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Degradation properties and ion release characteristics of Resilon® and phosphate glass/polycaprolactone composites

Aims (1) To characterize the degradation pattern and ion release characteristics of bioactive glass/polycaprolactone composites; (2) To compare the degradation behaviour of composites containing phosphate glass to that of Resilon®; (3) To determine the potential to control the degradation of the composites by modifying the iron content of phosphate glasses.
Methodology The degradation behaviour of Resilon® and 7 different polycaprolactone/phosphate glass composites (0.2 glass volume fraction), each containing a different glass composition (from the following range of variants: $[CaO]_{0.40}$-$[Na_2O]_{0.05–0.09}$-$[Fe_2O_3]_{0.01–0.05}$-$[P_2O_5]_{0.5}$) was determined in two aqueous solutions (buffered distilled water, HBSS (Gibco, Paisley, UK). Three replicates were performed per composition or material over a seven-day period using weight change measurement (Mettler Toledo AG204, Ca, USA), ion chromatography (Dionex, Camberley, UK), light microscopy (LM) and scanning electron microscopy (SEM). The change in weight was measured and a relative ranking of the samples was obtained.
Results An initial increase in weight of the samples was followed by loss for all phosphate glass/polycaprolactone composites, whilst the Resilon® remained relatively constant. Ion chromatography showed that the phosphate glass/polycaprolactone composites released amounts of iron, sodium, calcium and phosphate ions into solution depending upon the glass composition. The Resilon® samples released only sodium and calcium ions that tended to plateau after 2–4 days. The visual methods (LM, SEM) showed precipitate formation on the surface of the phosphate glass/polycaprolactone composites and in contrast, little on the surface of Resilon®. The precipitate was also evident on the semi-protected specimens (to simulate canal conditions) of Resilon® but was denser at the margins; in contrast, the precipitate on the phosphate glass/polycaprolactone was confined to the free margins and was more evident with a width of 300–700 μm.
Conclusions The degradation pattern of the composites containing phosphate glass was different from that of Resilon®. The phosphate glass/polycaprolactone composites encouraged precipitate formation on the material surface, particularly when semi-protected. Phosphate glass/polycaprolactone composites may have potential as bioactive root fillings.

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Bacterial community analysis of different apical segments of the root and periapical tissues by Denaturing Gradient Gel Electrophoresis

Aim To analyse the bacterial community in different apical segments of the root and periapical tissue by Denaturing Gradient Gel Electrophoresis (DGGE).
Methodology Fourteen roots of extracted teeth with apical but no marginal periodontitis, intact pulp chambers, absence of cracks and acute signs and symptoms, were evaluated after immediate cold storage upon extraction. The thawed tooth was decontaminated using validated protocols and then tissue samples were obtained from different apical parts of the roots. The apical 6 mm were divided into equal halves transversely; the canal in each half was filed to obtain canal lumen (CL) samples (CL1-apical 3 mm, CL2-coronal 3 mm); the remaining root portions were cryo-pulverised to yield apical root (AP) samples (AP1-apical, AP2-coronal). DNA was extracted from each of the 4 samples per root and the 16S rRNA genes were amplified by PCR. The amplicon profile was determined by DGGE (BioRad Laboratories, Hercules, USA). The electrophoretic banding patterns were imported into Adobe Photoshop® and aligned. Presence or absence of the unique patterns were converted to binary data and used to create similarity matrices (using PAUP, Sinauer Associates, Sunderland, USA), which were depicted (using TREEVIEW) as dendrograms to show the relatedness of the recovered taxa. A non-parametric Wilcoxon Signed Ranks Test was used to compare bacterial richness among the samples.
Results The PCR was positive in 49/59 of the samples; all negative results were in the cryopulverised group. DGGE revealed 27 different band patterns (taxa) amongst all samples. The band profiles suggested that each apical infection was unique in its distribution along the canal length. Taxa in the cryo-pulverised samples were a sub-set of those in the canal lumen. There were no significant differences in bacterial richness between the apical and coronal samples (CL1 vs. CL2; AP1 vs. AP2), whilst the canal lumen samples (CL1, CL2) had significantly ($P < 0.05$) higher bacterial richness than their corresponding cryo-pulverised root samples (AP1, AP2). Bacterial fingerprinting revealed a unique band profile in the apical 3 mm of 2/14 (14%) CL samples.
Conclusions DGGE can provide a simple means to analyse the distribution of bacterial communities within an infected root canal.

