

Dental Plaque Dissolving Agents: An In Vitro Study

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Abstract

Introduction: Dental plaque is an organic material, mainly comprising the polysaccharide dextran. There are some organic solvents which might be able to dissolve the plaque dextran with minimum harm to the body along with effective plaque reduction. This in vitro study aims to explore the effectiveness of two agents in dissolving dental plaque.

Methods: The study used 4% acetic acid, 50% glycerine and glass distilled water without adding any solute as control solvents. The same solvents by adding pure dextran at different concentrations were used as standard solutions. Test solutions were prepared by mixing equal amount of collected pooled dental plaque with the same solvents. Phenol sulphuric acid method was used to estimate the level of dissolved dextran. Multi calibration graphs were plotted using standard solution and dissolution of plaque dextran was estimated by extra plotting from graph as well as by factor (slope) calculation.

Results: Both calculation and graphical methods showed similar results, where glass distilled water showed 14.80 %, vinegar (acetic acid 4%) showed 25.47 %, glycerine showed 18.79% dissolution of plaque dextran respectively; when it was used at 50% concentration.

Conclusion: All the three solvents showed substantial dissolution of plaque dextran. Further in vitro and in vivo studies have to be performed for confirmation and clinical implication.

Keywords: Acetic Acid, Dental Plaque, Glycerin, Solvents.

Introduction:

Dental plaque is defined clinically as a structured, resilient yellow-greyish substance that adheres tenaciously to the intraoral hard surfaces, including removable and fixed restoration.¹ It comprises of microorganisms and intercellular matrix which are associated with a range of oral diseases including dental caries and periodontal diseases. Elimination of dental plaque is considered as basic requirement in preventing these diseases.²⁻⁵

Mechanical plaque control and chemical plaque control are two methods of plaque control. Mechanical plaque control involves tooth brushing, flossing, oral prophylaxis etc. whereas chemical

plaque control mainly involves antibacterial agents, used as an adjunct. The most common oral hygiene aid used to improve the oral health of an individual is toothbrush.³⁻⁶

Although tooth paste and brush has effective role in plaque control, it has certain limitations like technique sensitive, expensive, difficulty to use for physically and mentally handicapped persons, and can cause abrasion of the tooth and recession of the gingiva.⁷ There exists a chance of cross contamination of toothbrush when it is exposed to the oral cavity and external environment and these contaminated toothbrushes might play a role in systemic and oral diseases.⁸⁻¹⁰

Chemical plaque control agents mainly concentrate on antimicrobial properties. They reduce the plaque formation by bactericidal action. However, it also has side effects and it prohibits the normal commensals along with the pathogenic microorganisms. Thus, there is need to think of an alternative measure for effective plaque control.

Dental plaque consists of bacterial population and dental plaque intercellular matrix. Bacteria involve both aerobic as well as anaerobic form and dental plaque intercellular matrix is made up of organic and inorganic components. Dental plaque bacteria produce dextran by utilising sucrose in the oral cavity, which helps the microorganisms to adhere to the host tissue. It is hypothesized that if dextran is removed effectively, dental plaque will not be able to adhere to the tissues. Thereby effective plaque control may be achieved.

Dextran is a complex, branched glucan (polysaccharide made of many glucose molecules) composed of chains of varying lengths (from 3 to 2000 kilo Daltons) and it is soluble in solvents like methyl sulphide, formamide, ethylene glycol, glycerol.¹¹ Dextran can also hydrolyzed with acids.¹² Organic solvents like glycerol and acetic acid may have the property to dissolve the plaque dextran.

Distilled water, glycerol and acetic acid are known domestic substances and polar solvents for day to day use. Dextran is a polar compound and it dissolves well in polar solvents based on the rule of thumb of solubility.¹⁴ Thereby aim of this study is to assess the efficiency of these solvents on plaque dextran.

