

Original Article

An *in vitro* evaluation of the compressive strength and shear bond strength of resin-modified glass-ionomer cement containing purified powder of *Salvia officinalis*

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ABSTRACT

Background: In this study, the effect of adding purified powder of *Salvia officinalis* on the mechanical properties and bonding ability of resin-modified glass ionomer (RMGI) cement is investigated.

Materials and Methods: In this *in vitro* study Purified powder of *S. officinalis* with particles smaller than 50 μ , in weight percentages of 0.5%, 0.75%, 1%, and 1.25%, was added to RMGI powder (GC Fuji II LC, GC USA). Fifty samples in five groups of control and percentages that considered, from the powder of GI combined with liquid, according to the manufacturer instructions and prepared samples with 4 mm \times 6 mm dimensions and placing in distilled water in an incubator with temperature 37°C for 24 h. Fifty sound extracted upper premolars were collected and divided into five groups. The dentinal cross-sections on the occlusal surfaces of teeth were prepared and assessed under a stereomicroscope with \times 16. Then, samples with 2 \times 4 dimensions from RMGI was prepared on dentin surface and keeping in moisture environment in an incubator with temperature 37°C for 24 h. Compressive strength and shear bond strength test done with a universal testing machine at the rate of 0.5 mm/min. For the assessment of mode of failure, stereomicroscope with a digital camera and \times 20 was used, and photographs from bonding surface were taken. After collecting data, the ANOVA test was used for comparing shear bond strength between groups and Kruskal–Wallis test was used for comparing compressive strength. The Chi-square test was used for comparing the mode of failure between groups. The significance level for all tests was considered 5%.

Results: According to the statistical analysis, compressive strength ($P = 0.486$) and shear bond strength ($P = 0.076$) were not significantly different between the five groups. The distribution of mode of failure was not significantly different between groups ($P = 0.663$).

Conclusion: The addition of purified powder of *S. officinalis* to RMGI powder does not affect compressive strength and shear bond strength of RMGI cement.

Key Words: Bond strength, compressive strength, glass ionomer, *Salvia officinalis*

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INTRODUCTION

Administration of modern concepts in preventive dentistry, availability of new dental materials,

and improved education of the target populations have greatly improved the quality of oral health

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in different societies. On the other hand, having a microbial nature, dental caries is still a challenge which annually imposes major costs to the individuals and the whole health-care system.^[1,2] The development of smart dental materials with bioactive antimicrobial functions could enormously assist in almost full control of carious lesions besides the mechanical removal as the traditional method of treatment.^[3,4]

Introduction and development of glass-ionomer cements (GICs) during the years has provided dentists with a biocompatible, esthetic self-adhesive restorative material.^[5,6] GICs are indicated as a restorative, luting, or lining material. Their inherent ability to release and reuptake fluoride ion has been claimed to inhibit caries initiation or progression, but there is no clear scientific evidence to support this caries inhibitory potential.^[7,8]

The fact that ions can readily travel in and out of the GICs offers the opportunity to dope the cement with other soluble antimicrobials. In this regard, many investigations have evaluated the possibility of incorporation of a direct antimicrobial agent into GICs, innovating an esthetically pleasing, self-adhesive dental material which can control residual caries, decrease the incidence of recurrent caries and provide a microbial seal under direct or indirect restorations.^[7-9]

Takahashi *et al.* evaluated the antibacterial, physical, and bonding properties of a GIC containing chlorhexidine diacetate. They showed that incorporation of 1% chlorhexidine diacetate could provide antibacterial properties to GIC while keeping the other properties in the range.^[10]

In another study, Hook *et al.* investigated a GIC modified with nanoparticles of chlorhexidine hexametaphosphate. They concluded that this modified type of GIC has antibacterial properties on a dose-dependent base. Substitutions up to 5% appeared to have no significant deleterious effect on the tensile strength of cement.^[7]

In another experiment by de Castilho *et al.*, a resin-modified glass ionomer (RMGI) functionalized with doxycycline hyclate was studied. This antibiotic is active against many oral pathogenic bacteria and also shows an anti-MMps activity. The study showed positive antimicrobial results without modifying the biological and mechanical characteristics of dental material.^[11]

Positive properties of herbal extracts with a long history of use in traditional and modern medicine could be a new area of research in the production of preventive and restorative dental materials. Being safer and cheaper while showing the same effects are the main advantages.^[12]

