



Title	Antibiotic sensitivity of putative pathogens in Chinese periodontal patients
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2617 LonqoVital® in the Treatment of Sjögren's Syndrome. A PEDERSEN*, N GERNER, I. PALMVANG and M HØIER-MADSEN (Dental and Ophthalmol Dept., Bispebjerg Hospital, Statens Serum Inst., Copenhagen, Denmark)

LonqoVital® (LV) is a herbal based tablet enriched with the recommended doses of vitamins. Previous studies have demonstrated a preventive effect of the tablets on recurrent aphthous ulcers (*J Oral Pathol Med* 19 371-5, 1990) and in a reduction of gum bleeding (unpublished observations). The clinical benefit in both these groups of patients could possibly be ascribed to an augmentation of cellular immune competence. The purpose of the present study was to investigate the effect of 4 months' daily intake of LV on secondary and primary disease activity markers in 40 patients with Sjögren's syndrome (SS) in a placebo-controlled, double-blind, randomised clinical, 8 months' cross-over study. Gr.A received LV during the first 4 months and Gr.B LV during the last 4 months. Wilcoxon matched-pair signed rank test and Mann-Whitney U-test were applied on intra-intergroup data, respectively. Unstimulated salivary flow rate increased during the LV period in Gr.A ($p < 0.001$). Stimulated salivary flow rate increased during the subsequent 4 months on placebo in Gr.A ($p < 0.05$), and in Gr.B in the LV period ($p < 0.05$). Rose bengal score decreased in Gr B during ($p < 0.01$) and in Gr.A after the LV intake ($p < 0.05$). During the last 4 months both groups responded by increased serum levels of the pancreatic fraction of α -amylase (Gr.A: $p < 0.0001$; Gr.B: $p < 0.01$), of IgM (Gr.A and B: $p < 0.001$), and levels of circulating immune complexes decreased (Gr.A: $p < 0.05$; Gr.B: $p < 0.001$). It is concluded that LV has a beneficial and prolonged effect on some primary and secondary disease markers in SS. Supported by Paramedical A/S, Denmark.

2618 Histamine Receptor 2 may be a pacemaker for Neutrophil activation W.K.KIM-PARK*, M.A.MOORE AND M.J.KOWOLIK (Indiana University School of Dentistry, Dept. of Periodontics, Indianapolis, IN 46202, USA)

There is continuing debate over the relative importance of the H_1 and H_2 histamine receptors on neutrophil granulocytes, in relation to the generation of reactive oxygen species. To determine the roles of histamine subreceptors, histamine and its subreceptor-ligands were tested in primed human neutrophils. Human neutrophils were separated in our laboratory by a standard method (Kim-Park et al. *Ann NY Acad Sci* 832: 394-404, 1997). The respiratory burst activity was measured by Luminescent-dependent Chemiluminescence (CL) and compared by a two-tailed paired Student's t-test. Histamine alone did not significantly alter the CL generation in primed human neutrophils, but H_1 -antagonist (diphenhydramine, 10^{-6} M) pretreatment inhibited histamine-mediated or FMLP-mediated CL significantly ($p < 0.0001$) while the H_2 -antagonist, cimetidine (10^{-6} M), reduced the FMLP-mediated CL generation by 40%. H_2 -antagonist alone increased the control CL, indicating a disinhibition of H_1 -receptor mediated stimulation. The degree of disinhibition by the H_2 -antagonist was similar to that of a protein kinase A (PKA) inhibitor, indicating an involvement of the c-AMP pathway. When the cells were treated with the H_1 -antagonist and H_2 -agonist, dB-c-AMP, simultaneously, the degree of inhibition of CL was the same as with the H_1 -antagonist only. Considering a similar sensitivity of CL generation to EGTA-, H_1 -antagonist- or quacranine-treatment ($p < 0.0001$) with histamine, the H_2 -receptor mediated increases in Ca^{2+} may activate the respiratory burst activity via the PLC cascade. It also may activate the residual H_1 -receptor via the c-AMP pathway even in the presence of the H_2 -antagonist, opposing the H_1 -site activity. We conclude that histamine receptor 1-mediated neutrophil activation via the PLC pathway may be upstream of the H_2 -site and is regulated via H_2 -mediated activation of PKA, as a negative feedback mechanism, when the increases in Ca^{2+} by the H_1 -receptor is excessive.

