

IMPACT OF DIFFERENT CONCENTRATIONS OF SALVIA OFFICINALIS ON THE FLEXURAL STRENGTH AND MICROHARDNESS OF RESIN-MODIFIED GLASS-IONOMER CEMENTS

Dr. Sultana Ashraf Ali  
MD trainee, Department Of Dental Materials, Liaquat College of Medicine and Dentistry.  
[sultana.ashraf106@gmail.com](mailto:sultana.ashraf106@gmail.com)

Dr. Asad Farooq  
Associate Professor, Department Of Dental Materials, Liaquat College of Medicine and Dentistry. [drasadfarooq@gmail.com](mailto:drasadfarooq@gmail.com)

Dr. Ishma Binte Saeed  
MS trainee, Department Of Periodontology, Liaquat College of Medicine and Dentistry.  
[dr.ishma.saeed@gmail.com](mailto:dr.ishma.saeed@gmail.com)

Dr. Sobia Tariq  
MDS Trainee, Department Of Oral Surgery, Ziauddin University.  
[Sobia\\_bds15@hotmail.com](mailto:Sobia_bds15@hotmail.com)

Abstract

| Author Details                    |
|-----------------------------------|
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| Corresponding E-mails & Authors*: |

Resin modified glass-ionomer cement (RMGIC) has become popular in restorative dentistry due to its adhesive properties to dental hard tissues, fluoride release and ease of handling when compared with conventional glass-ionomer cement. However, flexural strength and the surface hardness and mechanical durability over time are still points of concern. Sage (Salvia officinalis) is rich in phenolic compounds, flavonoids and essential oil constituents that have been reported to have antioxidant, antimicrobial and anti-inflammatory properties. The inclusion in RMGIC might offer a way to bioactive restorative materials, without losing mechanical properties. The objective of this thesis was to check the flexural strength and microhardness changes of RMGIC after adding Salvia officinalis powder with different concentrations. The methods used were by comparing a control RMGIC group with RMGIC with 2%, 5% and 7% Salvia officinalis powder by weight. There were 100 valid flexural strength samples provided in the sample collection: 25 samples in each group. The flexural strength was determined by three-point bending test using the equation  $\sigma = 3FL / 2bh^2$ , where F is fracture load, L is supporting span, b is width and h is thickness. Descriptive statistics were used to analyse the data, followed by the Shapiro-

Wilk normality test, Levene's test for homogeneity, one-way ANOVA, followed by Kruskal-Wallis's sensitivity testing and pairwise comparisons. Available bench-note readings and specimen images were used to describe microhardness. Mean flexural strength was 10.15 MPa for control, 10.44 MPa for 2% sage, 10.43 MPa for 5% sage and 10.32 MPa for 7% sage. Normality and homogeneity were met. There was no significant difference between the groups based on one-way ANOVA ( $p = 0.732$ ). This was corroborated by Kruskal-Wallis's testing ( $p = 0.720$ ). The available microhardness readings were descriptive and incomplete and thus, there was no definitive conclusion regarding the microhardness. Discussion: *Salvia officinalis* powder with the concentration of 2%, 5% and 7% was not significantly reduce flexural strength of RMGIC. Mechanically, the modified groups were similar to the control group in bending. Prior to clinical interpretation, complete microhardness data and antibacterial testing is recommended.

**Keywords:** Resin-modified glass-ionomer cement; *Salvia officinalis*; sage; flexural strength; microhardness; dental materials.

## 1. Introduction

Restorative dental materials are required to function in a demanding biological and mechanical environment. A material should be handled and placed predictably, bond or be retained satisfactorily, tolerate moisture and chemical challenge, and withstand repeated loading and provide an acceptable surface that is clinically acceptable. The relevance of GICs is the chemical interaction with enamel and dentine, the release of fluoride and their compatibility with conservative restorative treatments. Unlike materials retaining fluoride exclusively through micromechanical retention, they are capable of releasing and recharging fluoride through ionic bond to mineralised tissue. However, there are drawbacks to using conventional GICs such as early moisture sensitivity, brittleness, comparatively low fracture resistance and lower wear resistance compared to many resin-composite systems (Sidhu and Nicholson, 2016; Park and Kang, 2020).

The aim of the development of resin-modified glass-ionomer cements (RGIC) was to incorporate desirable properties of conventional GICs and enhance handling and initial mechanical properties. In RMGIC, the acid-base reaction between the fluoroaluminosilicate glass and polyalkenoic acid is a traditional reaction that takes place simultaneously with light-activated polymerisation of resin components. This material is thus not just a resin with glass particles but also a multiphase system where the reactive glass, water, polyacid, resin monomers, photoinitiators and developing ionic matrix interact. The resin reaction - early strength, command setting; acid-base reaction

and subsequent maturation - adhesion, Fluoride-related behaviour (Park and Kang, 2020; Ghilotti et al., 2023). This dual setting process is clinically applicable, but could mean that small formulation and water balance changes, changes in light transmission and change in powder-to-liquid ratio can affect performance.

In cervical restorations, paediatric dentistry, liners, bases and in selected minimally invasive restorative procedures, where adhesion and fluoride release are important, perfect isolation is often difficult, and in these cases RMGIC is used. Additional factors that affect long-term clinical use are resistance to fracture, surface degradation and wear. Flexural strength is important because restorations are subjected to tensile and compressive stresses during bending. Defects on tensile surface are particularly relevant for brittle materials, and a material which is satisfactory in compression can fail in tensile with the presence of defects. The surface hardness is a distinct but related property of resistance to indentation, scratching and early wear. Research comparing products made of glass ionomer cement has found significant variations in flexural strength and microhardness, which can be attributed to the differences in the formulation, particle distribution, porosity, resin content and curing behaviour of the products (Rêgo, Butler and Santos, 2022; Bilge, Aşar and İpek, 2024; Lile et al., 2025).

