

bio'Xtracts

Clinical Evidence



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Patient comfort and compliance

bioXtra® Gel – Consumer Study.

July 2000: data on file Bio-X Healthcare.

One hundred dry mouth volunteers were recruited (90 completed the study) to compare their regular remedy for dry mouth with the bioXtra® system (this involving their use of the toothpaste / mouthwash for normal oral hygiene and the gel for 'comfort'). Their opinions were captured using a simple questionnaire. The study was conducted in the UK.

The increased viscosity of bioXtra® Moisturising Gel appears to have achieved the objective of creating greater patient acceptance – more than 80% of respondents noted a longer duration of lubrication when compared with their 'regular' remedy, approaching 90% perceived a greater feeling of moisture and approximately 75% reported greater mouth comfort.

Use of bioXtra® Moisturising Gel in Japanese Dry Mouth Patients.

Dr. Satoshi Nishimura et al, Department of Oral Maxillofacial Surgery, Nihon University School of Dentistry, Tokyo, Kanazawa, Japan, May 2002.

This study aims to record the subjective perceptions of comfort and relief from dry mouth of 7 patients suffering with dry oral cavity as a result of a systemic disorder. All patients' oral condition and function was evaluated prior to the trial. Subsequent examinations were conducted at the end of weeks 1, 2 and 4. Patients applied bioXtra® Moisturising Gel to the oral cavity whenever the mucosa felt dry. All patients reported an improvement in the feeling of dryness, especially at night. All patients except 1 wished to continue with the treatment.

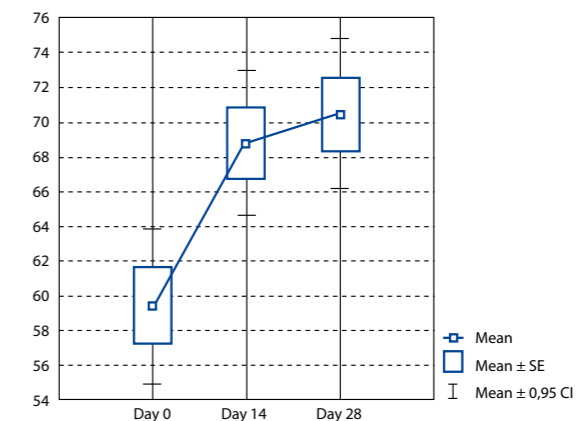
Efficacy of the bioXtra® dry mouth care system in the treatment of radiotherapy-induced xerostomia.

Piet Dirix & Sandra Nuyts & Vincent Vander Poorten & Pierre Delaere & Walter Van den Bogaert
Support Care Cancer (2008) 16:171–179

Xerostomia is a common complication of radiotherapy for head and neck cancer. Symptomatic treatment consists of stringent oral hygiene to prevent oral infections and saliva substitutes to increase comfort. The aim of the study was to evaluate the clinical effectiveness of the bioXtra® (BX) dry mouth care system. A xerostomia questionnaire consisting of 3 parts (xerostomia symptom score, quality of life (QoL) survey and visual analogue scale (VAS)) was completed by 34 patients suffering from radiation-induced xerostomia, before and after 4 weeks of treatment with the bioXtra® moisturizing gel, toothpaste and mouthwash. The bioXtra® products significantly diminished the most common symptoms of xerostomia. Mean VAS score at the start of treatment was 59.8. After treatment, this decreased to 36.4 (p<0.001). Twenty-six patients (77%) responded to treatment, 11 of these patients (32%) reported a major improvement. Quality of life significantly improved under treatment: mean QoL score at the start was 59.4; this increased to 70.5 (p<0.001). None of the 34 patients reported any adverse effects and all but 1 patient found the BX dry mouth care system easy to use.

Quality of Life score:

Mean QoL score at the start of treatment was 59.4.
This increased to 70.5 after 28 days of BX dry mouth care system (mean difference was -11.1, 95% CI [-28.7 to 6.6], P<0.001)



How effective are oral hygiene products for patients with Xerostomia?

S. A. Vasina, A. V. Lapatina. The department for the prevention of oral diseases at the Moscow State University of Medicine and Dentistry (MGMCU). Translated from Russian. DENTAL FORUM #1[19]2006

Forty volunteers took part in the clinical trial, all of whom due to associated pathologies (diabetes mellitus) or having taken certain medicines, suffered from symptoms of xerostomia. After an initial oral examination and instruction in oral hygiene rules, the trial participants were divided into 2 groups of twenty. Patients in Group I were advised to clean their teeth with bioXtra® toothpaste twice a day (in the morning and before going to bed) for no less than three minutes, and the Group II were told to use bioXtra® mouthrinse for thirty seconds after cleaning their teeth, as well as during the day if they experienced any dryness in the mouth. The duration of the clinical trial was set at 2 months. The patients were given oral check-ups on a regular basis once a month, during which the following points were assessed:

