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Antimicrobial And Mechanical Properties of Glass Ionomer Cements Containing Different Antimicrobial Agents: An In Vitro Study

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KEYWORDS

Antibacterial properties; Minimum Inhibitory Concentration (MIC); Mechanical properties; Shear-peel band strength; Glass Ionomer Cement (GIC)

ABSTRACT:

Aim: To evaluate the antimicrobial and mechanical properties of Ethanolic extract of Propolis and Aloe Vera extract to Glass Ionomer cement.

Materials and Methods: Cement was divided into seven groups: one with original Composition, three with 10%, 20%, and 30% EEP and three with 10% , 20% and 30% Aloe Vera extract added to the liquid and then manipulated. Each group in antimicrobial assay had 20 specimens. One hundred and forty premolars banded with conventional GIC and GIC with different dilutions of Ethanol Extract of Propolis (EEP) and Aloe Vera extract were used for the mechanical assay with each group containing 20 teeth. Krushkal Wallis test was used to assess Shear peel Bond Strength (SPBS) and Minimum Inhibitory Concentration (MIC) values and ascertain whether the samples have same group source as an origin or not by matching the medians of the groups and pairwise comparison was done using Mann Whitney U test to assess entities in pairs to judge which pair has greater quantitative property. Level of statistical significance was set at 0.05.

Results: GIC with 30% EEP was the most effective in inhibition of Streptococcus mutans growth as compared to all the groups. It showed a median of 800 colony forming units as compared to conventional GIC which showed a staggering median of 74000 colony forming units. No significant difference was seen in terms of Shear peel bond Strength (SPBS) , ($P > .05$). The mean for maximum load necessary to deband was recorded for conventional GIC was found to be 1.4615 megapascals and that of GIC containing 30% ethanolic extract of propolis (which had the least MIC value and colony count, thus exhibiting highest antibacterial property) was

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1.654 megapascals.

Conclusions: Addition of EEP and Aloe Vera extract increased antibacterial properties without negatively modifying the mechanical properties of conventional GIC.

I. INTRODUCTION

During the orthodontic treatment, the plaque accumulation primarily around brackets and at the cervical margins of the bands leads to enamel demineralization, caries and hyperplastic gingivitis in these areas due to patient's difficulty in maintaining oral hygiene.[1]

Glass ionomer cements (GICs) are the most commonly used material for band cementation. This material exhibits a continuous release of fluoride, which creates antimicrobial effect[2-4] against a small spectrum of microorganisms with a low bactericide potential. Therefore, GIC may not prevent plaque proliferation and development of caries and periodontal disease in some patients.[3-5] Caries is a disease usually associated with Streptococcus mutans. Hence, it is important to evaluate the interaction of orthodontic cements with these bacteria.

Since the antimicrobial property of Glass Ionomer cement is not adequate to stop plaque proliferation around the cemented band, thus incorporation of antimicrobial agents in Glass Ionomer cement and its banding in Orthodontic treatment should be studied as it would help in preventing periodontal diseases by decreasing plaque proliferation.

Propolis, a natural substance produced by honeybees, which has been widely consumed in the folk medicine since ancient times, seems to be a promising ingredient of topical formulations due to its multidirectional biological properties.[6] Apart from antibacterial activity, various studies have demonstrated that propolis has other beneficial properties, such as antioxidative, antifungal, antiviral, and anti-inflammatory ones.[7,8] Additionally, antiproliferative action in human tumor cell lines has been observed.[9-12]

It is confirmed[13-15] that ethanolic extracts of propolis (EEP) demonstrates antimicrobial activity against Grampositive cocci of *Streptococcusmutans*, a facultative anaerobic bacterium commonly found in human oral cavity (saliva and dental plaque) and a main contributor to tooth decay caused by biofilm formation.

Aloe vera gel has shown inhibitory effects on certain bacteria such as Streptococcus pyogenes and Enterococcus faecalis. Although several studies have demonstrated Propolis's and Aloe Vera's variable activity against different bacteria and its antibacterial activity on oral microorganisms, very few researchers have studied its effect on the mechanical properties of oral appliances.

This study was undertaken to evaluate the antimicrobial and mechanical properties of GIC when mixed with different concentrations of ethanolic extracts of propolis(EEP) and aloe vera extract. The antimicrobial properties of GIC containing different concentrations of EEP and aloe vera extract were evaluated by establishing the absolute colony count of S.mutans and the Minimum Inhibitory Concentration(MIC).Minimum Inhibitory Concentration(MIC) is the lowest concentration of chemical which prevents visible growth of bacterium.

For Mechanical properties, banding of extracted premolars, for orthodontic purpose, was done with GIC containing different concentrations of EEP and aloe vera extract. Stainless steel orthodontic premolar bands with attachments on the proximal sides were fitted and seated around the teeth, including adaptation of the margins with a band seater. An Instron testing machine was used for measuring the shear peel bond strength (SPBS) at a crosshead speed of 1 mm/min by recording the maximum load necessary to deband.

