



<b>Title</b>	<b>Detecting anions of human saliva by ion chromatography</b>
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<b>Citation</b>	<b>78th General Session and Exhibition of the International Association for Dental Research, Washington DC, USA, 5-8 April 2000, v. 79 n. Sp Iss, p. 519</b>
<b>Issued Date</b>	<b>2000</b>
<b>URL</b>	<b><a href="http://hdl.handle.net/10722/53748">http://hdl.handle.net/10722/53748</a></b>
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**3001** Tray delivery of Potassium Nitrate-Fluoride to reduce bleaching sensitivity. VB HAYWOOD\*, WF CAUGHMAN, KB FRAZIER, ML MYERS. Medical College of Georgia, Augusta, GA.

Tooth sensitivity during at-home bleaching is a major deterrent to successful completion of treatment. The purpose of this study was to evaluate the use of potassium nitrate and fluoride for sensitivity treatment. 30 patients bleached their teeth at night with 10% carbamide peroxide (Platinum™, Colgate Oral Pharm) in a custom-fitted tray. The bleaching tray was a rigid experimental tray with a non-scaled, no-reservoir design for which sensitivity was expected. Tooth sensitivity encountered was treated with the application of a gel containing 5% potassium nitrate and 1000 ppm sodium fluoride in the bleaching tray. If the patient experienced sensitivity, they applied the gel as needed for 10-30 minutes in the tray to continue bleaching if possible. Records and patient interviews were used to determine the number of patients needing the gel, the success in sensitivity reduction, the length of treatment time, the number of applications, and ability to complete bleaching treatment. 16 out of 30 patients (53%) experienced tooth sensitivity. 12 of 16 patients used the gel to continue bleaching. Of that 12, 11 patients (92%) reported reduction in sensitivity and completed bleaching successfully. Treatment times ranged from 10 minutes before bleaching to 30 minutes before and after, with 10 minutes being the most frequent treatment time. The number of applications ranged from 1 to continuous treatment. Since total bleaching times were not identical, there was a wide range in number of applications. Some patients were able to complete bleaching with a few gel applications for sensitivity and no further treatment. Other patients were unable to continue bleaching unless they continued using the gel daily. The use of a 5% potassium nitrate-fluoride gel applied in the tray as needed for tooth sensitivity associated with at-home vital bleaching reduced tooth sensitivity in a majority of patients and allowed most patients to continue bleaching to completion. Supported by Colgate Oral Pharmaceuticals #11-12-02-1260-82

**3002** Cysteine-induced oral production of volatile sulfur compounds (VSC). G. ROLLA\*, G. JONSKI and A.R. YOUNG (Dental Faculty, University of Oslo, Norway).

It is known that oral rinses with low concentrations of aqueous solutions of cysteine cause an immediate formation of very high concentrations of VSC in the oral cavity. Furthermore, it is well established that hydrogen sulfide (HS) and methyl mercaptan (MM) are the dominant VSCs in halitosis originating from the oral cavity. The aim of the present study was to examine the nature of the VSCs that are formed after cysteine rinses. A test panel of 25 subjects of both sexes aged from 12-70 years, with varying levels of oral hygiene and of periodontal conditions, rinsed with 3 and 6 mmol/l of L-cysteine for 30 s. The subjects were asked to keep their mouths closed for 90 s after which a 10-ml sample of mouth air was taken, injected directly into a Shimadzu 14 B gas chromatograph, and processed according to standard procedures. It was found that exclusively HS was produced after cysteine rinses by the test panel, regardless of age, sex and periodontal conditions. It has been suggested that the amount of VSC formation upon a cysteine rinse measured by a halimeter could have prognostic value regarding development of periodontal disease. As periodontal disease is associated with MM in mouth air and since this compound appears to have the highest potential for tissue destruction (and the most unpleasant odor), it is suggested that the use of cysteine rinse alone for prognostic tests as described above have limitations.

**3003** The effect of aqueous solutions of metal salts on volatile sulfur compounds (VSC) production from human saliva *in vitro*. A. YOUNG, G. JONSKI and G. ROLLA (Dental Faculty, University of Oslo, Norway).

