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Surface roughness of nanohybrid composite resin after exposure of basil leaf extract solution

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Abstract

Background: Dental caries is a chronic dental disease due to microbial metabolic activity, which causes tooth demineralization. Dental caries prevalence in Indonesia in 2018 was 45.3%. The prevalence value of dental caries can be reduced by restoring teeth and maintaining oral hygiene. One of the dental restoration materials is nanohybrid composite resin. Basil leaf extract solution (*Ocimum sanctum*) has the potential to be used as a mouthwash because it contains eugenol and polyphenols, which have antibacterial properties against *Streptococcus mutans* at concentrations of 2% and 4% to improve oral hygiene and prevent primary and secondary caries. Secondary caries occurs due to bacteria's attachment and penetration of restorative materials with a rough surface. The surface roughness of the composite increased after exposure to eugenol and polyphenols.

Purpose: To determine the effect of exposure to a solution of *Ocimum sanctum* leaf extract concentration of 2% and 4% on the surface roughness of nanohybrid composite resin.

Methods: Plant determination, phytochemical test, acidity test, immersion for five days, and surface roughness test.

Results: *Ocimum sanctum* leaf extract contains 0.21% eugenol and 5.11% polyphenols, and the extract solution has a pH of 4.7. The mean increase in surface roughness on the top and bottom surfaces of the nanohybrid composites after exposure to 2% and 4% *Ocimum sanctum* leaf extract solutions was significantly different from exposure to distilled water ($p<0.05$).

Conclusion: Exposure to *Ocimum sanctum* leaf extract solution increased the surface roughness of nanohybrid composite resin.

Keywords: Nanohybrid Composite Resin; *Ocimum sanctum*; Surface Roughness; Basil leaf extract solution

1. Introduction

Dental caries is a chronic dental disease due to microbial metabolic activity on the tooth surface for a specific time which causes tooth demineralization. Microbes in dental biofilm produce acidic metabolic waste. Metabolic waste acids demineralize enamel, dentine, and cementum resulting in carious lesions [1]. The prevalence of dental caries in Indonesia in 2018 was 45.3%, and the prevalence in East Java was 42.44%. The high prevalence of dental caries is not

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in line with the index of filled teeth. The caries index of filled teeth in East Java Province was only 3.82% [2]. The prevalence of dental caries can be reduced through prevention by improving oral hygiene and restoration treatment using dental restoration materials [3].

One of the natural ingredients for herbal medicine is basil (*Ocimum sanctum*). The contents of basil leaves identified include alkaloids, saponins, flavonoids, essential oils, and tannins. The content of essential oil in basil leaves is around 0.7%, with the main components being 71% eugenol and 20% methyl eugenol [4]. The potency of basil content can be used to improve oral hygiene and prevent dental caries [5]. Basil leaf extract has strong antibacterial properties. *Ocimum sanctum* leaf extract solution showed antibacterial properties against *Streptococcus mutans* at concentrations of 1%, 2%, and 4%, with an optimum concentration of 2% [6]. *Ocimum sanctum* leaf extract 4% solution showed maximum antimicrobial activity against dental biofilm microbes such as *Streptococcus mutans*, *Staphylococcus aureus*, and *Escherichia coli* [7].

Dental caries can be treated using a restorative material, namely composite resin. Many companies have made composite resins by combining micro-size resins mixed with other nano-sized fillers called nanohybrid resins [8]. Nanohybrid composite resin has a relatively small and delicate particle size of 0.04 µm. Nanohybrid composite resin has advantages such as easy polishing, high esthetics, low surface roughness, and high compressive strength so that this composite can be used for anterior and posterior teeth [9]

Basil leaf extract solution could be a mouthwash used to improve oral hygiene. However, its content contains active chemical compounds that can affect the physical properties of restorative materials. Research conducted by Chandra et al. proved that eugenol compounds could increase the surface roughness of nanohybrid composite resins [10]. Increased surface roughness of composites can lead to problems such as discoloration, plaque accumulation, gingival irritation, and secondary caries [11]. Secondary caries is caused by bacteria invading the microcracks between the surface of the restorative material and the prepared cavity [12]. According to in vitro and in vivo studies, the surface roughness of restorations has a top threshold value of 0.2 µm. This value aims to prevent the adhesion of bacteria that cause caries [13].

