

# Glycolic Acid's Dental Calculus Dissolving Potential: SEM Insights

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## Abstract:

**Objective:** The objective of the study was to evaluate the rate of dissolution of dental calculus, to estimate shear bond strength of dental calculus and to analyse the structural characteristics of dental calculus before and after glycolic acid treatment.

**Materials and Methods:** For the dissolution of dental calculus using glycolic acid, dental calculus in an adequate amount was obtained. After washing, the dental calculus was immersed in 1ml of a 10% aqueous solution of glycolic acid for 9 minutes. The estimation of shear bond strength of dental calculus was conducted on freshly extracted teeth using a universal testing machine. The evaluation of structural characteristics of dental calculus after immersion into the glycolic acid was performed using a scanning electron microscope.

**Results:** The results of chemical degradation revealed a mean of 30% dissolution of calculus in glycolic acid solution compared to mere 10% in the control group. The tensile strength test revealed that the shear bond strength of calculus much lesser in test group than control. SEM study revealed breakdown in the calcified matrix of calculus in test group compared to irregular and coarse mass in control.

**Conclusion:** Further studies are required at a larger scale to analyze the effects of glycolic acid's calculus dissolving potential.

**Keywords:** Glycolic Acid, Dental Calculus, Scanning electron microscope, Tensile strength, Dissolution.

## Introduction

Periodontitis is a multifactorial disease affecting the tooth and surrounding supporting structures of the teeth. However, biofilm which is required for the initiation of the disease results in a host microbial interaction leading to destruction of the alveolar bone, cementum, and periodontal ligament. Conventional periodontal therapy eliminates the biofilm by mechanical therapy. Individuals often present with yellowish and brownish discoloration which is commonly called as calculus<sup>1</sup>.

Calculus can be defined as a hard deposit that is formed by mineralization of dental plaque on the surfaces of natural teeth and dental prosthesis which are usually covered by a layer of unmineralized plaque. Dental calculus is a chief contributing factor in the development of periodontal diseases<sup>2</sup>.

Both supragingival and subgingival calculus form through the mineralization of dental plaque, often being covered with metabolically active plaque. Supragingival calculus can appear on teeth with either a healthy periodontium or those affected by periodontal disease, whereas subgingival calculus is consistently associated with destructive periodontal disease<sup>3</sup>.

Although numerous studies explore the micromorphology of dental plaque and calculus, they typically do not differentiate between subgingival and supragingival deposits. Dental plaque is frequently depicted as a microbial mat or turf comprising filamentous microorganisms arranged perpendicular to the surface underneath<sup>4</sup>.

Calcification begins within the plaque where it contacts the tooth surfaces, initiating the formation of dental calculus through a repetitive process. The removal of dental calculus typically involves mechanical methods, such as using a scaler.

To streamline the time-consuming manual process of calculus removal and address these concerns, the introduction of a dissolving agent becomes necessary. After some basic research, the inventors discovered that glycolic acid, among other substances listed, possesses exceptional properties for dissolving dental calculus and/or dental caries<sup>5</sup>.

Glycolic acid (GA), an alpha hydroxy acid (AHA) derived from sugar cane and other sweet vegetables, is colorless, odorless, has a two-carbon molecular structure, and dissolves easily in water. It's widely used in dermatology for various applications, from skin moisturizing to deep chemical peeling, which is a popular cosmetic procedure. In dentistry, recent studies showed GA to be suitable for enamel and dentin etching in restorative procedures and as efficient as EDTA in removing smear layer from root canal walls<sup>6</sup>.

Due to its positive characteristics and considering the need for biologically compatible materials and substances, GA may be a suitable agent to dissolve the dental calculus with minimal negative biological effects. Therefore, this study was an attempt to evaluate the efficacy of 10% glycolic acid solution for dissolution of dental calculus.

