

1817 The Role of Mast Cells in Periodontal Disease
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The discharge of heparin into the local environment due to the disruption of the gingival mast cells in periodontal disease is one of the factors simulating alveolar bone resorption. The aim of the present study was to estimate the activity of the mast cells in different stages of periodontal disease. Gingival biopsies were obtained during periodontal surgery from 24 patients of 27 to 45 years. Control specimens from 7 early lesion specimens from 5 established lesion specimens from 4 and advanced lesion specimens from 8 patients were stained by Pappenheim. The activity of mast cells was estimated in three stages using degranulation index by V Valdes (Männiste et al Phlebology Tallinn 1980 p 11). The granulation index was as follows: in case of intact periodontal tissues 1.28 ± 0.12 , in early lesion of periodontal disease 1.58 ± 0.11 in case of established lesion - 1.77 ± 0.10 in case of advanced lesion - 1.99 ± 0.14 . The difference between the groups was significant ($p < 0.05$). We conclude that the granulation index was increased proportionally to the stage of periodontal disease. This study was supported by Estonian Science Foundation, Grant 19.

1818 Cell Interaction between periodontal ligament fibroblasts and Malasse z epithelial rests in vitro T Inoue* S Hashimoto Y Enokiya and M Shimono Department of Pathology Tokyo Dental College Chiba Japan

The purpose of this study is to investigate the interaction between cells from Malasse z epithelial rests (MER) and fibroblasts of periodontal ligament (PLF) co cultured in vitro. Both MER and PLF derived from porcine periodontal ligament were isolated and subcultured on type I collagen coated culture dish in a MEM supplemented with antibiotics and 15% fetal bovine serum. Cultured MER and PLF were divided into next three groups and co-cultured: 1) ERM cultured on the multilayer of PLF 2) PLF cultured on the multilayer of ERM 3) ERM cultured with PLF on the same cell numbers of 2×10^5 cells. Cells were observed by phase contrast microscope. LM SEM TEM confocal laser microscopy and cell growth and occupation ratio were also investigated. When MER were cultured alone they appeared to form typical pavement like sheets and exhibited slow proliferation curve. When PLF were cultured alone they arranged in the characteristic wavy stromal pattern and had a marked increasing with arithmetical series of proliferation. MER were grown well on the multilayers of PLF but PLF were not grown on the multilayers of MER. When MER were co-cultured with PLF occupation ratios of MER were increased day by day: 3 75 ± 1.21 after 2 days 7 30 ± 5.70 after 4 days 34 62 ± 8.10 after 6 days and 57 82 ± 13.85 after 9 days of co-culture. These occupation ratios of the MER in later time period were significantly greater than those of early time periods ($p < 0.001$). Ultrastructurally only a few desmosome like structures but not basement membrane were detected between MER and PLF. These results suggest that MER inhibit the growth of the PLF in vitro. MER start growing on the connective tissue by stimuli such as endodontic lesions, and this may become lining epithelium of odontogenic cyst in vivo.

1819 Do Gingival Keratinocytes Express the CD14 Lipopolysaccharide Receptor? M S TONETTI* M SCHUSTER M IMBODEN and N P LANG (School of Dental Medicine, University of Bern, Switzerland)

Epithelia are an integral active component of gingival host defenses. Keratinocyte (KC) activation can be induced by a variety of interactions with bacteria. KC exposure to LPS "in vitro" however did not increase ICAM 1 and IL 8 expression above basal levels. The aims of this investigation were: i) to study "in situ" CD14 expression on junctional epithelium (JE) KC in healthy gingiva and ii) to establish the levels of CD14 expression in gingival KC cultures. 6 µm cryostat sections from a total of 12 gingival biopsies were stained with a sensitive 3 stage immunoperoxidase technique using the UCHM-1 anti CD14 mAb. Sporadic positive cells possibly mononuclear phagocytes were detected in the JE. KC in the JE and other gingival epithelia were consistently negative. In the connective tissue both leukocytes capillary loops and fibroblast like cells were CD14+. Expression of CD14 protein and mRNA on KC cell cultures was determined by FACS analysis (UCHM 1 and MY4 mAb) and RT PCR (primer set yielding a 529 bp product). CD14 mRNA was detected in all tested cultures (N=12). Median channel fluorescence of gingival KC scanned after indirect immunofluorescence staining with anti CD14 mAb was 49 (range 42-58). This was significantly higher than the isotype control fluorescence of 34 (range 33-34). CD14 complement mediated cytotoxicity of KC cultures resulted in a 12±6% decrease in cell viability. It is concluded that CD14 expression was below detection level in gingival biopsies. Gingival KC cultures express low levels of CD14 mRNA and protein. The biologic significance of such expression remains to be elucidated. Supported by Swiss National Science Foundation Grant #32 37763 93 Clinical Research Foundation University of Bern.

