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The Effect of Horn Beetle Nano Chitosan (*Xylotrupes gideon*) on the Surface Roughness of Glass-Ionomer Cement

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Keywords: Glass ionomer cement, nano chitosan horn beetle, surface roughness

Abstract. The purpose of this study was to evaluate the effect of adding horn beetle nano chitosan (NCH) to Glass Ionomer Cement (GIC) liquid on surface roughness. The evaluation was done before and after the addition of NCH and from the length of time soaking in artificial saliva. The disc sample with a diameter of 6 mm and a height of 3 mm of total 40 samples were divided into four groups: the GIC control group, GIC modified NCH 0.5%, 1%, and 2%. Each sample was immersed for 24 hours and seven days in artificial saliva. Surface roughness before and after immersion was measured using the Surtronic S-100 Series Surface Roughness Tester. Data analysis was performed using a one-way ANOVA test to determine the effect of adding horn beetle NCH and paired *t*-test to determine the effect of immersion time on the surface roughness of GIC. Although the highest surface roughness values were found in sample GIC modified NCH 1% (2.51±0.86 for 24 hours) and in sample GIC modified NCH 2% (2.64±0.84 for 7 days), there was no significant differences for both the surface roughness with the addition of NCH (p > 0.05) and the length of immersed time (p > 0.05). As the conclusion, there is no effect of horn beetle nano chitosan to GIC surface roughness. However, the addition of horn beetle NCH presented rougher surfaces after immersion. These experiments can help predict the performance of these materials under clinical conditions.

Introduction

Glass Ionomer Cement (GIC) is a tooth-colored restorative material used in dentistry. GIC is used widely because of its ability to release fluoride which prevents caries, biocompatible, and it has similar thermal expansion with tooth structure. On the other hand, GIC is also brittle because it has low acid resistance so it's easy to erode, dissolves, and cause surface roughness, high surface tension between components and weak adhesion between components so that the mechanical properties of GIC are relatively poor [1].

Surface roughness is an irregular surface characteristic of restorative materials and can be a retention of plaque accumulation to trigger caries. Glass Ionomer Cement is a restorative material that has a fairly high surface roughness value. In its development, the weakness of GIC filling material was improved by adding materials that could increase the GIC resistance. Materials added to improve the weaknesses of GIC are in the form of metals and resins [2]. Other materials developed to increase GIC resistance are chitosan natural materials [3].

Chitosan is a natural polymer which is very abundant in nature. Chitosan as a natural polymer has good characteristics, such as biodegradability, non-toxicity, adsorption, and antimicrobial properties because it can inhibit pathogenic bacteria and spoilage microorganisms including fungi, gram-

positive bacteria, and gram-negative bacteria. Chitosan can be made from insects and crustacean animal such as crab and shrimp. The advantage of chitosan natural ingredients compared to other materials is that chitosan is a natural material that is easily obtained, has economic value, and its activities can be applied in the fields of fisheries, agriculture, industrial environment, beauty, pharmacy, health, and food. Chitosan mixed in GIC manipulation can reduce GIC surface tension. This occurs when powder mixes with liquid GIC, chitosan's particle will be involved in increasing acid-base reactions with liquid GIC so that more salt bridges and cross-linking are formed. After reacting, the chitosan's particle will be adsorbed on the GIC matrix and fill between the glass particles. In addition, it can also increase GIC resistance to erosion due to acidic solutions, thereby reducing GIC surface roughness values [4].

Indonesia is a country known for its abundant marine products. As a food commodity that has economic value, so far, crustaceans are only used for their meat as a mixture for making crackers, shrimp paste or animal feed [5]. In addition to shrimp and crabs, insects can be also used as chitosan. Insect chitosan is derived from the head, skin or carapace. Insects are the largest group of animals, which is about 75% of the total number of living things. Some insects are beneficial but many are very harmful because they damage crops and spread disease to humans and livestock. One of the insects that is found widely in Indonesia (Bogor, Jawa Barat) and harms humans is horn beetle (*Xylotrupes gideon*) which mostly damages the shoots of coconut plants [5-6].

