Dalhousie University, Faculty of Dentistry

MARCH 1992

Dental Research News

Research Development Office, (902) 494-1675

VOLUME VI, NUMBER 3.

Embarrassments

The Research News would like to apologise to Dr. Robin Howell for a typographical error in the table on page 6 of the February edition. Unfortunately the table listed his Oral Biology grant funds from federal agencies for 1986-89 as biomaterials research. Further investigation has found an additional $21,991, which was also obtained by Dr. Howell from MRC. This error and oversight has been traced to the overworked night editor and he has been severely repremanded.

The 20 abstracts submitted to the IADR meeting are itemized on pages 3 to 12. The topics clearly indicate the broad base of our research which has developed rapidly over recent years. If all abstracts are accepted we shall have 37 papers presented at the 1992 AADR/IADR meetings. The 20 abstracts might be regarded as an embarrassment. The prediction in the January Dental Research News that our 200th abstract would be presented in Chicago looks to be in grave danger of being incorrect. The old saying that making predictions is a chancy business especially when it involves the future is certainly true. The Dental Research News would be delighted to be proved wrong and would love to celebrate the 200th paper from New Scotland in Old Scotland next July.

Another Industrial Grant for Dentistry

Within 24 hours of learning of the success of the MRC University/Industry Grant, Elliott Sutow had the excellent news that an Industry supported grant had been successful. The grant of about $25,000 will be directed towards evaluating the potential corrosion degradation and cytotoxicity of an endodontic system. Research in our own and other laboratories has shown that many of the materials which are utilized in the body have not been adequately tested for biological effects. It is most encouraging that at least one company realize the importance of adequate evaluation of biomaterials.

From Strength to Strength

Our research grant success goes from strength to strength. An industrial grant worth about $25,000 was the second successful grant to be obtained in 24 hours for the Dental Faculty. The project aims to evaluate the level of corrosion and cytotoxicity for a dental endodontic biomaterial system. The success of this grant is very encouraging. To bring in industrial funding during the current tight economic climate is a wonderful achievement. Elliott Sutow is to be congratulated in negotiating this grant at this difficult time. Each grant success in any subject area enriches and enhances the reputation of our Faculty and thus increases the potential for further success. Elliott Sutow is the principal investigator on this endodontic biomaterials project together with colleagues, Ken Zakariasen, Choong Foong and Derek Jones. Some of the basic and complex corrosion research pioneered by Elliott Sutow over the past 15 years does have a place in the real practical world after all. The following thoughts from De Witt Stetten, Jr. are applicable to the application of some of Elliott's corrosion work.

Planting Seedlings

...."the popular image of the unfettered scientist who is described in such terms as 'head-in-the-clouds,' "ivory-towered,' and 'impractical dreamer.' It is my experience that none of these epithets are any more applicable to the basic scientist than they might be to the plant economist employed by a major lumbering company who spends his time collecting and planting seedlings of trees in a recently lumbered tract. He cannot know at the time that he plants the seedlings what will be constructed out of the ultimate tree that will grow there. However, he has every reason to be assured that something useful will result." De Witt Stetten, Jr.

Correction

Below is an updated and corrected version of the table previously published on page 6 in the February edition of the Dental Research News. It should be noted that this table may still contain errors which may arise due the nature of reporting the federal research funding over different time periods.

Research Funding 1959-92

Date	Agency	Amount	Discipline
1959	NRC	$2,350	Oral Surg
1960	NRC	Not Known	Perio
1969/71	MRC	$14,000	Oral Biol
1972/74	MRC	$15,000	Oral Biol
1976/78	MRC	$60,000	Biomater
1979/80	NHRDP	$28,218	Oral Biol
1979/81	MRC	$55,042	Biomater
1982/84	MRC	$108,433	Biomater
1985/88	MRC	$170,802	Biomater
1985/90	MRC	$147,093	Oral Biol
1988/91	MRC	$712,591	Prog. Grt
1989	MRC	$18,333	Prog. Grt
1989/90	NHRDP	$141,872	Ped.Com
1991/92	MRC	$169,700	Prog. Grt
1991/92	NHRDP	$120,080	Ped.Com
1991/95	MRC	$743,156	Oral Biol
1992/95	MRC/Ind	$544,530	Biomater

