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Special Edition - Review of Research

Research Review for 1992

For the past three years a *special edition of the November Research News* has been devoted to a review of our ongoing faculty research based upon abstracts submitted to the IADR/AADR meetings. The following review on pages 2-19 is based upon abstracts which have been submitted for the IADR/CADR meeting in 1993. It should be made clear that this is not by any means a complete review of all of the research being conducted in our faculty. This review does not cover the educational research which is currently being undertaken, which may be presented at the AADS meeting. The object of the review of research is to make available to our Faculty and the Dalhousie community an indication of the type and diversity of research which is currently being undertaken. Any faculty members who would like to provide a report on their own specific area of research not covered in this review should forward this on a disk to the Dental Research Development Office.

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Dalhousie Faculty of Dentistry have submitted a record number of Abstracts to the IADR meeting in Chicago.

35 Abstracts a record for a single meeting

71st General Meeting and Exhibition of the IADR
22nd Annual Meeting and Exhibition of the AADR
The 17th Annual Meeting of the CADR

$ave Money

Did you know that by becoming a member of IADR/CADR you can save about $160 in registration fee for the Chicago meeting.

IADR/CADR Abstracts for 1993

The 35 abstracts submitted to the IADR meeting are itemized on pages 2 to 19. 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 35 papers presented at the 1993 IADR/CADR meeting. The 35 abstracts might be regarded as an embarrassment. The January 1992 Dental Research News predicted that our 200th abstract would be presented at the IADR meeting in Chicago. It is now clear that we will in all probability be presenting our 225th abstract. Fittingly this will occur on the 25th anniversary of our first IADR abstract presentation. The old saying that making predictions is a chancy business especially when it involves the future is certainly true. The years 1992/93 are truly record years for our faculty. The following pages provide the full text of the 35 abstracts which have been submitted to the IADR international dental meeting next March. The abstracts are sequenced in alphabetical order based on the presenter. 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. What is encouraging is the fact that 12 of these projects are in collaboration with other institutions or Departments (not counting Marquette).

Detecting CO_2 Laser Enamel Effects by 3D-Imaged Scanned Laser Fluorescence. J. R. BARRON*, K. L. ZAKARIASEN, B. E. PATON.
Faculty of Dentistry and Dept. Physics, Dalhousie University.

Dental Research has shown that human enamel can be altered by CO_2 laser energy to make it less susceptible to subsurface demineralization. The levels of laser energy required to produce this effect do not cause visible change on the enamel surface. Also, it has been shown that laser-induced fluorescence of normal enamel differs for lased and carious enamel. The objective of this study was to determine, utilizing 3D laser-induced fluorescence images, if differences could be observed between non-lased enamel and lased enamel at different laser energy levels. 5 non-carious third molars were selected (Bu surfaces). Two windows on each specimen were scanned utilizing a scanning laser fluorescence spectrophotometer. A scan area of 1.0 mm^2 was used consisting of 20 parallel scan lines, 1.0 mm in length spaced 50 μm apart. Measurements were taken at 50 μm intervals (20/line), resulting in 400 points/scan. 9 windows were lased (3 each at 0.5, 1.5 and 2.5 watts, 0.15 sec). Each specimen was re-scanned. 3D images were generated for each scan. From the 3D images, the laser irradiated area could easily be identified and isolated. The lased scans were statistically compared (t-test) with the corresponding pre-lased scan baseline areas. Each t-test showed a significant difference ($P < .02$) for each specimen tested. <u>These preliminary results suggest that CO_2 lased enamel may be identified using 3D-imaged scanned laser fluorescence.</u> Further research is ongoing utilizing other imaging techniques, an extended range of power levels and other types of lasers.

Caries Incidence in Patients and Non-patients in a Satellite Clinic.

I. C. BENNETT and A. I. ISMAIL. Faculty of Dentistry, Dalhousie University.

Dalhousie University operates a small satellite clinic in a Grade school in a low socio-economic area of Dartmouth NS. In the fall of 1990 the authors conducted a screening examination of all the 138 children in Grades 1, 2 and 3 at the school to test criteria for identification of children with high caries status. The examination recorded the number of decayed and filled surfaces and divided the children into four categories, **a) High caries risk**, cavities or restorations (not including incipient decay) in at least one surface of the anterior teeth, or, for Grade 1 children, three posterior surfaces with decay, or, for Grade 2 children, four posterior surfaces with decay, or, for Grade 3 children, six posterior surfaces with decay, **b) Low caries risk,** any child not in the High Risk group and not in the groups without any incipient decay or cavities, **c) Incipient,** incipient decay only, on the smooth surfaces of the teeth and **d) No decay,** no sign of any dental caries on the smooth surfaces, even incipient decay. All children of this age are eligible for free dental care sponsored by the provincial government. Half of the children seen were or had been patients at the Dalhousie clinic. The distribution of High Risk (26 patients, 25 non-patients), Low Risk(29 patients, 29 non-patients) and Incipient or No Decay patients (15 patients, 14 non-patients), was almost identical. The number of affected surfaces was 311 in patients and 385 in non-patients (5.65 and 7.13 surfaces per affected child). 46% of Grade 1 children were classified in the high risk group. The mean number of surfaces with incipient decay was significantly higher than the children in Grade 3. The group of children treated at the Dalhousie clinic had 1.48 fewer surfaces per affected child than the children treated elsewhere or not treated at all. This difference is not statistically significant, probably due to the small sample size. It was concluded that caries in primary teeth is still a significant problem in the children examined.

Effects of Fluoride, CO_2 Laser and dH_20 on Enamel Demineralization.

T. L. BORAN*, M. NICHOLS, and K. L. ZAKARIASEN. Faculty of Dentistry.

Our previous research has shown that low level CO_2 laser irradiation and fluoride on smooth surface enamel reduced sub-surface demineralization. This study examined the effect of dH_20 cycling on enamel treated with CO_2 laser radiation and fluoride subsequent to the demineralizing process. Twenty extracted third molars were selected and each tooth was covered with acid resistant varnish except for four windows on the buccal surface 1.5 mm in diameter, one control, and three experimental (fluoride, laser radiation, fluoride and laser radiation). 1.5 watts @ 0.15 sec with a 1.5 mm focal spot and NaF (2%) with 4 min. application was used. Ten teeth (Group A) were immersed in ten Caté solution (2.2m MCa^{++} and PO_4, 50mM acetic acid, .5 p.p.m F @ constant pH=4.3 for 12 days). The other ten teeth (Group B) were cycled for 12 days in dH_20 before the demineralizing process. 100μ hard tissue sections were prepared and embibed in dH20, photographed under a polarized light microscope and quantified with a planimeter. The data was analyzed by ANOVA and Duncan's Multiple Range Test for each group and Student T-tests for differences between groups. Means (mm^2) and standard deviations were as follows: Group A Control - 1.00 (.23); (Laser) - .59 (.13); (F) - .53 (.14); (F and Laser) - .77 (.17). Group B Control - .80 (.25); (Laser) - .31 (.12); (F) - .33 (.23); (F and Laser) - .56 (.23). All experimental conditions resulted in significantly smaller zones of demineralization than the control ($p=.05$). It would also appear a significant difference exists between group A (noncycled) and Group B (cycled in dH_20)($p=.05$). Research is ongoing to determine the parameters to increase consistency.

Pre-treatment and Post-treatment Dental Aesthetic Index Scores in Orthodontic Patients. W.K. LOBB, A.I. ISMAIL, D.A. BOWSER*
(Faculty of Dentistry, Dalhousie University).

This study evaluates pre-treatment and post-treatment esthetic differences in orthodontic patients, as measured by the Dental Aesthetic Index (DAI) (Cons et al., 1986). A total of 979 records for orthodontic patients treated between 1968 and 1989 were reviewed. Criteria for inclusion in the study included the presence of pre-treatment and post-treatment dental casts suitable for determination of the Dental Aesthetic Index. One hundred and thirty one patient records were included. There were 90 females and 41 males. Patient's age at start of treatment ranged from 81 months to 508 months. The DAI value improved (reduced) in the majority of the patients (85.5%) but for some (14.5%) it either remained the same or showed an increase between pre-treatment and post-treatment DAI values (treatment failure). There was no significant differences in the rate of treatment failure for males or females. Relating treatment failures to the components of the DAI through a logistic regression analysis identified "Extraction during treatment", and "Diastema size in mm.", as factors associated with treatment failure.($p \leq 0.050$) In conclusion, 14.5% of the patients showed no change, or a negative difference in pre-treatment and post-treatment DAI scores which for purposes of this study indicate treatment failure. Orthodontic treatment of patients with a pre-treatment diastema and/or extraction of teeth during treatment was more likely to fail.