The development of antimicrobial agents, fibres, nanoparticles and natural products into GIC and RMGIC has been driven by the interest of bioactive restorative materials. The goal is generally to add a second life to a product, but not to rule out the use of the original physical properties needed to serve their purpose. It's a tricky balance. An additive can also suppress microorganisms or adjust ion release, but also can provide some reactive glass, improve porosity, impact the light transmission, affect water uptake or produce weak interfaces. Studies on chlorhexidine, cetrimide, triclosan, antibiotics, and plant derived agents show that it is dependant on concentration, solubility, particle form, and compatibility with the base cement. Increased concentration can be beneficial for the loading of active ingredient as well as for mechanical deterioration (Yadiki et al., 2016; da Mota Martins et al., 2022; Gaur et al., 2026). So, the biological promise needs to be understood in conjunction with mechanical reliability.

*Salvia officinalis* L., commonly referred as sage, belongs to the Lamiaceae family and is well known for its medicinal and culinary applications. The preparations contain some compounds like rosmarinic acid, caffeic acid, carnosic acid, flavonoids, terpenoids, 1,8-cineole, camphor and borneol as essential-oils constituents. These compounds can be present in various amounts and forms depending on the botanical source and processing and formulation, and have been reported with antioxidant, antimicrobial, and

anti-inflammatory properties (Ghorbani and Esmaeilizadeh, 2017; Jakovljević et al., 2019; Maggini et al., 2024). Sage has been used in the oral health field in mouthwashes, extracts and experimental materials. Biological plausibility is given in clinical and laboratory reports regarding the effect against plaque associated organisms or oral inflammation, however, the delivery system and the concentration are important and the results obtained with a mouthwash or an essential oil cannot be assumed to apply to the cured restorative cement (Fadhil et al., 2024; Anwar et al., 2025).

Laboratory studies with purified *Salvia officinalis* powder are most direct evidence for the products that have been modified with sage. Shahriari et al. (2019) observed antibacterial activity when a small amount of sage powder was added to the conventional GIC. Forouzanmehr, Barekatin and Shahtalebi (2020) tested the effect of different concentrations of sage powder, 0.5%, 0.75%, 1.0% and 1.25%, on an RMGIC, and found no significant difference in compressive strength or shear bond strength. The results indicate that low levels are mechanically tolerated under certain conditions. They do not, however, define the effect of increased concentration and neither do they answer the question as to whether flexural strength and surface hardness is retained. The compressive strength is primarily related to resistance to crushing, while flexural strength is more susceptible to flaws on the tensile side, particle agglomeration and lack of matrix continuity.

There is a possibility of a concentration dependent response. Finely dispersed organic particles can be included in the powder phase without inducing a significant structural change, at low or moderate levels. They could be only partially surrounded by the matrix or they could occupy space. At higher loading rates, the same particles can result in agglomeration, more voids, less glass reactivity and stress concentration sites. The use of polymer composites with lower or intermediate amounts of the filler has been shown to be beneficial in the case of less beneficial dispersion, which is not possible to make a numerical transfer to RMGIC, as the matrices and the setting reactions are different (Poranek et al., 2025). Therefore, testing multiple concentrations of a product with a control of the same product is more informative than testing a single experimental formulation.

The literature consequently leaves three related gaps. Firstly, there is limited flexural-strength data of sage-modified RMGIC. Secondly, it is not known enough about the microhardness after the incorporation of graded sage. Third, biological and mechanical outcomes are seldom tested in the same formulations and it is thus challenging to identify a concentration that ensures retaining material characteristics and giving a meaningful antimicrobial action. In the present study, part of this deficiency

was overcome by comparing unmodified RMGIC with RMGIC with various contents (2%, 5% and 7% by weight) of *Salvia officinalis* powder. The main purpose was the assessment of three point bending flexural strength. The second goal was to characterize the microhardness data from the laboratory records, whilst acknowledging that the microhardness data set was not complete. It was hypothesized that the control and sage modified groups will have no difference in flexural strength.

## 2. Materials And Methods

### Study Design And Setting

A controlled laboratory experiment was performed by creating an experimental in vitro design to compare one unmodified RMGIC group with three sage-modified groups. The choice of in vitro testing was made because the preparation and storage of specimens, loading and data analysis could be standardised before any biological or clinical evaluation of the modified material. Laboratory work was related to the Pakistan Council of Scientific and Industrial Research laboratories and Junior Operative Laboratory at Liaquat College of Medicine and Dentistry. The study synopsis outlined a 1 year after institutional clearance for procurement of materials, preparation of specimens, testing, data collection and analysis.

There were flexural-strength and microhardness specimens in the original study plan. There were 100 flexural-strength observations, 25 in each of four groups, available for the present manuscript. A series of readings from a bench note was planned but was limited; microhardness testing was planned for balanced groups. Therefore, flexural strength was considered as a full primary outcome and microhardness was reported as an exploratory descriptive outcome. This distinction was made prior to the results being interpreted because of the desire to not make the mistake of comparing an incomplete and unbalanced observation as a fully powered comparison.

### Materials And Experimental Groups

The base material was used as a powder and liquid combination of GC Gold Label light-cured universal restorative glass ionomer cement. *Salvia officinalis* powder was the powder used as an experimental additive. Laboratory records indicated the plant material was dried sage to be included in the cement powder. Four different formulations were tested; Group 1, unmodified RMGIC (0% sage; control); Group 2, RMGIC with 2% sage powder by weight; Group 3, RMGIC with 5% sage powder by weight; and Group 4, RMGIC with 7% sage powder by weight. 25 flexural specimens were used for each group.

Lower, intermediate and high levels of incorporation were represented by the selected concentrations within the range of the experiments. Concentrations were determined by

weight against the powder part and it was prepared using an analytical balance. The study records did not include a comprehensive botanical authentication report, particle size distribution, moisture content or quantitative phytochemical profile of the sage powder. The factors mentioned are therefore recognized as formulation variables and not as standardized properties. When submitting to a certain journal, the product lot information, curing-light output and a verified nominal bar length should be verified by referring to the original laboratory records.

