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EFFECT OF DIFFERENT REMINERALIZATION AGENTS ON ENAMEL EROSION. AN IN-VITRO STUDY, AGENTS SUCH AS TOOTH MOUSSE PLUS, REMIN PRO, FLUOR PROTECTOR AND BIFLUORIDE 10

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Abstract

Aims and Objectives: The aim of this in-vitro study is to evaluate the effect of different remineralizing agents on enamel erosion.

Materials and Methods: A total of 100 extracted human maxillary and mandibular molar teeth were selected and cleaned with the ultrasonic scaler. The molars were sectioned using a disc cutting bur from the CEJ and the roots were removed from all the teeth. A 2 mm thickness of buccal surface was taken from all the teeth. The entire exposed surface was covered with clear nail polish to decrease the chance of accelerated demineralization caused by exposure of the dentin. Initial weights of all the teeth were taken. The teeth were stored in normal saline using different containers. The normal saline was decanted out of the containers just prior to commencement of the study. The teeth were then dipped in vinegar for 7 days for disinfection. Once the disinfection of the teeth was completed, the vinegar was decanted followed by its replacement with artificial saliva to all the containers to act as natural saliva. All the teeth were now randomly divided into five groups namely Group A, Group B, Group C, Group D and Group E. 20 teeth were allocated to each

group and were kept in the container labelled as the remineralizing agent going to be used in it.

Results: There is statistically significant difference present in remineralization in various groups. In this research after the treatment, Bifluoride 10 and Fluor Protector showed a significant difference between the experimental and control group. Remin Pro and Tooth Mousse Plus did not show a significant difference but the control group had a higher calcium concentration compared to the experimental group.

Conclusion: This research elucidates the nuanced effects of remineralization agents on calcium dynamics within artificial saliva. The comparative analysis between experimental groups and the protective role of fluoride underscore the clinical relevance of these findings. While acknowledging the in-vitro limitations, the observed potential for remineralizing agents to contribute to enamel remineralization suggests promising avenues for future research and clinical applications.

Keywords: Atomic Absorption Spectrophotometry, Remineralization Agents, Artificial Saliva, Dentin Remineralization, Fluoride, Remin Pro, Fluor Protector, Tooth Mousse Plus, Bifluoride 10

Introduction: Dental erosion is a chronic, localized loss of mineralized tooth structure resulting from their dissolution by non-bacterial acids or from the action of chelating substances on tooth surfaces.[1]

The clinically observed erosive lesions are smooth-surfaced, scoop like, or cup-like losses of dental tissues, a thin strip of undamaged enamel is often found at the edge of the gum, which can be justified by the remineralizing action of gingival crevicular fluid. When teeth undergo dental erosion, the most commonly involved sites are the buccal and lingual surfaces of the upper incisors, which appear smooth and shiny with a generalized loss of anatomy. The erosion of incisors can lead to thinning of the incisal edges and may gradually involve much deeper dental tissue layers leading to the exposure of the dentine tubules and ultimately the pulp.[2]

Dental erosion is of two types, based on causative factors it can be differentiated as:

- The erosion caused by extrinsic factors is also called exogenous dental erosion, where the erosive factors that cause the dental lesions come from food, drink, drugs, or the surrounding environment.
- The erosion caused by intrinsic factors is also called endogenous dental erosion, where the factor that dissolves the mineralized dental structures is hydrochloric acid originating from within the stomach and duodenum.[3,4]

Requirements of an ideal remineralising agent[5]: The requirements of remineralizing agents are:

- Diffuses into the subsurface, or delivers calcium and phosphate into the subsurface.
- Does not deliver an excess of calcium.
- Does not favour calculus formation.
- Works at an acidic pH.
- Works on xerostomic patients.
- Boosts the remineralizing properties of saliva.

Indications for remineralising agents [Zero, 2006][5]

- An adjunct preventive therapy to reduce caries in high risk patients
- To reduced calcification in orthodontic patients
- To repair enamel in cases involving white spot lesions
- Orthodontic decalcification or fluorosis or before and after teeth whitening and desensitizing sensitive teeth.

