



Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.

## ORIGINAL ARTICLE

# USE OF MOUTHWASH AND DENTIFRICE CONTAINING AN ANTIMICROBIAL PHTHALOCYANINE DERIVATIVE FOR THE REDUCTION OF CLINICAL SYMPTOMS OF COVID-19: A RANDOMIZED TRIPLE-BLIND CLINICAL TRIAL

MARCELO LUPION POLETI<sup>a</sup>, DANIELLE GREGÓRIO<sup>b</sup>, ALISSON GABRIEL IDELFONSO BISTAFFA<sup>b</sup>, KAREN BARROS PARRON FERNANDES<sup>c,d</sup>, FABIANO VIEIRA VILHENA<sup>e</sup>, PAULO SÉRGIO DA SILVA SANTOS<sup>f</sup>, ANDRÉA NAME COLADO SIMÃO<sup>g</sup>, MARCELL ALYSSON BATISTI LOZOVYOY<sup>g</sup>, BERENICE TOMOKO TATIBANA<sup>a</sup>, AND THAIS MARIA FREIRE FERNANDES<sup>b</sup>

<sup>a</sup>Federal Institute of Paraná, Londrina, PR, Brazil

<sup>b</sup>University of North Paraná (UNOPAR), Londrina, PR, Brazil

<sup>c</sup>School of Medicine, Pontifical Catholic University of Parana (PUCPR), Londrina, PR, Brazil

<sup>d</sup>Department of Health Sciences, Université du Québec à Chicoutimi (UQAC), Chicoutimi, Québec, Canada

<sup>e</sup>Trials - Oral Health & Technologies, Bauru, SP, Brazil

<sup>f</sup>Department of Surgery, Stomatology, Pathology, and Radiology, Bauru School of Dentistry, University of São Paulo, Bauru, SP, Brazil

<sup>g</sup>Research Laboratory in Applied Immunology, Department of Pathology, Clinical Analysis and Toxicology, State University of Londrina, Londrina, PR, Brazil

## ABSTRACT

### Purpose

This clinical trial aimed to evaluate the use of mouthwash and dentifrice containing an antimicrobial phthalocyanine derivative (APD) to reduce the clinical symptoms in patients with COVID-19.

### Methods

This randomized, triple-blind clinical trial enrolled 134 patients aged 18 years or older who underwent COVID-19 testing through the use of nasopharyngeal swab RT-qPCR in a reference center for the diagnosis of COVID-19, had no clinical contraindications to mouthwash and gargle, and had access to cell phones with communication applications. According to the use of a mouthwash and dentifrice containing antimicrobial phthalocyanine derivatives (APD), patients were randomly assigned (1:1) to the APD or non-APD (control) group. All participants were instructed to floss twice a day, brush teeth for 2 minutes 3 times a day, and gargle/rinse (5 mL) for 1 min/3 times a day for 7 days. An online questionnaire was sent to collect data on the clinical symptoms of COVID-19 3 times: T0 (baseline before using the oral hygiene products), T3 (3 days after), and T7 (7 days after). The investigators, patients, and outcome assessors were blinded to group assignment. The Mann-Whitney, Chi-Square, Fisher's exact, and Cochran's tests were used according to the nature of the variables studied, with the level of significance set at  $P < .05$ .

### Results

No statistically significant difference was found in the prevalence of symptoms between groups at baseline. A statistically significant reduction in clinical symptoms was found in the control group (fatigue, shortness of breath, hoarse voice,

Corresponding author: Marcelo Lupion Poleti. Federal Institute of Paraná Rua João XXIII, 600 Jardim Judith, 86.060-370, Londrina, PR, Brazil.  
E-mail: [marcelo\\_poleti@yahoo.com.br](mailto:marcelo_poleti@yahoo.com.br)

## KEYWORDS

Covid-19, Mouthwash, Dentifrices, Clinical symptoms

**CONFLICTS OF INTEREST:** The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper. Dr. Vilhena has a patent pending. The other authors have no conflicts of interest to declare

**SOURCE OF FUNDING:** This work was supported by TRIALS – Oral Health & Technologies.

Received 16 November 2021; revised 7 September 2022; accepted 11 September 2022;

J Evid Base Dent Pract 2022; [101777]

1532-3382/\$36.00

© 2022 Elsevier Inc.

All rights reserved.

doi: <https://doi.org/10.1016/j.jebdp.2022.101777>

sore throat, nasal congestion, and chest pain) and APD group (cough, fatigue, shortness of breath, hyposmia/anosmia, dysgeusia, hoarse voice, sore throat, nasal congestion, chest pain, diarrhea, and irritability/confusion) during the follow-up period. There were statistically significant differences, with a higher prevalence of symptoms in the control group at T3 and T7. Dysgeusia, sore throat, and irritability/confusion were less prevalent in the APD group at T3, and shortness of breath, hyposmia/anosmia, dysgeusia, hoarse voice, sore throat, diarrhea, and irritability/confusion were more prevalent in the control group at T7.

### Conclusions

Based on this methodology, the results demonstrated that the regular use of mouthwash and dentifrice-containing APD had a positive impact on the clinical symptoms, as reported by patients with COVID-19.