- any local irritation or allergic reactions caused by regular use of bioXtra® products;
 - the patients' oral hygiene levels – evaluated using changes in the Patient Hygiene Performance Index PHP (Podshadley, Haley, 1968)
 - the condition of the periodontal tissue - using the Gingivitis Index GI (Loe H., Silness J., 1963), in order to determine the extent of gum inflammation;
 - speed of salivation – by collecting patients' stimulated saliva in a measuring tube before and after applying BioXtra products;
- As part of the trial the participants were questioned in order to evaluate the organoleptic properties of the bioXtra® product range. A statistical analysis of the results obtained from the clinical trial was done using the Student method. The results of the trial revealed that the hygiene treatments had very good anti-inflammatory action, seen through a reduction in the Gingivitis Index scores compared with the start of the trial (46% reduction in patients who used the toothpaste, and 34% in those using the mouthrinse). During the final oral examination of the participants, a fall in the severity of signs of inflammation of the periodontal tissue was seen, and fewer patients complained of bleeding gums when cleaning their teeth or eating hard food. All patients who had used the bioXtra® range of oral healthcare products commented on a subtle pleasant fresh taste and smell, and feelings of comfort and freshness in the mouth for a long time after use. During the two months there were no cases of localised irritation or allergic reactions caused by these oral healthcare products. Therefore, regular use of these special treatments by patients with reduced salivation leads to a significant improvement in their oral hygiene levels and the condition of their periodontal tissue.

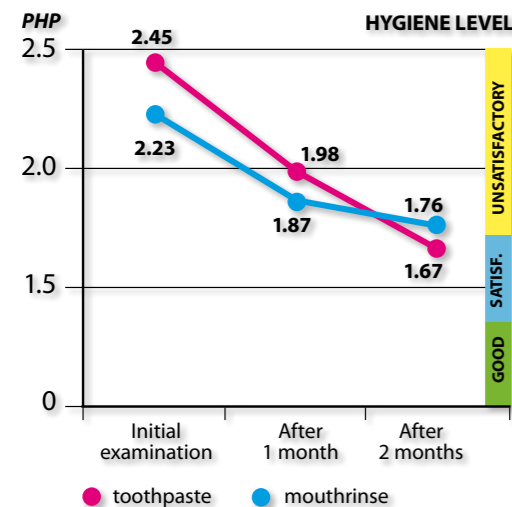


Fig. 1
Changes in hygiene index scores after using bioXtra® toothpaste and mouthrinse

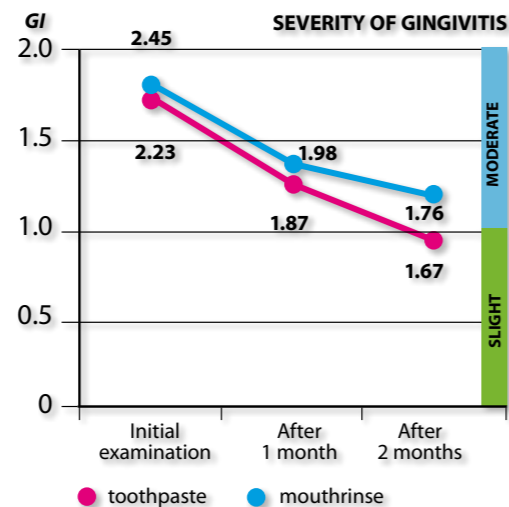


Fig. 2
Changes in gingivitis index scores after using bioXtra® toothpaste and mouthrinse

Comparative studies

bioXtra® versus biotène®: Biofilm Control

Novel Methods for the Treatment of Infectious Diseases Involving Microbial Biofilms.

Perraudin J-P, Courtois Ph, Free University of Brussels, Journ. Int. Assoc. Periodontology, Oct, 2001 (Abstracts) Le Monde Dentaire June, 2002 ; Vol 112, pp 5-9.

Two different aspects of the microflora colonisation can be distinguished. The first consists of an isolated form observed mainly in saliva. The second consists of a colonized form which can be observed in dental plaque or on mucosal surfaces.

The use of antimicrobial agents for the prevention and treatment of periodontal and dental plaque – related diseases is attracting considerable interest within the dental profession.

The present study compares the effectiveness in vivo of two toothpaste formulations: Toothpaste A (biotène®) - containing the Lactoperoxidase System producing OSCN⁻ (hypothiocyanite), and a Toothpaste B (bioXtra®) - containing Lactoferrin, Lysozyme, Lactoperoxidase System and Immunoglobulins as antimicrobial agents, and a mixture of Growth (or Wound Healing) Factors. Results indicate Toothpaste A (biotène®) has an inhibitory effect on a limited number of the planktonic bacteria present in the patients' saliva.

Conversely, Toothpaste B (bioXtra®), containing a mixture of antibacterial agents in conjunction with Immunoglobulins and Growth Factors was shown to be instrumental in removing colonized (biofilm) bacteria and to have a greater effect in controlling the subsequent higher proportion of planktonic bacteria found in the saliva.

The antibacterial activity for both formulations was calculated by an ATP assessment of the bacteria versus the number of bacteria present.

Removal of the biofilm bacteria was demonstrated by observing the number of planktonic bacteria in the saliva of the patients using the Toothpaste B (bioXtra®) compared to the number of bacteria in the saliva of the patients using Toothpaste A (biotène®).

The action of Toothpaste B (bioXtra®) on the biofilm bacteria was also characterised by a reduction in dental plaque and lactic acid production.