Materials and Methods: A total of 100 extracted human maxillary and mandibular molar teeth were selected and cleaned with the ultrasonic scaler. The molars were sectioned using a disc cutting bur from the CEJ and the roots were removed from all the teeth. A 2 mm

thickness of buccal surface was taken from all the teeth. The entire exposed surface was covered with clear nail polish to decrease the chance of accelerated demineralization caused by exposure of the dentin. Initial weights of all the teeth were taken. The teeth were stored in normal saline using different containers. The normal saline was decanted out of the containers just prior to commencement of the study. The teeth were then dipped in vinegar

for 7 days for disinfection. Once the disinfection of the teeth was completed, the vinegar was decanted followed by its replacement with artificial saliva to all the containers to act as natural saliva. All the teeth were now randomly divided into five groups namely Group A (control group, n=20), Group B (experimental group, n=20) Tooth mousse plus (GC), Group C (experimental group, n=20) Remin pro (Voco), Group D (experimental group, n=20) Fluor protector (Ivoclar) and Group E (experimental group, n=20) Bifluoride10 (Voco). 20 teeth were allocated to each group and were kept in the container labelled as the remineralizing agent going to be used in it.

After each tooth was completely covered with the respective material, the teeth were allowed to sit undisturbed for five minutes. The teeth were then placed back in the original labelled container with artificial saliva for 30 minutes as suggested by the manufacturer. The artificial saliva was then decanted and 30 ml of soft drink was added to each container for 10 minutes (Long A. 2016)[6.] After 10 minutes the soft drink was collected inside each container and was analysed to determine the absorption of calcium content using Atomic Absorption Spectrophotometer. After the adjustment of the instrumental parameters by a professional the soft drink was aspirated into a flame that was lined up with a light beam that was generated by lamp specific for calcium. A detector measured the intensity of the beam of light and calculated the absorbance by flame photometer. The absorption of all the samples was recorded and compared to find the parts per million of calcium in each solution.

Result: Data was analyzed using SPSS version 23. Descriptives, One way anova along with post hoc tukey test was done for inter group comparison.

Table1:Comparison of mean remineralization in various groups

Group	N	Minimum	Maximum	Mean	Std. Deviation	F value	P value
Tooth Mousse plus	20	.84	.96	9060	.04285	166.681	<0.001*
Remin Pro	20	.74	.88	.8030	.04725		
Fluor protector	20	.51	.66	.5990	.05281		
Bifluoride 10	20	.45	.58	.5130	.04256		
Control	20	.90	1.60	1.2700	.21051		

**-highly significant (p<0.001)

Inference: There is statistically significant difference present in remineralization in various groups. One way anova signifies overall comparison to know the individual comparisons post hoc test should be done.