TOTAL *$3,051,110

Patterns of Discomfort following Orthodontic Appliance Adjustment.
W. LOBB*, K.L. ZAKARIASEN, P. MCGRATH, K.A. ZAKARIASEN, M. MCMINN (Dalhousie Univ., Faculty of Dentistry.
Few studies have investigated discomfort following orthodontic adjustments, nor identified patient efforts to cope. The purpose of this investigation was to determine patterns of discomfort. 29 patients recorded discomfort levels for 4 days after adjustments. These patients (group A) were part of a larger randomized study involving 72 patients from two orthodontic practises. 25 of the 29 (86%) reported discomfort. 10 (34%) reported use of coping methods (e.g. Tylenol, soft diet). A significant decrease in the median discomfort values was noted between Day 1 and Day 4 (peak values during first 2 days- ANOVA,$p=.05$). There were no significant differences in levels relative to type or number of adjustments. The mean of the median scores (5 point scale, 0 to 4) was 1.46 (Day 1), 1.18 (Day 2). Two experimental coping strategy groups ($B:nB=23, C:nC=20$) were also studied. No significant differences from the control group (A) were found. In Group B, 19 of 23 (83%) reported discomfort, and in Group C, 20 of 20 (100%). The means of the median scores were: B- 1.00 (Day 1), 1.11 (Day 2) and C- 1.64 (Day 1), 1.21 (Day 2). Overall, 64 of 72 (89%) experienced discomfort. From these results, it would appear that most patients undergoing orthodontic treatment experience some post-adjustment discomfort, especially during the first two days following adjustments. Since the control group exhibited a low use of coping strategies, and yet the incidence of discomfort is reported to be frequent, further study is necessary to determine why coping methods are not more frequently used. Also, while Group B (gum chewing) did not show sig. lower discomfort scores, the trends in this direction, and patient perception of derived benefit, indicates further study. Support by CFAO & CFDE.

Relation Between Fixture Length And Implant Failure. P.K. MOY* (UCLA Dental Implant Center, Los Angeles, CA.) and C.A. BAIN (Dalhousie University.
To date only certain locations (various authors), poor bone quality (Jaffin & Berman 1991), and 7mm unloaded fixtures (Friberg et al 1991) have been positively identified as predisposing to implant failure. The purpose of this study is to evaluate the relationship of fixture length to implant failure in 2179 consecutively placed Branemark implants.
2179 Branemark implants placed by one operator (PKM) in 540 patients over a 6 year period were evaluated for failure. Most implants were loaded at the time of evaluation. Implant removal or radiographic bone loss >50% were used as criteria for failure. Records of fixture length were available for all implants. ANOVA of 120 failed implants revealed a statistically significant trend ($p<0.001$) in increasing implant failure with decreasing implant length. Failure rates varied from 0.76% in 264 18mm fixture to 13.07% in 199 7mm fixtures. When different areas of the mouth were analysed separately shorter implants always showed significantly higher failure rates.
It is concluded that implant failure decreases as fixture length increases and that, where only short fixtures can be placed, patients should be advised of the poorer prognosis and of surgical options which allow placement of longer fixtures.

Relation Between Smoking And Failure Rates of Dental Implants. C.A. BAIN* (Dalhousie University, N.S. Canada) and P.K. MOY (UCLA Dental Implant Center, Los Angeles, CA.)

Previous studies have identified poor bone quality (Jaffin & Berman 1991) and short implant length (Friberg et al 1991) as factors predisposing to implant failure. Smoking has been shown to compromise wound healing in plastic, cardio-vascular and obstetric surgery. The purpose of this study is to investigate the relationship between smoking and failure rates in dental implants. 2194 consecutive Branemark implants placed by one of the authors in 540 patients between 1984 and 1990 were evaluated (mean 3.2 years). Criteria for failure were, i. implant removal; ii. over 50% radiographic bone loss. Overall failure rate of 5.92% was consistent with other studies, however when patients were divided into smokers and non-smokers it was found that failures were significantly higher in smokers (11.28%) than in non-smokers (4.76%), ANOVA $p<0.00005$. When evaluated by sextant it was found that smokers had significantly higher failure rates in all regions except the posterior mandible (Post. Maxilla $p<0.005$; Ant. Maxilla $p<0.00001$; Ant. Mandible $p<0.001$).

It is concluded that smoking increases the risk of failure of dental implants and that a smoking cessation protocol should be implemented around the time of surgery.

Attitudes and Practices of Dentists in a Legalized Denturist Environment. D.V. CHAYTOR*, and D. POEL (Dalhousie University, Nova Scotia, CANADA)

The purpose of this study was to determine the perceptions, attitudes and practices of dentists in a province approaching the 20th year of legalization of denturists for complete denture service directly to the public. The information is important to dental leaders, health care planners and educators. A ten-page survey was mailed in the fall of 1991 on behalf of the Provincial Dental Association to active members for anonymous response. 224 (59%) dentists responded. Results were analyzed using the Statistical Package for the Social Sciences (SPSS). 91% of respondents were general practitioners, 82% had graduated between 1971 and 1990. All but 1% reported doing removable prosthodontics. 64% believed the presence of denturists reduced the complete denture component of their practices; 55% reported a reduction in removable partial dentures. Most importantly, results revealed the current relationships between dentists and denturists and the relevant attitudes of dentists. 47% had received denturist referrals. 48% of dentists reported directing patients to a denturist. The majority (55%) of the respondents stated that referrals between dentists and denturists should be accepted as ethical. 27% of respondents believed denturists need more training but 66% had no opinion. 47% stated that denturists should have access to continuing education designed for dentists and dental auxiliaries and 68% said dentists should participate in the training of denturists. Dental Association files, newspaper reports and public records indicate that dentists' opposition to the legalization of denturists had been strong. The results of this survey show a shift in attitudes and practices in favour of professional cooperation in education and clinical practice.