This study was supported by the MRC (Canada),Farqueson Scholarship Program and the Canadian Association of Orthodontists through the Canadian Foundation for the Advancement of Orthodontics.

Characterization of Plasticizers for Methacrylate Polymers in Terms of Creep. D. W. JONES, M. CHIAROT* and J. A. JOHNSON.
(Faculty of Dentistry, Dalhousie University).

We have previously reported values for creep of commercial and experimental materials (AADR 1992). A significant relationship was found between creep and plasticizer efficiency. Our data has shown dibutyl sebacate (DBS) to have the lowest leachability and it was also the most efficient plasticizer.

Objective: To further study the effect of quantity and type of plasticizer on the creep for a polymer-gel ethyl methacrylate soft polymer system. Three plasticizers were compared, dibutyl phthalate (DBP), dibutyl sebacate (DBS) and butyl phthalyl butyl glycolate (BPBG). Creep in compression of experimental materials (n = 3) containing 70, 75 and 80% plasticizer were compared. The creep apparatus applied a stress of 15 KPa to the disc specimen (3 mm thick X 21.5 mm diam.). The specimens were aged at 37±0.5°C for 24 hours prior to testing in water at 37±0.5 °C. At 180 and 360 min. DBS (75-80%) and DBP (80%) produced significantly higher creep (P= 0.05). Polymers containing BPBG produced significantly less creep. The experimental materials containing 75 and 80% DBS as a plasticizer produced the highest mean creep values. Creep increased as a function of quantity of plasticizer. However, a greater increase in creep occured between 70 to 75 % than between 75 to 80% plasticizer. The data agrees well with the previously established efficiency of DBS for lowering the glass transition temperature of methacrylate soft polymer systems.

This research was supported by Canadian MRC grant PG45.

Evaluation of Serum and Tissue Levals of Beta-Carotene. Q. DAO*, G. KAUGARS, R. BRANT, W. RILEY, S. SILVERMAN, JR., J. LOVAS, V. SINGH.
(Med. Coll. of Va., UC - San Fran., Dalhousie University and , Hoffmann-LaRoche).

Low levels of dietary and serum beta-carotene (BC have been associated with an increased probability of developing certain types of cancer. The primary purpose of this investigation was to ascertain if a correlation exists between serum and tissue levels of BC. A strong correlation would allow serum to be used for the measurement of BC without the surgical trauma of tissue collection. Serum and tissue specimens were obtained from 56 patients prior to their entry into a supplementation trial for the treatment of their oral leukoplakia. The tissue specimens were obtained from normal oral mucosa. Blood and tissue specimens were analyzed by conventional HPLC techniques. The mean value (±SD) for the serum BC was 24.4(±26.4) μg/dl, and the mean for tissue BC was 1.5(±1.4) μg/gm. The Pearson correlation between the serum and tissue levels was 0.51, but rose to 0.63 ($P<0.002$) if males were excluded. Based upon our data in regard to this correlation, the following equation was generated:

Tissue BC = -0.47 + serum BC(0.02) + k
(K = 1.03 for males, k = 2.03 for females)

This equation permits an accurate extrapolation of tissue BC levels from serum data.

Supported by NIH DE09523-02 and Hoffmann-LaRoche, Inc.

Thermal Expansion Characterization of Composite Materials.

D. W. JONES, A. S. RIZKALLA, M. DAVIDSON* E. J. SUTOW & G. C. HALL.
(Faculty of Dentistry, Dalhousie University).

Objective: To compare the thermal expansion (α) and glass transition temperature (Tg) for 10 commercial composites. Specimens (cylinders 3.5 X 5.0 ± 0.2 mm) were tested (n = 3) in two conditions: i) dry(D) and ii) wet(W). Hydrated specimens were tested following 2 weeks storage in distilled water at 37 ± 1°C. The measuring probe applied a force of 100 mN. Specimens were heated from 10 to 150°C at a scanning rate of 10°C/min. The values for Tg(D) ranged from 48 ± 5 to 77 ± 2°C and Tg(W) ranged from 43 ± 2 to 63 ± 1°C. The values for $\alpha(D) < Tg$ ranged from 1.1 ± 0.2 to 6.8 ± 0.3 (X 10^{-5}/°C), $\alpha(W) < Tg$ from 2.9 ± 0.3 to 8.8 ± 0.1 (X 10^{-5}/°C). $\alpha(D) > Tg$ ranged from 2.9 ± 0.3 to 8.0 ± 0.5 (X 10^{-5}/°C) and $\alpha(W) > Tg$ 3.7 ± 0.4 to 12 ± 0.9 (X 10^{-5}/°C). Reduction in Tg was observed for 8 out of 10 materials comparing dry vs. wet specimens ($P = 0.05$). Increases in $\alpha < Tg$ for five out of 10 materials was observed when comparing dry vs. wet specimens ($P = 0.05$). Increases in $\alpha > Tg$ for 7 out of 10 materials was observed when comparing dry vs. wet specimens ($P = 0.05$). The materials with the highest filler content gave significantly lower thermal expansion ($P< 0.05$). Thermal expansion (above and below Tg) and Tg of composites containing higher % of resin matrix were significantly affected by storage in water.

This research was supported by MRC (Canada) Grant PG45.

Assimilation of anionic amino acids by *Porphyromonas gingivalis* and *Prevotella intermedia.*

R. L. WHITE, H. N. SHAH, A. L. DOHERTY*, R. SETH and S. E. GHARBIA
(Depts. Chemistry & Oral Biology, Dalhousie University).

Nitrogenous substrates such as amino acids are catabolized by most periodontal pathogens. Among these, we have shown previously that anionic amino acids such as aspartate and glutamate and their corresponding amides are among the favoured substrates. In the present study we examined the uptake of these amino acids over 5 days by 6 strains each of *P. gingivalis* and *P. intermedia*. Incorporation was measured in solutions containing a mixture of 20 amino acids (1mM) and was analyzed by HPLC after precolumn derivatization with o-phthalaldehyde. All strains of *P. intermedia* incorporated significant concentrations (> 80%) of both aspartate (Asp) and asparagine (Asn) with a net production of glutamate (Glu) up to 0.3mM. By contrast, *P. gingivalis* converted 20% of the utilized Asn to Asp with a net production of Glu. The increased concentration of Asp produced correlated with the higher levels of asparaginase activity of *P. gingivalis* compared to *P. intermedia*. Assayed at pH 8.0, *P. gingivalis* strains had an apparent Km of ~ 5 x 10^{-4} M whereas *P. intermedia* enzyme had a Km value of ~2.0 x 10^{-4} M. Further experiments indicate that the low incorporation of Asp might be due to saturation of the free monomer which is made available by the high activity of asparaginase in *P. gingivalis*. These results suggest that the net production of Glu is most likely due to a transamination or a reductive amination reaction involving 2-oxoglutarate during the initial stages of Asn catabolism. Anionic amino acids are major catabolic substrates, however, their utilization and mechanism of degradation vary among periodontal pathogens.

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

Agar Overlay Cytotoxicity Testing of an Endodontic Obturating Device.