Chitosan modification has been done chemically and physically. Physical modification of chitosan is carried out to expand its utilization by changing the particle size of chitosan into nano chitosan. Chitosan nanoparticles can increase their ability to form bonds with other chemical substances. The development of chitosan into nano-forms can also improve the function of chitosan, one of which is inhibiting enamel demineralization [7].

Glass Ionomer Cement has a stable compressive strength at 24 hours of hardening. Based on previous research conducted by Nicholson, it was proven that the GIC experienced the most effective maturation process in compressive strength at a time duration of 7 days was 199.4 ± 12.0 MPa. In addition, this study stated that the compressive strength value of GIC at 7 days was 24.8 ± 14.3 MPa with the addition of acetic acid 45% in GIC [8]. Therefore, this study will be different from other research studies on modification of GIC with other chitosan, namely the manufacture of modified GIC samples of nano chitosan *Xylotrupes gideon* without the addition of acetic acid solvent.

Materials and Methods

The research was conducted at the DMT CORE Laboratory, Faculty of Dentistry, Universitas Trisakti. Chitosan as raw material medicine formulations derived from demineralization, deproteinization, decolorization, deacetylation of exoskeletons *Xylotrupes gideon*, which has undergone physical modifications become nano chitosan [5]. Preliminary research was carried out in accordance with the methods listed. Preparation of GIC (GC Fuji Type IX, GC Corporation, Tokyo, Japan) without the addition of NCH and GIC with the addition of NCH were carried out. First, GIC was prepared without the addition of nano chitosan as a control group (GIC Control). Then the GIC group was made with the addition of chitosan where chitosan was added to the liquid GIC in three different concentrations, namely 0.5 wt% (GIC+NCH 0.5%); 1wt% (GIC+NCH 1%); 2wt% (GIC+NCH 2wt%) were mixed with GIC powder and manipulated manually according to the manufacturer's recommendations [9].

GIC is manipulated according to the manufacturer's instructions and put under pressure into a mold with a thickness and diameter that has been made according to the specification standard. The cement is put into the mold with a plastic spatula and compacted with the help of a cement stopper. After completely filling the mold, the celluloid strip was pressed onto it with the help of a 2 kg weight for 5 minutes. After completion, the sample dimensions were measured using a digital caliper. Then, the samples were immersed in artificial saliva for 24 hours and 7 days and put in an incubator at 37°C, then the dimensions were measured using a digital caliper.

The total number of samples were 40 samples which were made in pairs for 24 hours and 7 days immersed time. Each sample was made using mold with diameter of 6 mm and thickness of 3 mm, in

accordance with the ISO 9917-1:2007 standard. The surface roughness test was carried out on 40 samples which were divided into eight groups, namely the 24 hours immersion group consisting of GIC Control, GIC+NCH 0.5 wt%, GIC+NCH 1 wt%, GIC+NCH 2 wt% groups and the 7 days immersion group consisting of from GIC Control, GIC+NCH 0.5%, GIC+NCH 1%, GIC+NCH 2% groups. The samples were tested using a Surface Roughness Tester. Surface roughness testing was carried out using a Surface Roughness Tester (*Surtronic S-100 Series*) with the same measurement area in each test. Measurement of surface roughness was done by placing the sample on a glass plate and the sample was fixed using double-sided tape so that its position remains stable during measurement. Then the Surface Roughness Tester detector was placed on the specimen at an angle of 90° and the stylus was placed on the marked end of the specimen.

Analysis of the mean of each group was performed using SPSS software. The average value of each test group was subjected to one-way ANOVA statistical test. For data analysis based on the immersion time, a paired t-test would be carried out.

Results

Fig. 1 showed there were no statistically significance differences of GIC control group and GIC modification nano chitosan group (one-way ANOVA, p > 0.05) and no differences of 24 hours and 7 days immersed time (paired t-test, p > 0.05). Normality test using the Kolmogorv-Smirnov test showed that the data of surface roughness test was 0.200 (p > 0.05). The homogeneity test of surface roughness data was 0.152 (p > 0.05). These results showed that the data of surface roughness test normally distributed and homogeneous.