2619 Antibiotic sensitivity of putative pathogens in Chinese periodontal patients K.Y.ZEE*, D.H.LEE and L.P.SAMARANAYAKE (Faculty of Dentistry, The University of Hong Kong)

The aim of the present study was to investigate the antibiotic sensitivity of the putative periodontal pathogens cultivable in Chinese patients suffering from advanced periodontal disease. Subjects with at least one tooth with severe periodontal involvement scheduled for extraction and without taking any antibiotics for at least 3 months were recruited. Subgingival plaque sample was obtained from each tooth by inserting 3 sterile paper points into the bottom of the pocket before extraction. Each sample was dispensed in reduced transfer fluid and cultured on non-selective media using anaerobic techniques to obtain pure isolates. After subculturing, all pure isolates were identified based on morphology, chemical and biochemical tests. Five commonly used antibiotics in Hong Kong i.e. minocycline, tetracycline, amoxicillin, erythromycin and metronidazole were selected and test against the identified species using the Ciest® system. A total of 24 samples were obtained from 19 subjects. From the 24 samples, 33 isolates containing 11 species were tested against the 5 antibiotics. Some of the MIC (μ g/mL) values of the more important species were shown below.

2620 Dentin Permeability *In Vitro* after Application of Tartaric Acid Solutions. R. MONGIORGI, M.L. TEDALI, A. LUCCHESI, S. CHERSONI, C. PRATI (Univ. of Bologna and Ferrara, ITALY)

Introduction: Preliminary investigations demonstrated that tartrate salts solutions are able to create a layer of non-homogeneous crystals able to close dentinal tubules and to reduce fluid flow rate calculated using a pressure apparatus. **Purpose:** The aim of this study was to evaluate the ability of a new solution constituted by tartaric acid (TA) solutions 0.1M (pH 3.5) to reduce *in vitro* the fluid flow, also defined as dentin permeability (L_p). Dentin discs from human molars were prepared and treated with 1M EDTA for 2 mins to remove the smear layer from the surface and to calculate the maximum rate for each disc (to which an arbitrary value of 100% was assigned). Discs were connected with the pressure apparatus working at 0.5 psi. A new smear layer was re-created in half of the samples. Solution was applied for 2 mins, washed with water for 30 seconds and fluid flow rate re-measured. 35% phosphoric acid was then re-applied for 2 mins, washed and fluid flow rate re-calculated. SEM analysis was obtained for several samples.

Group A EDTA 100±0.1	plus TA solution 68.5±23.1	plus phosph acid 67.1±29.7
Group B EDTA 100±0.1	smear layer 58.7±44.7	plus TA solution 34.07±22.4
		plus phosph acid 57.23±22

Results: (mean fluid flow rate ±SE, Student t test; N= 24 for each group):

SEM showed numerous crystals inside dentinal tubules. **Conclusions:** the application of TA solution reduced the fluid flow rate and modified smear layer morphology. This study suggests that TA solution may protect dentin from diet acids and may reduce dentin hypersensitivity.

	Minocycline	Tetracycline	Amoxicillin	Erythromycin	Metronidazole
<i>A. a.</i>	0.38	0.75	>256	>256	8
<i>Fusobacterium</i> spp	>256	>256	>256	>256	>256
<i>P. gingivalis</i>	<0.016	<0.016	<0.016	<0.016	<0.016
<i>P. intermedia</i>	<0.016	0.016-0.125	0.016-0.38	<0.016	<0.016
<i>Selenomonas</i>	8	24	0.125	1.5	2
<i>Campylobacter</i> spp	0.25-0.5	0.125-0.25	0.032-0.094	0.094-0.25	0.094-0.38

