Virgin Coconut Oil Inhibits Candida Albicans Growth In-vitro

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Abstract: *Candida albicans* is the commensal organism act as a normal flora of the human body that can turn into pathogens. Virgin Coconut Oil (VCO) is an oil obtained from coconut meat (*Cocos nucifera l.*). The purpose of this research is to obtain the inhibitory power of pure coconut oil (virgin coconut oil) to *Candida albicans* growth in-vitro. Preparation of VCO was set up in four concentrations, i.e. VCO 100%, 50%, 25%, 12.5%, fluconazole and distilled water as the control. The VCO antifungal test is performed through diffusion using the disc diffusion method (Kirby and Bauer test). VCO 50% has an average resistor of 5 cm larger than the average inhibition power of VCO 100% that is 4.5 cm, 1 cm on VCO 25% and 0.7 cm on VCO 12.5%. The inhibitory power of the fluconazole drug discs showed significantly different inhibitory power results (P-value <0.05) compared with VCO with 100%, 50%, 25%, 12.5% concentration, and distilled water. While the results of Mann Whitney-Test all VCO concentrations did not show differences in inhibitory results on the growth of *C. albicans* (P> 0.05), except in VCO concentrations 50% compared with 12.5% and distilled water (p <0.05). All VCO concentrations can inhibit the growth of *Candida albicans* in-vitro starting from concentration but the inhibitory power of the VCO test group is not as effective as the inhibitory power of the fluconazole drug disc.

1 INTRODUCTION

Candidiasis is a fungal disease that is caused by *Candida sp*. The most common cause is *Candida albicans* species. *Candida albicans* infection may affect the mouth, vagina, skin, nails, respiratory tract, and at some point can also cause septicemia, endocarditis, or meningitis if not adequately treated (Kuswadji, 2007).

Candida albicans is one of the commensal organisms that act as a normal flora of the human body especially in the gastrointestinal mucous membranes (24%) and the vaginal mucosa (5-11%) and this species is in normal state, therefore it is harmless to our body (Kayser, 2010). However, in the event of a disruption such as a weak immune system (in the case of HIV-AIDS), *Candida albicans*, originally a normal flora in the body, may become pathogenic, causing various diseases, such as vulvovaginal candidiasis. Based on the results of the study, three out of four women had at least once experienced vulvovaginal candidiasis in their lives (Hidalgo, 2012).

Pure coconut oil or Virgin Coconut Oil (VCO) is an oil obtained from old and fresh coconut meat (*Cocos nucifera l.*) processed by squeezing with or without water addition, with or without heating that is not more than 60oC and safe for human consumption (SNI, 2008). Based on the research, the content of lauric acid and capric acid in VCO can kill *C. albicans* by destroying the *C. albicans* plasma membrane and making its cytoplasm shattered and shrunk. Therefore, the VCO is possible when used as an infectious treatment caused by *Candida albicans* that infects the skin and mucosa, and is also possible to be used as an antibiotic therapy for long periods of time (Bergsson, 2001). Another research results on the antimicrobial power of VCO indicate that VCO has minimum inhibitory level and minimum killing rate of 25% by dilution and diffusion method (Nurjannah, 2012).

Based on the results of research conducted by Kabara (2005) showed that lauric acid contained in VCO has a bacteriostatic effect on gram-positive bacteria and also *Candida albicans* fungi. Where the researcher used fatty acid and its derivatives as antimicrobial substances tested on 9 gram-positive bacteria, i.e. *S. aureus, S. epidermidis, Streptococcus beta-hemolytic group A,*

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Streptococcus beta-hemolytic group non-A, Streptococci group D, Corynebacteria, Micrococcus sp., Nocardia asteroides, Pneumococci, and also *Candida albicans*. In the treatment of *Candida albicans*, the result obtained was the lauric acid having a minimum inhibitory level of 2.49 micromol/ml, the capric acid had a minimum inhibitory level of 2.9 micromol/ml, while the caprylic acid had no minimum inhibitory content.