Salvia officinalis, a herb with extensive therapeutic and preventive properties, is a perennial evergreen plant naturalized in Iran.^[13-15] It has a long history of use in the treatment of oral and pharyngeal infections and inflammations.^[12,13] The extract has been shown to inhibit *Streptococcus mutans* and *Lactobacillus* species as the main families of bacteria involved in the caries process.^[12] Superior properties compared to common antibiotics and synthetic antimicrobials make it the substitution of choice.^[14]

Hamidpour *et al.* presented a comprehensive analysis of the botanical, chemical, and pharmacological aspects of *S. officinalis* has. The findings of the study support the view that the hydroalcoholic extract of *S. officinalis* has a growth inhibitory effect on some dental caries causing bacteria such as *S. mutans*, *Lactobacillus rhamnosus*, and *Actinomyces viscosus*.^[15] Based on this study and the global interest in using traditional treatments instead of chemical solutions, *S. officinalis* with its bactericidal effect could be a natural remedy for the treatment of diseases affecting mouth and teeth.^[15,16]

Beheshti-Rouy *et al.* evaluated the clinical effectiveness of a mouthwash containing 1% *S. officinalis* extract on the reduction of *S. mutans* in dental plaque in a group of school-aged children. Sage extract mouth rinse exerted antibacterial action against *S. mutans* in dental plaque.^[13]

Shahriari *et al.* investigated the anti-*S. mutans* and anti-*Lactobacillus casei* properties of GIC modified with extract powder of *S. officinalis*. *S. officinalis* containing GIC have direct inhibitory activities against *S. mutans* and *L. casei* in a dose-response manner.^[17]

The aim of the present study is to investigate the effect of the addition of *S. officinalis* extract on compressive strength and dentin bonding ability of RMGI.

MATERIALS AND METHODS

In this *in vitro* study, Some high-quality dried leaves of *S. officinalis* is chopped and fragmented into small pieces and filtered through a #40 mesh

(Sina Lab. Inst., Tehran, Iran). Each 50 g of leaves are soaked in 1500 ml of solvent (50% water, 50% ethanol 96%) in a shaker apparatus (Heidolph Unmax, Schwabach, Germany) at 90 rpm for 48 h. Thereafter, the solution is passed through a strainer and then transferred to a rotary evaporator apparatus (Heidolph WD2000; Schwabach, Germany) to separate the solvent from the extract. The purified extract is then dried by applying the freeze-drying technique in three stages over 1 week. The final extract powder is stored in sealed vial at low temperatures to be used in the next steps. To filter the particles, the same size as the range of GIC powder, which is to be $<50\ \mu$, the powder is grinded and again filtered through a #270 laboratory mesh (Sina Lab. Inst., Tehran, Iran). The procedure is performed under the supervision of a pharmacology professor at the main laboratory of the School of Pharmacy, Isfahan University of Medical Sciences and Health Services, Isfahan, Iran.

A conventional powder and liquid Fuji II LC GIC (GC Corporation, Tokyo, Japan) are used as the control group. Experimental GIC samples are prepared by incorporating *S. officinalis* extract powder into the powder component of Fuji II LC GIC (GC Corporations, Tokyo, Japan) at 0.5%, 0.75%, 1%, and 1.25% weight concentration levels using a digital weight scale (METTLER AE200, Switzerland).

According to the standard ISO 9917 (Dental water-based cements 1991) and similar studies,^[1,4,11] cylindrical samples with a diameter of 4 mm and a length of 6 mm were prepared using plastic molds. According to the manufacturer's instructions, powder/liquid of glass ionomer was mixed on the glass slab and then, using a spatula, the mixture is moved into the mold placed on glass slab, and after ensuring that the mold was completely filled, and the air bubbles were removed, the surface of the sample was flattened with a glass slide and light-cured from each side for 20 s with the intensity of 1200 mW/cm² (LED Curing Light, Dentamerica, USA). Ten specimens were prepared for each group. After keeping the samples for 1 h in a wet environment, the specimens were immersed in distilled water and kept in an incubator at 37°C for 24 h. The specimens were extracted from the mold, and the compressive strength test was performed at a speed of 0.5 mm/min on the specimens using an Electromechanical Universal Testing Machine (K-21046, Walter + Bai, Switzerland). The numbers obtained were the maximum force input on

the sample up to the moment of failure in terms of the Newton, by dividing on the cross-section of the samples in mm, the compressive strength in mpa is obtained.