W. C. FOONG*, E. J. SUTOW, K. L. ZAKARIASEN, P. HIDI, D. W. JONES
(Faculty of Dentistry, Dalhousie University).

The Thermafil Endodontic Obturator uses various carrier materials for warm gutta-percha placement. Carriers remain in the canal and the tooth is restored as indicated. The objective of this study was to determine the cytotoxicity of the leachable components of the carrier materials using a recommended ISO method (ISO, Technical Report, #7405, 1984). Four carrier materials were evaluated: 1. Ti-alloy (Ti-6Al-4V), 2. modified Type 302 series stainless steel, 3. Vectra (Grade A950) and 4. Polysulfone (Grade UDEL (R) MG-11). Filter paper impregnated with either dibutyltin diacetate (DBTD) or DMEM were used as the positive and negative controls, respectively. Carrier test samples (n = 5, per test material), positive (n = 6) and negative (n = 6) controls were placed in contact with an agar layer overlaying a confluent monolayer of L929 fibroblasts cells stained with neutral red vital stain. After 24 hour incubation, the effect of leachable toxic substances was evaluated with respect to the extent of the zone of discoloration (Z) under and around the test sample and the degree of lysis (L) in the zone. The magnitude of the response is reported in terms of a "response index", R = Z/L. All the test materials and the negative control resulted in a response index (R)= 0/0. The positive control, (DBTD), consistently produced a zone of discoloration and lysis, the average response index (R) = 4/4.5. Based on z test for proportions, the test materials and the DBTD group are significantly different at $P<0.01$. Conclusion: the Ti-alloy, stainless steel, Vectra and Polysulfone carriers were not cytotoxic.

Effect of Separators on Surface Detail Reproduction of Flexible Dies.

R. B. PRICE, J. D. GERROW* and A. T. HARVEY.
(Faculty of Dentistry, Dalhousie University).

Recently it has been suggested that flexible die materials be used to simplify the fabrication of indirect composite acrylic resin inlays. The flexible die material is used instead of a gypsum product to produce a die on which the inlay is fabricated. This study compared the surface detail reproduction of potential flexible die materials (EOS, Impregum F, Panasil Medium, Extrude Extra and Extrude Medium) when used in combination with four impression materials (EOS, Proof, Panasil Light, and Extrude Light) to a control system made using Reprosil Light and Die Keen. Five test dies of each of the combinations were made in random order using the A.D.A. apparatus for compatibility testing. Materials were prepared according to manufacturers' instructions. Where indicated a Teflon/Silicone spray or the EOS system separator were used between the impression and die materials. The dies were evaluated under 10X magnification, using low-angle illumination. Dies were rated from 1 to 6 based on the reproduction of the 20μ line. Friedman's ANOVA and RIDIT analysis ($P \leq .05$) supported the following conclusions: (1) The Impregum F/Extrude Light die/impression combination produced the best surface detail which was better than the control dies. (2) The Proof/Extrude Medium die/impression combination was not significantly different from the control dies. (3) All remaining die/impression combinations required the use of a separator and had worse surface detail reproduction than the control dies. This study was partially supported by Kerr Manf. Co.

Conjugal transfer of Tn*4351* into *Prevotella intermedia* and *Porphyromonas gingivalis*. S. E. GHARBIA* and H. N. SHAH.

(Faculty of Dentistry, Dalhousie University)

Virulence of putative periodontal pathogens is multifactorial. To determine the possible role of each potential component in disease development, its isolation and purification is essential. The deletion of the activity of such factors from the parent cell by transposon insertion mutagenesis provides an alternative approach. In the present study we describe a procedure for the insertion of Transposon Tn*4351* into *P. gingivalis* ATCC33277 and *P. intermedia* ATCC 25611. *E. coli* HB101 which harbours R751::Ω4 (Shoemaker *et al*, 1986; J.Bacteriol. 165:929-36) was grown in LB containing 10 mg/ml tetracycline to the late log phase. Cells were harvested and diluted to 1×10^4 cell/ml in Trypticase soy broth (TSB) and 30 ml spread over the surface of *P. gingivalis* blood agar cultures (bacterial count of ~10^5). Plates were incubated aerobically for 2 hours at 37°C, then transferred to an anaerobic atmosphere for 24 hours. Each culture was harvested aseptically and suspended in 20 ml TSB containing 70 mg/ml gentamicin and 10 mg/ml erythromycin and incubated anaerobically for 16 h, after which 10-fold serial dilutions were plated on TSB-agar containing gentamicin and erythromycin. Cultures were incubated anaerobically for 5 days prior to screening for transposition. The rate of transconjugation of *P. gingivalis* was 4.5×10^{-5}, whereas the rate for *P. intermedia* was two folds higher. The recipient viable count for *P. intermedia* was 80% of the original count while only 60% transconjugants were recovered from *P. gingivalis*. Using this procedure transconjugants. deficient in several properties have been isolated. (Supported by MRC Grant # DG-411).

Effects of Various Cleaning Devices on Implant Abutments.
C. A. BAIN, S. GREEN*, G. G. B. BAIN, D. PRICE.
(Faculty of Dentistry, Dalhousie University and Sacred Heart, Nova Scotia).

The softness of CP Titanium has led to the development of various cleaning devices which are recommended for abutment cleaning. The purpose of this study is to evaluate the surface roughness associated with 4 cleaning devices. New 10 mm Branemark abutments were mounted in a jig exposing one third of the circumference. Each section was exposed to one cleaning method for an in vitro period considered equivalent to 5 years normal homecare and/or professional maintenance. Devices evaluated were Nobelpharma and Hu Friedy plastic scalers; Prophyjet air polisher and Rotadent toothbrush. 5 sections were treated using each method and 3 untreated control surfaces were also evaluated. Representative uncoated Scanning EMs at 1000x magnification were taken and 14 evaluators were asked to rank the micrographs on a 1 to 4 scale of "smooth" to "very rough"; only the control was identified as a reference. Means, SD and ANOVA were calculated. The Hu Friedy instrument produced a significantly rougher surface (mean 3.43 ± .51) than Prophyjet (2.86 ± 1.46) $p<0.05$; Rotadent (2.36 ± .50) $p<0.05$ and Nobelpharma instrument (1.35 ± .50) $p<0.01$. It is concluded that the various devices tested have significantly different effects on surface roughness and that any new device should be carefully tested in vitro prior to marketing. (Partial support from Nobelpharma Canada.)

Polymer Bead Size and Distribution of Synthesized and Commercial Methacrylates. G. C. HALL*, D. W. JONES and P. HURLEY.
(Faculty of Dentistry, Dalhousie University).

OBJECTIVE: To compare particle size & distribution of 11 commercial & 3 experimental batches of polymer beads of higher methacrylate soft polymer materials. Particle size and distribution of powders were determined by low-angle forward scattered light from a laser beam (Microtrac Model 9200). Containers of commercial materials were rolled before sampling the powders. The surfactant Alipal at 2% was found to be the best suspending agent. After sonication a pipette was used to place the samples (n= 5) into the Microtrac circulator. Four commercial materials were available with different lot numbers, these gave variations in particle size distribution from lot to lot. However, larger variations in particle size distribution occurred in multiple repeat samples (n= 5) from the same batch, due to sampling technique. This repeat sampling gave a mean particle size of 68.18 ± 16.44 µm. Particle size of commercial materials at 10, 50, 90 volume % ranged from, 27.51, 46.26, 74.61 µm to, 42.38, 81.79, 149.19 µm. The 11 commercial powders gave a mean average particle size of 57.80 ± 13.54 µm. Experimental polymers (Jones *et. al.* Abstract #2425 IADR 1991) had a mean size of 49.54 ± 8.78 µm. Batches B39 and 64 with identical polymerization yield, gave mean particle sizes of 52.31 and 56.6µm respectively despite a difference of 144 RPM's in the stirring rate during manufacturing. CONCLUSIONS: Riffling techniques should be employed for selecting samples for particle size analysis. Synthesized polymers were produced having mean particle size not significantly different to commercial materials $P<0.05$). Suported by MRC Grant PG45.

Effect of Liposomal Encapsulation on Retinoic Acid Activity.