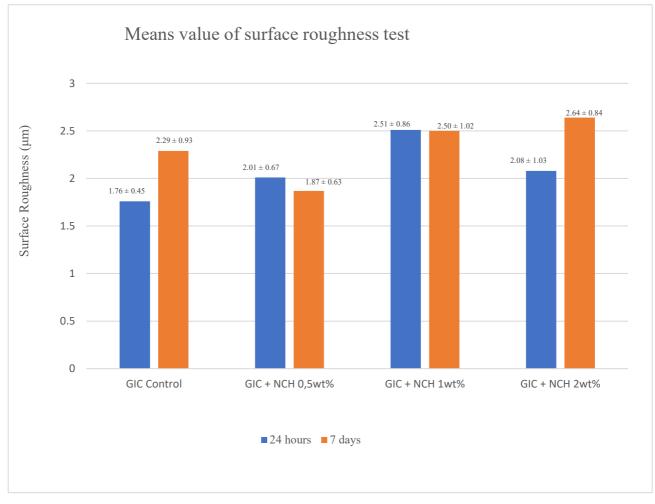


Figure 1. Surface roughness value of GIC Control, GIC+NCH 0.5 wt%, GIC+NCH 1 wt%, GIC+NCH 2 wt% with 24 hours and 7 days immersion time (p > 0.05, n=10)

Discussion

Glass ionomer cement is a restorative material that is widely used by dentists because of its advantages such as minimal preparation, fluoride release, good flexural strength and biocompatible with tissues. Disadvantages of GIC are brittle and easily eroded, causing increase in surface roughness. Surface roughness in restorations can cause discoloration of fillings and accumulation of dental plaque which if left untreated can result in tooth decay and supporting tissue in the long term. In addition, the increase in surface roughness also causes susceptibility to microbial colonization which can increase the risk of occurrence of oral disease and indications of deterioration of restorative materials [10].

Glass ionomer cement is a restorative material that will experience changes in surface roughness in long-term use due to contamination from saliva and acid [9]. The surface roughness could make interaction with bacterial adhesion that still physico-chemically happen from certain distance by the attractive forces of van der Waal's bonding and also the forces of electrostatic repulsive [11]. Parameter value for measuring surface roughness used is the average diameter roughness (Ra). According to Bollen et al. [11], the critical surface roughness value for bacterial colonization of restorative materials is $0.2 \mu m$, if the surface roughness value is above $0.2 \mu m$, it has the potential to increase the attachment of bacteria and dental plaque which can increase the risk of caries. which can control the surface roughness of the restorative material such as the size, shape, and homogeneity of the particles [12]. The larger of the particle size of the materials, the higher results of surface roughness.

According to Bala O et al. [13], a small particle size will produce a lower surface roughness compared to a larger particle size. The small particles as in nano measurement give a better ability in improving physicochemical properties that already offers advantages in such various applications [5]. It was stated in the previous study from Soygun et.al [14] that CH-modified GIC was promising candidate fort he future. This GIC modification study with horn beetle chitosan was not polished to determine the actual surface roughness value of the restorative materials. Therefore, it is possible that the surface roughness value was quite high both on immersion in artificial saliva for 24 hours and 7 days. There is no significant difference in the statistical analysis test (p > 0.05).

Study conducted by Tjandrawinata R et al. [10] determined the GIC group that received the coating had a greater hardness value that was statistically significantly higher (t-test, p < 0.05) than the GIC group with no coating. The hardness may be influence the surface roughness of GIC. To improve the physical properties of the GIC, polishing and the application of coating added to the restoration surface can be carried out.

Soygun et al. [14] examined the addition of 5% and 10% CH to GIC liquid did not increase the surface roughness of GIC. In a study by Silva and Zuanon [15] which evaluated surface roughness values after hardening of four different traditional GICs used in Atraumatic Restorative Treatment (ART), it was reported that with the exception of the Vitro molar GIC group, all the other groups had roughness values below the clinically accepted Ra value determined by Bollen et al [11]. The mixture between CH and GIC could happen from hydroxyl and acetamide group of CH that form hydrogen bonds with powder's hydroxyl groups and polyacrylic acid's carboxylic acid group. The interfacial tension could be reduce and later it could improve the mechanical properties of GIC. The lowest Ra values of GIC could lead mixture to become homogenous from their chemical bonding [16]. Sharafeddin, et.al. stated that the CH solution is a water-soluble cationic polyelectrolyte since the amine groups are protonated and charged positively. It can also be called as strong base due to its amino group, which the NH₂ group of CH and the functional group of the GIC (OH group and C = O group) could interact as their reaction mechanism. If CH concentration is increased then its molecules could react with each other instead of reacting with the GIC components [16].

