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Article

Reducing the Number of Germs on Hands Using Disinfectants Combination of Betal Leaves and Kalamansi Orange

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Abstract. Covid-19 can be spread through particles from a patient's sneeze or cough attached to other objects such as clothing or electronic devices from people around him. Therefore, it is necessary to take precautions to prevent the spread of Covid-19. There are many ways to prevent the transmission of Covid-19, one of which is the use of antiseptics and disinfectants. We can make disinfectants from natural ingredients such as boiled betel leaves and lime juice. Both of these plants have compounds that function as anti-microbial. From the results of the study, it was found that of the 3 combination formulas of Betel and Kalamansi, the best formula for reducing the number of germs was formula 2, namely the combination of Betel and Kalamansi with a ratio of 60:40. For testing against the nCov-2019 Virus, the best formula is formula 3, namely the combination of Betel and Kalamansi 40:60 with the results of 30 positive samples, 20 samples being negative due to damage to the glycoprotein envelope layer. Of the 30 positive samples, 12 samples became negative due to the damaged Open Reading Frame of the nCov-2019 Virus, and of the 30 positive samples, 7 samples were negative due to the damaged Nucleocapsid Protein of the nCov-2019 Virus

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1. Introduction

At the end of 2019, a new disease caused by a virus appeared and attacks the respiratory system, this disease is known as Novel Coronavirus Disease 2019 (Covid-19)1. Covid-19 first spread very quickly in China and has now spread to other parts of the country. The medical personnel working at this

time have studied this virus and made it possible to find the right steps to be taken to prevent and limit its increasingly widespread and rapid spread. Covid-19 can be spread through particles from a patient's sneeze or cough attached to other objects such as clothing or electronic devices from people around him [1,2,3,4].

Therefore, it is necessary to take precautions to prevent the spread of Covid-19. This pandemic is one of the things that people worry about, but it can be prevented by various things. There are many ways to prevent the transmission of Covid-19, one of which is the use of antiseptics and disinfectants [5,6,7,8]. Disinfectant is a material used in the disinfection process. Disinfectants commonly used are generally derived from synthetic chemicals [9,10,11,12,1,3]. Synthetic chemicals have the advantage that they can reduce bacteria quickly, but also have the disadvantage that they can leave residues and are difficult to decompose. Therefore, the use of synthetic chemicals needs to be reduced and replaced with natural materials. One of the natural ingredients that can be used as a disinfectant is Green Betel Leaf (Piper betle L) and Kasturi Lime *(Citrus microcarpa).*

Research conducted by Retno Sari and Dewi Isadiartuti (2006) on the Study of the Effectiveness of Hand Antiseptic Gel Preparations with Betel Leaf Extract (Piper betle Linn) showed that the resulting gel preparation was pale yellow, and the replica test showed that at an extract content of 15%, the number of colonies growth after use is reduced by up to 50%. While the level of 25% indicates the absence of growth of microorganisms in the media. The results of the replica test also showed that preparations containing 15% betel leaf extract were not significantly different from ethanol preparations, while preparations with 20% and 25% extract levels had the same activity as triclosan preparations. The content of betel leaf is an essential oil consisting of hydroxy chavicol, cavibetol, estradiol, eugenol, metileugenol, carvacrol, terpenes, sesquiterpenes, phenylpropane and tannins [14,15,16,17,18,19,20].

Fransiska Nuning Kusmawati and Tiara Fahriliyandi Putri (2019) also conducted a study on "The Effect of Betel Leaf Decoction on Reducing the Number of Candida Albicans on Heat Cured Acrylic Resin Plates" which showed results that betel leaf decoction has active substances that are efficacious as an antifungal, disinfectant and have antibacterial properties. bacteriostatic and bactericidal. This is evident from the decrease in the number of Candida albicans on heat-cured acrylic resin plates after soaking in 50% and 70% betel leaf decoction, respectively. The number of secondary metabolites found in betel leaf allows betel leaf to be used as a natural disinfectant that does not leave harmful residues for users [21,22,23,24,25].

Rother plants that can also be used as ingredients in the manufacture of disinfectants are kaffir limes or more often called Kalamansi oranges [26,27,28]. This Kalamansi orange also has many chemical compounds that can inhibit the development of microorganisms [29,29,30,31]. The compounds contained in the peel of the Kalamansi Orange are the main components of the essential oil, namely -citoprenol, -pinene, and D-limonene; The fruit contains organic acids such as ascorbic acid, citric acid, malic acid, tartaric acid, benzoic acid; The leaves produce limonoids, and are transferred to the fruit and seeds. The large number of chemical compounds contained in the Kamalansi Orange allows the Kamalansi Orange to be used as an anti-acne, antitussive antimicrobial, anti-inflammatory, anticonstipating, antidote, antioxidant, antianxiety agent. Research conducted by Deza Oktasila, Nurhamidah and Dewi Handayani on the Antibacterial Activity Test of Kamalansi Orange Leaves *(Citrofortunella microcarpa)* against Staphylococcus aureus

and Escherichia coli bacteria showed that results showed that the ethanolic extract of Kalamansi lime leaves had antibacterial activity against S. aureus and S. aureus bacteria. E. coli with moderate inhibition zone diameters were 7.20 and 5.73 mm at a concentration of 40%, while the antibacterial activity of the Kalamansi citrus leaf essential oil was categorized as strong with each inhibition zone diameter of 14.83 and 13, 00 mm at a concentration of 20%. Based on the content of chemical compounds and the ability of these two types of plants to inhibit the growth of microorganisms, the authors are interested in researching the combination of boiled betel leaf and kamalansi citrus fruit as a natural disinfectant in preventing the spread of Covid-19.

