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SEM Analysis of the Effectiveness of the Acid Etching on Cat Enamel

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Abstract—The aim of this paper is to summarize the literature on micromorphology and composition of the enamel of the cat and present an original experiment by SEM on how it responds to the etching with ortophosphoric acid for the time recommended in the veterinary literature (30", 45", 60"), derived from research and experience on human enamel; 21 teeth of cat were randomly divided into three groups of 7 (A, B, C): Group A was subjected to etching for 30 seconds by means of orthophosphoric acid to 40% on a circular area with diameter of about 2mm of the enamel coronal; the Groups B and C had the same treatment but, respectively, for 45 and 60 seconds. The samples obtained were observed by SEM to constant magnification of 1000x framing, in particular, the border area between enamel exposed and not exposed to etching to highlight differences. The images were subjected to the analysis of three blinded experienced operators in electron microscopy. In the enamel of the cat the etching for the times considered is not optimally effective for the purpose adhesives and the presence of a thick prismless layer could explain this situation. To improve this condition may clinically in the likeness of what is proposed for the enamel of human deciduous teeth: a bevel or a chamfer of 1 mm on the contour of the cavity to discover the prismatic enamel and increase the

Keywords—SEM, Cat, Enamel.

I. INTRODUCTION

ALWAYS more frequently in the domestic cat, dental restorations are made after fractures or FORL [1] using composite and adhesive systems originally designed for man. The substrates of the adhesion are, notoriously, the enamel and dentin (more rarely cement), [2] but do not seem to exist in the veterinary literature micromorphological specific works on the relationship between these hard tissues of the tooth of the cat with the adhesive/composite systems used clinically. To date, optical microscopy and scanning electron microscopy (SEM) have amply described both the growth [3]-[5] of the structure and the composition of the mature enamel of the cat. About the latter, in particular, the enamel surface of the cat tends to appear smooth, with noticeable deficiency of perikimata and prisms exposed in the surface [5]; this smooth surface would

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be formed "as a result of slowed incremental growth of enamel prisms by ameloblasts, which is associated with a change in the shape of the Tomes process, resulting in a lack of distinction between pitfloor and interpit enamel". Often, however, seem to present small areas of lesion with exposure of prismatic "cobbled" enamel that would be related to wear and trauma in the course of life. The thickness of the enamel varies, in the cat, from 0.1 to 1 mm and has three layers:

- A prismless surface layer, which shows a thickness varying from 5 to 20 microns and appears to cover all surfaces of the tooth with partial exception of the cervical areas:
- An intermediate layer with parallel prisms only in some sites:
- A deep layer, with parallel prisms and prominent bands of Hunter-Schreger, in which the diameter of the individual prisms seems to vary significantly and the course suddenly changes with respect to that of the surrounding prisms.

However, in cross-section the diameter of the prisms is on average 5 microns, similar to that the human. The composition of the enamel of the cat is, of course, based on calcium and phosphorus as in all mammals and in man, but seem to be formed even high concentrations (<5% of the total elemental composition) of fluorides, sodium and magnesium in addition to the combined presence (<0.1%) of sulfur, potassium and iron combined. Two interesting considerations that emerge from the literature in this respect are that the surface layer of the enamel manifests the lowest concentration of calcium and that there are differences in composition between males and females.

It is therefore evident that the enamel of the cat while much resembling to that of humans is not equal, however, in the veterinary literature are recommended for etching times and concentrations of orthophosphoric acid equal to those used for the enamel of permanent teeth of man. According to these acquisitions, the hypothesis of this work is that the enamel of the cat reacts to acid etching (the same concentrations of ortophosphoric acid and the same time) unlike that of the human permanent teeth.

II. MATERIALS AND METHODS

The study involved the use of 21 teeth from domestic cats died from various causes, including 12 canines and 9 premolars. Once extracted, the teeth were cleaned of the main organic residues in their roots by curettes and then immersed for 12 hours in a 5% aqueous solution of sodium hypochlorite. Once extracted from the solution, the samples were thoroughly

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washed by a jet of water and dried by means of air jet for 30-45 seconds. The samples were then divided randomly into three groups of 7. Each group has been identified by the letters A, B and C.

- Group A Each of the seven samples was subjected to etching for 30 seconds with phosphoric acid gel (40% Acid Etch H Schein Inc., NY USA) placed on a circular area (diameter about 2 mm) of the surface of the crown. At the end of the etching each sample was washed with abundant spray of air-water and dried by prolonged (30 seconds) air jet.
- Group B Each of the seven samples was treated as those of group A with the only variant that the etching time was 45 seconds.
- Group C Each of the seven samples was treated as those of groups A and B with the only variant that the etching time was 60 seconds.

All the samples obtained were then coated with a Carbon atomized layer of 300Å by means of sputtering machinery KQ150R sputter. The samples thus prepared were then subjected to observation with SEM EGF LEO 1525 to constant magnification of 1000 x framing, in particular, for each sample an area of surely unetched enamel, the border area between enamel exposed and not exposed to etching, an area of enamel situated centrally with respect to the part of etched enamel. Some images were also carried out with magnifications ranging from 50 to 7000 x relating to particular characteristics of the surfaces observed.