Table 1. *Experimental groups used for flexural-strength testing.*

| Group          | Sage concentration | Material                          | n  |
|----------------|--------------------|-----------------------------------|----|
| Control        | 0%                 | Unmodified RMGIC                  | 25 |
| Experimental 1 | 2% w/w             | RMGIC + Salvia officinalis powder | 25 |
| Experimental 2 | 5% w/w             | RMGIC + Salvia officinalis powder | 25 |
| Experimental 3 | 7% w/w             | RMGIC + Salvia officinalis powder | 25 |

**Specimen Preparation**

For the control, RMGIC powder and liquid were prepared as per the manufacturer's guidelines. The amount of sage powder needed was mixed into the RMGIC powder before the addition of liquid for each experimental group. The weighing process was very critical as it could alter the concentration if the mass of the powder is misweighed, which could in turn affect the powder-to-liquid relationship. The same general sequence of manipulation was employed in each group to reduce the variation due to the operators and mixing.

Flexural testing specimens were prepared in split moulds in the rectangular shape. The mixed material was placed in the mould, and covered with a strip of cellophane or glass surface to ensure a smooth specimen face and then light-cured with an LED curing unit following the study protocol. Excess material was removed after the cure and demoulding process and specimens were visually inspected. Obviously cracked, void, bubble specimens as well as those with irregular margins and unacceptable dimensions were excluded as possible artificial fracture origins. Individual specimens were measured for width and thickness and the results were incorporated into the strength calculation. A support span of 21 mm was recorded on the laboratory spreadsheet for each flexural test, however, the actual nominal mould length should be confirmed with the original equipment record since the thesis narrative included varying dimensional terminology.

Specimens which were accepted were stored at 37°C in distilled water for 24 hours prior to being tested. Early hydration, resin post-curing and glass-ionomer maturation could be performed in this condition while achieving a standardised short

term end point. It was not a long-term exposure to saliva, changes in temperature, acid challenge or cyclic mastication. The specimens were prepared as microhardness specimens – described in the study synopsis as approximately 9.5 mm diameter and 1 mm thickness – for a flat surface on which the indenter would be used.

### Flexural-Strength Testing

The flexural strength was determined by three points bending in Instron universal testing machine. Two supports were used and loading was done in the center of each bar up to failure. The support span was 21 mm and the crosshead speed was 0.5 mm/min. The fracture load (F) was measured in Newtons. The width, b, and thickness, h, of the individual specimens and flexural strength, sigma, were measured in millimetres and megapascals, respectively, and calculated as follows:  $\sigma = 3FL/(2bh^2)$  where L is the support span. The thickness of the specimens was raised to the power of 2 in the denominator, making it especially crucial to accurately measure the dimension of the specimens and to correctly orient the specimens.

The three-point bending test was chosen due to the fact that the test applied tensile stress to the lower surface of the specimen while the upper surface of the specimen was put under compressive stress. Therefore voids, roughness, poor particle-matrix integration and other defects may not be as visible in a compression test and may be more visible in this test. Fracture calculation was validated with the recorded load and dimensions during data cleaning.

### Exploratory Microhardness Assessment

According to the study synopsis Vickers microhardness was to be measured in a diamond indenter under a load of 100 g for a dwell time of 15 s. In this technique a disc specimen is focused under the microscope and indented under a controlled load on the tester stage. The length of the diagonals of the resulting indentation are measured and the Vickers hardness value is calculated. If multiple indentations are averaged within each individual specimen, and if the statistical analysis is to be made at the level of the specimen mean rather than each indentation, the ideal approach would be to take the average of the multiple indentations to obtain the specimen mean.

Those were the only bench-note readings for the present manuscript - four for the control group, four for the 2%, seven for the 5%, and ten for the 7%. The notes used the labels of 0.02%, 0.05% and 0.07% for the experimental groups, which were understood in the thesis as representing the proposed 2%, 5% and 7% formulations. The numbers in each group were not complete or balanced and all documentation was not available for the specimens planned for each group, so no inferential comparison was

made. Values have been summarised just to provide direction and range of available observations.

### Data Management And Statistical Analysis

The spreadsheet for flexural strength had sample identifier, group, concentration, test type, width, thickness, support span, applied load and calculated strength. The data were subsequently summarized and discarded, in addition to being empty, any row that was not a flexural-strength observation. Counting was done for group counts and numerical fields. A balanced four group design was used with 25 observations in each group in the final data set.

Sample size, mean, standard deviation (SD), median, minimum, maximum and 95% confidence interval (CI) were determined for each group. The Shapiro-Wilk test was used to evaluate the normality in each group separately. Levene's test was used to test for homogeneity of variance. One-way analysis of variance was employed as the main comparison of the four group means, as the assumptions were satisfied. Additionally, a non-parametric sensitivity analysis was conducted using a Kruskal-Wallis test. Standardised mean differences (Cohen's d) were computed for pairwise independent-samples t tests with Bonferroni correction. A level of statistical significance of  $p < 0.05$  was used. Descriptive statistical measures only were used to summarise the microhardness bench-note data.

### Ethical Considerations

The experiment did not include human subjects (indistinct persons, patient information), or animals. The thesis stated that an IRB was obtained or not through the institution. No approval or exemption identifier is included in the manuscript supplied and should be added before the manuscript is submitted to the journal. The experimental material was not used in clinical practice and the results were not interpreted as evidence of safety of the patients or clinical effectiveness.

## 3. Results

### Flexural-Strength Dataset And Descriptive Findings

A total of 100 observations (25 control and 25 trials for each concentration of sage) were included in the cleaned data. Mean flexural strength was 10.15 MPa in the control group, 10.44 MPa in the 2% group, 10.43 MPa in the 5% group and 10.32 MPa in the 7% group. The standard deviations were comparable for the two and ranged from 1.03 to 1.09 MPa. The experimental means were 0.17-0.29 MPa above the control means, but the differences were not great when compared to the within-group variation. The ranges of medians were similar and the ranges observed overlapped significantly.

All four groups were found to be clustered around the mean plot with SD bars. The 2% group had the highest mean while the 5% group was very close, the 7% group and the control group were the next highest. A boxplot also showed the variance in the IQR and a similar distribution. No experimental group showed an obvious increase or decrease in dispersion, which would suggest a large concentration related effect of weakening or strengthening.

