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Antimicrobial Effect of Cinnamon Oil Against Oral Microorganisms

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Abstract

Background/Objectives: There has been a growing interest in the antimicrobial activity of natural extracts that inhibit the growth of microorganisms without side effects. Therefore, we evaluated the antibacterial effects of cinnamon oil against *oral microorganisms* existing in dental plaque.

Methods/Statistical analysis: Dental plaque was formed on bovine specimens using a growth medium for 6 days, followed by treatment with either 2.5% cinnamon oil, 0.12% chlorhexidine gluconate, or 10% dimethyl sulfoxide for 5 min, twice per day. On day 7, each specimen was imaged using quantitative light-induced fluorescence–digital to assess microbial activity of dental plaque. A one-way analysis of variance and the Scheffe *post hoc* test were performed to analyze the differences in red/green ratio (R/G values) of dental plaque between the three treatment groups.

Findings: The average R/G values of dental plaque was the lowest in the 2.5% cinnamon oil group (1.180 ± 0.010), and these values were significantly different from those of the chlorhexidine gluconate group (1.249 ± 0.008) ($p<0.001$). Furthermore, the changes in the mean R/G values of each specimen showed statistically significant differences over time ($p<0.05$). Thus, we suggest that cinnamon oil is an effective antimicrobial agent that inhibits the proliferation of anaerobic bacteria in dental plaque.

Improvements/Applications: Further studies should compare and analyze the antimicrobial activity of cinnamon oil against *oral microorganisms* at different concentrations.

Keywords: *Antimicrobial agent, Antimicrobial effect, Cinnamon oil, Oral microorganisms, Oral diseases*

Introduction

According to the 2018 Statistics Korea report, dental caries and periodontal disease are among the top 10 outpatient diseases for which Koreans visit medical institution[1], and these easily preventable conditions are costly to both patients and the national healthcare system. Therefore, the government and dental professionals should work to develop a variety of intervention strategies to effectively prevent these diseases.

Dental caries and periodontal diseases are the main diseases that lead to tooth extraction, and their main cause is dental plaque, which contains more than 700 microbial species[2]. Dental plaque is a complex mass of microorganisms with high viscosity created when various oral bacteria get attached to salivary glycoproteins thinly attached to the tooth surface[3]. If dental plaque is not removed periodically, new bacteria will keep attaching to its surface, which makes the dental plaque more mature[4]. Eventually, mature dental plaque becomes the cause of demineralization of the tooth or inflammation of periodontal tissue[4]. Therefore, for the prevention of oral diseases, dental plaque accumulated on tooth surfaces should be thoroughly removed periodically.

The methods of controlling dental plaque are divided into mechanical methods such as tooth-brushing and chemical methods such as oral rinsing[5]. Although tooth-brushing is the most basic method to control dental plaque, many people do not use proper tooth-brushing

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technique and fail to effectively remove dental plaque[5]. Chemical methods are effective for removing interdental plaque that cannot be removed by tooth-brushing[6]. Hence, chemical methods are necessary for effective control of dental plaque.

Unfortunately, however, chlorhexidine gluconate, the most commonly used antimicrobial agent, can have side effects such as extrinsic dental staining or oral dryness, despite its effectiveness at inhibiting dental plaque formation[7,8]. In addition, when used constantly, its ability to inhibit dental plaque formation can diminish due to antibiotic resistance[9].

Recently, there has been increasing interest in essential oils extracted from herbs such as sweet basil as an alternative to chlorhexidine gluconate[10]. In particular, several studies have found that cinnamon oil has antibacterial effects on various bacteria, including *Staphylococcus aureus*[10,11]. Therefore, in this study, we analyze the antibacterial effects of cinnamon oil against *oral microorganisms* present in dental plaque using quantitative light-induced fluorescence–digital (QLF-D).

Method

This study was approved by the Ethics Committee of Gachon University (No. 1044396–201904-HR-057–01). An *in vitro* study was performed to evaluate the antimicrobial effects of cinnamon oil against *oral microorganisms*. All experimental processes in this study are based on previous studies[12-14].

First, cinnamon oil extracted from *Cinnamomum zeylanicum* via steam distillation was purchased from doTERRA (USA). Based on the results of a preliminary solubility test, the concentration of cinnamon oil to be used in the study was 2.5%. Tooth specimens (6 mm in diameter × 5 mm in thickness) were made using bovine incisors without cracks or white spots. Human stimulated saliva was collected from one healthy individual who had no active oral diseases and who had not used any antibiotics within the prior 3 months. The collected saliva was inoculated into each specimen, placed on a 24-well plate, and then incubated for 4 h (10% CO_2, 37°C). Then the saliva was aspirated from the wells, and a pre-made growth medium (0.5% sucrose and basal medium mucin) was added to the plate[12]. The plates were incubated for 6 days (10% CO_2, 37°C), with the growth medium replaced at the same time each day. During the 6 days, each specimen was treated with either 2.5% cinnamon oil (experimental group), 0.12% chlorhexidine gluconate (positive control group), or 10% dimethyl sulfoxide (negative control group) for 5 min, twice per day.