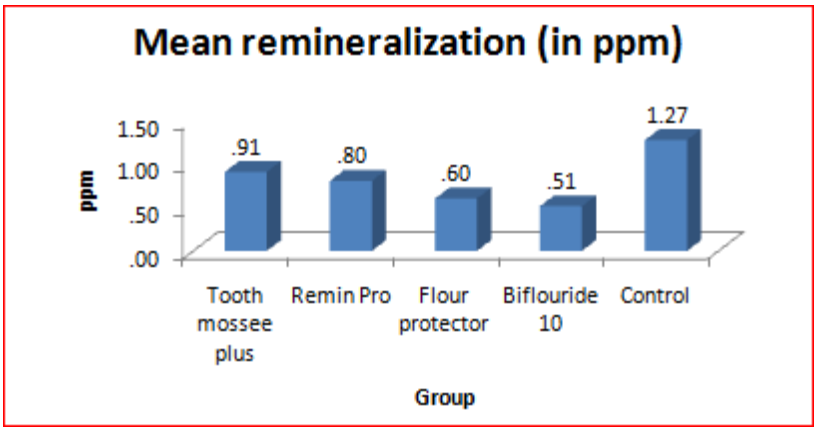


Table2: Individual pairwise comparison of remineralization

Comparison between		Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Tooth Mousse Plus	Remin Pro	0.103	.03255	0.002*	.0384	.1676
	Fluor Protector	0.307	.03255	<0.001**	.2424	.3716
	Bifluoride 10	0.393	.03255	<0.001**	.3284	.4576
	Control	0.364	.03255	<0.001**	-.4286	-.2994
Remin Pro	Fluor Protector	0.204	.03255	<0.001**	.1394	.2686
	Bifluoride 10	0.29	.03255	<0.001**	.2254	.3546
	Control	0.467	.03255	<0.001**	-.5316	-.4024
Fluor Protector	Bifluoride 10	0.086	.03255	0.010*	.0214	.1506
	Control	0.671	.03255	<0.001**	-.7356	-.6064
Bifluoride10	Control	0.757	.03255	<0.001**	-.8216	-.6924

**-Highly significant (p<0.001)

There is statistically significant difference present in all comparisons.

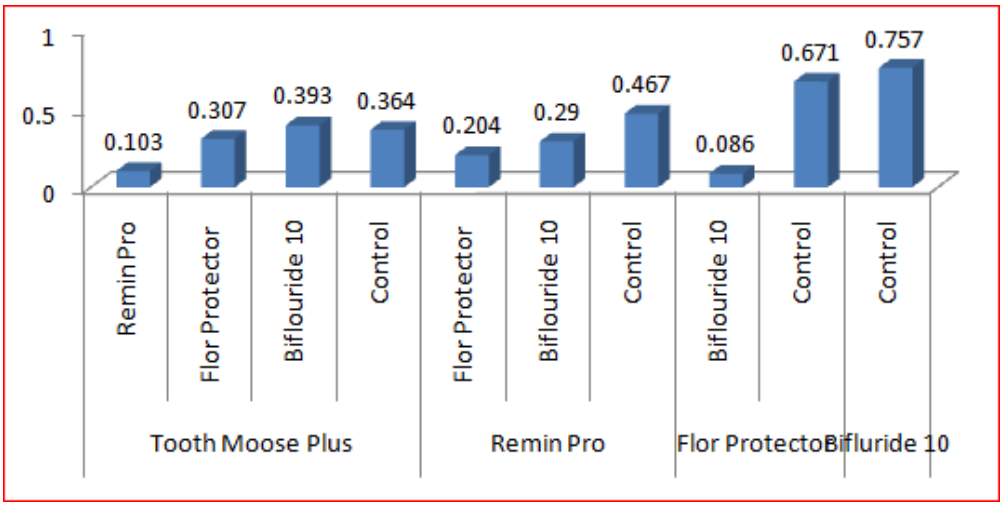
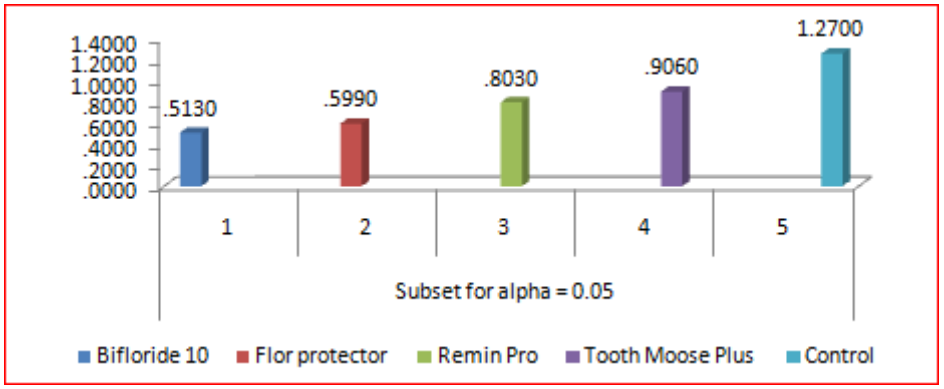