Some of the organisms yielded MIC values comparable to those in the literature. However, due to the sparsity of data, further work is required to establish antibiotic trends of periodontal pathogens from the region. **In conclusion,** the results suggested that a careful selection of antibiotic may be necessary during the treatment of periodontal diseases in Chinese patients. (This study was supported by HKICRCG Grant No. 10201262)

2621 *In vitro* mucosal model predictive of bioadhesives in the mouth. D.PATEL¹, S.STEVENSON, A.SMITH¹, N.GRIST¹, P.BARNETT², J.SMART¹ (School of Pharmacy and Biomedical Sciences, University of Portsmouth, U.K. SmithKline Beecham Consumer Healthcare, Weybridge, U.K)

The formulation of a drug/carrier complex that can be distributed and retained for extended periods throughout the oral cavity would be advantageous in the treatment of local conditions, such as aphthous stomatitis, oral candidiasis, and gingivitis. The aim of this study was to develop an *in vitro* system to allow the prediction of *in vivo* performance of bioadhesive agents, such as solutions of polymeric drug carriers in the oral cavity. Polymer adsorption onto human buccal cell surfaces was investigated using a lectin inhibition technique involving an avidin-biotin complex and a colourimetric detection system. 0.5% w/v polymer solutions in isotonic saline (pH 7.6) were left in contact with a standardized number of freshly collected human buccal cells (from healthy volunteers aged between 19-40 years), at 30°C. The cells were then subsequently exposed to 10 mg L⁻¹ biotinylated lectin from *Canaavalia ensiformis* and 5 mg L⁻¹ streptavidin peroxidase at 30°C. Thirty three polymer solutions were screened (including anionic, cationic and non-ionic polymers), N=5, each time against an isotonic saline control. Polymer adsorption in terms of masking of lectin binding sites was measured and expressed as a percentage reduction in the rate of o-phenylenediamine oxidation over 1 min at an absorbance of 414nm. Chitosan, polyacrylamide and cetylpyridinium chloride gave significantly greater masking of lectin binding sites ($p < 0.05$ multiple comparison-Tukey test) on the surface of buccal cells, with chitosan showing over 86% masking of lectin binding sites. This assay was confirmed using direct staining techniques (involving Alcian Blue and eosin stain). It was concluded that this assay can be used to reliably assess polymer adhesion to the buccal mucosa, and therefore develop more effective retentive polymer/drug formulations to treat localised disorders of the oral cavity. This study was supported by SmithKline Beecham Consumer Healthcare, Weybridge, UK.

2622 Cutaneous Hyperalgesia Following a Mild Cold Injury on Human Skin. C.R. BERGEY*, K.C. KAJANDER, D.A. SIMONE (University of Minnesota, Schools of Dentistry and Medicine, Minneapolis, MN, USA)

Hyperalgesia to cold is often present following nerve and tissue injuries, whereby gentle cooling of the skin produces a painful sensation. The underlying peripheral neural mechanisms that mediate cold hyperalgesia are poorly understood. It is possible Aδ and/or C-fiber nociceptors become sensitized to cold stimuli (reduced threshold and increased response to suprathreshold stimuli). In this study, we developed a model of cutaneous hyperalgesia produced by applying a cold conditioning stimulus (CS), -15°C for 20 sec, to the skin. The subjects were asked to estimate the magnitude and quality of cold and heat pain sensation evoked by a wide range of stimulus temperatures using a 1 cm² Pelletier type thermode before and beginning 5 minutes after the CS. Cold (28 to -4°C) and heat (38 - 48°C) stimuli (5 sec duration) were delivered from a base temperature of 30°C. Magnitude estimates of pain sensation (heat and cold) were normalized. The CS produced hyperalgesia to cold, heat and mechanical stimulation. Cold pain threshold decreased an average of 10.0 ± 3.9°C (n=10) ($p < 0.0001$) and the threshold for heat pain decreased 4.7 ± 0.9°C (n=6) ($p < 0.02$). In addition, magnitude estimates of pain evoked by suprathreshold cold and heat stimuli increased significantly after the CS, and the quality of cold pain often changed to a burning sensation. The CS also produced a surrounding area of secondary hyperalgesia to mechanical stimuli that measured 46.1 ± 28.8 cm² (n=6). **In conclusion,** hyperalgesia to thermal and mechanical stimuli develops following a mild cold injury. This model may be useful to study neural mechanisms that underlie cold hyperalgesia. This study was supported by grants from the American Academy of Orofacial Pain and NIH (NS 31223)

