RESEARCH PROTOCOL

The Effect of Chlorhexidine Gluconate 0.2% Mouthwash and 1% Povidone Iodine on SGTF and Non-SGTF SARS-CoV-2 (Clinical trials on Covid-19 positive patients with mild to asymptomatic symptoms)

Introduction

Since the emergence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) in December 2019 until now, more than 458 million people have been infected with SARS-CoV-2, and this number continues to increase every day. Data from the World Health Organization (WHO) states that by February 2022, more than 6 million people had died due to Coronavirus disease 2019 (Covid-19). This makes SARS-CoV-2 a disease capable of spreading rapidly and largely causing respiratory disorders. Transmission of SARS-CoV-2 can occur through respiratory processes or through coughing and sneezing by infected individuals. Additionally, transmission of SARS-CoV-2 can also occur through direct contact with contaminated surfaces followed by touching the nose, mouth, and eyes.[1–5] Patients suspected of being infected with SARS-CoV-2 must immediately undergo laboratory testing for confirmation and as an initial step in preventing transmission. The laboratory test in question is Reverse Transcriptase-polymerase chain reaction (RT-PCR), which currently serves as the diagnostic modality for detecting confirmed cases of SARS-CoV-2.[6–8] RT-PCR results can illustrate specific characteristics of certain SARS-CoV-2 variants. Among them, when the S gene (Spike glycoprotein) is not detected (SGTF) and other target genes besides the S gene are detected (Non-SGTF). The failure to detect the S gene (S Gene Target Failure) is due to an amino acid deletion in the S gene, commonly referred to as SGTF, and is found in the Omicron variant. The Non-SGTF group may represent other variants of the SARS-CoV-2 virus. The Omicron variant is currently the most frequently encountered and spreads more easily compared to other variants. Through SGTF and Non-SGTF, we can more specifically classify the SARS-CoV-2 variants. Thus, SGTF can serve as an initial screening method for detecting SARS-CoV-2 variants that have undergone mutations.[9–13] Certain variants of SARS-CoV-2 may have a high transmission rate, particularly through the oral cavity, especially from the saliva of infected individuals. This makes the oral cavity one of the primary transmission routes for SARS-CoV-2. According to research conducted by To et al., SARS-CoV-2 can be detected in saliva. This further confirms that transmission can occur

through saliva droplets.[14] Due to the high potential of SARS-CoV-2 presence in the oral cavity, preventive measures are necessary to avoid transmission from infected patients to healthcare workers, especially oral and maxillofacial surgeons whose field of work involves the oral cavity. These preventive measures can be implemented before the oral and maxillofacial surgeon performs treatment or surgical procedures on the patient. One form of prevention is the administration of mouthwash to the patient prior to procedures performed by healthcare workers. This preventive measure is expected to reduce the number of SARS-CoV-2 viruses in the oral cavity, thereby lowering the risk of transmission. Huang et al. explained that chlorhexidine gluconate effectively (86.0%) reduces SARS-CoV-2 in the oropharynx.[5] Chlorhexidine gluconate is a broad-spectrum antiseptic that works against gram-positive and gram-negative bacteria, both aerobic and anaerobic, by increasing cell wall permeability, eventually causing cell lysis. Yoon et al. also described the effectiveness of chlorhexidine gluconate in suppressing SARS-CoV-2.[15] In addition, the Center for Disease Control and Prevention (CDC) recommends the use of povidone-iodine-based mouthwash prior to performing procedures within the oral cavity. The use of 1% povidone-iodine has also been proven effective in reducing the amount of SARS-CoV-2 virus in the oral cavity, as reported by Anderson et al.[16,17] Although the use of chlorhexidine gluconate and povidone-iodine mouthwashes has been proven in vitro to reduce SARS-CoV-2 in the oral cavity, there are still few clinical studies conducted directly on infected patients, including those in the SGTF and Non-SGTF groups. Therefore, the authors aim to evaluate the effect of using 0.2% chlorhexidine gluconate and 1% povidone-iodine, as well as compare both, on viral load through CT values of RT-PCR in the SGTF and Non-SGTF groups of SARS-CoV-2.

Identification of problems

Based on the background described above, the problem identified in this study is the high risk of SARS-CoV-2 transmission. This risk can be mitigated by gargling with 1% povidone-iodine prior to procedures, as recommended by the American Dental Association. This preventive measure is expected to reduce the viral load of SARS-CoV-2 in the oral cavity, thereby helping to prevent transmission, particularly from infected patients to healthcare workers, especially oral and maxillofacial surgeons. The researcher is interested in evaluating the effect of gargling with 1% povidone-iodine.

Research Questions

1. What is the effect of gargling with 0.2% chlorhexidine gluconate on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2?
2. What is the effect of gargling with 1% povidone-iodine on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2?
3. What are the differences between gargling with 0.2% chlorhexidine gluconate and gargling with 1% povidone-iodine on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2?
4. What is the relationship between the SGTF and Non-SGTF groups of SARS-CoV-2 and the CT value of RT-PCR?

Research Objectives

1. Aim

To evaluate the effect of gargling with 0.2% chlorhexidine gluconate and 1% povidone-iodine on viral load through CT values of RT-PCR in SGTF and Non-SGTF SARS-CoV-2.

2. Objectives

- To determine the effect of gargling with 0.2% chlorhexidine gluconate on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2.

- To determine the effect of gargling with 1% povidone-iodine on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2.

