



Demonstration of Antimicrobial Activity of Commercial Oolong Tea and Green Tea Against Pathogenic Bacteria

Md. Sakil Munna^{1,*}, Samia Quaiyum², Helena Forunato²

¹Department of Microbiology, Stamford University Bangladesh, Dhaka, Bangladesh

²Department of Natural History Science, Hokkaido University, Sapporo, Japan

Email address:

sakilmunna@gmail.com (Md. S. Munna)

*Corresponding author

To cite this article:

Md. Sakil Munna, Samia Quaiyum, Helena Forunato. Demonstration of Antimicrobial Activity of Commercial Oolong Tea and Green Tea Against Pathogenic Bacteria. *American Journal of Plant Biology*. Vol. 3, No. 1, 2018, pp. 8-11. doi: 10.11648/j.ajpb.20180301.12

Received: December 17, 2017; **Accepted:** January 29, 2018; **Published:** March 2, 2018

Abstract: While an array of plants has long been used for the preparation of medicines, tea plants can be used for disease medication since they have been reported to possess anti-bacterial attributes. Present study emphasized on the assessment of antimicrobial activity of tea against *Bacillus* spp. (SkB01), *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01), *Salmonella* spp. (SkS01), *Vibrio* spp. (SkV01). Oolong tea and green tea were used to determine the antimicrobial activity employing minimal inhibitory concentration (MIC) methods. The results clearly illustrated interesting antimicrobial potentials of both experimented teas against the tested microorganisms. Oolong tea exhibited the anti-bacterial activity against *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01) and *Salmonella* spp. (SkS01). As well, green tea exhibited the anti-bacterial activity against *Bacillus* spp. (SkB01) and *Vibrio* spp. (SkV01). The in vitro anti-bacterial activity of oolong tea and green tea against the bacterial pathogens revealed most of the tea plants to be effective against the growth and survival of the pathogenic bacteria.

Keywords: Oolong Tea, Green Tea, Antimicrobial Activity, Pathogenic Bacteria, Natural Medicines

1. Introduction

For centuries tea is a very popular drink world-wide and has been consumed as a beverage and valued for its medicinal properties [1]. It is manufactured from the plant *Camellia sinensis*, are cultivated in more than 30 countries, under subtropical and tropical conditions [1-2]. There are mainly four arrays of tea produced world-wide, i.e., oolong, white, green, and black [2-5]. Purified catechin fractions from white tea and black tea can inhibit the growth of many bacterial species and possess anticarcinogenic properties [2, 4]. Besides, oolong tea from partially fermented leaves are most popular in China, Taiwan and Japan; and green tea is most popular in China, Japan, and Korea [2]. Green tea and oolong tea passing through various manufacturing processes [2-5, 7]. Green tea is produced by steaming or panning (on a country basis) with no fermentation to prevent catechin oxidation by polyphenol oxidase and retain their green color [2-5, 7]. Oolong tea is semi-fermented and has a unique feature of freshness of green tea and the fragrance of black tea [2-5,7]. Oolong tea

has been used for various types of activities, i.e., sharpen thinking skills and improvement of mental alertness, cancer prevention, tooth decay, osteoporosis, and heart disease [6-10]. Besides, green tea has been shown antioxidant potentiality, anticancer property, antiatherogenic function, and also hepatoprotective ability [7, 11-17]. Green tea is also known for its antimicrobial activity against many microorganisms. Previously several studies revealed that moderate daily consumption of green tea killed *Staphylococcus aureus*, *Vibrio parahemolyticus*, *Clostridium perfringens*, *Bacillus cereus*, *Pseudomonas shigelloides* and etc [9, 18-32]. These earlier studies led us to broaden the research interest in tea to assess the antimicrobial activity of oolong tea and green tea against our laboratory stock culture of *Bacillus* spp. (SkB01), *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01), *Salmonella* spp. (SkS01), *Vibrio* spp. (SkV01).