2623 Acids Evoke the Wiping Response in Frogs at Different pHs D.T.HAMAMOTO*, M.W.FORKEY*, W.L.DAVIS*, and K.C.KAJANDER^{1,2}. (Departments of Oral Science¹, and Cell Biology & Neuroanatomy², and Graduate Program in Neuroscience³, University of Minnesota, Minneapolis, MN, USA)

Application of acetic acid (AA) to hindlimb skin of frogs decreases subepithelial pH and evokes a quick wiping of the exposed skin. In previous studies, 90% of frogs tested responded to AA at pH 2.20. However, sulfuric acid (SA) and formic acid (FA) at pH 2.20, did not evoke the wiping response. Different acids may require different pHs to evoke the wiping response. By comparing these differences in pHs with differences in properties of these acids we hope to gain insight into how acids evoke the wiping response. Thus, the purpose of this study was to determine, for several acids (AA, FA, oxalic (OA), SA, and hydrochloric (HCl)), the pH required to evoke the wiping response in frogs. For each acid, a series of 11 solutions was made by serially diluting concentrated acid. A single drop of each solution in a series was applied starting with the least acidic solution until the frog wiped its hindlimb. The solution that evoked the wiping response was recorded as the threshold solution. The pH of the threshold solution differed between the acids (ANOVA, $P < 0.05$). The pH of the threshold solution for AA was the highest (pH 2.42) while the pH of the threshold solution for HCl was the lowest (pH 0.83). The pH of the threshold solutions for FA (pH 1.91), OA (pH 1.41), and SA (pH 0.97) were intermediate. Thus, several acids in addition to AA can evoke the wiping response in frogs, however, the pH required differs between acids. These acids differ in their dissociation constants (Ka) with AA having the smallest Ka and HCl having the largest Ka. This suggests that the level of dissociation or a related property (e.g. osmolality or ionic strength) may be responsible for evoking the wiping response. This research was supported by grants from the National Institutes of Health (NS33908 and DE00270).

2624 Evidence for Galanin in Nerve Fibers and Nerve Endings in the Gingiva of Rats. Y. KORKMAZ*, M.A. BAUMANN, F.F. EIFINGER, H. SCHRÖDER (Dental School and Institute for Anatomy, Univ. of Cologne, Germany)

The presence of substance P (SP) and calcitonin gene-related peptide (CGRP) in the epithelium and lamina propria of the gingiva and co-localization of neuropeptide galanin (GAL) with SP and CGRP in the dorsal horn neurons of the spinal cord and in the small neurons of the trigeminal ganglion raise the possibility for an existence of GAL in the epithelium and lamina propria of the gingiva. This experiment was designed to test this hypothesis in the molar gingiva of 12 week old Wistar rats [n=12]. Tissues were perfusion- and post-fixed, decalcified in 4N formic acid, frozen, sectioned at 50 μ m and immunoreacted as free-floating sections with rabbit antiserum against galanin [1,2000] using the avidin-biotin peroxidase complex method. Light microscopic observation of sections demonstrated the presence of GAL-immunoreactivity (GAL-IR) in the gingiva of the rat. GAL-IR-immunoreactive (GAL-ir) elements were usually distributed around blood vessels of various sizes in the lamina propria and single processes and endings were also observed beneath the epithelium and in the proppal-epithelial junction. These results may provide a morphological basis for possible actions and interactions of GAL in the gingiva. We conclude that the perivascular distribution of GAL-IR is compatible with a role for GAL in hemodynamic regulation. Furthermore, the existence of GAL-ir elements in the proppal-epithelial junction would suggest a possible involvement of GAL as a regulator in the modulation of nociception.