Figure 1: Quantity of plaque (mgr)

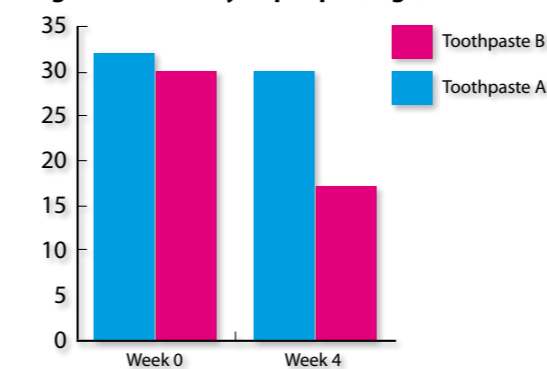


Figure 2: Lactic acid (µgr/mgr)

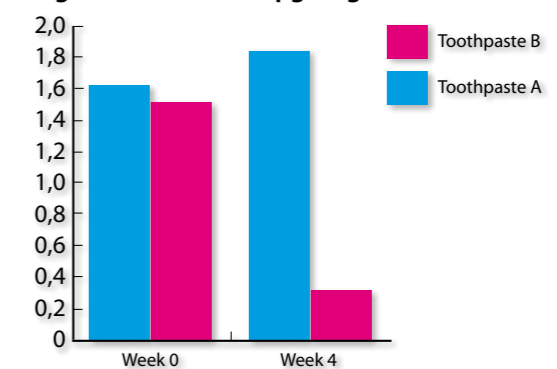


Figure 3: Total bacteria

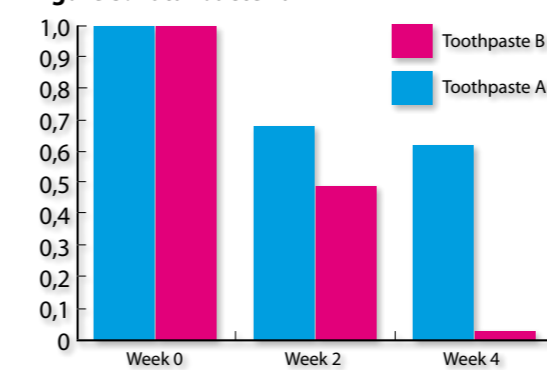
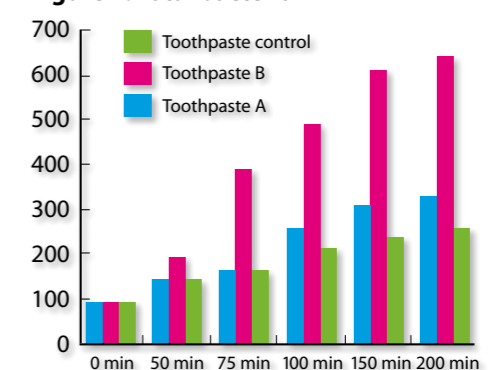


Figure 4: Total bacteria



bioXtra® toothpaste versus placebo: Efficacy in Oral Health

Biofilm inhibition and antimicrobial activity of a dentifrice containing salivary substitutes.

S Hatti, S Ravindra, A Satpathy, RD Kulkarni, MV Parande (2007), Department of Periodontics, SDM College of Dental Sciences and Hospital, Sattur, Dharwad, Karnataka, India; Department of Microbiology, SDM College of Medical Sciences and Hospital, Sattur, Dharwad, Karnataka, India; International Journal of Dental Hygiene 5 (4), 218–224. Nov. 2007.

Background/aims: A healthy mouth harbours the continuous combined action of a salivary defence system with that of a salivary peroxidase system, containing lactoferrin, lysozyme, immunoglobulin and growth factors. This system maintains neutral pH and creates an oral environment where harmful bacteria are inhibited, thus preventing the formation of biofilms. The objective of this clinico-microbiological trial was to evaluate the anti-plaque effect of a dentifrice containing salivary substitutes (bioXtra®), compared with a placebo-control dentifrice (commercially available fluoridated toothpaste marketed by Colgate-Palmolive, Mumbai, India) and to assess the effect of dentifrice on oral bacterial count.

Methods: The design was a randomized controlled, double-blind, parallel study comparing a placebo-dentifrice to a dentifrice formulation containing salivary substitutes. Toothpaste slurry rinses were used over a 96-h period by 20 volunteers who refrained from all other oral hygiene procedures. Commercially available fluoride toothpaste was used as control. Plaque was scored and unstimulated salivary samples were collected at day 0 and after 4 days. A microbiological analysis was carried out for the salivary samples. Data were analyzed by using Student's t-tests.

Results: There was a statistically significant mean difference in plaque scores after using test paste (1.19 + 0.31) in comparison with those using placebo toothpaste (1.95 + 0.33). The difference between mean increase in colony forming units for the test and the placebo group was (25.2 + 8) × 10⁵ and (17.5 + 6.01) × 10⁵, respectively, which was statistically significant.

Conclusions: The findings of the study support the hypothesis that toothpaste containing salivary substitutes (bioXtra®) prevents dental biofilm formation and exhibits antimicrobial property when compared with a placebo dentifrice (fluoridated toothpaste marketed by Colgate-Palmolive).

bioXtra® versus biotène®: Patient Compliance

A double-blind, crossover study of biotène Oralbalance® and bioXtra® systems as salivary substitutes in patients with post-radiotherapy xerostomia.

Shahdad S A, Taylor C, Barclay SC, Steen I N & Preshaw P M, (2005) European Journal of Cancer Care 1, 319–326.

This study assessed the efficacy of the bioXtra® (BX) and biotène Oralbalance® (OB) systems in the treatment of post-radiotherapy xerostomia. In a double-blind, crossover study, 20 patients with post-radiotherapy xerostomia were randomly allocated to receive either OB then BX, or vice versa, each product for 2 weeks, with a 1 week wash-out period in between. Subject-based dry mouth scores derived from 100-mm visual analogue scales were recorded at days 0 and 14 of each 2-week period, together with subjective perception of changes in dry mouth symptoms. Both treatments were effective, resulting in reduction of visual analogue scale scores from day 0–14. Between-groups comparisons identified that BX achieved significantly better improvements compared with OB for the perception of dry mouth and improvements in speech and was also rated as more pleasant to use than OB (P<0.05). In conclusion, both treatments were effective in alleviating the symptoms of post-radiotherapy xerostomia, although BX achieved superiority in some of the outcomes assessed compared.