III. RESULTS

The 63 images were obtained at 1000 magnification and 12 images at magnifications ranging from 50 to 7000 x. Of the 63 pictures related to the 21 samples, 21 are presented on the border area between exposed and unexposed to enamel etching. The 21 images were then subjected to the blinded assessment of three experienced operators (two dentists and a Vet), that have assessed the validity of the enamel etching according to the scale

- 3 Effective
- 2 Fair
- 1 Poor
- $\boldsymbol{0}$ Questionable or nothing

The ratings were then brought back to the 3 groups of 7 samples and the averages were obtained that were of 0.67 for group A (etching than 60 seconds), of 0.43 for group B (etching than 45 seconds) and of 0.43 for the group C (etching than 30 seconds). In all three groups the average is located, then, between poor and questionable even if the group A (etching than 60 seconds) has the best behavior of the other two.

IV. DISCUSSION

The ability to act with orthophosphoric acid etching on enamel effective cat for the times and concentrations proposals for the permanent human enamel is poor. The reason for this is probably to be ascribed to the presence of a thick aprismatic layer; which, moreover, possesses a decreased concentration of calcium [5], element on which it is preferentially occur in the chemical attack of orthophosphoric acid [6]. In support of this hypothesis, there is a similar situation which occurs in human deciduous teeth where the presence of a substantial aprismatic layer [7]-[9] has led first to hypothesize opportunities for different times of action of the etchant [10]-[13] and, subsequently, to add to them of mechanical pre-treatment of the enamel surface able to place in evidence the prismatic component [14]-[15]. In even more recent times, the clinic has overcome this problem by proposing the use of bevel or chamfer in the enamel margin of the cavity [16].

V.CONCLUSIONS

From a clinical point of view it seems possible to say that the acid etching on time and in the manner currently used in veterinary dentistry is not optimally effective against for adhesion to enamel cat and the probable cause for this may lie in the presence of a thick prismless layer. Given these acquisitions, the possible clinical solutions are:

- Increase the exposure time and / or the concentration of overcoming the barrier formed by the prismless enamel (remember, however, that this type of solution has proved ineffective in human deciduous, failing to eliminate the prismless thickness in a timeframe with the clinic)
- Establish, as occurs in the human reconstructions, a bevelling or a chamfer of the extension of about 1mm on the contour of the cavity to discover the prismatic enamel, attachable then in an ideal manner etching, and increase at the same time the surface small tea of accession. In a future contribution will test the hypothesis that, by a rugosimeter, it is possible to define a statistically reliable difference between the bonding surfaces obtained by enamel cat etched / not mechanically prepared and enamel cat prepared and subsequently etched.

REFERENCES

- S. Holmstrom, P. Frost, E. Eisner Veterinary Dental Techniques (3rd ed.) Saunders, Philadelphia (2004);
- [2] DuPont G A Radiographic Evaluation and Treatment of Feline Dental Resorptive Lesions - Veterinary Clinics of North America: Small Animal Practice 35 (4), July 2005, 943–962;
- [3] GorrelC Small Animal Dentistry in Saunders Solutions in Veterinary Practice (Nind F. Ed)) – Saunders Elsevier , Philadelphia, 2008;
- [4] Skobe Z, Prostak KS, Trombly PL (1985) Scanning electron microscope study of cat and dog enamel structure. J Morph 184, 195–203;
- [5] Boyde A (1964) The structure and development of mammalian enamel. PhD thesis, University of London;
- [6] Kallenbach E (1976) Fine structure of differentiating ameloblasts in the kitten. Am J Anat 145, 283–317;
- [7] Hayashi Y (1983) Crystal growth in matrix vesicles of permanent tooth germs in kittens. ActaAnat (Basel) 116, 62–68;
- [8] Sasaki T, Debari K, Higashi S (1984) Energy-dispersive X-ray microanalysis and scanning electron microscopy of developing and mature cat enamel. Arch Oral Biol 29, 431–436;
- [9] Boyde A, Fortelius M, Lester KS, Martin LB (1988) Basis of the structure and development of mammalian enamel as seen by scanning electron microscopy. Scanning Microsc 2, 1479–1490;
- [10] DeLaurier A., Boyde A., Horton M. A. Price J. S. Analysis of the surface characteristics and mineralization status of feline teeth using scanning electron microscopy J. Anat. (2006) 209, pp 655–669;

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ISSN: 2415-6612 Vol:8, No:6, 2014

- [11] Crossley DA (1995) Tooth enamel thickness in the mature dentition of domestic dogs and cats – preliminary study. J Vet Dent 12, 111–113;
- [12] Boyde A (1989) Enamel. In Handbook of Microscopic Anatomy (edsOksche A, Vollrath L), Vol. V/6, pp. 309–473. Berlin: Springer Verlag;
- [13] Boyde A, Reith EJ (1983) Cyclical uptake pattern of tetracycline in post-secretory maturation phase enamel demonstrated in rooted teeth.Calcif Tissue Intl 35, 762–766;
- [14] Boyde A (1969) Electron microscopic observations relating to the nature and development of prism decussation in mammalian dental enamel. Bull Group IntRechSciStomatol 12, 151–207;
- [15] Hayashi K, Kiba H (1989) Microhardness of enamel and dentin of cat premolar teeth. Jpn J Vet Sci 51, 1033-1035;
- [16] Colley PA, Verstraete FJM, Kass PH, Schiffman P (2002) Elemental composition of teeth with and without odontoclastic resorption lesions in cats. Am J Vet Res 63, 546–550.

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