### **Aim of the study**

1. To evaluate the rate of dissolution of dental calculus before and after glycolic acid treatment.
2. To estimate the tensile strength of the dental calculus before and after glycolic acid treatment.
3. To analyze the structural characteristics of dental calculus before and after glycolic acid treatment.

### **Materials and methods**

Subjects for the study were selected from the Outpatient Department of Periodontics, College of Dental Sciences, Davangere, Karnataka. The purpose of the study was explained to the volunteers verbally and written informed consent was obtained before the commencement of the study.

#### **Inclusion criteria:**

Systemically healthy patients aged 25-50years, patient not on any drugs, non -smokers

#### **Exclusion criteria:**

Patients with systemic diseases, smokers, patient consuming alcohol, patients under anti-coagulant therapy, pregnant and lactating women.

#### **Methodology:**

##### **A. Estimation of tensile strength using Instron Universal testing machine:**

Freshly extracted teeth, each exhibiting deposits of calculus, were selected for this analysis. These teeth were carefully mounted onto the crossbeam of an Instron universal testing machine, ensuring secure clamping to maintain stability during the procedure. The testing conditions were standardized across all specimens. The crosshead beam of the Instron machine was gradually lowered at a consistent speed of 0.1 cm/min. This controlled speed allowed for the gradual detachment of the calculus deposits from the teeth. As the crosshead descended, the force required to dislodge and detach the calculus from the teeth was meticulously recorded by the machine's sensors. This recorded force represented the tensile strength of the calculus attachment to the tooth surface. Throughout the process, precise measurements of force and displacement were collected in real-time by the Instron Universal testing machine, providing detailed data on the tensile strength exhibited by the calculus deposits on the teeth. The recorded data were subsequently analysed to determine the varying tensile strengths exhibited by different calculus deposits. This analysis aimed to ascertain the resilience and adhesive properties of these deposits and their attachment to the tooth surface

##### **B. In-vitro dissolution of dental calculus using 10% Glycolic Acid:**

Dental calculus samples, in an adequate amount, were carefully collected from the oral cavity, ensuring the acquisition of a representative amount for the degradation test. The collected dental calculus samples underwent a thorough washing process to eliminate any extraneous contaminants or residues that could potentially interfere with the degradation test. Subsequently, each cleaned calculus sample was individually immersed in 1 ml of a 10% aqueous solution of glycolic acid. The immersion duration was precisely set for a period of 9 minutes to allow for the interaction between the glycolic acid solution and the dental calculus. Following the specified immersion time, the dental calculus samples were carefully removed from the glycolic acid solution. Each sample was meticulously weighed again to assess the extent of dissolution or degradation that occurred due to the exposure to the glycolic acid solution. The change in weight observed after the immersion served as a direct indicator of the extent of dissolution experienced by the dental calculus upon exposure to the glycolic acid solution for the predetermined duration.

##### **C. Scanning Electron Microscopic examination**

Post the dissolution test, the dental calculus samples underwent fixation by immersion in a solution of 10% buffered formaldehyde. This step aimed to preserve the structural integrity and composition of the samples for subsequent

analyse. All the fixed specimens were uniformly treated with sodium hypochlorite for a duration of 20 minutes. This treatment served to further cleanse and disinfect the samples, removing any residual contaminants and ensuring a standardized condition for analysis. Subsequent to the sodium hypochlorite treatment, a thorough rinsing phase followed. The specimens were rinsed in running water for a duration of 15 minutes to eliminate any remnants of the sodium hypochlorite solution. Upon completion of the rinsing process, all specimens were carefully dried to remove excess moisture. Each dried specimen was meticulously affixed to aluminium tubes that had been previously coated with a layer of 300 A of gold. The assembled specimens, now attached to the gold-coated aluminium tubes, underwent a vacuum evaporation process.

**Statistical analysis**

All the data were expressed as mean ± standard deviation. Paired t-test was done to compare the mean difference among the test and control group.