Table 4. Mean (SD) VAS scores of additional continuous variables recorded at day 14 by treatment group*

	Biotène Oralbalance	bioXtra	P
How effective was the product system at relieving your dry mouth?	45.3 (31.2)	65.5 (27.4)	<0.01
How pleasant was the taste of the toothpaste?	57.6 (26.4)	76.3 (24.3)	<0.01
How pleasant was the taste of the mouthwash?	61.7 (21.2)	78.7 (24.5)	0.02
How pleasant was the taste of the gel?	52.8 (29.2)	70.0 (34.4)	0.03
How pleasant did your mouth feel after using the gel?	54.1 (26.5)	67.2 (31.0)	0.10
How pleasant did your mouth feel after the whole product system?	63.8 (26.7)	73.1 (26.7)	0.06

Active Ingredients

Colostrum

Colostrum Treatment of Aphthous Ulcers on the Oral Mucosa - Oral findings in patients with primary Sjögren's syndrome.

Pedersen AM, Andersen TL, Reibel J, Holmstrup P, Nauntofte B. Department of Oral Physiology, Anatomy, Pathology and Medicine, School of Dentistry, University of Copenhagen, Denmark. Clin. Oral Investig. 2002 Mar; 6(1): 11-20.

Bovine colostrum is rich in antimicrobial substances and growth factors. The purpose of this open study was to examine and compare the interventional effects of daily use of bovine colostrum-containing oral hygiene products (CHP) on oral symptoms and findings in 20 patients with primary Sjögren's syndrome (pSS) and 20 age-matched patients with oral lichen planus (OLP). Objective oral measures and self-assessment of oral symptoms and general health were conducted before and after 90 days' use of CHP. The pSS patients had more systemic diseases, medication intake, oral dryness, poorer general health and lower salivary secretion than the OLP patients, who had the highest plaque index (PI) and the most mucosal soreness. Oral dryness and soreness were correlated to general health. In both patient groups unstimulated whole saliva flow rate (UWS) had increased, PI and periodontal pocket depth (PPD) were reduced, and general health and oral dryness and soreness had improved after using CHP. A decrease in hyphae was found in candida smears from both groups and in blastospores in OLP. A reduction in the extension of the mucosal lesions was observed in 15 OLP patients. Results suggested beneficial effects of intervention with CHP on oral symptoms, general health, UWS, PI, PPD and candidal load in two patient groups—pSS and OLP—representing different oral symptomatology.

Effects of Bovine Immune and Non-immune Whey Preparations on the Composition and pH Response of Human Dental Plaque.

Loimaranta V, Laine M, Soderling E, Vasara E, Rokka S, Marnila P, Korhonen H, Tossavainen O, Tenovu J. Institute of Dentistry and Turku Immunology Centre, University of Turku, Finland. Eur J Oral Sci 1999, Aug; 107 (4): 244–50.

Colostrum products from non-immunized cows (CP) and cows immunized with mutans streptococci (IP) were used as mouthrinses in a short-term human study. The acidogenic potential of the products was tested and found to be negligible in vivo before application to subsequent rinsing tests. At first all the participants received a professional tooth cleaning, after which they rinsed with one of the solutions (IP, CP, water) three times per day for 3 days. After each rinsing period the resting pH and decrease in plaque pH after sucrose challenge was determined, the amount of plaque was estimated, and all available plaque was collected. No significant differences were recorded in the composition or in the amounts of accumulated plaque. The resting pH values of plaques with low 'innate' pH were increased after the IP rinsing period. Surprisingly the lowest pH values after sucrose challenge were recorded in the IP plaques. The number of cultivable facultative flora or total streptococci were not affected by rinsings, but the relative number of mutans streptococci significantly decreased after the IP rinsing period when compared to the CP period. Thus the short term rinsing indicates favourable effects of bovine immune whey on human dental plaque.

Generation of Bovine Immune Colostrum against Streptococcus Mutans and Streptococcus Sobrinus and its Effect on Glucose uptake and Extracellular Polysaccharide Formation by Mutans Streptococci.

Loimaranta V, Tenuovo J, Virtanen S, Marnila P, Syvaaja E-L, Tubasela T, Korhonen H. Vaccine, Vol 15, No 11, pp 1261–1268, 1997.

Due to potential side effects of active immunisation by cariogenic mutans streptococci oral administration of passively-derived antibodies could be a more acceptable way to reduce colonisation and virulence of these microorganisms in human dentition. The aim of this study was to produce antistreptococcal immunoglobulins into bovine colostrums and explore the possible antibacterial mechanisms of these immunoglobulins against S. mutans. Specific serum IgG antibodies to whole cell antigens of both S. mutans and S. sobrinus increased rapidly in cows during immunisation and were high also in the final whey-product. Results indicate that bovine colostrum have a significant inhibitory potential against S. mutans, apparently dependent on the presence of specific IgG antibodies against S. mutans and S. sobrinus.

Possible oral health effect of colostrum containing dentifrices.

P Barkvoll. University of Oslo, Norway. IADR. Seq #45 - Oral Medicine & Pathology, 17 September 2005 RAI Congress Center Lounge Upper Floor-5.