Determination of Serum & Gingival Drug Levels Following Localized Minocycline Application L.M. DELOREY*, C.A. BAIN, P. YEUNG & S. MOSHER (Dalhousie University, Halifax, N. S. CANADA)

The purpose of this study was to provide data on serum and gingival crevicular fluid (GCF) levels of minocycline following a single dose subgingival application of 2% minocycline ointment to patients with moderate to severe adult periodontal disease. One test site per quadrant (5-7mm), in each of 10 healthy patients (mean age 46) was treated with minocycline ointment. Venous blood samples were collected at pre dose time and 1, 2, 4, 6, 24, and 48 hours after the administration of the drug. Plasma was extracted in a refrigerated centrifuge within 1 hour of collection and frozen at -20°C until assayed. GCF was collected using filter papers placed at the gingival margin and the volume by weight was determined at pre-dose time, 15, & 20 minutes, and at 1, 2, 4, 6, 24, 48, 72, 96, and 120 hours. Minocycline hydrochloride was assayed in the test samples using high performance liquid chromatography (HPLC). The results of this study showed that from 1 to 24 hours 90% of the patients had a detectable plasma concentration of the drug, (mean 0.024 ±0.006 ug./ml.), while no drug was found in plasma for any patient at the 48 hour period. One patient had no detectable drug levels at any time period (Asian). All patients showed a GCF drug readings at 6 hours (mean 55.3±58.6 ug./ml.) and at 24 hours, 70% of the patients showed a mean drug level of 36.6±61.3 ug./ml. CGF drug levels were found in some test sites in all patients from 48 to 120 hours, however, some of these readings were below the sensitivity range of the HPLC. It may be concluded that this drug delivery system provides a localized method of chemotherapeutic treatment with little systemic involvement. Further research is ongoing to evaluate the bacterial flora of the pocket pre and post localized delivery of 2% miniocycline. (Lederle Intl. Grant # R2-52-6585).

Subcutaneous Implantation of Experimental Denture Soft Polymers. B.B. HARSANYI, W.C. FOONG, R.E.HOWELL, P. HIDI & D.W. JONES. Dalhousie .

We have previously shown the effects of a commercial soft polymer, C, with a leaching plasticizer (Harsanyi *et al.* 91;Jones *et al.* 88). This study compares the effects of two new, experimental polymers, A&B, with a non-leaching plasticizer, to those of C, a negative and a positive control material(N&P) and the surgical injury (S).Material A was a heat-cured polymer of ethyl and lauryl methacrylate(m); B a room temperature gelling copolymer of ethyl and lauryl m. Both had dibutyl sebacate as plasticizer (pla). C contained poly(ethyl)- and (methyl)m as powder and dibutyl phthalate as pla. For N, the powder of C was blended with ethanol without pla; P contained powder C with dibutyltin diacetate. Four subcutaneous pockets were prepared surgically in the back of each of 12 guinea pigs. One was left empty; the others were implanted with A, B, C, or N. Animals were killed 7 days post-implantation.Two examiners scored each feature from O(absent) to 4(severe) in a blind fashion. Scores were evaluated by ridit, Kruskal-Wallis and Mann-Whitney-U tests. Only P showed gross pathology. Microscopic rankings were: edema S=B<A=C<N<P; necrosis S=B, S<A, B=A=C=N<P; peri-implant/incision infiltrate S=B, S<A=C=N<P; distant subcutaneous infiltrate S=B, S<A=C, B=A, B<C=N<P; distant subepithelial infiltrate S=B=C=A<P; muscle injury S=B=N=A=C<P. Test material B showed no greater reaction than the surgical insult. A was of equal or lesser irritancy than the commercial and negative control polymers, which in turn were less irritant than the positive control. MRC (Canada)PG-45.

Enamel Optical Property Differentials: Reflected Laser Light and Laser-Induced Fluorescence. ZAKARIASEN, K.L.*, BARRON, J., PATON, B., (Dalhousie University, Halifax, Nova Scotia, Canada).

