REMINERALIZING POTENTIAL OF SALIVA ON PATIENTS HAVING DENTAL EROSIONS DUE TO WINE CONSUMPTION

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ABSTRACT

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Introduction: The aim of the study is to assess the action of a remineralizing product (calcium-phosphate-fluoride-based varnish) on the saliva remineralization capacity on patients having dental erosions caused by frequent wine consumption.

Methodology: The study group was made up of 15 patients who are frequent wine consumers. Unstimulated saliva was collected on the same day and at the same hour for each patient. A total amount of 0.5 ml of unstimulated saliva was placed on a glass plate, dried for 30 minutes in a thermostat at +37˚C, and then studied using a Nikon Eclipse E 600. The images were saved and stored on a computer. The IMK index was determined using the formula: IMK= number of the network areas filled with crystals/number of the network areas projected on the entire saliva drop. The treatment plan for each patient included a five-week application of MI Varnish (GC Corporation) once a week. After 5 weeks IMK values were recorded again.

Results: The mean value of IMK increased from 0.33 before treatment to 0.83 after treatment. The distribution of micro-crystallization categories varied from 86% Type II before treatment to 93% Type I after treatment.

Conclusions: The fluor local treatment that uses varnishes containing casein phosphopeptide, tri-calcium phosphate, amorphous calcium phosphate and fluoride, increase the saliva remineralising potential and can be recommended both as preventive therapy and to counteract the erosive effect of acid oral environment on patients with dental erosions related to the frequent consumption of wine. Local treatment with GC Recaldent MI Varnish increased the remineralizing potential of saliva on patients with dental erosions due to wine consumption.

Keywords: erosion, wine, salivary micro-crystallization index, remineralization.
processes related to dental erosions. The success of dental erosions therapy depends on the preventive and therapeutic strategy that must be focused on the risk assessment, oral hygiene practices, diet, lifestyle, medical factors. The long-term success is related to the possibilities to improve the saliva environment quality and regular application of remineralising protocols. Some studies proved the effectiveness of products based on calcium and phosphate in the erosion lesion onset.

Initially, dental erosion is associated with enamel loss of a few micrometers, a process known as demineralization. Demineralization is associated to a decrease in the calcium and phosphate ions concentration. In time, the loss can affect the entire enamel layer. Factors like the remineralisation process and acquired pellicle formation can reduce the rate of dental erosion. De- and remineralisation processes alternate on long time intervals.

Remineralization is influenced by the concentration of calcium and phosphate ions in saliva. Previous studies showed a direct correlation between the remineralizing capacity of saliva and the aspect of saliva crystals on microscopy analysis. Patients having high remineralizing capacity of saliva presented, in 93.5% of the cases, an aspect of fern-shaped crystals, with the highest crystals concentration in the middle of saliva drop. Patients with low remineralising capacity of saliva presented no such this structure or it was diffuse in 87% of the cases and saliva presented a few crystals in the visual field or some needle-shaped crystals, homogeneously distributed or grouped to the periphery of the saliva drop.

Dental erosion is influenced by a lot of internal and external factors. The most important factor is considered the frequent consumption of beverages, including cola-like drinks, high acid drinks and wine. The frequent consumption of wine (alcoholism) or professional exposure to wines represent major factors in the dental erosions onset. The erosive potential of red wine, white wine, and champagne is related to the presence of lactic acid, citric acid, malic acid, and tartaric acid.

The treatment of dental erosions is based on the removal of etiological factors and the use of remineralizing products containing calcium, phosphate, fluoride, magnesium, and zinc. The aim of study was to assess the action of a remineralizing product (calcium-phosphate-fluoride based varnish) on the remineralizing capacity of saliva on patients having dental erosions related to frequent wine consumption.