This study will examine the exposure of *Ocimum sanctum* leaf extract solutions with concentrations of 2% and 4%, which can potentially be used as mouthwashes on the surface roughness of nanohybrid composites. The results of the study will provide information to the public regarding the effect of exposure to the chemical properties of *Ocimum sanctum* leaf extract solution on the physical properties of the surface roughness of nanohybrid composite resins.

2. Material and methods

This research is considered experimental laboratory research with pre-test and post-test designs.

The sample used a nanohybrid composite resin type Filtek Z250XT Shade A2, 3M ESPE Dental product, St. Paul, MN, USA. The total sample is 21 and divided into three groups of 7 samples each. The group was divided into a control group immersed in distilled water, treatment group 1 immersed in *Ocimum sanctum* leaf extract 2% solution, and treatment group 2 immersed in *Ocimum sanctum* leaf extract 4% solution.

Nanohybrid composite resin was made in the form of tablets with a diameter of 10 mm and a thickness of 2 mm. Samples were made with a sample mold made of acrylic in the form of a plate containing wells measuring 10 mm in diameter and 2 mm in thickness which were exposed to LED light curing (I-Led DBA Woodpecker) for 20 seconds with blue light with a wavelength of 465 nm. The sample's surface must be flat, smooth, and shiny, so use celluloid strips at the top and bottom of the mold and give 1 kg of pressure. Then, the samples were stored in 300 mL distilled water for 24 hours at 37°C in an incubator to obtain post-setting polymerization [14].

The thick extract of *Ocimum sanctum* was obtained by maceration using 42 mL of 96% ethanol. The treatment solution was prepared by dissolving 0.4 mL and 0.8 mL of the extract using distilled water to a volume of 20 mL. Solutions of 2% and 4% *Ocimum sanctum* leaf extract were measured for their degree of acidity using a digital pH meter (Mettler Toledo pH Meters) [15].

The samples were measured for top and bottom surface roughness before treatment using a surface roughness tester (Mitutoyo SJ-201). Each sample is immersed in a 20 mL test solution in a 30 mL plastic jar. The sample is hung with sewing thread. Samples were soaked for 6 hours per day with 12 cycles every 30 minutes in an incubator with a temperature of 37°C. The soaking cycle was carried out with samples every 30 minutes removed from immersion, dried with tissue paper, then put into 20 ml of distilled water for 60 seconds and dried afterward. The sample is put back into the test solution and stored in the incubator for the next cycle. Samples were stored in distilled water in an incubator at 37°C for 18 hours. Immersion was repeated for five days using the test solution, which was renewed daily. The samples were measured for top and bottom surface roughness after treatment using a surface roughness tester (Mitutoyo SJ-201) [14].

The result data is in the form of surface roughness values measured using the Ra (Roughness) parameters, with micrometers (μm) units. Surface roughness measurement results were analyzed using One-Way ANOVA to see any significant differences. Tukey HSD test to compare the results between treatment groups. The degree of research confidence is 95%.

3. Results and discussion

Plant determination is an activity to identify plants to be used for research. Plant determination aims to avoid errors in the collection and use of the material to be studied. Determination and identification of the *Ocimum sanctum* plant were obtained from the Laboratory of Herbal Materia Medica. *Ocimum sanctum* has several common names in Indonesia, such as purple basil, purple basil, and lampes. The morphology of *Ocimum sanctum* leaf is oval, single, pointed tip, a serrated edge, pinnate spine, 14-16 mm long, 3-6 mm wide, stalk length ±1 cm, and green in color. The results of the determination of *Ocimum sanctum* plants have a special determination key in the form of code 1b -2b -3b -4b -6b -7b -9b -10b -11b -12b -13b -14a -16a -239b -243b -244b -248b -249b -250b -266b -267b -273b -276b -278b -279b -282a: Lamiceae-1a-2b-4b-6b-7b: Ocimum-8b: *O. sanctum*. *Ocimum sanctum* has the following taxonomic classification:

- Kingdom: Plantae
- Division: Angiospermae
- Class: Dicotyledonae
- Kelas: Asteridae
- Order: Lamiales
- Family: Lamiaceae / Labiatae
- Genus: Ocimum
- Species: *Ocimum sanctum* L.

