

The Antibacterial Activity Test Comparison of Green and Black Tea Ethanol Extract (*Camellia sinensis*) Against *Propionibacterium acnes*

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ABSTRACT

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A study was conducted in connection with the comparison of antibacterial activity tests between ethanol extracts from green tea leaves and black tea leaves (*Camellia sinensis*) against *Propionibacterium acnes*. There were obtained by a softening technique that was softened for 24 hours. The filtrate was then concentrated to give a concentrated extract and evaporated ethanol content. The concentrated extract was then diluted with distilled water to extract 5%, 10%, and 15% concentration variations. The test method for inhibitory activity was the disc method using distilled water as a negative blank. The results obtained with blank, 5%, 10% and 15% green tea were 0.525; 7.05 and 7.95 mm and black tea ethanol extract were 0; 2.50; 3.20 and 5.30 mm. The conclusion of this study is that the concentration of the extract is directly proportional to its inhibitory power, and the green tea ethanol extract is more potent than the black tea ethanol extract, so both can be used as antibacterial agents against *P. acnes*.

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1. Introduction

Acne is the most common inflammatory skin disease in which high percentage of teenagers temporarily suffer and lower percentage acne continues also to adulthood. Acne is specified by hyperactivity of sebaceous glands (Svensson et al., 2018; Sinha et al., 2014). *Propionibacterium acnes* (*P. acnes*) is a bacterial pathogen that is closely related in the pathophysiology of the common skin disease acne vulgaris (McLaughlin et al., 2019; Dessinioti & Katsambas, 2017; Omer et al., 2017). *P. acnes* are recently renamed *Cutibacterium acnes* along with other major pathophysiological factors, and inflammation has long been associated with the cause of acne. Recent advances have helped to understand the role of *P. acnes* in acne. There is no quantitative difference in the skin of *P. acnes* from patients with acne compared to controls, but *P. Acnes* phylogenetic groups have different genetic and phenotypic characteristics, *P. acnes* biofilms are more common in acne, and different phylogenetic types can cause different immune responses in acne. (Platsidaki & Dessinioti, 2018)

According to (Heckmann et al., 2018), research on *P. acnes* continues despite various skin preparation techniques. Studies on antimicrobial susceptibility and phylogenetic analysis of *P. acnes* in Japanese acne patients were conducted in 2013 and 2015 in (Nakase et al., 2017). The best known is how to utilize natural resources that contain plants as the active ingredient in herbs. Those natural activities help people overcome their health problems (Setijo, 2020). One of the most popular plants uses tea leaves. Different studies have shown that tea has many therapeutic benefits including as antimicrobial agent (Kalalinia et al., 2020). Tea leaves, a popular drink around the world, have many benefits, including skin problems that caused *P. acnes* (Saeed et al., 2017). Antibacterial agents contained in white tea have been studied against *P. acnes* (Robbani, 2019). The

antibacterial activity of *Camellia sinensis* against skin-related microbial pathogens was also studied, and green tea was superior to other teas (Sharma & Pundir, 2019). Green tea and other tea polyphenols were also investigated, especially for sebum production and scab vulgaris. (Saric et al., 2016). The effects of green tea extract seemed the significant results showed the reduction acne to woman during four weeks in their noses, perioral areas, and chins. (Lu & Hsu, 2016).

Tea plants thrive in the highlands of Sidamanik District, North Sumatera . The good inhibitory activity against *Escherichia coli* of green tea labeled Juma from Sidamanik District had been shown. (Fahmi et al., 2022). In this research, we compare both of green and black tea extract from Simanaik District which more effective agains *P. acnes*.

2. Method

Equipment used is autoclaves, stir bars, hot plates, incubators (Mettler), calipers, layered airflow cabinets, micropipettes, tweezers, eyelet needles, vortex, and other glassware (beakers, petri dishes, reaction tubes, Erlenmeyer flasks, etc.). The materials used are nutrient agar medium (Oxoid), nutrient broth (Oxoid), blanks, distilled water, *P. acnes* strain, green tea ethanol extract, black tea ethanol extract.

2.1. Preparation of green and black tea ethanol extract

The dried green and black tea were macerated with ethanol 96 % for 24 hours. After that, their filtrates were taken and then concentrated to be concentrated extracts and evaporated ethanol content until run out and obtained thick extracts. The thick extracts were diluted with distilled water to be extracts with 5 %, 10 %, and 15 % concentration variations.

2.2. Preparation of bacterial culture

Preparation of bacterial culture *P. acnes* were removed with a round needle, sterilized with glow, then transplanted onto the surface of nutrient medium, scratched and tilted, and incubated at 37 °C. for 24 hours. (Dirjen et al., 1995)

2.3 Bacterial inoculation production

Bacterial colonies were removed from the culture stock using sterile eyelets, then suspended in tubes containing 10 ml of nutrient medium and incubated at 37 °C for 24 hours until turbidity was comparable to standard Mc. Farland. (Dirjen et al., 1995)

2.4 Antibacterial activity test

Place a total of 0.1 ml of bacterial inoculum in a sterile Petri dish, then pour the nutrient agar medium into a 15 ml Petri dish, then shake the Petri dish on the table surface to homogenize the medium and bacterial suspension and can be integrated. Disc paper was given at the concentration of each test solution (5, 10 and 15 % concentration variations of black and green tea ethanol extract) and then placed on the surface of solid medium. Samples were incubated at 37 °C for 18-24 hours. The diameters of the suppression zone formed around the paper disc were observed and measured with a caliper. (Dirjen et al., 1995)

3. Result And Discussion

The inhibit zone diameter between green tea and black tea ethanol extract had been formed and listed in the figures below. Figure 1 shows the inhibition diameter of green tea ethanol extract with concentration variations are 5%, 10 % and 15 % with negative blank uses distilled water. This work is done two repetitions.