Conclusion

In this in-vitro study, GIC modified nano chitosan was produced with the addition of nano chitosan horn beetle (*Xylotrupes gideon*) to Glass Ionomer Cement (GIC). The surface roughness results of GIC in artificial saliva of this study are presented that there were no statistically significance differences between control group and modification group. These materials under clinical conditions. A protective action against the carious process with significant surface damage due to erosion may be expected.

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The Effect of Horn Beetle Nano Chitosan (*Xylotrupes gideon*) on the Surface Roughness of Glass-lonomer Cement

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Keywords: Glass ionomer cement, nano chitosan horn beetle, surface roughness

Abstract. The purpose of this study was to evaluate the effect of adding horn beetle nano chitosan (NCH) to Glass Ionomer Cement (GIC) liquid on surface roughness. The evaluation was done before and after the addition of NCH and from the length of time soaking in artificial saliva. The disc sample with a diameter of 6 mm and a height of 3 mm of total 40 samples were divided into four groups: the GIC control group, GIC modified NCH 0.5%, 1%, and 2%. Each sample was immersed for 24 hours and seven days in artificial saliva. Surface roughness before and after immersion was measured using the Surtronic S-100 Series Surface Roughness Tester. Data analysis was performed using a one-way ANOVA test to determine the effect of adding horn beetle NCH and paired *t*-test to determine the effect of immersion time on the surface roughness of GIC. Although the highest surface roughness values were found in sample GIC modified NCH 1% (2.51±0.86 for 24 hours) and in sample GIC modified NCH 2% (2.64±0.84 for 7 days), there was no significant differences for both the surface roughness with the addition of NCH (p > 0.05) and the length of immersed time (p > 0.05). As the conclusion, there is no effect of horn beetle nano chitosan to GIC surface roughness. However, the addition of horn beetle NCH presented rougher surfaces after immersion. These experiments can help predict the performance of these materials under clinical conditions.

Introduction

Glass Ionomer Cement (GIC) is a tooth-colored restorative material used in dentistry. GIC is used widely because of its ability to release fluoride which prevents caries, biocompatible, and it has similar thermal expansion with tooth structure. On the other hand, GIC is also brittle because it has low acid resistance so it's easy to erode, dissolves, and cause surface roughness, high surface tension between components and weak adhesion between components so that the mechanical properties of GIC are relatively poor [1].

Surface roughness is an irregular surface characteristic of restorative materials and can be a retention of plaque accumulation to trigger caries. Glass Ionomer Cement is a restorative material that has a fairly high surface roughness value. In its development, the weakness of GIC filling material was improved by adding materials that could increase the GIC resistance. Materials added to improve the weaknesses of GIC are in the form of metals and resins [2]. Other materials developed to increase GIC resistance are chitosan natural materials [3].

Chitosan is a natural polymer which is very abundant in nature. Chitosan as a natural polymer has good characteristics, such as biodegradability, non-toxicity, adsorption, and antimicrobial properties because it can inhibit pathogenic bacteria and spoilage microorganisms including fungi, gram-

All rights reserved. No part of contents of this paper may be reproduced or transmitted in any form or by any means without the written permission of Trans Tech Publications Ltd, www.scientific.net. (#597854856-27/08/22,10:58:57) positive bacteria, and gram-negative bacteria. Chitosan can be made from insects and crustacean animal such as crab and shrimp. The advantage of chitosan natural ingredients compared to other materials is that chitosan is a natural material that is easily obtained, has economic value, and its activities can be applied in the fields of fisheries, agriculture, industrial environment, beauty, pharmacy, health, and food. Chitosan mixed in GIC manipulation can reduce GIC surface tension. This occurs when powder mixes with liquid GIC, chitosan's particle will be involved in increasing acid-base reactions with liquid GIC so that more salt bridges and cross-linking are formed. After reacting, the chitosan's particle will be adsorbed on the GIC matrix and fill between the glass particles. In addition, it can also increase GIC resistance to erosion due to acidic solutions, thereby reducing GIC surface roughness values [4].