2. Materials and Methods

The On the basis of strain availability, laboratory stock

cultures of *Bacillus* spp. (SkB01), *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01), *Salmonella* spp. (SkS01), *Vibrio* spp. (SkV01) were used. Mueller Hinton agar (MHA) (Sigma-Aldrich Corporation, USA), Mueller Hinton broth (Sigma-Aldrich Corporation, USA) were used [33]. Pre-cultures were prepared by inoculating 5 ml Mueller Hinton broth by a loopful of a colony from the freshly prepared bacterial culture plates, followed by incubation at 37 °C in static condition up to 24 hours [33]. 25 grams of oolong and green tea were separately mixed up with 225 ml of water and prepared with 93 to 96 °C (200 to 205 °F) water (not boiling) and steeped 3–10 minutes and leave it for 30 minutes to lower the temperature around 35 °C. To observe the antibacterial effect on cell growth employing minimal inhibitory concentration (MIC) methods [33-34], different concentrations of tea (oolong and green) including 0 µl (control), 100 µl and 1000 µl were used. The optical density at 600 nm (OD₆₀₀) was monitored after 24 hours of incubation periods. All experiments were conducted in triplicates. Statistical analysis regarding bacterial growth was performed by determining the P

value (~0.3) through t test.

3. Results

In our study, when *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01) and *Salmonella* spp. (SkS01) cells were grown separately with 100 µl and 1000 µl of concentration of oolong tea, a significant decrease in the cell turbidity were observed compared to control (0 µl) (shown in Table 1). Nonetheless, *Bacillus* spp. (SkB01) and *Vibrio* spp. (SkV01) and *Pseudomonas* spp. (SkP01) exhibit a significant decrease in the cell turbidity, when grown separately with 100 µl and 1000 µl of concentration of green tea compared to control (0 µl) as shown in table 2. Curiously, the data clearly illustrated that commercial oolong Tea has the anti-bacterial activity against *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01) and *Salmonella* spp. (SkS01). As well, commercial green tea has the anti-bacterial activity against *Bacillus* spp. (SkB01) and *Vibrio* spp. (SkV01) (Table 1 and Table 2).

Table 1. Minimum Inhibitory Concentration (MIC) of Oolong Tea.

Concentration of Oolong Tea	Optical Density at 600 nm					
	SkB01	SkE01	SkK01	SkP01	SkS01	SkV01
0 µl	1.851	1.911	1.574	2.110	1.074	1.130
100 µl	1.805	0.715	0.597	0.637	0.853	1.117
1000 µl	1.783	0.360	0.271	0.379	0.778	1.051

Table 2. Minimum Inhibitory Concentration (MIC) of Green Tea.

Concentration of Green Tea	Optical Density at 600 nm					
	SkB01	SkE01	SkK01	SkP01	SkS01	SkV01
0 µl	1.851	1.911	1.574	2.110	1.074	1.130
100 µl	0.797	1.403	1.253	0.581	1.417	0.641
1000 µl	0.591	1.203	0.853	0.315	1.463	0.273

4. Discussion

Our earlier studies revealed the growth and proliferation of pathogenic bacteria and anti-bacterial traits in the commonly available salad vegetables and herbal medicines in Bangladesh. [35,36-40]. In all instances, the reports showed the anti-bacterial features of natural products suggesting them appropriate substitute of synthetic medicines [35, 36, 38-40]. The prospect of tea is a very popular drink all over the world [2, 4, 5,29]. Tea has been shown to have a wide range of beneficial effects including catabolism of catecholamines, strengthening capillaries, exerting an anti-inflammatory effect, acting as an antioxidant, inhibiting angiotensin-converting enzyme and inhibiting the growth of implanted malignant cells [2,4,5]. To our knowledge, demonstration of antimicrobial activity of commercial oolong and green tea against pathogenic bacteria is not that frequent, which in turn, may pose a persuasive natural medicine [6-8, 11-14, 16, 17, 21, 37-40]. Our current study clearly illustrated antimicrobial potentials of both experimented teas against *E. coli* (SkE01), *Klebsiella* spp. (SkK01), *Pseudomonas* spp. (SkP01), *Salmonella* spp. (SkS01), *Bacillus* spp. (SkB01) and *Vibrio*

spp. (SkV01). There had been studies of the antibacterial effects of tea against clinical isolates of methicillin-resistant *Staphylococcus aureus* (MRSA), *Helicobacter pylori*, *Staphylococcus aureus*, *Vibrio parahaemolyticus*, *Clostridium perfringens*, *Bacillus cereus* and *Pleisomonas shigelloides* were evaluated [6-32], which is in agreement with our current study.