Oral malodor or bad breath arises from the production of volatile sulfur compounds produced as a result of degradation of whole saliva. It is known that metal ions such as Zn<sup>2+</sup> and Cu<sup>2+</sup> used in oral care products exhibit anti-VSC effects *in vivo*. The aim of the present study was to examine and compare the effect of the salts of Zn, Cu, Sn, Hg, Pb and Cd on VSC production *in vitro*. The hypothesis to be tested was that the metal ion with the greatest affinity for sulfur results in the greatest reduction in VSC production. 10-µl of the metal chloride solutions was added to test tubes containing 1-ml of freshly collected human whole saliva and incubated overnight at 37 °C. The solutions contained 7.34 mM or 22.02 mM of the metals (7.34 mM ZnCl<sub>2</sub> = 0.1 % ZnCl<sub>2</sub>). Controls containing saliva without any additions were included. After incubation the tubes were shaken and the saliva headspace was analysed for VSCs directly in a 14B Shimadzu gas chromatograph. At the low concentration, Cu<sup>2+</sup> and Hg<sup>2+</sup> gave the best results, reducing VSC production dramatically, while Sn<sup>2+</sup>, Cd<sup>2+</sup> and Zn<sup>2+</sup> were effective at the higher concentration. Pb<sup>2+</sup> did not appear to exhibit any positive effects in this system. Apart from the results for lead, these results confirm the hypothesis, although copper and mercury presumably have a greater antibacterial effect and may also operate by this mechanism.

**3004** Sulphide Probe Analyses of Plaque Treated with an Oxidising Mouthrinse. E LYNGH\*, B MILLS\*, C SILWOOD\*, A CLAXSON\*, M GROOTVELD\* (Cons. Dent & Infl. Res. Grp, Barts & the Royal London SMD, QMW, London E1 2AD, UK).

The nature, rate and extent of salivary reductant consumption by oxidants present in anti-halitosis products reflects their oxidising capacity, a parameter of much relevance to their therapeutic and aesthetic actions. Therefore, we have evaluated the influence of a chlorine dioxide-generating mouthrinse [1] on the sulphide (S<sup>2-</sup>) content of human plaque using a novel microelectrode system [2] for measuring S<sup>2-</sup> levels at the tip of a periodontal probe (meter readings are inversely proportional to S<sup>2-</sup> levels). Measurements were made on a range of standards between 0.002 mM and 0.2 mM, S<sup>2-</sup> and plots of response vs concentration were linear (r = -0.99). Baseline S<sup>2-</sup> levels were recorded at 5 sites on the posterior one third of the tongue, in approximal and subgingival plaque on 4 molar teeth and 4 incisor teeth from volunteers (n = 10), prior to the use of the mouthrinse, as well as at periods of 5 min, 1 hr and 24 hr subsequent to this treatment. Results acquired demonstrated that oxidants in the mouthrinse gave rise to reduced S<sup>2-</sup> readings for at least 24 hours. Tongue S<sup>2-</sup> readings (mean±SE) changed from -0.189±0.008 to -0.113±0.005 (5 min), -0.124±0.005 (1 hr) and -0.117±0.003 (24 hr) (p<0.01 for each comparison). Approximal molar S<sup>2-</sup> (mean±SE) changed from -0.334±0.033 to -0.162±0.015 (5 min), -0.119±0.012 (1 hr) and -0.155±0.008 (24 hrs) (p<0.01). Subgingival molar S<sup>2-</sup> (mean±SE) were modified from -0.307±0.022 to -0.151±0.006 (5 min), -0.144±0.007 (1 hr) and -0.170±0.007 (24 hrs) (p<0.01). S<sup>2-</sup> probe analyses of plaque provides valuable molecular information regarding the oxidising capacity and actions of oral health care products. Since S<sup>2-</sup> is associated with halitosis and periodontal disease processes, their oxidative consumption by oxohalogen oxidants represent a useful therapeutic role for oral health care products containing these agents. [1] retrADENT™, Rowpar Pharmaceuticals, AZ, USA. [2] Diamond General Corporation, Ann Arbor, Michigan, USA.