1820 "In situ" Detection of DNA Breaks in Healthy Human Gingiva D CORTELLINI* M S TONETTI and N P LANG (School of Dental Medicine University of Bern Switzerland)

Programmed cell death (apoptosis) is a physiologic process characterized by the presence of DNA fragmentation. It plays a fundamental role during tissue development and homeostasis and is particularly evident in tissues with high turnover rates. Gingival epithelia are thought to express some of the fastest turnover rates. In the gingiva however apoptosis has not been examined so far. Aim of this study was to evaluate in situ the presence and distribution of cells with DNA breaks in healthy human gingiva. 6 µm cryostat sections from 12 healthy gingiva biopsies were fixed in acetone and processed for TdT mediated dUTP biotin nick end labeling (TUNEL). Biotin 16 dUTP (4µM) was incorporated at the 3' end of DNA breaks by incubation with 0.4U/µL terminal deoxynucleotidyl transferase (TdT). The signal was amplified with avidin biotin horseradish peroxidase complexes and detected using DAB as chromogenic substrate. Positive (pre incubation with DNase) and negative (omission of TdT) controls were run with each series. Results indicated that DNA breaks were consistently present in the junctional epithelium. Only sporadic cells were positive in the superficial layers of the sulcular and oral epithelia. DNA breaks were also evident in the infiltrated connective tissue (ICT) where the median density of positive cells was 265 cells/mm² (133-495). It is concluded that DNA breaks detectable by the TUNEL technique are present in normal human gingiva. Highest expression was in the junctional epithelium and the ICT. Supported by Swiss National Science Foundation Grant #32-37763 93 Clinical Research Foundation and Italian Society of Periodontology.

1821 Immunocompetent cells in periodontal tissues following experimental tooth movement V VANDVSKA-RADUNOVIC* I HALS KVINNSLAND S KVINNSLAND R JONSSON University of Bergen Norway

The aims of the study were to evaluate the distribution of immunocompetent cells in periodontal tissues and to quantify their recruitment incident to orthodontic tooth movement. The first right maxillary molar was moved mesially for 1, 3, 7, and 14 days in 27 young rats. The left side served as control. At the end of each experimental period the animals were perfused. The jaws were excised, postfixed, demineralized and sagittally sectioned at 10-12 µm on a cryostat microtome. The immunohistochemical procedure was carried out on alternate serial sections using the avidin biotin peroxidase method. Antibodies against CD4+ (helper T lymphocytes), CD11b+ (macrophages dendritic cells), CD43+ (T lymphocytes granulocytes) and Ia antigen expressing cells were used. On the control side mean counts of immunolabelled cells in the periodontal ligament (PDL) showed the highest distribution of CD11b+ (243.4 ± 52.4 SD) and Ia antigen expressing cells (128.0 ± 19.1 SD) while CD4+ and CD43+ cells were scarcely found, expressing high variability (41.1 ± 24.4 SD and 4.5 ± 3.1 SD respectively). Significant increase in the number of CD11b+ cells in the PDL of the orthodontically moved first molars was found at all experimental periods ($p < 0.05$). CD43+ cells showed significant increase on the experimental side at 1, 7 and 14 days ($p < 0.05$), while Ia antigen expressing cells had significant increase at 3 and 7 days ($p < 0.05$). For CD4+ cells no significant differences were found at any of the experimental periods. These data indicate that experimental tooth movement leads to increased recruitment of immunocompetent cells which mainly belong to the mononuclear phagocytic system. This study was supported by Colgate Norge.