The phytochemical content in the *Ocimum sanctum* viscous extract, which was extracted by the maceration method using 96% ethanol, is shown in table 1. The phytochemical test was carried out at the Industrial Research and Consulting Agency. *Ocimum sanctum* viscous extract was made in a concentrated solution of 2% and 4%, with a pH range of 4.65-4.77, which is classified as a weak acid.

Table 1 Phytochemical test results for active compounds in *Ocimum sanctum* leaf extract

No	Bioactive Compounds	%
1.	Eugenol	0.21
2.	Polyphenols	5.11
3.	Saponins	4.08
4.	Flavonoids	6.01
5.	Tannins	2.88
6.	Essential oils	2.26
7.	Alkaloids	4.32

Ocimum sanctum leaf extract contains essential oils, flavonoids, tannins, saponins, alkaloids, iron phosphorus, sulfur, and vitamins A & C. The extracts have antibacterial properties against bacteria that cause caries and plaque formation, especially *Streptococcus mutans*. Basil essential oil contains eugenol and polyphenols. Eugenol and polyphenols are phenolic compounds that have an antiseptic effect and work by damaging cell membranes. This basil leaf extract can be formulated in solution form with 2% and 4% [7].

The results of this study focused on the increase in the surface roughness value of nanohybrid composites after being exposed to 2% and 4% *Ocimum sanctum* leaf extract solutions, as shown in Figure 1. The normality test results using the Shapiro-Wilk test showed that the data were normally distributed ($p>0.05$). Furthermore, the Lavene test homogeneity test showed a homogeneous diversity of data (p>0.05). Then, the One-Way ANOVA statistical test with p<0.05, which means that there was a significant difference in the surface roughness of the nanohybrid composite resin between treatment groups. Finally, the Post Hoc statistical test using the Tukey HSD test obtained a significance level between treatment groups which can be seen in table 2.

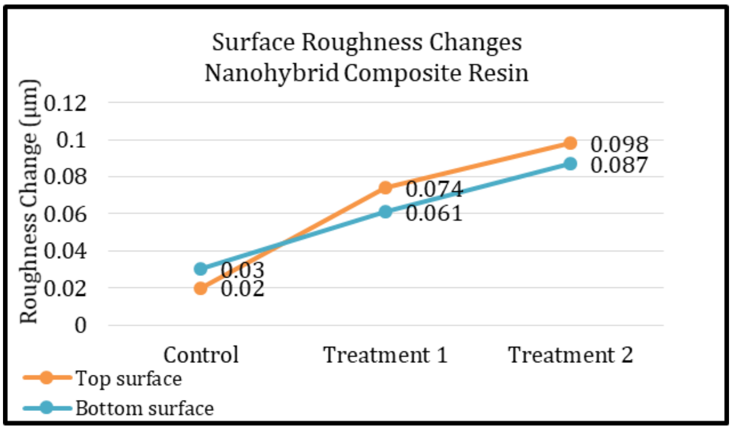

Figure 1 Graph of the average value of changes in the surface roughness of the nanohybrid composite after treatment

Changes in the average value of the top surface roughness of the nanohybrid composites, which were exposed to a direct light cure beam, and the bottom surface, which was not exposed to a direct light cure beam, had no different significant values. Changes in surface roughness could be seen from one of the control group's immersion results, which had a value of 0.02 µm on the top and 0.03 µm on the bottom. This result is because the sample thickness is only 2 mm, and the irradiation distance is <5 mm, which allows the light cure beam to optimally polymerize the nanohybrid composite resin throughout the sample mold from the top surface to the bottom surface [16].

The results showed that the change in the surface roughness value of the nanohybrid composite resin in the control group was not large with an estimated test time of 5 years of use *Ocimum sanctum* leaf extract solutions (one-minute daily use). The surface roughness change was only 0.020 µm on the top surface and 0.030 µm on the bottom. This result indicates that the absorption and degradation processes of the control group nanohybrid composites were low. The results of this study are consistent with the ability of the nanohybrid composite resin to prevent water absorption because it has a denser inorganic filler with a concentration of 68% and a composite matrix composed by a combination of Bis-EMA, UDMA, Bis-GMA, and TEGDMA. The combination of Bis-GMA and UDMA monomers will produce a crosslink bond that is strong, rigid, and durable. Bis-EMA, Bis-GMA, and TEGDMA monomers have low water absorption, which helps composite resins last longer [17].