B. B. HARSANYI*, P. HIDI, B. SINGH and M. MEZEI.
(Collage of Pharmacy and Faculty of Dentistry, Dalhousie University).

We have previously shown that liposomes increase local and decrease systemic drug concentrations. In this study, we compare the biologic effects of retinoic acid, RA, in a liposomal and a conventional dosage form. Liposomal RA was prepared following a method patented by Mezei; the conventional dosage form was a cream. Both contained 0.05% RA. Nine albino guinea pigs were divided into three groups. Group 1 was treated with liposomal, group 2 with conventional RA; group 3 received no drug (control). The dorsal surfaces of both ears and six areas on the shaved skin of the back were painted daily ; each guinea pig had sites treated for 1,2,3,4,5, 11 and 14 days when killed 24 hrs after the last treatment on day 15. Skin specimens were processed for histology and evaluated in a blind fashion. Epidermal thickness (ET) was measured by image analysis. Clinically, group 1 showed mild redness of the ears from day 7 -11, group 2 marked redness and scaling from day 4-11. Microscopically, group 1 showed no pathology, but group 2 had marked psoriasiform changes. In group 1, ET increased to 209 % of control by day 5, peaked at 361% on day 7 and decreased to 248% by day 12. In group 2, ET reached 208% of control by day 4, peaked at 344% on day 6, and decreased to 233% by day 12. As tested by ANOVA, both RA groups were significantly ($p<0.01$) higher than control, but did not differ from each other at treated sites. However, the untreated ventral aspects of ears in group 2 also showed increased ET at $p<0.05$ while group 1 did not. Liposomal encapsulation did not change pharmacologic activity, but limited drug diffusion and decreased adverse side effects. MRC- MA-6664.

A Comparison of Marginal Adaptation of Indirect Composite Resin Inlays.

J. D. GERROW, R. B. PRICE, and A. T. HARVEY*.
(Faculty of Dentistry, Dalhousie University).

Several one appointment indirect techniques have recently been suggested for the fabrication of composite acrylic resin inlays. Some of these techniques involve the use of flexible die materials. This study compared the marginal adaptation of composite inlays made using 6 impression/die systems(EOS, Extrude/ImpregumF, Extrude/Extrude Extra, Extrude/Extrude Medium, Proof/Extrude Medium, Extrude/Suprastone). Gold inlays fabricated using a gypsum die system(Extrude/Suprastone) served as controls. In random order, using manufacturers' instructions, 10 impressions were made of a metal master die with a Class II composite resin inlay preparation for each test system. The impressions were poured in the designated die material. Composite resin(Herculite) inlays were made and finished under magnification on each test die. The 5 gold inlay controls were fabricated using traditional techniques. After standardized seating of the inlays on the master die, the marginal opening at three previously determined and standardized sites was measured using a measuring microscope. ANOVA and Scheffé F-test supported the following conclusions. (1) There was no statistically significant difference in marginal adaptation at 2 of the measured points(buccal and distal occlusal). (2) Inlays made using the Extrude/Extrude Extra system had the worst marginal adaptation at the third measurement point(gingival). Although not statistically significant, the test systems had larger mean marginal openings and ranges than the control system. This study was supported by Kerr Manufacturing Co.

Surface Area and Size Analysis of Composite Filler Particles.

A. S. RIZKALLA, D. W. JONES, P. HURLEY*, E. J. SUTOW & G. C. HALL.
(Faculty of Dentistry, Dalhousie University).

Objective: To compare surface area and particle size of fillers for 10 commercial composites. Particle size distribution was determined by a light scattering technique (SRA Microtrac), and surface area by nitrogen adsorption. Filler particles were extracted from the polymer matrix by a solvent, and the remaining organic material was thermally decomposed. Specimens for surface area measurement were degassed for 60 min at 150°C, prior to nitrogen adsorption. Filler content was determined by weight (n = 3). Filler ranged from 52.56 ± 0.04 to 85.72 ± 0.13 weight (%). Surface area values (n = 3) varied from 3.8 ± 0.1 to 67.7 ± 1.0 m^2/g. Mean particle size (n = 3) varied from 1.30 ± 0.02 to 13.30 ± 1.10 μm. A Student-Newman-Keuls rank order test separated mean surface area for 10 materials into 7 different groups (P= 0.05). Mean particle size separated into 6 groups (P= 0.05) and weight % of filler separated into 5 groups (p= 0.05). A correlation was found between previously established dynamic Young's modulus (E) and % filler (P< 0.001). Correlations were also found between E and range of particle size/mean particle size (P<0.01) and between Poisson's ratio and range of particle size/mean particle size (P< 0.001). A correlation was also found between E and the product of the range of particle size/mean particle size X % wt. filler (P< 0.001). No correlation was found between particle size and surface area. Conclusion: Significant differences in weight, size distribution and surface area of filler particles were observed amongst the ten materials. Supported by MRC PG 45.

Sealant Decisions in Children: A Two-year Follow-up.

A. I. ISMAIL*, P. GAGNON, J. M. BRODEUR, M. OLIVIER
(Faculty of Dentistry, Dalhousie University, Laval University, & Hôpital St-Luc, Montreal).

The purpose of this longitudinal study is to correlate between tooth status and restorative treatments in a universal children's dental insurance program in Quebec which does not cover sealants. Out of a sample of 911 randomly selected children, 6-9 years of age, in 1990, in Montreal, 816 were re-examined in 1991 and 733 in 1992. The occlusal surfaces of first peramanent molars (fpms) were classified into: sound, non-cavitated pits and fissures caries, arrested and active cavitated caries. This analysis included only the 733 children with 2 year data. Out of 4398 surfaces of fpms 11.62% were sealed and 10.9% were filled during the 2 years. Out of those sealed after 2 years, 80.2% (411) were placed on sound tooth surfaces in 1990 and 11.9%(61) and 7.2%(37) were placed on teeth erupted after 1990 and non-cavitated carious surfaces, respectively. The retention rates of sealants in 1990 were 93.2% and 86.0% after 1 and 2 years, respectively. Non-cavitated carious surfaces were more likely to be restored than sealed. Only the probability of restoring a non-cavitated fpm was significantly higher than that of a sound fpm (P < 0.05). Sound fpms in 1990 were 3.83 more likely to be restored after 2 years than sealed fpms. Children of parents with a university or college degree had significantly higher probability of receiving sealants. Caries status (free vs high caries) was not associated with probability of receiving new sealants. Regular dental attenders were twice more likely to receive sealants. After two years, more new sealants were placed on fpms than fillings. Sealants were effective in reducing the need for restorations.

The study is supported by an NHRDP grant 6605-1340-CD.

Correlation of Microprobe and AAS Analysis of Glass Biomaterials.

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We are currently synthesizing a range of glass compositions by wet chemical methods. A need exists to determine the final bulk chemical composition for the various glass batches, since the chemical composition can affect important physical properties. Two methods of analysis were used; i) electron microprobe analysis (EMP), using a JEOL 733 microprobe and ii) atomic absorption spectroscopy (AAS) using a Perkin-Elmer 2380 system. Analysis of the surface of a solid glass sample by EMP is more rapid than the use of AAS which analyzes an aspirated solution of an acid digested glass sample. Objective: To compare the chemical analysis results of seven different alkali silicate glasses using EMP (n= 5) and AAS (n= 3) test analysis methods. Regression analysis gave a good correlation between the Si content in each glass when analyzed by the two methods ($P<0.01$). A further good correlation was found between the K values obtained using the two analytical methods ($P< 0.01$). However, there was no correlation obtained in the case of the two analysis methods for Na, at $P< 0.05$. A Student-Newman-Keuls rank order test could not separate mean values for Na, K or Si obtained by the two analysis methods for the glasses ($P= 0.01$). Conclusion: The microprobe (surface) and atomic absorption spectroscopy (bulk) analysis methods indicated essentially the same chemical composition for a range of different glasses.

This research was supported by MRC (Canada) Grant PG45.

A Clinical Study of Pre-treatment Pain in Endodontic Patients . W.K. LOBB*, K. L. ZAKARIASEN, P. J. MCGRATH, D. L. BARKLEY, P. A. GRIFFITH.

(Faculty of Dentistry, Dalhousie and Marquette Universities)

While some reports exist, the nature of pre-treatment, treatment, and post-treatment endodontic pain has not been fully elucidated. This research is designed to study pain patterns in endodontic patients before, during, and after therapy. This initial report describes the sample demographics, and pre-treatment pain patterns. 177 consecutive, first appointment endodontic patients completed pre-treatment and post-treatment questionnaires. In addition, the endodontist completed a questionnaire for each patient. 105 (59.3%) were female and 72 (40.7%) were male, the mean ages being 43.3 years (female) and 48.2 years (male) with a range of 16 to 90. Subjects responded to questions regarding oral pain by rating their pain on a 6 point scale. Of the 177 subjects, 31 (17.5%± 5.6 - 95% C.I.) reported pain in the mouth at the first appointment (mean duration was 11.8 days, range = 0 to 31 days). The pain was constant in 19 (61.3%± 17.4) of 31 subjects. Of the 31, 26 (83.9%± 13.2) reported thermally induced pain, and 23 (74.2%± 15.7) reported pain on occlusion. Of the 31, 24 (77.4%± 14.7) had pain which was considered to be significant (3 or greater, 3 = "Pain- I can't ignore it but I can do my usual activities"). 12 of 31 (38.7%±17.3) reported use of non-prescription analgesics to manage pain, and 6 of 31 (19.4%±13.9) reported use of prescription analgesics. These results indicate that nearly one in five endodontic patients experience pre-treatment pain, and that when it occurs, it is over an extended time period, is constant, and thermally and/or occlusally induced in the majority of subject. It appears that over half of the patients reporting pain, used non-prescription or prescription analgesics. This ongoing research seeks to develop a predictive model for management of post-treatment endodontic pain.