## INTRODUCTION

Oral health care has been reported as one of the challenges of COVID-19.<sup>1-5</sup> Recent surveys have shown that global neglect of dental care has affected the population mainly due to the lack of self-care, which directly impacts general health.<sup>1,5-9</sup> Since the involvement of the mouth in the pathophysiology of COVID-19<sup>2</sup> as a reservoir of SARS-CoV-2, many studies have been carried out.

Antiseptic products have already been used in oral health care. Moreover, during the pandemic, it was hypothesized that these products could be used as adjuvants in the prevention of COVID-19 infection, by reducing the oral viral load of SARS-CoV-2.<sup>10-12</sup> Recent studies have demonstrated promising clinical and laboratory findings such as intraoral reduction of viral load of SARS-CoV-2, virucidal efficacy against salivary SARS-CoV-2, anti-inflammatory effectiveness, and possible benefit for COVID-19 with evidence-based approach.<sup>10-16</sup> In contrast, the daily use of certain oral hygiene virucidal products could cause adverse in patients exposed to long-term povidone iodine (thyroid problems), and chlorhexidine (alteration in taste, staining of teeth, and calculus formation).<sup>17-18</sup>

In the pandemic context, few oral antimicrobial products have been investigated for the maintenance of oral hygiene and as a measure to prevent COVID-19, especially those containing chlorhexidine, hydrogen peroxide, povidone-iodine, and, more recently, an antimicrobial phthalocyanine derivative (APD).<sup>19,20</sup> Phthalocyanine derivative is a compound with oxidizing properties, with principle of promoting self-activation and continues production of reactive oxygen in the presence of molecular oxygen. This production of reactive oxygen with phthalocyanine derivative can occur either associated with photodynamic therapy or free.<sup>15</sup> In previous studies using APD, the authors reported promising outcomes such as reduction of clinical symptoms (sore throats, mouth ulcers, and cough) and viral load of SARS-CoV-2.<sup>19, 21-23</sup> However, as this is a new technology, there are no results from clinical trials with APD in patients in the early stage of the COVID-19 disease.

In the current study, we tested the hypothesis that the use of a mouthwash and dentifrice containing APD would reduce the clinical symptoms of COVID-19 during the early stage of the disease.

## PATIENTS AND METHODS

### Trial Design

This was a randomized (1:1), triple-blind, parallel-group clinical trial, conducted in a reference center that performed the diagnosis of COVID-19. The study protocol was approved by the National Commission of Research Ethics, Brazil, on October 22, 2020 (# 35,530,020.1.0000.8156), upon acquisition of the Londrina Municipal Health Authority, and was registered in the Brazilian Registry of Clinical Trials (REBEC; RBR-8 x 8 g36). This study was performed in accordance with the Consolidated Standards of Reporting Trials (CONSORT).<sup>24</sup>

### Participants

Eligible participants were adult patients aged 18 years or older who underwent COVID-19 testing through nasopharyngeal swab RT-qPCR, had no clinical contraindications to the use of mouthwashes and gargle, and had access to cell phones that had communication applications (WhatsApp). The exclusion criteria were those with negative COVID-19 test results, those who did not answer the clinical questionnaires, and those who did not use oral products as recommended. The recruitment of participants in this study took place at a reference center for the diagnosis of COVID-19 in Londrina, Brazil, from November 6, 2020, to November 19, 2020. Londrina had 580.000 habitants and a 7-day moving average of 124 new COVID-19 cases when this study was performed.

Sample size calculation was performed based on the APD group (time to clinical improvement, defined as the length of stay in the hospital) of a previous study with hospitalized patients<sup>21</sup> with an alpha level of 0.5 and a beta value of 0.2 to attain 80% test power; 40 patients were required for each group. However, considering that approximately 80% of the COVID-19 tests were negative at the referral center for this

study, 250 patients were needed for each arm in October 2020.

### Randomization and Blinding

Randomization was performed by an external investigator (T. F.) with no clinical involvement in the trial using a computer program (Excel 2007, Microsoft Windows, Microsoft, Chicago, IL), and was stratified by a group with a 1:1 allocation using random block sizes of 20. This investigator inserted the codes according to the randomization schedule in pre-packaged kits with a toothbrush, a floss, and an experimental mouthwash and dentifrice, both with APD (APD group) and non-APD (control group). These kits were identical in appearance, taste, smell, and method of use, and the allocation sequence was kept blinded by the other investigators at all times. After conducting a swab test at the reference center and obtaining the patient's consent, the kits were delivered to patients by 2 investigators (D. G. and A. B.) who assigned the participants to the interventions. The kits were kept blinded to the patients at all times during the clinical trial. Additionally, the analysis of the data and statistical treatment were performed by an external investigator with no clinical involvement in the trial (K. F.), who was unable to identify the intervention groups. Therefore, the investigators, patients, and outcome assessors were blinded to group assignment.

### Interventions

The patients were divided into 2 groups:

- Control group (N = 250): oral hygiene was performed with an experimental mouthwash and dentifrice, both free of APD.
- The APD group (N = 250): oral hygiene was performed with an experimental mouthwash and dentifrice, both containing APD.

Videos with instructions on oral hygiene were sent via WhatsApp. All participants were instructed to floss twice a day, use the same amount of dentifrice, brush the teeth and tongue for 2 min/3 times a day, and then gargle/rinse with 5 mL of mouthwash for 1 min/3 times a day. They were also instructed to wait for 30 minutes after using oral hygiene products to eat or drink anything. This oral hygiene protocol was started on the morning of the first day after swab collection and continued for 7 days. The participants did not use any other oral hygiene products during the study period.