Colostrum is milk that is produced during the first days post partum and it contains high concentrations of antibodies, enzymes and nutrients that protect the neonate against infection. Pedersen et al. (2002) recommended use of colostrum containing oral hygiene products for individuals suffering from oral medical problems. Objectives: To examine the clinical effect of three colostrum/enzyme

containing dentifrices without sodium lauryl sulfate (SLS) with two appropriate controls to the number of oral ulcers in a group of patients suffering from multiple recurrent aphthous ulcers (RAU). Other oral health parameters such as plaque index and gingival bleeding point were also to be investigated. Methods: All dentifrices were tested in a test period of 8 weeks. Each participant tested all five dentifrices. A wash out period of one week between each test period was necessary. 75 patients with RAU participated in this double-blind, crossover study. Only patients with at least one aphthous ulcer during a 6-week period were accepted. Friedman's non-parametric test was applied to assess evidence of overall differences between the dentifrices tested, followed by the non-parametric Wilcoxon matched-pairs signed-ranks test. Results: Brushing with the control, a 1.2 % SLS containing toothpaste resulted in a mean number of 9.23 new ulcers during the experimental period. The experimental colostrum containing toothpastes resulted in 3.98, 4.01 and 4.34 new ulcers respectively during the same period ($p < 0.05$). The plaque index was reduced significantly when the participants brushed with the colostrum/enzyme containing dentifrices compared to both controls. The colostrum containing dentifrices reduced the plaque index with 19.42 %, (mean value) compared to the controls with a percentage reduction measured to 5.67 %. Conclusion: It might be suggested that incorporation of bovine milk-derived colostrum into human oral health care products could be a serious and commercially active attempt to enhance and restore saliva's own antimicrobial capacity.

Growth Factors

Growth Factors and their Implications for Clinicians: a Brief Review.

McGeachie J, Tennant M, Department of Anatomy and Human Biology, University of Western Australia. *Aust Dent J* 1997; 42 (6): 375-80.

Growth factors play a vital role in both homeostasis and disease. In recent years considerable research has revealed the importance of growth factors in biology and they are now becoming incorporated in the clinical literature. Growth factors are peptides (protein fractions) that transmit signals within and between cells. They were discovered in the early 1960s as growth stimulants in tissue culture. It is now evident that growth factors play a comprehensive role in the modulation of tissue growth and development. The modes of action of growth factors are discussed with examples to pertinent clinical dentistry.

Lactoperoxidase System

Candida albicans inhibition by thiocyanate / H_2O_2 / peroxidase system in oral gel.

M. Ahariz, H. Dewèvre, J.-P. Perraudin, Ph. Courtois, Belgian Society for Fundamental and Clinical Physiology and Pharmacology, Leuven, November 17th 2007.

Introduction: In the presence of hydrogen peroxide (H_2O_2), lactoperoxidase (L) system produces hypothiocyanite (OSCN⁻) from thiocyanate (SCN⁻) and hypoiodite (OI⁻) from iodide (I⁻). In vitro, OI⁻ inhibits the growth of *Candida albicans* more efficiently than OSCN⁻ (Majerus & Courtois, 1992). However, lactoperoxidase affinity for SCN⁻ is higher than for I⁻ and induces in the oral cavity a preferential oxidation of salivary thiocyanate, making inefficient the use of iodide / L system in oral care products.

Objectives: This study aimed to document in vitro the competition between both substrates (SCN⁻ and I⁻), and to evaluate the effect of combining the SCN⁻ / peroxidase system with other exocrine proteins (lysozyme, lactoferrin, colostrum extract) in order to inhibit *Candida* growth.

Conclusion: In conclusion, the presence of SCN⁻ in saliva makes incorporation of I⁻ in anti-*Candida* oral gel useless since SCN⁻/I⁻ competition for lactoperoxidase favours the endogenous salivary substrate. However, incorporation of G/GOD/SCN⁻/L with other antimicrobial exocrine proteins in gel can delay yeast growth, opening other ways for further investigations.

Susceptibility of Anaerobic Micro-organisms to Hypothiocyanite produced by Lactoperoxidase.

Courtois Ph, Majerus P, Labbe M, Vanden Abbeele A, Yourassowsky E, Pourtois M., Free University of Brussels, Faculty of Medicine, Laboratory of Stomatology, Laboratory of Microbiology.

The susceptibility of Capnocytophaga ochracea, Eikenella corrodens, Eubacterium yurii, Fusobacterium nucleatum, Peptostreptococcus micros, Prevotella intermedia, Selenomonas sputigena, Wolinella recta to hypothiocyanite (OSCN⁻) produced by the lactoperoxidase system was tested. Results showed a decrease of bacterial survival rate after OSCN⁻ exposure, with an intra- and inter-species variability of 0 to 95% for C.ochracea, 34 to 100% for E. corrodens, 0-83% for E. yurii, 1-15% for F.nucleatum, 8-61% for P. micros, 0-100% for P.intermedia, 0-44% for S sputigena and 0-8% for W.recta. The survival rate did not correlate with the NADH/OSCN⁻ oxidoreductase activity present in lysed bacteria ($r = 0.3248$; $N = 15$; NS).

Treatment of Aphthous Patients by Enhancement of the Salivary Peroxidase System.

Hoogendorn H, Piessens JP. *J.Oral Path* 1987; 16: 425-427.