Table 2. *Descriptive statistics for flexural strength by sage concentration.*

| Concentration | n  | Mean (MPa) | SD   | Median | Minimum | Maximum | 95% CI      |
|---------------|----|------------|------|--------|---------|---------|-------------|
| 0% (control)  | 25 | 10.15      | 1.09 | 10.12  | 8.62    | 12.05   | 9.69-10.60  |
| 2%            | 25 | 10.44      | 1.04 | 10.61  | 8.72    | 12.16   | 10.01-10.87 |
| 5%            | 25 | 10.43      | 1.03 | 10.53  | 8.60    | 12.03   | 10.01-10.86 |
| 7%            | 25 | 10.32      | 1.06 | 10.30  | 8.86    | 12.18   | 9.88-10.76  |

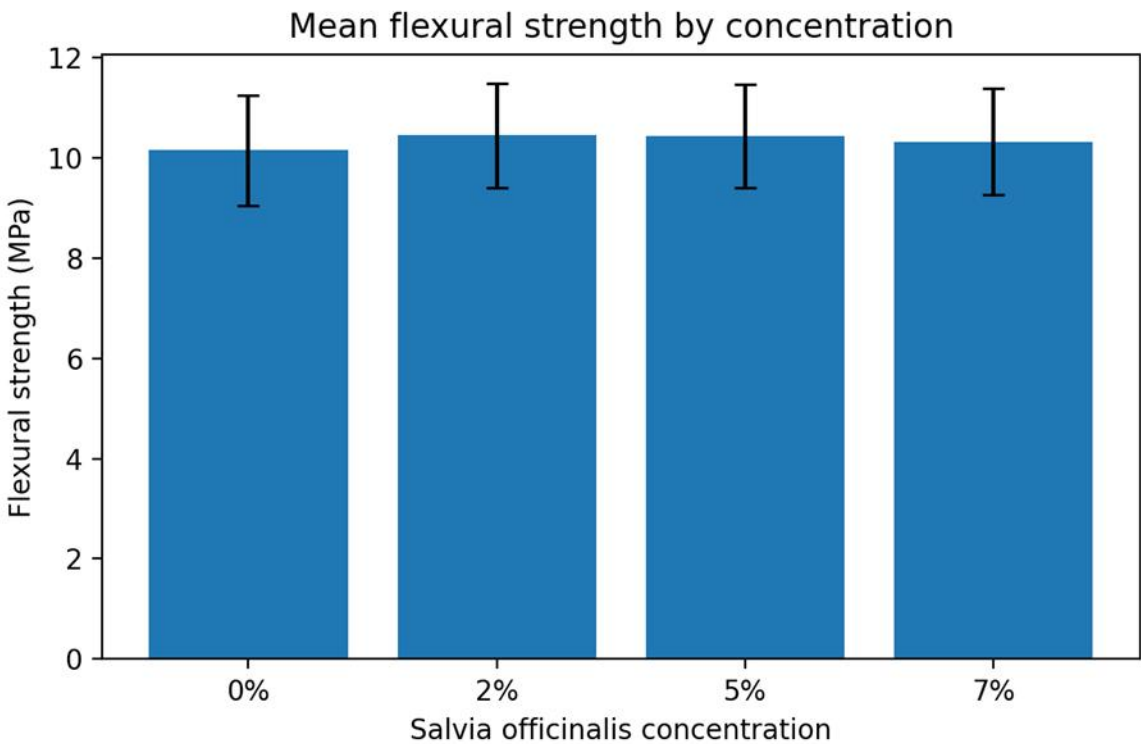


Figure 1. *Mean flexural strength by Salvia officinalis concentration. Error bars represent standard deviation*

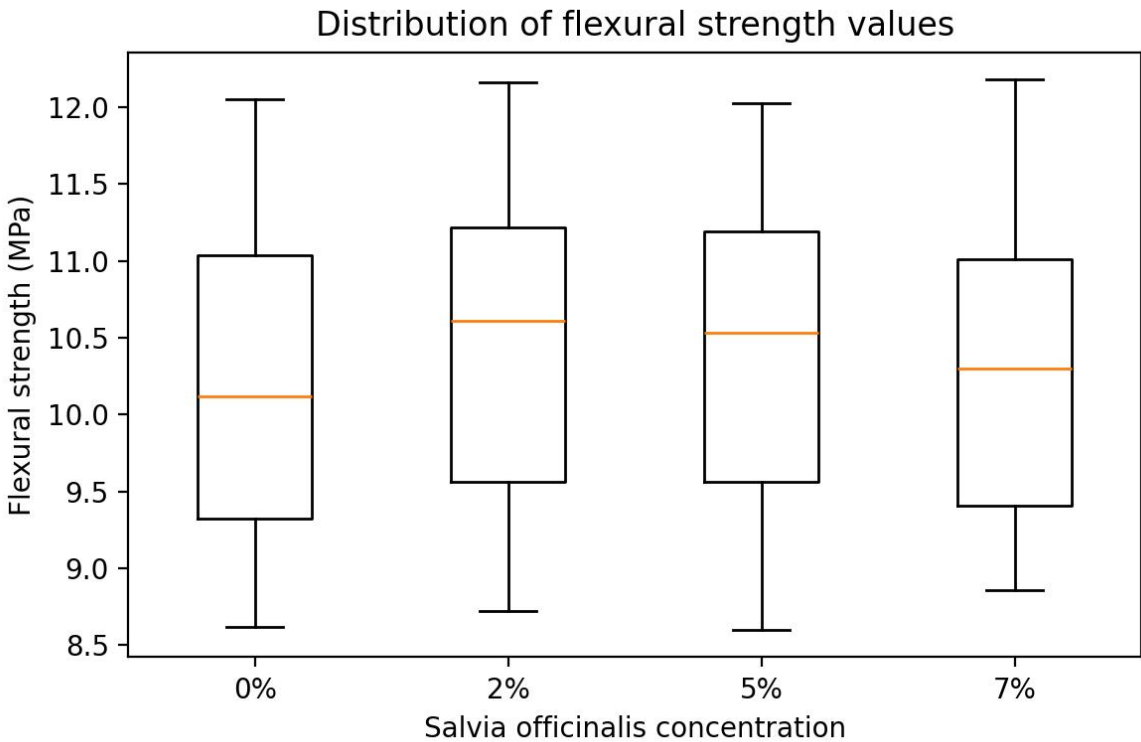


**Figure 2. Distribution of flexural-strength values by *Salvia officinalis* concentration**

**Distributional Assumptions And Overall Comparison**

Shapiro-Wilk p values were 0.089 for control, 0.095 for 2% sage, 0.351 for 5% sage and 0.072 for 7% sage. There was no statistical evidence of non-normality (the number was not below 0.05) within each group. The result of the Levene's test was 0.289, with p = 0.833, which provides a basis for assuming that there is homogeneous variance.