### Data Collection

For the analysis of clinical symptoms, a research instrument was used to collect the self-reports of the episodes, in their natural environment, through an application, according to a previous study.<sup>25</sup> Each participant previously received (at the baseline) instructions for responding to links via mobile messages via WhatsApp. The questionnaire was sent to collect

the demographic characteristics of the patients, clinical data on COVID-19 (symptoms), and oral hygiene product compliance using the Mentimeter system (Mentimeter AB, Stockholm, Sweden) 3 times: T0 (baseline, before using the oral hygiene products), T3 (3 days after), and T7 (7 days after). All questionnaires were answered in the evening and identified using a randomization code.

For the diagnosis of COVID-19, the samples were processed in 2 laboratories (Central Laboratory of Paraná State - Lacen, Curitiba, PR, Brazil, and Research Laboratory in Applied Immunology, Department of Pathology, Clinical Analysis and Toxicology, State University of Londrina, Londrina, PR, Brazil), following the same protocol. The analysis of RNA viral load was performed by RT-PCR using TaqPath COVID-19 multiplex Real-Time RT-PCR test for detection of 3 viral genes (ORF1ab, N, and S genes). Cycle threshold values  $\leq 37$  for 2 or more genes were considered positive for COVID-19.

The primary outcome of the effectiveness of the mouthwash and dentifrice-containing APD for the COVID-19 patients was the comparison of patients who achieved an improvement in clinical symptoms from baseline (T0) to T7.

### Statistical Analysis

The Statistical Package for Social Sciences (version 18.0) was used to conduct all analyses, establishing a confidence interval of 95% and a level of significance of 5% ( $P < .05$ ) for all tests.

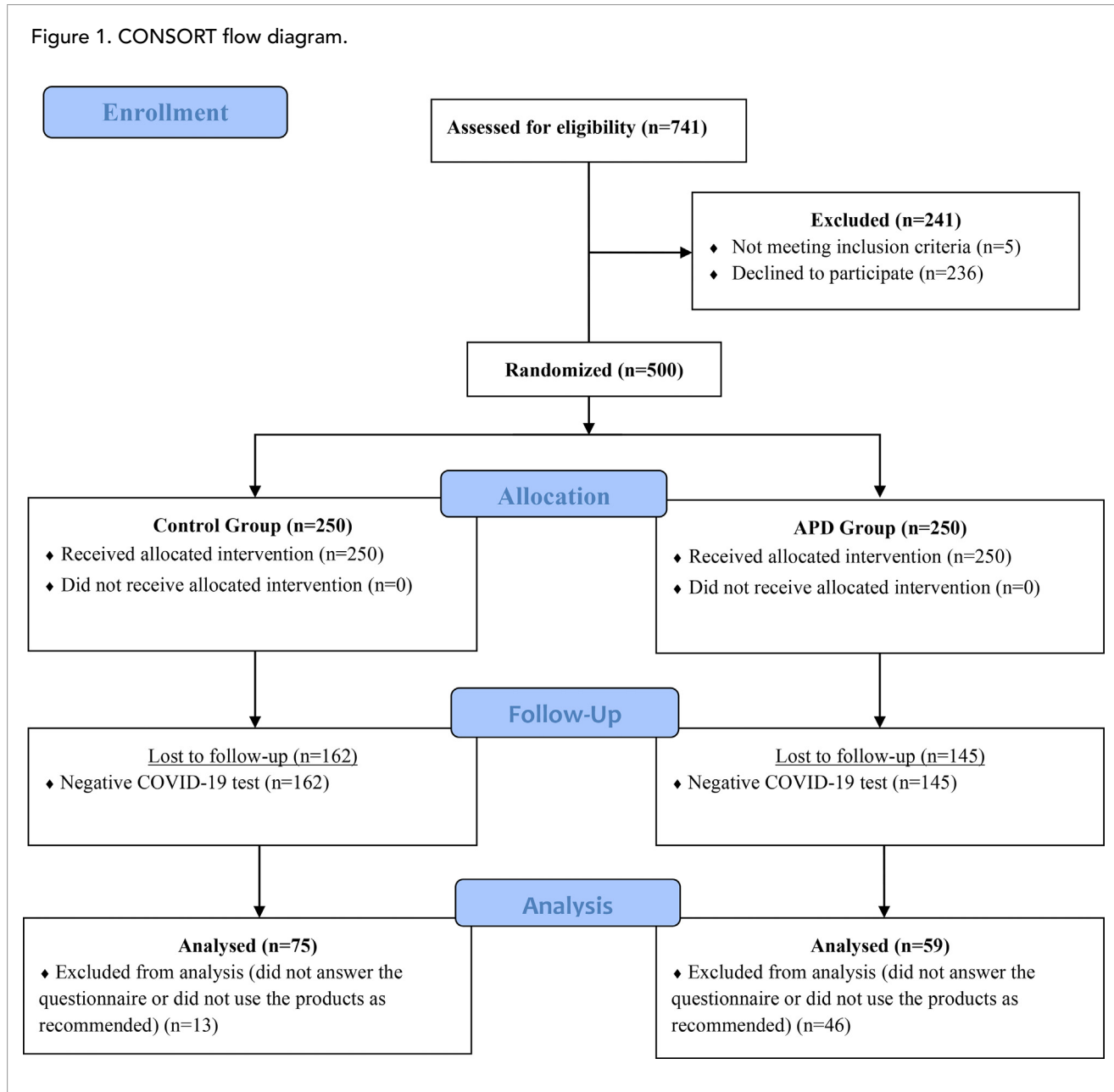
To compare the demographic characteristics among the groups, the Mann-Whitney test was used to compare the age, BMI, and the period between symptom onset and swab collection, and the *Chi-Square* test was used to compare the results as per the sex. Additionally, the *Chi-Square* test or exact Fisher test was used to compare the signs and symptom prevalence between the groups at baseline. Additionally, Cochran's test was used to compare the signs and symptoms within the treatment groups during the follow-up period.