### 2. Methodology

The study group was made up of 15 patients who are frequent wine consumers (minimum 5 times a day), age 30-50, having dental erosions. The patients were selected from patients treated in the Dental Clinic of the Dental Medicine Faculty, “Gr. T. Popa” University of Medicine and Pharmacy, Jassy. After clinical examination the remineralizing capacity of saliva was evaluated using micro-crystallization index (IMK). For IMK evaluation, unstimulated saliva was collected in a test tube before 12 a.m. From this saliva, 0.5 ml were applied on a microscope slide; the saliva liquid was dried for 30 minutes in 37°C temperature. The microscopy analysis was performed using Nikon Eclipse E 600, and images were recorded and stored in a computer. The IMK micro-crystallization index was calculated using the following formula:

\[
\text{IMK} = \frac{\text{number of points of numbering grill projected on crystals}}{\text{number of points of numbering grill projected on saliva drop}}
\]

The IMK values were divided in three types, namely high, moderate and low level of crystallization. The basic criteria for the crystals evaluation were the diameter, shape and number. The mean values of IMK between 0.6 and 1 were included in type I of crystallization, values between 0.4 and 0.6 were included in type II and values between 0 and 0.4 were included in type III. The treatment plan for each patient included a weekly application of MI Varnish (GC Corporation, Tokyo, Japan) for 5 weeks. Saliva was evaluated at the baseline and after 5 weeks.

### 3. Results

The IMK values before and after treatment are presented in Table 1. The mean value of IMK increased from 0.33 before treatment to 0.83 after treatment after treatment.

The distribution of the micro-crystallization categories varied from 86% Type II before treatment to 93% Type I after treatment. The types of micro-crystallization were represented by needle-shaped, fern-shaped, tree-shaped, flake-shaped, oval-shaped or cubic-shaped crystals, as well as their multiple points or combinations (Figs 1-6). Before treatment 86% of the IMK values were included in type II category of micro-crystallization and 14% in type III. After treatment 93% of the IMK values were included in type I and 7% in type II. The IMK values before and after treatment were statistically analyzed using the Wilcoxon test. Statistically significant results were obtained when comparing the IMK values before and after treatment (p<0.05).

### 4. Discussion

Saliva acts as a buffer agent for various beverages. However beverages like fruit juices or wine are resistant to the acid neutralizing action of saliva and have the capacity to prolong the time of...
**Table 1.** IMK values at baseline and after 5 weeks of treatment using MI Varnish

<table>
<thead>
<tr>
<th>IMK values</th>
<th>Mean</th>
<th>STDEV</th>
</tr>
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<tbody>
<tr>
<td>initial</td>
<td>0.3</td>
<td>0.2</td>
</tr>
<tr>
<td>after</td>
<td>0.4</td>
<td>0.3</td>
</tr>
<tr>
<td>treat.</td>
<td>0.5</td>
<td>0.4</td>
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<td></td>
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<td></td>
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<td>0.2</td>
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<td>0.3</td>
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<tr>
<td></td>
<td>0.4</td>
<td>0.4</td>
</tr>
<tr>
<td></td>
<td>0.333</td>
<td>0.097</td>
</tr>
</tbody>
</table>

**Figures 1-6.** Examples of saliva micro-crystallization aspects

**Figure 1.** IMK = 0.2

**Figure 2.** IMK = 0.8

**Figure 3.** IMK = 0.4

**Figure 4.** IMK = 0.8

**Figure 5.** IMK = 0.3

**Figure 6.** IMK = 0.8
pH dropping. The development, evolution and prognostic of dental erosion depend on the frequency and duration of the acid exposure. The protective biological factors against erosive factors are saliva and acquired pellicle.\textsuperscript{11-13} The remineralizing ability of saliva is influenced both by the minerals levels and the quality and levels of salivary mucins. Mucins are proteins that participate in the saliva bio-crystallization processes and influence directly the increase and development of salivary biocrystals. Salivary mucins also influence the diameter and shape of the anorganic deposits and the type of crystallization in IMK test. Lennon AM\textit{et al.}\textsuperscript{34} assessed in vitro the protective ability of a casein-calcium phosphate tooth against dental erosiions. The researchers concluded that 12500 ppm AMF gels supply the most effective protection against dental erosion. The mechanism by which the products containing casein-calcium phosphate reduce erosion is not clearly established yet. Except for the prevention of dental hard tissues demineralization, it was suggested that the product also remineralizes (repairs) eroded enamel and dentine crystals. Ramalingam observed a superficial granular structure formed on the enamel surface. It is highly possible for these structures to represent remineralized enamel crystals.\textsuperscript{11}

