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Efficacy of different concentrations of glycerol in dissolving dental plaque

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Abstract
Glycerol is one of the effective plaque dissolving agents, which dissolves the important plaque adhering polysaccharide called dextran. The study used 25%, 50%, 75% and 90% glycerine and glass-distilled-water without adding any solute as control solvents and adding pure-dextran [Himedia, India] at 10%, 20%, 30%, 40%, 50% and 60% concentrations were used as standard solutions. Test solutions were prepared by mixing equal amount of collected pooled-dental plaque with the same solvents. Colorimetric Phenol-sulphuric-acid method was used to estimate the level of dissolved dextran using spectrophotometer [Shimadzu-UV-1700, Japan]. Both calculations by factor (slope) analysis and Multi calibration graphical methods showed similar results, where glass distilled water showed 14.80 %, glycerine showed 12.30%, 18.79%, 30.10% and 30.94% dissolution of plaque dextran at 25%, 50%, 75% and 90% respectively. By observing the properties, swishing of 50-75% of glycerol for 3-5min considered to be ideal for the use of dental plaque control.

Keywords: Plaque dissolving agents, glycerine, solvent

Introduction
Dental Plaque embraces of microorganisms and intercellular matrix which are associated with a series of oral diseases including dental caries and periodontal diseases. Removal of dental plaque is considered as basic requirement in preventing these diseases [1-5].
Mechanical and chemical plaque controls are the presently practicing methods of plaque control. Mechanical plaque control includes tooth brushing, flossing, oral prophylaxis etc. whereas chemical plaque control primarily encompasses antibacterial mediator, which used as an adjunct. The most common oral hygiene aid used to improve the oral health of an individual is toothbrush [1, 4-6].
Dental plaque bacteria produce dextran by consuming sucrose in the oral cavity, which benefits the microorganisms to adhere to the host tissue. It is theorized that if dextran is removed effectively, dental plaque will not be able to adhere to the tissues. Thus effective plaque control will be attained.
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 [7]. Dextran can also hydrolyzed with acids [8]. Organic solvents like glycerol have the property to dissolve the plaque dextran [2].
Glycerol is a polyol compound, and polar solvent. Dextran is a polar compound and it dissolves well in polar solvents based on the rule of thumb of solubility [2]. Glycerol of different concentration shows different solubility property. Thereby aim of this study is to assess the ideal concentration of glycerol as solvent on plaque dextran.

Material and Methods
A pool of Supra-gingival dental plaque was collected distinctly using a Columbia scaler in a test tube comprising 2ml of ice-cold glass distilled water and was sent to the Department of Biochemistry where it was sediment by cold centrifugation at 5000rpm for 5 minutes and preserved at -20° Centigrade temperature. Control, standard and test solutions were prepared.

Biochemical Procedures

Standard Solution Preparation

Pure dextran [Himedia, India], 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, 25%, 50% 75% and 90% glycerine. Glass distilled water, 25%, 50% 75%, 90% 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 – 10 degree Centigrade for 5 minutes. Supernatant fluids were discarded and sediments of pooled dental plaque were utilized for further scrutiny. Pool of collected plaque samples were weighed into 5 equal portions of 100 mg each and were transferred to five different test tubes containing

a. Glass distilled water,
b. 25% Glycerine,
c. 50% Glycerine
d. 75% Glycerine
e. 90% Glycerine

These solutions were vortexed for accurate 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 [9]

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.

Procedure

Four ml of each standard control and test samples were collected in separate test tubes to which freshly prepared 50µl of phenol reagent was added. 75% 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 1 & 2).

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 predicted by extra plotting from graph as well as by factor (slope) analysis (Tables 1 & 2 and Graph No.1, 2, 3, 4, and 5).

Results

Calculation of dissolved dextran by slope or factor analysis

The slope or the factor of the standard solution was calculated by using linear equation formula Y= mX+c where m(slope)= (Y2-Y1)/(X2-X1), c = constant. The results from calculation found that glass distilled water showed 14.80 % dissolution of plaque dextran; 25% Glycerol has shown12.30 % dissolution and glycerine of 50%, 75% and 90% showed 18.79%, 30.10% and 30.94% dissolution respectively.

Calculation of results by graph analysis

When plotting the graph by keeping 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 matching percentage of plaque dextran existing in the solution was measured. (Graph 1, 2, 3, 4, and 5).