Research indicates that CO_2 laser irradiation can raise enamel resistance to caries, and that the optical properties of normal and carious enamel are different, which may be useful for caries diagnosis and research. The objective of this initial study was to determine whether differentials in reflected laser light and laser-induced fluorescence could be observed between lased and non-lased enamel, and between non-carious and carious enamel. A fluorometer, using HeNe (reflection) and HeCd lasers (fluorescence), has been designed. A scan of 0.09 mm^2 was used, consisting of six parallel scan lines, 300 mm in length spaced 50 mm apart. Measurements are made at 5 mm intervals (60 per line). Each of the 60 measurements was averaged with the corresponding measurement on the 5 remaining lines resulting in 60 mean values. Specimens were Bu surfaces of third molars. Four specimens contained adjacent normal and carious enamel areas (carious area created in art. caries sys.). Two specimens contained a normal enamel area adjacent to an area CO_2 lased. Each area was scanned. Reflection/fluorescence intensities were determined and graphed as running intensity sums. For the two lased/non-lased specimens, the HeNe reflected light was greater for the non-lased area, but the fluorescence intensity sums were greater for the lased sites. For the four non-carious/carious specimens, the non-carious areas reflected more HeNe light, while the carious areas showed higher fluorescence intensity sums. <u>These preliminary results indicate that enamel optical property differentials appear to exist with respect to HeNe laser reflected light and HeCd laser-induced fluorescence.</u>

Comparison of Combined Hand/Sonic Techniques with Traditional Step-back Filing. ZAKARIASEN, K.L., ZAKARIASEN, K.A.*, MCMINN, M., (Dalhousie)

Research has shown that combined hand/sonic enlargement can result in properly shaped canals. This research is designed to compare curved canals shaped by step-back filing (A, B) with those shaped by hand/sonic techniques (C, D, E) using three different hand files and one sonic system (M-M Sonic Air 1500). The five groups studied were: A - S.S. Hed., B - Titan. Hed., C - S.S. Hed./Sonics, D - Titan. Hed./Sonics, E - Heliap./Sonics. Forty-five #101 Endo-vu blocks were accessed, randomized into 5 groups, and preflared to 20 mm with hand files, or with hand files/sonics. Apical preparations were completed to #25 at 23 mm. Canal flaring was continued with either step-back filing or sonics until a D11T spreader would freely penetrate to 22 mm. During sonic instrumentation, #1 Rispisonic files were used around curvatures. One operator prepared all canals. Instrumentation times and preferences were recorded. Canal preparations were randomly evaluated blind for apical transportation, ledge formation, and hour-glass shaping of the canal. No significant differences were found between the 5 groups for these factors, all groups being essentially error-free with regard to these criteria. The mean preparation times (min.) were: Step-back = 13.5, Hand/Sonic = 11.0, significant at $p < .05$. The hand/sonic technique was preferred in all 9 comparisons made between groups (Heliapical files/sonics preferred in 5/9). <u>Under the conditions of this study, the results indicate that all 5 techniques can provide satisfactory canal preparations with minimal ledging, apical transportation, or "hour-glass" shaping, that the hand/sonic technique is faster and that the hand/sonic techniques may have characteristics which encourage operator preference.</u>

2-Dimensional Laser Scanned Fluorescence and Reflected Laser Light Intensity Repeatability. BARRON J.R.*, ZAKARIASEN K.L., PATON B.E. Optical and laser technology offer promising techniques for detection and quantification of dental caries. Studies show that the fluorescence and reflected intensity of carious enamel is significantly different from non-carious. Fluorescence and reflected laser light intensity from enamel have been studied using an experimental scanning system. Utilizing the optical properties of enamel, while providing an X-Y scanning mechanism, allows the collection of 2-dimensional fluorescence and reflected intensity data from enamel. For fluorescence studies, a 25 mW He-Cd laser beam is utilized and a 1 mW He-Ne laser is used as a pointer for sample alignment and as the optical source for reflected intensity studies. Fluorescence or reflected intensity is measured on a photomultiplier tube. This study was designed to determine the repeatability of 2-dimensional fluorescence and reflected intensity scans. 11 non-carious third molars were selected for this study. A 0.09 mm^2 area was scanned 5 times in succession on each specimen using both fluorescence and reflected. The scan area consisted of 6 parallel scan lines, each 300 μm in length, spaced 50 μm apart. Measurements were made on each scan line at 5 μm intervals. Values from a scan were averaged resulting in one value for each scan. It was found that the repeatability of both fluorescence and intensity were acceptable with values of 0.87 ± 0.003 (S.D.) to 1.54 ± 0.082 and 0.24 ± 0.017 to 2.48 ± 0.657 respectively. A Bartlett test for equality of variances shows that fluorescence is significantly ($p<0.001$) better than reflected intensity repeatability . It would appear from these results that, although both fluorescence and reflected intensity repeatability are acceptable, fluorescence provides significantly better repeatability.