The one-way ANOVA yielded F = 0.430 with p = 0.732. The null hypothesis of Equal Mean flexural strength was therefore not rejected. A Kruskal-Wallis sensitivity analysis also proved to be non-significant (H = 1.339, p = 0.720), which was also consistent with the above interpretation, which did not rely on normality.

**Table 3. Assumption checks and overall comparisons for flexural strength**

| Analysis     | Group/statistic | Value     | P value | Interpretation                          |
|--------------|-----------------|-----------|---------|-----------------------------------------|
| Shapiro-Wilk | Control         | W = 0.930 | 0.089   | No significant departure from normality |
| Shapiro-Wilk | 2% sage         | W = 0.932 | 0.095   | No significant departure from normality |

|                |            |       |       |       |                                         |
|----------------|------------|-------|-------|-------|-----------------------------------------|
| Shapiro-Wilk   | 5% sage    | W     | =     | 0.351 | No significant departure from normality |
|                |            | 0.957 |       |       |                                         |
| Shapiro-Wilk   | 7% sage    | W     | =     | 0.072 | No significant departure from normality |
|                |            | 0.926 |       |       |                                         |
| Levene's test  | All groups | 0.289 | 0.833 |       | Homogeneity supported                   |
| One-way ANOVA  | All groups | F     | =     | 0.732 | Not significant                         |
|                |            | 0.430 |       |       |                                         |
| Kruskal-Wallis | All groups | H     | =     | 0.720 | Not significant                         |
|                |            | 1.339 |       |       |                                         |

Pairwise Comparisons And Effect Sizes

None of the pairwise comparisons were statistically significant. All six comparisons had Bonferroni adjusted p values of 1.000. The difference (Cohen's d) between control and 2%, 5% and 7% sage was -0.28, -0.27 and -0.16 respectively, all small differences favouring the sage treatments. The effect size ranged from 0.01 to 0.12 value when comparisons were made between experimental groups. The direction of the observed, therefore, did not provide any evidence that any concentration was mechanically superior. The main outcome was that all the formulations exhibited similar early flexural properties.

Table 4. *Pairwise comparisons for flexural strength*

| Comparison | t statistic | Unadjusted p | Bonferroni p | Cohen's d |
|------------|-------------|--------------|--------------|-----------|
| 0% vs 2%   | -0.991      | 0.327        | 1.000        | -0.28     |
| 0% vs 5%   | -0.953      | 0.346        | 1.000        | -0.27     |
| 0% vs 7%   | -0.561      | 0.577        | 1.000        | -0.16     |
| 2% vs 5%   | 0.045       | 0.964        | 1.000        | 0.01      |
| 2% vs 7%   | 0.430       | 0.669        | 1.000        | 0.12      |
| 5% vs 7%   | 0.388       | 0.700        | 1.000        | 0.11      |

Exploratory Microhardness Observations

The limited microhardness bench notes yielded a mean (SD) of 78.75 (2.50) for control, 80.00 (4.08) for 2% sage, 74.29 (4.50) for 5% sage and 83.30 (4.35) for 7% sage. The descriptive mean values of these seem to be lower at 5% than at 7%. The number of samples ranged from 4 to 10, however, an equal number of samples were not collected, and the records were not adequate to verify samples through testing. Therefore, the figure and table are indicative and are not intended to be used for claiming a concentration effect on the microhardness.

Table 5. Available microhardness bench-note readings (descriptive only).

| Group         | n  | Mean  | SD   | Median | Minimum | Maximum |
|---------------|----|-------|------|--------|---------|---------|
| Control/blank | 4  | 78.75 | 2.50 | 80.00  | 75.00   | 80.00   |
| 2% sage       | 4  | 80.00 | 4.08 | 80.00  | 75.00   | 85.00   |
| 5% sage       | 7  | 74.29 | 4.50 | 75.00  | 70.00   | 80.00   |
| 7% sage       | 10 | 83.30 | 4.35 | 85.00  | 75.00   | 90.00   |