Should the Drinking Water of Truro, Nova Scotia, be Fluoridated?

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An epidemiological assessment of differences in caries and fluorosis prevalences between children in Truro (< 0.1 ppm) and Kentville (fluoridated at 1.1 ppm in 1991), Nova Scotia, Canada, was completed in 1991. Out of a total of 429 children, in grades 5 and 6, in the 2 towns in 1991, 219 (51%) were examined. Parents answered a self-administered questionnaire. The examination criteria differentiated between non-cavitated and cavitated carious lesions. Dental fluorosis was measured using the TSIF index. Examiner agreement was excellent. Of the children examined, 80 (36.5%) drank water (fluoridated or non-fluoridated) from municipal water systems during the first 6 years of life. The percentage difference in mean DMFS scores between children in the fluoridated and non-fluoridated groups is 17% (delta DMFS1 = 0.7) when non-cavitated carious lesions are included and 39% (delta DMFS2= = 1.1) when they are excluded. The differences are not statistically significant. The significant risk factors associated with the DMFS1 and DMFS2 scores identified by a stepwise multiple regression analysis are: education level of the father, sex, and number of years of reported use of toothpaste during the first 6 years of life. Dental fluorosis (mainly TSIF score of 1) was present in 41.5% and 69.2% of the children in the non-fluoridated and fluoridated groups, respectively. Residence in a fluoridated area and the educational status of the mother were positively associated with fluorosis status. It was concluded that water fluoridation be recommended as a part of an overall plan to educate the public and the medical and dental professions with respect to the proper and safe use of fluoride products.

Tarnish Resistance of Three Polishing Methods for Cast Alloys.

E. J. SUTOW, G. DOYLE, A. S. RIZKALLA and D. MacNEIL.*
(Faculty of Dentistry, Dalhousie University)..

Previous studies have shown that a customized polishing method was rated clinically superior for surface finish compared with two conventional methods (IADR, Abst. #1641, 1991). However, the customized method provided the lowest sulfide tarnish resistance for a Cu-Al alloy (AADR, Abst. #1584, 1992). The objective of the present study was to evaluate the three polishing methods for their influence on tarnish resistance in sodium chloride and chlorhexidine solutions. Four representative alloys were cast as discs and polished using the following methods: 1. Brasseler white and Dedco green rubber wheels, Degussa pre-polishing paste and a commercial sink cleaner (customized method); 2. Shofu gold polishing kit; 3. fine emery, fine garnet, medium and fine cuttle sandpaper discs, tripoli and rouge. The alloys were: A. Type III gold; B. high gold content ceramo-metal; C. Ni-Cr-Be; and D. Type III Cu-Al. Ten specimens of each alloy were tested for each polishing technique (N=30). For the sodium chloride solution, the Cu-Al alloy was tested at 0.009%, while the remaining alloys were tested at 0.9%. For the chlorhexidene solution, all alloys were tested at 0.2%. Specimens were immersed for 72 h, at 37°C. Tarnish was assessed by a colorimeter, using the CIELAB system. The ΔE^* data were statistically analyzed using a Duncan multiple comparison test ($p=0.05$). Analysis showed that the tarnish resistance rankings of the three finishing methods were alloy and tarnish solution dependent. It was concluded that finishing Method 3 gave the best overall tarnish resistance to the four alloys in the two solutions.

Comparison of Hand and Hand/Sonic Techniques with an Alternative Instrumentation System.

K. A. ZAKARIASEN, K. L. ZAKARIASEN, M. M. MCMINN*.
(Faculty of Dentistry, Dalhousie and Marquette Universities).

Research has shown that hand and hand/sonic enlargement techniques can result in properly shaped canals. This research is designed to compare curved canals shaped by step-back and hand/sonic filing (A, B) with those shaped by a hand/mechanical technique (C, D, E, F) using 4 different file combinations in an Endo-Lift contra-angle. The six groups studied were A) Step-Back Hed., B) Hed./Sonics, C) Hed./Endo-Lift (15, 35 K-flex), D) Hed./Endo-Lift (15, 35 Hed.), E) Hed./Endo-Lift (15 K-flex, 35 Hed.), F) Hed./Endo-Lift (15,25,35 Hed.). Sixty #101 Endo-vu blocks were accessed, randomized into 6 groups and preflared to 20 mm with hand files, hand/sonics or hand/Endo-Lift. Apical preparations were completed to #25 at 23 mm. Canal flaring was continued with step-back, sonic or Endo-Lift filing until a D11T spreader would freely penetrate to 22 mm. One operator prepared all canals and instrumentation times were recorded. Canal preparations were randomly evaluated blind for apical transportation, ledge formation, and hour-glass shaping of the canal. The mean preparation times range from 7.1 to 8.2 min. with no significant differences ($p = .05$) being observed. No ledging occurred in any groups. Some occurrences of apical transportation and hour-glass shaping were observed, but these were infrequent and no significant differences ($p = .05$) were observed between groups. Under the conditions of this study, the results indicate that step-back, hand/sonic and hand/Endo-Lift techniques can all provide satisfactory canal preparations with no or minimal ledging, apical transportation and hour-glass shaping of the canal space.

Effect of Plasticizers on the Permeability of Hamster Oral Mucosa.

W. C. FOONG, J. POTVIN*, P. HIDI, D. W. JONES.
(Faculty of Dentistry, Dalhousie University)

Previous studies have shown that plasticizers such as dibutyl phthalate (DBP) can leach out of denture soft polymers (Jones *et al.*, 1988). We have shown that dibutyl sebacate (DBS) is a more efficient plasticiser than DBP and it exhibits a significantly lower leachability than DBP (Hall *et al.*, 1991). However, there are no data on the effect of these plasticizers on the permeability of oral mucosa. This study examined the effect of two plasticizers on the *in vitro* water permeability of normal or traumatized (tape stripped, Pink *et al.*, J. App. Biomater. 1991) hamster pouch mucosa. Hamster pouches were divided into four groups: untreated normal control (N); DBP treated normal (NDBP); DBS treated normal (NDBS); untreated traumatized (T); n=6 per group. Mucosa specimens were mounted in a glass diffusion cell system. The mucosa was pretreated with 20 µL DBP or DBS for 0.5 hour. Excess plasticizer was removed and permeability to tritiated water after 5, 15, 30, 60, 120 and 240 mins at 37 °C determined. All groups achieved a steady-state after 120 to 240 mins. Traumatized (T) and plasticizer treated (NDBS, NDBP) mucosa resulted in higher permeability constant (Kp) than normal untreated mucosa ($P< 0.05$, ANOVA). There were no significant difference in Kp between traumatized and plasticizer treated (NDBP, NDBS) mucosa. Conclusion: DBP and DBS increase water permeability of hamster pouch mucosa.

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Effect of Supervised Home Bleaching on the Oral Health.

