After keeping them overnight their extracts was used for checking antibacterial activity.

Determination of antibacterial activity :

The antibacterial was determined by well- diffusion method.

RESEARCH FINDINGS AND ANALYSIS

The results obtained from the present investigation as well as relevant discussion have been summarized under following heads :

Identification of isolated bacterial species :

Bacterial species identified from the Bergey's manual was gram positive rod shaped and gram negative coccus shaped which included *Neisseria* spp., *Micrococcus* spp., *Bacillus* spp., *Staphylococcus* spp. and *Klebsiella* spp. Among these bacterial species *Klebsilla* spp., *Bacillus* spp., *Micrococcus* spp. and *Staphylococcus* spp., showed negative test for casein hydrolysis while *Neisseria* spp. showed positive test while bacterial species BS2 showed positive test for urease, Simmon citrate and catalase test and thus, the bacterial genus identified from Bergey's manual was *Bacillus* spp. and *Neisseria* spp. (Table 1).

Antibacterial activity of Lipton green tea on bacteria BS1, *Neisseria* spp. :

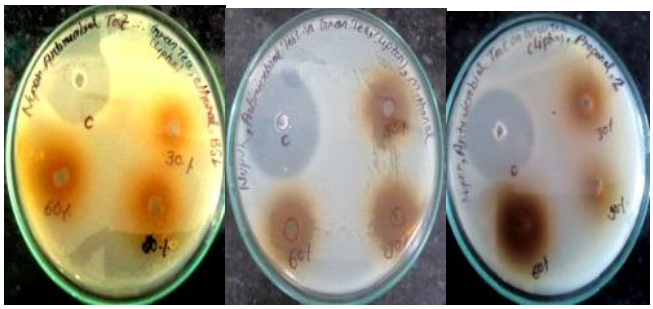

Fig. 1: Antibacterial activity of green tea in ethanol, methanol and propanol extract on BS1,*Neisseria* spp.

Antibacterial activity of Lipton green tea on BS2, *Bacillus* spp. :

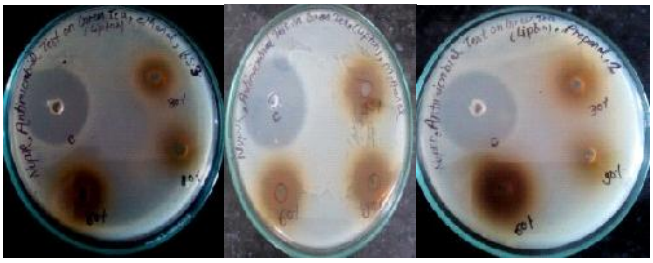

Fig. 2: Antibacterial activity of green tea in ethanol, methanol and propanol extract on BS2, *Bacillus* spp.

Table 1: Biochemical test for identification of isolated bacterial species

1.	MR/VP test	Positive/negative	Positive/negative
2.	Casein hydrolysis test	Negative	Negative
3.	Simmon citrate agar test	Positive	Positive
4.	Indole test	Negative	Negative
5.	Catalase test	Positive	Positive
6.	Sugar fermentation test	Negative	Negative

Fig. 1, 2 and 3 shows the maximum inhibition zone in 60 per cent (3 cm) ethanol on BS1 and BS2 and 90 per cent (2 cm) ethanol on BS1 and minimum zone of inhibition in 90 per cent ethanol on BS2.

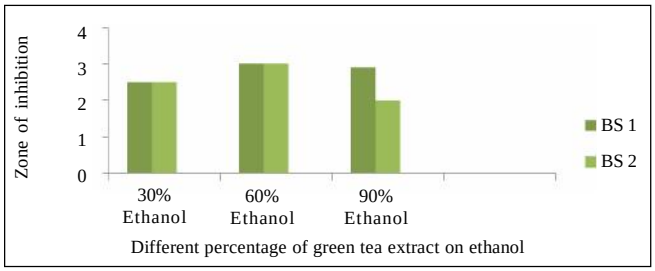

Fig. 3: Antibacterial effect of green tea (Lipton) extract in ethanol on BS1 and BS2

Fig. 4 shows the maximum zone of inhibition in 30 per cent ethanol on BS1 and minimum zone of inhibition shows in 90 per cent methanol on BS2 and Fig. 5 shows the maximum zone of inhibition in 60 per cent propanol on BS1 and minmum zone of inhibition shows in 90 per cent propanol on BS2.