Our study proved that local treatment with \textsc{recaldent\textsuperscript{TM}} (CPP-ACP) MI Varnish\textsuperscript{TM} influences the remineralizing potential of saliva for patients with dental erosions by increasing the mean IMK value after treatment. The crystallization types also improved significantly. In our study, prior to treatment, most of the patients presented the highest crystals concentration in the middle of saliva droplet. Other studies showed that patients having dental erosions presented separated crystals in the form of a branch or stem, placed relatively evenly over the whole surface of the dried droplet or a large amount of separated stellate crystals of an oval and irregular shape located isometrically.\textsuperscript{35}

The major components of the varnish that we tested were casein phosphopeptide, calcium phosphate, amorphous calcium phosphate and fluoride. They can contribute to increase the remineralising potential of saliva and to arrest the erosive effect. Previous studies showed that other products having similar components (Tooth Mousse, GC Corporation, Tokyo, Japan) have good control on tooth erosion, possibly due to anticariogenic remineralizing agent represented by casein phosphopeptide-amorphous calcium phosphate (CPP-ACP) nanocomplex.\textsuperscript{36}

The reduction in tooth erosion induced by white wine consumption was clearly demonstrated by Manton (2010) and Piekarz C (2008).\textsuperscript{9,10} In the absence of a device focused on the early detection of dental erosions, the clinical aspect and the practitioner’s experience contribute to the accurate detection in the early stages.\textsuperscript{6,9,13,34}

IMK evaluation is an affordable test to monitor the effectiveness of the non-operative, preventive methods. The methodology is simple, accessible and inexpensive. The principal components of this varnish, casein phosphopeptide (CPP-ACP) calcium phosphate, amorphous calcium phosphate and fluoride, can contribute to the increase of saliva remineralising potential and to arresting the erosive effect. GC Recaldent MI Varnish can be recommended to counteract the erosive effect of wine and acid beverages.

5. Conclusions
The use of varnishes containing casein phosphopeptide, calcium phosphate, amorphous calcium phosphate and fluoride increases the remineralizing potential of saliva for patients having dental erosions related to frequent wine consumption.

Acknowledgments
The authors declare no conflict of interest related to this study. There are no conflicts of interest and no financial interests to be disclosed.

REFERENCES

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CV

Dr. Nicoleta Tofan graduated College of Dentary Technique (2002) at Grigore Ghica Voda, Jassy and Faculty of Dental Medicine at “Gr. T. Popa” University of Medicine and Pharmacy, Jassy, Romania (September 2012).
She also participated at Intensive Training Program in Aesthetic Medicine the Injections Module at QClinic Cluj (May 2014) with Professor Adrian Avram and achieved the necessary skills and techniques for performing the Injections with Hyaluronic Acid.
Also she attended at the Worksop “Global Approach in Facial Rejuvenation by Injecting Hyaluronic Acid” organised by the Romanian Society of Dermatology in May 2016.
Dr. Nicoleta Tofan has published 2 articles in Chemistry Magazine (December 2015) and in Romanian Journal of Oral Rehabilitation.

Questions

Researches of Leus showed that the patients with low remineralizing capacity of saliva presented in microscopy
- a. A „fern” aspect of saliva crystals;
- b. A few crystals in visual field or needle-shaped crystals;
- c. The highest crystals concentration in the middle of saliva drop;
- d. A “corncob” aspect of saliva crystals.

The erosive potential of red wine, white wine, and champagne is related to the presence of:
- a. Lactic acid, phosphoric acid, tartaric acid;
- b. Citric acid, phosphoric acid, malic acid;
- c. Lactic acid, citric acid, malic acid, tartaric acid;
- d. Stearic acid, linoleic acid, malic acid.

The use of MI Varnish containing casein phosphopeptide, calcium phosphate, amorphous calcium phosphate and fluoride:
- a. Increased only the mean IMK values;
- b. Had no effect on IMK values;
- c. Decreased the type of saliva crystallization;
- d. Increased the mean IMK values and type of saliva crystallization.

The types of saliva micro-crystallization were represented by:
- a. Needle-shaped crystals;
- b. Tree-shaped crystals;
- c. Cubic-shaped crystals;
- d. All the previous answers are correct.