The inhibition of glucose-stimulated acid production by indigenous bacteria in human saliva is not achieved by the addition of up to 250uM hydrogen peroxide in vitro. However, in the presence of 2×10^{-4} % of hydroxyquinone and the same amount of Zn,

acid production is immediately terminated by addition of peroxide to only 25uM. No inhibition is observed when any one of these components is omitted. On the basis of these observations, a mouthrinse containing the same concentrations of hydroxyquinoline and Zn was prepared. Hydrogen peroxide was provided by including glucose oxidase and amyloglucosidase. This mouthrinse was used in a pilot clinical study of 64 patients subject to severe aphthous attacks which were not previously relieved by the use of a peroxidogenic toothpaste. After a two months period, during which these patients rinsed twice daily with 5ml of the mouthrinse, 45 patients reported relief of their symptoms. Of the remaining 19 patients, 17 reported no effect of using the mouthrinse, while 2 reported an exacerbation of their symptoms. The results of this study suggest that the mouthrinse may be effective for treating patients who suffer from severe aphthous attacks.

Inhibition of Streptococcus Mutans by the Lactoperoxidase Antimicrobial System.

Edwin L Thomas, Kate A Pera, Keith W Smith, Alice K Chwang. Dept Biochemistry, St Judes Childrens Research Hospital, Memphis, Tennessee. *Infect and Immunity*, Feb 1983. p 767- 778.

Inhibition of bacterial metabolism by the lactoperoxidase (LP)-hydrogen peroxide (H_2O_2)-thiocyanate system was studied with representatives of serotypes a through g of *Streptococcus mutans*. The aims were to determine whether the amount of H_2O_2 released from these catalase-negative bacteria is sufficient to activate the LP system and whether these oral bacteria are resistant to inhibition by the LP system, which is active in human saliva. When the washed, stationary phase cells were incubated aerobically with LP, thiocyanate, and glucose (Glc), greater than 90% inhibition of Glc utilization and lactate production was obtained with strains that released large amounts of H_2O_2 (BHT, FA-I, OMZ 176): 20 to 50% inhibition was obtained with strains that released about half as much H_2O_2 (B-13, Ingbritt); and no inhibition was obtained by strains that released only small amounts of H_2O_2 (AHT, HS-6, GS-5, LM-7, OMZ-175, 6715-15). Inhibition was most effective at pH 5, whereas release of H_2O_2 and accumulation of the inhibitor (hypothiocyanite ion) were highest at pH 8. With H_2O_2 releasing cells from early stationary phase, preincubation with Glc abolished inhibition, though it did not influence H_2O_2 release. Cells harvested 24 h later were depleted of sulfhydryl compounds or disulfide compounds (reduced or oxidised glutathione, cysteine or cystine). This preincubation increased cell sulfhydryl content but had no effect on H_2O_2 release. All strains were inhibited when incubated with LP, thiocyanate, and added (exogenous) H_2O_2 . Smaller amounts of H_2O_2 were required to inhibit at pH 5 and larger amounts were required to inhibit cells preincubated with Glc or with Glc and sulfhydryl or disulfide compounds. The results indicate that pH, amount of H_2O_2 , cell sulfhydryl content and stored carbohydrate content determine susceptibility to inhibition.

Determination of the production of OSCN⁻ in an enzyme based toothpaste and gel for use in dry mouth sufferers in vitro.

Date on file, Biopole S.A., January 2006, Les Isnes, Belgium.

Lactoperoxidase system generated hypothiocyanite ions (OSCN⁻) and hypothiocyanous (HOSCN) are inhibitory against a number of oral bacteria. HOSCN/OSCN⁻ are the molecules produced by the Salivary Peroxidase system in all healthy mouths and which, in conjunction with other oral systems, helps maintain healthy oral conditions by regulating the presence of bacteria and facilitating the mouth's natural ecosystem. Commercially available products bioXtra[®] toothpaste and bioXtra[®] gel comprise the complete lactoperoxidase system. The aim of this study was to assess the levels of HOSCN/OSCN⁻ generated by two bioXtra[®] products – bioXtra[®] Moisturising Gel and bioXtra[®] Mild Toothpaste containing the lactoperoxidase system and to check if there are any inhibitory effects due to the presence of the saliva.

1 gr of bioXtra[®] paste was added to tubes containing 2 ml non-sterilized human whole saliva which was previously incubated at 37°C. After a short period of time (15 seconds to 2 minutes), a 100 µl aliquot was directly withdrawn for analysis of the amounts of HOSCN/OSCN⁻ generated. After a longer period (3 minutes to 360 minutes), a 100µl aliquot was added to the tubes and the tubes were vortexed for 30 sec. Experiments with 1gr of the same paste and the same gel but without containing the lactoperoxidase system served as controls. The peroxidase activity and thiocyanate concentration was determined of in the non-sterilized saliva used for each test before the addition of the bioXtra[®] products. The HOSCN/OSCN⁻ yield ranged from 200 – 300 µM for the bioXtra[®] toothpaste and from 15 to 300µM for the bioXtra[®] gel while the salivary levels of HOSCN/OSCN⁻ before the addition of each bioXtra[®] products were respectively between 10 to 40µM and 2 to 5 µM respectively. Our results show that it is possible to significantly increase the levels of the lactoperoxidase system generated antimicrobial component HOSCN/OSCN⁻ in saliva by means of a dentifrice or a gel. Toothpastes and gel are important vehicles for delivery of different antimicrobial agents.