**3005** Proton (1H) NMR Investigations of the Antioxidant Actions of a Dentifrice. M. GROOTVELD\*, B. MILLS, C. SILWOOD, A. CLAXSON, E. LYNGH (Barts & the Royal London SMD, QMW, London E1 2AD, UK).

Reactive oxygen-derived radical species (RORS) have been implicated in the pathogenesis of inflammatory gum diseases (IGDs) and their reactions with biomolecules can give rise to the generation of 'unnatural' catabolites with the potential to propagate periodontal diseases. We tested the ability of a novel dentifrice, Dentavite containing an antioxidant matrix (Pycnogenol®) to suppress the potentially deleterious hydroxyl radical (OH•) and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>)-induced damage to salivary biomolecules. Saliva specimens collected from a total of 12 volunteers were centrifuged, the clear supernatant removed and divided into two 2.50 ml portions. To the first of these was added 0.25 ml of an aqueous supernatant derived from centrifugation of the dentifrice investigated (I) whilst the second was treated with an equivalent volume of doubly-distilled H<sub>2</sub>O. Each sample was then further sub-divided into three 0.60 ml portions: the first of these was treated with H<sub>2</sub>O<sub>2</sub> (2.00 x 10<sup>-3</sup> mol dm<sup>-3</sup>), the second with a source of OH radical, and the third served as an untreated control. Each sample was equilibrated at 37°C for a period of 30 min and the nature and extent of RORS-mediated oxidative damage to salivary biomolecules was monitored by high resolution 1H NMR analysis. The dentifrice supernatant was found to inhibit (1) the OH radical-mediated depolymerisation of salivary glycosaminoglycans to oligosaccharide fragments, (2) the oxidation of lactate to pyruvate and then to acetate and CO<sub>2</sub> by H<sub>2</sub>O<sub>2</sub>, (3) generation of pro-inflammatory formate from the attack of OH radical on carbohydrates in general. We conclude that the dentifrice tested protects salivary biomolecules against RORS-induced oxidative damage, a phenomenon relevant to its therapeutic potential. Supported by Appl. Dent. Science, Mass., USA.

**3006** Peculiarities of salivary ions' secretion in dogs, rats and humans. I.A. PETROVITCH, R.P. PODOROZNAJA, SOMASUNDARAM SUBRAMANIAN\* (Moscow Medico-Stomatological Univ., Moscow Medical Acad., Moscow, Russia.)

We studied the peculiarity of salivary gland (SG) secretion by comparison of ion levels in saliva and blood of humans, dogs and rats. In humans the concentration of stable ions were determined spectrophotometrically or with titration, time dynamics of isotopes were studied radiometrically in dogs after intravenous injection and in rats after subcutaneous injection. Stimulated parotid saliva in humans was received with Leshly's capsula, in dogs with Pavlov's fistula from externally exposed ducts of parotid gland. In rats stimulated mixed saliva was obtained. Each experiment was repeated for 5 to 33 times. Statistically confirmed difference p<0.05. In humans the concentration of stable thiocyanate in saliva was 3 to 10 times higher than in blood. The level of [<sup>125</sup>I]thiocyanate in dogs was 4 to 25 times higher in saliva than in blood, but in rats on the contrary it was 1.5 to 3 times less in saliva than in blood after the injection of KCN<sup>125</sup>S. We observed that SG concentrate stable iodine in saliva 10 to 80 times more than in blood in humans and dogs, but in rats SG do not concentrate <sup>127</sup>I after injection of Na<sup>127</sup>I. In humans the level of stable Cl in saliva is 2 to 5 times less than in blood, in rats concentration of <sup>36</sup>Cl was lesser in blood, but in dogs β-radiation of <sup>36</sup>Cl was almost the same in both fluids after injection of Na<sup>36</sup>Cl. In humans salivary levels of stable Ca was 2 to 4 times less in saliva than in blood, in dogs <sup>45</sup>Ca was 2 to 4 times more in saliva, in rats radioactivity of <sup>45</sup>Ca was almost the same in both fluids after injection of <sup>45</sup>CaCl<sub>2</sub>. Blood capillaries, acinar cells and ducts of salivary glands determine the selectivity of (morpho-functional) system which we call as hemato-salivary barrier, its function has definite peculiarities in different mammals.