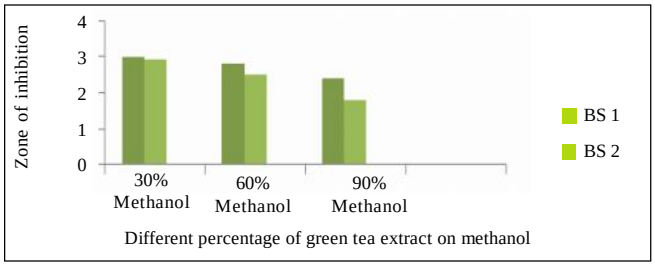

Fig. 4: Antibacterial effect of green tea (Lipton) extract in methanol on BS1 and BS2

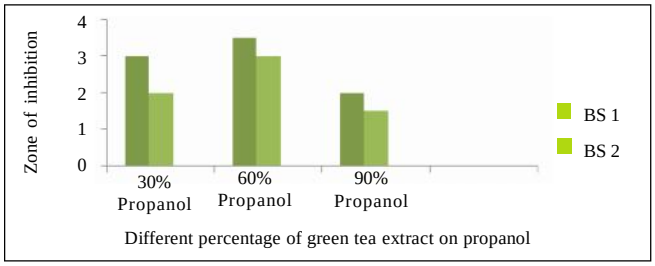

Fig. 5: Antibacterial effect of green tea (Lipton) extract in propanol on BS1 and BS2

Antibacterial activity of green tea (Tetley) on bacteria BS1, *Neisseria* spp.

Antibacterial activity of green tea (Tetley) in on bacteria BS2, *Bacillus* spp.

Fig. 6 shows the maximum inhibition zone on 30 per cent ethanol and minimum zone of inhibition on 90 per cent propanol in BS1 whereas Fig. 7 shows the maximum inhibition zone on 30 per cent ethanol and minimum zone of inhibition on 30 per cent propanol in BS2.

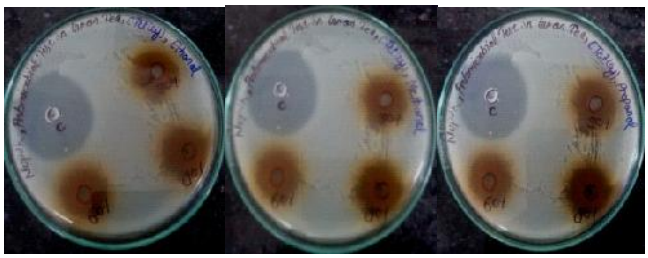

Fig. 6: Antibacterial activity of green tea in ethanol, methanol and propanol extract on BS1, *Neisseria* spp.

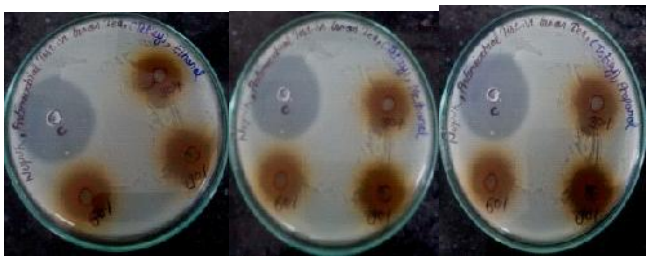

Fig. 7: Antibacterial activity of green tea in ethanol, methanol and propanol extract on BS2, *Bacillus* spp.

Fig. 8 shows the maximum zone of inhibition in 30 per cent ethanol on BS1 and BS2 (3 cm) minimum zone of inhibition shows in 90 per cent ethanol on BS2 (1.5 cm) and Fig. 9 shows the maximum zone of inhibition in

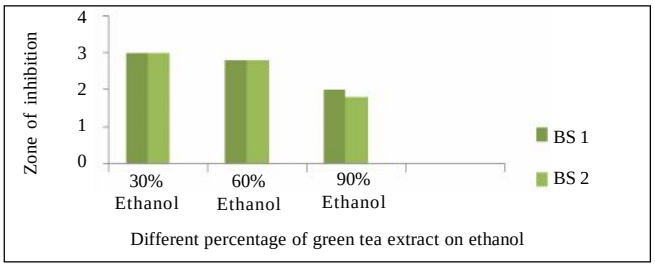

Fig. 8: Antibacterial effect of green tea (Tetley) extract in ethanol on BS1 and BS2

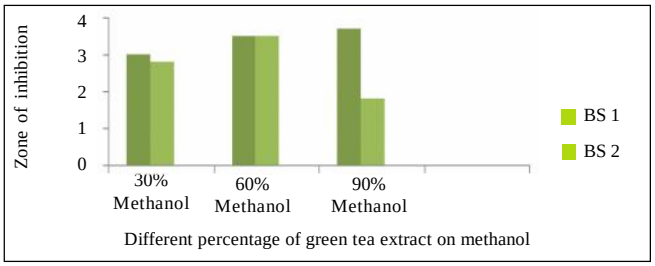

Fig. 9: Antibacterial effect of green tea (Tetley) extract in methanol on BS1 and BS2

90 per cent methanol on BS1 (3.5 cm) and minmum zone of inhibition shows in 90 methanol on BS2 (1.5cm).

Fig. 10 shows the maximum zone of inhibition in 90 per cent propanol on BS1 (3.1 cm) minimum zone of inhibition shows in 30 per cent propanol on BS2 (1.4 cm).