16S-rRNA gene restriction/enzyme patterns; clonal variation among periodontal pathogens. AJ. ROCKWELL, SE. GHARBIA and HN. SHAH (Depart. of Oral Biology, Dalhousie University.
The application of chemotaxonomic tests such as menaquinone, polar lipid, cellular fatty acids, peptidoglycan, DNA base composition and hybridization and enzymic analyses have had a considerable impact in improving the circumscription of many periodontopathic bacteria. However, it is now clear that within taxa intraspecies heterogeneity exists which, if resolved, may help define the role of several species in periodontal diseases. In the present study, we continue to apply a variety of enyzme electrophoretic analysis and use a plasmid with the 16S-ribosomal RNA gene insert to probe the genomic DNA of such periodontal pathogens as *P. gingivalis, P. intermedia, L. buccalis, T.denticola* and *Fusobacterium* species. Within all taxa heterogeneity was observed both in enzymic and 16S-ribosomal RNA gene restriction patterns. Thus, 3 centers of variation were found within *P. gingivalis, P. intermedia* and *L. buccalis* and 2 in each of *T. denticola* and *F. nucleatum* subspecies *nucleatum.* Interestingly the present tests clearly separated human from animal isolates of *F. necrophorum.* This study has so far confirmed the suspected intraspecies diversity of several taxa and should now facilitate a clearer understanding of the ecological and clinical signifance of this clonal variation. (This work was supported by MRC Grant Number DG-411)

Reactions surrounding the metabolism of glutamate by *Fusobacter-ium nucleatum*. S.E. GHARBIA, H.N. SHAH & R. WHITE (Depts. of Oral Biology & Chemistry, Dalhousie University.

Glutamate represents more than 50% of the free amino acid pool of human dental plaque. Consequently several putative periodontal pathogens can metabolise this monomer. We have recently reported that *Fusobacterium* species can ferment glutamate via the 2-oxoglutarate, 4-aminobutyrate or mesaconate pathways to produce acetate and butyrate. In the present study, we utilize ^{13}C-NMR and reverse-phase HPLC to study further reactions surrounding glutamate catabolism. Cell-suspension experiments revealed that the fermentation of basic (cationic) amino acids such as arginine, histidine and ornithine led to intermediates of the 2-oxoglutarate pathway, such that their subsequent catabolism resulted in the production of acetate and butyrate. ^{13}C-NMR studies confirmed earlier experiments involving enzyme assays and are consistent with operation of the 2-oxoglutarate pathway. The possible routes by which one of the major intermediates of this pathway, 2-hydroxyglutarate, is converted to crotonyl-CoA and subsequently acetate and butyrate may involve glutaconate or vinyl acetate. Data have so far indicated that the uptake of basic amino acids is profoundly affected by the concentration of glutamate available in the environment. These studies should help to clarify the complex control mechanisms involved in regulating the utilization of amino acids by specific periodontal pathogens.

(This study was supported by MRC Grant Number DG-411).

Construction of a DNA probe for *Prevotella intermedia*. J. CLOW*, H.N. SHAH, A.J. ROCKWELL and S.E. GHARBIA (Dept of Oral Biology, Dalhousie.

Prevotella intermedia has been implicated as an etiological agent of various forms of periodontal disease. The aim of this study was therefore to construct and test the specificity and sensitivity of a nucleic acid probe for this species for subsequent clinical studies. Because of its stability, high cell copy number and species-specific variable "V" regions of the ribosomal RNA, we utilized the primary 16S-rRNA sequence of *P. intermedia* to construct a 52 *mer* oligonucleotide derived from the V7 region. In addition, primers based on the conserved flanking sequences of the target region were synthesized for direct amplification using a Programmable MJR Thermal Cycler. The results of this study revealed that DNA from *P. intermedia* hybridized to its homologous probe without cross-hybridization to other periodontal pathogens such as *P. gingivalis, P. oralis, P. melaninogenica, F. nucleatum, F. periodonticum, L. buccalis* or oral spirochetes. The sensitivity of the probe was high (<1 pg) and detection was rapid (<2 days). Plaque taken from sites showing clinical signs of periodontal disease were alkaline hydrolyzed, mixed with the primers and amplified by a PCR technique. The method used allowed detection of *P. intermedia* directly from whole plaque. These results obviate the need for lengthy, laborious and complex bacteriological tests which can take several weeks. Studies of this nature bridges the gap between laboratory based molecular genetic techniques and clinical diagnostic practice, helps elucidate the ecological role of specific pathogens in response to disease treatment and, in the long term assist in identifying patients at high risk of periodontitis. (This study was supported by MRC Grant Number DG-411).

^{14}C-Methyl-lysine release from protein hydrolysates by plaque bacteria. HN. SHAH, SE GHARBIA, A THOMSOM & S SREEDHARAN (Oral Biology, Dalhousie Univ; Adelaide Univ AUSTRALIA & QMW London Univ).