Moreover, the amounts of HOSCN/OSCN⁻ generated are high enough to exert an antibacterial effect on the isolated bacteria but not enough against the biofilm bacteria. In addition, the bioXtra[®] toothpaste and bioXtra[®] gel may offer an option for pain relief, and medium-term restoration of a more normal ability to eat and drink, for patients with painful oral lesions of a variety of aetiologies.

Lysozyme, Lactoferrin & Lactoperoxidase System

Salivary Lysozyme, Lactoferrin and Peroxidases: Antibacterial Effects on Cariogenic Bacteria and Clinical Applications in Preventive Dentistry.

Tenovuo J, Lumikari M, Soukka T. Department of Cariology, University of Turku, Finland. *Proc Finn Dent Soc* 1991; 87 (2): 197-20.

Many antibacterial agents in human saliva are known to have bacteriostatic or bactericidal effects on cariogenic bacteria, in particular against *Streptococcus mutans*. Studies have usually been conducted with purified agents (proteins) in vitro. Very little proof exists to show that they also have an effect on oral cariogenic bacteria in vivo. Recent studies have shown that some salivary systems can act synergistically against *Streptococcus mutans*.

Such synergistic antibacterial activity is likely to exist in the human mouth. Attempts to enhance the anticariogenic properties of saliva have been made by adding antimicrobial proteins such as peroxidase, lactoferrin and lysozyme to oral health products. Although clinical evidence is still limited, the idea of using such antimicrobial agents – ‘natural antibiotics’ – rather than synthetic agents against cariogenic bacteria seems promising.

Lactoferrin

A Component of Innate Immunity prevents Bacterial Biofilm Development.

Pradeep K Singh, Matthew R, Parsek E, Peter Greenberg and Michael J Walsh, Department of Internal Medicine, Department of Microbiology, Department of Physiology and Biophysics, Howard Hughes Medical Institute, University of Iowa, Iowa City USA. *Nature* Vol. 417 May 2002; 552.

Antimicrobial factors form one arm of the innate immune system, which protects mucosal surfaces from bacterial infection. These factors can rapidly kill bacteria deposited on mucosal surfaces and prevent acute invasive infections. In many chronic infections, however, bacteria live in biofilms, which are distinct, matrix-encased communities specialised for surface persistence. The transition from a free-living, independent existence to a biofilm lifestyle can be devastating, because biofilms notoriously resist killing by host defence mechanisms and antibiotics. We hypothesized that the innate immune system possesses specific activity to protect against biofilm infections. Here we show that lactoferrin, a ubiquitous and abundant constituent of human external secretions, blocks biofilm development by the opportunistic pathogen *Pseudomonas aeruginosa*. This occurs at lactoferrin concentrations below those that kill or prevent growth. By chelating iron, lactoferrin stimulates twitching, a specialised form of surface motility, causing the bacteria to wander across the surface instead of forming cell clusters and biofilm. These findings reveal a specific anti-biofilm defence mechanism acting at a critical juncture in biofilm development, the time bacteria stop roaming as individuals and aggregate into durable communities.

Xylitol

Comparison of Erythritol and Xylitol Saliva Stimulants in the Control of Dental Plaque and Mutans Streptococci.

K.K. Mäkinen, K.P. Isotupa, T. Kivilompolo, P.L. Mäkinen, J. Toivanen, E. Söderling International Institute for Preventive Dentistry, University of Turku, and Pello Public Health Center, Pello, Finland. *Caries Res*, 2001; 35:129-135.

The effect of 2-month usage of saliva-stimulating pastils containing either erythritol or xylitol was studied in a cohort of 30 subjects assigned to the respective polyol groups (n = 15). The daily consumption level of both polyols was 5.2 g, used in 5 daily chewing episodes. The mean weight of total plaque mass (collectable during a standard period of 3 min from all available tooth surfaces) was reduced significantly in the xylitol-group, while no such effect was observed in the erythritol-group. This reduction in plaque mass was accompanied by a significant reduction in the turbidity readings (A660) of aqueous plaque suspensions; no such effect was observed in the erythritol-group. However, plaque protein levels did not differ between baseline and endpoint in either polyol group. The plaque and salivary levels of *Streptococcus mutans* and plaque levels of total streptococci were reduced significantly in the xylitol-group, while no such effect was detected in the erythritol-group. However, either polyol regimen had no effect on plaque levels of *S. sobrinus*. The results suggest that systematic use of xylitol-containing saliva stimulants may be more effective in controlling some oral-hygiene-related and caries-associated parameters than similar use of erythritol-containing products. The results also speak for a special relationship between xylitol and *S. mutans*. However, owing to the great potential of erythritol as a caries-reducing agent - based on the tetritol nature of erythritol - the present laboratory results should be considered preliminary and subject to verifying clinical studies.