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Effect of canal preparation and residual root filling material on root impedance

Aim To investigate the effect of root canal preparation and residual root filling material during root canal re-treatment, on the impedance characteristics of extracted, human roots, using an *in vitro* model.
Methodology Thirty extracted, human single-rooted teeth were mounted (with roots immersed in 0.1 mmol KCl) and used in a custom-made apparatus constructed from Perspex/acrylic that allowed strict temperature control at 25 °C. Impedance measurements of the roots were made with a file in the root canal acting as the internal electrode, using a Frequency Response Analyser (Voltech TF 2000, Voltech instruments Ltd, Didcot, UK). The measurements were made under three different canal conditions: (1) *before* chemo-mechanical preparation; (2) *after* chemo-mechanical preparation with rotary nickel-titanium GT files (#30 apical size / 0.08 taper) and irrigation with 2.5% sodium hypochlorite and 17% EDTA solutions; (3) *after* root filling removal with instruments and chloroform to re-establish patency (following placement of gutta-percha and zinc oxide/eugenol sealer using the Continuous wave technique). The measurements were taken at 0, 0.5, 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10 mm coronal to the apical terminus and also 0.5 & 1 mm past the apical terminus. Impedance values were calculated and viewed using Nyquist plots. Comparisons were made within each tooth between measurement points along the length of the canal, as well as under the different canal conditions. Equivalent circuit modelling was performed to determine circuit composition in teeth and surrounding tissues.
Results The impedance decreased from the coronal to the apical levels and showed a rapid drop at the apical terminus under all canal conditions. Chemo-mechanical preparation of the root canal decreased the impedance, while residual root canal filling material on the canal walls resulted in a substantial increase in impedance. Equivalent circuit modelling showed that the equivalent circuit remained consistent at the tested positions within the canal regardless of canal condition but the circuit component values changed with the impedance.
Conclusion Impedance decreased in a corono-apical direction, with a marked drop at the apical canal terminus. Chemo-mechanical preparation decreased the root impedance and residual root canal filling material on the canal walls substantially increased the impedance. The findings help explain the behaviour of apex locators under tested conditions.

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In situ immunocytochemical colloidal gold probing of three bacterial species in the root canal system of teeth associated with apical periodontitis

Aims (1) To establish a protocol for immunocytochemical colloidal gold labelling of bacteria in tooth samples; (2) To use the method to map the distribution of specific microorganisms (*Porphyromonas gingivalis, Fusobacterium nucleatum, Treponema denticola*) in extracted human teeth associated with apical periodontitis.
Methodology Twenty carious teeth with apical but no marginal periodontitis were carefully extracted and immediately fixed with either 0.5% glutaraldehyde or 4% paraformaldehyde (both in 0.1 M cacodylate) and processed for electron microscopy. Primary antibodies tested for sensitivity and specificity gave positive results with pure cultures of *Fusobacterium nucleatum* ATCC 25586, *Treponema denticola* ATCC 35405 and *Porphyromonas gingivalis* ATCC W50 (American Type Culture Collection, Rockville, Maryland, USA) – positive controls. Sixteen teeth showing bacterial presence in light microscopy, were processed for transmission electron microscopy (TEM). Ultra-thin sections, cut from each third of the root were labelled with anti-bodies for the selected species. Controls without the primary antibody confirmed absence of labelling of tissue components by the secondary antibody (negative controls). Recognizable bacterial cells associated with the probes were taken as a positive result; the number of labelled bacterial cells was enumerated for each of the coronal, middle and apical thirds.
Results Necrotic or degenerating tissue was evident in one or more segments of the root. The bacterial presence in 16/20 samples was evident mainly as a bacterial biofilm of variable thickness, comprising of cocci, rods and filaments, sometimes accompanied by polymorphs in the apical third. The bacterial presence was generally heavier in the coronal two-thirds but sometimes uniformly so along the entire length. Immunocytochemical probing revealed all three species in the sample teeth, though *Porphyromonas gingivalis* was the most prevalent. No obvious trends were discernible with regard to distribution along the length of the canal, although that of *F. nucleatum* and *T. denticola* was similar.
Conclusions The protocol for immunocytochemical colloidal gold labelling of bacteria in human teeth was established. Bacterial biofilms were commonly thicker coronally than apically. *Porphyromonas gingivalis, Fusobacterium nucleatum* and *Treponema denticola* were detected in most root segments but the distribution did not follow a discernible pattern. *P. gingivalis* was the most prevalent.