To deal with missing data, strategies were carried out regarding outcomes under the assumptions of missing at random or not at random. The chosen method was the last observation carried forward, which is a type of single imputation, as it substitutes the censored data for the last observed value, under the assumption that this value likely has not changed. Indeed, this conservative method is widely accepted and even recommended by the FDA, since it mimics real-life scenarios of non-compliance of the patients.<sup>26,27</sup>

## RESULTS

Of the 741 individuals assessed for eligibility, 500 were enrolled in the study. After confirming the diagnosis of COVID-19 using RT-PCR, 307 negative cases were lost to follow-up,

Figure 1. CONSORT flow diagram.



and 59 were excluded from analysis because they did not answer the clinical questionnaires or did not use the products as recommended. As shown in the study flowchart (Figure 1), 134 individuals were assessed and all the symptoms were compared during the follow-up period.

Table 1 shows that the groups were paired as per the demographic and clinical characteristics, early pharmacological treatment use for COVID-19, and the possible occurrence of symptoms in relatives of participants enrolled in the study.

Table 2 shows the symptoms observed at T0, T3 and T7. No statistically significant difference was found in the prevalence

of symptoms between groups at baseline. The most prevalent symptoms were fatigue, shortness of breath, hoarse voice, dysgeusia, and hyposmia/anosmia, with a similar prevalence in both groups at baseline; those with a prevalence higher than 20% at the initial assessment were included in the analysis. When comparing the prevalence of symptoms within the groups, dysgeusia, sore throat, irritability/confusion were less prevalent in the APD group at T3. Additionally, at the follow up at day T7, shortness of breath, hyposmia/anosmia, dysgeusia, hoarse voice, sore throat, diarrhea, and irritability/confusion were more prevalent in the control group.

**Table 1. Demographic and clinical characteristics of the study population (n = 134).**

Variables	Control group (n = 75)	APD group (n = 59)	P-value
Age, y (Median ± Interquartile range)	34.2 ± 20.9	35.5 ± 21.4	0.47 <sup>a</sup>
BMI (Median ± Interquartile range)	25.6 ± 6.6	26.7 ± 6.3	0.25 <sup>a</sup>
Sex			
Female, n (percentage)	48 (64.0)	35 (59.3)	0.58 <sup>b</sup>
Male, n (percentage)	27 (36.0)	24 (40.7)	
Polypharmacy use			
Yes, n (percentage)	05 (6.7)	02 (3.4)	0.46 <sup>c</sup>
No, n (percentage)	70 (93.3)	57 (96.6)	
Early pharmacological treatment for COVID			
Yes, n (percentage)	22 (29.3)	19 (32.2)	0.85 <sup>b</sup>
No, n (percentage)	53 (70.7)	40 (67.8)	
Symptoms' presence in relatives after treatment			
Yes, n (percentage)	08 (10.7)	04 (6.8)	0.55 <sup>c</sup>
No, n (percentage)	67 (89.3)	55 (93.2)	
Period between symptoms onset and swab collection, days (Median ± Interquartile range)	4 ± 3	4.5 ± 2	0.33 <sup>a</sup>

BMI: body mass index; APD: the antimicrobial phthalocyanine derivative.  
<sup>a</sup> Mann-Whitney test  
<sup>b</sup> chi-square test  
<sup>c</sup> exact Fisher test;

Additionally, [Table 3](#) depict the major symptom variation after the intervention in the control and APD groups. A reduction in symptoms (fatigue, shortness of breath, hoarse voice, sore throat, nasal congestion, and chest pain) was observed in the control group. In contrast, according to Cochran's test, a marked reduction in all major symptoms was observed in the APD group intervention. No side effects of oral hygiene products were reported in this study.

[Table 4](#) shows a comparison of symptom prevalence during follow-up between the control and APD groups. There were statistically significant differences, with a higher prevalence of symptoms in the control group at T3 and T7.