II. MATERIALS AND METHODS

For Antimicrobial Assay Fabrication of Glass Ionomer Cement specimens:

A total number of 140 specimens were fabricated. Test specimens were divided into 7 groups based on the

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concentration of Ethanolic Extract of Propolis and Aloe Vera in Glass ionomer cement.

Fabrication of Glass Ionomer Cement with different dilutions of EEP and Aloe Vera Extract specimens:

For 10% dilution, 0.3 ml of EEP or Aloe Vera Extract was mixed with 1 drop of liquid of GIC ,than this liquid was than mixed with 1 unit powder of GIC. So, for 20% dilution 0.6 ml of both antimicrobial agents were used and 0.9 ml for 30% dilution.

Group 1 - Conventional GIC

Group 2 - GIC containing 10% ethanolic extract of propolis

Group 3 - GIC containing 20% ethanolic extract of propolis

Group 4 - GIC containing 30% ethanolic extract of propolis

Group 5 - GIC containing 10% of aloe vera extract

Group 6 - GIC containing 20% of aloe vera extract

Group 7 - GIC containing 30% of aloe vera extract

Each group comprised of 20 specimens. After processing using standardized technique, the specimens were stored in distilled water until the experiments took place.

Minimum Inhibitory Concentration

The turbidity of S.mutans inoculum was adjusted to 0.5 Macfarland standards by using Brain Heart Infusion broth. Microtitre plate was labeled according to the groups and filled with Brain Heart Infusion broth containing S.mutans by using a sterile dropper (Fig 1). All the samples from their respective groups were placed in the labeled wells by using a forcep and microtitre plate was incubated at 37 degree Celsius for 24 hours (Fig 2&3).After 24 hours broth was subcultured on to the blood agar using inoculating loop (Fig.4) and incubated at 37 degree Celsius for 24 hours. Colonies were counted and microbial growth of each group were compared after 24 hours.

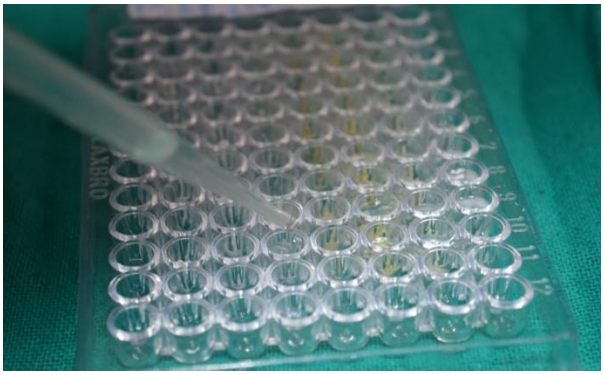

Fig. 1 - Microtitre plate filled with Brain Heart Infusion broth containing S.mutans by using a sterile dropper.

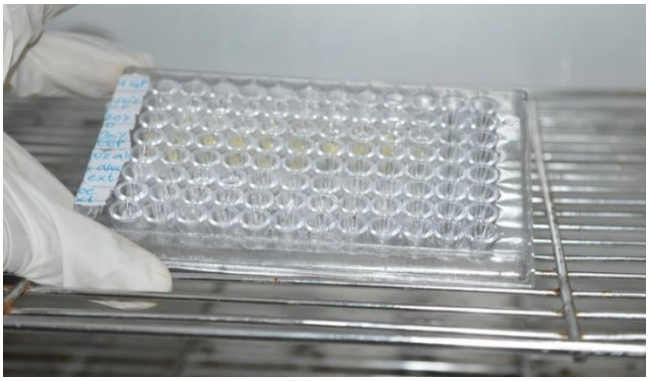

Fig. 2 - Samples from their respective groups were placed in the labeled wells by using a forcep and microtitre plate was incubated at 37 degree Celsius for 24 hours.

Fig. 3 - Microtitre plate incubated at 37 degree Celsius for 24 hours.

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Fig. 4 - Broth subcultured on to the blood agar using inoculating loop.

For mechanical Assay

140 human permanent premolars extracted for orthodontic reasons without any cracks, restorations, surface defects and caries were collected for the study. Hydrogen peroxide was the solution of choice for storage. After extraction, the teeth were stored for 1 week and kept in room temperature until the experiments take place. The sample were divided into seven groups for the mechanical assay. Each group had 20 premolars. The premolars were randomly divided into seven groups. These teeth were then placed in plastic model and filled with cold-cure acrylic resin. Stainless steel orthodontic premolar bands with buccal attachments were fitted and seated around the teeth, including adaptation of the margins with a band seater. Bands selected were placed by one researcher to eliminate any operator bias as far as band positioning and fitting were concerned. Each band was cemented in place with its specific cement. To avoid adverse influence on results excess cement was removed from the occlusal and cervical margins of the band.