On day 7 of incubation, each specimen was photographed using QLF-D to assess microbial activity of dental plaque. QLF-D detects porphyrins produced by oral bacteria using 405 nm blue light. The blue light conditions were set as follows: shutter speed 1/30 s, aperture 5.0, and ISO 1,600[15]. The average red/green ratio (R/G values) of the images was calculated using ImageJ (NIH, MD, USA)[13]. A higher R/G values indicate high anaerobic bacterial activity.

A one-way analysis of variance (ANOVA) and the Scheffe *post hoc* test were performed to analyze differences in R/G values of dental plaque between the three treatment groups. A repeated measures ANOVA was conducted to compare the changes in the mean R/G values of dental plaque of each treatment group over time. A p-value < 0.05 was chosen to indicate a significant difference between groups.

Result and Discussion

R/G values were the lowest in the 2.5% cinnamon oil group (1.180 ± 0.010), and there was a significant difference between this group and the chlorhexidine gluconate group (1.249 ± 0.008), with the values of the 10% dimethyl sulfoxide group being highest (1.461 ± 0.003; $p < 0.001$) [Table 1]. The red fluorescence of each specimen is shown in Figure 1. A higher red fluorescence intensity indicates heightened activity of anaerobic bacteria. As a result of the repeated measures ANOVA, the changes in the mean R/G values of each specimen showed statistically significant differences over time ($p<0.05$) [Figure 2]. In particular, the R/G change was the smallest in the specimen which was treated with 2.5 cinnamon oil.

We used cinnamon oil extracted by steam distillation, because this method extracts the highest concentration of antimicrobial components[16], and the 2.5% cinnamon oil was found to have a greater antimicrobial effect against *oral microorganisms* than 0.12% chlorhexidine gluconate and 10% dimethyl sulfoxide. Our results suggest that cinnamon oil can be used to inhibit the formation of the pathogenic bacteria that cause dental diseases as effectively as chlorhexidine gluconate, which is commonly used in clinical practice.

As an increasing number of studies have confirmed the antimicrobial effects of cinnamon oil, in-depth research to amplify the antimicrobial activity of the oil has begun. One study reported that cinnamon oil's antimicrobial effect increases when combined with other essential oils, such as thyme oil[17]. Another recent study proposed the combination of cinnamon and antibiotics as an alternative therapeutic application that not only decreases the side effects of antibiotics but also enhances their curative effects on infectious diseases[18]. Although the antimicrobial effects of cinnamon oil against various microorganisms such as *S. aureus* have come to light, the mechanism behind them have not been fully elucidated. A few studies have suggested that cinnamaldehyde, a major component of cinnamon oil, causes microleakage of bacterial cell membranes[19,20]. Moreover, a study found that not only cinnamaldehyde but also eugenol, another component of cinnamon oil, has antibacterial activity[21]. Our study revealed, using QLF-D, that cinnamon oil inhibits the proliferation of anaerobic bacteria in dental plaque. Further research is needed to clarify the antimicrobial mechanism of cinnamon oil against pathogenic *oral microorganisms*. Furthermore, future studies should analyze and compare the antimicrobial activity of cinnamon oil against *oral microorganisms* according to concentration. Finally, it is necessary to search for other surfactants with high biocompatibility aside from dimethyl sulfoxide that can be used as a chemical solvent for cinnamon oil.

Table 1: R/G values according to treatment group (at 7 day)

Treatment group	R/G values	
	Mean ± SD	F(p)
2.5% Cinnamon oil	1.180 ± 0.010[a]	621.277 (<0.001)
0.12% Chlorhexidine gluconate	1.249 ± 0.008[b]	
10% Dimethyl sulfoxide	1.461 ± 0.003[c]	

[a,b,c]The different characters are significant by Scheffe's multiple comparison at α=0.05.

A: 2.5% cinnamon oil, B: 0.12% chlorhexidine gluconate, C: 10% dimethyl sulfoxide

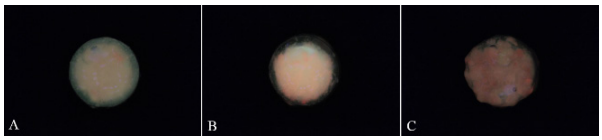

Figure 1. Red fluorescence of each specimen (at 7day)

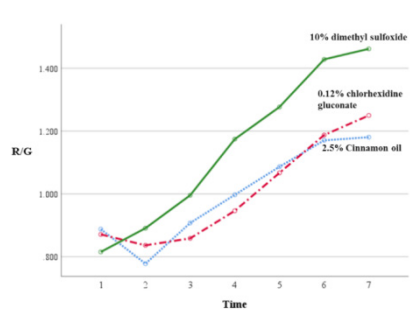

Figure 2. Changes in the mean R/G values of each specimen over time

Conclusion

Cinnamon oil is a natural substance that has a strong antibacterial effect against *oral microorganisms* without the side effects associated with chlorhexidine gluconate. This finding suggests that cinnamon oil has the potential to be developed as a preventive agent to prevent oral diseases. Further studies should analyze the antimicrobial effect of cinnamon oil against *oral microorganisms* at different concentrations. Furthermore, it is necessary to search for other surfactants with high biocompatibility aside from DMSO that can be used as a chemical solvent for cinnamon oil.

Ethical Clearance: Not required

Source of Funding: Self

Conflict of Interest: Nil

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