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A simulated 'biofilm' *ex vivo* model to evaluate the influence of canal dimension and irrigation variables on the efficacy of irrigation

Aims (1) To devise an *ex vivo* model to test the efficacy of irrigation in removing a simulated 'biofilm' in root canals; (2)

to test the influence of root canal dimensions, repeated irrigation and mode of irrigation (static and dynamic) on removal of the simulated 'biofilm' using the model.

Methodology Forty human teeth with single straight canals were randomly allocated to two groups for static (n = 20) or dynamic (n = 20) irrigation. The root canals were prepared to different apical sizes (20, 40) and tapers (0.04, 0.08). The teeth were split longitudinally to give two halves. Stained collagen was applied on the canal surfaces in a standard manner and then the tooth was re-assembled in a silicone matrix for dynamic or static irrigation. Digital images of the canal surface were taken before and after irrigation with 9 mL, 18 mL, 27 mL & 36 mL solution. Static irrigation consisted of simple placement of an irrigation needle (gauge 30, Max-I-Prob, Maillefer-Dentsply, Ballaigues, Switzerland), in a fixed orientation 4 mm short of the working length. Dynamic irrigation consisted of the addition of push-pull agitation of the placed irrigant with a well-fitting, tapered gutta-percha point (SybronEndo, Orange, USA) matching the canal dimensions. The percentage of area of canal surface covered with stained collagen was quantified using a software package (ipWin4®). The data were analysed using paired-t tests and linear regression models.

Results All the five explanatory variables: 'volume of irrigant used', 'mode of irrigation', 'orientation of side port of needle', 'coronal-apical level of canal' and 'root canal dimension' had significant ($P < 0.001$) influence on outcome of irrigation. The corono-apical level of canal emerged as the most dominating factor. After irrigation, the apical third had 19.9% and 33.8% less area covered with the simulated 'biofilm' than the middle and coronal thirds, respectively.

Conclusions The test model established that all the explanatory factors had significant influence on the efficacy of irrigation. The stained organic collagen applied on the canal surface could not be removed completely by either static or dynamic irrigation. The dominant influencing factors were ranked in the following order of decreasing priority: corono-apical level of root canal, apical size and taper of canal preparation, and dynamic/static irrigation.

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Mandibular block or buccal infiltration for pulp anaesthesia in molars?

Aim To compare outcome data from the same volunteers participating in two randomised controlled double blind crossover studies: (1) Efficacy of articaine or lidocaine buccal infiltration on mandibular molar pulp anaesthesia, (2) Efficacy of slow or rapid lidocaine inferior alveolar nerve block (IANB) on mandibular molar pulp anaesthesia, to gain insights into the relative effectiveness of these two approaches.