R. B. PRICE* and J. D. STERRETT
(Faculty of Dentistry, Dalhousie University).

While supervised home bleaching to lighten teeth has become popular, little is known of the effect of carbamide peroxide (CP) on the oral tissues. The purpose of this study was to assess the i) oral soft tissue/gastro-intestinal ii) periodontal and iii) microscopic enamel effects of a 10% CP solution (Opalescence) in a custom mouthguard for 14 nights in 9 subjects. A questionnaire was used to evaluate oral and gastro-intestinal changes. Periodontal assessment was made of the probing depths on teeth 1.4 to 2.4 and the crevicular fluid flow (CFF) was measured using the Periotron. Replicate impressions of the tooth surfaces were assessed using the SEM to see if any changes occured during bleaching. RESULTS: One subject experienced TMJ pain and discontinued home bleaching after 4 nights. The remaining 8 subjects continued home bleaching for 14 nights and experienced no oral soft tissue/gastro-intestinal problems apart from a temporary increase in tooth sensitivity to cold. All 8 subjects perceived their teeth to be lighter at the end of the study although 4 subjects hoped that their teeth would be lighter. Scheffé F-test for multiple comparisons ($P \leq 0.01$) showed that the CFF and periodontal pocket depths were not significantly affected by 14 nights of home bleaching using 10% CP. No visible changes were found between the pre and post bleaching SEM photographs of the teeth. It was concluded that for the parameters assessed, there were minimal side effects after 14 nights of bleaching using 10% CP.

This study was partially supported by Ultradent Inc.

Indentation Fracture Toughness of Glass Biomaterials: Computational Methods.

A. S. Rizkalla*, D. W. Jones, R. P. Miller, and E. J. Sutow.
(Faculty of Dentistry, Dalhousie University).

We have a need to determine fracture toughness (K_{Ic}) for glasses synthesized by wet chemistry. OBJECTIVE: To evaluate 8 formulas from the literature for calculating K_{Ic} from indentation cracks in the Palmqvist regime. METHODS were as used previously (Rizkalla *et al.* IADR 1992), Vickers indention was used to produce cracking (n= 15) and Knoop indentation (n= 5) for 'true hardness' (H_0) for 5 glasses (2 experimental & 3 commercial glasses). Young's modulus (n= 3) was obtained using an ultrasonic method. RESULTS: K_{Ic} values using 8 formulas ranged from 1.02 ± 0.05-1.89 ± 0.03 for an 8 component experimental glass; 0.95 ± 0.03-1.48 ± 0.15 for a 3 component experimental glass; 1.60 ± 0.02-3.52 ± 0.14 for fused silica; 1.05 ± 0.03-1.47 ± 0.03 for soda lime glass and 1.16 ± 0.02-2.31 ± 0.07 for a borosilicate glass. Dimensional analysis of relationships for Palmqvist cracks ($0.25 \leq l/a \leq 2.5$) and median cracks ($c/a \geq 2.5$) showed that Blendell (B), Niihara *et al.* (N) & Evans (E) are in good agreement for the 5 glasses tested (P= 0.01). The 5 other formulas showed higher values for K_{Ic} and produced different rankings (P= 0.05). A Student-Newman-Keuls rank order test did not separate K_{Ic} means for 2 out of 5 glasses using formulas B, N and E (P= 0.05). For two other glasses no separation occured for formulas B and E (P= 0.05). For the fifth glass no separation occured for formulas E & N. CONCLUSION: All 5 glasses evaluated were in the Palmqvist crack regime, in such cases, formulas B, N & E are recommended to be used for calculating the indentation fracture toughness (K_{Ic}). MRC, PG 45.

Aspects of subgingival plaque ecology; biodiversity and functional interaction of species. H. N. SHAH* and S. E. GHARBIA
(Depts. Oral Biology, and Microbiology and Immunology, Dalhousie University).

Chemosystematic and molecular taxonomic clarification of several subgingival plaque bacterial species now provide a firm basis for studies on the interrelationships between microbial biodiversity and ecological processes. In the present study we present preliminary data on such interactions utilizing ribosomal RNA gene restriction analysis and a range of physiological functions such as activities of proteinases, peptidases, lipases, racemases, dehydrogenases, electron transport components and amino acid fermentation patterns among strains of *P. gingivalis, F. nucleatum, T. denticola, P. intermedia* and *L. buccalis* using previously described methods (Shah et al. Chemotaxonomic methods, in Anaerobic Microbiology, a Practical Approach, IRL Press, 1991, pp 65-100). In all cases biodiversity was clearly evident and expressed mainly through extracellular enzyme activities whereas intracellular components such as dehydrogenases (eg. malate and glutamate dehydrogenases) and electron transport complexes (eg cytochromes and menaquinones) remained highly conserved within species. Extracellular enzymes affected the functional capacity of species and substrate processing. Subtle variation in amino acid fermentation patterns were evident that reflected both species diversity and strain adaptation. These results demonstrate that biodiversity is a feature common to all key periodontal pathogens and is likely to exhibit a profound effect on the functional capacity of bacterial communities in vivo. (This work was supported by MRC Grant # DG-411).

Long-Term Subcutaneous Tissue Reaction to Experimental Denture Soft Polymers.
B. SINGH*, B. B. HARSANYI, P. HIDI and D. W. JONES.
(Faculty of Dentistry, Dalhousie University).

We have previously shown that two new denture soft polymers, A & B, produced no ill effects by seven days . In this study, we compare the 28-day effects of the experimental polymers to those of a commercial soft liner,C and a negative and a positive control material, N & P. Material A is a heat-cured polymer of ethyl and lauryl methacrylate(m); B a room temperature gelling copolymer of ethyl and lauryl m. Both use dibutyl sebacate as plasticizer. C containes poly(ethyl)- and (methyl)m as powder and dibutyl phthalate as plasticizer. For N, the powder of C is blended with ethanol; P containes powder C with dibutyl tin diacetate. Ten guinea pigs were used; each received four implants: A,B,C and N or P. Tissue reaction was determined by implant rejection, histologic features and width of the inflammatory-fibrous capsule measured with an image analyzer. 5/5 P and 2/10 C implants were rejected, while all others remained *in situ*. B implants were surrounded by fibrous, the others by granulation tissue. The Student-Newman-Keuls test ranked maximal capsule widths as B=A=N <C and minimal capsule widths as B<A=N=C at $p<0.05$. Polymer B is the least irritant. Polymer A is no more irritant than the negative control. Both experimental materials are less irritant than the commercial polymer.

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Further Evidence to Support the Cysteine Proteinase Nature of Gingivain.

S. SREEDHARAN*, H. N. SHAH, S. E. GHARBIA, and K. BROCKLEHURST
(Dalhousie University, & QMW, University of London, UK).

Porphyromonas gingivalis produces a plethora of putative virulence factors of which the extracellular proteinase is considered to be particularly significant. Despite major advances in the molecular cloning of the genes encoding this enzyme, its expression and sequence determination, the nature of the enzyme remains controversial. Most workers consider the enzyme to be a trypsin-like serine proteinase. We have previously purified the enzyme of strain W83 and shown by its method of isolation and reactivity characteristics towards 2,2' - dipyridyl disulphide that this enzyme (referred to as "gingivain") is a cysteine proteinase. In the present study evidence, based on interactions with the inhibitor L-3-carboxy-trans-2,3-epoxypropionyl-leucylamido-(4-guanidino) butane (E-64), is presented that gingivain may be similar to clostripain, the cysteine proteinase from *Clostridium histolyticum* rather than to papain. The catalytic activity of gingivain towards a-N-benzoyl-L-arginine-4-nitroanilide (L-BAPNA) was studied in the absence and in the presence of E-64 by recording the increase in A_{410} and quantified by using Δe_{410} = 8800 $M^{-1}cm^{-1}$. In the absence of E-64, and with [L-BAPNA] = 10-130μM, Km was determined as 30μM in Tris/HCl buffer pH 7.4, I 0.1M, at 25°C. Addition of E-64 produced instantaneous inhibition which was not followed by subsequent time-dependent inhibition. The extent of the instantaneous inhibition increased with increase in [E-64]. Kinetic data with [L-BAPNA] = 10-130μM in the presence of 40μM E-64 when combined with the analogous data collected in the absence of E-64 demonstrated the competitive nature of the inhibition of gingivain and a Dixon plot with [E-64] = 10-100μM and [L-BAPNA] = 10, 50 and 110μM provided a value of Ki = 15μM. This type of inhibition is similar to that of clostripain and provides further support for the cysteine proteinase nature of gingivain. This work was supported by MRC Grants DG-411 (Canada) and 99004592 II SD (UK).

Epithelial Remnants in the Crestal Periodontium of the Primary and Permanent Dentition of Beagle Dogs.

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(Dalhousie and Gothenburg Universities).