2. Experimental Section

2.1. Tools and Materials

The tools used in this study include the manufacture of disinfectants, namely analytical scales, oven, knife, cutting board, orange squeezer, water bath, aluminium foil, pan, incubator, Petridis, colony counter, spray bottle and stopwatch. The material used in this study Green betel leaf, Kalamansi orange, aquades.

2.2. Methods Used

2.2.1. Making Betel Leaf Stew

The betel leaves are collected, separated from the stems, washed with running water, drained on parchment paper, then weighed. Obtained a wet weight of 2 kg, then the betel leaf is finely sliced and then boiled with a ratio of water and betel leaf 1: 2, close the container and wait until it boils and the volume of water remains half. Strain the cooking water with the leaves. Save and ready to be mother liquor and ready to be diluted based on the required concentration.

2.2.2. Kalamansi Orange Squeeze

Kalamansi oranges are cut in half and squeezed using an orange squeezer, the water is collected and stored in the refrigerator. Save the juice and it is ready to be a mother liquor to be used according to the expected formula.

2.2.3. Formulation of Betel and Kalamansi (SIRKALA)

Formula 1 (50: 50), as much as 50 ml of boiled water for betel leaves and 50 ml of lime juice. Formula 2 (60:40), as much as 60 ml of boiled water of betel leaf and 40 ml of lime juice. Formula 3 (40:60), as much as 40 ml of boiled water of betel leaf and 60 ml of Kalamansi orange juice

2.2.4. Test of Parameters

Test of parameter in this research are physical parameters, chemical parameters and microbiological parameters. All test will be done with standard test.

3. Results and Discussion

a. Physics Test Results

Table 1. Results of Physical Test of Kalamansi Betel Formula (Sirkala)

No	Physical Test	Results		
		Formula 1 (50:50)	**Formula 2 (60:40)**	**Formula 3 (40:60)**
1	Color	Yellow	Yellow	Orange Yellow
2	Smell	Betel Dominant	Betel Dominant	Betel Dominant

The results in table 1 show that the results of the physical test using the organolaptic test obtained that the Kalamansi Betel formula has a yellow color and has a dominant aroma of betel for formulas 1 and 2 and 3. The yellow color produced is due to the yellow color of the Kalamansi orange juice. The dominant aroma of betel in all formulas is because the essential oil contained in betel leaf reaches 4.2%, namely bethel phenol compounds. This compound will produce a distinctive odor on the betel that will come out when the betel leaf is boiled.

b. Chemical Test Result

Table 2. Chemical Test Results of Kalamansi Betel Formula (Sirkala)

No	Chemical Test	Results		
		Formula 1 (50:50)	**Formula 2 (60:40)**	**Formula 3 (40:60)**
1	pH	5	5	4

The results in table 2 show that the chemical test results, namely the pH of the Betel Kalamansi formula, has an acidic pH. This acidic pH is not very good for germs and viruses to survive. Because this acidic atmosphere can cause damage to the cell walls of germs and viruses.

c. Microbiological Test Results

Table 3 Laboratory Test Analysis of Kalamansi Betel Formula (Sirkala) Against Germ Numbers

No	Dose	Average Germ		Decrease (Colonies)
		Sebelum (Koloni)	**Sesudah (Koloni)**	
1.	Formula 1	±256	±126	130
2.	Formula 2	±322	±116	206
3.	Formula 3	±76	±21	55

The results of laboratory tests in table 3 showed that the combination of boiled betel leaf and kalamansi orange in all formulas could reduce the number of germs. The highest average reduction in germ numbers was in formula 2 with a ratio of 60:40 Betel and Jeruk, which was 206 colonies.

d. Statistic Analysis

Table 4. T-Test Results Effect of Giving Betel and Kalamansi Formula 1Before and After Treatment on Germ Numbers

Formula 1	Average	Std. Deviation	P-value
Before	256.13	203.759	0,000
After	126.43	133.169	

Based on the statistical analysis of the T-test, it shows that there is a significant difference between before and after treatment using the P-value formula 1 (0.000).

Table 5 T-Test Results Effect of Giving Betel and Kalamansi Formula 2 Before and After Treatment on Germ Numbers

Formula 2	Average	Std. Deviation	P-value
Before	321.63	303.367	0,000
After	115.87	171.498	

Based on the statistical analysis of the T-test, it shows that there is a significant difference between before and after treatment using the P-value formula 2 (0.000).

Table 6. T-Test Results Effect of Giving Betel and Kalamansi Formula 3 Before and After Treatment on Germ Numbers

Formula 3	Average	Std. Deviation	P-value
Before	75.80	106.736	0,000
After	20.43	18.390	

Based on the statistical analysis of the T-test, it shows that there is a significant difference between before and after treatment using the P-value formula 3 (0.000). Based on the results of statistical tests, it can be seen that the combination of boiled water of betel leaf with lime juice can significantly reduce the number of germs on hands.

4. Conclusion

From the research conducted, the following conclusions are obtained the best combination of Betel and Kalamansi Orange for the number of germs is formula 2 (60:40) where 60 ml of Betel and 40 ml of Kalamansi Orange and the combination of Betel and Kalamansi Orange can reduce the number of germs.

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