The bands were cemented around the premolars using glass ionomer cement as follows:

Group 1 - Sample banded with Conventional GIC

Group 2 - Sample banded with GIC containing 10% ethanolic extract of propolis

Group 3 - Sample banded with GIC containing 20% ethanolic extract of propolis

Group 4 - Sample banded with GIC containing 30% ethanolic extract of propolis

Group 5 - Sample banded with GIC containing 10% of aloe vera extract

Group 6 - Sample banded with GIC containing 20% of aloe vera extract

Group 7 - Sample banded with GIC containing 30% of aloe vera extract

Each band was cemented in place with its specific cement prepared by mixing GIC powder with appropriate liquid as assigned to the different groups. All specimens were then stored in distilled water at 37 degree C for 24 hours before band retention was measured because maximum band retention is obtained 24 hours after cementation. An Instron testing machine was used for the shear peel bond strength (SPBS) test at a crosshead speed of 1 mm/min. The force was applied using a 0.9-mm diameter stainless steel wire from attachments on the buccal surface of the bands (Fig 5). The maximum load necessary to deband was recorded using the instron testing machine (Fig 6).

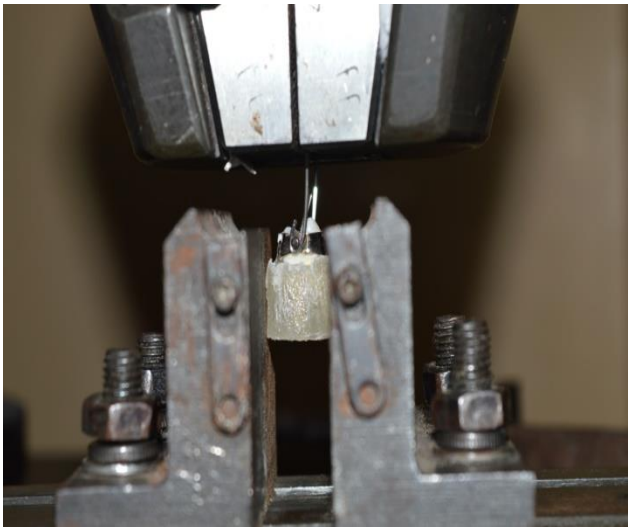

Fig 5 - The force was applied using a 0.9-mm diameter stainless steel wire from attachments on the buccal surface of the bands.

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Fig 6 - The maximum load necessary to deband was recorded using the instron testing machine.

Sample Size Estimation

Sample size estimation was done by using **GPower software (version 3.0).** Sample size was estimated for F test.

A minimum total sample size of 140 was found to be sufficient for an alpha of 0.05, power of 80 %, 0.32 as effect size (as assessed from similar study). Sample size was further divided as 20 in seven study groups.

F tests - ANOVA: Fixed effects, omnibus, one-way

Analysis: A priori: Compute required sample size

Input:

Effect size f	=	0.32
α err prob	=	0.05
Power (1-β err prob)	=	0.80
Number of groups	=	7

Output:

Noncentrality parameter λ	=	14.3360000
Critical F	=	2.1674232
Numerator df	=	6
Denominator df	=	133
Total sample size	=	140
Actual power	=	0.8018417

Statistical Analysis

The data were analysed using Statistical Package for Social Sciences(SPSS) version 21.All the variables were continuous which were summarized as mean, median and standard deviation. Shapiro Wilk test was used to check the normality of the data.Data was found to be normal. Krushkal Wallis test was used to assess Shear peel Bond Strength(SPBS) and Minimum Inhibitory Concentration(MIC) values and acertain weather the samples have same group source as an origin or not by matching the medians of the groups. Pairwise comparison was done using Mann Whitney U test to assess entities in pairs to judge which pair has greater quantitative property. Level of statistical significance was set at 0.05.

III. RESULTS:

The data was collected, compiled and statistically analyzed. Table 1 depicts median, IQR, range and MIC of colony forming units of microbial colony for all 7 groups. Median values of colony forming units of microbial colony are depicted in graph1.

Table 1 :- Microbial colony and MICs of Groups Against the S. Mutans Strain

Group	Median	IQR	Range		MIC
			Minimum	Maximum	
Conventional GIC	74000 cfu	18750 cfu	50000 cfu	100000 cfu	50000 cfu/ml
GIC containing 10% ethanolic extract of propolis	39000 cfu	19000 cfu	30000 cfu	49000 cfu	30000 cfu/ml

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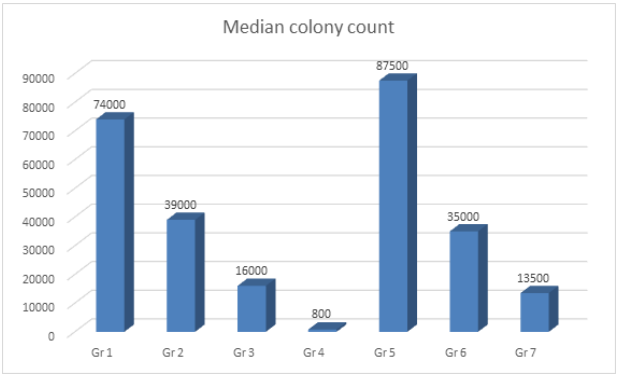
GIC containing 20% ethanolic extract of propolis	16000 cfu	12750 cfu	8000 cfu	29000 cfu	8000 cfu/ml
GIC containing 30% ethanolic extract of propolis	800 cfu	200 cfu	500 cfu	1000 cfu	500 cfu/ml
GIC containing 10% of aloe vera extract	87500 cfu	17500 cfu	60000 cfu	100000 cfu	60000 cfu/ml
GIC containing 20% of aloe vera extract	35000 cfu	20000 cfu	20000 cfu	59000 cfu	20000 cfu/ml
GIC containing 30% of aloe vera extract	13500 cfu	4750 cfu	10000 cfu	19000 cfu	10000 cfu/ml
P^a value	<0.001, S				
P^b of post hoc pairwise comparison	4<3,7<2,6<1<5				