Material and Methods:

The study was conducted in the Department of Public Health Dentistry, SDM College of Dental Sciences & Hospital, Dharwad, India and the Department of Biochemistry, SDM College of Medical Sciences & Hospital, Dharwad, India.

A pool of dental plaque was collected separately using a Columbia scaler in a test tube containing 2ml of chilled glass distilled water and was sent to the Department of Biochemistry where it was sedimented by cold centrifugation at 5000rpm for 5 minutes and preserved at -20° Centigrade temperature. Three

solutions were prepared-control, standard and test solutions.

Biochemical procedures:

Standard solution preparation

Pure dextran[‡], molecular weight (MW) 10,000 (REF – RM736-5G) was obtained. Various standard solutions of 10 mg%, 20 mg%, 30 mg%, 40mg%,50mg% and 60 mg% of standard pure dextran was prepared in glass distilled water, 4% acetic acid and 50% glycerine. Glass distilled water, 4% acetic acid and 50% glycerine in glass distilled water taken as controls.

Test solution preparation

The pool of dental plaque samples were re-suspended in chilled distilled water and centrifuged for 5000 rpm at 5 – 10⁰ Centigrade for 5 minutes. Supernatant was discarded and sediments of pooled dental plaque were used for further analysis. Pool of collected plaque samples were weighed into 3 equal portions of 100 mg each and were transferred to three different test tubes containing

- a) Glass distilled water,
- b) 4% Acetic acid,
- c) 50% Glycerine

These were vortexed for proper mixing and dissolution for 2 minutes. Then it was subjected to centrifugation of 5000 revolutions per minuets (rpm) for 10 min. Supernatant fluid was separately collected and analysed for dissolved dextran content.

Biochemical analysis for dissolved dextran content:

Dextran estimation by Phenol sulphuric acid method

Principle:

Carbohydrate was treated with 75% sulphuric acid at elevated temperature to form furfuraldehyde and their condensation with phenol at ambient temperature formed chromogen which is estimated at 475 – 480 nanometer(nm) of wavelength of light.

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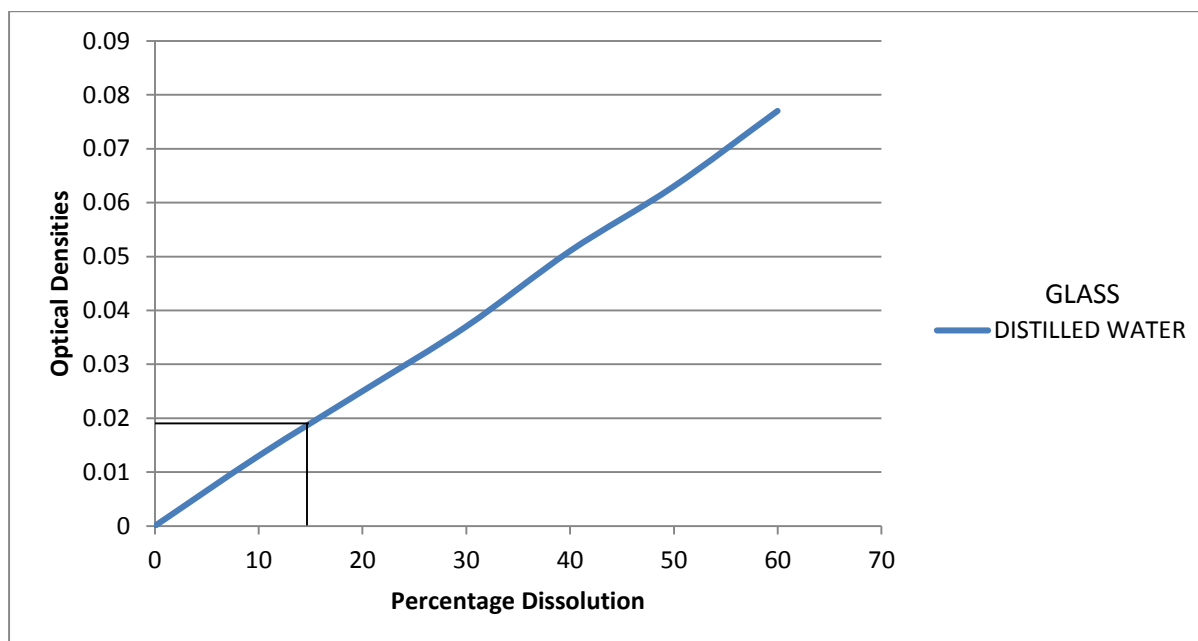
Table No. I: Optical Densities of Standard Solution