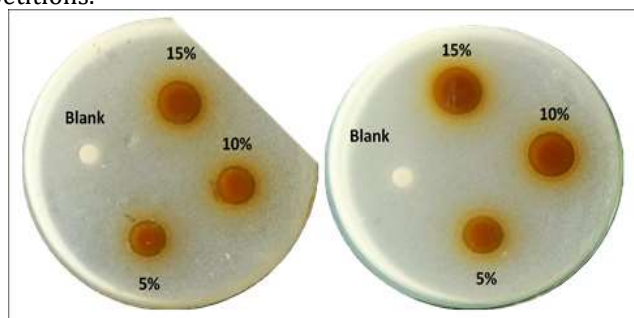


Figure1. GTEE against *P. acnes* (two repetitions)

Meanwhile, Figure 2 shows the inhibition diameter of black tea ethanol extract with concentration variations are 5%, 10 % and 15 % with negative blank uses distilled water. This work is also done two repetitions.

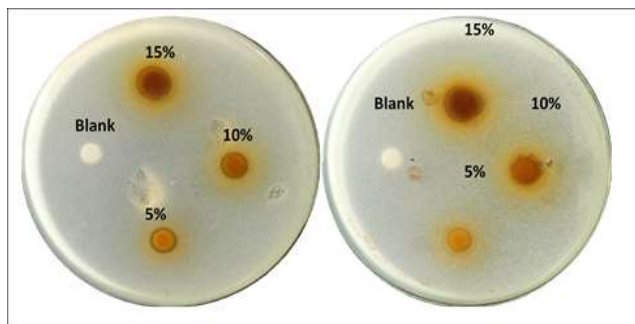


Figure 2. BTEE against *P. acnes* (two repetitions)

For inhibition activity detail, let us see the table below.

TABLE 1
COMPARISON OF INHIBITION DIAMETERS OF GREEN TEA AND BLACK TEA ETHANOL EXTRACT

C (%)	R	ID (mm)	
		BTEE against <i>P. acnes</i>	GTEE against <i>P. acnes</i>
Blank	P1	6	6
	P2	6	6
Avarage ± ID		0	0
15%	P1	11,1	13,7
	P2	11,5	14,2
Avarage ± ID		5,30	7,95
10%	P1	9,0	13,0
	P2	9,4	13,1
Avarage ± ID		3,2	7,05
5%	P1	8,6	10,6
	P2	8,4	11,9
Avarage ± ID		2,5	5,25

Note: C is sample concentration, R is repetitions, BTEE is Black Tea Ethanol Extract, GTEE is Green Tea Ethanol Extract, and ID is named as Inhibition Diameter (The calculation of ID is Total Diameter – Blank Diameter). For further detail, let us see the diagram below:

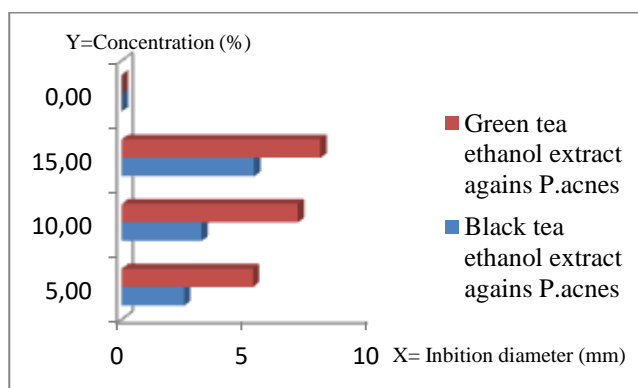


Figure 3. Antibacterial activity comparison diagram of black and green tea ethanol extract against *P. acnes*

In the antibacterial activity test by (David & Stout, 1971), antibacterial activity was classified as weak if the suppression zone was less than 5 mm, moderate if the suppression zone was 5 until 10 mm, and it is classified as strong. The inhibition zone is 11 until 20 mm, and if the inhibition

zone is 20 until 30 mm, it is classified as very strong. The formation of the inhibition zone was due to the antibacterial content contained in the ethanol extract of tea leaves, and at the same concentration, the antibacterial activity of the ethanol extract of green tea was higher than that of the ethanol extract of black tea. Green tea was higher than black tea due to the active chemical content of EGCG. This is due to the difference in the manufacturing process, in which the tea leaves are dried and wilted to make green tea without fermenting, whereas the tea leaves are fermented using the oxidation reaction of the phenolate enzyme contained in the tea leaves. According to (Xu & Chen, 2002).

The statement is referred to (Song & Seong, 2007), they studied the proportion of catechins between green tea and black tea is 30: 5%, and the proportion of oxidized phenol compounds between green tea and black tea is 0: 25%.

4. Conclusion

Inhibition tests of ethanol extracts from green tea leaves and black tea leaves (*Camellia Sinensis*) against *Propionibacterium acnes* were compared. The results obtained with blank, 5%, 10% and 15% green tea were 0. 5.25; 7.05 and 7.95 mm and black tea ethanol extract were 0.2.50; 3.20 and 5.30 mm. The conclusion of this study is that the concentration of the extract is directly proportional to its inhibitory power, and the ethanol extract from green tea is stronger than the ethanol extract from black tea, so both can be used as antibacterial agents against *P. acnes*.

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