According to standard ISO CD TR 11405 and same studies^[18,19] to assess the shear bond strength to dentin, upper premolar teeth were extracted for orthodontic treatment were collected. After cleaning the teeth with brush and checking the absence of decay and structural anomalies, they were kept in 1% thymol solution. Then, the teeth were randomly divided into five groups of ten. The occlusal surface of all specimens was cut with a disk to prepare the dentinal sample. To ensure that dentin cuts were obtained without the presence of enamel and no pulp exposure, the samples were examined under a $\times 16$ magnification Trinocular Zoom stereo microscope (MBX-10, Labo America Inc., Russia). To simulate the clinical conditions, the dentin surface was roughened using diamond with medium particles. The teeth were molded using self-cure acrylic resin in plastic molds. Then plastic molds with a diameter of 2 mm and length of 4 mm were fixed on the dentin surface, and powder/liquid of glass ionomer was mixed on the glass slab and then, using a spatula, the mixture was moved into the mold, and after ensuring that the mold was completely filled and the air bubbles were removed, light cured from each side for 20 s (LED Curing Light, Dentamerica, USA). The specimens were kept in a 100% humidity medium for 24 h at 37°C in an incubator. The specimens were extracted from the mold, and the shear bond strength test was performed using a universal testing machine at a speed of 0.5 mm/min. The force was applied with a wide edge blade at the sample/dentin joint. The maximum force input on the sample up to the moment of sample separation in terms of the Newton is obtained, by dividing on the cross-section of the samples in mm; the Shear bond strength in mpa is obtained. To investigate the type of failure, a $\times 20$ stereomicroscope equipped with a digital camera (SMP 200, HP, USA) was used, and the photo was taken from the bond surface [Figures 1-3].

Statistical analysis

After collecting data using the IBM SPSS 22.0 software (SPSS Inc., IL, USA), the Kolmogorov–Smirnov test was performed to verify the normal distribution of data, that in both compressive strength and shear bond strength were not significant and the distribution of data was normal. Then,

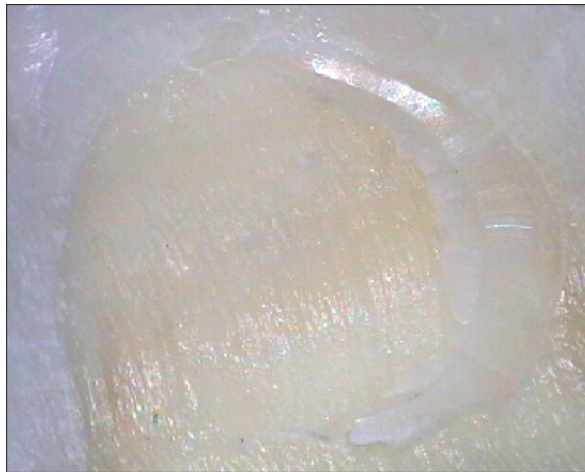


Figure 1: Mix failure of resin-modified glass ionomer sample bonded to dentin bonding surface.



Figure 2: Mix failure of resin-modified glass ionomer sample bonded to dentin bonding surface.

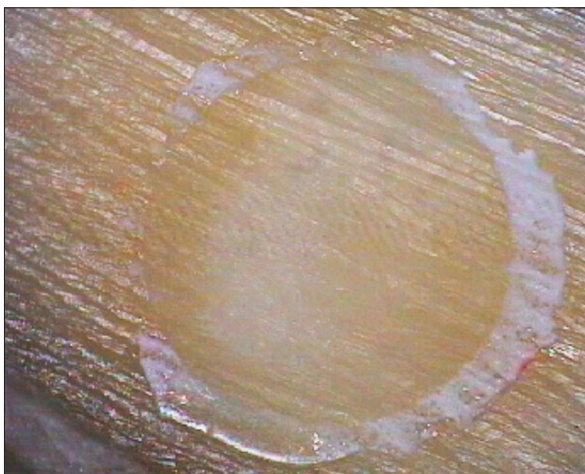


Figure 3: Adhesive failure of resin-modified glass ionomer sample bonded to dentin bonding surface.