[a]Kruskal Wallis test, [b]Mann Whitney U test

P value was found to be statistically significant since it was less than 0.05

Note:- Median, minimum and maximum range are in cfu (colony forming unit)



Graph 1:- Bar Graph depicting the median colony count of the seven groups containing the S.Mutans strain

The turbidity of S.mutans inoculum was adjusted to 0.5 Macfarland (10^5 cfu/ml) standard by using Brain Heart Infusion broth. Microtitre plates were labeled according to the groups and filled with Brain Heart Infusion broth containing S.mutans by using a sterile dropper. All the samples from their respective groups were placed in the labeled wells by using a forcep and microtitre plate was incubated at 37 degree Celsius for 24 hours.

After 24 hours broth containing the samples were subcultured on to the blood agar using inoculating loop and incubated at 37 degree Celsius for 24 hours.

After 24 hours colonies were counted and microbial growth of each group was compared.

Growth of the the gram positive bacteria was found to be the least in GIC containing 30% ethanolic extract of propolis as compared to the rest of the groups.

The MIC value of conventional GIC was found to be very high as compared to the rest of the groups(Table1).

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Table 2 depicts the comparison of all the groups for antimicrobial assay.

Statistically significant difference was found in the P values of the each groups by applying Kruskal Wallis test(Table 2).

Post hoc pair comparison was done using Mann Whitney U test.

Table 2 :- P Values of all the 7 groups of the Antimicrobial Assay

	Gr 2	Gr 3	Gr 4	Gr 5	Gr 6	Gr 7
Gr 1	<0.001	<0.001	<0.001	0.018	<0.001	<0.001
Gr 2	-	<0.001	<0.001	<0.001	0.398	<0.001
Gr 3	<0.001	-	<0.001	<0.001	<0.001	0.183
Gr 4	<0.001	<0.001	-	<0.001	<0.001	<0.001
Gr 5	<0.001	<0.001	<0.001	-	<0.001	<0.001
Gr 6	0.398	<0.001	<0.001	<0.001	-	<0.001

Following was concluded from the post hoc pair comparision

Gp 4<Gp3,Gp7<Gp2,Gp6<Gp1<Gp5

The Antibacterial effect of propolis has a significant P value of 0.001 against all the groups.

Despite giving a reddish tinge and a slight odour to GIC,the antibacterial effect of 30% EEP were significantly higher than the rest.

Even as compared to GIC containing 30% of aloe vera extract,the MIC value of GIC containing 30% ethanolic extract of propolis is much less i.e 500 cfu/ml(Table 1). 30% ethanolic extract of propolis is statistically more significant than 30% of aloe vera extract(Table 2).

GIC containing 10% and 20% aloe vera had a significantly high colony count(Graph 1) and thus lacked high antibacterial activity like propolis.It was also statistically less potent than propolis.

30% ethanolic extract of propolis had the least colony count with median of 800 cfu/ml as compared to 13500 cfu/ml of GIC containing 30% of aloe vera extract(Graph 1). 30% ethanolic extract of propolis was found to more statistically significant than 30% of aloe vera extract.

The control group containing conventional GIC showed very high MIC value i.e 50000 cfu/ml as compared with the rest of the groups.

The antibacterial property of conventional GIC was very low whereas the antibacterial property of GIC was increased by adding different concentrations of propolis extract and Aloe Vera extract.

The MIC value of the GIC containing 30% ethanolic extract of propolis was the least with 500 cfu/ml when compared with the rest of the six groups.The median of the antimicrobial colonies of the group containing GIC with 30% ethanolic extract of propolis was 800 cfu/ml with 200 Interquartile rangle,according to Kruskal Wallis test (Table 1)

Out of both the 10% dilutions of the antibacterial agents, EEP was found to be more potent than Aloe Vera extract. Even in 20% and 30% dilutions, EEP was found to be more potent than Aloe vera extract. EEP resulted in fewer bacterial colony count,thus resulting in low MIC value. EEP was found to be more statistically potent than Aloe Vera

GIC containing 10% EEP showed statistically significant difference in colony count when compared to GIC containing 10% Aloe Vera extract. Mann whitney test for post hoc comparision showed Gp2<GP5 with P value being <0.0001

GIC containing 20% EEP also showed statistically significant difference in colony count when compared to GIC containing 20% Aloe Vera extract. Mann whitney test for post hoc comparision showed Gp3<GP6 with P value being <0.0001

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Same was seen in GIC containing 30% EEP as it showed statistically significant difference in colony count when compared to GIC containing 30% Aloe Vera extract.Mann whitney test for post hoc comparision showed Gp4<GP7 with P value being <0.0001

When 10% , 20% and 30% dilutions of EEP in GIC were compared for the bacterial colony count,statistical difference was seen among the 3 groups.