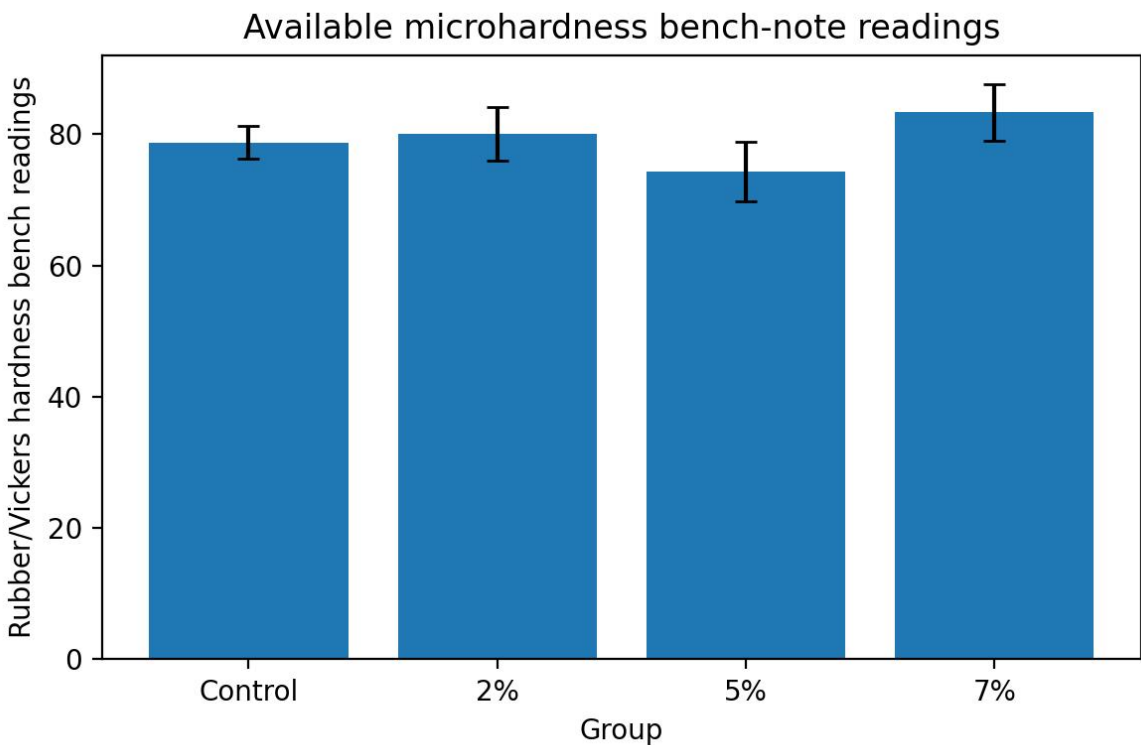


Figure 3. Available microhardness bench-note means with standard-deviation bars

4. Discussion

The main result of this study is that there was no statistically significant difference between the early flexural strength of the tested RMGIC when the *Salvia officinalis* powder was added in various concentrations, 2%, 5% and 7% of the total weight. The means for all groups were not significantly different from each other (difference < 0.3 MPa), the standard deviations were similar, and the distributions overlapped. The ANOVA and Kruskal-Wallis analyses both agreed and the pairwise effect sizes were small. These results do not allow the rejection of the null hypothesis in terms of flexural strength. They cannot be taken as proof of short-term mechanical compatibility in the

study conditions, but rather as proof of mechanical compatibility of the material in the study conditions within a short period of time.

The difference between these is significant for experimental study of dental materials. The mean of a modified group could be numerically higher than that of the control group, but the group may not show a reliable improvement. The mean of the 2% and 5% groups were greatest but not significant compared with the difference between specimens. All confidence intervals were overlapping. The data thus do not warrant selection of 2% or 5% for better strength but do suggest that there is no clear flexural penalty at either of these concentrations. Likewise, the 7% mean was not significantly lower than the control, and did not exhibit the marked loss of continuity in the matrix that might have been anticipated if the higher loading was to have caused severe disruption of matrix continuity.

The results are generally in line with the most directly comparable previous study. Forouzanmehr, Barekatin and Shahtalebi (2020) found that the compressive strength and shear bond strength of an RMGIC were not significantly different after short-term storage when the powder of sage was incorporated at concentrations of 0.5%-1.25%. The present work expands the range of concentrations and investigates another mechanical endpoint. The agreement with respect to the compression, bonding and flexural results indicates that sage can be used in at least some formulations of RMGIC, but it is not recommended to assume equivalence. This depends on the base material, the method of processing the plants into powder, the geometry of the specimens, the method used for curing the specimens, and the statistical power of the experiment; absence of a significant difference is not the same as the formal equivalence of the groups within a prespecified margin.

Flexural testing provides additional data which is not available from compressive testing data alone. In three-point bending, the specimen has the greatest tensile stress on one surface. Brittle or heterogeneous materials can fail due to defects on their surface or internal defects which grow in tension. If the compressive behaviour is relatively unchanged but the voids are created or the continuous area of the matrix is reduced due to the presence of organic particles, they may serve as weak inclusions. That this does not lead to a significant flexural reduction in the current set of data indicates that the sage particles, at the concentration and processing conditions used, did not cause a noticeable defect population severe enough to change the mean bending strength. This does not preclude very small changes in fracture toughness, fatigue behaviour and reliability not measured.

Preservation of flexural strength may be attributed to several mechanisms. First, the powder added could have been at a level that would not have caused a significant loss of continuity within the RMGIC matrix. It is about 7% higher than the previous sage-RMGIC research but still maintains the majority of the powder phase of the original restorative material. Secondly, the particles may not have been large enough to form clusters. Third, the decrease in reactive glass and/or the setting change may have been small compared to normal variation of hand-mixed specimens. Lastly, there is a limited packing or void-filling effect at lower concentrations. It is impossible to differentiate between these explanations using the data presented here, as particle-size analysis, microscopy, porosity measurement and chemical characterisation were not performed.

Caution should be exercised with the concentration pattern. The means increased from control to 2%, were only slightly changed at 5%, and decreased slightly at 7%, but all remained significantly higher than control. This is similar to the pattern that could be expected if there were an intermediate optimum and a slight decrease at higher levels of filler. Poranek et al. (2025) saw a concentration-dependent performance of the filler, derived from sage, in polyurethane composites, as well as an improved performance at certain lower and intermediate concentrations when there was more agglomeration. This work is related to a different matrix, and therefore cannot confirm a mechanism in RMGIC. It does, however, illustrate the reasons for studying dispersion and interfacial structure instead of making the assumption that the response to the concentration of plant-powder will be linear.

Simple predictions are also complicated by the chemical and physical complexity of RMGIC. The early properties are related to light-induced resin polymerisation and the acid-base reaction and ionic maturation takes place over time. An additive may have a number of effects. They can absorb water, scatter or attenuate curing light, affect the actual powder-to-liquid ratio or react with the acidic material. On the other hand, if the hydroxyl- and carboxyl-containing phytochemicals are present at low levels, they will not affect the network. The literature review of thermal and setting properties of resin containing glass-ionomer materials shows that they are dependent on each other and not independent (Park and Kang, 2020; Aleksandra Maletin et al., 2023). The current flexural measurement is an end-point measurement and cannot show if the underlying setting kinetics was unaltered or not.