Table3: Order of mean remineralization (post Hoc Tukey test)

Group	N	Subset for alpha=0.05				
		1	2	3	4	5
Bifluoride10	20	.5130				
Fluor protector	20		.5990			
Remin Pro	20			.8030		
Tooth Mousse Plus	20				.9060	
Control	20					1.2700

Order of remineralization

Bifluoride10> Fluor Protector>Remin Pro> Tooth Mousse Plus > Control



Discussion: For many years dental erosion was thought to consist of a one-way progressive demineralization of enamel crystallites followed by degradation of dentin, leading to cavity formation. Dental hard tissues are constantly undergoing cycles of demineralization (when pH is low) and remineralization (when conditions favor) leading to variations in mineral status of the teeth throughout the day.[1,2]

In **1998, McIntyre et al,**[3] showed how the initial demineralization of tooth enamel isa reversible process, and that it occurs alongside a process of remineralization, maintaining a

balance between the two. However, at low pH, created by bacterial acids, this equilibrium may shift to favor the demineralization process.

Differential Effects of Remineralization Agents: Among the tested agents, Bifluoride 10 exhibited a notably higher effectiveness in reducing enamel erosion, as evidenced by the lowest ppm values. This finding suggests that the composition of Bifluoride 10 may confer enhanced remineralization properties compared to the other agents under the studied conditions.

Jayarajan et al (2011)[7]stated that CPP ACP can consume the acid generated during enamel lesion remineralization by generating more calcium and phosphate ions, including CaHPO4, thus maintaining the high concentration gradient into the lesion. The present study findings are in consistent with the study conducted by **Diamanti et al (2010)[8]**where he compared the remineralization efficacy of Fluoride and Nova Min and showed both can inhibit demineralization and enhance remineralization with fluoride having more remineralizing efficacy.

In this research after the treatment, Bifluoride 10 and Fluor Protector showed a significant difference between the treatment and control group. The sample size (20in each group) was adequate to show that the treatment group had a significantly lower concentration of calcium compared to the control group. Remin Pro and Tooth Mousse Plus did not show a significant difference but the control group had a higher calcium concentration compared to the treatment group.

Bifluoride 10 and Remin Pro are fluoride varnishes that are recommended by dentists to be applied every six months at a cleaning. Fluoride in the varnish binds to the calcium ions on the surface of the tooth and binds the calcium ions together. Binding the ions together forms a stronger surface layer which in turn decreases the rate of demineralization **Lata, et al (2010).[9]**

The results show that the treatment group has a lower concentration of calcium compared to the control group. The concentration of calcium in the treatment group decreases from the first treatment itself. Remin Pro and Tooth Mousse Plus contain a lower concentration of fluoride compared to a varnish. These two protective agents are intended to be used by a patient daily.

Conclusion: Remineralizing agents provide a potential avenue for the management of tooth demineralisation, especially that caused by dental caries. This can lead to a paradigm shift in the management of these diseases; from limiting the damage caused, by excavation and restoration; to, one day, potentially reversing the hard tissue destruction, followed by minimal restorative management. Several synthetic and naturally-derived materials have been tried for this purpose, and many of these have shown a lot of promise as remineralizing agents. Perhaps, using multiple such agents in conjunction with each other may prove more beneficial in obtaining remineralization. Fluorides were the first such agents, used for the remineralisation of enamel. While fluoride ions play an integral part in the demineralisation-remineralisation process, helping in the long-term repair of subsurface demineralization; they take years to take effect in vivo, and are of limited use in cavitated lesions. Among new materials, casein phosphopeptide-containing materials seem to show the greatest promise in terms of remineralisation of cavitated and noncavitated lesions. Although several of these remineralizing agents have shown quite a bit of promise, with their current level of effectiveness, they still remain just an adjuvant to conventional, more invasive methods for the management of dental caries.

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