Mann whitney test for post hoc comparision showed Gp4<Gp3<Gp2 with P value being <0.0001.

10% , 20% and 30% dilutions of Aloe Vera extract in GIC were compared for the bacterial colony count,statistical difference was seen among the 3 groups.

Mann whitney test for post hoc comparision showed Gp7<Gp6<Gp5 with P value being <0.0001.(Table 2)

GIC containing 10% ethanolic extract of propolis showed statistically no difference in bacterial colony count when compared with GIC containing 20% Aloe Vera extract.The P value according to post hoc comparisions by Mann Whitney test was 0.398(Table 2).

Table 3 depicts mean, standard deviation and range of shear peel band strength for all 7 groups.

Table 3 : - Mean ,Standard deviation and Range of Shear Peel Band Strength values for all 7 groups

Shear Peel Band Strength (Megapascals)					
Group	N	Mean	Std. Deviation	Range	
				Minimum	Maximum
Conventional GIC	20	1.4615	0.90209	0.39	2.96
GIC containing 10% ethanolic extract of propolis	20	1.4755	0.79360	0.57	2.81
GIC containing 20% ethanolic extract of propolis	20	1.6615	0.75901	0.69	2.91
GIC containing 30% ethanolic extract of propolis	20	1.6540	0.80796	0.51	2.91
GIC containing 10% of aloe vera extract	20	1.4590	0.87159	0.43	2.94
GIC containing 20% of aloe vera extract	20	1.5095	0.86903	0.47	2.95
GIC containing 30% of aloe vera extract	20	1.3320	0.80539	0.48	2.85
P[a] value	0.686, NS				

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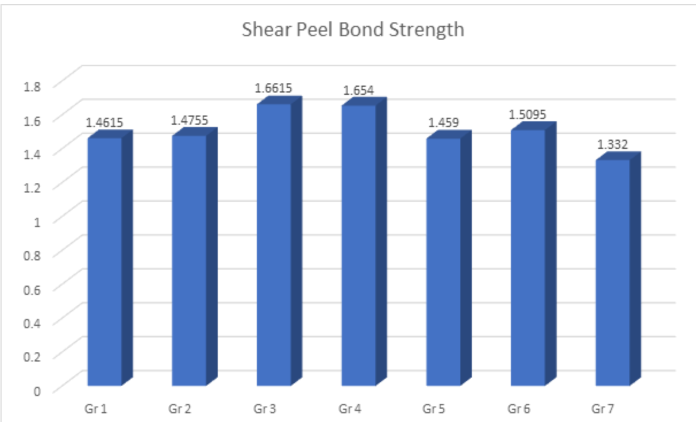
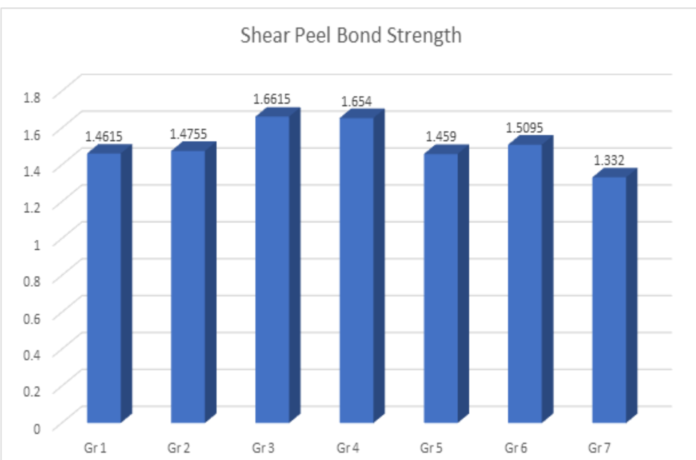
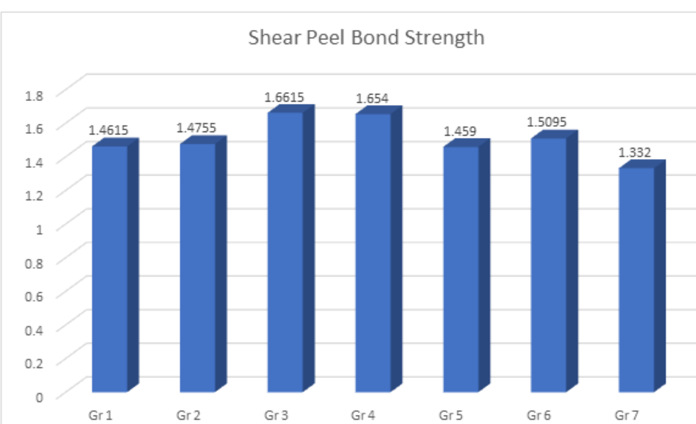
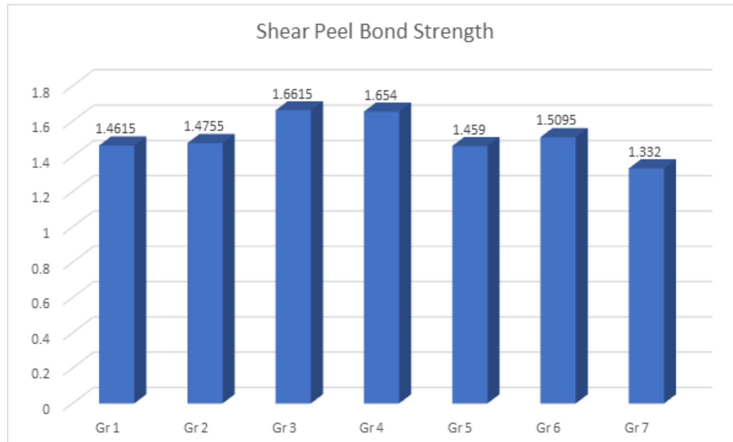
[a]Kruskal Wallis test