Levene's test was used to evaluate the homogeneity of variances. This test was not significant for shear

bond strength data ($P = 0.782$), and the ANOVA test was used to compare the shear bond strength of the groups. However, Levene's test for the compressive strength data was statistically significant ($P = 0.031$), and the Kruskal–Wallis test was used to compare the compressive strength of the groups. The Chi-square test was used to compare the failure type in the shear bond strength test. The significance level for all tests was considered 5%.

RESULTS

According to Kruskal–Wallis test, the compressive strength between the five groups was not statistically significant ($P = 0.486$), and the addition of the *S. officinalis* extract to the RMGI cement powder did not affect the compressive strength of it [Table 1]. According to the ANOVA test, the shear bond strength of dentin between the five groups was not statistically significant ($P = 0.076$), and the addition of the *S. officinalis* extract to the RMGI cement powder did not affect the bond strength of the dentin [Table 2]. According to the Chi-square test, failure type in dentin bonding was adhesive and mixed, and it was not significantly different among groups ($P = 0.663$) [Table 3].

DISCUSSION

The ability of dental materials to prevent the formation of recurrent caries is an important clinical feature. Glass ionomer has been used for more than 30 years, and its major ability to control decay is well known because of releasing fluoride and adhesion into dentin structure.^[1]

The release of fluoride and low pH of glass ionomer during the setting is well known. However, the results of previous studies on antibacterial effects caused by fluoride and low pH of this substance are contradicting, and a decrease in bacterial count due to the use of glass ionomer is not reliable.^[1,20,21]

The view of “controlled release therapeutic systems” with the aim of releasing specified amounts of a drug for a specific time is not new, and combining antibacterial agents, especially chlorhexidine, with restorative materials has already been investigated.^[1]

RMGI is used for the restoration of posterior teeth and base under extensive restorations that, in these cases, the strength of material against heavy occlusal forces should be sufficient. These properties of material are investigated by its compressive strength.^[4]

Table 1: Compressive strength (mpa) of the RMGI containing purified powder of *Salvia officinalis*

Concentration (% weight)	n	Mean	SD	SE	95% CI for mean		Minimum	Maximum
					Lower bound	Upper bound		
0 (control)	10	185.7600	50.08191	15.10026	152.1145	219.4055	124.17	270.80
0.5	10	175.1190	37.21701	11.76905	148.4956	201.7424	118.24	218.05
0.75	10	199.5730	24.61774	7.78481	181.9625	217.1835	155.57	236.32
1	10	176.7890	34.01375	10.75609	152.4570	201.1210	121.22	224.17
1.25	10	174.8960	27.89117	8.81996	154.9439	194.8481	141.96	220.65
Total	50	182.4927	36.02689	5.04477	172.3600	192.6255	118.24	270.80

SD: Standard deviation, SE: Standard error, CI: Confidence interval, RMGI: Resin-modified glass ionomer

Table 2: Shear bond strength (mpa) of the RMGI containing purified powder of *Salvia officinalis*

Concentration (% weight)	n	Mean	SD	SE	95% CI for mean		Minimum	Maximum
					Lower bound	Upper bound		
0 (control)	10	7.4540	2.80587	0.88729	5.4468	9.4612	2.71	12.81
0.5	10	5.5160	2.61011	0.82539	3.6488	7.3832	2.24	11.14
0.75	10	5.0000	2.15359	0.68103	3.4594	6.5406	1.62	8.22
1	10	4.0880	2.42519	0.76691	2.3531	5.8229	1.41	8.76
1.25	10	6.6340	3.69349	1.16799	3.9918	9.2762	2.43	14.92
Total	50	5.7384	2.92861	0.41417	4.9061	6.5707	1.41	14.92

SD: Standard deviation, SE: Standard error, CI: Confidence interval, RMGI: Resin- modified glass ionomer

Table 3: Type and percentage of dentin bonding failure of the RMGI containing purified powder of *Salvia officinalis*

Group (% weight)	Mode of failure count (%)		Total (%)
	Adhesive	Mix	
0	4 (40)	6 (60)	10 (100)
0.5	4 (40)	6 (60)	10 (100)
0.75	5 (50)	5 (50)	10 (100)
1	5 (50)	5 (50)	10 (100)
1.25	7 (70)	3 (30)	10 (100)
Total (%)	25 (50)	25 (50)	50 (100)

RMGI: Resin- modified glass ionomer

After evaluating the antibacterial effect of RMGI containing extract of *S. officinalis*,^[17] the present study evaluated compressive strength and shear bond strength of RMGI containing purified powder of *S. officinalis*. According to the results of this study, the addition of purified powder of *S. officinalis* to GIC powder did not have a negative effect on compressive strength and shear bond strength. It was also determined that with increasing weight percent of salvia extract combined with glass ionomer powder, there was no significant difference in compressive strength and shear bond strength to dentin between groups and in comparison with the control group.