## DISCUSSION

In light of the ongoing discussion on COVID-19 and oral health management,<sup>2,4,28-30</sup> the overarching aim of this triple-blind randomized clinical trial was to evaluate the use of mouthwash and dentifrice containing APD to reduce clinical symptoms in patients with COVID-19. The results of this

study indicated a reduction in clinical symptoms that were self-reported by patients at home, while corresponding data was collected through a questionnaire.<sup>25</sup> The prevalence of symptoms was examined at baseline (T0), 3 (T3), and 7 (T7) days after using APD in oral hygiene products and compared to the control group. These results are in accordance with clinical and laboratory evidence built over this pandemic period on the beneficial effect of using APD in oral care products against COVID-19 in the studies samples.<sup>14-16,20-23</sup>

In one of these APD studies, the authors reported that hospitalized patients with COVID-19 experienced faster recovery and hospital discharge without disease progression after using an adjuvant APD rinse protocol.<sup>21</sup> The same protocol was used by non-hospitalized patients diagnosed with COVID-19 and was reported in 2 case series.<sup>22,23</sup> In these studies, the patients experienced a reduction in clinical symptoms such as sore throat, cough, and mouth sores and became asymptomatic after a few days of use APD rinse protocol. In addition, a pilot study showed a reduction in the viral load of

Table 2. Comparison of the prevalence of symptoms among groups at T0, T3 and T7 days of follow-up.

Symptoms observed	T0			T3			T7		
	Control group (n = 75)	APD group (n = 59)	P-value	Control group (n = 75)	APD group (n = 59)	P-value	Control group (n = 75)	APD group (n = 59)	P-value
	Yes (%)	Yes (%)		Yes (%)	Yes (%)		Yes (%)	Yes (%)	
Cough	26.7	35.6	0.27	16.0	22.0	0.37	18.7	16.9	0.79
Fatigue	61.3	54.2	0.69	46.7	39.0	0.37	32.0	23.7	0.29
Shortness of breath	53.3	54.3	0.43	42.7	28.8	0.09	37.3	20.3	0.03
Hyposmia/ Anosmia	44.0	52.5	0.33	52.0	44.1	0.36	49.3	32.2	0.04
Dysgeusia	45.3	50.8	0.66	49.3	30.5	0.03	41.3	22.0	0.02
Hoarse voice	54.7	57.6	0.73	48.0	32.2	0.07	33.3	15.3	0.02
Sore throat	38.7	30.5	0.33	29.3	13.6	0.03	14.7	1.7	0.01
Nasal congestion	32.0	28.8	0.69	26.7	16.9	0.18	14.7	8.5	0.27
Chest pain	24.0	28.8	0.53	16.0	10.2	0.32	10.7	12.1	0.80
Diarrhea	21.3	22.0	0.92	18.7	8.5	0.09	13.3	0.0	0.01
Irritability/confusion	29.3	28.8	0.95	29.3	10.2	0.01	20.0	6.8	0.03

Data are shown as n and percentage; P: Chi-Square test; statistical significance set at  $P < .05$ ; APD: the antimicrobial phthalocyanine derivative.

SARS-CoV-2 after brushing teeth with a dentifrice containing APD,<sup>14</sup> and an in vitro study demonstrated SARS-CoV-2 inactivation on the use of a mouthwash (90%) and dentifrice (99.99%), both containing APD.

Understanding the pathophysiology of COVID-19 with the entry of SARS-CoV-2 through the upper airway, affinities for the nasal and oral mucosae, and the salivary glands as reservoirs of the virus,<sup>2,31</sup> is important for conducting investigations in our study. A recent study found the presence of SARS-CoV-2 in periodontal tissue and concluded that periodontal tissue can be a target for SARS-CoV-2 and contribute to the presence of the virus in saliva.<sup>3</sup> In another study,<sup>32</sup> the authors recommend that oral hygiene be maintained, if not improved, during COVID-19 to reduce bacterial load in the oral cavity and the potential risk of superinfection. The authors also stated that poor oral hygiene is considered a risk factor for complications of the disease, especially in patients with comorbidities. Thus, the habit of oral hygiene through mechanical cleaning should be more rigorously associated with chemical action against microorganisms, such as SARS-CoV-2.

In the current study, no statistically significant difference was found in the prevalence of clinical symptoms between

groups at baseline (Tables 1 and 2). There was a significant reduction in 6 self-reported symptoms in the control group during the follow-up period and a significant improvement ( $P < 0.05$ ) in all major COVID-19 symptoms in the APD group (Table 3). Both groups presented with symptom reduction; however, the APD group showed a significant reduction during the follow-up period (94.9%-49.2%, Table 4) and these results may be related to the mechanical, antimicrobial, anti-inflammatory, and tissue regeneration actions.<sup>15,16,20-23</sup> Based on these studies, we did not include a placebo group in this study because the timing of the pandemic required a clinical study to provide direct benefits to all the research participants. Moreover, according to the safety outcomes, no side effects of the oral hygiene protocol were reported by the patients. Therefore, the use of oral hygiene as a strategy to reduce COVID-19 symptoms should be considered.

Evidence suggests that mechanical oral hygiene can reduce the viral load on the mouth and oropharynx, prevent upper respiratory tract infections, and reduce infectivity.<sup>8,11,33,34</sup> However, in our study, we believe that mechanical action was an adjunct that potentiated the action of APD in reducing the load of SARS-CoV-2, which was clinically demonstrated

Table 3. Major prevalence of symptoms during follow-up period in the control and APD groups.