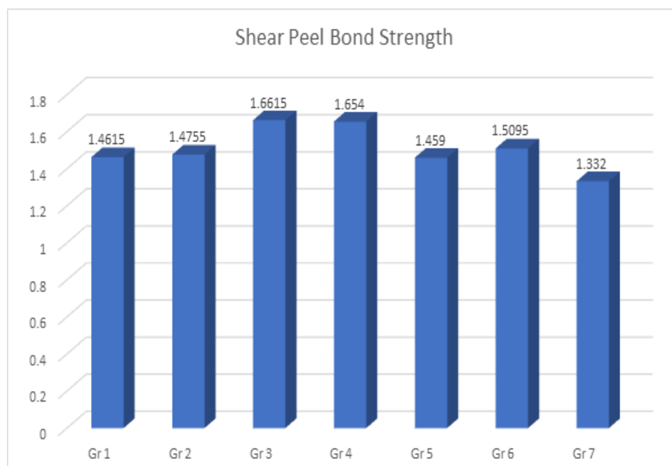
Mean values of Shear peel band strength for all 7 groups are depicted in graph 2.











Graph 2 :- Bar Graph depicting the median Shear Peel Band Strength of conventional GIC and GIC diluted with different concentrations of Aloe Vera and Ethanol extract of Propolis following debanding by the Universal Testing machine

Each band was cemented in place with its specific cement prepared by mixing GIC powder with appropriate liquid as assigned to the different groups. All specimens were then stored in distilled water at 37 degree C for 24 hours before band retention was measured because maximum band retention is obtained 24 hours after cementation. An Instron testing machine was used for the shear peel band strength (SPBS) test at a crosshead speed of 1 mm/min. The force was applied using a 0.9-mm diameter

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stainless steel wire from attachments on the buccal and lingual surface of the bands. The maximum load necessary to deband was recorded.

Since each group had in total 20 premolars banded,the mean for conventional GIC was found to be 1.4615 megapascals and that of GIC containing 30% ethanolic extract of propolis(which had the least MIC value and colony count,thus exhibiting highest antibacterial property) was 1.654 megapascals(Table 3,Graph 2)

The mean values of Shear Peel Band strength of samples with different concentrations of GIC and Aloe Vera as compared with samples of conventional GIC was found to be statistically insignificant with P value being 0.686(Table 3)

Table 4 depicts the comparison of all the groups for mechanical assay.

Table 4 :- P Values of all the 7 groups of the Mechanical Assay

	Gr 2	Gr 3	Gr 4	Gr 5	Gr 6	Gr 7
Gr 1	0.904	0.265	0.277	0.989	0.383	0.758
Gr 2	-	0.314	0.565	0.989	0.904	0.495
Gr 3	0.314	-	0.738	0.341	0.602	0.081
Gr 4	0.565	0.738	-	0.314	0.445	0.211
Gr 5	0.989	0.989	0.341	-	0.265	0.968
Gr 6	0.383	0.904	0.602	0.445	-	0.583

Statistically no significant difference was found in the P values of the each groups by applying Kruskal Wallis test(Table 4).

Post hoc pair comparison was done using Mann Whitney U test.

IV. DISCUSSION

Chlorhexidine, antibiotics, and titanium dioxide nanoparticles were added to GIC in previous studies. Adding chlorhexidine to GIC increased the antibacterial properties and had no deleterious effect on the mechanical properties (diameter tensile strength, shear bond strength) of GIC.[3,18,19]

Cariogenic microbacteria include S mutans, Lactobacillus, and some Actinomyces species. There is a significant increase in the levels of cariogenic bacteria in the saliva and plaque of patients undergoing fixed appliance treatment.[20]

However, during the initial phase of caries growth, S mutans is the most frequently associated microorganism.[21] S mutans was used in this study to examine antibacterial activity because it is considered to be the primary organism responsible for enamel demineralization.

The turbidity of S.mutans inoculum was adjusted to 0.5 Macfarland (10^5 cfu/ml) standard by using Brain Heart Infusion broth. Microtitre plates were labeled according to the groups and filled with Brain Heart Infusion broth containing S.mutans by using a sterile dropper.