The results with graphical method found that Glass distilled water showed ≈ 15% dissolution of plaque dextran; glycerine showed ≈ 19%, 30% and 31%dissolution when it was used at 50%, 75% and 90% concentration.

Optical densities of test solutions showed similar results in both the methods.

Discussion

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 [2]. Dental plaque bacteria create dextran by consuming sucrose in the oral cavity, which helps the microorganisms to stick to to the host tissue. 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 [2]. Hence the need of an alternative method to efficient plaque control by dissolving the plaque contents.

To limit the biochemical reaction in detached dental plaque; it is collected in 2ml of chilled glass distilled water and immediately transported to biochemical laboratory and stored in -20 degree centigrade temperature [2, 10].

Previous preliminary in-vitro study on plaque dissolving agents, glycerine showed effective plaque dissolving efficacy [2]. Glycerol, as a polar molecule, is capable of dissolving many polar organic compounds such as inorganic salts, acids, bases, enzymes and transition metal complexes. Based on its polarity, glycerol is therefore immiscible with non-polar compounds [2, 11]. Glass distilled Water and other ionic compounds share very similar traits with glycerol, and are used to facilitate the isolation of reaction products. Glass distilled water is the first choice of solvent based on its chemical, physical, biological properties and also plays an important role from environmental, economic and safety point of view [11]. It shows negligible soluble property on many plaque induced organic compound and organo – metallic compound, which limits its application as plaque dissolving agent [2, 11] Eventhogh distilled water considered as universal solvent [2] glycerol is advantageous when dissolving hydrophobic substrates produced by plaque microorganisms [11].

Dextran is a polar compound; hence as the rule of solubility get easily soluble with glycerol and distilled water. pH does not affect the solubility significantly, hence even in extreme acidic medium of plaque will not affect the dissolution of the dextran. Molecular weight of the dextran and its structure in a molecular form in inversely associated with solubility [7].

In the present study plaque dextran dissolve more in 75% concentration than that of the rest. It clearly shows at the initial stage of 25% concentration dissolution slope is below 20% this may be due to less available polar molecules of glycerine to dissolve the plaque dextran. Similar slow but gradual increase observed in 25 to 50% of glycerol. 50% to 75% of glycerol dissolved around 35% of whole plaque dextran. 75% to 90% the rate or slope of dissolution remains almost similar to that of 75%. The graph appears to be sigmoidal with proper lag, log and plateau phase. (Graph 6).

Conclusion

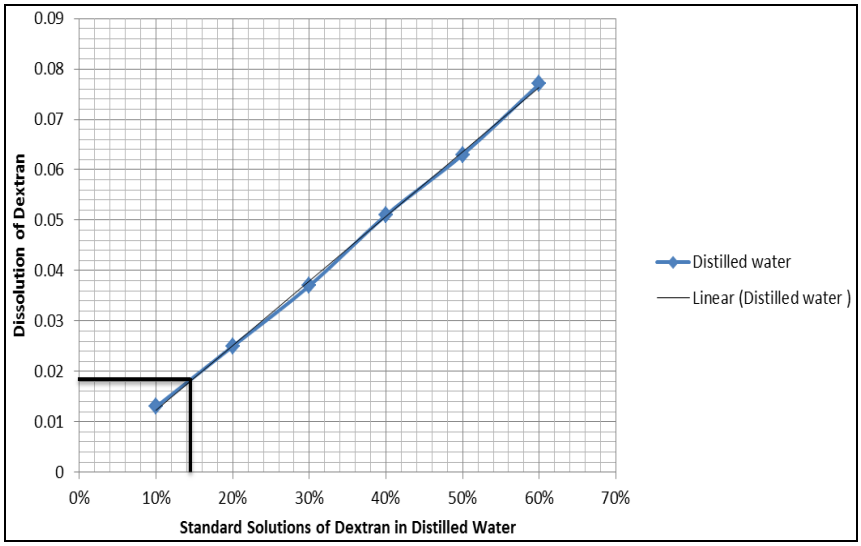
Physical property of glycerol shows as the concentration increases, viscosity of the solvent increases which gives maximum resistance in swishing or cleaning the oral cavity. By observing the characteristic sigmoid curve of solubility and its physical property, swishing of 50 - 75% of glycerol for 3-5min considered to be ideal for the use of dental plaque control.