Symptoms observed	Control group				APD group			
	T0 (n = 75)	T3 (n = 75)	T7 (n = 75)	P-value	T0 (n = 59)	T3 (n = 59)	T7 (n = 59)	P-value
	Yes (%)	Yes (%)	Yes (%)		Yes (%)	Yes (%)	Yes (%)	
Cough	26.7	16.0	18.7	0.10	35.6	22.0	16.9	0.01
Fatigue	61.3	46.7	32.0	0.00	54.2	39.0	23.7	0.00
Shortness of breath	53.3	42.7	37.3	0.04	54.3	28.8	20.3	0.00
Hyposmia/Anosmia	44.0	52.0	49.3	0.31	52.5	44.1	32.2	0.02
Dysgeusia	45.3	49.3	41.3	0.38	50.8	30.5	22.0	0.00
Hoarse voice	54.7	48.0	33.3	0.00	57.6	32.2	15.3	0.00
Sore throat	38.7	29.3	14.7	0.00	30.5	13.6	1.7	0.00
Nasal congestion	32.0	26.7	14.7	0.01	28.8	16.9	8.5	0.00
Chest pain	24.0	16.0	10.7	0.03	23.7	10.2	12.1	0.00
Diarrhea	21.3	18.7	13.3	0.34	22.0	8.5	0.0	0.00
Irritability/confusion	29.3	29.3	20.0	0.11	28.8	10.2	6.8	0.00

Data are shown as n and percentage; P: Cochran's test; Statistical significance was set at  $P < .05$ .

Table 4. Prevalence of symptoms during follow-up period between groups.

Group	T0 (%)	T3 (%)	T7 (%)
Control	94.7	82.7	69.3
APD	94.9	64.4	49.2
P-value	0.95	0.02	0.02

Data are shown as n and percentage; P: Chi-Square test; statistical significance set at  $P < .05$ ; APD: the antimicrobial phthalocyanine derivative.

by the greater reduction in self-reported symptoms by patients in the APD group. Thus, as previously reported, the use of APD in oral care products could positively contribute to the improvement of clinical symptomatology in patients with COVID-19.<sup>22</sup>

In addition to mouth and pharyngeal symptoms, diarrhea could happen during COVID-19 infection since SARS-CoV-2 can reach and replicate in intestinal epithelia direct from the mouth.<sup>35</sup> In this study, diarrhea was self-reported by pa-

tients in both groups at baseline, and at the end of 1 week (T7) none of the patients who used the oral hygiene APD protocol presented the symptom, unlike the group that did not use it (Table 2). In the Control Group, of the 16 initial cases, 02 remained with diarrhea and 07 new cases appeared during the 7-day follow-up period (data not shown). Thus, based on self-related of the patients, we believe that the oral hygiene protocol may have contributed to the non-appearance of new cases of diarrhea during the evaluated period, from the reduction of the SARS-CoV-2 viral load in the mouth and pharynx of the patients.

The main limitation of this study was that the clinical data on COVID-19 symptomatology were collected from an electronic system at home. Despite the ease of obtaining information considering the context of the pandemic, without face-to-face interaction with the patients, we could not exclude any symptoms not exclusively associated with COVID-19. Another limitation was that the individual outcomes of the patients taking systemic medications were not evaluated. Despite the homogeneity of the samples at baseline regarding medication use, symptom relief may also be related to supportive medications. The third limitation was compliance with certifying the correct use of oral hygiene products. As it was also conducted electronically, the data



from this study were entirely dependent on the fidelity of participants' responses. Finally, the lack of a placebo group was also a limitation of the study. Despite these limitations, the significant reduction in symptoms during the follow-up period revealed a promising path for the use of oral hygiene care.

The COVID-19 pandemic has presented moments of exacerbation and improvement, and currently, the arrival of new variants of the virus shows that the fight against SARS-CoV-2 should be maintained. Additionally, recent studies demonstrate the effects of mouthwash in reducing the viral load in the saliva of infected individuals. Therefore, the strategy of using oral hygiene products containing antimicrobial agents is an important adjuvant against SARS-CoV-2.

## CONCLUSION

Clinical symptoms were reduced in the control group (fatigue, shortness of breath, hoarse voice, sore throat, nasal congestion, and chest pain) and APD group (cough, fatigue, shortness of breath, hyposmia/anosmia, dysgeusia, hoarse voice, sore throat, nasal congestion, chest pain, diarrhea, and irritability/confusion) during the follow-up period. Dysgeusia, sore throat, and irritability/confusion were less prevalent in the APD group at T3, and shortness of breath, hyposmia/anosmia, dysgeusia, hoarse voice, sore throat, diarrhea, and irritability/confusion were more prevalent in the control group at T7.

Based on this methodology, the results demonstrated that the regular use of mouthwash and dentifrice-containing APD had a positive impact on the clinical symptoms reported by patients with COVID-19.

## ACKNOWLEDGMENT

The authors gratefully acknowledge the Londrina Municipal Health Authority for their support in this study.

## CREDIT AUTHOR STATEMENT

**Marcelo Lupion Poletia:** Conceptualization; Data curation; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Resources; Software; Supervision; Validation; Visualization; Writing - original draft; Writing - review & editing. **Danielle Gregório:** Data curation; Formal analysis; Investigation; Writing - review & editing. **Alisson Gabriel Idelfonso Bistaffab:** Data curation; Formal analysis; Investigation; Writing - review & editing. **Karen Barros Parron Fernandesc:** Formal analysis; Investigation; Software; Writing - original draft; Writing - review & editing. **Fabiano Vieira Vilhenae:** Funding acquisition; Resources; Writing - review & editing. **Paulo Sérgio da Silva Santosf:** Investigation; Methodology; Resources; Software; Validation; Writing - original draft; Writing - review & editing. **André a Name**

**Colado Simão:** Investigation; Methodology; Software; Validation; Writing - review & editing. **Marcell Alysson Batisti Lovoyog:** Investigation; Methodology; Software; Validation; Writing - review & editing. **Berenice Tomoko Tatibanaa:** Investigation; Methodology; Resources; Software; Validation; Writing - review & editing.