In the same studies to produce antibacterial properties for glass ionomer, different antibiotics, and antibacterial agents were added to glass ionomer or RMGI, and physical, mechanical, and antimicrobial

activity of samples were investigated.^[1,11,22] The results of these studies were similar to the present study, and the addition of small amounts of antibacterial agents had inhibitory effects on caries-producing bacteria without negative effects on mechanical properties of the substance.^[1,11,22]

However, the use of antibiotics and antibacterial agents has the potential for creating side effects and antibiotic-resistant bacteria.^[11] In the present study, due to the use of the herbal drug with known history, there is no likelihood of these complications.^[14,15]

Decrease in mechanical properties can be due to a slight change in the powder/liquid ratio following the addition of antimicrobial agent at high concentrations, but similar to the present study, at low concentrations (below 2%) without any negative mechanical changes, the antimicrobial effect of substance still exists.^[1,10,17,22]

In the study of Becci *et al.*, the effect of adding chlorhexidine on the bond strength of GIC to healthy dentin and dentin affected by decay was investigated. The concentrations of 0.5% and 1% of chlorhexidine diacetate, increased antibacterial activity of cement and has similar bond strength in comparison with pure glass ionomer, but at a concentration of 2%, the bond strength was decreased.^[18]

The chemical bond of glass ionomer to mineralized tooth tissue is due to ionic exchange between carboxylic

groups of acid polyacrylic and calcium hydroxyapatite ions.^[23,24] Because of its cationic properties, chlorhexidine salts interact with the reaction of polyacrylic acid and glass particles, and the mechanical and bonding properties of the agent are affected.^[18]

In this study, RMGI was used. Given that the bond strength of RMGI is more than common glass ionomer, and its amount is acceptable for bonding durability to the tooth structure, the addition of antimicrobial agents in low amounts cannot affect the bond strength to the tooth structure.^[8,25,26]

In the study of Hatunoğlu *et al.*, the effect of adding an ethanolic extract of propolis on antibacterial properties and bond strength of GIC used to attach orthodontic bands was investigated.^[27] The results were similar to the present study and the addition of materials with the natural base has no adverse effect on the properties of glass ionomer.

The chemical composition of *S. officinalis* extract consists mainly of 1,8-cineole, camphor, borneole, α -pinene, and camphene. There are also flavonoids and polyphenolic compounds such as carnosic acid, rosmarinic acid, and caffeic acid present in this herb.^[14,15] These compounds have OH and COOH functional groups in their chemical formulas. The presence of these active groups leads to antioxidant properties and the release of harmful free radicals.^[15] Functional groups OH and COOH are also present in the chemical structure of glass ionomer, which participates in the acid-base reaction between polyacrylic acid and glass particles as well as glass ionomer bond to calcium in the tooth structure.^[5,23,24] The similarity of chemical formula of these compounds with functional groups present in GIC can be explained that there is no significant reduction in compressive strength and bond strength to dentin of samples containing extract of the herb. It seems that the addition of *S. officinalis* extract at low concentrations does not alter the chemical structure of glass ionomer and does not adversely affect the properties of the main ingredient and may be due to the presence of compounds having OH functional groups that are increasing bond strength of the material to the tooth structure. Of course, further studies are needed on possible chemical reactions that have been made in combining these two substances together.

Adhesive and mixed failure of bonding to dentin was seen in the samples. However, there was no significantly difference between the groups.

CONCLUSION

According to the results of this study, the ability to apply occlusal loads on the glass ionomer having the extract of *S. officinalis* and the inherent bond strength to the tooth structure has not decreased.

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Conflicts of interest

The authors of this manuscript declare that they have no conflicts of interest, real or perceived, financial or non-financial in this article.

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