Table 1: Optical Densities of standard solution

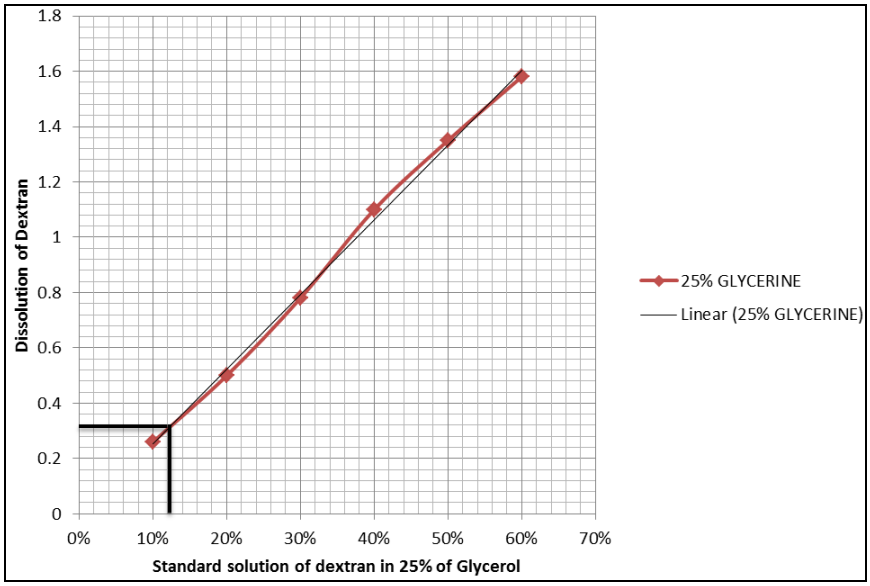
	10%	20%	30%	40%	50%	60%
Distilled water	0.013	0.025	0.037	0.051	0.063	0.077
25% GLYCERINE	0.26	0.5	0.78	1.1	1.35	1.58
50% GLYCERINE	0.29	0.54	0.82	1.11	1.39	1.66
75%GLYCERINE	0.34	0.62	0.86	1.18	1.46	1.76
90% GLYCERINE	0.37	0.67	0.92	1.2	1.49	1.79

Table 2: Optical densities of Test solution

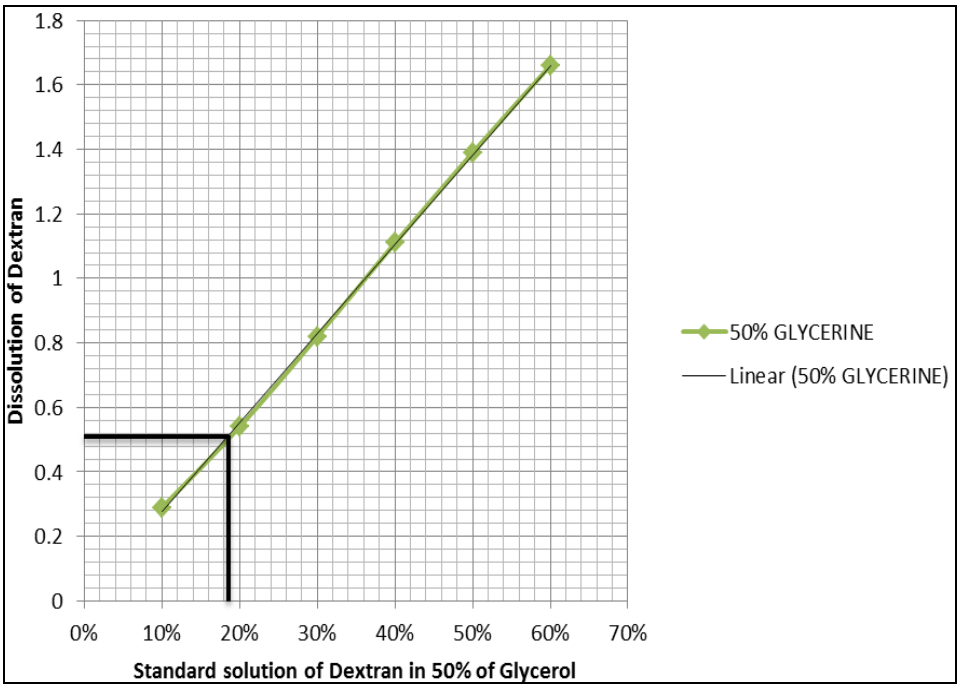
	A	B	C	Mean
Distilled water	0.019	0.018	0.019	0.018667
25% Glycerine	0.37	0.39	0.36	0.373333
50%Glycerine	0.52	0.52	0.52	0.52
75% Glycerine	0.87	0.91	0.89	0.89
90%glycerine	0.91	0.94	0.92	0.923333