Another probable factor is the standardisation of the mixing. The properties of glass ionomer are highly dependent on the powder-to-liquid ratio, the volume of air and the operator's technique. The amount of powder can affect handling and mechanical reliability – as demonstrated by Mulder (2018) – and the dispensing and

mixing systems can affect a number of properties of RMGIC luting materials – as shown by Sulaiman et al. (2018). The solid composition changed in the current study whenever sage powder was added, although this change was not always necessary, and may have influenced the ratio of reactive glass to liquid. The comparable standard deviations indicate that the procedure introduced very little extra source of variance however, working time, viscosity and void volume were not measured in the study. It is suggested that mixing time, curing-light irradiance and powder mass for each batch should be recorded for future work.

The absolute flexural-strength values should be regarded in the context of the specific study, and not compared directly to values reported for other commercial products that were not part of the study. Rêgo, Butler and Santos (2022) and Bilge, Aşar and İpek (2024) found significant variations between various glass-ionomer-based restorative materials, which were due to the proprietary chemistry, filler technology, resin used and specimen preparation methods. Strength may also vary due to differences in bar geometry, loading rate, and storage duration and support span. The internal purpose of the current control was thus to compare all the sage groups with the unmodified version of the same material prepared and tested in the same laboratory. The dimensions and material lot information of the specimen should be checked prior to external publication for exact reporting, which is necessary for reproducibility.

During the exploration microhardness observations a definite conclusion cannot be drawn. The available means were not monotonic in that the 5% group was less than the control group, while the 7% group was greater than the control group. This trend may represent a genuine response of the material, or it may be due to the surface variation or to sample instability due to the small and unequal groups. Microhardness is a surface preparation, indenter site, resin conversion, water exposure, and ongoing acid-base maturation sensitive property. The surface hardness and its roughness changes as a function of material composition and environmental exposure are shown in Biancalana et al. (2019) and Saadat, Moradian and Mirshekari (2021). Data must be balanced, having verified Vickers values for each of the desired specimens, before statistical comparison can be made.

The hardness analysis is completed, as flexural strength and microhardness are different aspects of the performance of the material. A formulation can be highly bend resistant and soft, or it can be extremely surface hard, but be prone to crack propagation. De Mendonça et al. (2021) reported that the hardness can be different at various depths and light penetration for restorative materials, while Ivica et al. (2024) indicated that some mechanical properties can be enhanced with an additive, and

hardness and/or ion release can be changed. The short-term bending strength may not change significantly, but the optical transmission or water uptake performance of sage powder may change. Therefore, the results obtained in the current flexural test must not be substituted for a comprehensive evaluation of surface properties.

The biological basis for the use of sage is also not tested in the present specimens. The extracts of *S. officinalis* possess several bioactive compounds and studies have indicated antimicrobial or antifungal activity in oral health-related applications (Ghorbani and Esmaeilzadeh, 2017; Shahriari et al., 2019; Fadhil et al., 2024). But the delivery system of incorporation into a cured cement is different. Active constituents need to be stable, be released or act at the surface of the material, and attain a biologically relevant concentration. The diffusion might be limited by resin polymerisation and the glass-ionomer matrix and/or over time might be affected by leaching. The mechanical preservation is thus a first requirement and not proof of antimicrobial activity.

The next steps would be to perform the same testing with the 0%, 2%, 5% and 7% formulations against cariogenic organisms in both release-based and surface-associated models. Agar diffusion can be used to show whether soluble compounds will be able to pass through the agar or not, but will favour agents that diffuse well and is not a recognised biofilm. A greater amount of biomass in the biofilm, as well as viable counts and metabolic activity on the specimen surface would constitute more compelling evidence. Multispecies models are more clinically relevant, while *Streptococcus mutans* and *Lactobacilli* are logical targets as initial targets. Testing at multiple ages would determine if there is a measurable effect over time or if the effect fades as soluble materials are used up.

The same time should be used to evaluate biological safety. Safety cannot be assumed with natural origin and some constituents of sage essential oil and phenolics may be concentration-dependent in terms of their effect on pulp, gingival or oral cells. Before clinical use, study of the cytotoxicity of eluates, the release kinetics, colour stability and taste and allergy related issues should be performed. Formulation evaluation should also include determination of fluoride release, water sorption, solubility and dimensional stability. If plant components leach from the material or the resin phase plasticises after long water exposure, it may change after 24 hours.

For RMGIC, ageing is important due to the changes in properties that occur following the initial light curing. Some properties may be enhanced by continued ionic maturation, others by water uptake, hydrolysis and repeated thermal stresses. Early mechanical compatibility is the only endpoint currently available for the 24-hour

endpoint. More realistic challenges would be extended water or artificial saliva storage, thermocycling, pH cycling and mechanical fatigue. Multiple hardness tests could be used to determine the impact of sage on surface degradation or on maturation. If the groups were equal at the beginning, then fracture testing after ageing would reveal if the groups differentiate over time.

Mechanical findings would be explained by microstructural analysis. The distribution of particles, agglomeration, voids and crack paths could be revealed in the scanning electron image of polished and fractured surfaces. Energy-dispersive analysis may be able to differentiate plant inclusions from glass particles and micro-computed tomography may allow the measurement of internal porosity. Spectroscopic techniques could be used to determine whether there are chemical interactions between the matrix and sage constituents. Reproducibility would be enhanced by particle size measurement and by having a documented sieving or milling procedure. If these studies are based on the same percentage by name, they could be testing different physical forms of the same additive.

The statistical interpretation of the data should be highlighted. It is not a conventional null-hypothesis test that asks whether the observed data provide sufficient evidence of a difference; in fact, it does not prove that the materials are equivalent. The design included 25 samples per group which allowed the description of the central tendency and variability within the available samples, but did not have a priori minimum clinically important difference or equivalence margin defined. Equivalence or non-inferiority methods may be used for a future confirmatory study, with a pre-specified maximum reduction in flexural strength. This method would respond to a more clinically relevant question – does a sage formulation maintain strength in a range that is deemed acceptable for the intended use?