5. Conclusion

The present investigation can be concluded that commercial oolong tea and green tea were found to be effective in inhibiting the pathogenic microorganisms through antibacterial studies, which opening a promising path of clinical applications in the preparation of specific and natural antibacterial remedies. But further research is required for isolation and identification of the biologically active compounds present in commercial oolong tea and green tea.

Acknowledgements

Authors are thankful to the Department of Natural History Science, Hokkaido University, Japan for logistic supports.

Conflict of Interest

The authors declare that they have no conflict of interest.

References

- [1] R. Soukand, C. L. Quave, A. Pieroni, M. Pardo-de-Santayana, J. Tardío, R. Kalle, L. Luczaj, I. Svanberg, V. Kolosova, L. Aceituno-Mata, G. Menendez-Baceta, I. Kołodziejska-Degórska, E. R. Pirożnikow, Petkevičius, A. Hajdari, B. Mustafa “Plants used for making recreational tea in Europe: a review based on specific research sites” *J Ethnobiol Ethnomed*, vol. 9, pp.58. 2013.
- [2] J. M. Hamilton-Miller “Antimicrobial properties of tea (*Camellia sinensis* L.)” *Antimicrob Agents Chemother*, vol. 39, pp. 2375–2377. 1995.
- [3] M. Toda, S. Okubo, R. Ohnishi, T. Shimamura “Antibacterial and bactericidal activities of Japanese green tea” *Jap J Bacteriol*, vol. 44, pp. 669–672. 1989.
- [4] E. Ryu, D. C. Blenden, D. Wendall “The inhibition of growth of selected bacteria by incorporating powdered tea in the medium” *Int J Zoonoses*, vol. 9, pp. 73-77.1982.
- [5] C. V. Stagg, D. J. Millin “The nutritional and therapeutic value of tea: a review” *J Sci Food Agric*, vol. 26, pp. 1439-59.1975.
- [6] M. Weerawatanakorn, W. Hung, M. Pan, S. Li, D. Li, X. Wan, C. Ho “Chemistry and health beneficial effects of oolong tea and theasinensins” *Food Sci Human Wellness*, vol. 4, pp. 133–146. 2015.
- [7] E. W. C. Chan, E. Y. Soh, P. P. Tie, Y. P. Law “Antioxidant and antibacterial properties of green, black, and herbal teas of *Camellia sinensis*” *Pharmacognosy Res*, vol. 3, pp. 266–272. 2011.
- [8] A. Nkubana, Q. He “A comparative study of antioxidant activity between black tea from Rwandan highlands with green and oolong teas from China” *Int J Food Safety Nutr Public Health*, vol. 1, pp.159–166. 2008.
- [9] X. Su, J. Duan, Y. Jiang, J. Shi, Y. Kakuda “Effects of soaking conditions on the antioxidant potentials of oolong tea” *J Food Compos Anal*, vol. 19, pp. 348–353. 2006.
- [10] H. Sasaki, M. Matsumoto, T. Tanaka, M. Maeda, M. Nakai, S. Hamada, T. Ooshima “Antibacterial activity of polyphenol components in oolong tea extract against *Streptococcus mutans*” *Caries Res*, vol. 38, pp. 2-8. 2004.
- [11] W. C. Reygaert “The antimicrobial possibilities of green tea” *Frontiers Microbiol*, vol. 5, pp. 434. 2014.
- [12] P. W. Taylor, J. M. T. Hamilton-Miller, P. D. Stapleton “Antimicrobial properties of green tea catechins” *Food Sci Technol Bulletin*, vol. 2, pp. 71–81. 2005.
- [13] H. H. S. Chow, I. A. Hakim, D. R. Vining, J. A. Crowell, J. Ranger-Moore, W. M. Chew, C. A. Celaya, S. R. Rodney, Y. Hara, D. S. Alberts “Effects of dosing condition on the oral bioavailability of green tea catechins after single-dose administration of Polyphenon E in healthy individuals” *Clinical Cancer Res*, vol. 11, pp. 4627–4633. 2005.