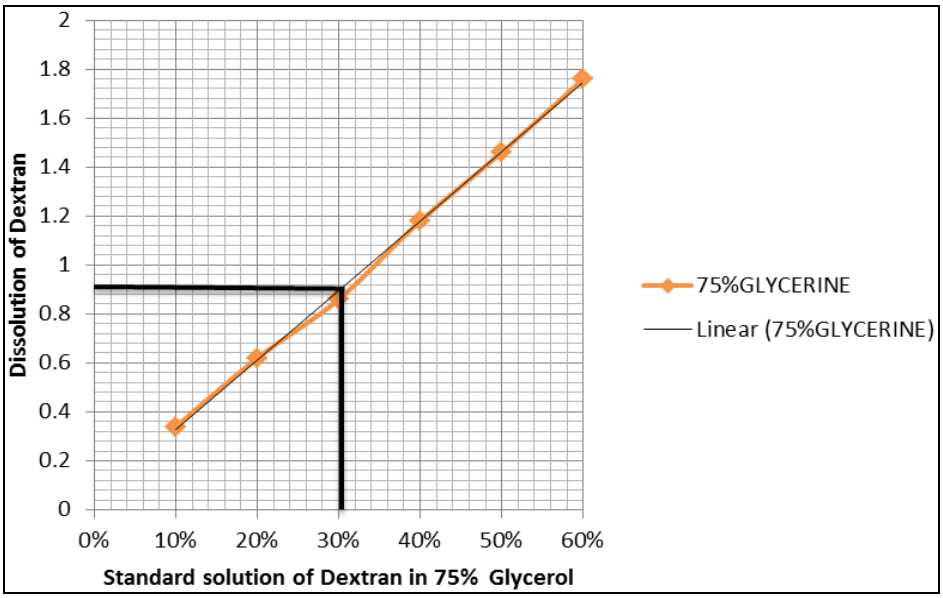
Graph 1: Dissolution of Plaque dextran in Distilled water



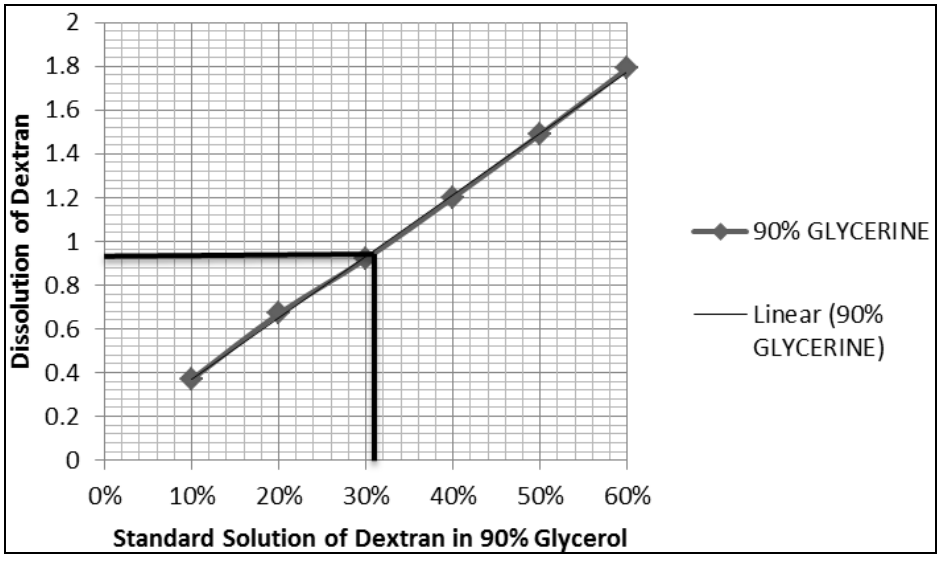
Graph 2: Dissolution of Plaque dextran in 25% glycerol