Even if there were not equivalence testing, the range of the small differences is informative. Control vs sage Cohen's  $d$  values ranged from -0.16 to -0.28, and differences between the experimental concentrations were close to zero. Such estimates contradict a significant short term impact, either positive or negative. However, they were not given in the thesis and a small mean effect at 24 hours may turn into a more significant effect during ageing or re-loading. Confidence intervals for effect sizes would provide greater precision of replication and aid the assessment of whether there was a truly negligible effect or whether it was poorly estimated.

Another key to reproducibility is to clearly document the geometry of specimens and calculation pathway. The degree of flexural strength depends on support span, width, thickness and surface quality and thickness is squared. Thus, a slight error in the

measurement can affect the derived value. A strong point of the individual-dimension approach utilized in the spreadsheet is a journal article should report mould dimensions, dimensional tolerance, measurement instrument, loading-nose and support radii, specimen orientation and edges finished or polished. This information enables another laboratory to reproduce the stress distribution and not simply take the same formula.

Other clinically significant effects of sage addition were not dealt with here. The color, translucency, radiopacity, working time, setting time, viscosity, and surface texture of the plant powder may change, or it may be impossible to get a void-free restoration. This can impact on acceptance and placement despite maintaining flexural strength. The opaqueness or darkness of a visible cervical restoration may not be desirable, but may be more important if placed in a liner or base. On the other hand, changed handling might have an effect on porosity, which would affect strength in clinical use. Objective colour measurement, radiopacity, handling assessment and surface-roughness testing should be incorporated into future formulation studies, together with mechanical outcomes.

This "best group" should be determined by using a multi-criteria approach, not by choosing the group with the highest average score on one test. One formulation might be more effective in terms of antimicrobial activity, but less effective with respect to colour or bond strength; another formulation might have high bond strength, but low antimicrobial activity. In a rational development programme, concentration ranges are first screened and candidates with suitable characteristics (e.g. flexural strength, hardness, adhesion, fluoride release, cytocompatibility and antimicrobial activity) are then selected. The chosen formulation may then be further subjected to ageing, fatigue, animal-free biological testing prior to any clinical feasibility test. This staged approach minimises the risk of pushing a formulation because only one favourable endpoint is recorded.

The need for transparent reporting is particularly relevant due to the use of a thesis dataset that was compiled from spreadsheets, images of the equipment and bench notes. The flexural file is raw while the microhardness documentation is not. The authors are encouraged to archive the cleaned data set, data dictionary, statistical output, material batch records and original testing sheets before submitting. The analysis code or software settings should also be retained. If possible, de-identifying the data set and analysis file and providing them as supplementary material would be helpful for the editors and readers to verify the reported means and tests and would support confidence in the negative primary result.

There are a number of strengths with the study. It used a balanced flexural dataset with 25 observations per group, included three experimental concentrations and compared each formulation with an unmodified control from the same base material. The dimensions of each individual specimen were used in the flexural calculation, and parametric and non-parametric overall tests yielded the same results. Reporting effect size in addition to a non-significant p value minimized the risk of concluding that the formulations were the same. The figures and descriptive statistics give a clear picture of the close group distributions.

There are significant restrictions and limits of interpretation. The work was done in vitro and examined at an early, 24-hour time period. The botanical identity, moisture content, particle-size distribution and phytochemical composition of the sage powder were not standardized. Actual bar length and a few product and curing aspects must be verified from the original laboratory record. Microscopy, porosity, fracture toughness, fatigue, wear, bond strength, fluoride release, water sorption and antibacterial activity were not evaluated. No formal sample size calculation or equivalence margins were available and thus non-significance cannot be used to infer equivalence. The most significant issue was that the microhardness data set was incomplete and unbalanced, which does not allow the drawing of any inferential conclusions.

These limitations also affect the claims and titles. The study includes a comprehensive analysis of flexural strength, whereas the analysis of microhardness is exploratory. The work is certainly not a definitive study of both properties. Hardness observations should be considered preliminary and flexural strength should be emphasized as the most scientifically defensible. The plan for a microhardness dataset may be feasible if the intended dataset is completed, allowing for a revision of the manuscript with both outcomes as primary or secondary endpoints or with distinct primary and secondary outcomes.

In terms of clinical-development, retention of flexural strength is good but not enough. Two issues that could be significant for sage-modified RMGIC, would be the ability to attain meaningful biological activity and to preserve the handling, adhesion, surface quality and durability. This Experiment shows that there was no detectable short term flexural disadvantage with 2%-7% sage. They don't specify an optimum concentration. In making a choice, the optimal would be determined by integration of mechanical, antimicrobial, cytotoxic, aesthetic and ageing data, not by the maximum numerical flexural mean.

In general, this study can be considered as a compatibility screening in the early stage of a staged material development program. In the next stage, all components must be

completely characterised, the specimen geometry is to be controlled, the curing process is to be controlled and the statistical analysis is to be planned, and the sage powder has to be fully characterised. Obviously, it should be flexible, it should include an antimicrobial test, bond strength, fluoride release, and ageing. Once one concentration exhibits these attributes and also demonstrates long-lasting antibiofilm properties with acceptable cytocompatibility, clinical relevance might then be pursued. In the absence of such evidence, it must be concluded at this time only that they are mechanically comparable in the current laboratory conditions.

## 5. Conclusion

In the limits of this in vitro study, 2%, 5% and 7% *Salvia officinalis* powder by weight did not significantly affect the 24-hour water storage flexural strength of the tested resin modified glass ionomer cement. Mechanical similarity to the unmodified control was consistently suggested by mean values, variability, distributional tests, and overall comparisons as well as by small effect sizes. The results are based on the short-term compatibility and not on reinforcing or equivalence. The observations performed in the microhardness had been very incomplete and unbalanced, so they are still exploratory. Prior to the clinical or commercial interpretation the formulation should undergo full microhardness testing, specimen and material documentation, botanical and particle standardisation, antimicrobial and cytotoxicity assessment, bond and surface-property testing, fluoride and water behaviour analysis and long term ageing.

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