## SUPPLEMENTARY MATERIALS

Supplementary material associated with this article can be found, in the online version, at doi:[10.1016/j.jebdp.2022.101777](https://doi.org/10.1016/j.jebdp.2022.101777).

## REFERENCES

1. Aida J, Fukai K, Watt RG. Global neglect of dental coverage in universal health coverage systems and Japan's broad coverage. *Int Dent J.* 2021;71:454–457. doi:[10.1016/j.identj.2020.12.027](https://doi.org/10.1016/j.identj.2020.12.027).
2. Huang N, Pérez P, Kato T, et al. SARS-CoV-2 infection of the oral cavity and saliva. *Nat Med.* 2021;27:892–903. doi:[10.1038/s41591-021-01296-8](https://doi.org/10.1038/s41591-021-01296-8).
3. Matuck BF, Dolhnikoff M, Maia GVA, et al. Periodontal tissues are targets for SARS-CoV-2: a post-mortem study. *J Oral Microbiol.* 2020;13:1848135–1848135. doi:[10.1080/20002297.2020.1848135](https://doi.org/10.1080/20002297.2020.1848135).
4. Matuck BF, Dolhnikoff M, Duarte-Neto AN, et al. Salivary glands are a target for SARS-CoV-2: a source for saliva contamination. *J Pathol.* 2021;254:239–243. doi:[10.1002/path.5679](https://doi.org/10.1002/path.5679).
5. Pepsodent WOHD 2021. Bangladesh research summary report 2021: attitudes, behaviors and experiences of oral health during the covid-19 pandemic. Accessed 06 September 2022. [https://www.pepsodent.com/sk-eu/content/dam/brands/smile/bd/en/smile\\_fact\\_sheet\\_bangladesh\\_final-38049334.pdf](https://www.pepsodent.com/sk-eu/content/dam/brands/smile/bd/en/smile_fact_sheet_bangladesh_final-38049334.pdf).
6. Lu M, Xuan S, Wang Z. Oral microbiota: a new view of body health. *Food Science and Human Wellness.* 2019;8:8–15. doi:[10.1016/j.fshw.2018.12.001](https://doi.org/10.1016/j.fshw.2018.12.001).
7. Zhang S, Liu C, Zhang C, Jiang H, Tai B, Du M. Impact of COVID-19 on the oral health of adults in Wuhan and China: results of a nationwide online cross-sectional questionnaire survey. *BMC Oral Health.* 2021;21:1–7. doi:[10.1186/s12903-021-01533-z](https://doi.org/10.1186/s12903-021-01533-z).
8. González-Olmo MJ, Delgado-Ramos B, Ruiz-Guillén A, Romero-Maroto M, Carrillo-Díaz M. Oral hygiene habits and possible transmission of COVID-19 among cohabitants. *BMC Oral Health.* 2020;20:1–7. doi:[10.1186/s12903-020-01274-5](https://doi.org/10.1186/s12903-020-01274-5).
9. Pinzan-Vercelino CR, Freitas KM, Girão VM, da Silva DO, Peloso RM, Pinzan A. Does the use of face masks during the COVID-19 pandemic impact on oral hygiene habits, oral conditions, reasons to seek dental care and esthetic concerns? *J Clin Exp Dent.* 2021;13:e369–e375. doi:[10.4317/jced.57798](https://doi.org/10.4317/jced.57798).
10. Elzein R, Abdel-Sater F, Fakhreddine S, et al. In vivo evaluation of the virucidal efficacy of chlorhexidine and povidone-iodine mouthwashes against salivary SARS-CoV-2. A randomized-controlled clinical trial. *J Evid Based Dent Pract.* 2021;21:101584. doi:[10.1016/j.jebdp.2021.101584](https://doi.org/10.1016/j.jebdp.2021.101584).