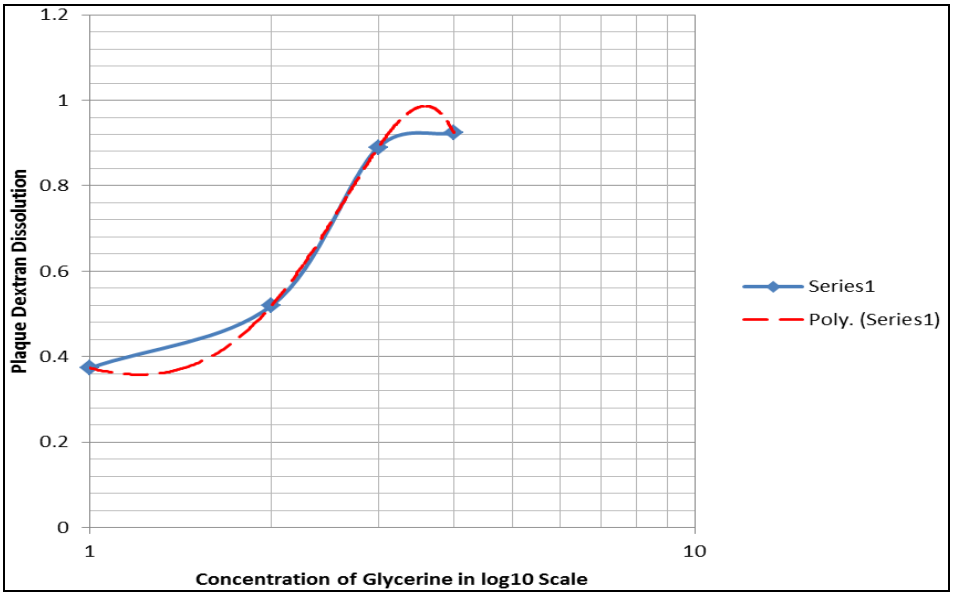
Graph 3: Dissolution of Plaque dextran in 50% glycerol



Graph 4: Dissolution of Plaque dextran in 75% glycerol



Graph 5: Dissolution of Plaque dextran in 90% glycerol



Graph 6: Dissolution of Plaque dextran at different concentrations of glycerol

References

1. Bergström J, Batwa S, Al-Otaibi BMF. The effectiveness of chewing stick miswak on plaque removal. Saudi Dental Journal. 2006; 18(3).
2. Bhat M, Prasad K, Trivedi D, Acharya A. Dental plaque dissolving agents: An *in vitro* study. Int J Adv Health Sci. 2014; 1(3):1-7.
3. Prasad KV, Sreenivasan PK, Patil S, Chhabra KG, Javali SB, DeVizio W. Removal of dental plaque from different regions of the mouth after a 1-minute episode of mechanical oral hygiene. American journal of dentistry. 2011; 24(1):60.
4. Terézhalmy G, Biesbrock A, Walters P, Grender J, Bartizek R. Clinical evaluation of brushing time and plaque removal potential of two manual toothbrushes. International journal of dental hygiene. 2008; 6(4):321-7.
5. Van der Weijden G, Hioe K. A systematic review of the effectiveness of self-performed mechanical plaque removal in adults with gingivitis using a manual toothbrush. Journal of clinical periodontology. 2005; 32(s6):214-28.
6. Deshmukh J, Vandana K, Chandrashekar K, Savitha B. Clinical evaluation of an ionic tooth brush on oral hygiene status, gingival status, and microbial parameter. Indian Journal of Dental Research. 2006; 17(2):74.
7. Dextran Chemistry, Physical Properties [Internet]. 2018. Available from: https://www.dextran.com/about-dextran/dextran-chemistry/physical-properties.
8. Misaki A, Torii M, Sawai T, Goldstein IJ. Structure of the dextran of Leuconostoc mesenteroides B-1355. Carbohydrate research. 1980; 84(2):273-85.
9. Mecozzi M. Estimation of total carbohydrate amount in environmental samples by the phenol–sulphuric acid method assisted by multivariate calibration. Chemometrics and Intelligent Laboratory Systems. 2005; 79(1-2):84-90.
10. Aledo JC, Jiménez-Riveres S, Tena M. The effect of temperature on the enzyme-catalyzed reaction: insights from thermodynamics. Journal of Chemical Education. 2010; 87(3):296-8.
11. Wolfson A, Dlugy C, Shotland Y. Glycerol as a green solvent for high product yields and selectivities. Environmental Chemistry Letters. 2007; 5(2):67-71.