	10%	20%	30%	40%	50%	60%
Distilled water	0.013	0.025	0.037	0.051	0.063	0.077
4% acetic acid	0.012	0.025	0.036	0.048	0.061	0.073
50% glycerine	0.29	0.54	0.82	1.1	1.39	1.66

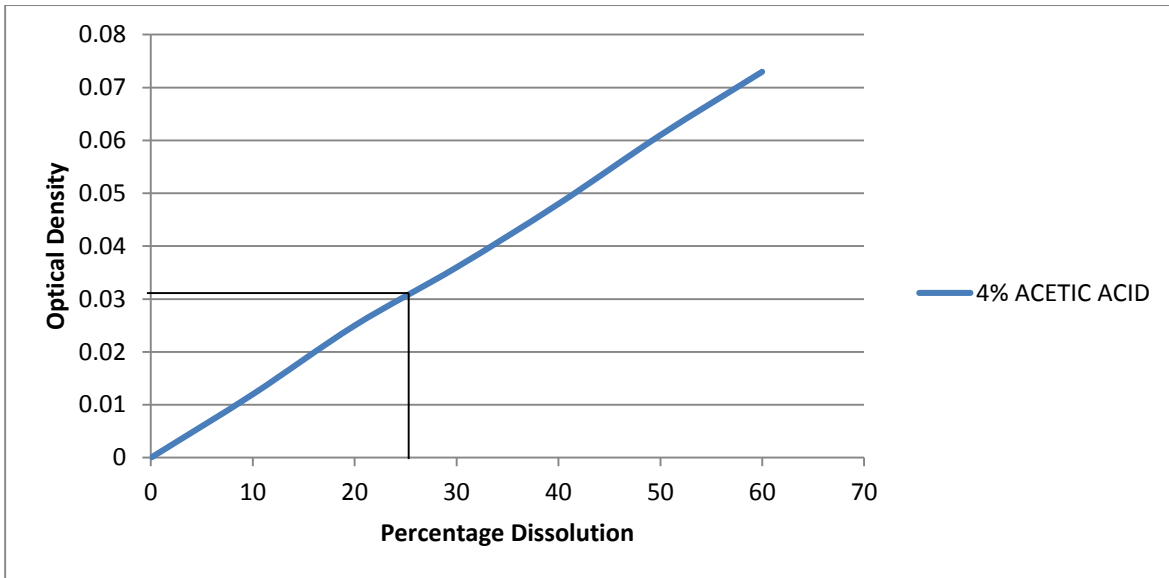
Table II: Optical Densities of Test Solution

	A	B	C	MEAN
Distilled water	0.019	0.018	0.019	0.019
4% acetic acid	0.031	0.03	0.031	0.031
50% glycerine	0.52	0.52	0.52	0.52