The purpose of this study was to compare epithelial remnants (EPRs) of the crestal periodontal tissues from the primary and permanent dentition of beagle dogs. The material consisted of 7 beagle dogs which had been raised under similar conditions. Since birth the dogs had been subjected to professional prophylaxis 4x/year. At the age of 10 weeks (primary dentition) and 15 months (permanent dentition) biopsies were obtained from the contralateral mandibular $_{02}P$, $_{03}P$ and P_3, P_4 premolar regions and prepared for histologic analysis. Two areas, (1) the supracrestal region and (2) the marginal periodontal ligament region, were identified. The supracrestal region was further divided into four equal compartments. The histologic parameters studied included the (i) EPR frequency (ii) EPR size (iii) EPR-Root Distance (iv) EPR-Bone Distance (v) epithelial cell area. No difference was observed in the number, size and relative location of EPR units in the supracrestal region (total and various compartments) between the two dentitions . A similar finding was observed for EPR units of the periodontal ligament region for all parameters except EPR-Bone distance. No difference was noted in the cell area of EPRs between either dentition for either region. Epithelial remnants of the supracrestal region in both groups of dogs tended to be more frequent, larger and positioned further from the root surface than the EPRs of the periodontal ligament region. It was concluded that EPRs are a normal constituent of the periodontal tissues of the primary dentition of the beagle dog and appear to be similar to those found in the permanent dentition of young dogs.

Corrosion Evaluation of Alloys Used for an Endodontic Filling Device.

E. J. SUTOW*, K. L. ZAKARIASEN, W. C. FOONG, D. W. JONES and G. C. HALL.
(Faculty of Dentistry, Dalhousie University).

The metallic carriers used in Thermafil Endodontic Obturators are either a modified Type 302 stainless steel or Ti-6Al-4V. In the absence or loss of an apical seal, release of metallic ions is a potential biocompatibility problem. It was the objective of this study to evaluate the crevice corrosion behavior of these carriers using long-term and accelerated testing. For long-term testing, five gutta-percha free carriers of each alloy were mounted in epoxy blocks and wet-ground (600 grit SiC) to expose the maximum longitudinal section. Small diameter wires of stainless steel (ASTM F138-86) (n=5) and high purity Ti (n=5) were used as controls. They were mounted and surface finished in the same manner as test specimens. Approximately 52% of the area of each carrier and control specimen was then covered with a Plexiglas crevice former. Specimens were placed individually in 50 mL of 0.9% aqueous NaCl solution, at 37°C. Elemental analysis (ppb) of the corrosive solutions was conducted at 2, 6 and 10 weeks, using Inductively Coupled Plasma-Mass Spectroscopy (ICP-MS). Test solutions were replaced completely with 50 mL of fresh electrolyte at each measurement time. Testing will continue for 42 additional weeks. Carriers (n=5) and controls (n=5) of each alloy were also tested according to a modified ASTM Designation: F 746-87, for critical potential (CP) for crevice corrosion. All data were analyzed using the Wilcoxon two-sample rank sum test. The corrosion rates (ICP-MS) for the stainless steel and Ti-6Al-4V alloy carriers were not significantly different from their respective controls ($p \geq 0.11$). Accelerated testing of the Ti materials yielded CP's >+800 mV (SCE). The stainless steel alloys had CP's that were not significantly different ($p=0.16$).

Studies on glutamate catabolism via the 2-oxoglutarate pathway in *Fusobacterium nucleatum*.

R. L. WHITE*, S. E. GHARBIA, R. SETH, A. L.. DOHERTY, M. RAMEZANI, and H. N. SHAH
(Departments of Chemistry and Oral Biology, Dalhousie University).

We have shown previously that three pathways exist for the dismutation of glutamate to acetate and butyrate in *Fusobacterium* species. These experiments were carried out using ^{14}C- labeled glutamate and assay of key enzymes of these pathways. Among these species, *F. nucleatum*, a key periodontal pathogen, appears to catabolize glutamate primarily via the 2-oxoglutarate pathway whereas other species utilize two or three pathways for glutamate degradation. In the present study, the end products of glutamate metabolism were isolated as their Na^+ salts by addition of 3M NaOH to give a final pH 9.5 prior to lyophilization of the spent broth. The lyophilized residue was refluxed in ethanol with r-bromophenacyl bromide and the resulting ρ-bromophenacyl esters were separated by silica gel chromatography using a solvent system comprising methylene chloride and carbon tetrachloride. The separated phenacyl esters were subjected to NMR analysis at 62.9 MH_z using a Bruker AC250 spectrometer. The high ^{13}C enrichment (8.6 times natural abundance) observed in the C-1 of butyrate is consistent with the 2-oxoglutarate pathway, whereas the smaller enrichment (2.6 times natural abundance) at C-3 of butyrate is most likely due to the synthesis of butyrate from acetyl CoA. These results further support the existence of the 2-oxoglutarate pathway in *F. nucleatum* but additional experiments involving glutamate labeled at positions 2,3 or 4 are required to exclude the occurrence of other catabolic pathways.

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Validity of Caries Diagnosis in Pits and Fissures. A. ISMAIL, D. WOTTON*, R. ANDERSON, R. BANNERMAN,T. BORAN, G. DOYLE, R. MacDONALD and W. MacINNIS. (Faculty of Dentistry, Dalhousie University)

The purpose of the in vitro study is to test the validity of visual only, visual with magification, and visual-tactile examinations of dental caries in pits and fissures of posterior teeth. 37 extracted teeth provided surfaces with sound, sound and stained, questionable, non-cavitated or cavitated carious. To identify the area to be examined drawings of each tooth were enlarged and the areas to be examined were indicated. Six dentists examined the teeth independently. The first 2 dentists visually classified each area into one of the six categories. The second 2 dentists used a magnifying lens (2.5X) and the last 2 dentists used a visual and tactile method to classify the tooth surfaces. The examinations were repeated after one week. Visual examinations were completed before the visual-tactile examinations to reduce the potential of physical damage to the tooth surfaces. After examination, the teeth were sectioned using a hard tissue microtome and examined under a polarized light (40X). Poor agreement was found between the clinical examinations and histological diagnosis (Kappa ranged between 0.18 and 0.36). The percentage of clinically sound tooth surfaces diagnosed with caries in enamel ranged between 68.0% and 88.9%. No differences in agreement with histological diagnosis or misdiagnosis of sound surfaces were found among the three methods. The percentage of sound surfaces diagnosed histologically as carious in dentin ranged between 5.6% and 28.0%. The agreement between clinical status of cavitated carious surface and histological examination ranged between 81.3% and 100.0%. Diagnosis of pit and fissure caries is problematic and the visual-tactile method is not more valid than visual examination alone.

A Comparison of Root Canal Leakage Study Methods. K. A. ZAKARIASEN*, K. L. ZAKARIASEN, M. M. MCMINN, (Dalhousie and Marquette Universities)

Research indicates that the various leakage study methods may not produce the same results when comparing obturation techniques. This research compares the results derived from three methods comparing leakage for two obturation techniques. Forty single canal teeth were selected, the crowns removed and the canals prepared. The roots were coated, except for the apical foramina and coronal canal openings, and randomly distributed into 2 groups of 20. One group was obturated with lat. cond., the other with an experimental technique. Each group was divided into 2 groups of 10 each. A lat. cond. and an exper. group were submerged (under vacuum) in 1% methylene blue (M.B.) dye for 7 days. Apical and coronal linear dye penetrations were then measured, the roots dissolved in HNO_3 and spectrophotometric determinations of leakage volume calculated. The second lat. cond. and exper. groups were submerged (under vacuum) in India ink (I.I.) for 7 days followed by clearing and linear measurement of both ap. and cor. leakage. The M.B. linear, M.B. volumetric and I.I. linear methods all indicated no significant differences ($p = .05$) in leakage between lat. cond. and exper. methods. However, the p values for the lat. cond. and exper. group comparisons for the three methods showed large variation and the correlation co-efficients for linear and volumetric values were only $r = .197$ (lat. cond.) and $r = .626$ (exper. group). In addition, linear dye penetrations shown by M.B. were significantly greater than those shown by I.I. ($p = .002$ and $p = .004$). The results indicate that (within the same groups and between comparable groups) the three study methods can provide very different data regarding root canal leakage. This could have profound effects on the conclusions drawn from leakage studies.