Indonesia is a country known for its abundant marine products. As a food commodity that has economic value, so far, crustaceans are only used for their meat as a mixture for making crackers, shrimp paste or animal feed [5]. In addition to shrimp and crabs, insects can be also used as chitosan. Insect chitosan is derived from the head, skin or carapace. Insects are the largest group of animals, which is about 75% of the total number of living things. Some insects are beneficial but many are very harmful because they damage crops and spread disease to humans and livestock. One of the insects that is found widely in Indonesia (Bogor, Jawa Barat) and harms humans is horn beetle (*Xylotrupes gideon*) which mostly damages the shoots of coconut plants [5-6].

Chitosan modification has been done chemically and physically. Physical modification of chitosan is carried out to expand its utilization by changing the particle size of chitosan into nano chitosan. Chitosan nanoparticles can increase their ability to form bonds with other chemical substances. The development of chitosan into nano-forms can also improve the function of chitosan, one of which is inhibiting enamel demineralization [7].

Glass Ionomer Cement has a stable compressive strength at 24 hours of hardening. Based on previous research conducted by Nicholson, it was proven that the GIC experienced the most effective maturation process in compressive strength at a time duration of 7 days was 199.4 ± 12.0 MPa. In addition, this study stated that the compressive strength value of GIC at 7 days was 24.8 ± 14.3 MPa with the addition of acetic acid 45% in GIC [8]. Therefore, this study will be different from other research studies on modification of GIC with other chitosan, namely the manufacture of modified GIC samples of nano chitosan *Xylotrupes gideon* without the addition of acetic acid solvent.

Materials and Methods

The research was conducted at the DMT CORE Laboratory, Faculty of Dentistry, Universitas Trisakti. Chitosan as raw material medicine formulations derived from demineralization, deproteinization, decolorization, deacetylation of exoskeletons *Xylotrupes gideon*, which has undergone physical modifications become nano chipsan [5]. Preliminary research was carried out in accordance with the methods listed. Preparation of GIC (GC Fuji Type IX, GC Corporation, Tokyo, Japan) without the addition of NCH and GIC with the addition of NCH were carried out. First, GIC was prepared without the addition of nano chitosan as a control group (GIC Control). Then the GIC group was made with the addition of chitosan ahere chitosan was added to the liquid GIC in three different concentrations, namely 0.5 wt% (GIC+NCH 0.5%); 1wt% (GIC+NCH 1%); 2wt% (GIC+NCH 2wt%) were mixed with GIC powder and manipulated manually according to the manufacturer's recommendations [9].

GIC is manipulated according to the manufacturer's instructions and put under pressure into a mold with a thickness and diameter that has been made according to the specification standard. The cement is put into the mold with a plastic spatula and compacted with the help of a cement stopper. After completely filling the mold, the celluloid strip was pressed onto it with the help of a 2 kg weight for 5 minutes. After completion, the sample dimensions were measured using a digital caliper. Then, the samples were immersed in artificial saliva for 24 hours and 7 days and put in an incubator at 37°C, then the dimensions were measured using a digital caliper.

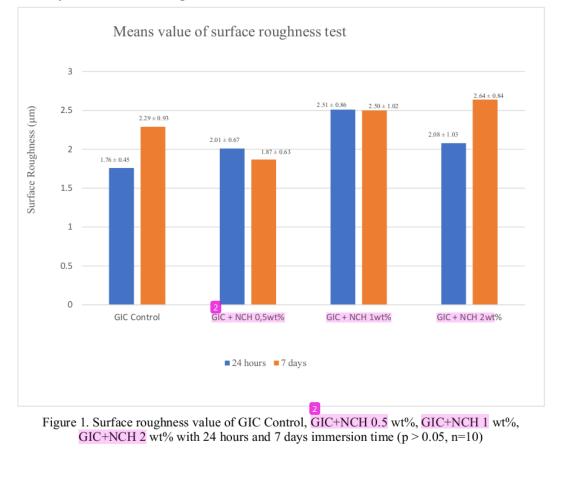
The total number of samples were 40 samples which were made in pairs for 24 hours and 7 days immersed time. Each sample was made using mold with diameter of 6 mm and thickness of 3 mm, in