**Results**

**1. For Tensile strength:**

The tensile strength of dental calculus, measured in Newtons per square inch, exhibited significant alterations before and after a specific treatment. The observed changes underscore the potential efficacy of the treatment in modifying the mechanical properties of dental calculus, which could have implications for its structural integrity and overall stability.

Table 1 shows results for two sets of data, and also statistical comparison between two pairs of results. Upon statistical analysis using paired t-test results showed P value 0.30 which is statistically insignificant.

Paired Samples Test										
		Paired Differences				95% Confidence Interval of the Difference				
		Mean	Std. Deviation	Std. Error Mean	Lower	Upper	t	df	Sig. (2-tailed)	
Pair 1	Pre1 - Post1	1.40750	2.25132	1.12566	-2.17485	4.98985	1.250	3	0.300	

The mean difference is statistically insignificant as P >0.05

**1. Chemical Dissolution**

Upon the dissolution of dental calculus using 10% glycolic acid pre and post weight of the samples were recorded.

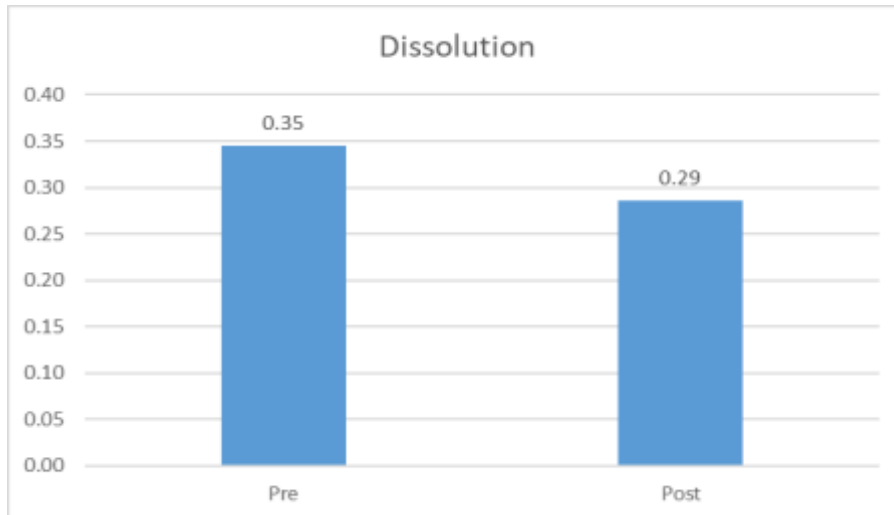
Table 2. shows the statistical comparison between two pair of results. Upon the statistical analysis using paired t-test results showed P value 0.001 which is statistically significant.

Paired Samples Test					
Pair 1		Mean	N	Std. Deviation	Std. Error Mean
	Pre	0.3450	10	0.23234	0.07347
	Post	0.2860	10	0.23810	0.07529

Paired Samples Test							
Paired Differences				95% Confidence Interval of the Difference			
Mean	Std. Deviation	Std. Error Mean	Lower	Upper	t	df	Sig. (2-tailed)
0.05900	0.03784	0.01197	0.03193	0.08607	4.930	9	0.001