11. Schürmann M, Aljube M, Tiemann C, Sudhoff H. Mouthrinses against SARS-CoV-2: anti-inflammatory effectivity and a clinical pilot study. *Eur Arch Otorhinolaryngol.* 2021;278:5059–5067. doi:10.1007/s00405-021-06873-8.
12. Moosavi MS, Aminishakib P, Ansari M. Antiviral mouthwashes: possible benefit for COVID-19 with evidence-based approach. *J Oral Microbiol.* 2020;12:1794363. doi:10.1080/20002297.2020.1794363.
13. Silva A, Azevedo M, Sampaio-Maia B, Sousa-Pinto B. The effect of mouthrinses on severe acute respiratory syndrome coronavirus 2 viral load: a systematic review. *J Am Dent Assoc.* 2022;153:635–648 e16. doi:10.1016/j.adaj.2021.12.007.
14. Poleti ML, Gregório D, Bistaffa AGI, et al. Toothbrushing with a dentifrice containing antimicrobial phthalocyanine derivative for the intraoral reduction of viral load of SARS-CoV-2: a pilot study. *Res Sq. Preprint (version 1).* 2021. doi:10.21203/rs.3.rs-690819/v1.
15. Santos CA, da Fonseca Orcina B, Reia VB, et al. Virucidal activity of the antiseptic mouthwash and dental gel containing anionic phthalocyanine derivative: in vitro study. *Clin Cosmet Investig Dent.* 2021;13:269–274. doi:10.2147/CCIDE.S315419.
16. Santos P, Orcina B, Alves L, Oliveira R, Zangrando M, Vilhena F. A recommendation of PHTALOX mouthwash for preventing infection and progression of COVID-19. *Acta Scient Dent Sci.* 2020;4:111–112. doi:10.31080/ASDS.2020.04.0991.
17. Schürmann M, Aljube M, Tiemann C, Sudhoff H. Mouthrinses against SARS-CoV-2: anti-inflammatory effectivity and a clinical pilot study. *Eur Arch Otorhinolaryngol.* 2021;278:5059–5067. doi:10.1007/s00405-021-06873-8.
18. Zanatta FB, Antoniazzi RP, Rösing CK. Staining and calculus formation after 0.12% chlorhexidine rinses in plaque-free and plaque covered surfaces: a randomized trial. *J Appl Oral Sci.* 2010;18:515–521. doi:10.1590/s1678-77572010000500015.
19. Santos CA, Vilhena FV, Zangrando MSR. Adjuvant use of phthalocyanine derivative for calculus control. *Int J Case Rep Images.* 2021;12:1–4. doi:10.5348/101220Z01CS2021CI.
20. Vilhena FV, Brito Reia VC, da Fonseca Orcina B, et al. The use of antiviral phthalocyanine mouthwash as a preventive measure against COVID-19. *GMS Hyg Infect Control.* 2021;16 Doc24. doi:10.3205/dgkh000395.
21. da Silva Santos PS, da Fonseca Orcina B, Machado RRG, et al. Beneficial effects of a mouthwash containing an antiviral phthalocyanine derivative on the length of hospital stay for COVID-19: randomized trial. *Sci Rep.* 2021;11:1–10. doi:10.1038/s41598-021-99013-5.
22. BdF Orcina, Vilhena FV, Oliveira RC, et al. A phthalocyanine derivate mouthwash to gargling/rinsing as an option to reduce clinical symptoms of COVID-19: case series. *Clin Cosmet Investig Dent.* 2021;13:47–50. doi:10.2147/CCIDE.S295423.
23. BdF Orcina, PSdS Santos. Oral manifestation COVID-19 and the rapid resolution of symptoms post-phtalox treatment: a case series. *Int J Odontostomat.* 2021;15:67–70. doi:10.4067/S0718-381X2021000100067.
24. Moher D, Hopewell S, Schulz KF, et al. CONSORT 2010 explanation and elaboration: updated guidelines for reporting parallel group randomised trials. *Int J Surg.* 2012;10:28–55. doi:10.1016/j.ijsu.2011.10.001.
25. Menni C, Valdes AM, Freidin MB, et al. Real-time tracking of self-reported symptoms to predict potential COVID-19. *Nat Med.* 2020;26:1037–1040. doi:10.1038/s41591-020-0916-2.
26. Haukoos JS, Newgard CD. Advanced statistics: missing data in clinical research – part 1: an introduction and conceptual framework. *Acad Emerg Med.* 2007;14:662–668. doi:10.1111/j.1553-2712.2007.tb01855.x.
27. Fregni F, Illigens B. *Critical Thinking in Clinical Research: Applied Theory and Practice Using Case Studies.* New York, NY: Oxford University Press; 2018.
28. Imai K, Tanaka H. SARS-CoV-2 infection and significance of oral health management in the era of “the new normal with COVID-19”. *Int J Mol Sci.* 2021;22:1–17. doi:10.3390/ijms22126527.
29. Iranmanesh B, Khalili M, Amiri R, Zartab H, Aflatoonian M. Oral manifestations of COVID-19 disease: a review article. *Dermatol Ther.* 2021;34:e14578. doi:10.1111/dth.14578.
30. Kamel AHM, Basuoni A, Salem ZA, AbuBakr N. The impact of oral health status on COVID-19 severity, recovery period and C-reactive protein values. *Br Dent J.* 2021:1–7. doi:10.1038/s41415-021-2656-1.
31. Jafer MA, Hazazi MA, Mashi MH, et al. COVID-19 and Periodontitis: a reality to live with. *J Contemp Dent Pract.* 2020;21:1398–1403.
32. Sampson V, Kamona N, Sampson A. Could there be a link between oral hygiene and the severity of SARS-CoV-2 infections? *Br Dent J.* 2020;228:971–975. doi:10.1038/s41415-020-1747-8.
33. Satomura K, Kitamura T, Kawamura T, et al. Prevention of upper respiratory tract infections by gargling: a randomized trial. *Am J Prev Med.* 2005;29:302–307. doi:10.1016/j.amepre.2005.06.013.
34. Nagatake T, Ahmed K, Oishi K. Prevention of respiratory infections by povidone-iodine gargle. *Dermatology.* 2002;204:32–36. doi:10.1159/000057722.
35. D’Amico F, Baumgart DC, Danese S, Peyrin-Biroulet L. Diarrhea during COVID-19 infection: pathogenesis, epidemiology, prevention, and management. *Clin Gastroenterol Hepatol.* 2020;18:1663–1672. doi:10.1016/j.cgh.2020.04.001.