accordance with the ISO 9917-1:2007 standard. The surface roughness test was carried out on 40 samples which vere divided into eight groups, namely the 24 hours immersion group consisting of GIC Control, GIC+NCH 0.5 wt%, GIC+NCH 1 at%, GIC+NCH 2 wt% groups and the 7 days immersion group consisting of from GIC Control, GIC+NCH 0.5%, GIC+NCH 1%, GIC+NCH 2% groups. The samples were tested using a Surface Roughness Tester. Surface roughness testing was carried out using a Surface Roughness Tester (*Surtronic S-100 Series*) with the same measurement area in each test. Measurement of surface roughness was done by placing the sample on a glass plate and the sample was fixed using double-sided tape so that its position remains stable during measurement. Then the Surface Roughness Tester detector was placed on the specimen at an angle of 90° and the stylus was placed on the marked end of the specimen.

Analysis of the mean of each group was performed using SPSS software. The average value of each test group was subjected to one-way ANOVA statistical test. For data analysis based on the immersion time, a paired t-test would be carried out.

Results

Fig. 1 showed there were no statistically significance differences of GIC control group and GIC modification nano chitosan group (one-way ANOVA, p > 0.05) and no differences of 24 hours and 7 days immersed time (paired t-test, p > 0.05). Normality test using the Kolmogorv-Smirnov test showed that the data of surface roughness test was 0.200 (p > 0.05). The homogeneity test of surface roughness data was 0.152 (p > 0.05). These results showed that the data of surface roughness test normally distributed and homogeneous.



Discussion

Glass ionomer cement is a restorative material that is widely used by dentists because of its advantages such as minimal preparation, fluoride release, good flexural strength and biocompatible with tissues. Disadvantages of GIC are brittle and easily eroded, causing increase in surface roughness. Surface roughness in restorations can cause discoloration of fillings and accumulation of dental plaque which if left untreated can result in tooth decay and supporting tissue in the long term. In addition, the increase in surface roughness also causes susceptibility to microbial colonization which can increase the risk of occurrence of oral disease and indications of deterioration of restorative materials [10].

Glass ionomer cement is a restorative material that will experience changes in surface roughness in long-term use due to contamination from saliva and acid [9]. The surface roughness could make interaction with bacterial adhesion that still physico-chemically happen from certain distance by the attractive forces of van der Waal's bonding and also the forces of electrostatic repulsive [11]. Parameter value for measuring surface roughness used is the average diameter roughness (Ra). According to Bollen et al. [11], the critical surface roughness value for bacterial colonization of restorative materials is $0.2 \mu m$, if the surface roughness value is above $0.2 \mu m$, it has the potential to increase the attachment of bacteria and dental plaque which can increase the risk of caries. which can control the surface roughness of the restorative material such as the size, shape, and homogeneity of the particles [12]. The larger of the particle size of the materials, the higher results of surface roughness.

According to Bala O et al. [13], a small particle size will produce a lower surface roughness compared to a larger particle size. The small particles as in nano measurement give a better ability in improving physicochemical properties that already offers advantages in such various applications [5]. It was stated in the previous study from Soygun et.al [14] that CH-modified GIC was promising candidate fort he future. This GIC modification study with horn beetle chitosan was not polished to determine the actual surface roughness value of the restorative ng terials. Therefore, it is possible that the surface roughness value was quite high both on immersion in artificial saliva for 24 hours and 7 days. There is no significant difference in the statistical analysis test (p > 0.05).

Study conducted by Tjandrawinata R et al. [10] determined the GIC group that received the coating had a greater hardness value that was statistically significantly higher (t-test, p < 0.05) than the GIC group with no coating. The hardness may be influence the surface roughness of GIC. To improve the physical properties of the GIC, polishing and the application of coating added to the restoration surface can be carried out.