The ability of a putative pathogen to hydrolyse native proteins directly influences its pathogenic potential. A variety of methods have therefore been used to measure bacterial proteolysis. In the present study, albumin and casein, labelled by reductive methylation using [^{14}C] formaldehyde and sodium borohydride were used to measure the capacity of several putative periodontal pathogens to digest these substrates. Several non-oral species such as *B. fragilis*, *F. necrophorum* and *P. corporis* were included for comparison. All species hydrolyzed these substrates but to different extents. *T. denticola*, *P. gingivalis* and *P. heparinolytica* hydrolysed both substrates linearly over the 3 hours tested whereas the remaining species required an initial period of incubation before maximum rates of activities were achieved. Specific activities were 206, 175 and 102 for albumin and87, 127 and 134 for casein respecively. When the hydrolytic activities of all species were compared after 4 hours (the time taken to reach maximum hydrolysis), oral species (except *T. denticola*) such as *P. gingivalis*, *F. nucleatum* and *P. intermedia* hydrolysed casein more rapidly than albumin. With the exception of *B. fragilis*, non-oral species such as *P. corporis*, *B. levii* or *B. macacae* digested both albumin and casein at approximately similar rates. These results suggest that [^{14}C]-casein is an excellent test substrate for determining proteolysis by oral bacteria. Furthermore, the method used is appropriate for labeling host-derived proteins for studies on periodontal tissue degradation. (This work was supported by MRC Grant # DG-411).

The use impedance measurements to study amino acid and peptide utilization by periodontal pathogens. S.E. GHARBIA and H.N. SHAH (Oral Biology, Dalhousie University.

We have shown previously that energy assimilation by subgingival plaque bacteria occurs mainly through the catabolism of nitrogenous substrates such as amino acids and peptides. The present study builds upon this data by taking advantage of recent developments in the measurement of impedance in biological systems. Here the growth response of *P. gingivalis*, *F. nucleatum* and *T. denticola* to peptides (supplied as trypticase) and amino acids (as casamino acids) was measured over 24 h by monitoring changes in AC conductivity at 37°C. All speices utilized peptides preferentially to amino acids consistent with results obtained previously by conventional growth response experiments. The linear growth response to trypticase for *P. gingivalis* was 12.26 h whereas both *F. nucleatum* and *T. denticola* was greater than 24 h. By contrast, there was more uniformity in the utilization of amino acids of casamino acids, previously shown to be a poor growth substrate. In all cases these results, which would take from several days to weeks to obtain, were completed within 24 h. Furthermore, subtle differences in growth patterns such as the ability of *F. nucleatum* to metabolise its storage glycopolymers before utilizing amino acids were clearly evident. The present method provides an excellent means of studying bacterial growth kinetics and delineating bacterial/substrate specificities of both synthetic and natural substrates.

(This work was supported by MRC Grant Number DG-411).

Studies on the nutrional function of gingivain produced by *Porphyromonas gingivalis*. HN. SHAH, SE. GHARBIA, S. SREEDHARAN and K.BROCKLEHURST (Dalhousie Univ, & QMW, Univ. London, UK).

We have recently purified the extracellular proteinase of *P. gingivalis* (W83) and shown by both its method of isolation together with titration of the catalytic site cysteine and pH dependent stopped-flow kinetics that the enzyme is not trypsin-like as widely reported, but a cysteine proteinase for which the name gingivain has been proposed. Here we report our preliminary findings on the nutritional function of this enzyme. Gingivain was purified using covalent chromatography by thiol-disulphide interchange. Purified gingivain was then used to digest a variety of protein hydrolysates (eg. trypticase and proteose peptone) which we have shown previously to support bacterial growth. The resulting peptides were separated and the growth response of *P. gingivalis* and other oral species to each fraction was compared with the corresponding native substrate. In all cases, gingivain-digested peptides had a marked effect on their cell-yield (*ca.* 30-50% increase). In addition, gingivain incubated with erythrocytes under reduced conditions caused lysis and rapid release of protohaem (<2 hours), which was subsequently used for cytochrome biosynthesis. Thus apart from its biological role in the disruption of host-defence mechanisms, gingivain, by virtue of its location on the outer membrane of cells/vesicles is able to hydrolyse proteins to release peptides which support the growth of *P. gingivalis* and other species such as *F.nucleatum.*

(This work was supported by MRC Grants DG-411 and 99004592 II SD)

Desiccation/Water Sorption Effects on Intercuspal Relationship in Complete Dentures. O. SYKORA*, E. SUTOW and S. RICHARDSON, (Dalhousie University, Faculty of Dentistry, Halifax, CANADA).