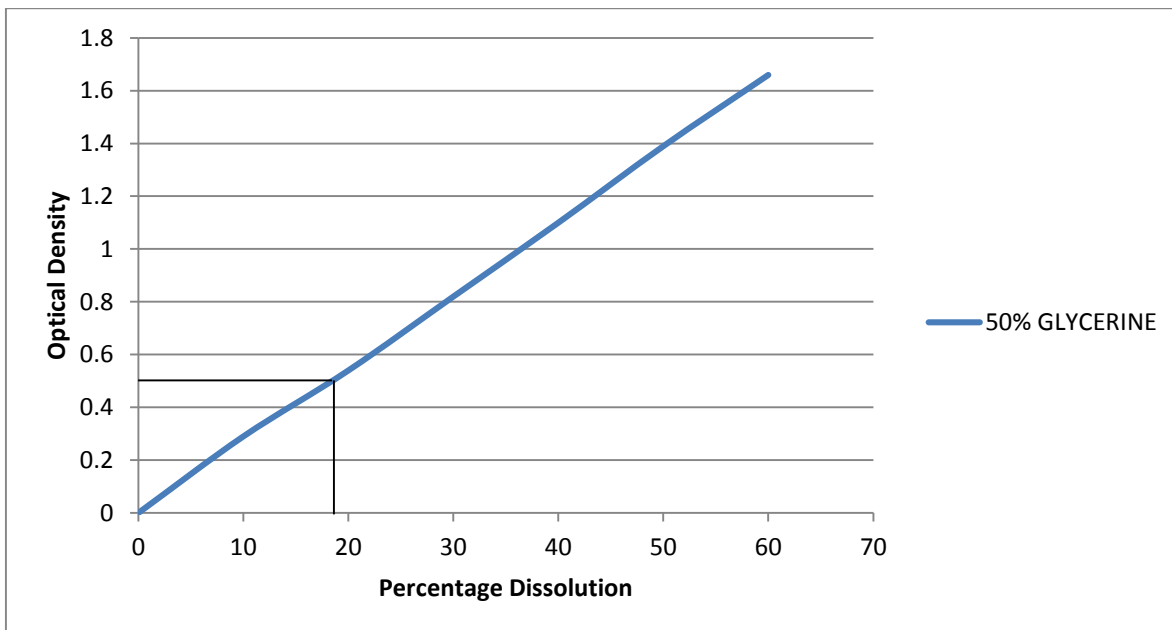
Graph No. I: Optical densities of pure dextran dissolved in glass distilled water



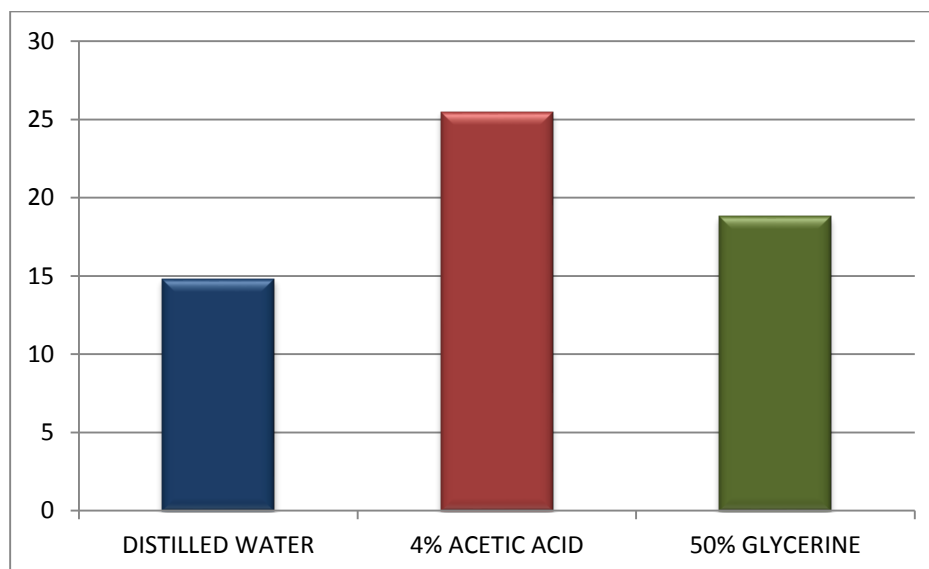
Graph No. II: Optical densities of pure dextran dissolved in 4% acetic acid



Graph No. III: Optical densities of pure dextran dissolved in 50% Glycerine



Graph No. IV: Percentage dissolution of glass distilled water, 4% Acetic acid and 50% glycerol.