Soygun et al. [14] examined the addition of 5% and 10% CH to GIC liquid did not increase the surface roughness of GIC. In a study by Silva and Zuanon [15] which evaluated surface roughness values after hardening of four different traditional GICs used in Atraumatic Restorative Treatment (ART), it was reported that with the exception of the Vitro molar GIC group, all the other groups had roughness values below the clinically accepted Ra value determined by Bollen et al [11]. The mixture between CH and GIC could happen from hydroxyl and acetamide group of CH that form hydrogen bonds with powder's hydroxyl groups and polyacrylic acid's carboxylic acid group. The interfacial tension could be reduce and later it could improve the mechanical properties of GIC. The lowest Ra values of GIC could lead mature to become homogenous from their chemical bonding [16]. Sharafeddin, et.al. stated that the CH solution is a water-soluble cationic polyelectrolyte since the amine groups are proported and charged positively. It can also be called as strong base due to its amino group, which the NH₂ group of CH and the functional group of the GIC (OH group and C = O group) could interact as the greaction mechanism. If CH concentration is increased then its molecules could react with each other instead of reacting with the GIC components [16].

Conclusion

In this in-vitro study, GIC modified nano chitosan was produced with the addition of nano chitosan horn beetle (*Xylotrupes gideon*) to Glass Ionomer Cement (GIC). The surface roughness results of GIC in artificial saliva of this study are presented that there were no statistically significance differences between control group and modification group. These materials under clinical conditions. A protective action against the carious process with significant surface damage due to erosion may be expected.

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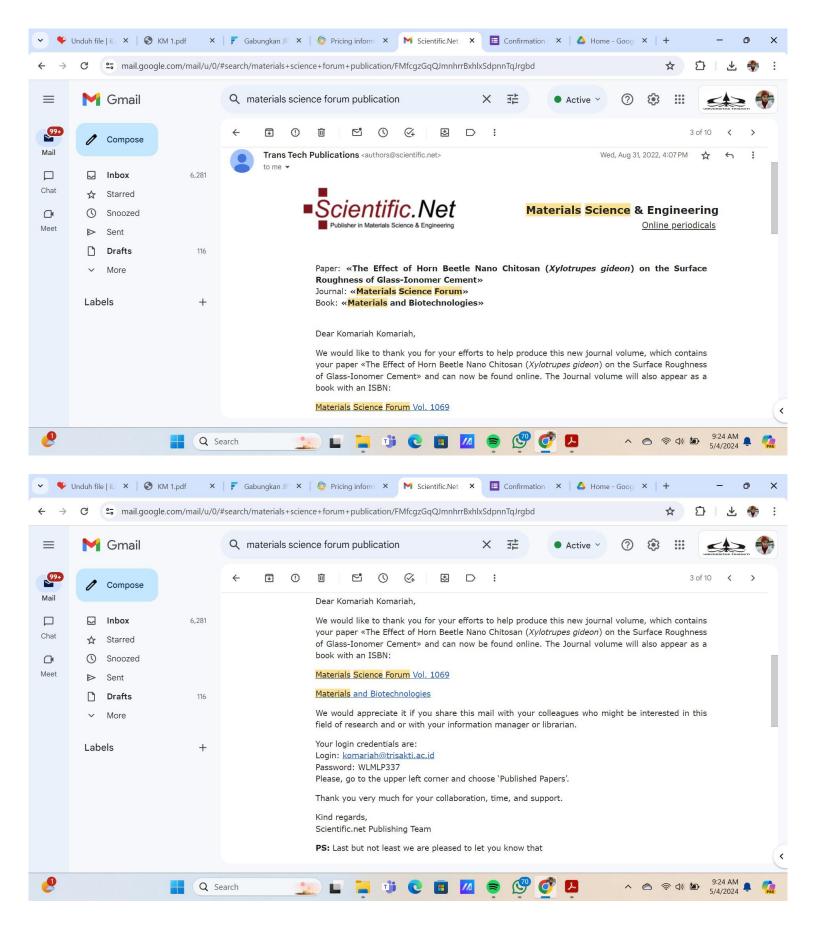
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