**3007** Detecting Anions of Human Saliva by Ion Chromatography. Z.F. CHEN,\* B.W. DARVELL, V.W.H. LEUNG (Faculty of Dentistry, The University of Hong Kong).

This study was to investigate the behaviour of 10 anions (see Table) reported as components of human saliva under various ion chromatographic conditions. Reagent grade chemicals and deionized water (18 megohm-cm at 23 °C) were used to prepare solutions with ion concentrations ranging from 1 to 100 mg/L which were analysed on an ion chromatograph (DX-100, Dionex, CA, USA) equipped with an anion guard column (IonPac AG-4A), anion separator column (IonPac AS-4A), suppressor (ASRS-1) and a conductivity detector. The flow rate of the mobile phase was set at 2.0 mL/min. Data acquisition and analysis programs (DaqBook, Iotech, OH, USA; PeakFit 4.0, SPSS, IL, USA) were used for retention times with 2 eluents: (1) 1.7 mM NaHCO<sub>3</sub> - 1.8 mM Na<sub>2</sub>CO<sub>3</sub>; (2) 10 mM Na<sub>2</sub>B<sub>4</sub>O<sub>7</sub>. 15 runs for each ion were made for at least 5 concentrations. Retention time was unaffected by concentration. Fluoride and lactate were co-eluted in both eluents while iodide and sulphate were co-eluted in Eluent 2. These results show the feasibility of detecting multiple anions in human saliva using IC. Eluent 1 seems a better choice for human saliva than Eluent 2. However, another eluent or stationary phase needs to be found to resolve fluoride and lactate. Supported by HKU Grad. Sch. Res. Stud. Support Fund.

Anion	Retention time of anions (mins)									
	F <sup>-</sup>	Lac	Cl <sup>-</sup>	NO <sub>2</sub> <sup>-</sup>	B <sup>-</sup>	NO <sub>3</sub> <sup>-</sup>	HPO <sub>4</sub> <sup>-</sup>	SO <sub>4</sub> <sup>2-</sup>	I <sup>-</sup>	SCN <sup>-</sup>
Eluent1	0.938	0.954	1.400	1.664	2.455	2.796	4.394	5.781	10.81	18.80
= s.d.	0.004	0.008	0.007	0.007	0.012	0.020	0.028	0.053	0.16	1.38
Eluent2	1.240	1.317	2.725	3.568	6.178	7.089	21.27	32.37	33.08	64.35
= s.d.	0.025	0.005	0.072	0.076	0.158	0.361	0.60	1.78	1.21	4.03

**3008** Tannic Acid Pigmentation in Human Saliva: Mechanisms. G.R. GERMAINE\*, S.J. JOHNSON and J.R. MILLER (Department of Oral Science, School of Dentistry, Univ of MN, Minneapolis, MN, USA)

Human whole saliva (WS) and parotid fluid increase the rate of pigment formation from tannic acid (TA). The overall objective of this study is to describe the biochemical basis of salivary augmentation of TA pigmentation. TA and other desired substances were prepared in 50-100mM potassium phosphate adjusted to the desired pH, incubated anaerobically to allow TA transformation followed by aerobic incubation to monitor pigment development at 405nm. Paper chromatography of anaerobic reaction mixtures revealed TA, an unknown compound (Px) of high mobility and a cluster of low mobility substances. Gallic acid also yields Px and the low mobility cluster. Px is strongly associated (p<0.00004) with high pigmentation rates (>0.44/hr). Px appears similar, if not identical, to pyrogallol (PYG) based upon: (i) the same mobility in two-dimensional chromatography, and (ii) the chromatographic species generated from TA or GA also appear to be produced by PYG. Salivary promotion of Px formation and pigmentation are heat (100C/10 min) and protease sensitive. Thus, salivary promotion of TA pigmentation appears to depend upon enzymatic processes and involve several intermediates. Supported by MDRCBB.