Procedure

Four ml of each standard control and test samples were collected in separate test tubes to which freshly prepared 50 μ l of phenol reagent was added. Concentrated sulphuric acid was added along the side of each test tube in cold water bath. Solutions were vortexed and optical densities of these solutions were taken within 5 min with a spectrophotometer[§] of 480nm wavelength of light. Zero level was adjusted with the control solutions (Tables I&II).

Multi calibration graph was plotted for each of the solvent separately at 10 mg%, 20 mg%, 30 mg%, 40mg%, 50mg% and 60 mg% concentration. Dissolution of plaque dextran was estimated by extra plotting from graph as well as by factor (slope) calculation (Tables I & II and Graph No.s I,II &III).

Results:

Results were obtained in the form of calculation method and by graphical method.

Calculated result by slope or factor:

The slope or the factor of the standard solution was calculated by using linear equation formula $Y = mX + c$ where $m(\text{slope}) = (Y_2 - Y_1) / (X_2 - X_1)$, $c =$

[§] UV - 1700, Shimadzu, Japan

constant. The results from calculation found that glass distilled water showed 14.80 % dissolution of plaque dextran; 4% acetic acid has shown 25.47 % dissolution and glycerine showed 18.79% dissolution when it was used at 50% concentration (Graph No. IV).

Calculation of results by graph:

By keeping the y axis as optical density and x axis as percentage solution of pure dextran, optical densities of standard solutions was plotted. By using this graph corresponding percentage of plaque dextran present in the solution were measured. (Graph No.s I,II & III).

The results with graphical method found that Glass distilled water showed \approx 15% dissolution of plaque dextran; 4% acetic acid showed \approx 25% dissolution and glycerine showed \approx 19% dissolution when it was used at 50% concentration.

Almost similar optical densities of test solutions were obtained from both the methods.

Discussion:

Most of the oral problems are plaque related and plaque control practice is done either with mechanical, chemical or combination of both, which

may not prove to be effective at all times. So the authors thought of an alternative method to efficient plaque control by dissolving the plaque contents.

In this preliminary in-vitro study, the authors used vinegar (4% acetic acid) and 50% glycerine as a plaque dissolving agent because a) both are polar solvents b) harmless to the body c) used in day to day life d) economical; which was compared with glass distilled water.

Dissolution of plaque dextran was observed in all the three solutions, out of which 4% acetic acid shows maximum dissolution, followed by glycerine and distilled water respectively. Along with the dissolving capacity, vinegar also has bactericidal action.¹² which gives a strong synergistic effect on plaque control. Acetic acid is known for its unpalatable taste and demineralisation¹⁵⁻¹⁷ activity of enamel. Diluting the vinegar might become palatable and show least harm to the tissues, but it might affect the dissolving capacity as well.

Distilled water is the universal solvent;¹⁸ it dissolves most of the polar solutes and has the capability of dissolving the plaque dextran with least harm to the tissues. so it can be recommended to swish water after every meal and snacks. It not only removes the plaque mechanically, but also dissolves it.

Glycerine also showed dissolution of plaque dextran. Due to its sweet taste it might be accepted by the subjects. But further studies have to be performed to check the efficiency at different concentrations and viscosity with different temperatures.

As public health aspect these materials are inexpensive, less technique sensitive compared to tooth paste and brush so even physically and mentally challenged population can use. This is a preliminary study providing limited information; more research should be directed in this direction.

Limitations of the study:

The dextran analysis by phenol sulphuric acid method is not an ideal method to evaluate the dissolution of plaque dextran. As there was no chemical method for quantitative measurement of dissolved plaque dextran, phenol sulphuric acid method was used and it is recommended that large sample size to substantiate the data.

Conclusions:

In conclusion, all the three solvents showed substantial dissolution of plaque dextran. But vinegar showed more dissolution and glass distilled water showed the least. Further in vitro and in vivo studies have to be performed for the confirmation and effective implication.

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