- [14] D. Chen, K. G. Daniel, D. J. Kuhn, A. Kazi, M. Bhuiyan, L. Li, Z. Wang, S. B. Wan, W. H. Lam, T. H. Chan, Q. P. Dou “Green tea and tea polyphenols in cancer prevention” *Front Biosci*, vol. 9, pp. 2618–2631. 2004.
- [15] M. Hirasawa, K. Takada, M. Makimura, S. Otake “Improvement of periodontal status by green tea catechin using a local delivery system: a clinical pilot study” *J Periodontal Res*. vol. 37, pp. 433–438. 2002.
- [16] Isogai E, Isogai H, Hirose K, Hayashi S, Oguma K (2001) In vivo synergy between green tea extract and levofloxacin against enterohemorrhagic *Escherichia coli* 0157 infection. *Curr Microbiol*, 42: 248-51.
- [17] H. Graham “Green tea composition, consumption and polyphenol chemistry” *Preventive Med*, vol. 21, pp. 334–350. 1992.
- [18] T. I. Mbata, L. Debiao, A. Saikia “Antibacterial activity of the crude extract of chinese green tea (*Camellia sinensis*) On *Listeria monocytogenes*” *Int J Microbiol*, vol. 2. 2006.
- [19] A. Saikia, T. I. Mbata, L. Debiao “Antibacterial activity of the crude extract of Chinese green tea (*Camellia sinensis*) on *Listeria monocytogene*” *Int J Microbiol*, vol. 3. 2006.
- [20] L. Jian, L. P. Xie, A. H. Lee, C. W. Binns “Protective effect of green tea against prostate cancer: I. Kubo, H. Muroi, M. Himejima (1992) “Antimicrobial activity of green tea flavor components and their combination effects” *J Agric Food Chem*, vol. 40, pp. 245–248. 2004.
- [21] M. Hirasawa, K. Takada “Multiple effects of green tea catechin on the antifungal activity of antimycotics against *Candida albicans*” *J Antimicrob Chemother*, vol.53, pp. 225-229. 2004.
- [22] K. Matsunaga, T. W. Klein, H. Friedman, Y. Yamamoto “Epigallocatechin gallate, a potential immunomodulatory agent of tea components, diminishes cigarette smoke condensate-induced suppression of anti-Legionella pneumophila activity and cytokine responses of alveolar macrophages” *J Immunol*, vol. 9, pp. 864-71. 2002.
- [23] M. Zhang, C. W. Binns, A. H. Lee AH “Tea consumption and ovarian cancer risk: a case-control study in China” *Cancer Epidemiol Biomarkers Prev*, vol. 11, pp. 713–718. 2002.
- [24] E. Isogai, H. Isogai, K. Hirose, S. Hayashi, K. Oguma “In vivo synergy between green tea extract and levofloxacin against enterohemorrhagic *Escherichia coli* O157 infection” *Current Microbiol*, vol. 42, pp. 248–251. 2001.
- [25] S. Gupta, K. Hastak, N. Ahmad, J. S. Lewin, H. Mukhtar “Inhibition of prostate carcinogenesis in TRAMP mice by oral infusion of green tea polyphenols” *Proc Natl Acad Sci USA*, vol. 98. 2001
- [26] I. A. Hakim, R. B. Harris, U. M. Weisgerber UM “Tea intake and squamous cell carcinoma of the skin: influence of type of tea beverages” *Cancer Epidemiol Biomarkers Prev*, vol. 9, pp. 727–31. 2000.
- [27] K. Mabe, M. Yamada, T. Takahashi “In vitro and in vivo activities of tea catechins against *Helicobacter pylori*” *Am J Microbiol*, vol. 43, pp. 1788-1791. 1999.
- [28] K. Nakachi, K. Imai, K. Suga “Epidemiological evidence for prevention of cancer and cardiovascular disease by drinking green tea. In: T. Yoshikawa, ed. Food factors for cancer prevention” Tokyo: Springer- Verlag, pp.105-08. 1997.