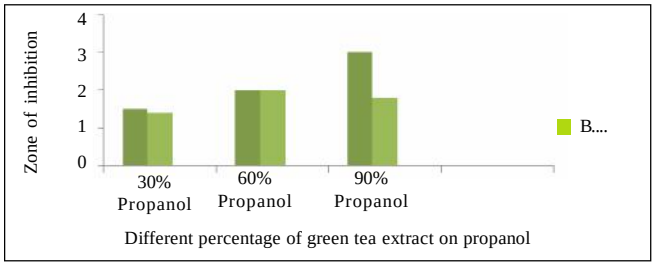

Fig. 10: Antibacterial effect of green tea (Tetley) extract in propanol on BS1 and BS2

Fig. 11: Antibacterial activity of normal tea in ethanol, methanol and propanol extract on BS1,*Neissiera* spp.

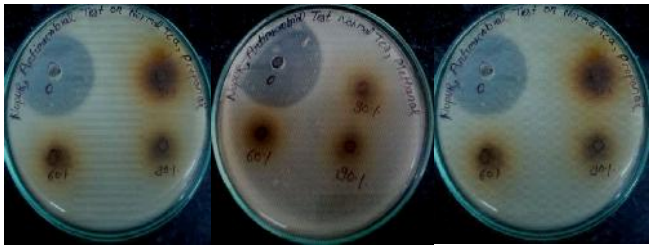

Fig. 12: Antibacterial activity of normal tea in ethanol, methanol and propanol extract on BS2, *Bacillus* spp.

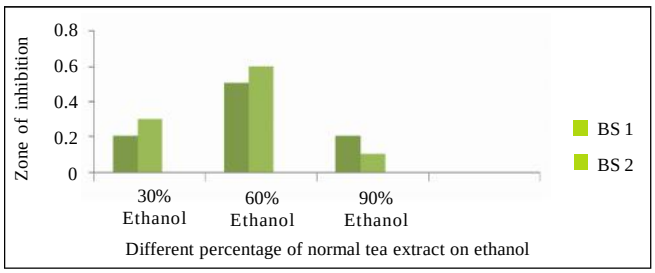

Fig. 13: Antibacterial effect of normal tea extract in ethanol on BS1 and BS2

Antibacterial activity of normal tea on bacteria BS1, *Neisseria* spp.

Fig. 14 shows the maximum zone of inhibition in 90 per cent methanol on BS1(0.4cm) and minimum zone of inhibition shows in 30 per cent methanol on BS1 and BS2 (0.1cm) and Fig. 15 shows the maximum zone of inhibition in 30 per cent propanol on BS1 and minmum zone of inhibition shows in 90 per cent propanol on BS2 (0.2 cm). Green tea may lower blood low-density lipoprotein and total cholesterol levels, though the studies were of short duration and it is not known if these effects result in fewer deaths and evidence does not support green tea's reducing coronary artery disease risk. Several randomized controlled trials suggest green tea can reduce body fat by a small amount for a short time, though it is not certain if the reduction would be meaningful for most people (Magalhaes *et al.*, 2009). It was found that certain catechins found in green tea taken at levels many hundreds of times greater than what could be obtained from even very high tea consumption may actually damage DNA (Ferrazzano, 2011). Hirasawa *et al.*(2006) suggested that green tea consumption may be a possible preventative and treatment for Parkinson's disease. Green tea contains polyphenols that can protect dopaminergic neurons from malfunctioning. Green tea may interfere with the anti-cancer drug Bortezomib (Velcade) and other boronic acid-based proteasome inhibitors (Xiao *et al.*, 2000; Sinija and Mishra, 2008).

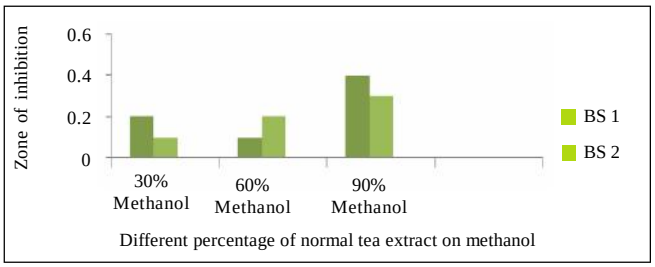

Fig. 14: Antibacterial effect of normal tea extract in methanol on BS1 and BS2

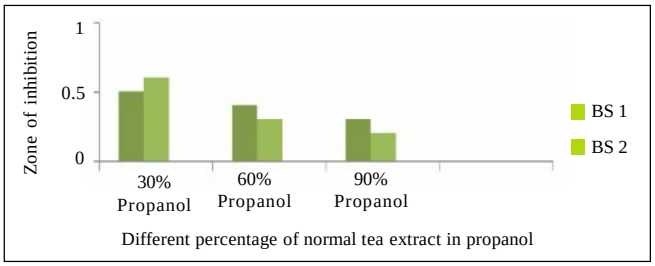

Fig. 15: Antibacterial effect of normal tea extract in propanol on BS1 and BS2

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