Table 2 Post Hoc test results (Tukey HSD) changes in surface roughness of nanohybrid composite resin after treatment

Surface	Test group	Control	Treatment 1	Treatment 2
Top surface	Control	-	-	-
	Treatment 1	0.000*	-	-
	Treatment 2	0.000*	0.671	-
Bottom surface	Control	-	-	-
	Treatment 1	0.032*	-	-
	Treatment 2	0.000*	0.790	-

*p<0.05 (significantly different)

The results of the Post Hoc test (Tukey HSD) in Table 2 showed that the surface roughness of the top and bottom of the nanohybrid composite resin in the control group was significantly different from the treatment group 1 (p<0.05) and the treatment group 2 (p<0.05). This result shows that the treatment group has a more significant change in the roughness value of the nanohybrid composite resin than the control group. The comparison between the top and bottom surface roughness values of the nanohybrid composite resin did not differ significantly in treatment group 1 and treatment group 2 ($p>0.05$). This comparison shows that the surface roughness values of the two groups treated are relatively the same even though the concentration of the solution is different.

This study showed an increase in the mean surface roughness of the nanohybrid composites that were given treatments 1 and 2 with values of 0.074 µm and 0.098 µm on the top surface, these values were more significant than the control, which only increased by 0.02 µm on the top surface. The research results were analyzed using the One-Way ANOVA test and were significantly different ($p<0.05$). These results indicate an influence of the mildly acidic *Ocimum sanctum* extract solution on the surface roughness of the composite.

The surface roughness of the nanohybrid composites increased in this study due to the *Ocimum sanctum* leaf extract solution, which is mildly acidic due to the presence of eugenol and polyphenol content so that the test solution has a pH of 4.65-4.77. Eugenol and polyphenols have mild acidic properties because they belong to the phenol compound group. *Ocimum sanctum* leaf extract solution has a charge value because it releases H^+ ions from hydroxyl groups (OH^- ions). H^+ ions in acidic compounds are dominant, so their effect on the environment is more dominant than OH^- ions. Free H^+ ions can react with carbon double bonds (C=) in the nanohybrid composite resin polymer matrix to produce broken carbon double bonds. The carbon double bonds in the matrix broken in large quantities cause a degradation process of the matrix [10]. The matrix degradation process makes the inorganic filler particles on the surface of the composite easily detached. Loose inorganic filler on the composite surface makes small cracks in the composite so that the composite surface roughness value increases [18].

Free OH^- ions in *Ocimum sanctum* leaf extract solution can enter and affect the bonding of matrix polymers (Bis-GMA, UDMA, TEGDMA and Bis-EMA6) in chains composed of oxygen elements. The results of the OH^- reaction into the matrix make the siloxane bond chains in the matrix break and form silanol (Si-OH) and silicon oxide (Si-O) compounds. Si-O experiences electron disorientation so that Si-O can react when it meets water to produce products in the form of Si-OH and OH- compounds. The produced OH^- ions make it possible to carry out the reaction with the composite polymer matrix bonds again so that they can continuously break the siloxane bonds to form a cycle. This cycle will not stop if the nanohybrid composite resin is immersed in a liquid. The process of this cycle that supports the degradation of inorganic fillers and polymer matrices in nanohybrid composite resins causes a change in the surface of the composite, this change in the surface of the composite results in an increase in surface roughness [10].

The difference in solution concentration in treatment 1 and treatment 2 to changes in the mean value of the composite surface roughness was not significant on the top and bottom surfaces according to the Tukey HSD test analysis ($p=0.671$ and $p=0.790$). This result is because the small difference in concentration did not change the pH value of the solution significantly, the average pH of the treatment solution 1 was 4.72 and the treatment solution 2 was 4.68. This result indicates that the small difference in solution concentration is not the main factor that increases the surface roughness of the nanohybrid composite resin but the solution's acidity.

4. Conclusion

The results of this study can be concluded that exposure to a solution of basil leaf extract (*Ocimum sanctum*) concentrations of 2% and 4% proved to increase the surface roughness of the nanohybrid composite resin to greater than exposure to distilled water.

Compliance with ethical standards

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Disclosure of conflict of interest

All authors declare that they have no conflicts of interest.

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