According to Bergsson (2001), the sensitivity of *Candida albicans* to fatty acids and some of its monoglycerides tested by short-acting inactivation, and ultrathin were studied using an electron transmission microscope (TEM) after being given a capric acid. The results showed that the capric acid (C-10) caused the quickest and most effective killings of all three *C. albicans* strains tested, leaving the cytoplasm in an irregular and shrunken state due to the disruption or destruction of the *Candida albicans* plasma membrane. Lauric acid (C-12), is the most active at lower concentrations and after incubation time is longer. Here's a picture of electron microscope *Candida albicans* morphology after being given capric acid.

The objective of this research is to see the inhibitory power of virgin coconut oil to the growth of *Candida albicans* in-vitro.

2 METHOD

The type of this research is laboratory experiments. This study examined the minimum inhibitory levels of virgin coconut oil (Virgin Coconut Oil) on the growth of Candida albicans in vitro. The research was conducted at the Microbiology Laboratory, Faculty of Medicine, University of North Sumatera. The sample of this study is the culture of *Candida albicans* fungus taken from the Microbiology Laboratory, University of North Sumatera.

The culture of *Candida albicans* is done by making the germ suspension made by taking the culture result (+) with osse, then diluted with 0.9% NaCl sterile and adjusted to 0.5 McFarland solution. The germ solution is taken with a sterile cotton swab, emphasized on the edge of the tube until it does not drip when removed. Then the cotton swab was evenly applied on the surface of the Saboraud Dextrose Agar medium and waited until it dried. After drying, the disc to be tested is taken with sterile tweezers and placed on Saboraud Dextrose Agar media for 24 media. Then incubated for 24 hours at 37^{o}C.

VCO preparations were made into four concentrations. A total of 12 sterile 3 ml volume reaction vessels were provided for 3 series VCO dilutions. For each series of dilutions prepared 4 sterile reaction tubes, then numbered from 1 to 4. As much as 2 ml VCO is inserted in tube 1 (for 100% VCO concentration). Then as much as 1 ml VCO is inserted in tube no.2 and mixed with tween 80 as much as 1 ml (total volume 2 ml) then stirred until homogeneous (for making VCO concentration 50%). The VCO has then added as much as 0.5 ml on tube number 3, then mixed with tween 80 as much as 1.5 ml, then mixed until homogeneous (to make VCO concentration 25%). Then as much as VCO 0.25 is inserted into tube number 4, then mixed with tween 80 as much as 1.75 ml (for making VCO concentration 12.5%). Furthermore, blank disc paper is inserted into each test material cylinder (100% VCO, 50%, 25%, 12.5%, distilled water) for 10 minutes for the solution to be absorbed into the disc paper properly.

3 RESULT

The VCO antifungal test is performed by diffusion using the disc diffusion method (Kirby and Bauer test). Each sabouraud dextrose agar medium that has been planted with *Candida albicans* affixed to each paper disc containing 100%, 50%, 25%, 12.5% VCO, distilled water, and fluconazole drug solutions. The preparation is made up of 4 media and each medium is repeated 4 times.

Table 1: The inhibitory power of all test groups on the growth of *Candida albicans* in-vitro

Group	Mean inhibitory zone diameter (mm)	Standard deviation (mm)
Fluconazole	17.7	1.78
VCO 100%	4.5	6.22
VCO 50%	5	1.76
VCO 25%	1	2.23
VCO 12.5%	0.7	1.56
Distilled water	0	0

Then all treated media were incubated for 24 hours at 37 ° C in the incubator. Then perform the drag zone measurements by using the sliding term

on the clear area that occurs around the hole and measured with the measuring paper. The clear area is the diameter of the growing resistance of the fungus being tested. Then interpret the results of the measurement of the clear zone.