Refining Scanned Laser Fluorescence as an Enamel Research Method.
K. L. ZAKARIASEN*, J. R. BARRON, B. E. PATON.
(Dalhousie and Marquette Universities).

Laser-induced fluorescence has potential for both caries research and diagnosis. However, research has shown that precise control is necessary for accurate enamel comparisons. Such factors as focus, number of sampling points, scan matrix size and reproducible placement of the scan matrix can all have substantial effects on results. This research seeks to achieve reproducibility by closely matching enamel window and scan matrix sizes, focus control and high sampling frequency within matrices. Five non-carious third molars were mounted in acrylic and ground so that a flat Bu enamel surface was created. Each enamel area was covered with acid-resistant varnish except for a window approx. 1.0 mm^2. Each specimen was mounted in the fluorometer for scanned laser fluorescence analysis using a 1.0 mm^2 scan matrix. Readings were taken at 50 μ intervals for a total of 400 readings per matrix. Each specimen was re-mounted and re-analyzed in the fluorometer. Each specimen was lased (1.5 W, 1.5 mm f.s., 0.15 sec.) and reanalyzed as per above by fluorometer. Running-sums of line means were calculated for each scan and graphed. In all samples, the curves for non-lased enamel were lower and widely separated from those for lased enamel. In each specimen, the non-lased curves were either coincident or nearly so. Lased curves showed similar characteristics. ANOVA comparing scans for each specimen showed significant differences ($p < .001$) between non-lased and lased enamel scans. The results indicate that ensuring constant focus, repeatable orientation of scan matrices and high sampling frequency collectively can lead to a high degree of scanned laser fluorescence reproducibility.

Optimizing conditions for bio-electrical impedance measurements for physiological studies on *Porphyromonas gingivalis*. H. N. SHAH, ZHANG* MIN, GHARBIA S. E
(Depts. Oral Biology & Biology, Dalhousie University)

Preliminary experiments using bioelectrical impedance measurements to study the utilization of amino acids/peptides among putative periodontal pathogens have revealed specific patterns of conductivity curves. This system utilized a dual electrode cell and a fixed frequency of 10 KH_z. It is clear, however, that improvements can be made to the system to eliminate such problems as electrode polarization. In the present study we have constructed a multiple electrode cell and examined some of the parameters which affect the accurate recording of conductimetric changes. The response of *Porphyromonas gingivalis* to nitrogenous substrates was used to monitor these effects. Fluorescamine labeling of substrates was used to confirm substrate concentrations (Gharbia & Shah, Curr. Microbiol, (1991),**22**,159-163). A LCR (HP 4284A) meter which recorded 40 simultaneous readings in a frequency range of 100 H_z to 800 KHz was used to measure conductivity. The effect of ionic strength of the buffer used on medium impedance spectrum was extracted from the resistance curve in relation to the molarity of the buffering system. The results showed that bioimpedance properties varied with the buffer concentration. Thus in 0.1 M Tris, there was significant reactance below 1KHz, whereas at 0.01 M, reactance existed both at low and high frequencies. Higher ionic strengths increased electrode polarization and therefore affected the medium's apparent impedance. The electrode polarization was considerably reduced by using a multiple electrode cell with inter-electrode spacing (cm) of 1.7, 3.6 and 5.5 for a 8 cm cell. Although essentially similar trends to the physiological properties of this species were obtained, it was concluded that both frequency and electrode polarization should be considered in impedance measurements, and that the use of a multiple electrode cell greatly increases the accuracy of bioimpedance measurements. (This work was supported by MRC Grant # DG-411).

Marco Chiarot Top of the Table.

Marco Chiarot 4th year DDS student won the national competition for table clinics held by the ten Canadian Dental Schools. Marco's topic was based upon his research which involves using finite elemental analysis to study stress distribution in materials and tissues. Marco's Faculty supervisor for the table clinic was Jim Johnson. Marco has been involved in research with the Division of Biomaterials for the past 3 years. During this time he has studied biocompatibility of materials using cell culture, the creep (plastic flow) of soft polymers and (computer) finite analysis of different cavity designs relative to the stress transferred to the tooth structure from the restoration. Marco has worked on these research projects with supervisors Choong Foong, Jim Johnson, Derek Jones and Graduate Student Brian Smythe. Marco's reward for winning the national Table Clinic competition in Calgary was a trip to compete for the North American title in Orlando. Marco is one of twelve students from Dalhousie who hope to be presenting research papers at the IADR meeting in Chicago next March. Marco's success shows what is possible for those students who have developed a strong interested in research.

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Success

A Faculty member at age 25 has about 73,000 hours of work ahead of them before retirement at age 65. Unless of course they are a workaholic in which case they may have 150,000 hours. How much they broaden their scope of knowledge and how much they add to the knowledge base, reflects their degree of success as an academic. Most of our Faculty members have less than 36,000 hours left. Think about it. How many hours do you have left? Today is the first day of the rest of your life. No age or time of life, no position or circumstance, has a monopoly on success. Any age is the right age to start doing! Stewart B. Johnson once said "Our business in life is not to get ahead of others, but to get ahead of ourselves." Remember that when everybody is somebody, then nobody is anybody. Remember also that success is never final and failure never fatal. It's courage that counts. Those who choose the beginning of a road, also choose it's destination. Are you heading in the right direction?

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Only the Beginning?

When we look at our excellent achivements in research, we should perhaps heed the words of Winston Churchill, who wrote: "Now this is not the end. It is not even the beginning of the end. But it is, perhaps, the end of the beginning."

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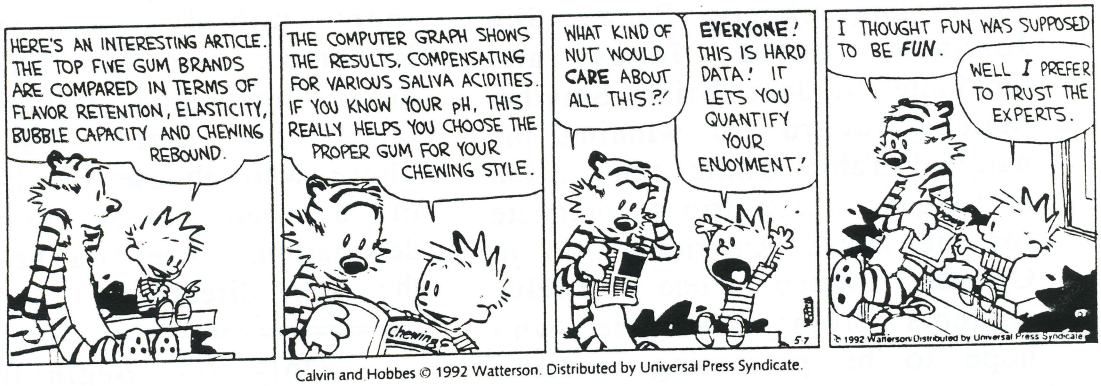
Wrigley Research Funds

Wrigley Canada Inc. has provided the CFDE with funding to annually sponsor awards to support dental students working on a research project. Preference is given to those projects involved with the benefits of salivary stimulation and caries reduction. However, CFDE will entertain other innovative research ideas linked to the benefits of chewing sugarfree gum. The 1993 competition will be the fourth year that the funding has been made available. In 1992 awards were granted to students at the Universities of Alberta, Dalhousie and Manitoba for their summer student research projects. A total of three awards of $3,333 per student will again be available for the summer of 1993. The submission must be made by a faculty member with the name of a student who will be involved. The application should provide an outline of the scope of the project, the background, research objectives, methodology, hypothesis and the significance and timeliness of the project. The deadline for the application to be in Ottawa at the CFDE office is the 28th February 1993. Further details can be obtained from the Dental Research Development Office. As a help in stimulating ideas a draft proposal for a research project involving 'GUM' has been developed by the RDO and is reproduced below.

How to Make Your Research Dollars Stretch a Little Further.



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An Idea for Your Wrigley Research Project

Hard to Understand

"Genius is the ability to reduce the complicated to the simple." -
- C.W. Ceram

Mindful

"Man's mind streached by a new idea never goes back to its original dimensions."
Oliver Wendell Holmes.