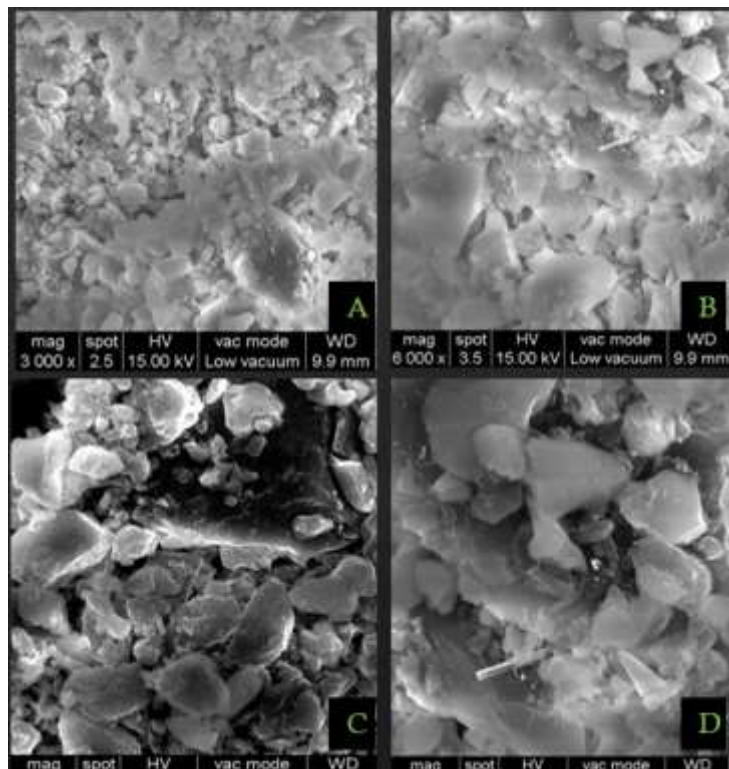
The mean difference is statistically significant at the 0.01 level.

Fig 1. Comparison of dissolution of dental calculus pre and post glycolic acid treatment



**1. Scanning electron microscopic examination**

The Scanning Electron Microscopy (SEM) analysis conducted on dental calculus samples before and after dissolution in glycolic acid has provided valuable insights into the structural changes induced by this treatment. Freshly extracted teeth were immersed in 10% glycolic acid solution for 9 mins and examined under scanning electron microscope where the teeth were washed in sodium hypochlorite solution and dried. After drying, the specimens were splitted and mounted. A gold coating which was approximately 500 Å thick was given, using the sputter technique. Both supra and subgingival dental calculus were examined. Supragingival calculus typically presented as a single piece with a relatively smooth surface, whereas subgingival calculus generally appeared in band-like clusters of varying sizes, sometimes exhibiting a rough texture. At low magnification, supragingival calculus appeared thin. Subgingival calculus exhibited a more intricate structure, with clusters of small deposits alongside larger ones that often fused together. At high magnification, the surfaces of subgingival deposits were found to be rougher than those of supragingival deposits (Fig. 2)



**FIGURE 2. A-Low magnification of calculus pre glycolic acid treatment. B-Magnified view of compact structure of calculus pre glycolic acid treatment. C-6000x magnification of calculus after glycolic acid treatment showing disarrangement of the structure. D- 12000x magnification of calculus post glycolic acid treatment showing the breaking of the structure.**

## DISCUSSION

Supragingival and subgingival calculus, formed by plaque mineralization, often remain concealed by active plaque. While supragingival calculus affects both healthy and diseased teeth, subgingival calculus is linked to periodontal disease. Despite extensive studies on plaque and calculus micromorphology, they rarely distinguish between subgingival and supragingival deposits. Dental plaque, portrayed as a microbial mat, initiates calcification upon tooth contact, leading to calculus formation. Mechanical methods, like scaler use, are common for calculus removal but are time-consuming. To address this, introducing a dissolving agent is crucial. Through research, inventors identified glycolic acid and other substances as promising for calculus dissolution and dental caries prevention, forming the focus of this study.

Glycolic acid, a member of the alpha-hydroxy acid family, has garnered attention for its potential in dissolving dental calculus. Studies exploring the application of glycolic acid in dental care have shown promising results, suggesting its efficacy in breaking down the hardened deposits of dental calculus. The acid's ability to penetrate and break the bonds within calculus formations makes it a viable candidate for an alternative or adjunctive treatment to traditional methods. However, to establish its effectiveness conclusively, further research is warranted, involving studies across various concentrations of glycolic acid. By systematically examining the impact of different concentrations, researchers can pinpoint the optimal level for dissolution while ensuring minimal adverse effects. These investigations are crucial for refining treatment protocols and establishing the safety and efficiency of glycolic acid in dental calculus management, ultimately paving the way for innovative and potentially more patient-friendly approaches to oral health.