- To determine the differences between gargling with 0.2% chlorhexidine gluconate and gargling with 1% povidone-iodine on the CT value of RT-PCR in SGTF and Non-SGTF SARS-CoV-2

Research Benefits

1. Benefits to Patients and Society

- This study is expected to serve as a guideline for the general public as one of the methods to reduce the risk of transmission from patients infected with SARS-CoV-2. It is also hoped that this study can contribute to supporting the healing process as an adjunctive therapy for patients infected with SARS-CoV-2 in both the SGTF and Non-SGTF groups.

2. Benefits for Educational Institutions

- The benefits of this research are expected to provide an alternative mouthwash formulation that can be recommended for use prior to oral surgical procedures in patients infected with SARS-CoV-2, in order to reduce the risk of SARS-CoV-2 transmission in both the SGTF and Non-SGTF groups.

3. Benefits for Researchers

- This study is expected to serve as a guideline for preparation prior to performing oral surgical procedures in patients infected with SARS-CoV-2, in order to reduce the risk of SARS-CoV-2 transmission to healthcare workers, particularly to oral and maxillofacial surgeons. The preventive measure of gargling is also expected to serve as

a reference for oral and maxillofacial surgeons in preparing patients before performing treatment procedures in both the SGTF and Non-SGTF groups.

RESEARCH METHODS

Research Design

The design of this study is a quasi-experimental study aimed at evaluating the effect of gargling with 0.2% chlorhexidine gluconate and 1% povidone-iodine on viral load through CT values of RT-PCR in SGTF and Non-SGTF groups of SARS-CoV-2.

Research Place and Time

This study was conducted in the Microbiology Laboratory of RSUP Persahabatan from September to December 2022.

Research Subjects

The population in this study consists of all patients infected with SARS-CoV-2. The research subjects are SARS-CoV-2-infected patients at RSUP Persahabatan who are either asymptomatic or exhibit mild symptoms, and who have been confirmed positive through testing at RSUP Persahabatan.

The number of research subjects was determined using G-Power 3.1.9.7 software, with an effect size of 1.1, resulting in a calculated requirement of 13 subjects in each experimental group (with an error rate of 5% and power of 80%).

The researcher set the number of subjects in each group at 15 patients. Thus, the total number of research subjects is 45 patients. All subjects were divided into three groups: treatment group 1 (gargling with 0.2% chlorhexidine gluconate), treatment group 2 (gargling with 1% povidone-iodine), and the control group (gargling with mineral water).

Inclusion and Exclusion Criteria

Inclusion Criteria

1. SARS-CoV-2-infected patients who have been confirmed positive through a single RT-PCR examination.
2. Patients within less than 3 days after being confirmed positive for SARS-CoV-2.
3. RT-PCR value ≤ 30.
4. Outpatients with mild symptoms or asymptomatic.

Exclusion Criteria

1. Patients with comorbidities or underlying medical conditions.
2. Patients with a history of allergy to povidone-iodine or chlorhexidine gluconate mouthwash.
3. Pregnant patients.
4. Patients who are unwilling to participate as research subjects.

Statistical Analysis

After the data were collected, consisting of CT values from RT-PCR both before and after the treatment, normality testing (Shapiro-Wilk test) was performed. If the data were found to be normally distributed, parametric testing using One-way ANOVA would be conducted. If the data were not normally distributed, nonparametric testing using the Friedman test would be carried out. Data analysis was performed using SPSS Statistics Version 22.0.

Research Ethics

All patient data in this study will be kept confidential and will be used solely for the purposes of this research. The researcher will obtain informed consent from patients who agree to participate in this study. The informed consent form, following the format used at RSUP Persahabatan, includes the research objectives, research benefits, procedures, procedure methods, risks, complications/side effects, actions to be taken in the event of complications/side effects, and compensation in case of complications/side effects, as detailed in the appendix. This study has passed ethical review by the Health Research Ethics Committee of RSUP Persahabatan with approval letter number: No.73/KEPK-RSUPP/08/2022.

Operational Definitions

No	Variable	Operational Definition	Measurement	Unit of Measure	Scale
1	Gargling	A process involving the repetitive movement of 15 mL of liquid within the oral	Patient recording was categorized into three groups: the 0.2% chlorhexidine gluconate group,	Group gargling with 0.2% chlorhexidine gluconate	Categorical (Nominal)

		cavity for 30 seconds and in the posterior throat for 30 seconds, performed by the research subjects three times a day for 5 days.	the 1% povidone-iodine group, and the mineral water group.	Group gargling with 1% povidone-iodine Group gargling with mineral water	
2	CT Value of SARS-CoV-2 RT-PCR	The number of thermal cycles required for the fluorescent signal to cross the threshold in an RT-PCR machine when detecting the presence of SARS-CoV-2 genetic material from oropharyngeal specimens collected by trained personnel at RSUP Persahabatan.	The value obtained from the Bioneer Exicycler™ 96 RT-PCR machine (EXI96V4-OSN-BXA012) using mBioCov–19 reagents.	Cycle	Numeric (Interval)
3	SGTF and Non-SGTF	A characteristic pattern from RT-PCR examination results where target genes other than the S gene are detected,	The RT-PCR results are categorized into two groups: the SGTF group and the Non-SGTF group.	S gene not detected (SGTF) S gene detected (Non-SGTF)	Categorical (Nominal)

	while the S gene is not detected.

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