All the samples from their respective groups were placed in the labeled wells by using a forcep and microtitre plate was incubated at 37 degree Celsius for 24 hours (Fig. 1)

After 24 hours broth containing the samples were subcultured on to the blood agar using inoculating loop and incubated at 37 degree Celsius for 24 hours(Fig 2)

After 24 hours colonies were counted and microbial growth of each group was compared.

Adding antibiotics to GIC enhanced antibacterial activity, but Yesilyurt et al.[22] showed that the antibiotic negatively affected the mechanical properties (compressive and bond strength) of GIC. Castilho et al[18] found that antibiotics have negative effects on compressive strength, but the finding was not statistically significant.

Elsaka et al.[23] observed that titanium dioxide nanoparticles improved the antibacterial properties of GIC without affecting the band strength insignificantly. This study showed that adding EEP to GIC increased antibacterial activity of GIC against S mutans without affecting the shear peel bond strength.

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The MIC value of the GIC containing 30% ethanolic extract of propolis was the least with 500 cfu/ml when compared with the rest of the six groups. The median of the antimicrobial colonies of the group containing GIC with 30% ethanolic extract of propolis was 800 cfu/ml with 200 Interquartile rangle, according to Kruskal Wallis test (Table 1).

Control group(Group 1) showed the highest bacterial colony count as compared to the rest the groups with the exception of GIC containing 10% of aloe vera extract, which showed hardly any difference in the bacterial colony count despite the addition of 10% Aloe Vera extract. This group which was labeled Group 5 had a median of 87500 cfu/ml with MIC value being 60000 cfu/ml.

GIC containing 30% EEP as it showed statistically significant difference in colony count when compared to GIC containing 30% Aloe Vera extract. Mann whitney test for post hoc comparision showed Gp4<GP7 with P value being <0.001

Aloe vera extract showed weak antibacterial activity as compared with EEP. This might be the result of low concentration of its antibacterial components compared to nutrient polysaccharides which could prevent the microorganism to be fully in touch with the Aloe vera active components. Aloe vera's pharmacotherapeutic and cosmetic properties have been studied since long time ago [16].

As a comparison between Aloe vera and propolis, the antimicrobial effect of Aloe vera was less than alcoholic extracts of propolis and its obtained MIC on *S.mutans* was more than propolis(Table1).

Evaluation mechanical properties of different concentrations of Ethanol Extract of Propolis and Aloe Vera extract was done by measuring SPBS .An Instron testing machine was used for the shear peel bond strength (SPBS) test at a crosshead speed of 1 mm/min. The force was applied using a 0.9-mm diameter stainless steel wire from attachments on the buccal surface of the bands. The maximum load necessary to deband was recorded using the instron testing machine. Results of the mechanical test revealed that adding EEP was statistically insignificant effect on SPBS (Fig 3 & 4).

Hatunoglu et al[25] also found no statistical significant increase in the Shear peel bond strength of Propolis in different dilutions whereas there was statistically significant increase in its antibacterial effect.

The study by Hatunoglu et al[25] was in agreement with this study were as there were contrasting results in the study by Troca et al.[24]

There are very few studies about how propolis and Aloe Vera affect the mechanical characteristics of cements. After the experience from this study, it was found that by adding propolis and Aloe Vera to the liquid of GIC makes the liquid less viscous and prolongs the working time.

However, the longer working time was at the low level, so this would not create any problems during the clinical application. These results should encourage the use of propolis and Aloe Vera in clinical practice. A study for the evaluation and comparision of the working time of Propolis and Aloe Vera when mixed with GIC is recommended to get an accurate and significant idea for its clinical application.

Shear peel bond strength (SPBS) is of great clinical value. After noticing increase in antimicrobial activity by adding 30% Ethanolic extract of Propolis in GIC , shear peel bond strength was tested with the help of Instron test machine . It concluded that the bond strength wasn't affected. The shear peel bond strength is of utmost importance because if the orthodontic band comes off due to poor bond strength than levelling and alignment will be lost thus adversely affecting the orthodontic treatment.

30% Ethanolic extract of propolis when mixed with GIC shows significant decrease in microbial activity thus should be considered in banding of molars as it results in less plaque accumulation

V. CONCLUSION

1) Both, GIC containing Ethanolic Extract of Propolis and GIC containing Aloe Vera Extract, showed a greater antibacterial effect than Conventional GIC.

2) GIC containing Ethanolic Extract of Propolis showed higher antibacterial effect than GIC containing Aloe vera extract and on comparison of mechanical properties, the shear peel bond strength(SPBS) was comparable in all the

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groups of GIC irrespective of addition of EEP/Aloe vera extract.

REFERENCES:

[1] Mizrahi E. Enamel demineralization following orthodontic treatment Am J Orthod 1982;82:62–7.

[2] Millett DT, Doubleday B, Alatsaris M, Love J. Chlorhexidine modified glass ionomer for band cementation? An in vitro study. J Orthod 2005;32:36–42.

[3] Farret MM, de Lima EM, Mota EG, Oshima HM, Barth V, de Oliveira SD. Can we add chlorhexidine into glass ionomer cements for band cementation? Angle Orthod 2011;81:496–502.

[4] Millett DT, Duff S, Morrison L, Cummings A, Gilmour WH. In vitro comparison of orthodontic band cements. Am J Orthod Dentofacial Orthop 2003;123:15–20.