1822 Effects of Advanced Periodontal Disease in Human Dental Pulp TR S AGUIAR* J E A MATTOZ G C TRISTÃO, F E PUSTIGLIONI, S KON (Federal Fluminense University and Sao Paulo University Brasil)

The common trend today is to accept periodontal pathology as a consequence of pulpal problem, while periodontal pathology causing pulp damage is still a controversial issue. The purpose of this research is to study the possibility of pulp damage caused by advanced periodontal disease. We selected 30 unradicular teeth showing the following characteristics: caries free, no abrasion or erosion, bone destruction up to the apex, and no periodontal treatment at that site. The teeth were extracted, divided in two (root and crown) at CEJ level, and placed in 10% buffered formalin for seven days. Decalcification was obtained by an equal solution of 20% sodium citrate and 45% formic acid. Slides were obtained by HE and Gomori-Trichrome and studied under light microscopy (1000X). Histologic results have shown no change at the coronal portion of dental pulp. Changes like pulp atrophy, pulp fibrosis, alteration in odontoblastic layer, diffuse calcification and localized inflammatory infiltrate, could be observed at the root portion. From the results obtained we concluded that: 1) The coronal portion showed a 100% normal tissues and cells. 2) The radicular portion showed several modifications: 2.1) isolated fibrosis in the entire pulp chamber; 2.2) 76% of fibrosis and diffuse calcification present at the medial and apical third; and 2.3) apical fibrosis associated with inflammatory infiltrate in only two cases.

1823 Therapeutic effects of chlorhexidine digluconate (CHX) in laborers with untreated gingivitis EF Corbet* O W Tam K Y Zee C M Lo A Mombelli M Wong N P Lang Universities of Hong Kong and Bern Switzerland

The prophylactic effects of CHX are well documented but information on the effects of CHX in untreated subjects is lacking. The purpose of the study was to evaluate the therapeutic effects of a 0.12% CHX solution on (Peridex®) in subjects with marked gingivitis and abundant calculus. 60 subjects were recruited from a knitting factory in the Province of Guangzhou, People's Republic of China. At baseline and after 3 months the oral hygiene status was assessed by the Plaque Index (PI) and the Calculus Surface Severity Index (CSSI) Systems. The evaluation of gingival health or disease included an assessment of the conditions on the basis of the criteria of the Gingival Index (GI) and the tendency to bleed on probing (BOP). The study was designed as a double blind, placebo controlled clinical trial. The subjects were group matched according to the initial GI and they then rinsed twice daily under supervision with either CHX or placebo. After 3 months the test group (CHX, n=13) showed significantly decreased mean PI, GI and BOP while the discoloration index (DI) had increased. The control group (Placebo, n=26) also showed decreased mean PI, but increased mean GI. The test and control group differed significantly at 3 months in mean GI, BOP percentages and mean DI. An increase in the proportion of GI=0 scores from 11% to 29% and a decrease in the proportion of GI=2 scores from 50% to 36% in the test group documented that CHX has a therapeutic effect on untreated, marked gingivitis with abundant calculus. Supported by the Clinical Research Foundation (CRF) University of Bern, Switzerland.

1824 Antiplaque Efficacy of Dentifrices in a 4-Day Plaque Regrowth Model N CONFORTI* C HOVLIAAS DELOZIER* S MANKODI B KOHUT (DPT Laka, Worth FL and Warner Lambert Co, Morris Plains NJ)

One hundred and twenty three (123) healthy adult subjects completed this clinical study to determine the relative antiplaque efficacies of 2 Cool Mint Listene dentifrices [CMLD] (gel [CMLG] and paste [CMLP]) during a 4-day period with no other oral hygiene procedures. Subjects were randomly assigned to either 1 of 2 CMLD groups, a positive control antiseptic mouthrinse (AM), or a 5% hydroalcohol negative control mouthrinse (CM). The CMLD were tested in the form of slurries with which subjects rinsed for 60 seconds. Slurry rinses were prepared by mixing 1 part dentifrice with 3 parts tap water. Following baseline oral soft tissue and plaque examinations, subjects received a thorough dental prophylaxis to remove all supragingival plaque and then rinsed twice daily under supervision with their randomly assigned treatment for 4 days. No other form of oral hygiene was permitted during this 4-day period. On day 5, subjects received an oral soft tissue examination and plaque levels were scored using the Turesky modification of the Quigley Hein Plaque Index. Used as the only oral hygiene measure, CMLG and CMLP were statistically significantly ($p < 0.01$) different from the CM with plaque reductions of 21.05% and 21.80% respectively. AM was statistically significantly ($p < 0.01$) different from the CM with a plaque reduction of 25.19%. There was no statistically significant difference between CMLG and CMLP. This clinical screening study demonstrated that the 2 CMLD, used as a slurry, were significantly effective in reducing plaque over a 4-day period of no oral hygiene. (Supported in part by a grant from the Warner Lambert Company).