Methodology Healthy adult volunteers (n = 27) received IANBs of 2 mL lidocaine 2% with 1 : 80 000 epinephrine slowly over 60 s or rapidly over 15 s, in a randomised order separated by at least 1 week. In a separate trial, the same volunteers received buccal infiltrations of 1.8 mL 2% lidocaine and 4% articaine (both with 1 : 100 000 epinephrine) in randomised order adjacent to a mandibular first molar, separated by at least 1 week. Seventeen volunteers received IANBs first. First molar pulp sensitivity was assessed electronically before injection and at 2 minute intervals until 10 minutes, at 20 and 30 minutes post-injection. Changes from baseline response and number of episodes of no sensation on maximal stimulation (80 μA) were analysed by McNemar, and Paired t Tests ($P < 0.05$).

Results Change from the baseline pulp tester reading at first sensation was significantly greater after articaine than lidocaine buccal infiltration (mean 25.5, SD 22.2 and mean 15.4, SD 21.2 respectively, paired sample test, $t = 6.6$ $P < 0.001$). The change was also greater after slow than rapid IANB (mean 24.5, SD 17.2 and mean 20.2, SD 18.3 respectively, paired sample test, $t = 2.7$ $P = 0.007$). There was no significant difference in the change from baseline pulp tester readings at first sensation when articaine buccal infiltration was compared with slow IANB (paired sample test, $t = 0.5$ $P = 0.63$). Articaine buccal infiltration produced more episodes of no response to maximal pulp stimulation (80 μA) than lidocaine infiltration (96 cases vs. 52 cases respectively, McNemar test $P < 0.001$). Similarly, slow IANB produced more episodes of negative response than rapid IANB (80 cases vs. 58 cases respectively, McNemar test $P = 0.002$). There was no significant difference between the number of episodes of negative pulp response between articaine buccal infiltration and slow IANB (McNemar test $P = 0.097$).

Conclusions Data from two randomised controlled double blind crossover studies in healthy volunteers suggests that buccal infiltration with articaine may be as effective as IANB with lidocaine for mandibular molar pulp anaesthesia.

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Quality of root canal treatments performed by undergraduate dental students on single-rooted teeth in Glasgow Dental School and Hospital

Aim The aim of this investigation was to assess the technical quality of root canal treatments performed by undergraduate dental students on single rooted teeth at the Glasgow Dental Hospital and School.

Methodology Utilising student records, case notes of those patients who underwent root canal treatment by undergraduate dental students in the Glasgow Dental Hospital and School in 2003–2004 were retrieved. The first 100 case notes to meet the selection criteria were chosen for investigation. The radiographs were examined with even illumination in a darkened room at x2 magnification by two observers. Root canal fillings were classified according to the relationship of the root canal filling to the apex of the tooth. They were classified as: 'Acceptable', where the root canal filling material was within the root canal system and within 2 mm of the radiographic apex; 'Under-filled' where the root canal filling material was >2 mm short of the radiographic apex; 'Over-filled' where the root canal filling material was extruded

beyond the radiographic apex. From the radiographs, the presence of fractured instruments, voids, or perforations was noted. The obturation technique was recorded from the case notes.

Results A total of hundred teeth were examined. Seventy-two teeth were from the maxilla, and twenty-eight were from the mandible. All canals had been prepared using a balanced force technique, and canals were obturated using cold lateral compaction with gutta percha and sealer. There was no evidence of fractured instruments. One tooth (1%), had a perforation and twenty teeth (20%), had voids present within the root canal filling. Of the remaining 79 teeth, 63 (80%) were classified as being 'acceptable', 4 (5%) were classified as being 'under-filled', and 12 (15%) were classified as being 'over-filled'. The root canal treatments had been carried out by thirty-five students in their fourth year and sixty-five students in their fifth year.

Conclusion This study has shown that in Glasgow Dental School, 63% of root canal treatments carried out on single rooted teeth by undergraduate students were obturated to an appropriate length with no voids, perforations or fractured instruments.