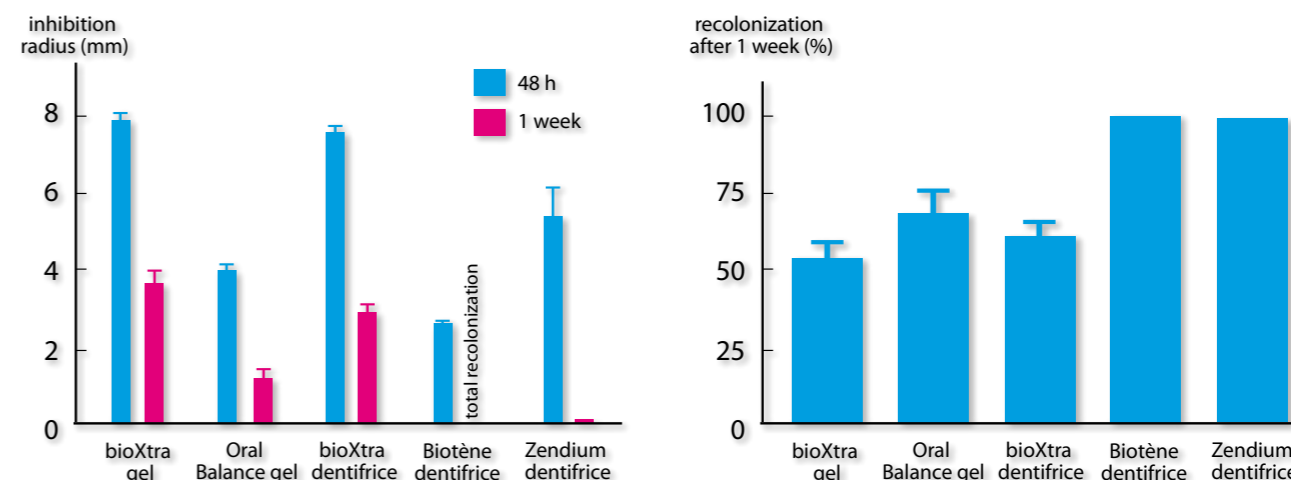
Efficacy against *Candida albicans*

Effect of saliva-mimicking oral care compounds upon *Candida albicans* ATCC 10231.

Ahariz M., Courtois P. Communication at the Meeting of the Belgian Society of Fundamental and Clinical Physiology and Pharmacology. Leuven (Belgium), 20th November, 2004. abstract in *Plügers Arch. - Eur J. Physiol.* 449: R1-R13, 2005.

This investigation aimed to evaluate, by a radial diffusion test, the effect of 5 different oral care products containing proteins from bovine milk against *Candida albicans*. For this purpose, 25 µl of yeast suspension (*Candida albicans* ATCC 10231 in 30 g/l Sabouraud liquid medium) adjusted to a 600 nm absorbance of 0.800 was dispersed on Sabouraud/ Gentamycin/ Chloramphenicol agar solid medium in centrally gel-punched plates and then 1g of oral care product was deposited in the central well. Inhibition radius

after a 48-hours incubation was significantly higher with oral care products containing lactoperoxidase, lactoferrin, lysozyme plus colostrum whey factors. Inhibition has been shown reversed after a one-week incubation at 37°C but in a less extent when oral care products contain lactoperoxidase system, Lactoferrin, lysozyme plus colostrums whey factors. In conclusion, toothpaste (bioXtra®) and oral gel (bioXtra®) containing colostrum whey factors besides antimicrobial proteins have shown a higher and a longer candida activity than other oral care products incorporating only the last ones.



Sodium Lauryl Sulfate (SLS)

Sodium lauryl sulfate and recurrent aphthous ulcers - A preliminary study.

Herlofson BB, Barkvoll P, Department of Oral Surgery and Oral Medicine. Dental Faculty, University of Oslo, Oslo, Norway, *Acta Odontol Scand* 1994; 52: 257-259. Oslo.

Sodium lauryl sulfite (SLS), asynthetic detergent commonly used in dentifrices, is an effective denaturant. The aim of the present study was to investigate the effect of SLS in patients with recurrent aphthous ulcers (RAU). Ten patients with multiple minor RAU participated in the study. The mean incidence of RAU was 17.8 during a 3-month period before the study. The patients used a dentifrice containing 1.2% SLS for a test period of 3 months followed by a dentifrice without SLS for the same time period. The results showed a statistically significant decrease in the number of aphthous ulcers from 14.3 after using the SLS-containing dentifrice to 5.1 ulcers after brushing with the SLS-free dentifrice (p<0.05). It is suggested that the denaturing effect of SLS on the oral mucin layer, with exposure of the underlying epithelium, induces an increased incidence of recurrent aphthous ulcers.

Importance of Saliva in Oral Health

Antimicrobial Function of Human Saliva – How Important is it for Oral Health?

Tenovuo J. Institute of Dentistry and Turku Immunology Centre, University of Turku, Finland. *Acta Odontol. Scand.* 1998 Oct; 56(5): 250-6.

Human saliva contains a number of physical physiochemical and chemical agent that protect oral tissues against noxious compounds, in particular those produced by various microorganisms. Among such protective factors, the flushing effect of saliva flow is the most important one, not only because it so effectively removes exogenous and endogenous microorganisms and their products into the gut but also because a steady supply of saliva guarantees continuous presence of both non-immune and immune factors in the mouth. A great number of studies with controversial results have been published regarding various individual agents and their possible association to oral health, particularly to dental caries. It appears that no single chemical agent is far more important than the others. For example patients with IgA deficiency have normal levels of non-immune defence factors and often display a compensatory increase in the other immunoglobulin isotypes. The concerted action of all agents in whole saliva, both saliva- and serum-derived, provides a multi-functional protective network that is collapsed only if salivary flow rate is substantially reduced. In this mixture of defence factors, many show additive or even synergistic interactions against oral pathogens. Increased knowledge of molecular functions of various agents has made it possible to prepare oral hygiene products that include host-derived antimicrobial agents instead of synthetic agents. Although clinical efficacy of some products is still unsatisfactory and poorly described, new technologies, for example the production of specific antibodies against oral pathogens, may considerably improve the anti-microbial power of these products.