The result of the inhibitory zone diameter of all VCO concentrations above is then analyzed by using SPSS with the initial stage determining the normality and homogeneity of the data. From the normality and homogeneity test, the result is that the data distribution is not normal (p> 0.05). Furthermore, non-parametric analysis of the Kruskal Wallis method was performed. The results obtained are p <0.05 (ie 0.006), this indicates that all the test groups there is no significant difference.

Mann Whitney-Test was then performed by comparing each test group. Then the results obtained are Fluconazole drug disc's inhibitory effect showed significantly different inhibitory power results (P <0.05) compared with VCO 100%, 50%, 25%, 12.5%, and distilled water. While the results of Mann Whitney-Test all VCO concentrations did not show differences in inhibitory results on the growth of *C. albicans* (P> 0.05), except in VCO concentrations 50% compared with VCO 12.5% and distilled water (p <0.05) (Table 2).

Table 2: Differences of inhibitory zone results on the growth of *C. albicans.*

Group	Comparison	P-value
Flukonazol	VCO 100%	0.020 (p<0.05)
	VCO 50%	0.020 (p<0.05)
	VCO 25%	0.018 (p<0.05)
	VCO 12,5 %	0.018 (p<0.05)
	Distilled water	0.014 (p<0.05)
VCO 100%	VCO 50%	1.000 (p>0.05)
	VCO 25%	0.321 (p>0.05)
	VCO 12,5 %	0.321 (p>0.05)
	Distilled water	0.131 (p>0.05)
VCO 50 %	VCO 25%	0.069 (p>0.05)
	VCO 12,5 %	0.037 (p<0.05)
	Distilled water	0.013 (p<0.05)
VCO 25%	VCO 12,5 %	0.850 (p>0.05)
	Distilled water	0.317 (p>0.05)
VCO 12,5 %	Distilled water	0.317 (p>0.05)

4 DISCUSSION

VCO concentrations of 100%, 50%, 25%, and 12.5% on average can inhibit the growth of *Candida albicans* fungi. The results obtained are in accordance with the results of research conducted by Diana,(2009) who also conducted research on the antifungal effects of VCO against *C. albicans*. Based on the results he obtained, the Minimum Depression Level (KHM) VCO 100% was 14 mm, VCO 50%: 11 mm, VCO 25%: 10 mm and VCO 12.5%: 8mm. Other studies have also been conducted by Nurjannah (2012) about the antimicrobial power of VCO against *C. albicans* with the tube dilution method. The results show that VCO has anti mycotic power against *C. albicans* with Minimum Fungicidal Concentrations (MFCs) and Minimum Inhibitory Concentrations (MICs) are 25%.

However, other studies show that VCO cannot inhibit the growth of C. albicans in-vitro (Yulian, 2007) (S.S Dewi, T Aryadi, 2010). According to research the factors that influence is due to the poor solubility of VCO in the air (Yulian, 2007). Because in making the concentration of VCO that is by mixing VCO with tween 80 and distilled water with ratio 1:1:2.

The use of tween 80 in this study was as a solvent VCO in the manufacture of concentrations. Tween 80 is a nonionic surfactant and emulsifier derived from polyethoxylated sorbitan and oleic acid. Surfactants are molecules that have hydrophilic groups and lipophilic groups. Can dissolve in the form of air or oil. Tween 80 can lower the voltage between drugs and medium and various micelles. This building will be carried away by micelles dissolved in the medium (Zulkarnain, 2008)

In this study, four repetitions were conducted, it aims to estimate the variations of the experimental error, to estimate the standard error of the treatment rate, to improve the test provision, to extend the precision of the experimental conclusions through the selection and use of experimental units which is more varied. If the number of replications increases, then the alleged population mean value through the midpoint of the observed treatment becomes more thorough. The research error is a measure of diversity among all observations derived from the experimental units that received the same treatment (Raupong, 2011).

5 CONCLUSIONS

All VCO concentrations can inhibit the growth of *Candida albicans* in-vitro from VCO concentrations of 12.5% to 100% VCO. However, the inhibitory power of the VCO test group is not as effective as the inhibitory power of the fluconazole drug disc.

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