[5] Hoszek A, Ericson D. In vitro fluoride release and the antibacterial effect of glass ionomers containing chlorhexidine gluconate. Oper Dent 2008;33:696–701.

[6] Topcuoglu N, Ozan F, Ozyurt M, Kulekci G. In vitro antibacterial effects of glass ionomer cement containing ethanolic extract of propolis on Streptococcus mutans. Eur J Dent 2012;6:428-33.

[7] S. Kumazawa, T. Hamasaka, and T. Nakayama, "Antioxidant activity of propolis of various geographic origins," *Food Chemistry*2004;84(3); 329-39.

[8] A. Kujumgiev, I. Tsvetkova, Y. Serkedjieva, V. Bankova, R. Christov, and S. Popov, "Antibacterial, antifungal and antiviral activity of propolis of different geographic origin," *Journal of Ethnopharmacology* 1999;64(3); 235–40.

[9] A. Russo, V. Cardile, F. Sanchez, N. Troncoso, A. Vanella, and J. A. Garbarino, "Chilean propolis: antioxidant activity and antiproliferative action in human tumor cell lines," *Life Sciences* 2004;76(5);545–58.

[10] D. Sawicka, H. Car,M. H. Borawska, and J. Nikli´nski, "Theanticancer activity of propolis," *Folia Histochemica et Cytobiologica*, 2012;50(1) ;25–37.

[11] E. Szliszka, Z. Czuba, J. Bronikowska, A. Mertas, A. Paradysz, and W. Krol, "Ethanolic extract of propolis augments TRAILinduced apoptotic death in prostate cancer cells," *Evidence- Based Complementary and Alternative Medicine* 2011;11;1-11.

[12] E. Szliszka, Z. P. Czuba, K. Jernas, and W. Kr´ol, "Dietary flavonoids sensitize HeLa cells to tumor necrosis factor-related apoptosis-inducing ligand (TRAIL)," *International Journal of Molecular Sciences*2008;9(1);56–64.

[13] J. M. Sforcin, A. Fernandes, C. A. M. Lopes, V. Bankova, and S. R. C. Funari, "Seasonal effect on Brazilian propolis antibacterial activity," *Journal of Ethnopharmacology* 2000;73(1);243– 49.

[14] G. P. Rezende, F. C. Pimenta, and L. R. Costa, "Antimicrobial activity of two Brazilian commercial propolis extracts," *Brazilian Journal of Oral Sciences* 2006;5;967–97.

[15] F. A. Santos, E. M. A. Bastos, M. Uzeda et al., "Antibacterial activity of Brazilian propolis and fractions against oral anaerobic bacteria," *Journal of Ethnopharmacology* 2002;80(1);1–7.

[16] Ehsani M , Marashi MA, Zabihi E , Issazadeh M , Khafri. A Comparison between Antibacterial Activity of Propolis and Aloe vera on Enterococcus faecalis (an *In Vitro* Study). IJMCM 2013;2:110-7.

[17] Y. K. Park, M. H. Koo, J. A. S. Abreu, M. Ikegaki, J. A. Cury, and P. L. Rosalen, "Antimicrobial activity of propolis on oral microorganisms," *Current Microbiology* 1998; 36 (1); 24–28.

[18] Castilho AR, Duque C, Negrini Tde C, et al. Mechanical and biological characterization of resin-modified glass-ionomer cement containing doxycycline hyclate. Arch Oral Biol. 2012; 57:131–8.

[19] Sanders BJ, Gregory RL, Moore K, Avery DR. Antibacterial and physical properties of resin modified glass-ionomers combined with chlorhexidine. J Oral Rehabil. 2002;29:553-8.

[20] Feres M, Figueiredo LC, Barreto IM, Coelho MH, Araujo MW, Cortelli SC.In vitro antimicrobial activity of plant extracts and propolis in saliva samples of healthy and periodontally-involved subjects. J Int Acad Periodontol. 2005;7:90–6.

[21] Turkun LS, Turkun M, Ertugrul F, Ates M, Brugger S. Long-Term Antibacterial Effects and Physical Properties of a Chlorhexidine Containing Glass Ionomer Cement. *J* Esthet Restor *Dent 2008;*20:29–45.

[22] Yesilyurt C, Er K, Tasdemir T, Buruk K, Celik D. Antibacterial activity and physical properties of

glass-ionomer cements containing antibiotics. Oper Dent. 2009;34:18–23

[23] Elsaka SE, Hamouda IM, Swain MV. Titanium dioxide nanoparticles addition to a conventional glass-ionomer restorative: influence on physical and antibacterial properties. J Dent. 2011;39:589–98.

[24] TROCA VBPB, Fernandes KPB, Terrile AE, Marcucci MC. Effect of green propolis addition to physical mechanical properties of glass ionomer cements. J Appl Oral Science 2011;19:100-5.

[25] Hatunoglu E, Ozturk F, Bilenler T, Aksakall S, Simsek N. Antibacterial and mechanical properties of propolis added to glass ionomer cement. Angle Orthod. 2014;84:368–3