Small dimensional changes in a completed prosthesis affect the occlusion and thus the stability of the denture. The purpose of this study was to determine the influence of desiccation/water sorption on the stability of completed prostheses. Ten complete dentures were processed on identically mounted casts made from master moulds. Five were processed by the trial-pack technique and five by the continuous injection technique. Cusped acrylic teeth, set to bilateral balance, had pins inserted to serve as markers for measuring the dimensional change in the distance between lingual and buccal cusps of mandibular and maxillary 2nd molars and in cuspids. Measurements were made with a Vernier caliper (0.025 mm). The prostheses were trimmed and polished and then immersed in water (23°C) for a period of 8 weeks. The prostheses were removed from the water and measurements were made after an 8 h, 24 h, 3 wk and 8 wk desiccation. The desiccated dentures were again immersed in a water bath (23°C) and measurements were made after 1 h, 24 h, 1 wk and 3 wk. A Student-Newman-Keuls multiple comparison test (p=0.05) was used to analyze the data. Results showed that for both materials there were significant differences among the measurement periods for water sorption, but not for desiccation. It was concluded that in terms of intercuspal relationships, the full effect of desiccation occurs within 8 hours, while the effect of water sorption continues for at least 1 to 3 weeks.

Elastic Moduli of Wet and Dry Experimental Composite Materials.
D. W. JONES*, A. S. RIZKALLA, E. J. SUTOW, & G. C. HALL.
(Dalhousie University, Halifax, Nova Scotia, Canada).

Objective: To determine effects of filler volume and water storage on elastic moduli for experimental composites. Dynamic (sonic) moduli (Young's shear and bulk) of elasticity and Poisson's ratio (n=3) were obtained for composite materials containing various silanized filler fractions (0, 6.4, 20.9, 38.9 and 48.1% Vol). Specimens were aged for 18 months (prolonged dry storage) prior to determining the moduli on dry specimens in air. Specimens were sequentially tested following 8, 15, 22 and 29 days storage in water @ 37°C. Fifteen specimens were evaluated on five separate occasions for a total of 75 determinations. A Student-Newman-Keuls rank order test was able to separate Young's modulus of 'dry' specimens with 48.1 % Vol filler (14.3±0.3 GPa) from those stored in water (8, 15 and 29 days; i.e., 13.6±0.3, 13.5±0.2 and 13.8±0.2 GPa, p= 0.05). The shear modulus dry condition for 20.9 and 48.1% Vol fractions (3.4±0.1 and 5.6±0.1 GPa) separated from all water stored specimens (3.0±0.9 to 3.2±0.1 and 5.3±0.1 to 5.4±0.1 GPa, p= 0.05). The values for bulk modulus did not show any significant difference between dry and wet storage (p= 0.05). Change in weight due to water uptake was not different for specimens stored for 8 to 29 days (p= 0.05). Conclusion: Significant correlations were obtained between volume filler fractions and elastic moduli (p<0.001). Water sorption did not have a major influence on elastic moduli for experimental composite materials up to 29 days storage in water. This research was supported by MRC (Canada) Grant PG45.

Prosthodontic Soft Polymers: Leachability Profiles of Sebacic Ester Plasticizers. G. C. HALL*, D. W. JONES and M. LANGMAN. (Faculty of Dentistry, Dalhousie University, Halifax, Nova Scotia, Canada).

Objective: Determination of leaching rate of four sebacic acid esters when used as plasticizers in experimental methacrylate soft polymers. Previous work (Jones *et al.*, IADR 1991, AADR 1992) has shown dibutyl sebacate to be a most efficient plasticizer. In the present work dimethyl (DMS); diethyl (DES); dibutyl (DBS) and diethyl-hexyl sebacate (DEHS) were used to plasticize a commercially available copolymer powder of 79.4% poly(ethyl methacrylate) and 20.6% poly(methyl methacrylate). Disc specimens (1 and 2 mm thickness) were produced in PTFE moulds from the polymer powder and a mixture of 50% plasticizer, 50% EtOH. A powder to liquid ratio of 1.5 was used. Specimens were pressed to the desired thickness against a PTFE coated slab. Pie shaped samples were cut, weighed and placed into 100 mL of distilled water. At nine time intervals up to 76 days the water was changed, extracted and prepared for gas-chromatographic analysis. Data was calculated as cumulative mg/g of polymer-gel. A Student-Newman-Keuls rank order test separated all the 1mm thick discs (P=0.05) with a range from 0.3623 ±.0692 to 46.8445± 2.3464 mg/g. The order was DBS<DEHS<DES<DMS respectively. SNK analyses (P=0.05) of both 1 and 2 mm thick specimens indicated that DBS leached at a significantly lower rate and DMS produced significantly greater leaching at both thicknesses. Conclusion: Dibutyl sebacate provides longer effective plasticization of ethyl methacrylate copolymers compared to other sebacates. MRC Grant PG 45.