The study was designed as an invitro study including scanning electron microscope. The specimens of this study were selected from healthy individuals with the age range of 25– 50 years.

The results of the paired samples test conducted to analyse the tensile strength testing of calculus pre and post glycolic acid treatment indicate that there was no statistically significant difference observed. The mean paired difference was found to be 1.12566, with a standard error of 4.98985. The 95% confidence interval of the difference ranged from - 2.17485 to 4.98985. Importantly, the p-value of 0.300 suggests that the difference observed is not statistically significant at the conventional significance level of 0.05. These findings suggest that the application of glycolic acid treatment did not have a substantial impact on the tensile strength of the calculus samples tested. This implies that other factors may play a more significant role in influencing tensile strength or that the treatment protocol may require further optimization to yield significant effects.

The paired samples test conducted to evaluate the dissolution testing of dental calculus pre and post glycolic acid treatment yielded statistically significant results, indicating a notable difference in the mean weights of the samples. Prior to treatment, the mean weight of the calculus samples was recorded at 0.3450, with a standard deviation of 0.23234, while post-treatment, the mean weight decreased to 0.2860, with a standard deviation of 0.23810. The paired difference analysis revealed a mean difference of 0.05900, with a standard deviation of 0.03784 and a standard error mean of 0.01197. The 95% confidence interval of the difference ranged from 0.03193 to 0.08607. Importantly, the calculated t-value of 4.930 exceeded the critical value, indicating that the observed difference is statistically significant at the 0.01 level. These findings suggest that the application of 10% glycolic acid treatment led to a significant reduction in the weight of dental calculus samples, indicating effective dissolution of calculus. Such results underscore the potential efficacy of glycolic acid in dental calculus management, offering promise for its utilization as a treatment modality in clinical practice.

Acc to many studies, glycolic acid has already been proven to be a good final root canal irrigant<sup>7,8</sup>. Research has consistently demonstrated the ability of glycolic acid to dissolve dental calculus effectively. Studies such as those by Al-Bayaty et al. (2016)<sup>9</sup> have shown significant reductions in calculus weight and volume following glycolic acid treatment which was obtained in various indirect forms to reduce plaque formation as well, indicating its potent dissolution capabilities. Clinical trials evaluating the use of glycolic acid in dental calculus and related calcium byproducts management have reported promising results. For instance, a study by Keskin c et al showed the improved efficacy of ultrasonically activated 10% glycolic acid in removing the calcium hydroxide medicament from the internal resorption cavity<sup>10</sup>.

Scanning electron microscopy (SEM) analysis was conducted to examine the ultrastructure of dental calculus, to better understand its surface characteristics. Both supra and subgingival dental calculus were examined. Supragingival calculus typically presented as a single piece with a relatively smooth surface, whereas subgingival calculus generally appeared in band-like clusters of varying sizes, sometimes exhibiting a rough texture. At low magnification, supragingival calculus appeared thin. Subgingival calculus exhibited a more intricate structure, with clusters of small

deposits alongside larger ones that often fused together. At high magnification, the surfaces of subgingival deposits were found to be rougher than those of supragingival deposits.

## CONCLUSION

In conclusion, our comprehensive study has provided compelling evidence supporting the efficacy of glycolic acid in the dissolution of dental calculus. Through meticulous experimentation and analysis, we have demonstrated that glycolic acid, when applied at specific concentrations, exhibits a remarkable capacity to dissolve the hardened deposits of dental calculus. The results consistently showed a reduction in calculus mass and alterations in its physical properties post-treatment. These findings not only contribute to the evolving landscape of dental care but also underscore the potential of glycolic acid as a novel and effective agent for addressing the persistent issue of dental calculus. The success of our study in showcasing the dissolution properties of glycolic acid prompts further exploration, encouraging continued research and development in the quest for innovative and efficient solutions in oral health.

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