- [29] M. Toda, S. Okubo, R. Hiyoshi, T. Shimamura “The bactericidal activity of tea and coffee” *Lett Appl Microbiol*, vol. 8, pp. 123-125. 1989.
- [30] Y. J. Ahn, T. Kawamura, M. Kim, T. Yamamoto, T. Mitsuoka “Tea polyphenols: selective growth inhibitors of *Clostridium* spp.” *Agric Biol Chem*, vol. 55, pp. 1425–1426. 1991.
- [31] K. S. Diker, M. Akan, G. Hascelik, M. Yurdakok “The bactericidal activity of tea against *Campylobacter jejuni* and *Campylobacter coli*” *Lett Appl Microbiol*, vol. 12, pp. 34-35. 1991.
- [32] Y. J. Ahn, S. Sakanaka, M. Kim, T. Kawamura, T. Fujisawa, T. Mitsuoka “Effects of green tea extracts on growth of intestinal bacteria” *Microb Ecol Health Dis*, vol. 3, pp. 335–338. 1990.
- [33] M. S. Munna, S. Humayun, R. Noor “Influence of heat shock and osmotic stresses on the growth and viability of *Saccharomyces cerevisiae* SUBSC01 Microbiology” *BMC Res Notes*, vol.8. 2016.
- [34] S. M. S. Alam, M. A Kalam, M. S. Munna, S. K. Munshi, R. Noor “Isolation of pathogenic microorganisms from burn patients admitted in Dhaka Medical College and Hospital and demonstration of their drug-resistance traits” *Asian Pac J Trop Dis*, vol. 4, pp. 402-407. 2014.
- [35] S. Quaiyum, N. I. Tanu, M. Sharmin, L. Paul, M. S. Munna, K. K. Das, M. Acharjee, R. Noor “Microbiological contamination and anti-bacterial traits of common oral herbal medicinal products within Dhaka Metropolis” *European J Med. Plants*, vol. 4. 2014.
- [36] T. Ahmed, N. J. Urmi, M. S. Munna, K. K. Das, M. Acharjee, M. M. Rahman, R. Noor “Assessment of microbiological proliferation and in vitro demonstration of the antimicrobial activity of the commonly available salad vegetables within Dhaka Metropolis, Bangladesh” *American J Agric For*, vol. 2, pp. 55-60. 2014.
- [37] P. Su, A. Henriksson, C. Nilsson, H. Mitchell “Synergistic effect of green tea extract and probiotics on the pathogenic bacteria, *Staphylococcus aureus* and *Streptococcus pyogenes*,” *J Microbiol and biotechnol*, vol. 24, pp. 1837-1842. 2008.
- [38] J. Gopal, M. Muthu, D. Paul, D.-H. Kim, S. Chun “Bactericidal activity of green tea extracts: the importance of catechin containing nano particles,” *Scientific Reports*, vol. 6. 2016.
- [39] W.-J. Zheng, X.-C. Wan, G.-H. Bao “Brick dark tea: a review of the manufacture, chemical constituents and bioconversion of the major chemical components during fermentation” *Phytochem. Rev*, vol. 14, pp. 499–523. 2015.
- [40] T. Atomssa, A. V. Gholap “Characterization of caffeine and determination of caffeine in tea leaves using UV-visible spectrometer” *African J of Appl Chem*, Vol. 7, pp. 22–31. 2015.