Relationship Between Chemical and Physical Properties for Na_2O-K_2O-SiO_2 Glasses. **A. S. RIZKALLA*, D. W. JONES, E. J. SUTOW, & R. P. Miller. (Dalhousie University, Halifax, N. S. Canada).**

Objective: To determine relationships between the physical and chemical properties of Na_2O-K_2O-SiO_2 glasses synthesized by wet chemical methods. Method: Calculations of nonbridging oxygen (NBO) were undertaken from chemical analysis data previously determined by AAS. Dynamic Young's modulus (E) was determined (n = 3) by an ultrasonic method and density (D) (n = 3) by water displacement. A Tukon Hardness tester gave both Vickers (H_v) and Knoop indentations (n = 3). Indentation (Vickers) fracture toughness K_{Ic} values (n = 3) were obtained using Blendell's equation.
Results: A series of 6 crack-free Knoop indentations were performed at different loads. Indentation length was plotted versus the square root of the load values. 'True hardness' (H_T) was determined from the slope of the line. Linear and nonlinear regression analyses between K_{Ic} and H_T, E, NBO and D were significant for all of the above variables (probability from $P< 0.001$ to $P< 0.01$). K_{Ic} using the H_v ranged from 0.98±0.03 to 2.06±0.01 MPa $m^{1/2}$ and using H_T from 0.84±0.05 to 1.68±0.02 MPa $m^{1/2}$. A Student-Newman-Keuls test separated K_{Ic} values determined from H_v and H_T ($P = 0.05$). Generalized equations were developed allowing the conclusion that K_{Ic}, E, and H_T were a function of chemical and physical properties of the glass compositions studied. Incorporation of H_T into calculations significantly reduced K_{Ic} values by 14 to 18%. Supported by MRC (Canada) Grant PG45.

Corrosion of Dental Amalgam: Influence of Finishing Technique. **E.J. SUTOW,* D.W. JONES, A.S. RIZKALLA and P. JOHNSON (Dalhousie University, Halifax, Nova Scotia, Canada)**

The corrosion resistance of metals can be enhanced by improving surface finish. It was the objective of this study to evaluate the corrosion of amalgam as a function of various finishing techniques. Circular specimens of Tytin (T) and Dispersalloy (D) were prepared by hand-packing and treated to one of the following eight finishing techniques: 1. carved, 2. carved and burnished, 3. carved, burnished and polished (disc, pumice, Amalgloss), 4. carved and polished, 5. immediate polish, 6. pre-carved burnished, 7. pre- and post-carved burnished and 8. carved, burnished and polished (Shofu polishing kit). Specimens were stored at 37°C and 7 d before testing. Corrosion testing was conducted in a Ringer's solution: pH=7, 37°C, stirred and exposed to the atmosphere. Specimens were polarized at -50 mV (S.C.E.) for 18 h. Seven specimens were corrosion tested for each finishing technique for each alloy (N=112). Four untested specimens for each finishing technique for each alloy (N=46) were examined for surface roughness using a profilometer. Correlation analysis of the results for alloy T showed no relationship between the current density at 18 h vs surface roughness (r=0.088, p=0.836). For alloy D, a good correlation was shown (r=0.847, p=0.009). Correlation analysis of surface roughness of T vs D, due to finishing technique, was r=0.927 (p=0.927 (p=0.001). It was concluded that corrosion of amalgam is not a simple function of surface smoothness.

Record Number

An exciting aspect of the 1992 statistics of the 62 abstract papers submitted or presented at IADR/AADR or AADS meetings is the record number of 25 educational course development and educational research papers which are also being presented at the 1992 AADS meeting. The year 1992 is a truly record year for our faculty with a record total of 62 papers submitted or presented at the three meetings. However, we should also not forget that at least 10 other papers are also being given or have been presented at other research meetings seminars and workshops during 1992.

Pages 3 to 12 provide the full text of the 20 abstracts which have been submitted by our Faculty to the IADR international dental meeting next July. These abstracts have been reproduced in full so that our faculty colleagues and other members of the Dalhousie community can see for themselves the wide range of subject matter which our research programmes cover as well as the high quality of the of the science involved. A total of 1723 papers were included in the programme for the AADR meeting in Boston. It looks as though the combined total for the two meetings this year will produce a further all time record number of papers. A number of interesting Lunch and learning sessions are being held at the AADR this year. Amongst the topics are:- "Computer-generated restorations," "Industry-University Alliances in Research," "Stress Analysis by Finite and Experimental Methods," "Periodontal Disease Susceptibility: The human twin model," and not forgetting the important "Lasers in Dentistry" by our own Ken Zakariasen.

One of the Greatest

"...I am convinced that Dalhousie is one of Canada's greatest universities. Dalhousie's eminence as a true university is based on the synergy it has achieved between teaching and research or scholarship."

Donald Betts

SMILE

Research Definitions

"A standard error" means that I have made the same mistake again.

"Further analysis of the data will be undertaken" means, I do not intend to conduct any further laboratory tests.

"A less discriminating statistical test was used" means, I needed to get a statistically significant result.

INNOVATION

"....scientists are gradually becoming more accepting of industry as an ally in pursuit of their research objectives. The success of the University-Industry programme sponsored by MRC is a good example of this interaction."

Barbara Sparrow, Parliamentary Secretary to Health Minister Bouchard.